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Investigating the effect of PrP genotype on production traits in Charollais sheep

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Introduction Prion protein (PrP) genotype controls susceptibility to scrapie in sheep (Baylis, 2005) and a number of other transmissible spongiform encephalopathies including Creutzfeldt-Jakob disease in humans. Sheep have five PrP alleles (ARQ, ARH, AHQ, VRQ and ARR) and up to 15 PrP genotypes. In an attempt to control and eventually eradicate scrapie the GB National Scrapie Program (NSP, 2004) promotes the use of rams with scrapie resistant PrP genotypes (e.g. ARR/ARR) and discourages the use of scrapie susceptible genotypes (e.g VRQ carriers). Little is known about the effects of PrP genotype on sheep biology in general and commercially relevant production traits in particular. This study aims to determine whether PrP genotype is associated with production traits in a number of UK commercial sheep breeds, and results are reported here for the Charollais breed.

Materials and methods The population studied comprised 29,574 lamb performance records from commercial Charollais flocks, collected from 1997 to 2004. Phenotypic measurements included live weight at 8 weeks (W8W) and scanning (USWT, ~20 weeks), ultrasonic muscle depth (UMD) and fat depth (UFD). All traits were normally distributed except UFD, which was normalised by a square root transform prior to analysis. PrP genotype at codons 131, 154 and 171 was obtained from the National Scrapie Plan Administration Centre and merged with performance records for each animal. A total of 3575 PrP genotypes were matched to performance records. Traits were analysed with mixed models using ASReml (Gilmour *et al.*, 2002). Fixed effects fitted for all traits were: flock code, year of birth, sex, litter size reared, dam age, 'genotyped or not' and PrP genotype. Age at weighing was fitted as a covariate for W8W and age at scanning was fitted for all ultrasonic traits. Birth date ('seasonal') was fitted as a covariate for all traits to capture climatic and other seasonal or management differences throughout the lambing season. Interactions were fitted for flock code x year and seasonal x year. Random effects included direct genetic, dam genetic, litter (LIT) and dam permanent environmental (PE) effects.

Results Genetic parameters from the best-fit model for each trait, as judged by the likelihood ratio test, are shown in Table 1; these were in broad agreement with published estimates. Ungenotyped animals had significantly lower values ($p<0.01$) for all traits, however the PrP genotype effect never reached significance. Trait predictions for each PrP genotype are shown in Table 2.

Table 1 Genetic parameters for the analysed traits

Trait		Random effects	σ_p	h_d^2 (se)	h_m^2 (se)	PE^2 (se)	LIT^2 (se)
W8W	kg	Direct + PE + litter + dam	15.08	0.12 (0.01)	0.04 (0.01)	0.07 (0.01)	0.28 (0.01)
USWT	kg	Direct + PE + litter	32.32	0.21 (0.02)	-	0.05 (0.01)	0.14 (0.01)
UMD	mm	Direct + litter + dam	6.614	0.25 (0.02)	0.02 (0.01)	-	0.09 (0.01)
SqrtUFD	$mm^{0.5}$	Direct +litter	0.113	0.27 (0.02)	-	-	0.12 (0.01)

σ_p ,phenotypic variance; h_d^2, direct heritability; h_m^2, maternal heritability; PE^2, ratio σ_{PE}/σ_p^2; LIT^2, ratio of LIT^2/σ_p^2

Table 2 Estimated means for each trait and PrP genotype

Genotype (n)/Trait	W8W (se)	SWT (se)	sqrtUFD (se)	UMD (se)
ARR/ARR (n=1813)	23.32 (0.12)	50.48 (0.20)	1.91 (0.01)	28.66 (0.10)
ARR/ARQ (n=1360)	23.18 (0.13)	50.28 (0.21)	1.89 (0.01)	28.75 (0.10)
ARR/VRQ (n= 139)	23.48 (0.33)	51.58 (0.59)	1.97 (0.04)	29.18 (0.27)
ARQ/ARQ (n= 197)	23.06 (0.27)	50.42 (0.45)	1.90 (0.03)	28.81 (0.21)
Minor† (n= 66)	22.47 (0.46)	49.96 (0.75)	1.94 (0.05)	28.96 (0.35)
Ungenotyped (n=25,999)	21.92 (0.08)	49.96 (0.12)	1.88 (0.01)	28.35 (0.06)

†several rare genotypes

Conclusions Significant evidence of differences between genotyped and ungenotyped animals was found, suggesting that breeders preferentially genotype better performing animals. However, differences in performance amongst genotyped animals were not significant. In particular, alterations to the PrP genotype frequencies [following the move from the ARQ to the ARR allele] in this breed will have negligible impacts on lamb performance in this breed. Further studies will examine other commercial sheep breeds and additional traits for associations between production traits and PrP genotype.

Acknowledgements We wish to thank our funders Defra, MLC and the sheep breeders involved in this study.

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Association between polymorphisms of the ovine prion protein gene and lamb performance traits of Scottish Blackface sheep

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Introduction Several studies investigating associations between prion protein (PrP) genotypes and performance traits in several sheep breeds have recently been published. Most of these studies have failed to give conclusive results due to small numbers of animals used or a potential genotyping bias as those animals selected for PrP genotyping were chosen based on their performance (e.g. De Vries *et al.*, 2005). Polymorphisms of the PrP gene has been linked with susceptibility to scrapie with the ARR allele generally associated with resistance and the VRQ with susceptibility. This study investigates the associations with PrP genotype for a wide range of lamb performance traits in experimental flocks of Scottish Blackface in which all animals have been PrP genotyped.

Materials and methods Data were obtained from two flocks raised under typical Scottish hill environmental and management practices. A total of 7,138 animals born in 1999 through 2004 with genotypes for polymorphisms at codons 136, 154 and 171 were utilised. Traits studied were live body weight (at birth, BW, marking, MW, weaning, WW, and before slaughter, SW), ultrasonic measurements (muscle and fat depths), carcass traits (weight, CW, and conformation class), age at slaughter (Age) and Computer Tomography (CT) predicted killing out percentage (KO%) and carcass components (weights of muscle, carcass fat, internal fat and bone). A linear mixed model with random direct and maternal additive genetic effects and maternal permanent and temporary environmental effects was fitted for all traits except for CT traits. The CT predicted traits were analysed with a simpler animal model with direct additive and maternal common environmental effects. Several fixed factors were tested for each trait. These factors were sex, type of birth and rearing, age of dam, flock, date of birth, grazing paddock at marking and at weaning, genetic line, age at measurement and carcass fat class and all their two way interactions when biologically sensible. Only significant fixed effects and PrP genotype were included in the final models. Five analyses differing in PrP genotype classification were carried out for each trait. In the first analysis animals were categorised into nine classes, one per genotype. In the other four analyses (one analysis per allele) animals were categorised into classes depending if they carried 2, 1 or 0 copies of a particular allele.

Results Fitting all nine genotypes revealed no significant effect of the PrP genotype on any of the traits (results not shown). Likewise, there was no significant effect of the number of ARR alleles on any of the studied traits (Table 1). Lambs homozygous for the ARQ allele were consistently lighter from birth to slaughter and had lighter CW than the heterozygous ARQ lambs, though the differences were only significant for BW and SW. Lambs without the AHQ allele had lighter live body and carcass weights than heterozygous AHQ lambs, reaching statistical significance for SW. For the VRQ allele, non-carrier lambs needed about 10 days less finishing time to attain the targeted fat class but their carcasses were ca. 0.50 kg lighter compared with VRQ carrier lambs. For the ultrasonic measurements and CT predicted KO% and weights of carcass components the differences between PrP genotypic classes were not statistically significant (results not shown).

Table 1 Least square means and their standard errors (SE) for live body weights and slaughter and carcass traits from analyses with different PrP genotypic classes[1,2]

Allele	No.	BW	MW	WW	SW	CW	Age	Records
ARR	**2**	3.52	16.50	26.32	37.98	16.69	206.2	651
	1	3.52	16.53	26.32	38.00	16.78	204.7	3128
	0	3.49	16.49	26.20	37.82	16.75	207.7	3359
ARQ	**2**	**3.48^{b}**	16.47	26.15	**37.64^{b}**	16.67	206.7	2517
	1	**3.52^{a}**	16.52	26.31	**38.05^{a}**	16.82	205.7	3482
	0	**3.52^{ab}**	16.54	26.37	**38.13^{ab}**	16.80	206.9	1139
AHQ	**2**	3.50	16.22	26.43	**38.40^{ab}**	17.11	213.5	49
	1	3.53	16.58	26.42	**38.37^{a}**	16.97	208.0	1091
	0	3.50	16.50	26.23	**37.87^{b}**	16.74	205.8	5998
VRQ	**1**	3.49	16.53	26.08	38.50	**17.21^{a}**	**216.4^{b}**	141
	0	3.51	16.50	26.27	37.91	**16.75^{b}**	**205.9^{a}**	6997
SE	**Min**	0.03	0.10	0.24	0.47	0.22	4.49	
	Max	0.08	0.34	0.55	0.83	0.40	8.19	

[1]Means within a column and allele class sharing a common superscript are not significantly different (P>0.05) with adjustment for multiple tests using Bonferroni correction. [2]BW, MW, WW, SW, CW are in kg and age in days.

Conclusions Although a few significant associations were found, PrP genotypes may be considered as not associated with the majority of the lamb traits studied. Most importantly, there was no significant association between performance traits and the ARR allele which is of particular importance in the current National Scrapie Plan policy (NSP).

Acknowledgements Financial support from Defra and SEERAD is greatly appreciated.

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Analysis of association between the PrP gene and live weight and slaughter traits in an experimental flock of Swaledale sheep in the UK

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Introduction Genetic selection based on polymorphisms identified at codons 136, 154 and 171 of the prion protein (PrP) gene is being implemented in scrapie eradication programs (e.g. Defra, 2005). These programs aim to remove the VRQ allele (associated with the highest incidence of classical scrapie) and increase the frequency of the ARR allele (which confers the highest level of resistance). However, concerns remain about how selection on the PrP gene would affect other economically important traits. The objective of this study was to investigate potential associations between the PrP alleles and lamb traits in an experimental flock of Swaledale sheep, a hill sheep breed.

Materials and Mmethods The dataset included animals born between 2000 and 2004 (all of which were PrP-genotyped) in a flock maintained by ADAS at Redesdale in Northumberland, UK. There were about 1,450 PrP-genotyped lambs with live weight records (in kg) at birth (BW), marking (at ~30 days old, MW) and weaning (at ~120 days old, WW), and 620 genotyped lambs with slaughter records. The latter included slaughter (non-fasting, SW) and carcass (CW) weights and age at slaughter (SA in days). The time of slaughter was determined by the assessment of fat cover on the live animal. Mixed model univariate analyses were carried out using ASReml (Gilmour *et al.*, 2002). The effect of the PrP gene and other fixed effects that were statistically significant were included in the final model for each trait. Significance of random effects (direct genetic, maternal genetic and maternal permanent environment) was determined based on the log likelihood ratio test. Five analyses differing in the classification of PrP genotype were done for each trait: 1) ten classes, one for each genotype; 2) ARR/ARR, ARR/XXX and XXX/XXX; 3) ARQ/ARQ, ARQ/XXX and XXX/XXX; 4) AHQ/AHQ, AHQ/XXX and XXX/XXX; and 5) VRQ/VRQ, VRQ/XXX and XXX/XXX. Allele XXX represents any non-ARR, non-ARQ, non-AHQ and non-VRQ allele for analyses 2, 3, 4 and 5, respectively.

Results Table 1 shows the number of lambs per genotype and the estimated means from analysis 1. The overall effect of PrP genotype was not significant (P>0.10) for any of the traits in this analysis. Analyses 2, 3, 4 and 5 also produced non-significant results as none of the contrasts between genotype classes reached the significance threshold derived from Bonferroni correction for multiple tests. As an example, contrast estimates from analysis 2 are shown in Table 2.

Table 1 Number of lambs and estimated means (and standard error) of analysed traits from analysis 1

	Number	BW	MW	WW	SW	CW	SA
ARR/ARR	375	3.32 (0.07)	12.4 (0.2)	28.6 (0.5)	38.5 (0.9)	17.1 (0.4)	236.8 (8.2)
ARR/ARQ	132	3.32 (0.07)	12.2 (0.2)	28.5 (0.5)	38.6 (0.9)	16.9 (0.4)	244.1 (8.4)
ARR/AHQ	179	3.36 (0.08)	12.5 (0.2)	28.7 (0.5)	38.7 (0.9)	17.0 (0.4)	248.6 (9.0)
ARR/VRQ	72	3.27 (0.09)	12.5 (0.3)	28.8 (0.6)	40.6 (1.1)	17.8 (0.5)	250.9 (10.3)
ARQ/ARQ	235	3.33 (0.07)	12.5 (0.2)	29.1 (0.5)	39.4 (0.9)	17.4 (0.4)	239.3 (8.1)
ARQ/AHQ	49	3.19 (0.10)	12.1 (0.3)	29.7 (0.6)	39.1 (1.1)	16.9 (0.5)	235.8 (10.9)
ARQ/VRQ	95	3.17 (0.09)	12.1 (0.3)	28.5 (0.5)	38.9 (1.1)	17.3 (0.5)	241.6 (10.1)
AHQ/AHQ	248	3.37 (0.07)	12.5 (0.2)	29.2 (0.5)	39.6 (0.9)	17.5 (0.4)	238.0 (8.6)
AHQ/VRQ	25	3.26 (0.13)	12.1 (0.4)	29.2 (0.8)	39.0 (1.4)	17.6 (0.6)	244.0 (13.8)
VRQ/VRQ	59	3.20 (0.11)	12.3 (0.3)	29.0 (0.6)	40.0 (1.1)	18.0 (0.5)	246.0 (11.0)

Table 2 Estimates of differences between means (and standard error of difference) from analysis 2

	BW	MW	WW	SW	CW	SA
ARR/ARR-ARR/XXX	-0.01 (0.05)	0.08 (0.14)	-0.05 (0.30)	-0.48 (0.49)	0.03 (0.23)	-9.85 (4.67)
ARR/XXX-XXX/XXX	0.01 (0.03)	-0.09 (0.10)	-0.44 (0.20)	-0.44 (0.34)	-0.31 (0.16)	7.26 (3.48)
ARR/ARR-XXX/XXX	0.01 (0.06)	-0.01 (0.14)	-0.50 (0.31)	-0.92 (0.51)	-0.28 (0.24)	-2.60 (4.65)

Conclusion This study indicates that there is no significant association between the PrP gene and the performance traits analysed. The results are valuable as all lambs born in the years of analysis were genotyped and the dataset consisted of a relatively high frequency of the rarer alleles (namely AHQ and VRQ). This contrasts with previous studies where the range of genotypes available was more limited.

Acknowledgements Financial support from Defra and SEERAD is greatly appreciated.

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The importance of animal-derived foods in supplying very long chain n-3 polyunsaturated fatty acids to the UK diet

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Introduction The benefits of adequate intakes of very long chain (VLC, carbon chain length ≥20) n-3 PUFA, in particular eicosapentaenoic acid (EPA, C20:5) and docosahexaenoic acid (DHA, C22:6) are well documented and crucially include a reduced risk of coronary heart disease. Although fish oils are a rich source of these fatty acids consumption of oily fish is low and it is now clear that *in vivo* synthesis of EPA and DHA from dietary α-linolenic acid is very limited, especially in men. This work assessed what the current dietary sources and intakes of EPA and DHA are, and how intake may be increased by enriching their concentration in foods of animal origin.

Materials and methods Estimates of mean consumption of fish, fish products, milk, meat and their products and eggs were obtained from several government and industry sources (MAFF, 2001; DEFRA, 2004; NDNS, 2003; SACN/COT, 2004; BEIS, 2005). Some of the data were based on whole families, some on adults only and some only represented a part of the total consumption of some products. In addition, some data were transformations of other data, e.g. SACN/COT (2004) adjusted earlier data (NDNS, 2003) on oily fish consumption downwards by excluding canned tuna. The data set arrived at for adults aimed to be representative of the present and include all the key contributions from animal products. Typical concentrations of VLC n-3 PUFA in normal and enriched foods were obtained from current studies in this laboratory and the literature.

Results The key sources contributing to the current mean VLC n-3 PUFA intake by adults (244 mg/person/day) are summarised in Table 1. The mean intake is in broad agreement with other estimates in the literature but is very much lower than the current recommended value of 450 mg/person/d (SACN/COT, 2004). Of the total mean EPA+DHA intake, approximately 54% is provided by oily fish although poultry meat makes a valuable contribution and contributes most (72%) of all meats. It is however critical to note that only 27 % of the adult population consume any oily fish and thus for the majority of the adult population the daily intake will at best be only 113 mg with about 40% of this from animal-derived foods. Table 1 also shows estimates of the dietary impact of enriching animal-derived foods with VLC n-3 PUFA by animal nutrition and indicates that milk (mainly via cheese), eggs and poultry meat could make very valuable contributions of about 70, 55 and 141 mg/person/day.

Table 1 Estimated mean intakes of EPA and DHA by adults in the UK from normal and enriched foods

Food			EPA+DHA in food (mg/g)		EPA+DHA intake (mg/d)	
Class	Sub-class	Intake (g/7d)	Normal	Enriched	Normal	Enriched
Fish	Oily	50	18.4	-	131	-
	Total fish	*217*			*199*	*199*
Meat	Beef & veal	245	0.116	0.293	4.1	10
	Poultry	369	0.50	2.3	26	121
	Total meat	*1103*			*36*	*141*
Eggs	Table & others	194	0.32	2.0	8.8	54
Milk	Semi-skimmed	870	0	0.105	0	13
	Cheese, full fat	97	0	1.7	0	24
	Total milk products	*1606*			*0*	*71*
				Grand total	***244***	***465***

Conclusion Although data on food consumption are difficult to interpret, it is clear that currently, intake of VLC n-3 PUFA by the UK adult population is substantially sub-optimal. In the vast majority of the population that consume no oily fish, animal-derived foods make a valuable and perhaps crucial contribution. Assuming current levels of consumption, enrichment of animal-derived foods with VLC n-3 PUFA by animal nutrition could play a major role in public health nutrition in the future. This will of course depend on a sustainable supply of VLC n-3 PUFA for use in animal diets and this process being accepted by consumers.

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Nutritional status of ewes in early and mid pregnancy: 1. Effects of plane of nutrition on ewe reproduction and offspring performance

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Introduction Alterations *in utero* result in adaptations that can alter the physiology of adult offspring a process known as "foetal programming". Recent studies have shown that in adolescent ewes, low planes of nutrition in early pregnancy result in better reproductive performance (Annett and Carson, 2004). In adult ewes, responses are less clear. Some studies indicate no effect of nutrition level in early pregnancy on subsequent lamb output (Annett and Carson, 2004), whereas others indicate that low planes of nutrition in early pregnancy, reduced lamb output at weaning (Smeaton *et al*., 1999). Differences between studies may be due to residual effects of early pregnancy feeding on mid pregnancy. Consequently, the aim of this study was to evaluate the effect of different planes of nutrition in early and mid pregnancy on maternal and offspring performance.

Materials and methods Ninety-nine multiparous Greyface and Texel x Greyface ewes (live weight 76.3, s.d. 6.3 kg; body condition score 3.8, s.d. 0.1), were assigned to one of 3 early pregnancy treatments balanced for live weight (LW), condition score (BCS), ewe breed and sire. Ewes were bred to a synchronised oestrus and from day 0 to 39 post mating (early pregnancy, EP) were fed grass nuts to provide either 175% (High, H), 100% (Medium, M) or 60% (Low, L) of predicted ME requirements for maintenance (AFRC, 1993). From day 40 to 90 (mid pregnancy, MP), ewes were further allocated to rations providing 80% (Medium, M) or 140% (High, H) of ME requirements. Within each treatment, ewes were either treated or not supplemented with selenium. Results for the effect of selenium will be presented on a subsequent paper (Muñoz *et al.* 2006). After day 90, all ewes were fed to meet requirements for late pregnancy. Throughout the study LW and BCS of ewes were recorded fortnightly. Lamb LW were assessed within 24 hour of birth, at 3 and 6 weeks of age, and fortnightly thereafter until weaning. The data were analysed using the Genstat REML procedure, looking at the main effects of the treatments while adjusting for the effect of ewe breed and sire. For lamb LW gain (regression), litter size and sex were also included. Categorical data were analysed by binary logistic regression using LogXact.

Results There were no significant interactions between early and mid pregnancy treatments therefore, only main effects are presented. Plane of nutrition during early and mid pregnancy resulted in significantly different ewe LW and BCS change from day 0 to 39 and day 40 to 90 of gestation (Table 1). The proportion of pregnant ewes (average values of 0.93) and litter size were similar, irrespective of treatment. Ewes on a L plane of nutrition during early pregnancy had a longer gestation compared with those on H plane ($P<0.05$), with ewes on a M plane intermediate. Lambs born from ewes with H plane of nutrition during early pregnancy achieved higher growth rates ($P < 0.05$) than lamb born from ewes on M or L plane diets. Rates of lamb mortality (including stillborns) were not affected by the treatments. Outputs of weaned lamb were similar for each of the treatments.

Table 1 Effect of early and mid pregnancy nutrition on ewe and lamb performance

	Early Pregnancy Nutrition					Mid Pregnancy Nutrition			
	L	M	H	S.E.	Sig	M	H	S.E.	Sig
Ewe live weight change (g/d)									
Day 0 - 39	-151[a]	-10[b]	151[c]	17.5	***	-	-	-	
Day 40 - 90	94[a]	55[b]	-1[c]	11.4	***	14	85	9.3	***
Day 91 -138	384	347	342	25.7		379	336	20.8	P=0.054
Ewe condition score at lambing	3.66	3.72	3.71	0.055		3.64	3.75	0.045	**
Ewe reproductive performance									
Litter size	2.00	1.98	2.00	0.200		1.95	2.04	0.161	
Length of gestation (days)	147.2[a]	146.8[ab]	146.1[b]	0.42	*	146.8	146.6	0.38	
Lamb viability and output									
Birth weight (kg)	5.45	5.23	5.44	0.126		5.40	5.35	0.101	
Growth rate to weaning (g/day)	278[a]	272[a]	293[b]	9.1	*	284	278	7.1	
Lambs weaned/ewe lambed	1.83	1.57	1.59	0.192		1.61	1.72	0.155	
Litter weaned (kg)/ewe lambed	71.2	60.0	63.4	7.84		63.0	66.7	6.31	

Conclusion In adult ewes, plane of nutrition in early and mid pregnancy had no effect on reproductive performance. However, a high plane diet during early pregnancy resulted in lambs with higher growth potential.

Acknowledgments This study is supported by the European Union Programme of High Level Scholarships for Latin America, (Alβan), scholarship Nº E04D047780CL and DARDNI.

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Lamb viability is improved by supplementing Docosahexaenoic acid for a specific time period during late gestation

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Introduction Previous investigations have shown lamb vigour to be improved by including long chain omega-3 fatty acids in maternal diets during late pregnancy (Capper *et al*., 2003; Dawson and Edgar 2005; Pickard *et al*., 2005). However, these trials fail to elucidate whether there is a period in late gestation when supplementing diets with omega-3s would be optimal. Omega-3 fatty acids, particularly docosahexaenoic acid (DHA), are found in high concentrations in brain and nervous tissue (Arbuckle and Innis, 1993), and are therefore required at times of neural and brain tissue growth. This study explored the effects of feeding an algal source of EFAs, with a high content of DHA, during different time periods on measures of lamb viability.

Materials and methods 48 twin-bearing English mule ewes were allocated to treatment groups, matched for condition score, at 9 weeks prior to predicted lambing date. Ewes were fed, during different time periods, either a control diet based on silage and a commercial ewe concentrate feed, or a similar diet containing algal biomass (AB, 18.9%DHA) to provide 12g DHA/ewe/day. There were 4 treatment groups: ewes fed solely on the control diet for 9 weeks prior to lambing (**C**); ewes fed the C diet for the first 3 weeks of the trial followed by 3 weeks on the AB diet, before being returned to C diet until lambing (**3wk**); ewes fed the C diet for the first 3 weeks of the trial followed by AB diet until lambing (**6wk**); and those receiving AB diet for 9 weeks up to parturition (**9wk**). After lambing all ewes received the standard concentrate. At parturition, the level of assistance (0 = no assistance, 1-3 scale for increasing intervention) required by each ewe, the time taken for lambs to stand and to suckle, and lamb birth weight were recorded. Lambs which had not suckled within 2 hours of birth were assisted to do so. Colostrum samples were taken from each ewe before lambs suckled. Lamb weights were monitored unitl weaning. The statistical unit was a pen of 3 ewes during gestation, but ewes lambed in individual pens. Analysis of variance was used to compare treatment effects on ewe and lamb measures, except for lamb time to suckle which was analysed non-parametrically using a Kruskal-Wallis test.

Results AB supplementation tended to increase gestation length, however this was not significant. There were no significant differences in the level of assistance given to ewes at birth or in lamb birth weight between groups, either with or without the inclusion of gestation length as a covariate. Lambs requiring assistance at birth were found to take longer to first stand (Mean level 0 = 17.9min, level 1 = 30.0min, SED 3.03, $P<0.01$). There were no differences between treatment groups in the time taken for lambs to stand after birth. Lambs born from ewes in the 3wk group suckled significantly sooner after birth than lambs born from ewes of the C group ($P<0.05$). Lamb growth rates to weaning did not differ significantly between treatments.

Table 1 Effects of feeding DHA-rich algal biomass during different periods in late gestation

	C	3wk	6wk	9wk	SEM	Sig
Gestation Length (d)[1]	146.1	146.9	148.1	148.3	0.79	NS
Birth wt (kg)[1]	5.6	5.1	5.0	5.2	0.26	NS
Time to stand (min)[2] §	20.8	20.3	22.7	21.4	4.12	NS
Median time to suckle (min)[2]	92.0	46.5	66.0	69.5	-	<0.05
24hr weight gain (kg/day)[1]	0.18	0.24	0.14	0.07	0.11	NS

[1]Analysed with ewe as the experimental unit [2]Analysed with lamb as the experimental unit
§ with level of assistance given at birth included in the model ($P<0.01$)

Conclusion Ewes fed algal biomass for 3, 6 and 9 weeks in late pregnancy gave rise to lambs that suckled sooner after birth than those of ewes in the C group. In a previous experiment (Pickard *et al.,* 2005) latency to stand was found to be reduced in lambs from mothers who received algal biomass for at least 6 weeks during pregnancy, however this result was not emulated here, where birth weights of control lambs were higher. Similarly to the present study, Capper *et al*. (2003) showed a decreased latency to suckle in lambs from ewes fed DHA from fish oil for 6 weeks before birth, and also found data on latency to stand to be indeterminate. Combining results from these experiments suggests that there may be a 3 week time window when foetal development is most susceptible to improvement by feeding DHA during gestation.

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Nutritional status of ewes in early and mid pregnancy: 2. Effect of selenium supplementation on ewe reproduction and offspring performance

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Introduction Nutritional programming during early foetal life may play an important role in determining lamb vigour at birth, the development of the immune system and subsequently, lamb viability. However, information on the effect of macro and micro nutrient status of ewes during early pregnancy is limited. To address this, a research programme has been initiated at this Institute to investigate the effect of plane of nutrition and mineral status on foetal and lamb development and viability. The aim of the current study was to evaluate the effect of selenium supplementation from 14 days pre- to 90 days post-mating on maternal and offspring performance.

Materials and methods Ninety-nine multiparous crossbred ewes, described by Munoz *et al.* (2006) were assigned to one of two treatments balanced for live weight (LW), condition score (BCS), ewe breed and sire. Ewes were bred to a synchronized oestrus and treatments where applied from day -14 to 90 post-mating and consisted of control (C), receiving no supplement; and selenium supplement (Se), receiving 1g of Selplex® (selenium intake of 0.5mg Se/ewe/day). Diets were based on grass nuts, offered at 3 different levels (high, medium and low plane of nutrition), barley (a vehicle for selenium) and straw, during the first 39 days of gestation and two different levels (medium and high) during mid pregnancy (Day 40 to 90) (Munoz *et al.*, 2006). After day 90 all ewes were fed to meet requirements for late pregnancy and supplemented with a multivitamin and mineral mix. Blood biochemistry, including glutathione peroxidase (GPx) activity, LW and BCS, were recorded fortnightly. At lambing, dystocia and lamb presentation (Dwyer *et al.,* 1999) were recorded. Lamb LW and GPx activity was assessed within 24 hour of birth, at 3 and 6 weeks of age, and fortnightly thereafter until weaning. The data were analysed using the Genstat REML procedure, looking at the main effects of the treatments while adjusting for the effect of ewe breed and sire. For lamb LW gain (regression), litter size and sex were also included. Categorical data were analysed by binary logistic regression using LogXact.

Results Mean GPx activity (99.1, s.d. 87 U/g HB) indicated that ewes at the start of the study had a low selenium status. Ewes supplemented during early and mid pregnancy showed higher (P<0.001) GPx activity at day 40, 90 and 140 post mating (Figure 1). For GPx activity there was an interaction between Se supplementation and mid pregnancy nutrition at day 90 of gestation (P<0.001), showing that Se treated ewes on a high plane of nutrition reached higher GPx levels then the medium plane diets (765 vs 657, S.E. 59.1), differences that were not observed in the C ewes (142 and 157, respectively; S.E. 59.1). In addition, GPx activity at birth of lambs born to Se treated ewes (Table 1) was significantly higher than those born to the C ewes (P<0.001), however these differences were not sustained to weaning. Se supplementation had no significant effect on ewe fertility (average pregnancy rate 0.93) or prolificacy. Supplemented ewes tended (P=0.058) to require less assistance at lambing than C ewes (0.26 vs 0.48; odds ratio (OR) 0.39). The proportion of malpresented lambs and lamb birth weight were not affected by Se treatment; however, lambs born from supplemented ewes reached higher growth rates (P<0.05) than C lambs. Supplementation significantly reduced perinatal mortality (0.03 vs 0.12): OR for mortality at 1 week in supplemented ewes was 0.25 (P<0.05); however, mortality at weaning was unaffected. There were no clinical signs of selenium deficiency in control sheep throughout the trial.

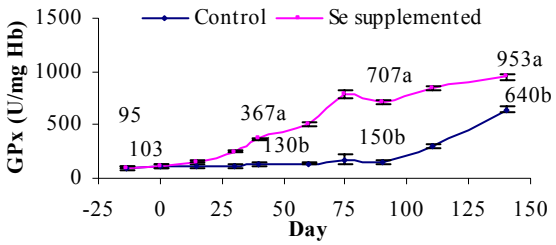

Figure 1 Changes in ewe GPx activity throughout pregnancy

Table 2 Effect of selenium supplementation in early and mid pregnancy on ewe and lamb performance

	Se Supplementation			
	C	Se	S.E.	Sig
Ewe performance				
Lambs born/ewe lambed	1.98	2.01	0.161	
Lambs weaned/ewe lambed	1.59	1.74	0.154	
Lamb performance				
Lamb GPx birth (U/g HB)	682	1099	35.1	***
Lamb GPx weaning	605	673	74.7	
Lamb birth weight (kg)	5.33	5.42	0.100	
Lamb growth rate (g/d)	272	290	7.1	*

Conclusions In adult ewes with low GPx activity, selenium supplementation from day -14 to 90 of pregnancy had no effect on reproductive performance. However, it resulted in lambs with higher viability and growth potential.

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The effect of supplementing pregnant ewes with marine algae or linseed on milk yield, milk composition and lamb growth rate

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Introduction Supplementing pregnant ewes with the *n*-3 polyunsaturated fatty acids (PUFA) found in marine algae and fish oil increases gestation length and improves lamb vigour (Capper *et al*., 2002). However, continuing fish oil supplementation into lactation reduces milk component yield and lamb growth rate (Capper, 2005). The objective of the current experiment was to investigate the potential carry-over effects of feeding marine algae during pregnancy on ewe milk yield and composition during lactation and subsequent lamb growth rate.

Materials and methods Forty-eight twin-bearing, and twelve triplet-bearing Suffolk X Mule ewes were blocked according to age, liveweight, litter size and condition score and allocated to one of six dietary strategies at 103 days of gestation. Ewes were individually penned and housed from day 103 of gestation to 28 days into lactation and fed one of three diets within the strategies. Diets fed during pregnancy contained marine algae (A; C22:6*n*-3 source; *n*=30), whole linseed (L; C18:3*n*-3 source; *n*=20) or Megalac (M; C16:0 source; *n*=10) as the principal fat source. At 12 hours *post partum*, 10 ewes previously fed algae and 10 ewes fed linseed were changed to the Megalac diet, and 10 ewes previously fed algae were changed to the linseed diet. The concentrates were isoenergetic, isonitrogenous and provided 83g fatty acids/kg DM. The concentrate allowance was increased from 0.65 kg/day (0.7 kg for triplet-bearing ewes) at day 103 to 1.15 kg/day (1.20 kg/day for triplets) at day 147 of gestation. Straw was offered *ad-libitum*. At 28 days *post partum* lambs were penned behind a mesh barrier to prevent suckling. Ewes were injected with one ml of oxytocin and hand-milked to empty the udder. Four hours later, the procedure was repeated, milk samples were taken and lambs returned to the ewe. Milk secretion rates were calculated from recorded four-hour milk yields and milk composition analysed by N.I.R.S. Lamb growth rate was estimated by regression applied to liveweight recorded at birth and at 7, 14, 21 and 28 days of age. Data were analysed as a randomised block design using ANOVA.

Results Milk secretion rate was significantly higher in ewes fed strategy AL or AM compared to strategies LL, LM or MM (Table 1). Milk fat concentration was lowest in ewes fed strategy AA and highest in ewes fed strategy MM (P<0.001). The combination of differences in milk yield and composition induced by diet resulted in ewes supplemented with strategies AL or AM having significantly higher milk fat yields than those fed strategies AA or LL. Milk protein concentrations were similar between treatments, although a tendency (P=0.083) for milk protein yield to be increased was observed in ewes offered strategies AL or AM. Furthermore, although lactose concentration was unaffected by treatment strategy, lactose yield was highest in ewes fed strategies AL or AM when compared to those fed LL, Lm or MM (P = 0.021). Lamb growth rate was unaffected by dietary strategy.

Table 1 Effects of supplementing pregnant ewes with various fatty acids sources during pregnancy on milk yield, milk composition and lamb growth

	Treatment Diet[a]							
	AA	AL	AM	LL	LM	MM	s.e.d.	*P*-value
Milk secretion rate (ml/hour)	109	120	126	92.6	92.6	91.3	12.26	0.018
Milk fat concentration (g/kg)	65.2	99.2	92.8	78.9	103	113	9.24	<0.001
Milk fat yield (g/hour)	7.18	11.9	11.6	7.35	9.64	10.4	1.617	0.021
Milk protein concentration (g/kg)	35.4	38.6	38.5	37.5	38.8	41.0	2.00	0.173
Milk protein yield (g/hour)	3.95	4.61	4.80	3.47	3.61	3.76	0.526	0.083
Milk lactose concentration (g/kg)	47.1	48.4	47.7	48.6	47.4	45.5	1.19	0.162
Milk lactose yield (g/hour)	5.26	5.84	6.06	4.49	4.50	4.16	0.631	0.021
Lamb growth rate (kg/day)	0.26	0.29	0.29	0.28	0.28	0.28	0.017	0.504

[a] AA: algae fed throughout pregnancy and lactation, AL: algae fed in pregnancy followed by linseed during lactation; AM: algae fed in pregnancy followed by Megalac in lactation; LL: linseed fed throughout pregnancy and lactation; LM: linseed fed in pregnancy followed by Megalac in lactation; MM: Megalac fed throughout pregnancy and lactation

Conclusions The decrease in milk fat yield conferred by algal supplementation during lactation was negated by dietary change. However, feeding algae during pregnancy exerted carry-over effects upon milk protein and lactose yields at 28 days after the dietary change. This suggests that milk component synthesis in lactation may be affected by the provision of PUFA *pre-partum*, although the biological mechanism for this is, as yet, unknown.

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Maternal nutrition effects on the birth weights of lambs sired by Texel rams that were either carriers or non-carriers of the Inverdale FecX^{I} prolificacy gene

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Introduction Productivity of hill ewe flocks in Britain is dependent on both the genotype and viability of lambs born, with birth weight being a significant determinant of their neonatal viability and subsequent growth. Hill ewes lambing in late Spring frequently endure conditions of moderate to severe feed restriction during early pregnancy in their coincident winter environment. The extent to which such restriction has a negative impact on lamb birthweight, especially where the sire genotype has been selected for improved lean tissue growth, or is offset by safe mobilisation of maternal body reserves will be crucial to subsequent survival of the offspring. Nutrient demands imposed by the foeto-placental unit(s) depend not only on stage of gestation but also on litter size. They may also be influenced by the genetic potential for muscle growth of the foetal genotype, the expression of which is influenced by early pregnancy maternal nutrition (Fahey *et al*., 2005). In the present study we examined whether provision of a diet meeting 0.5 versus 1.0 times ME requirements for maintenance of pregnant ewes up to Day 95 of gestation influenced the birth weights of lambs. A total of 134 ewes lambed, 30 of these with twins. Sires used were Texel rams that were either carriers or non-carriers of the Inverdale fecundity gene (*FecX^{I}*) gene.

Materials and methods Mature Scottish Blackface ewes of mean liveweight 46.5 kg (s.d. 4.75) and body condition score 2.3 (s.d. 0.46) were housed indoors on wood-chip bedding and offered a concentrate ration (DM 873 g/kg; crude protein 174 g/kg; ME 12 MJ/kg DM) in equal feeds at 08.00 h and 16.00 h daily; supplementary chopped hay (200 g/day) and *ad libitum* water were provided also. Ewes were inseminated via laparoscope with frozen-thawed pooled semen from either two Inverdale *FecX^{I}* carrier or two non-carrier Texel rams. Ewes in each 'sire' category were assigned either to a dietary regimen meeting 1.0 (Control, C) or 0.5 times (Restricted, R) ME requirements to Day 95 of pregnancy. Subsequently, all ewes were given the same ration but at levels adjusted to meet the demands of single- and twin-bearing ewes, identified via ultrasound scanning in mid-pregnancy. Between Weeks 4 and 17 of pregnancy, a sub-set of ewes (n=24 per dietary regimen) was blood-sampled to provide weekly non-esterified fatty acid (NEFA) profiles. Data for single males, single females and twins were analysed separately, via General Linear Model (Minitab; Version 14) with diet, sire and 'diet x sire' interaction as factors in the model. Pre-insemination dam liveweight and gestation length were covariates. NEFA data were compared using Repeated Measures analysis (Genstat; Version 8) with diet, sire and 'diet x week' in the model.

Results NEFA profiles (Figure 1) indicated that, during feed restriction, ewes mobilised body fat reserves (P<0.001) and this was discontinued when they were restored to their estimated feed requirements.

Plane of nutrition during the first 95 days of pregnancy had no significant effect on mean gestation length in twins (C vs R = 146.1 vs 146.6) but did influence it in singles (C vs R = 146.5 vs 147.3: P<0.01). Plane of nutrition up to Day 95 and sire genotype effects on birth weight are presented in Table 1. Dietary restriction reduced the birthweight of single male lambs and twin lambs of both genders, with sire having no significant effect. In contrast, among single females, diet was not a significant factor but sire genotype was, with those females that inherited the Inverdale gene being heavier than non-carrier counterparts. Since male lambs cannot inherit this X-linked gene from their sires, the contrasting effects of maternal nutrition on birth weight suggest that the prenatal growth of females carrying the *FecX^{I}* gene was less severely compromised in utero than male counterparts from the same sire.

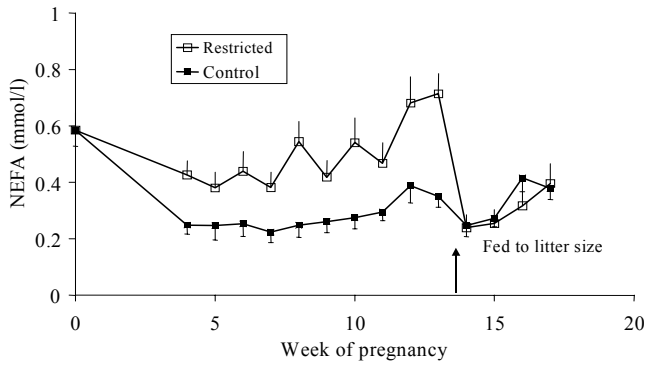

Figure 1 NEFA profiles in relation to maternal nutrition.

Table 1 Lamb birth weights (kg) in relation to maternal nutrition (C=Control and R=Restricted in the first 95 days of gestation) and Sire genotype. There was no Diet x Sire interaction.

Diet/Sire	C/Non-carrier	R/Non-carrier	C/Carrier	R/Carrier	Diet	Sire
Single females	4.32±0.21 (6)	4.36±0.20 (16)	5.20±0.23 (17)	5.02±0.22 (11)	ns	P<0.01
Single males	5.21±0.17 (19)	4.54±0.22 (12)	5.76±0.27 (11)	5.25±0.28 (12)	P<0.001	ns
Twins (m & f)	3.80±0.16 (10)	3.65±0.19 (20)	3.99±0.14 (18)	3.27±0.17 (12)	P<0.01	ns

Conclusions Although it is possible that birth weight advantages among their offspring reflect other features of Inverdale-carrier sires, these data raise the intriguing question as to whether foetuses with the *FecX^{I}* gene are less susceptible to undernutrition, perhaps via influences on placental function. This merits further investigation.

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Effect of different levels of iodine in the diet of pregnant ewes on the absorption of immunoglobulin G (IgG) from artificial colostrum in neonatal lambs

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Introduction The presence of very high levels of iodine in the diet of sheep in late pregnancy is subsequently associated with very low levels of immunoglobulins (Ig) in the blood of the lambs following consumption of colostrum (Boland *et al*., 2005). It remains to be established what minimum concentration of iodine in the diets of pregnant sheep affects IgG absorption, and whether the levels of iodine found in commercial concentrate feeds have an effect. The mechanism by which excessive iodine in the diet of pregnant sheep affects the absorption of Ig in lambs also remains to be established. The aims of this study were to determine the maximum dose of dietary iodine for late pregnant sheep that avoids the inhibition of IgG uptake from a standard quantity of colostrum given to neonatal lambs, and to examine the effects on thyroid hormone concentrations.

Materials and methods Forty-eight twin bearing Mule ewes (Blueface Leicester x Hardy Speckleface) mated with Texel rams and predicted to lamb in the same week were used. Six weeks before the mean expected lambing date, the ewes were allocated to one of four groups, and given a concentrate meal containing either 5.5, 9.9, 14.7 or 20.8 mg of iodine/kg DM. Iodine supplementation was with calcium iodate. The concentrate allocation was 0.22 kg DM/ewe/d, 6 weeks before lambing. This amount was increased at weekly intervals to 0.44, 0.65 and then 0.87 kg DM/ewe/d, and remained at 0.87 kg DM/ewe/d for the final three weeks before lambing. All four groups had free access to grass hay, liquid molasses and water *ad libitum*. The concentrations of iodine in the hay and molasses were 0.16 and 0.29 mg/kg DM. At lambing, the ewe and lambs were moved to a pen, and the lambs were prevented from suckling. Jugular blood samples were taken from each lamb 30 minutes and 24 hours after birth. The lambs were bottle-fed a total of 50g DM/kg liveweight of artificial ewe colostrum in four feeds at 6 hourly intervals during the first 18 hours of life. The colostrum contained 39 g IgG/kg DM. Plasma thyroxine (T4) and tri-iodothyronine (T3) concentrations were determined by radioimmunoassay; IgG concentrations were determined by ELISA. Differences between treatments were tested by analysis of variance, with lambs nested within ewes. When there was a significant overall effect, differences between individual treatments were determined using the least significant difference test.

Results Lamb plasma IgG and thyroid hormone concentrations are shown in Table 1. As the concentration of iodine in the concentrate increased, the concentration of IgG in the plasma decreased ($P<0.001$). Similarly, the efficiency of Ig absorption from that fed decreased as the iodine added to the concentrate increased ($P<0.001$). The reductions in the plasma concentration of IgG and in the efficiency of IgG absorption were significant at 14.7 mg/kg iodine in comparison to the lower inclusion levels ($P<0.05$). The plasma concentration of T3 in the lambs at birth and at 24 hours of age was not significantly affected by the dietary treatments. However, plasma concentrations of T4 were significantly higher in the lambs from ewes given the greater iodine inclusion rates, both at birth ($P<0.01$) and at 24 hours after birth ($P<0.05$). Plasma concentrations of insulin and cortisol were not affected by the treatments ($P>0.1$; results not shown).

Table 1 Plasma concentrations of IgG in lambs at 24 hours, and T3 and T4 at 30 minutes and 24 hours

	Iodine in Concentrate (mg/kg DM)				SED	*P*
	5.5	9.9	14.7	20.8		
IgG – 24 hours (g/l)	6.08^{a}	5.06^{a}	3.18^{b}	3.10^{b}	0.762	<0.001
IgG absorption efficiency (g/kg)	235^{a}	195^{a}	123^{b}	120^{b}	29.4	<0.001
T3 – Birth (ng/l)	4.09	3.46	4.81	4.41	0.538	0.068
T3 – 24 hours (ng/l)	5.86	5.53	6.20	6.38	0.793	0.725
T4 – Birth (μg/l)	10.96^{b}	11.61^{b}	14.69^{a}	14.90^{a}	1.18	0.002
T4 – 24 hours (μg/l)	9.96^{bc}	9.56^{c}	11.81^{a}	11.27^{ab}	0.719	0.011

$^{a, b, c}$Means within rows with different superscript letters are significantly different at $P<0.05$

Conclusions Iodine at 14.7 mg/kg in the concentrate significantly impaired the immune status of lambs in comparison to 9.9 mg/kg in the concentrate, following the consumption of a standard amount of colostrum. This effect was associated with elevated levels of T4. The mechanism by which iodine in the diet impairs the absorption of Ig is unknown, though it seems likely that the thyroid hormones are involved. In pregnant ewe concentrates iodine is typically included at approximately 10 mg/kg DM. In order to ensure that there is no negative effect on lamb IgG levels, there may now be justification for reviewing the iodine inclusion rates of pregnant ewe feeds, particularly in coastal areas that have high rates of iodine deposition from the sea.

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Verifying quantitative trait loci for muscle depth in UK commercial sheep flocks

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Introduction Although a considerable amount of research is being carried out in molecular genetics, few results have as yet found practical application in livestock breeding. In a previous study, quantitative trait loci (QTL) for growth and carcass traits were identified in commercial Texel and Suffolk sheep in the UK (Walling *et al.*, 2004) and in Charollais sheep (McRae *et al* 2005). QTL were identified for muscle depth on chromosome 18 for Texel Sheep and Chromosome 1 for Suffolk and Charollais sheep. The current study aims to verify that QTLs for muscle depth found in commercial sheep are segregating in independent populations, and implement the technology required for the integration of genetic markers into commercial sheep breeding programmes.

Materials and methods Commercial Texel, Charollais and Suffolk sheep with performance and pedigree recor-ding were studied. DNA was collected from nineteen half sib-families (average family size, 67), comprising eight Texel, six Charollais and five Suffolk families. Available traits included eight and 20 week weight, and ultrasonic muscle and fat depth, recorded on the third lumbar at scanning (20 weeks). Of interest in this study were muscle depth and muscularity, i.e. muscle depth was corrected for body weight. Genotyping was done using multiplex micro-satellite marker panels developed around the regions where QTLs were identified in previous studies. For Texels the panel comprised five microsatellites around the distal portion of chromosome 18 and a single multi-plex panel with two microsatelite markers for Charollais and three markers for Suffolk on chromosome 1 was used. Relative marker order and position was verified using Cri-Map. QTL analyses for muscle depth and muscu-larity used half-sib regression interval mapping using QTL Express (Seaton *et al*., 2002). Environmental effects accounted for were: flock, year, litter size born and reared, and age of dam with age at scanning as a covariate. The QTL and polygenic breeding values were also estimated using a variance component approach. This was achieved by firstly, estimating the proportion of genes identical-by-descent (IBD) between all individuals (Pong-Wong, *et al.*, 2001) at the QTL location. Then, estimated breeding values were obtained using BLUP-MAS.

Results Evidence for QTL for muscle depth or muscularity was found at the 0.01 and 0.05 chromosome-wide level in Texels and Charollais, and at the nominal 0.05 level in Suffolks, in the targeted regions. Families with significant evidence for segregating QTLs are shown in Table 1, along with the number of progeny genotyped and the estimated size of effects Evidence of segregating QTL was found in half of the families investigated, and also in families that were not closely related to those in which the QTL were first detected. Total (i.e. QTL + polygenic) breeding values were bimodal in families where the QTL effects were largest.

Table 1 Sire families with possible QTL segregating for muscle depth and muscularity† (QTL express analysis)

Breed	Sire	No. of	Muscle Depth			Muscularity†		
		Lambs	Estimate (mm)	S.E.	T-Value	Estimate (mm)	S.E.	T-Value
Texel	1	115	1.79	0.87	2.07	2.40	0.67	3.60
	2	43	4.29	1.40	3.07	2.41	1.08	2.23
	3	12	4.40	2.05	2.15	3.03	1.57	1.93
Charollais	1	49	1.42	0.70	2.04	1.40	0.76	1.85
	2	91	1.12	0.53	2.12	1.07	0.62	1.71
	3	51	4.10	1.84	2.23	2.84	1.21	2.36
Suffolk	1	69	3.15	1.74	1.81	2.40	1.43	1.68
	2	22	2.28	1.48	1.54	3.01	1.21	2.48
	3	30	2.45	1.23	1.99	1.15	1.02	1.23

†Muscularity (muscle shape) as defined as the depth of the muscle corrected for scan weight

Conclusion This study has successfully confirmed previously detected QTL for muscle depth and muscularity. Significant results in independent families suggest that the QTL may be widespread and not specific to individual families. The provision of estimated QTL EBV will enable the use of QTLs in selection programmes irrespective of the lack of knowledge of underlying genes.

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QTL affecting fatty acid composition of meat in Scottish Blackface sheep

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Introduction There has been an increased interest in recent year in manipulating meat fatty acid composition, because meat is a major source of fat in the diet - particularly saturated fatty acids, which have been implicated in diseases associated with modern life. Conversely, polyunsaturated fatty acids (PUFA), which lower blood cholesterol concentrations, are present at low levels in meat, especially those of the *n*-3 series which are beneficial to health. Sales of meat have remained static or fallen slightly in recent years (Wood *et al.*, 1999). This pressure on sales has caused a reappraisal of the factors which influence the appeal of meat to consumers, which together constitute 'quality', with one of these factors being the fatty acid composition. Like most animal production traits, fatty acid composition is influenced by both genetic and environmental factors, and the aim of the present study was to locate QTL affecting fatty acid composition in Scottish Blackface lamb meat.

Materials and methods The study was carried out from 2001 to 2003 on 300 8-month old male Scottish Blackface lambs from 9 half-sib families, ranging from 12 to 46 lambs per family. Lipids were extracted from 10g duplicate samples of lean muscle, separated into neutral and phospholipid, saponified, methylated, and individual fatty acids by column chromatography, and quantified as described by Demirel *et al*. (2004). DNA from each animal was extracted from blood samples; and genotypes for 150 microsatellite markers distributed over eight chromosomes (1, 2, 3, 5, 14, 18, 20, and 21) were determined. Linkage analyses were undertaken by multiple regression (Knott *et al.*, 1996), fitting year, management group, litter size and year by slaughter day as fixed effects. For each regression an *F*-ratio of the full model, including the inheritance probability, *vs*. the same model without the inheritance probability, was calculated across families. The location with the largest *F*-ratio signified the QTL location, and *F*-ratios exceeding the α=0.05 genome-wise threshold were declared significant. The one-LOD drop method was used to estimate the 95% confidence intervals. Lastly, the proportion of the genetic variance explained by the QTL was estimated as $\sigma^2_{QTL}/\sigma_g^2$, where σ_g^2 (i.e. genetic variance) was estimated from REML analyses of the same data.

Results Highly significant QTL have been observed for fatty acids in five chromosomal regions (Table 1), three for saturated, three for monounsaturated and 10 for polyunsaturated fatty acids. Eight significant QTLs were mapped to the same position on chromosome 21 (57-58 cM), with the same families segregating in all cases.

Table 1 QTL significant at 5% genome level

Trait		Chr†	Location (cM)††	*F*-ratio		Proportion of genetic variance
				5% threshold	1 *vs.* 0 QTL	
Saturated	Myristic acid	21	57 (41, 77)	3.02	**3.14**	0.050
	Palmitic acid	21	57 (42, 88)	2.94	**3.17**	0.077
	Stearic acid	21	58 (43, 85)	3.13	**3.25**	0.066
Monounsaturated	Oleic acid	21	58 (42, 85)	2.98	**3.23**	0.052
	Cis-Vaccenic acid	21	58 (43, 85)	2.99	**3.26**	0.185
	Gadoleic acid	21	21 (52, 106)	3.03	**3.49**	0.090
Polyunsaturated	Linoleic acid	21	58 (43, 85)	3.14	**3.22**	0.027
	Linolenic acid	2	269 (260, 297)	2.89	**3.97**	0.103
		21	57 (42, 88)	3.08	**3.17**	0.079
	CLA	3	159 (148, 173)	2.51	**3.22**	0.125
	Arachidonic acid	21	58 (45, 72)	2.41	**3.84**	0.111
	EPA	2	229 (213, 297)	2.70	**3.05**	0.054
		1	79 (60, 93)	3.00	**3.02**	0.051
	DPA	1	168 (156, 175)	3.19	**3.52**	0.039
		2	87 (77, 117)	3.32	**3.49**	0.038
		21	0 (17, 22)	2.66	**3.07**	0.033

† Chr=Chromosome. †† The one-LOD support intervals are given in parentheses.

Conclusions This study has been successful in detecting QTL related to fatty acid composition on sheep chromosomes 1, 2, 3, 5, 14, 18 and 21; the first such QTL in sheep. This may lead to new opportunities to genetically alter and improve the fatty acid profile in sheep meat.

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Metabolic regulation in bull calves is genetically related to fertility in their offspring

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Introduction Direct selection for fertility is difficult and yields little improvement because traditional parameters have low heritability and can only be measured in the mature female. A potential solution would be the use of an indirect selection criterion that is measurable in young bulls, is heritable and is genetically correlated to the fertility of the bull's female progeny. The objective of this study was in investigate the genetic variation in free fatty acids (FFA), glucose, growth hormone (GH) and insulin concentrations in plasma from Danish male dairy calves following overnight fasting and the genetic covariation with fertility in their daughters.

Materials and methods Plasma samples from 9 month old, fasted (overnight) male (n=800) Red Dane, Danish Holstein and Danish Jersey calves were used. For the current analyses, the mean of two samples, taken 30 mins apart were used. Free fatty acids and glucose were measured using a commercial assay kit based on enzyme colorimetry (Opera, Bayer Germany). Growth hormone and insulin were measured by time-resolved fluoro-immunometric assay (Løvendahl & Purup, 2002; Løvendahl *et al.*, 2003). Estimated breeding values (EBV) for female fertility (FertEBV) calculated by the Danish Cattle Federation (2003) were available for 666 of the calves (estimated from approximately 100 daughters each). The FertEBV links information on the three fertility measures which are: days from first to last insemination in heifers and cows and days from calving to first insemination in cows. Variance was stabilised and distribution properties improved using log-transformation of data for GH, Insulin and FFA. A univariate mixed model was fitted to the physiological data using the average information restricted maximum likelihood (AI-REML) method (Jensen *et al.*, 1997) in DMU, statistics computer software (Madsen & Jensen, 2002). Data for Danish Holstein (DH) alone and Danish Holstein, Red Dane + Danish Jersey (ALL) were analysed separately for each trait. Approximate genetic correlations (r_A) were estimated by correlating the EBVs for the hormone or metabolite with the FertEBV.

Results Heritabilities of glucose, FFA and insulin were moderate in DH and ALL and GH was low in DH and ALL (Table 1). Genetic correlations of FertEBV with glucose and FFA EBVs were negative and significant in DH and ALL.

Table 1 Estimated genetic correlations, heritability (h^2) and descriptive statistics for FertEBV and Glucose (mmol/l), FFA (mmol/l), GH (ng/ml) & Insulin (pmol/l)

Trait	n	Mean	h^2	s.e (h^2)	r_A	P-value
FertEBV	666	96.29				
DH-Glucose	473	4.94	0.19	0.08	-0.13	P<0.05
DH-FFA	475	6.31	0.20	0.08	-0.18	P<0.001
DH-GH	473	2.69	0.06	0.06	0.07	NS
DH-Insulin	477	32.79	0.19	0.08	-0.01	NS
ALL-Glucose	800	4.94	0.30	0.07	-0.14	P<0.001
ALL-FFA	803	6.30	0.17	0.06	-0.12	P<0.01
ALL-GH	803	2.56	0.11	0.06	-0.02	NS
ALL-Insulin	804	32.79	0.23	0.07	-0.01	NS

Conclusion This analysis has provided evidence to show that additive genetic variance is responsible for a substantial proportion of the phenotypic variation in a number of hormones in Danish male calves. Furthermore, these results indicate that glucose and FFA are negatively correlated to fertility. This indicates that on average, male calves with high glucose and FFA at 9 months have female offspring with reduced fertility. Glucose and FFA may therefore be of potential interest to dairy cattle selection programmes to improve female fertility, as a measurement in young bulls. This is of course providing that the high heritability estimates in the present study are confirmed, and the trait is shown to be correlated genetically to high reproductive efficiency in subsequent lactations.

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Measures of luteal activity postpartum to monitor the early postpartum fertility in the dairy cow

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Introduction A phenotypic and genetic decline in dairy cattle fertility has been reported in several countries simultaneous to the increase in genetic merit for production. In addition, an increase in atypical milk progesterone (MP_4) patterns, particularly extended luteal phases, has been observed by Royal *et al*. (2000). One such MP_4 parameter; the interval from calving to commencement of luteal activity (CLA), has been studied by Royal *et al.* (2002a,b; 2003) and is reported to have moderate heritability (h^2), an unfavourable genetic correlation (r_A)to milk yield (r_A; 0.36) and a favourable r_A to calving interval and interval to first service, respectively (0.39, 0.53). Although CLA is a good indicator of onset of luteal activity (LA) postpartum (pp), it does not take account of subsequent ovarian activity. The objective of this study was to investigate an alternative MP_4 parameter using the proportion of MP_4 samples representing LA within the first 60 days pp in both British and Swedish dairy cows.

Material and methods For the present study two MP_4 databases were used. A UK database, previously studied by Royal *et al*.(2000, 2002a,b, 2003; D1) and a Swedish database, also previously studied (Petersson *et al*., 2006; D2). MP_4 samples for D1 (n=1209 lactations) were collected thrice weekly between 1996 and 1999 from Holstein-Friesian cows. MP_4 samples for D2 (n=1049 lactations) were collected twice weekly between 1987 and 2002, from Swedish Red and White cows and Swedish Holsteins. The percentage of MP_4 samples representing luteal activity (PLA) was calculated using samples collected $\leq$ 60 days pp. PLA was calculated for different sampling intervals by using one sample per week (WPLA), one sample every second week (TWPLA) and one sample per month (MPLA). A genetic analysis was conducted using D1, mixed linear models were fitted to the data using restricted maximum likelihood method (ASREML and DMU). A model with fixed effects of herd, year, season, lactation, uterine infection and diet, fixed regression of percent heterosis and percent American Holstein and random effects of herd-year-season interaction, breeding value and the error term were applied.

Results The mean PLA (± se) for cows with delayed first ovulation was 13.5 ± 0.8 and 8.2 ± 0.7 for D1 and D2 respectively. For cows with a persistent corpus luteum in the first cycle (PCL) the mean was 60.2 ± 1.2 and 58.7 ± 2.0 in D1 and D2, respectively. For cows with no atypical MP_4 patterns, mean PLA was 51.7 ± 0.5 and 45.2 ± 0.6. for D1 and D2, respectively. The genetic analysis of D1 estimated h^2 for PLA of approximately 10% higher (see table 1) than that reported previously for CLA (0.16-0.21; Royal *et al*., 2002).

Table 1 Phenotypic means ± se and heritability (se) estimates for the different PLA measures in D1.

Measurement	Mean ± se	Heritability (se)
PLA (%)	47.3 ± 0.6	0.295 (0.058)
WPLA (%)	44.0 ± 0.6	0.247 (0.057)
TWPLA (%)	41.3 ± 0.6	0.201 (0.055)
MPLA (%)	43.8 ± 0.8	0.140 (0.055)

Conclusion The endocrine fertility parameter; PLA, investigated in this study shows potential for use in cattle breeding programmes to monitor and improve fertility. Phenotypically, in addition to encompassing CLA, it also takes account of atypical MP_4 profiles such as PCL. Genetically, it appears to have moderate to high levels of h^2 making it an attractive parameter for genetic improvement, providing the genetic covariation with other traits of economic importance is favourable. This is currently being investigated. Finally, although in theory a potentially attractive fertility parameter, it remains impractical to measure at a commercial level without the use of in-line MP_4 monitoring.

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The effect of genetic selection and feeding system on dairy cow fertility, measured using milk progesterone profiles

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Introduction The ability of high-yielding modern dairy cows to perform in the range of systems found on UK dairy farms and to be profitable and sustainable needs investigating. As part of a project to study the robustness of such animals, milk-progesterone profiling was used to investigate the reproductive performance of two genotypes of dairy cow on two production systems. The use of milk progesterone profiles to give a detailed picture of oestrus cycle activity and fertility in dairy cows has been described by Royal *et al*. (2000).

Material and methods Milk samples were collected three times per week (Monday, Wednesday and Friday), between September 2003 and August 2005, from newly calving cows on SAC's Crichton Royal herd. Samples were taken for the first four months of lactation. The herd comprises 200 cows in two genetic lines (a Selection Line bred for high milk solids and an unselected Control Line), each managed on two systems of production, a high forage system and a high concentrate system. The milk samples were analysed by enzyme linked immunoabsorbant assay (Ridgeway Science Limited) and milk progesterone levels recorded (ng/ml). Routine observations of heats, services and pregnancy diagnoses were also recorded. The following traits were calculated from the progesterone data using the methods outlined by Royal *et al*. (2000); calving to first luteal activity (d; CLA), average progesterone level (ng/ml; APL), length of the luteal phase (d; LLP), cycle length (d; CL) and length of the inter-luteal interval (d; LIL). Other traits were calculated from the farm data; day of first heat (d; DFH), day of first service (d; DFS), number of services per conception (NSC), number of cycles (NC) and number of heats (NH). All traits were log transformed and analysed using generalised linear models (GLM) comprising lactation number, year, genetic group and system as fixed effects where appropriate. Least-squares means were calculated from untransformed data.

Results There was no difference between the two genetic groups in any of the four oestrus cycle traits (APL, LLP, CL and LIL). The High Concentrate system had a shorter luteal phase (12.0 v 13.1d; $P<0.05$) and a longer inter-luteal interval (5.6 v 5.2d; $P<0.01$) than the High Forage group but the net effect was a similar cycle length. The least-squares means for a range of fertility traits for the two genetic groups and systems are shown in Table 1. The Control group commenced cycling activity 6 days sooner after calving than the Selection Line. A similar pattern was seen for the two systems with the High Forage system starting to cycle earlier after calving by 4 days. The two genetic groups had a similar number of services per conception, number of cycles and number of heats per lactation. No significant interactions were found between the main effects in these analyses.

Table 1 Effect of genetic group and production system on 6 measures of fertility (least-squares means).

	APL (ng/ml)	LLP (d)	CL (d)	LIL (d)	CLA (d)	DFH (d)	DFS (d)	NSC	NC	NH
Control	10.9	12.5	22.7	5.2	28*	63**	74	2.00	2.89	2.24
Selection line	10.7	12.7	23.2	5.5	34	75	79	2.02	2.66	2.09
High conc.	10.7	12.0*	22.5	5.6*	33*	72	78	1.89	2.83	2.00*
High forage	10.9	13.1	23.2	5.2	29	67	75	2.12	2.72	2.33

The probabilities of differences between the two genetic groups or between the two systems from the GLM analysis * $P<0.05$, ** $P<0.01$, *** $P<0.001$

Conclusion Measures of fertility are rarely made on different genetic groups or production systems without the confounding effects of farm, climate, soil type and management level. The particular combination of genetic groups and feeding systems at SAC's Crichton Royal farm allows such a comparison. Both selection for high milk production and the use of a high concentrate feed affected fertility in dairy cows. These groups had lower fertility in early lactation than non-selected cows and those on a high forage system respectively. However, oestrus cycle characteristics were not affected by genetic group but varied by feeding system. Progesterone profiling allows a more accurate picture of cow fertility to be studied than farm measurements alone.

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The relationship between body energy traits and functional traits in first lactation cows

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Introduction Many studies have shown the negative impact of high production on functional performance in the dairy cow (e.g. Dechow *et al.,* 2002). It is suggested that the increased capacity for milk production has resulted in the partial "shift" of nutrients away from maintaining functional fitness towards production output. Bull profiles for daughter body energy traits were produced using random regression techniques. The relationship of functional traits with body energy traits across 1st lactation was examined. If body energy is correlated with health and fertility it could indicate that high metabolic stress leads to deterioration in one or more of these traits.

Materials and methods Production and linear type classification data for 311,719 first lactation daughters of 11,736 sires were extracted from the national recording databases (NMR, CIS, HUK). Daily sire solutions for angularity, body depth, chest width, condition score and stature were estimated using random regression models (Wall *et al*., 2005). The daily sire solutions for type traits were used to estimate liveweight (LWT), daily energy content (EC) and cumulative energy balance (CEB) of sires' daughters across first lactation. Daily body energy breeding values were correlated with nationally estimated sire breeding values for somatic cell count (SCC), lifespan (LS), calving interval (CI) and non-return rate (NR). Genetic correlations were approximated from the PTA correlation by accounting for the reliability of the the PTAs.

Results There was large variation in the plots of individual sire profiles for daughter body energy (not shown) showing that the daughters of some bulls end up in positive energy balance and other complete their first lactation in negative energy balance. On average, the CEB was lowest on day 50 of lactation (–191 MJ). Results showed that daughters of the bulls with negative CEB profiles lost large amounts of body energy in early lactation and were unable to regain it during lactation, culminating in a negative CEB at the end of the lactation.

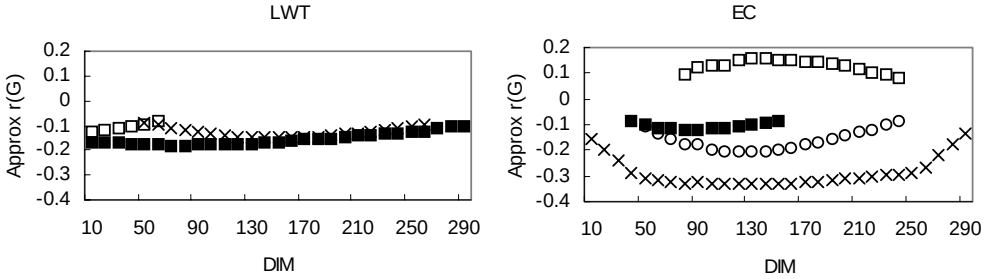

Figure 1 Genetic correlation between body energy and functional traits LS, □; SCC, ○; CI, ×; NR, ■). Results significantly different from zero are shown.

Figure 1 shows the approximate genetic correlation of LWT and EC with functional breeding values (LS, SCC, CI, and NR). Early lactation LWT was correlated with LS with lighter animals with higher growth rates in early lactation more likely to have a higher longevity. Animals that are still growing in early lactation will not have reached maturity by first calving and therefore there could be a partitioning of energy towards early lactation growth and away from milk production with favourable effects on overall survival in the herd. The genetic correlations of EC with the functional traits were similar to those seen with condition score (not shown) and the production and functional traits. This suggests that estimate of body energy content is heavily dependent on the fatness levels of the cow. This is further illustrated by the high correlation between EC and condition score breeding values on any particular day ranging from 0.96 to 0.99. The correlation of CEB with functional traits was lower than correlations of EC with functional traits. CEB was not significantly correlated with either fertility trait. The relationship of CEB with other functional traits was relatively consistent across lactation and was –0.13 with SCC and 0.13 with LS. This suggests that animals with negative CEB across first lactation are likely to have higher somatic cell counts and a shorter lifespan.

Conclusions There are differences between sires in how their daughters grow and utilise body energy across first lactation. These changes could be indicative of animals utilising body reserves to maintain lactation. There is a favourable relationship of liveweight and body energy content with functional traits which could be harnessed in selection with a broad breeding goal that results in improvements in production and functional traits.

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The effect of negative energy balance on expression of the ovarian insulin-like growth factor (IGF) system in the *post partum* dairy cow

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Introduction Subfertility in lactating cows has a multifactorial background, but a significant cause may be the negative energy balance (NEB) associated with early lactation. One effect of NEB is to delay the interval from calving to first ovulation. The likely candidates for mediating the effects of NEB on reproductive function are the metabolites and metabolic hormones that change in relation to physiological and nutritional state. Circulating IGF-I concentrations fall *post partum* and both IGF-I and insulin concentrations influence the interval from calving to first ovulation, follicular oestradiol production, and the number of ovulatory oestrogen-active follicles. Moreover, cows with higher IGF-I concentrations after calving generally ovulate within 35 days. Effects of circulating IGF-I on developing follicles are influenced by IGF receptor concentrations and local production of specific binding proteins (IGFBPs). We postulated that severe NEB compromises the early stages of follicular growth via alterations to the follicular IGF system.

Materials and methods 24 multiparous Holstein-Friesian cows were blocked 2 weeks prior to expected calving according to parity, body condition score (BCS), and previous lactation yield, and randomly allocated to 2 treatments designed to produce mild or severe NEB: MILD were fed *ad lib* grass silage with 8 kg day^{-1} of a 21% CP dairy concentrate and milked x1 daily; SEVERE were fed 25 kg day^{1} silage with 4 kg day^{-1} concentrate and milked x3 daily. Calculations of EB, based on the French NE system (Jarrige 1989), and measurements of BCS were used to select 6 cows from each group for slaughter which showed extremes in EB. Cows were slaughtered on day 6-7 of the first follicular wave after calving, based on daily transrectal ultrasonography. The ovary opposite to that containing the dominant follicle was collected, frozen at –80 degrees C and sectioned for *in situ* hybridisation. Expression patterns of mRNAs encoding follicular IGF-II, type I IGF receptor (IGF-IR), and the IGF binding proteins (IGFBP)-2, -5 and –6 were determined as described previously using oligonucleotide probes (Perks *et al*. 1999). Three sections per cow taken through the centre of the ovary were examined for each probe, two antisense and one sense (control). Optical density (OD) measurements for specific hybridisation to walls of individual small (S, 2.5–5 mm) and very small (VS, 1 - 2.5 mm) follicles were made from autoradiographs using image analysis. Health status of each follicle was assessed from serial sections stained with H & E to determine the morphology of the theca and granulosa. Data were analysed using mixed model analysis with treatment, size and health as factors and post hoc independent Students t-tests.

Results Ovaries were obtained on days 9-16 in the MILD and days 12-15 in the SEVERE groups, when the estimated EB was -2.7 ± 1.41 and -6.1 ± 1.03 UFL day^{-1} respectively and the circulating IGF-I concentrations were 51 ± 8 and 11 ± 1 ng ml^{-1}. Follicle numbers were made up of MILD: 31 VS follicles (12 healthy, 19 atretic) and 21 S follicles (2 healthy, 19 atretic). In the SEVERE group there were: 32 VS follicles (18 healthy, 14 atretic), and 23 S follicles (7 healthy, 16 atretic). Mean expression data (OD ± s.e.m.) for the study is provided in Table 1. mRNA encoding IGFBP-1 and IGFBP-3 was not detectable.

Table 1 Effect of treatment (mNEB / sNEB), follicle health (Healthy / Atretic), and follicle size (VS / S) on expression of the follicular IGF system

	mNEB †	sNEB †	Healthy ‡	Atretic ‡	VS § (1–2.5 mm)	S § (2.5–5 mm)
IGF-II	1.12 ± 0.124 [a]	0.92 ± 0.074 [b]	0.94 ± 0.061 [c]	0.71 ± 0.028 [d]	0.99 ± 0.068 [e]	0.76 ± 0.116 [f]
IGF1R	0.22 ± 0.035	0.21 ± 0.017	0.21 ± 0.017 [c]	0.17 ± 0.015 [d]	0.22 ± 0.018	0.18 ± 0.041
IGFBP-2	0.19 ± 0.025 [a]	0.12 ± 0.024 [b]	0.13 ± 0.016	0.13 ± 0.009	0.15 ± 0.020	0.11 ± 0.023
IGFBP-5	0.84 ± 0.076 [a]	0.60 ± 0.118 [b]	0.31 ± 0.153 [d]	0.56 ± 0.056 [c]	0.37 ± 0.229	0.20 ± 0.035
IGFBP-6	0.31 ± 0.055	0.34 ± 0.036	0.33 ± 0.030 [c] 0.15 ± 0.042 [d]	0.23 ± 0.037 [d] 0.26 ± 0.033 [c]	0.33 ± 0.030 [e]	0.15 ± 0.042 [f]

Means differ significantly (a>b, c>d, e>f; all P<0.05). † Follicles were VS & healthy except for IGFBP-5 (VS atretic).
‡ Follicles were 1–5 mm in diameter, except for IGFBP-6 where VS follicles are the upper values and S follicles the lower values. § Follicles were healthy

Conclusions The two treatments produced the desired differences in the depth of NEB experienced at the time of sample collection. As both circulating IGF-I and follicular IGF-II expression were lower in the SEVERE group, this would be likely to reduce both proliferation and oestradiol production in the VS follicles <2.5 mm. IGFBP-2 and IGFBP-5 expression were, however, also lower. As these IGFBPs are generally considered inhibitory, this would tend to have the opposite effect. Whereas expression of IGF-II and IGF1R decreased in atretic follicles, IGFBP-5 expression increased. IGFBP-6 expression was differentially altered by both health and size. We conclude that EB status alters locally produced components of the intra-follicular IGF system, thus potentially influencing both ovarian activity and fertility.

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mRNA expression of the insulin like growth factor family members in the liver of lactating cows under mild and severe negative energy balance

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Introduction The liver is the principle site of production of both insulin-like growth factor (IGF) and the associated binding proteins (IGFBPs). During lactation, tissue reserves are mobilised and gluconeogenesis in the liver is up-regulated. The high nutrient demands of lactogenesis result in a physiological state of negative energy balance (EB), a situation that has profound effects on the IGF system. It is also known that the IGF family members play an important role in fertility by acting on reproductive tissues. Genetic selection of dairy cows for high yield milk production has led to a decline in fertility and it is thus hypothesised that this disruption is a consequence of down-regulation of certain IGFs and/or their receptors and binding proteins. In this study a qPCR approach was used to quantify the expression levels of specific transcripts of the IGF family in the liver during different situations of mild or severe energy balance.

Materials and methods Multiparous Holstein-Friesian cows (n=24) were blocked two weeks prior to expected calving according to parity, body condition score (BCS), and previous lactation yield, and randomly allocated to 2 treatments designed to produce mild or severe NEB: Mild were fed *ad lib* grass silage with 8 kg day^{-1} of a 21% CP dairy concentrate and milked x1 daily; Severe were fed 25 kg day^{1} silage with 4 kg day^{-1} concentrate and milked x3 daily. Calculations of EB, based on the French NE system (Jarrige 1989), and measurements of BCS were used to select 6 cows from each group for slaughter which showed extremes in EB. Cows were slaughtered on day 6-7 of the first follicular wave after calving, based on daily transrectal ultrasonography. This was 9-16 days *post partum*. Liver samples were taken from each cow and total RNA was extracted in TRIzol reagent. All RNA samples were treated with DNase before converting to cDNA in a randomly primed reverse-transcription reaction. Oligonucleotide primers were designed for members of the IGF family, including their receptors and binding proteins and levels of these traits were determined using SYBR green qPCR: All liver cDNA samples were run in duplicate on the same plate to eliminate inter-assay variation and specificity of the reaction products was confirmed by melting curve analysis and gel electrophoresis. Levels of target transcripts were expressed as amount (fg) per microgram (μg) of reverse transcribed RNA and were compared between energy groups using independent samples t-test. As the liver is the major producer of circulating IGF-I, mean transcript levels of IGF-I were correlated with plasma IGF-I concentrations to further confirm assay validity.

Results Intra-assay variation over the entire quantitation range was between 5.4 and 16.4 % for all gene products examined with qPCR. Circulating levels of IGF-I positively correlated with liver transcript levels and were related to energy balance status as illustrated in Figure 1. Changes in mRNA expression are given in Table 1: IGF-I, IGF1R, IGF2R, IGFBP-3, -5, -6 and GH-R all showed evidence of down-regulation during severe energy balance while IGFBP-2 levels showed an up-regulation.

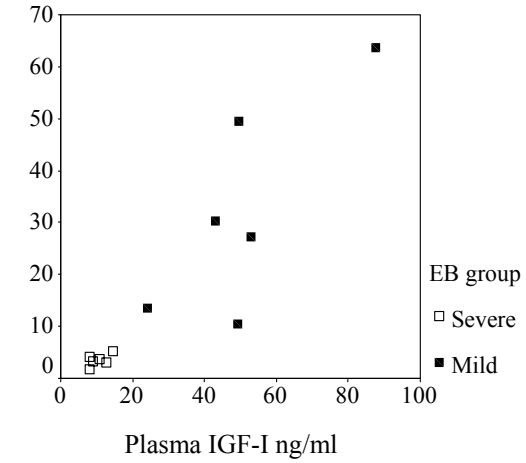

Figure 1 Relationship between liver IGF-I mRNA expression and circulating IGF-I in cows under different conditions of EB. Pearson correlation = 0.905, $P < 0.001$.

Table 1 Levels of liver mRNA expression obtained from cows subjected to different conditions of EB (mean ± S.E.M., n = 6 animals per group).

Gene	Mild EB	Severe EB	P
IGF-I	32.44 ± 8.44	3.55 ± 0.47	0.007*
IGF-II	190.96 ± 43.13	100.28 ± 17.65	0.080
IGF1R	0.78 ± 0.14	0.33 ± 0.06	0.007*
IGF2R	2.10 ± 0.35	1.03 ± 0.18	0.017*
IGFBP1	130.00 ± 59.49	85.60 ± 42.75	0.559
IGFBP2	58.43 ± 8.03	109.53 ± 10.58	0.004*
IGFBP3	18.58 ± 2.83	7.75 ± 1.04	0.005*
IGFBP4	78.74 ± 21.16	33.91 ± 3.35	0.063
IGFBP5	0.59 ± 0.10	0.21 ± 0.02	0.004*
IGFBP6	0.75 ± 0.09	0.36 ± 0.04	0.005*
IR-A	1.47 ± 0.24	0.94 ± 0.10	0.082
IR-B	2.97 ± 0.62	2.06 ± 0.22	0.192
GH-R	56.81 ± 11.51	18.83 ± 1.48	0.008*

*denotes significant difference. IGF type 1 and 2 receptor 1R & 2R; IGFBP1-6 insulin-like growth factor binding protein 1-6; IR insulin receptor -A, -B transcript variant A, B; GH-R growth hormone receptor.

Conclusions A decrease in IGF-I mRNA and corresponding plasma levels, as well as transcripts for the receptors, and several major binding proteins indicates that the system as a whole is down-regulated during severe negative EB. These changes in the liver are likely to alter the effect of the IGF system in peripheral reproductive tissues.

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Changes in caspase activity during the post mortem conditioning period and its relationship with shear force in porcine *longissimus dorsi* muscle

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Introduction Toughness is a determinant of meat quality and a common cause of unacceptability in meat products. Although calpain proteases are believed to be involved in meat tenderisation by post mortem (PM) degradation of proteins within muscle (Sensky *et al*., 2001), other proteolytic systems are likely to contribute to the process (Sentandreu *et al*., 2002). Caspases are proteases involved in protein degradation in apoptosis and are activated early in pathological events associated with hypoxia/ischaemia, not dissimilar to the hypoxic conditions in muscle after slaughter. Caspases cleave a number of proteins which are targeted by calpains during post mortem proteolysis and also degrade the calpain-specific inhibitor calpastatin. The caspase system has a hierarchy of initiating isoforms (such as caspases 8, 9 and 12) which activate effector caspases (such as 3 and 7) that cleave specific substrates. Our hypothesis is that caspase activity may contribute to early post mortem proteolysis and tenderisation, similar to the calpain system. The objective of this study was to investigate the relationship between changes in caspase activity during post mortem conditioning and shear force in pork.

Materials and methods Large White pigs (n=10) were slaughtered and core samples (2 g) of *longissimus dorsi* (LD) muscle were taken from the area of the last rib at 0, 2, 4, 8, 16, 32 and 192 h after slaughter. At 48 h duplicate chops were taken in the region proximal to the sampling area, vacuum-packed and conditioned at 1ºC for 6 d. Samples were snap frozen in liquid nitrogen and stored at -80ºC. Crushed core samples (1g) were homogenised in 3 ml extraction buffer (25mM HEPES (pH 7.5), 0.1% Triton X-100, 5mM $MgCl_2$, 2mM DTT, 74µM antipain, 0.15µM aprotinin, 1.3mM EDTA, 20µM leupeptin, 15µM pepstatin), centrifuged and the supernatant removed for caspase assays. Caspases 3 and 7 activities were measured using Apo-One Caspase 3/7 assay (Promega) and caspase 9 activity with Caspase-Glo luminescence assay (Promega) using a FluoStar Galaxy spectrometer (BMG). Shear force (SF) was determined in the conditioned chops using a Stevens CR Analyzer with Volokevich type jaws (Brown *et al*., 1998). Data were analysed using one-way analysis of variance.

Results Changes in caspase 3/7 and caspase 9 activity were similar, both peaking within the first 4 h after slaughter (Fig.1). By 16 h PM the majority of activity had already occurred, with activity at time points 0 to 16 h all being significantly greater than activity at 32 and 192 h ($P<0.001$). The relationship between caspase 9 and caspase 3/7 activities at all time points was examined and a positive correlation was found between them ($P<0.001$, $r = 0.91$) The relationships between shear force at 8 d and 0:32 h caspase 3/7 and 9 activity ratios were investigated and a significant correlation was found between 0:32 h caspase 9 activity ratio and shear force ($P<0.05$, $r = 0.68$) whilst there was a trend observed between the 0h:32h ratio of caspase 3/7 activity and shear force ($P<0.08$, $r = 0.62$) (Fig.2).

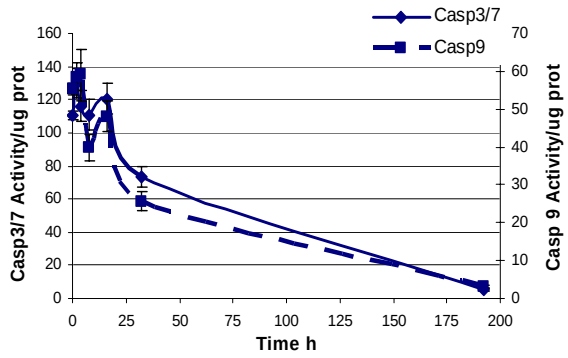

Figure 1 Caspase 3/7 and caspase 9 activity post mortem in porcine LD muscle

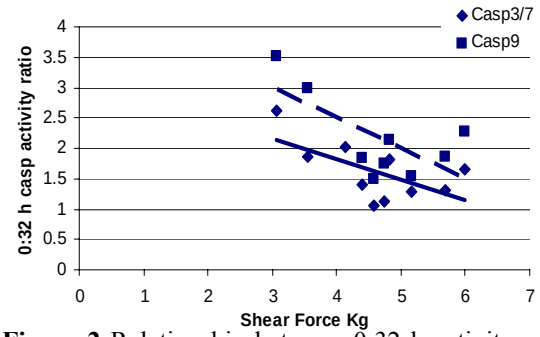

Figure 2 Relationship between 0:32 h activity ratio of caspase 3/7 (r=0.62) or caspase 9 (r=0.68) and SF at 8 d

Conclusion The data indicate that the activity of skeletal muscle caspases changes PM and both initiating (caspase 9) and activating (caspase3/7) members of the caspase proteolytic system are activated early in the meat conditioning period. *In vivo* activation of caspase 9 triggered by oxidative stress in the mitochondria leads to apoptosis through activation of caspase 3/7. The similar pattern of activity and the significant correlation observed between caspase 3/7 and caspase 9 activity observed in this study suggests that caspase 9 could be acting as initiatior caspase cleaving and activating executioner caspases 3 and 7, causing degradation of substrates in the early conditioning period. Shear force values at 8 d were negatively correlated to the 0:32 h activity ratio of caspases indicating that the greater the rate of decrease of both caspase 3/7 and caspase 9 activity over the first 32 h post mortem the more tender the meat. Caspases' activity significantly declined after the first 16 h PM and this together with their relationship with shear force could implicate caspases' involvement in early PM tenderisation.

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The effect of beef genotype, pelvic hanging technique and aging period on the eating quality of some hindquarter muscles

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Introduction The proportion of beef cattle originating from the suckler herd is projected to decrease, relative to the proportion from the dairy herd. Sinclair *et al*., (2001) found no difference between eating quality of Holstein and Charolais when hung tenderstretch. Most genotype comparisons in relation to meat quality have used the *longissimus dorsi* muscle and very few have considered more than one particular post slaughter process (ie hanging technique, aging period). The present study was undertaken to investigate the effect of genotype and post slaughter processing (hanging technique and aging time) on the eating quality of a range of hindquarter muscles.

Materials and methods The experiment involved a total of 40 steers, consisting of two genotypes, Holstein (100% Holstein, Hol) and Charolais (>75% Charolais, CH). All animals were offered grass silage *ad libitum* supplemented with 4.5 kg concentrate for 98 days prior to slaughter. After slaughter, one side of each carcass was randomly allocated to one of two hanging techniques, namely Achilles tendon (AT) or tenderstretch (TS carcass suspended through the aitch bone tendon from 0.75 to 48 hours post-mortem). The sirloin, rump and topside joints were removed from the carcasses of 12 animals of each genotype, and the *longissimus dorsi* (LDa anterior, LDp posterior), *biceps femoris* (BF), *gluteus medius* (GM) and *semimembranosus* (SM) muscles were dissected out 48 h post slaughter. Steaks 2.5 cm thick were cut across the longitudinal direction of the muscle fibres, vacuum packed and aged for either 7 or 21 days at 1^{o}C, then blast frozen and stored at -20^{o}C until required. Prior to eating quality assessment the samples were thawed out at 1^{o}C for 24 h and then allowed to equilibrate to ambient temperature for 2 h prior to grilling. The slices were grilled on a clam shell hotplate until considered 'well done' (78^{o}C internal). Assessments were undertaken by a consumer panel for tenderness, juiciness, flavour and overall liking using a line scale (0-100). The data were analysed using analysis of variance for the main effects of genotype, hanging techniques, ageing period, muscle and their interactions.

Results The effect of genotype, carcass hanging technique, ageing period and muscle on meat eating quality are presented in Table 1. The scores for all sensory attributes were significantly ($p<0.05$) higher for Hol relative to CH. Hanging the carcass TS resulted in significantly ($p<0.001$) higher values for all sensory attributes. All sensory attributes increased significantly ($p<0.001$) when aging was increased from 7 to 21 days. The sensory scores for tenderness, flavour and juiciness were higher in the anterior portion (LDa) than the posterior portion (LDp) of the sirloin. The statistically significant ($p<0.001$) differences due to muscles generally followed expected patterns with the LDa highest in all sensory attributes and the SM lowest. There were statistically significant two factor interactions between hanging technique & muscle for tenderness; between hanging technique & aging period for all attributes; between muscle & genotype for tenderness and juiciness; and between aging period & genotype for juiciness and overall liking. There were significant ($p<0.05$) three factor interactions between genotype, hanging technique and muscle for tenderness and overall quality. The tenderness of the BF was significantly greater for the Hol than the CH for both AT (56.2 & 46.5 respectively) and TS (64.3 & 57.1 respectively), this was not the case for the LDa. The tenderness of the LDa was significantly higher for the Hol than CH when hung AT (56.7 & 49.5 respectively) but not when hung TS (65.4 & 66.8 respectively).

Table 1 Effect of genotype, hanging technique, ageing period and muscle on eating quality

	Genotype				Hanging Technique				Aging Period				Muscle						
	CH	Hol	sed	Sig	AT	TS	sed	Sig	7d	21d	sed	Sig	LDa	LDp	BF	GM	SM	sed	Sig
Tenderness	52	56	2.05	*	51	57	1.06	***	52	55	0.69	***	61	55	56	56	41	1.4	***
Juiciness	48	55	1.69	***	50	54	0.85	***	50	53	0.69	***	56	52	56	51	43	1.1	***
Flavour	54	58	1.54	**	54	58	0.75	***	55	57	0.58	***	61	56	57	58	48	1.0	***
Overall	53	58	1.63	**	53	58	0.87	***	54	57	0.61	***	61	56	57	57	47	1.1	***

Sig statistical significance, * $p<0.05$, ** $p<0.01$, ***$p<0.001$, sed standard error of difference

Conclusion Overall, under these conditions, the eating quality of the Holstein meat was better than the Charolais meat. The results clearly show an improvement in meat eating quality by using the tenderstretch technique and extending the aging period. The interactions show that when comparing genotypes it is important to consider the post slaughter protocol, as this will affect the magnitude of difference between genotypes. The choice of muscle used for comparison is also critical.

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Pork tenderness: analysis of a promoter regulating calpastatin expression

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Introduction Unpredictable variations in meat toughness in the British pig herd remain a concern to the meat industry. Whilst genetics is clearly important, 50% of this variability can be accredited to environmental factors, such as stress. At slaughter the levels of calpastatin (encoded by the CAST gene), a specific inhibitor protein that regulates the calpain proteinases largely responsible for postmortem tenderization, are strongly related to meat toughness. Down-regulation of CAST expression in the days before slaughter therefore provide a practical opportunity to reduce toughness. There are at least three CAST gene promoters (1xa, 1xb and 1u) containing transcription factor motifs sensitive to β-adrenergic stimulation, which can be induced by physiological stress (Parr *et al*., 2004). The 1u promoter is the predominant form in porcine muscle and the resulting CAST mRNA transcripts are increased in animals treated with the β-agonist clenbuterol (Sensky *et al*., 2004) or in cells treated with the analogue dibutyryl cAMP (db cAMP) (Sensky *et al*., 2005). In this study, the functionality of the 1u promoter is further dissected by truncation studies and mutation of potential cAMP responsive elements (CRE).

Materials and methods Eight 1u promoter constructs in the pGL3 Basic vector (Promega) were generated (termed A, B, C, C1, C2, C3, C4 and D, in order of decreasing length), in which potential CREs and other transcriptional motifs were systematically eliminated (Figure 1). Three further constructs containing a mutation in one of the CRE regions of the A construct were also generated (Am1, Am3, Am4). pGL3 with a viral promoter (SV40) and the promoterless pGL3 Basic were used as positive and negative controls, respectively. The constructs were transiently transfected into a rat muscle cell line (L6G8) using GeneJuice reagent (Novagen). Immediately after transfection, the cells were treated with 2 mM db cAMP. Luciferase activities were measured 24 h after transfection using the Dual-Glo Luciferase Assay System (Promega). The data from 8 replicates per construct ± treatment were normalised to untreated pGL3 SV40 data and the relative expression of the different promoters was calculated. The effects of truncation, mutation and db cAMP treatment on each construct were analysed using Student's t-test for unpaired data.

Results Truncation of the 1u promoter construct resulted in virtually the complete loss of expression between constructs C and C1 ($p < 0.001$, Figure 2). This was almost totally recovered following further truncation to C2, with this construct being responsive to db cAMP ($p < 0.01$). Further truncation resulted in a stepwise reduction in expression. Mutations of two of the CREs in the A construct significantly reduced the level of CAST expression ($p < 0.001$), whilst mutation of the CRE most proximal to the transcription start site abolished expression completely. The construct with the mutation in the most distal CRE was still responsive to db cAMP ($p < 0.01$).

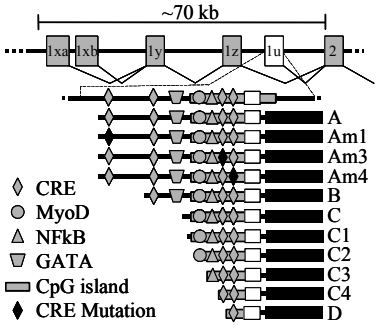

Figure 1 Calpastatin 1u promoter constructs and potential transcription factor motifs

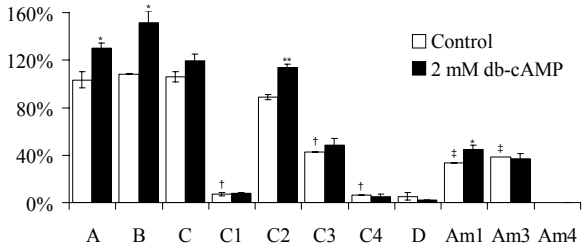

Figure 2 Relative expression of calpastatin 1u promoter constructs and the effects of dibutyryl cAMP. Levels of significance with respect to treatment (* $p < 0.05$, ** $p < 0.01$), truncation († $p < 0.001$) and mutation (‡ $p < 0.001$) are indicated

Conclusions With the notable exception of construct C1, sequential 5' truncation between constructs C and D indicate that the removal of different motifs result in a systematic reduction in calpastatin expression from the 1u promoter, implying that these regulatory motifs are required for full CAST expression. The loss of expression in C1 may be due to a negative enhancer being present in the sequence that is not normally functional. The responsiveness of the mutated Am1 construct to db cAMP suggests that not all the CREs need to be functional for cAMP to enhance cAMP-induced CAST expression, although mutating the CREs most proximal to the transcription start site does appear to either abolish CAST expression or the effect of cAMP. The high sensitivity of CAST mRNA expression to β-adrenergic signalling pathways may suggest a link between physiological stressors or growth promoter usage and toughness in pork. Natural variations or polymorphisms in the 1u promoter region may have profound affects on meat toughness and identification of such SNPs would be useful in marker-assisted selection or molecular prediction of ultimate toughness.

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Relationships between carcass characteristics and eating quality: influence of beef genotype and hanging method

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Introduction Recent data on meat quality indicates that dairy genotypes produce more tender meat than beef genotypes. The relationships between carcass parameters and instrumental measures of meat quality depend on the method of hanging (Lively *et al*., 2005). The present study was undertaken to evaluate the impact of some carcass parameters on meat eating quality of two genotypes when two carcass hanging methods are used.

Materials and methods The experiment involved a total of 40 steers, consisting of two genotypes, Holstein (100% Holstein, Hol) and Charolais (>75% Charolais, CH). All animals were offered grass silage *ad libitum* supplemented with 4.5 kg concentrate for 98 days prior to slaughter. At slaughter, one side of each carcass was randomly allocated to one of two hanging methods, namely Achilles tendon (AT) or tenderstretch (TS). After 48 hours post slaughter the sirloins were removed from the carcasses of 12 animals of each genotype for eating quality assessment. The *longissimus dorsi* was dissected out and cut into 3 equal sections, steaks 2.5 cm thick were prepared for eating quality assessment from each section as described by Lively *et al.,* (2006). Assessments were undertaken by a consumer panel for tenderness, juiciness, flavour and overall liking using a line scale (0-100) involving a total of 600 consumers. Relationships between carcass parameters and the eating quality of 7 day aged *longissimus dorsi* muscle from the anterior and posterior sections were tested for linear regressions for genotype differences within each of the two hanging methods.

Results Carcass weight and conformation were negatively correlated to tenderness when hung AT. Conformation was negatively correlated with juiciness, flavour and overall liking and fitted by a common line (i.e. similar intercept and slope) for the two genotypes. Age at slaughter and fat classification were positively correlated with juiciness, flavour and overall liking and fitted by parallel lines (different intercept, similar slope) for the each genotype. Carcass weight was positively correlated to juiciness and flavour when hung TS and fitted by parallel lines for each genotype. Carcass weight was negatively correlated to flavour and overall liking when hung AT and fitted by parallel lines for each genotype. Marbling score was positively correlated with juiciness and flavour and fitted by a common line for each genotype. The correlation of marbling score with overall liking when hung AT showed two distinct lines (i.e. different intercepts and slopes) for the two genotypes.

Table 1 Linear relationship between production / carcass parameters and meat eating quality

	Tenderness		Juiciness		Flavour		Overall liking	
	AT	TS	AT	TS	AT	TS	AT	TS
Age at slaughter								
Significance[1]	NS	NS	*	**	*	**	**	*
Slope[2]			+ve	+ve	+ve	+ve	+ve	+ve
% variance expalined			19.2	37.7	25.6	28.2	28.6	21.9
Carcass weight								
Significance[1]	*	NS	NS	**	*	*	**	NS
Slope[2]	-ve			+ve	-ve	+ve	-ve	
% variance expalined	11.7			30.9	24.9	24.0	28.0	
Conformation score								
Significance[1]	**	NS	**	**	*	**	***	**
Slope[2]	-ve		-ve	-ve	-ve	-ve	-ve	-ve
% variance expalined	20.5		22.8	21.5	28.3	22.2	34.2	24.5
Fat classification								
Significance[1]	NS	NS	*	**	*	***	**	**
Slope[2]			+ve	+ve	+ve	+ve	+ve	+ve
% variance expalined			19.2	37.3	25.5	44.8	28.3	33.2
Marbling score								
Significance[1]	NS	NS	*	**	*	**	**	**
Slope[2]			+ve	+ve	+ve	+ve		+ve
% variance expalined			20.5	25.2	20.7	21.9	36.7	24.0

[1]NS = not statistically significant; * = P<0.05; ** = P<0.01; *** = P<0.001; [2]-ve = negative; +ve = postive

Conclusion For the two genotypes represented in this study improving the conformation lead to a decrease in eating quality, whereas an increase in fat classification or marbling lead to an improvement in eating quality. Further work is required to establish whether the relationship with conformation represents the differences in conformation between the genotypes.

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The fatty acid composition of muscle fat and relationships to meat quality in Charolais steers: influence of level of red clover in the diet

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Introduction Legumes are important constituents of ruminant diets and are a vital part of low input and organic systems. Previous studies have established the high intake characteristics and animal production potential of red clover silage. Studies with milk produced from cows fed on red clover silage was noted to have higher levels of polyunsaturated fatty acids (PUFA) but had a reduced shelf life which could be ameliorated by feeding additional vitamin E (Al-Mabruk *et al*., 2004). This study considered the effects of incremental inclusion of red clover silage in the diet of beef cattle on the fatty acid composition of the *m*. *longissimus dorsi* and meat quality.

Materials and methods Thirty two Charolais steers (initial live weight 490 kg (s.e.d. 6.7)) were randomly allocated to one of four dietary treatments (each consisting of eight animals) differing in forage type (grass or red clover silage or a mixture) and concentrate type (differing in vitamin E content, C1 and C2 containing 25 and 500 mg/kg vitamin E, respectively). The four diets were (1) grass + C1 (GS); (2) grass and red clover mix (50:50 DM basis) + C1 (GRS); (3) red clover + C1 (RC) and (4) red clover + C2 (RCVitE). Forage was offered *ad libitum* and feed levels were adjusted weekly to maintain a forage:concentrate ratio of 70:30 on a DM basis. Animals were slaughtered after 100 days on treatment and samples of *m*. *longissimus thoracis et lumborum* (LTL) were taken at 48h post-mortem for fatty acid analysis. Colour (L*a*b*) and lipid oxidative shelf life were determined on meat conditioned for 10 days at 1^{o}C and then packed in a modified atmosphere for simulated retail display shelf-life at 4^{o}C. Eating quality was determined on meat conditioned for 10 days by a 10-member trained sensory panel. An ANOVA with diet as the main factor was used to analyse the data.

Results Half carcass weights were similar across treatments and averaged 194 kg (s.e.d. 5.9). Total muscle fatty acids, saturated (SFA) and monounsaturated fatty acids (MUFA) were not different (Table 1). Increasing the amount of red clover relative to grass silage in the diet significantly increased total PUFA content and in particular the 18:2*n*-6 and 18:3*n*-3, resulting in beneficially higher P:S ratio and lower *n*-6:*n*-3 ratio. TBARS increased and colour saturation decreased incrementally with increasing amount of clover in the diet. The concentration of vitamin E in the muscle also decreased with increasing amount of clover. However, feeding additional vitamin E (diet RCVitE) alleviated this problem. Sensory attributes were largely not influenced by diet, although red clover (RC) produced the most tender meat.

Table 1 Fatty acid content (mg/100g muscle) of *longissimus dorsi* and meat quality attributes of LTL.

	Forage type					
	Grass (G)	Grass/red clover (GRS)	Red clover (RC)	Red clover + vitamin E (RCVitE)	sed	P
	Fatty acid composition mg/100 g muscle					
Total fatty acids	3081	3639	4001	3074	0.36	NS
SFA	1300	1535	1643	1270	261.7	NS
MUFA	170.7	206.4	244.4	216.8	13.44	NS
PUFA	170.7^{a}	206.4^{b}	244.4^{c}	216.8^{bc}	13.44	0.001
18:2*n*-6	73.2^{a}	92.8^{b}	113.2^{c}	99.3^{b}	6.68	0.001
18:3*n*-3	22.5^{a}	34.1^{b}	50.7^{c}	37.5^{b}	3.83	0.001
P:S	0.07^{a}	0.09^{ab}	0.10^{bc}	0.12^{c}	0.01	0.01
n-6:*n*-3	3.28^{c}	2.73^{b}	2.30^{a}	2.66^{b}	0.15	0.001
TBARS (mg MDA/kg meat) day 7	0.64 ^{a}	1.31 ^{b}	4.88 ^{c}	1.13 ^{b}	0.45	0.001
Colour saturation day 7	22.9 ^{a}	21.5 ^{b}	20.3 ^{c}	22.6 ^{ab}	0.57	0.001
Vitamin E	3.47 ^{a}	2.92 ^{b}	1.80 ^{c}	3.32 ^{a}	0.18	0.001
Toughness (0-100 line scale)	46.5^{ab}	41.6^{a}	42.0^{a}	48.2^{b}	2.80	0.05
Beef flavour (0-100 line scale)	31.7	34.2	34.2	31.0	2.28	NS
Fishy (0-100 line scale)	0.6	0.2	1.1	0.4	0.5	NS

Conclusions Red clover increased the content of PUFA in meat and reduced colour shelf life. The latter was most likely associated with the low vitamin E content rather than the increased content of PUFA in the muscle, since a supra-nutritional supplement of vitamin E in the diet restored the vitamin E concentration and stability of the meat to that observed with other diets.

Acknowledgements This work was supported by Department for Environment Food and Rural Affairs.

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Discrimination of adipose tissue from different species using Raman spectroscopy

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Introduction The potential of Raman spectroscopy for the analysis of fats and oils has been recognised for some decades. The main advantages of the technique are that no sample preparation is required (allowing *in situ* or on-line studies) and that it can be applied to any physical state including gases, liquids, gels, amorphous solids and crystals. The power of Raman spectroscopy to discriminate between cooking oils has previously been demonstrated. Beattie *et al* (2004a) demonstrated the use of Raman spectroscopy for the characterisation of lipids in model systems and also in clarified butterfat (Beattie *et al* 2004b). The aim of this paper was to determine whether Raman spectroscopy could discriminate between the adipose tissue from different species and which characteristics of the lipids (i.e chain length , unsaturation) contributed to the discriminating functions.

Experimental Subcutaneous adipose tissue was dissected from above the *Longissimus dorsi* (LD) in beef, lamb and pork, and from above the breast muscle in chicken. A total of 255 spectra were recorded (84 Pork, 82 beef, 53 Lamb and 36 Chicken). Raman measurements were carried out using line-focused 785 nm excitation wavelength (typically 100-120 mW at the sample). The spectra were automatically baseline subtracted and normalised about the band at 1745 cm^{-1}, which arises from the carbonyl bond of the fatty acid chain. Partial Least Squares Discriminant Analysis (PLSDA) was used to generate a classification model based on 100 spectra, which was then tested on the remaining spectra. Chromatographic determination of fatty acid composition was carried out on sub-samples of adipose tissue. These samples were extracted using chloroform:methanol and fatty acid methyl esters prepared (BS 684:1980) and analysed by gas-liquid chromatography (Beattie *et al* 2004b).

Results Applying PLSDA to the GC data gave a correct classification rate of 98.6% in both cross validation and validation by the independent test set. In the most important discriminant factor there was clear separation between the Chicken and pork and the ruminant species, but some overlap between lamb and beef on the first discriminant axis. Further separation between the two ruminant species occurred on the second discriminant axis. Application of PLSDA to the Raman spectra again showed clear separation between the ruminant and non ruminant adipose tissue as the major discriminant factor. The bands in the spectra accounting for this separation are those spectral peaks corresponding to unsaturation and saturation. In addition to unsaturation /saturation bands, 7 further factors were required to fully account for the differences between the four species. These factors were primarily related to physical phenomena such as the conformation the fatty acid chain adopted and the cell packing.

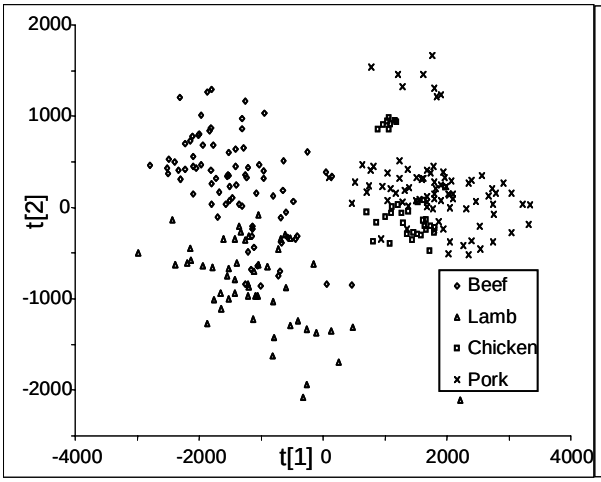

Figure 1. PLSDA score plot of the first two spectral components obtained from PLSDA of the Raman spectra of beef, lamb, chicken and pork adipose tissue.

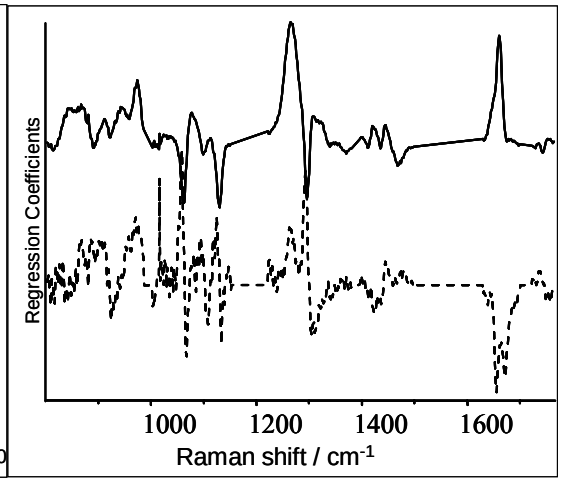

Figure 2. PLSDA loading plots of the first two spectral components used to produce the score plot Figure 1 from the Raman spectra.

Conclusion We have demonstrated the ability of Raman to distinguish between chicken, pork, beef and lamb adipose tissues in a rapid and non-destructive manner. PLSDA was determined to employ a more effective data reduction method than PCA (Principal Component Analysis) maximising the explanation of the separation of the different species.

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AGEWEAN - The effect of weaning age on the performance of sows and their progeny in the first parity

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Introduction The immediate postweaning period in pigs is often characterised by a reduced and variable food intake and poor growth and development, reducing lifetime performance. At present the effects of the postweaning growth check are reduced by the use of antibiotic growth promoters (AGPs), copper sulphate and zinc oxide to enhance the efficiency of feed conversion and hence maximise nutrient capture. However from January 2006 the routine use of in feed AGPs is to be banned and, due to concern over environmental pollution, levels of inclusion of heavy metals are likely to be further reduced. Weaning pigs from the sow at an older age, when their digestive systems are more mature, has been suggested as an approach to reduce the potential negative effect of the AGP ban on the national herd. The objective of the AGEWEAN programme of research is to investigate the effects of weaning age (4, 6 and 8 weeks) in both an indoor and outdoor lactation environment on biological and economic efficiency of a production system where diets contain no AGPs and lower levels of copper (<25ppm added) and zinc (<100ppm added).

Materials and methods The research was carried out at 6 separate experimental sites, chosen to represent a range of diverse geographical locations and production systems within the UK. Two sites were used to provide an outdoor lactation environment (total of 100 sows per weaning age treatment) and four sites to provide the indoor environment (total of 90 sows per treatment). The three weaning age treatments were 4 weeks (weaned at between 21 and 28 days of age), 6 weeks (35-42 days of age) and 8 weeks (49 to 56 days of age). On any given site contemporary blocks of gilts were introduced at the point of farrowing and will be followed through four consecutive parities. All progeny were monitored to weaning, 50% of progeny were monitored to 30kg live weight and 25% were monitored through to slaughter weight to generate data on lifetime health, growth and feed use. The statistical significance of site and treatment effects were analysed using a two way analysis of variance (Genstat), following data transformation where necessary

Results There were no significant effects of weaning age on the loss of back fat (at the P2 position) or bodyweight of the sows over the course of lactation. There were also no significant differences between treatments with respect to the number of piglets born alive, born dead, mean birth weight or total litter birth weight as treatments were not imposed until the point of farrowing. The physical performance of the progeny from the gilt litters during the lactation, weaner and grower-finisher stages can be seen in Table 1. There were no significant interactions between weaning age and lactation environment (indoor/outdoor).

Table 1 The physical performance of pigs from gilt litters weaned at 4, 6 and 8 weeks of age (DLWG – Daily liveweight gain, FCR – Feed conversion ratio).

	4 week		6 week		8 week		sed	Significance	
	In	Out	In	Out	In	Out		Out/In	Wean age
Lactation									
Sow food (kg/sow and litter)	143	160	243	274	364	394	32.22	*	***
Creep food (kg/litter) #	2.61	n/a	7.48	7.73	26.75	30.03	-	NS	***
Piglet wean wt (kg)	7.58	7.48	11.06	11.20	16.20	17.24	1.85	NS	***
Wean – 30kg									
DLWG (kg)	0.44	0.51	0.47	0.60	0.52	0.61	0.05	NS	***
Feed/pig/d (kg)	0.68	0.77	0.80	1.00	0.94	1.07	0.06	***	***
FCR	1.57	1.56	1.76	1.75	1.84	1.79	0.18	NS	***
30-90kg									
DLWG (kg)	0.78	0.75	0.78	0.76	0.80	0.74	0.09	NS	NS
Feed/p/d (kg)	1.98	2.26	1.90	2.30	1.94	2.23	0.16	NS	NS
FCR	2.56	3.06	2.47	3.12	2.47	3.13	0.21	*	NS
Lifetime									
DLWG birth-slaughter (kg)	0.61	0.61	0.59	0.62	0.59	0.60	0.05	NS	NS

#Mean resulting from back transformed statistical output

Conclusion
Whilst there were significant differences with regards to DLWG during the immediate postweaning period, there were no beneficial effects of increasing weaning age on the lifetime DLWG of the progeny. At this stage in the study there appears to be little to suggest that weaning pigs at older ages will help to reduce the potentially detrimental effects of the AGP ban. However the results presented here only represent the gilt parity and this study will continue to monitor the population of sows through four successive parities to assess lifetime performance and longevity of the sows in addition to the performance of progeny.

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Micronised whole rapeseed as a home-grown protein source for pigs weaned at different ages

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Introduction Oilseed rape is a protein crop that can be readily grown under UK conditions and has a good amino acid profile relative to other plant protein sources. Its use in diets for young pigs has been limited by concerns about its content of anti-nutritive factors, especially glucosinolate compounds, although these have been progressively reduced by plant breeders (Gill and Taylor, 1989). The whole seed contains a high level of digestible oil, making it an excellent energy source for piglets provided that the oil is made available by milling or heat treatment to rupture the seed coat. The forthcoming ban on use of in-feed anti-microbial growth promoters has renewed interest in rapeseed use because glucosinolate compounds can have antimicrobial effects (Fenwick and Heany, 1983), and because weaning age may be increased in future to reduce risk of health problems. In this circumstance, where regular intake of solid feed is established during lactation, the sensitivity to dietary rapeseed in the post weaning stage may be less critical. This experiment therefore evaluated the response in performance and health indicators of piglets weaned at different ages to diets with different rapeseed inclusion levels.

Materials and methods A split litter design with 9 pen replicates per treatment was used to investigate the response to 4 levels of inclusion of micronised whole rapeseed in the diet (0, 50, 100 or 200 g/kg; glucosinolate in raw seed = 21 μmol/g) during the first 4 weeks after weaning, when piglets were weaned by half-litter at either 4 or 6 weeks of age. After weaning, piglets were housed in litter matched groups of 4 in flat deck pens, and offered the experimental diets *ad libitum*. Rapeseed substituted for soya in diets formulated to provide the same net energy (4 week=11.3 MJ/kg, 6 week=11.1 MJ/kg) and ileal digestible lysine (4 week=1.42 g/kg, 6 week=1.29 g/kg). Liveweight and feed consumption were recorded weekly, and health and faecal consistency were recorded daily. Faecal samples were taken from 3 pigs/pen at 5 and 26 days after weaning for culture and enumeration of lactobacilli and coliform bacteria, and the activity of faecal extract in an *in vitro* pathogen inhibition test using *Salmonella enteriditis* was measured. Data were analysed by ANOVA according to the split-plot design, with weaning age, rapeseed inclusion and their interaction as treatment factors and replicate as a blocking factor.

Results Piglets weaned at 6 weeks of age had, as expected, higher feed intake and growth rate during the 4 weeks after weaning, but there were no significant interactions between weaning age and dietary rapeseed inclusion. Inclusion of rapeseed in the diet reduced feed intake and consequently liveweight gain, but had no significant effect on feed conversion efficiency. Pig health, faecal consistency and pen cleanliness were not affected by dietary rapeseed inclusion level. Absolute populations of lactobacilli, coliform bacteria and the ratio between these (an indicator of gut health) in faecal samples taken at 5 and 26d after weaning were not consistently affected by dietary rapeseed inclusion level. Piglets weaned at 6 weeks of age had lower faecal $\log_{10}$ coliform counts at day 5 (P=0.02) and day 26 (P=0.002) and a higher L:C ratio at day 26. Faecal extracts did not show significant differential activity in the pathogen inhibition test.

Table 1 Effect of inclusion level of micronised whole rapeseed (g/kg) on performance and health of piglets, weaned at four or six weeks of age, in the 28 days after weaning. Food intake (FI), liveweight gain (LWG), feed conversion ratio (FCR), ratio of $\log_{10}$ *Lactobacilli* to $\log_{10}$ *Coliform* bacteria in faeces (L:C), and specific growth rate of *Salmonella enteriditis* in an *in vitro* pathogen inhibition test of faecal extract (SSGR).

Weaning Age (W)	4 weeks				6 weeks					Sig		
Rape inclusion (R)	0	50	100	200	0	50	100	200	s.e.m	W	R	WxR
Weaning wt (kg)	8.8	8.8	8.8	8.8	14.4	14.4	14.4	14.4	0.09			
FI (g/d)	583	559	506	487	832	807	731	719	16.7	***	***	
LWG (g/d)	475	466	435	423	597	570	523	506	14.6	***	***	
FCR (feed/gain)	1.22	1.19	1.16	1.15	1.39	1.42	1.40	1.42	0.023	***		
L:C day 5	1.21	1.17	1.21	1.20	1.24	1.15	1.17	1.14	0.045			
L:C day 26	1.36	1.43	1.42	1.38	1.68	1.48	1.49	1.41	0.042	***	*	**
SSGR day 5	0.462	0.474	0.463	0.513	0.501	0.497	0.496	0.519	0.022			
SSGR day 26	0.502	0.508	0.420	0.487	0.421	0.426	0.489	0.444	0.025	0.06		*

Conclusion Micronised whole rapeseed has the potential to be a cost effective protein source in diets for weaned piglets, since inclusion levels greater than previously accepted can be used without adversely affecting health, feed conversion efficiency or feed cost per kg gain. However, rapeseed inclusion reduced feed intake, and hence liveweight gain, even after later weaning. Strategies to overcome this detrimental effect need to be developed.

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The effect of weaning age, feed crude protein level and experimental challenge with enterotoxigenic *Escherichia coli* on the health and performance of newly weaned pigs

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Introduction In-feed antimicrobial growth promoters (AGPs) have long been used as means of protection against enteric disorders such as post-weaning colibacillosis (PWC), a diarrhoeal disease of newly weaned pigs resulting from the colonisation and proliferation of enterotoxigenic *E. coli* (ETEC) in the small intestine. One of the strategies to enable the British pig industry to remain competitive in the global market whilst complying with the imminent removal of AGPs may be to increase weaning age and change the level of crude protein in the diet. If weaning age is increased, sensitivity to PWC may be reduced and pigs be better able to cope with higher levels of protein in the diet, which have been associated with an increased risk of PWC (Prohászka and Baron, 1980). The objective of the current experiment was to investigate the effects of experimental challenge, weaning age and dietary protein supply post-weaning on the health and performance of pigs in the absence of AGPs.

Materials and methods The experiment consisted of a 2 x 2 x 2 factorial combination of weaning age (4 vs 6 weeks), dietary protein content (H, 230g CP/kg vs L, 130g CP/kg) and experimental challenge (+ vs -). At weaning (day 0) 32 pigs from each weaning age were balanced for initial weight, sex and litter and offered *ad libitum* access to diet L or H. Both diets were formulated to contain 16.0 MJ DE/kg and were balanced for amino acid composition as a proportion of total protein, sodium and lactose content. On day 14 a standard grower ration was fed to all animals until 10 weeks of age. On day 3 post weaning half of the piglets were challenged *per os* with 10^9 ETEC (*E. coli* O149: K91). Individual feed intakes, weight gain and faecal consistency (assessed through subjective scoring) were recorded and fresh faecal samples were collected regularly to detect the presence and concentration of ETEC. On day 6 post weaning 4 pigs per treatment were slaughtered to assess gut pH and the ratio of lactobacilli to coliforms, both used as indicators of gut health. All animals were individually housed. Data were analysed using the REML procedure of Genstat with body weight at four weeks of age used as a covariate. The animal experiment committee of SAC approved this work.

Results ETEC shedding was significantly (P = 0.003) greater and persisted longer in the 4 week weaned pigs than those weaned at 6 weeks (Figure 1). There was no significant effect of CP level on number of ETEC shed. ETEC challenge had a detrimental effect on performance (Table 1), significantly decreasing average daily gain (ADG) (3-6 days) in both 4 and 6 week weaned animals. Challenged 4 week weaned pigs on the H diet demonstrated a larger decrease in ADG immediately post challenge compared to non-infected animals than those on the L diet, -42 and -25% respectively. This effect was smaller in the 6 week weaned pigs, -26 and -19% for diets H and L respectively. Pigs on the high CP diet had significantly looser faeces in the 3 days post challenge period (H = 1.7 vs L = 1.4; P = 0.023) and poorer gut health as indicated by an increase in the pH of proximal colon contents (H = 6.1 vs L =5.8; P = 0.012). Pigs weaned at 6 weeks significantly outperformed those weaned at 4 weeks (Table 1), although there was no significant difference in body weight at 10 weeks of age, weighing (± SD) 35.5 (± 4.87) and 35.2 (± 3.43) kg respectively.

Table 1 Effect of experimental treatment on pig performance

	ADFI (g/d)			ADG (g/d)		
Day	3-6	0-14	14-28	3-6	0-14	14-28
4L+	239	351	830	213	262	652
6L+	589	680	1392	378	498	1043
4H+	280	373	914	238	324	598
6H+	557	663	1303	382	600	954
4L-	318	433	962	285	305	729
6L-	599	763	1299	461	586	936
4H-	303	338	814	407	311	645
6H-	494	658	1331	517	627	950
S.E.D	29.6	41.0	56.9	53.2	35.8	52.3
	W***	W***	W*** LxCxW*	C* W*	W***	W***

Main effects * P < 0.05, *** P<0.001; where C = Challenge,
L = Protein Level and W = Weaning Age

Conclusion The experimental ETEC challenge had a greater impact on performance and persisted longer in the 4 week weaned pigs than those weaned at 6 weeks of age. In the absence of AGPs decreasing the level of dietary CP may help to prevent and minimise the effects of PWC especially in earlier weaned animals.

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Sodium butyrate improves small intestine structure regardless of zinc oxide supplementation

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Introduction Post weaning piglet performance is often associated with a growth check (LeDividich and Seve, 2000) which is a key concern to the pig industry. The ban of in-feed antimicrobial growth promoters (AGPs) has increased this concern. Sodium butyrate (NaB) has been shown to reduce intestinal inflammation and stimulate enterocyte growth in humans (Ogawa *et al*., 2003) and thus appears to be a possible alternative to AGPs in diets for weaned pigs. Zinc oxide (ZnO) is normally added to weaner diets to control post weaning diarrhoea. The objective of this experiment was to compare varying levels of dietary supplementation of NaB both with and without ZnO to determine whether NaB can improve piglet performance alone or in combination with ZnO.

Materials and methods Three hundred and eighty four crossbred piglets (JSR Healthbred) were weaned at a mean age of 27.8 ± 0.23 days (± SEM) and liveweight of 7.4 ± 0.07 kg. Piglets were weaned into 48 flat-deck pens (1.99m^2) in groups of 8 each balanced for litter origin, weaning weight and sex. Each group was offered a diet (15.7 MJ DE/kg, 207g/kg CP, 15g/kg lysine) containing either 0 or 3.1 g/kg ZnO and supplemented with either 0, 1.5, 3.0 or 6.0g NaB per kg of diet. Feed and water were provided *ad libitum* throughout the 13 day trial. Daily feed intake (FI) and piglet liveweight on days 0 and 13 following weaning were recorded. Average daily gain (ADG) and feed conversion ratio (FCR) were calculated. On days 6 and 13 of the trial 72 piglets (9 per treatment) were killed to measure villus height (vh) and crypt depth at 25, 50 and 75% of small intestine length. Data were analysed using the Polyanova procedures of Genstat 6.1.

Results Dietary ZnO significantly improved all aspects of pig performance and small intestine structure (P<0.001, Table 1). Sodium butyrate supplementation improved overall FCR (P<0.05) and villus height 25% along the small intestine was improved when all inclusion levels were considered together (P<0.05). There was no dose response in villus height to increasing levels of sodium butyrate. There was an interaction between ZnO and NaB for ADG, NaB consistently increased growth rate with increasing concentration of NaB in the presence of ZnO; without ZnO ADG increased at 1.5g/kg NaB inclusion but then declined with higher inclusion levels.

Table 1 Effect of varying dietary inclusion levels of sodium butyrate, with and without zinc oxide, on piglet performance and small intestine structure

	No ZnO				Plus ZnO						
NaB (g/kg):	**0**	**1.5**	**3**	**6**	**0**	**1.5**	**3**	**6**	**sem**	**P**	
FI (kg/d)	0.20	0.22	0.21	0.19	0.24	0.26	0.25	0.26	0.009	ZnO**	
ADG (kg/d)	0.15	0.19	0.16	0.15	0.21	0.22	0.23	0.25	0.012	ZnO**	ZxN#
FCR	1.39	1.20	1.29	1.27	1.13	1.19	1.07	1.06	0.034	ZnO**	NaB*
D13wt (kg)	9.2	9.6	9.3	9.3	10.0	10.3	10.4	10.4	0.18	ZnO**	
Villus ht 25% si (mm)	0.30	0.35	0.33	0.33	0.33	0.37	0.37	0.36	0.019	ZnO*	NaB#
50% si (mm)	0.33	0.38	0.34	0.34	0.40	0.38	0.41	0.39	0.015	ZnO**	

Key: # P<0.05 when all NaB treatments considered together, * P<0.05, **P<0.001, ZxN = interaction between ZnO and NaB

Conclusions Pigs grew poorly in this trial thus giving the supplements every opportunity to show their merit. Pigs responded well to ZnO as anticipated. Interestingly pigs continued to respond to NaB to high levels with ZnO but only showed a good response at the lowest level without ZnO. NaB did promote improved FCR at all levels of supplementation regardless of whether ZnO was present in the diet. It is likely that this improvement resulted from the enhanced villus structure in the proximal part of the small intestine. This experiment used the unprotected sodium salt form of NaB which rapidly dissociates in the small intestine. It would be interesting to investigate whether a protected form would dissociate more slowly and hence maintain its effects further down the tract.

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The effect of wheat endosperm texture on nutritional value for weaned piglets

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Introduction Wheat is a major component of piglet diets, representing a significant source of energy. Nutritional value of wheat can be influenced by genetic and environmental factors (Wiseman and Inborr, 1990). Previous work examining the effect of wheat cultivar on pig performance has failed to show a clear relationship with digestibility (Lewis *et al*., 1999), although selection of which wheats to evaluate has invariably been unstructured. The aim of the current study was to investigate the effect of wheat endosperm texture, in wheats which were closely related, on nutritional value for the newly weaned piglet.

Materials and Methods Twenty piglets weaned at 28 days of age were individually housed and fed *ad-libitum* one of two diets differing only in wheat of different endosperm texture (1 hard, 2 soft; using the SKCS single kernel characterisation system, data were 47.4 and 26.3 respectively). Wheats shared a common genetic background and did not contain the 1B1R characteristic. Diets contained wheat (ground through 1.5mm screen) at 580g/kg, were iso-nitrogenous and iso-energetic, and contained no antibiotic growth promoters. Experimental period was for 14 days, with piglets slaughtered at day 0 (n=2), and on days 2, 4, 6, 10 & 14 post weaning (n=4; 2 per diet). At slaughter, digesta samples (at 0.25, 0.5 & 0.75 along the small intestine) were analysed for pH and viscosity, and starch and nitrogen. The statistical model was a 2 (Diet) * 3 (Region) * 6 (Day) factorial.

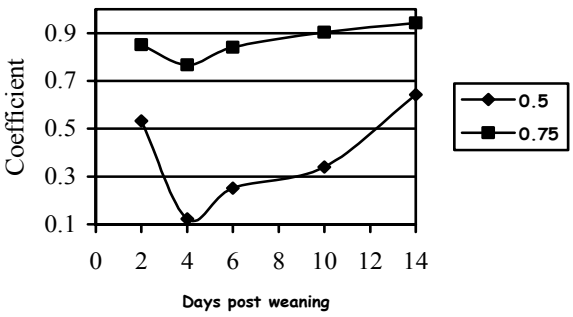
Results Piglets on the soft wheat diet exhibited greater voluntary feed intake (VFI) post 5 days (P=0.063) but not significantly greater daily live weight gains (DLWG) than those on the hard wheat (P=0.645). Although coefficients of nitrogen digestion increased significantly along the small intestine from the proximal to distal region for both groups (P = <0.001), animals fed soft wheat exhibited significantly greater coefficients within the 0.75 region than those fed hard wheat (Table 1). No effect of dietary treatment on digesta pH or starch digestibility was observed, although there was a reduction in starch digestibility in the immediate post-weaning period which was particularly noticeable in the 0.5 region (Figure 1). Piglets fed the diet based on soft wheat were found to have significantly less viscous digesta than those on the hard wheat diet (P=0.029).

Table 1 Performance and coefficient of nitrogen (N) digestibility.

	Hard	Soft	SED	*P*
VFI † (g/d)	589	647	30.8	*0.063*
DLWG (kg)	0.30	0.33	0.05	*0.645*
N*	0.65	0.74	0.01	*0.006*

† post 5 days * distal portion (0.75) of intestine

Figure 1 Effect of region (R) and day (D) on coefficient of starch digestibility



P<0.001 R; 0.012 Q D; 0.028 Q R*D
0.5 & 0.75 indicate region of small intestine.
There was virtually no starch digestibility at 0.25

Conclusions Under the conditions of the trial, wheat of soft endosperm appears to be of superior nutritional value. The less viscous digesta from piglets on the soft wheat diet and improved hydration of soft wheat components (confirmed with *in vitro* analyses) may also enhance digestibility. The reduction in starch digestibility immediately post-weaning, irrespective of diet and particularly pronounced in the distal duodenum, is a problem and may explain the post-weaning growth check and deterioration in health commonly associated with animals at this stage.

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The effect of varying solubility and inclusion level of dietary non starch polysaccharides on the performance and risk of post weaning enteric disorders in newly weaned pigs

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Introduction The removal of in-feed antimicrobial growth promoters (AGPs) is expected to place pigs at greater risk of post weaning enteric disorders (PWED), and accentuate the need for alternative, non-pharmaceutical, strategies for disease prevention. Evidence in the literature suggest that appropriate non-starch polysaccharides (NSPs) may be added to weaner diets in order to improve gut health and reduce the incidence of PWED (Bolduan *et al.,* 1988). NSP solubility may be an important factor in such potential health benefits with soluble NSPs that lead to high digesta viscosity actually increasing the occurrence and severity of PWED (Hopwood *et al.,* 2004). The objective of this experiment was to investigate effects of NSP solubility and inclusion rate on gut health and development, growth performance and the risk of PWED in the absence of effects on digesta viscosity.

Materials and methods A 2 x 3 factorial design experiment compared NSP type (soluble (sNSP) vs insoluble NSP (iNSP)) and inclusion level (Low (L) vs Medium (M) vs High (H)) with the addition of a negative (C) and positive control (C+). Sixty-four pigs were weaned at 27.6 ± 2.66 days (± S.D) of age and 9.0 ± 1.5 kg body weight were balanced for initial weight, sex and litter and offered *ad libitum* access to one of the eight dietary treatments. The increase in sNSP and iNSP was achieved by increasing inulin (a non-viscous sNSP source) and methylcellulose respectively from 50 g/kg in the L diets to 100 and 150 g/kg in the M and H diets respectively. The C diet contained both methylcellulose and inulin at 50 g/kg. The C+ diet was identical to the C diet with the addition of AGPs. All diets contained similar amounts of cereal (i.e. background NSP) and were balanced for CP (230g CP/kg), DE (16MJ DE/kg) and amino acid composition as a proportion of total protein. Individual feed intakes and faecal consistency was recorded daily. Animals were individually housed and slaughtered on day 14 for assessment of gut health and development. Data were analysed using the REML procedure of Genstat. The animal experiment committee of SAC approved this work.

Results NSP solubility had little effect on performance, although sNSP inclusion had a beneficial effect on gut health (lower pH and higher L:C ratio, see Table 1) and a lower faecal score. Increasing NSP inclusion significantly decreased the pH of caecal and proximal colon contents (Table 1), but did not affect faecal score. Both sNSP and increasing NSP levels resulted in relatively heavier caeca and colons and lighter small intestines (Table 1). Increasing levels of NSP only had a small penalty on pig performance, with empty body weight percentages of 87.9 and 86.7 on the L and H diets respectively (P = 0.067). The inclusion of AGPs had beneficial effects on performance, gut health and faecal score.

Table 1 Effect of NSP supply on empty organ weight (% of empty gastrointestinal tract (GIT)), pH values of digesta samples and the lacobacillus to coliform ratio (L:C). All samples were taken at slaughter on day 14.

	Empty organ weight (% GIT)				L:C		pH		
	Stomach	Small intestine	Caecum	Colon	Ileum	Colon	Ileum	Colon	Caecum
Hs	13.33	49.88	8.05	28.74	1.30	1.46	6.94	5.61	5.11
Hi	13.10	55.10	6.51	25.29	1.27	1.23	7.09	5.99	5.53
Ms	12.56	54.39	7.62	25.43	1.38	1.43	6.95	6.09	5.44
Mi	16.41	58.31	3.70	21.58	1.19	1.22	6.73	6.21	5.84
Ls	13.96	57.85	4.85	23.33	1.26	1.36	6.99	5.95	5.49
Li	12.98	58.09	5.68	23.25	1.28	1.21	7.06	6.18	5.76
C	11.45	55.31	6.08	27.16	1.27	1.36	6.86	5.99	5.46
C+	14.54	58.04	6.96	20.49	1.34	1.36	7.09	6.48	5.93
S.E.D	1.695	1.128	0.600	1.411	0.059	0.058	0.078	0.113	0.096
Response		S**, I***	S**, I** SxI***	S*, I* A**		S***	I*	S*, I*, A*	S***, I**, A***

Main effects * P < 0.05, *** P<0.001; where I = Inclusion level, S = Solubility and A = AGP

Conclusion These results suggest that in order to minimise the risk of PWED whilst maximising performance that diets containing sources of predominantly sNSP which do not affect viscosity should be recommended, whilst inclusion level should be kept to a minimum. However, as the pigs in this trial appeared to be at low risk to PWED, effects of NSP feeding under a higher infectious pressure remain to be assessed.

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Genetic parameters of egg and weight traits in Japanese quail

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Introduction Analysing egg and weight traits together may provide a new procedure to express the maternal effects in poultry. Maternal effects in poultry are atypical from those of mammals as any maternal effects on chicks, incubated artificially, must be the residual effect of dam reflected in egg characteristics at laying. Possible relationships between W (weekly body weight) and reproduction, including the existence of maternal effects, might be used to improve both meat and egg production. The aims of this study were to analyse the genetic parameters of liveweights and egg traits, and their relationship with maternal effects for a one-to-one sire and dam pedigree recorded and artificially incubated Japanese quail population.

Materials and methods A total of 1808 Japanese quail were used from the population as described by Saatci *et al.* (2003). It consisted of individually recorded birds, weighed weekly from hatching to 6 weeks of age (From W0 to W6). Each trait was analysed with animal as a random factor to fit the additive direct effect ($\sigma 2A$), the animal being the individual for which the trait was recorded, namely hen (dam) for egg traits and chick (progeny) for body weight (W). For egg traits, the permanent environmental variance associated with the animal ($\sigma 2PEA$) was included. Bivariate analyses were completed for all combinations of the traits using the ASREML programme. All the bivariate analyses included the covariance between the additive direct effects. The bivariate analyses between egg traits and W also included the covariance between the additive direct effect for the egg trait and the maternal genetic effect for BW (Gilmour *et al*. 1998).

Results Parameters obtained from the analyses are presented in Tables 1 and 2.

Table 1 Estimates of genetic parameters from bivariate analyses with correlations between direct genetic effects below the diagonal and correlations between the direct genetic effect for egg traits and the maternal genetic effect for weight traits above the diagonal. Heritabilities from univariate analyses are on the diagonal.

	Egg Height	Egg width	Egg weight	W0	W1	W2	W3	W4	W5	W6
Egg Height	*0.18*			0.51	0.86	0.80	0.81	0.78	0.68	0.78
Egg width	0.22	*0.22*		0.51	>1.0	0.92	>1.0	0.87	0.91	0.75
Egg weight	0.61	0.89	*0.25*	0.97	>1.0	0.90	>1.0	0.91	0.91	0.80
W0	0.27	0.27	0.17	0.07						
W1	0.01	-0.02	0.05	0.88	*0.18*					
W2	0.18	0.22	0.33	0.88	0.95	*0.19*				
W3	0.27	0.09	0.25	0.89	>1.0	0.96	*0.18*			
W4	0.10	0.24	0.21	0.90	0.94	0.90	0.91	*0.19*		
W5	0.29	0.19	0.14	0.85	0.90	0.78	0.79	0.88	*0.13*	
W6	0.23	0.27	0.22	0.83	0.77	0.58	0.65	0.73	0.94	*0.15*

Table 2 Estimates of genetic parameters from bivariate analyses with correlations between maternal genetic effects above the diagonal and correlations between permanent environmental effects below the diagonal. Maternal heritabilities for body weights (W0 to W6) from univariate analyses are on the diagonal.

	Egg Height	Egg width	Egg weight	W0	W1	W2	W3	W4	W5	W6
Egg width	0.70									
Egg weight	0.85	0.96								
W0	0.98	0.98	0.99	*0.74*	>1.0	0.96	1.00	0.86	0.91	>1.00
W1					*0.17*	0.96	0.91	0.85	0.88	>1.00
W2						*0.11*	0.94	0.92	0.99	>1.00
W3							*0.10*	1.00	0.99	>1.00
W4								*0.10*	>1.0	>1.00
W5									*0.12*	0.97
W6										*0.07*

Conclusions Egg traits and chick weights are influenced by a combination of genetic and permanent environmental effects. The strong maternal influences on BW are counter intuitive but probably reflect a residual effect of the dam through characteristics of the egg. Whatever, the biological reasons for these effects there are implications for the construction of selection indexes in Japanese quail, and possibly poultry in general. Such indexes, particularly if they address egg and bird traits as selection objectives, should include a consideration of maternal effects and the correlations between direct genetic and maternal genetic effects. The results also illustrate the strength of permanent environmental effects, thus reinforcing the importance of high standards of care and feeding.

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The application of random regression models in turkey egg production

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Introduction The common methodology for the genetic evaluation of the egg production in poultry is to use cumulative data. However, the source and the scale of variation in egg laying are not constant during the whole period and thus, longitudinal models might offer more accurate predictions. Random Regression Models (RRMs) allow for differences in the phenotypic trajectory within a population and for these to be decomposed for each individual into genetic and environmental components. A similar approach is widely used in dairy cattle, where RRMs have underpinned the development of test day models for genetic evaluation. The objective of this study was to investigate the application of RRMs in the egg production of turkeys.

Materials and methods Data included records for egg laying in a daily basis for a period of 140 days from a fully pedigreed population over five generations compromising a total of 2400 hens. The production period was divided into five 28-day periods and the sums of each period corresponded approximately to the monthly egg production. The models included a fixed regression function, derived from Ali and Schaeffer (1987), to account for the phenotypic trajectory of the average observations across the hens. One combined fixed effect was fitted that included the factors year, hatch within year, defining birth groups, and egg-laying pens. Two random effects were fitted, a permanent environmental and an additive genetic, were used. For each of the random effects a random regression function using Legendre polynomials was fitted. Several values for the order of the orthogonal polynomials were tested and the optimum was selected based on a log-likelihood test.

Results The fixed regression function fitted very effectively the average monthly egg production (Figure 1). The optimum order for the Legendre polynomials was found to be the cubic based on the log-likelihood tests and having the lowest residual term. The genetic parameter obtained can be visualised in the heritability profile presented in Figure 2. Genetic correlations between periods were also estimated.

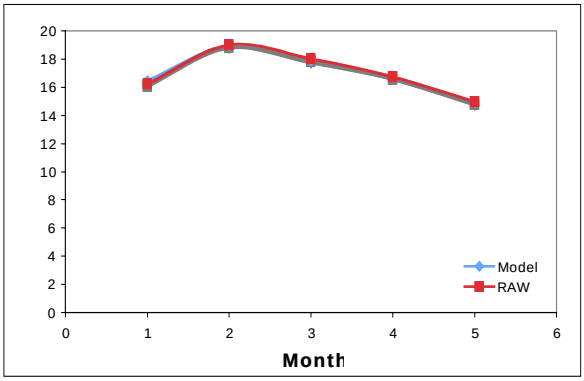

Figure 1 Modelling the monthly egg production

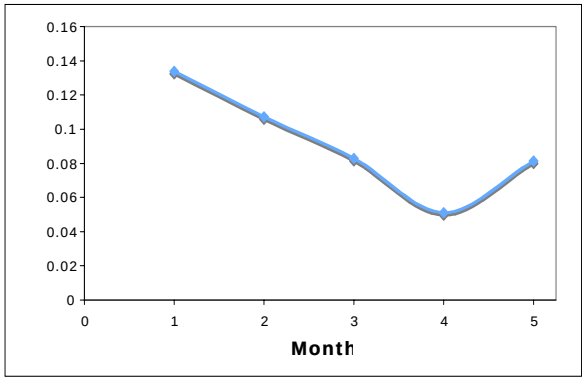

Figure 2 Heritability profile

Conclusions The use of RRMs has several advantages. A heritability profile can be plotted, offering information for the changes of the genetic variance over time for the whole production period. Thus, data collection and selection procedures can be optimised and it can be monitored which periods have records that are the richest in genetic information. RRMs are also efficient in the prediction of missing values, having a low mean square error when using a reduced dataset. In brief, the genetic evaluation for egg production is robust and offers an alternate method for the breeding values prediction.

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QTL mapping within commercial poultry populations using variance component methodology

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Aims and objectives Mapping quantitative trait loci (QTL) within commercial populations circumvents the need to verify segregation of QTL detected under experimental crosses. Variance component (VC) mapping combines information from all relationships within a pedigree to provide a breeding value for the animal and QTL with immediate potential for marker assisted selection. Variance components are estimated by modelling the covariance structure between relatives sharing alleles identical by descent (IBD). Commercial poultry data and simulation will be used as a model to explore how VC methodology behaves under varying population structures. Data is available from the Cobb Vantress broiler population to evaluate extensions to the additive QTL model. Computer simulation will be used to test models including additive, dominance, epistatic and imprinting effects over a range of pedigree structures including family sizes and number of generations.

Materials and methods Initial work focused upon using existing methods of within line QTL detection. Half-sib (HS) and variance component analyses (VCA) were implemented and compared within a two-generation pedigree. The aim was to investigate QTL in previously identified regions of chicken chromosomes 1, 4 and 5 relating to 40-day weight and conformation score. Data consisted of 100 full-sib broiler families (46 paternal half-sib families) with trait data for body-weight and conformation for 2,708 offspring, and genotypes for markers on chicken chromosomes 1, 4, and 5. HS QTL mapping was performed using the regression method in QTL Express for both paternal and maternal families. Confidence intervals and significance thresholds were estimated using bootstrapping and permutation analysis. VC mapping was done testing a novel module in QTL Express using MCMC to estimate identical by descent coefficients and ASREML to estimate QTL effects.

Results Chromosome 4 showed nominal significance for QTL affecting weight and conformation, and linkage was confirmed for both traits on chromosome 5. Table 1 compares power to detect QTL using half sib and variance component methods. Half sib analysis using dam as common parent appeared to be most powerful and generally explained the most variance.

Table 1 Proportion of within family variance explained by fitting a QTL for half sib families using different analyses.

Trait	Chr	V_w QTL (%)		
		sire	dam	VCA
Weight	1	0.003	0.003	0.04
	4	0.03*	0.03	0.03†
	5	0.002	0.06**	0.03
Conformation	1	0.02	0.03	0.02
Score	4	0.03*	0.02	0.02
	5	0.02*	0.09**	0.04††

sire and dam denote common parent, VCA refers to QTL heritability from variance component analysis
* pointwise significance $P < 0.05$, ** chromosome wide significance, *** genome wide significance
† nominal 5% significance assuming $\chi^2_{0.5}$, †† nominal 1% significance assuming $\chi^2_{0.5}$

Conclusions Results varied according to method of analysis and common parent in the half-sib method. Analysis of dam families gave the strongest evidence for segregation of QTL. VCA tended to detect effects segregating in either parent. In many cases half sib analysis was more powerful than the VCA, however when analysing only one parent a better picture can be garnered with the VC approach. Thus far, the VCA methods have only been used as a comparison in sib ships. There is a need to look at performance in more complex data sets. The VC approach has the advantage of direct application to a pedigree regardless of structure. The method provides estimates of the additive genetic variance, the variance due to the QTL at the test location and the likelihood value of the solution. It can also be used to provide breeding values for the QTL for all individuals in a population. Furthermore, the linear mixed model can be extended to include interactions within and between loci. Future work will involve the use of the method proposed by Pong-Wong *et al*., (2001) to estimate IBD matrices modelling underlying genotypic effects due to dominance, epistasis and sex specific or imprinting effects. Simulations to explore the effect of depth and size of pedigree on power to detect these effects should provide an insight to the feasibility of applying the new methodology to map and dissect complex traits within existing commercial populations.

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Effect of species and breed on the enrichment with long chain n-3 polyunsaturated fatty acids in edible poultry tissues from the conversion of dietary α-linolenic acid by broilers and turkeys

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Introduction The concentrations of the long chain n-3 polyunsaturated fatty acids (LC n-3 PUFA) eicosapentaenoic acid (C20:5, EPA) and docosahexaenoic acid (C22:6, DHA) in poultry meat can be increased (to some extent) by enriching poultry diets with these two acids. However, this relies almost entirely on the use of fish oil as there are no terrestrial sources of these acids. This raises the issue of both the sustainability of such an approach and also the consumer acceptability of such a reliance on fish oil in poultry diets. The conversion of α-linolenic acid (LNA) to EPA and DHA, though limited, is done by birds and so an alternative approach would be to enrich poultry diets with the more abundantly available LNA as a means of increasing the EPA and DHA content of poultry meat. The objective of this study was to investigate whether there was any effect of poultry species and breed on the enrichment of poultry meat with EPA and DHA in response to increased dietary concentrations of LNA.

Materials and methods Day old chicks (Ross 308, 36 and Cobb 500, 36) and turkey poults (Wrolstad, 24 and BUT T8, 24) were brooded as separate groups according to species and breed for 21 d (chicks) or eight weeks (poults). During this time they were fed proprietary compound feeds appropriate for their age. At the end of the brooding period, the birds were randomly assigned to one of three dietary treatments and housed in pens (four turkeys per pen) or cages (six broilers per cage). They were then fed one of three wheat-based diets, consisting of 50 g/kg (fresh weight) added oil. The oil was either vegetable oil, or partially replaced by linseed oil (400 or 800 g/kg oil). There were two replicates for every breed x diet interaction. Broilers were slaughtered at 42 d and turkeys at 16 weeks. Samples of white and dark meat, together with samples of the diets fed, were analysed for LNA, EPA and DHA. Within species and for each tissue (white or dark meat), a linear model was used to determine whether there was a straight-line effect of the proportion of LNA in the diet (% w/w total fatty acids) and the proportion (% w/w total fatty acids) of EPA or DHA in the meat using SAS. Species could not be compared statistically, but the estimates of the slopes could be qualitatively compared to estimate whether there was any evidence of a difference between them.

Results These are summarised in Table 1. Increasing the concentration of LNA in the diet of broilers or turkeys had no effect on the EPA content of their meat. However, there was a tendency ($P<0.1$) for the DHA content of turkey meat to decrease in response to increased dietary LNA content. The magnitude of this response, however, was very small. There was no evidence of any difference between breeds of broiler or turkey in their response to enriching the diet with LNA.

Table 1 Effect of poultry breed on the estimate of the slope describing the linear relationship between diet LNA content (% w/w total fatty acids) and meat EPA or DHA content (% w/w total fatty acids).

Acid	Turkey breed		SEM	P[1]		Broiler breed		SEM	P[1]	
	Wrolstad	BUT T8		L	LxB	Ross	Cobb		L	LxB
White meat										
EPA	-0.008	0.003	0.017	0.829	0.667	0.022	0.004	0.031	0.575	0.689
DHA	-0.110	-0.115	0.078	0.075	0.968	-0.031	-0.185	0.106	0.187	0.334
Dark meat										
EPA	0.026	0.012	0.016	0.119	0.532	0.016	-0.019	0.030	0.947	0.413
DHA	-0.029	-0.053	0.029	0.082	0.578	-0.007	-0.129	0.071	0.250	0.231

[1]L: significance of the linear relationship between diet LNA and meat EPA or DHA content; LxB: significance of the interaction between L and the breed of bird.

Conclusion Although both broilers and turkeys may convert some dietary LNA to either EPA or DHA, it would seem from these results that these acids are not then deposited in the edible tissues to any nutritionally meaningful extent. Indeed, the supplementation of the diet with LNA appears to slightly inhibit the deposition of DHA in both white and dark meat. Seeking to enrich the LC n-3 PUFA content of poultry meat by supplementing poultry diets with LNA therefore seems unlikely to be successful. There is also no evidence that this policy would be any more successful in turkeys rather than broilers, or if a particular commercial breed was selected. Attempts to increase human consumption of LC n-3 PUFA by enriching poultry meat with these acids should therefore focus on identifying and developing alternative sources of the LC n-3 PUFA.

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Effect of different levels of chromium nicotinate on growth performance and antibody titre responses to Newcastle and Avian Influenza disease in heat-stressed broiler chicks

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Introduction High ambient temperature reduces feed intake, live weight gain, and feed efficiency (Siegel, 1995), thus negatively influencing the performance of broilers. Heat stress has also been shown to suppress immune responses of broiler. Chromium is an essential nutrient required to promote the action of insulin in body tissues so that the body can use sugars, proteins and fats. Cr deficiency can disrupt carbohydrate and protein metabolism, reduce insulin sensitivity in peripheral tissues and impair growth rate (Vincent, 2000).Chromium is important in altering the immune response by immunostimulatory or immunosuppressive processes as shown by its effects on T and B lymphocytes, macrophages and cytokine production. Dietary chromium supplementation has been reported to have a positive effect on growth rate and feed efficiency of growing poultry under stress conditions (Sahin *et al*., 2002). Another reason for supplementing chromium is due to the fact that chromium is excreted excessively during stress (Anderson, 1994). Also the dietary requirement of livestock for chromium has not been defined yet (NRC, 1997). This study was to investigate the effects of different levels of Cr nicotinate on the growth performance and antibody titres against Newcastle and Influenza disease in broilers.

Materials and methods Two hundreds and Forty commercial broilers (Ross 308) allocated to four treatments in a completely randomized design. Treatments supplemented with 0 (control), 500, 1000 or 1500 ppb Cr in the form of chromium polynicotinate. Each treatment consisted of 4 pens with 15 birds in each pen. Birds were raised from 5 day old to 42 days of age, in controlled house with mean value of daily temperature 33 ± 3 °C and were fed *ad libitum* with a corn-soybean meal basal diet for starting (5-21d) and finishing (21-42d) periods. Feed intake and body weight were measured at 21 and 42 days of age. All birds were intramuscularly immunized with killed vaccine of Newcastle disease and Avian Influenza (H9 N2) at age of 8 days. At the same age (8d) live Newcastle disease vaccine (Nobilis ND Hitchner) was administered intraocularly. Live Newcastle disease vaccine (Nobilis ND Lasota) was administered orally (drinking water) on 22d. On days 18 and 30 blood samples were collected from the wing vein of twelve birds per treatment and serum antibody titres against Newcastle and Influenza diseases were determined by haemagglutination inhibition (HI) test and were expressed as the logarithm base 2.

Results The effects of chromium supplementation on performance are shown in table 1. Body weight of broilers fed supplemental chromium for 21 days increased significantly ($P<0.05$) but at 42d broilers fed 1000 and 1500 ppb Cr had more weight than control and 500 ppb Cr ($P<0.01$). Body weight gain of chicks fed 1000 and 1500ppb Cr significantly increased ($P<0.01$) in all of periods. Feed intake of broiler during 21 to 42d was not affected ($P>0.05$) by chromium supplementation but in 5 to 42d feed intake of broilers fed 1500 ppb Cr increased ($P<0.05$).No significant difference in feed conversion was observed ($P>0.05$).Table 1 lists the effects of supplemental chromium on antibody titres of broilers. Antibody against Newcastle disease (ND) at 18d was not affected by treatments. Dietary Cr supplement not significantly tended to increase antibody titre against ND at 30d ($P= 0.09$).Broilers fed chromium had higher antibody titres against Influenza disease at 30 d ($P< 0.01$).

Table1: The effects of supplemental chromium on performance and antibody titers against Newcastle and Avian Influenza in chicks

Treatments		Body weight (gram)		Weight gain (gram/day)			Feed intake (gram/day)			Newcastle (log_2 titer)		Influenza (log_2 titer)	
		21d	42d	5-21d	21-42d	5-42d	5-21d	21-42d	5-42d	18d	30d	18d	30d
Control		671b	2170b	36.9b	71.4b	56.5b	49.4b	133	94.7b	2.12	4	3.87	3.46b
Cr added (ppb)	500	692a	2176b	37.3b	70.7b	56.7b	51.4ab	132	97.1ab	2.25	4.67	4.17	4.54a
	1000	701a	2248a	38.8a	73.5a	58a	51.9a	132	97ab	2	4.58	3.96	4.25a
	1500	698a	2254a	38.6a	74.2a	58.7a	51.2ab	137	98.4a	2	4.62	4.08	4.29a
SE		6.692	7.633	0.415	0.519	0.382	0.633	1.423	0.923	0.173	0.151	0.121	0.119

Means within the same column without common superscripts differ significantly ($P<0.05$) SE: Standard Error

Conclusion The results of this experiment indicated that chromium supplementation improved live weight gain, feed intake and elevated antibody titre against Influenza disease in heat stressed broilers. Chromium may offer a potential protective management practice in preventing heat stress-related depression in performance of broilers.

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Effect of species and breed on the response by broilers and turkeys to increased dietary concentrations of n-3 polyunsaturated fatty acids in terms of the n-3 polyunsaturated fatty acid content of edible poultry tissues

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Introduction The potential to increase the n-3 polyunsaturated fatty acid (PUFA) content of poultry meat by enriching the poultry diet with n-3 PUFA is well established. However, the responsiveness to different fatty acids is variable (Rymer and Givens, 2005) and there is little information on the effect of species and breed of poultry. The objective of this study was to investigate the effect of poultry species and breed on the response to increased dietary concentrations of the n-3 PUFA α-linolenic acid (C18:3, LNA), eicosapentaenoic acid (C20:5, EPA) and docosahexaenoic acid (C22:6, DHA) in terms of the increased concentrations of these fatty acids in white and dark poultry meat.

Materials and methods Day old chicks (Ross 308, 72 and Cobb 500, 72) and turkey poults (Wrolstad, 48 and BUT T8, 48) were brooded as separate groups according to species and breed for 21 d (chicks) or eight weeks (poults). During this time they were fed proprietary compound feeds appropriate for their age. At the end of the brooding period, the birds were randomly assigned to one of six dietary treatments and housed in pens (four turkeys per pen) or cages (six broilers per cage). They were then fed one of six wheat-based diets, consisting of 50 g/kg (fresh weight) added oil. The oil was either vegetable oil, or partially replaced by linseed oil (400 or 800 g/kg oil), by fish oil (400 or 800 g/kg oil), or a mixture of the two (400 g/kg oil as linseed oil and 400 g/kg oil as fish oil). There were two replicates for every breed x diet interaction. Broilers were slaughtered at 42 d and turkeys at 16 weeks. Samples of white and dark meat, together with samples of the diets fed, were analysed for LNA, EPA and DHA. Within species and for each tissue (white or dark meat), a linear model was used to determine whether there was an effect of the proportion of an individual fatty acid in the diet (% w/w total fatty acids) on the proportion (% w/w total fatty acids) of that fatty acid in the meat. The interaction between this effect and breed was also determined using SAS. Species could not be compared statistically, but the estimates and standard errors of the slopes were qualitatively compared to estimate whether there was any evidence of a difference between them.

Results These are summarised in Table 1. In all cases there was a significant (P<0.001) linear relationship between the concentration of n-3 PUFA in the diet and in the meat. Wrolstad turkeys showed a greater response to LNA in their dark meat compared with BUT T8. Ross broilers were more responsive than Cobbs to LNA in the dark meat. There was some evidence (based on the estimates of the slopes and their standard errors) that turkey white meat was more responsive than broiler white meat to increased dietary concentrations of EPA.

Table 1 Effect of poultry breed on the estimate of the slope describing the linear relationship between diet and meat n-3 PUFA content

Acid	Turkey breed		SE	P[1]		Broiler breed		SE	P[1]	
	Wrolstad	BUT T8		L	LxB	Ross	Cobb		L	LxB
White meat										
LNA	0.481	0.447	0.125	<0.001	0.847	0.474	0.368	0.052	<0.001	0.164
EPA	0.498	0.510	0.082	<0.001	0.919	0.257	0.320	0.059	<0.001	0.461
DHA	0.728	0.884	0.203	<0.001	0.591	0.682	0.690	0.128	<0.001	0.967
Dark meat										
LNA	0.570	0.465	0.035	<0.001	0.046	0.557	0.449	0.022	<0.001	0.002
EPA	0.332	0.444	0.064	<0.001	0.229	0.317	0.307	0.041	<0.001	0.869
DHA	0.465	0.573	0.114	<0.001	0.514	0.396	0.406	0.086	<0.001	0.937

[1]L: significance of the linear relationship between diet and meat n-3 PUFA content; LxB: significance of the interaction between L and the breed of bird.

Conclusion There does appear to be some difference between species and breeds of poultry in their responsiveness to dietary n-3 PUFA. When seeking to enrich poultry meat with LNA, it would appear that Ross 308 broilers are more responsive than Cobb 500 and Wrolstad turkeys more responsive than BUT T8. Turkeys may also be more responsive than broilers to EPA. This variation could be valuable in attempts to achieve meaningful increases in the consumption by humans of long chain n-3 PUFA.

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Sire referencing strategies in hill breeds to improve weaning weight in lambs

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Introduction In sire referencing schemes (SRS) genetic links are formed among flocks by the mutual use of some rams (reference sires). These connections allow for across-flock genetic evaluations offering a larger pool of candidates for selection. Where SRS have been established in hill breeds, their guidelines have been adopted from terminal sire breeds. Hill flocks, however, are typically much larger in size than other pure breeds with more candidates to select among within individual flocks. Guidelines from other breeds may therefore be inappropriate. Using stochastic simulation, the objective of this study was to explore the effect of number and method [artificial insemination (AI) *vs.* natural service (NS)] of use of reference sires (RS) on genetic gain, inbreeding and connectedness for a lamb trait (weaning weight) in SRS tailored to characteristics of hill breeds.

Materials and methods Weaning weights in 17 flocks that varied in size from 100 to 700 breeding ewes were simulated. After 4 yr of random mating, the flocks joined a SRS. Selection ensued for 15 years using across-flock Best Linear Unbiased Prediction (BLUP) of breeding values for weaning weight (heritability of 0.29). Flocks connected by using 1, 2 or 3 rams by AI drawn from a team of 2, 4 or 6 RS, respectively. A natural service (NS) scenario in which each flock drew 2 rams from a larger team (34 RS) was also considered. Rams in the top 15% on BLUP breeding value were chosen as the RS, with the entire team replaced each year. Each RS was mated to 16 ewes. Surplus ewes were mated to homebred or unrelated purchased rams. In another scenario, no RS were used (within-flock selection). True breeding values and inbreeding coefficients were obtained for all animals born in each year. Connectedness was measured as the prediction error correlation of the average BLUP breeding values between pairs of flocks, based on Kennedy and Trus (1993). Each scenario was replicated 25 times.

Results In Table 1 genetic gain and inbreeding at year 15 of selection is shown for the five schemes. After 15 years, participation in SRS resulted in gains of 1.04 (NS, 2 RS) to 1.09 (AI, 2 RS) times as much as without reference sires' use (None). Much of that advantage accrued due to greater gains in the smaller flocks (100-300 ewes), although benefits persisted even in the largest flocks. Use of more RS via artificial insemination resulted in more gain yet the differences among the SRS were comparatively small. Inbreeding was higher with within-flock selection than with SRS, with lower inbreeding rates with natural service and when more RS were used. Even so, the average inbreeding coefficient remained low – less than 2.5% after 15 years. In Figure 1, the curvilinear rise in connectedness over time among the SRS scenarios is shown. Connectedness accumulated more quickly as more RS were used, and thus more ewes mated to RS. With natural service, connectedness lagged in early years yet accrued at a similar rate as the comparable AI scenario once the scheme became established.

Table 1 Genetic gain for weaning weight and inbreeding after 15 year of selection for different numbers and methods of use of reference sires (RS)

RS use		Genetic	Inbreeding
Method†	No.	gain (kg)	coefficient (%)
AI	1	6.75 (0.10)	1.97 (0.07)
	2	6.98 (0.11)	1.73 (0.08)
	3	6.75 (0.12)	1.31 (0.05)
NS	2	6.70 (0.10)	1.47 (0.05)
None	0	6.40 (0.13)	2.30 (0.10)

† RS use by artificial insemination (AI), natural service (NS) or no RS used (None)

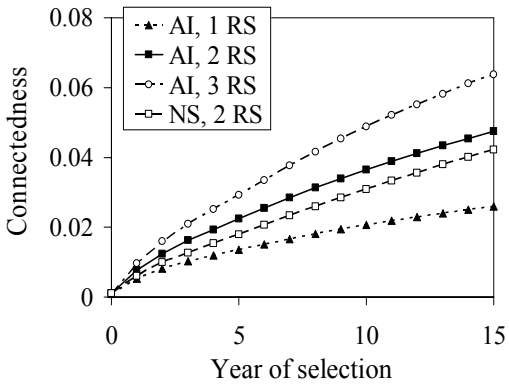

Figure 1 Connectedness by year throughout a 15-year time frame for four sire referencing schemes

Conclusions Sire referencing is clearly advantageous, with 2 RS mated by AI to 16 ewes achieving topmost gains in weaning weight when that the single goal. This strategy is currently used in hill breed SRS. The use of NS rams resulted in only small losses in gain. Where AI deemed unfeasible, the exchange of rams could be an option for hill schemes. Although there is no absolute measure for sufficient connectedness, it is strongly related to bias in genetic evaluations (Lewis *et al.*, 2005). An industry guideline of 0.025 for 'good' connectedness is currently used. Given that guideline, the use of 2 RS establishes sound genetic links within two to three generations.

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Comparison of crossbred lamb performance from four breeds of sire mated to Welsh Mountain ewes in the hill environment

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Introduction In order to increase farm incomes, hill sheep farmers must improve the productivity and quality of their flocks. Recent changes to farm support schemes have focused attention on a number of options, including a change of ewe breed, genetic improvement of the flock, the use of crossbred ewes, or crossbred lamb production from native hill breeds. A switch to crossbred lamb production could lead to an immediate increase in the quality of production from hill ewes whilst maintaining a dam that is adapted to the hill environment. The aim of this work was to evaluate the performance of crossbred lambs from Welsh Mountain ewes.

Materials and methods Over a three year period, 1800 Welsh Mountain ewes were mated to either Cheviot, Poll Dorset, Lleyn or Texel sires using laparoscopic AI. Single lambs were reared on the hill and twins were reared on improved upland pastures. All lambs were weighed at birth, five, ten and sixteen (weaning) weeks of age. Male lambs were finished on farm, and slaughtered at a target fat class of 3L. Prior to slaughter, ultrasonic muscle and fat depths were measured at the third lumbar vertebra and carcass weight, fat class (2-4L) and conformation score (EUROP recoded 1-5) were recorded post-slaughter. The fixed effects of sire breed, year, age of dam (2- 6 years), litter size (single, twin and triplet), sex (entire male or female, growth data only), fat class (slaughter data only) and grazing type (rough grazing, semi-improved and improved) and all two-way interactions were tested using the REML procedure of Genstat, with sire fitted as a random effect. Significant effects ($P<0.05$) were retained in the final analyses.

Results Small differences ($P<0.05$) were observed between sire breeds in mean birth weights (Table 1). However, by five weeks of age lambs sired by Texel and Poll Dorset rams were significantly ($P<0.01$) heavier than both Cheviot x and Lleyn x lambs and this weight advantage was even greater by sixteen weeks. Texel x lambs reached slaughter 12 days earlier than Poll Dorset x lambs and were 23 to 25 days younger at slaughter than the Cheviot x and Lleyn x lambs (Table 2). Poll Dorset x and Texel x lambs were significantly ($P<0.01$) heavier at slaughter than Lleyn x lambs, with Cheviot x being intermediate. Texel x lambs produced a higher mean carcass weight ($P<0.05$) than the Cheviot x and Lleyn x, and had significantly greater eye muscle depth than the other three crosses ($P<0.001$). There were no significant differences ($P>0.05$) between sire breeds in back fat depth or killing out proportion.

Table 1 Effect of sire breed on the mean live weights (kg) of lambs at birth, 5, 10 and 16 weeks of age.

Weight at:	Cheviot x	Dorset x	Lleyn x	Texel x	SED	Sig
Birth	3.5^{ab}	3.6^{a}	3.4^{b}	3.6^{a}	0.08	*
5 weeks	13.9^{a}	14.6^{b}	13.5^{a}	14.8^{b}	0.35	**
10 weeks	21.5^{a}	22.6^{b}	20.8^{a}	22.8^{b}	0.41	***
16 weeks	26.5^{a}	27.9^{b}	26.1^{a}	28.1^{b}	0.46	***

$^{a, b}$ Means within a row with different superscripts were significantly different ($P<0.05$)

Table 2 Effect of sire breed on the mean values for the slaughter attributes of crossbred lambs

	Cheviot x	Dorset x	Lleyn x	Texel x	SED	Sig
Slaughter age (days)	195.4^{a}	182.2^{b}	192.8^{a}	170.1^{c}	5.14	***
Slaughter Weight (kg)	38.7^{ab}	39.4^{b}	37.8^{a}	39.5^{b}	0.49	**
Muscle depth (mm)	25.6^{a}	25.2^{a}	25.0^{a}	26.6^{b}	0.38	***
Back Fat depth (mm)	2.78	2.87	2.78	2.74	0.093	NS
Carcass weight (kg)	16.9^{a}	17.0^{ab}	16.7^{a}	17.3^{b}	0.21	*
Killing out proportion	0.436	0.433	0.443	0.438	0.0039	NS

$^{a, b, c}$ Means within a row with different superscripts were significantly different ($P<0.05$)

Conclusions In comparison to the more conventional choice of Cheviot x Welsh Mountain for use on hill pastures, crossbred progeny from Texel and Poll Dorset rams showed higher growth rates and reached slaughter earlier, thus demonstrating the potential of these crossbreeds for use in the hill environment. Lleyn and Cheviot crossbred lambs had similar performance throughout. Further work is underway to evaluate the performance of crossbred ewes from these breeds.

Acknowledgement Funding from Hybu Cig Cymru - Meat Promotion Wales is gratefully acknowledged.

Selection for carcass and maternal traits: effects on lambing difficulties in Scottish Blackface hill ewes

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Introduction The incidence of lambing difficulties is known to be affected by factors such as sex, birth rank (single versus multiple), sire breed and dam breed of the lamb (e.g. Smith *et al*, 1977). Some of these difficulties are thought to be largely due to the link with lamb birth weight (e.g. heavier in males and singles, leading to increased dystocia). Age and parity of the ewe also influence dystocia, as does body condition score pre-lambing (Carson *et al.* 2001). The aim of this study was to examine whether selection of hill sheep using a breeding index designed to improve both carcass and maternal traits affects the incidence of lambing difficulties.

Materials and Methods Lambing records (n=3705) were collected over 4 years, between 2001 and 2004, from two Scottish Blackface flocks (~600 ewes/flock) on hill farms of differing severity (farm 1 less harsh climate and topography than farm 2). Approximately one third of each flock was born into each of three genetic lines: a selection line (S) selected since 1998 on an index to improve carcass and maternal traits; a control line (C) selected on average index values; an industry line (I) selected by normal commercial (visual) means (Conington *et al.*, 2005). Average index scores for each farm in 2004 are shown in Table 1. All three lines were run as one flock within each farm. Ewes were brought off the hill to lamb in fenced paddocks, where every lamb was tagged and recorded within 24 hours of birth. Ewes were only assisted with lambing by the shepherd if necessary and all assisted lambings were recorded. A total of 235 lambings were assisted, 57% of which were on farm 1. The data were analysed using a binomial generalised linear mixed model (GLMM) with a logit link function in Genstat (Lane and Payne, 1996). The fitted model (model 1) included: genetic line, birth rank, year, ewe age, lamb sex and whether the lamb was born dead or alive as fixed effects; ewe pre-mating weight, ewe pre-lambing condition score and lambing date as covariates; the interaction between ewe pre-mating weight and line; ewe sire and lamb sire as random effects. The model was then re-run (model 2) including lamb birth weight as a covariate and two-way interactions of lamb birth weight with sex, farm, and lambing date. Least-squares means for probability of lambing difficulties in each genetic line were tested for significant differences.

Results Ewes from the S line did not differ significantly from either the C or I line in their probability of lambing difficulty (Table 2). C line ewes had the lowest probability of lambing difficulty, which was significantly lower than that of the I line (highest probability). These relationships existed even after the model was adjusted for lamb birth weight (model 2), suggesting that factors other than birth weight (which increased lambing difficulty) caused the need for increased lambing assistance in the I line compared to the C line. Using both models, lambing difficulties were significantly affected by ewe sire and lamb sire (suggesting a genetic component), year, ewe age (highest at 2-years-old), lamb sex (male>female), still births (higher) and ewe pre-mating weight (higher weights = less assistance). However, no significant differences were observed due to farm, lambing date or pre-lambing condition score, probably due to management methods. Ewes producing twins required less assistance than ewes with single lambs. However, after adjusting for lamb birth weights this pattern was reversed. Birth weight increased lambing difficulties less with female lambs and at farm 2 (lighter lambs).

Table 1 Average index scores at each farm in 2004

Line	Farm 1	Farm 2
S	331	244
C	49	-37
I	181	49

Table 2 Least-squares means for probability of lambing difficulties within each genetic line

Line	Model 1	Model 2
S	0.069^{ab}	0.071^{ab}
C	0.048^{b}	0.049^{b}
I	0.099^{a}	0.106^{a}

Means within a column sharing a common character in their superscript are not significantly different (P>0.05)

Conclusions These results suggest that there has been no significant change in the incidence of ewes requiring assistance at lambing as a result of selecting Scottish Blackface ewes for improved carcass and maternal performance (line S), compared to a control line (C), or to a line selected by normal commercial (visual) methods (I). The higher incidence in the I line compared to C line may be due to visual selection for a particular body shape which also increases lambing difficulties. Monitoring of this trait should continue following additional generations of selection. Further studies on genetic relationships between lambing difficulties and other traits would be useful to help make future breeding decisions to improve productivity whilst maintaining high standards of animal welfare in hill flocks.

Acknowledgements Thanks to Defra for funding this project and A. McLaren and M. Steel for data collection.

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Evaluation of National Research Council 2001 and Cornell Pennsylvania Miner softwares for predicting dry matter intake of Holstein cows during the midlactation

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Introduction Predicting dry matter intake (DMI), precisely and accurately is important to prepare balanced rations when on farm estimates of feed intake are not available (Hayirli *et al*., 2003). Lack of accuracy in prediction of feed intake may result in nutrient underfeeding or overfeeding affecting to animal performance, animal health or dairy farm environmental impact (NRC2001). The 2001 "Nutrient Requirements of Dairy Cattle" recommended a DMI prediction equation based on animal factors that were evaluated with treatment means from experiments published in the Journal of Dairy Science between 1988 and 1998 (NRC2001). The variables used to evaluate DMI prediction are mean square prediction error (MSPE) and relative prediction error (RPE). MSPE accounted as $1/N\ (A-P)^2$ that N is number of comparisons, A is actual DMI monitored in farm, and P is predicted DMI in each software. The objective of this investigation was to evaluate the accuracy of National Research Council 2001 and Cornell Pennsylvania Miner softwares in predicting DMI of lactating dairy cows in midlactation.

Material and methods Sixteen lactating Holstein dairy cows (8 cows in second and 8 cows in third lactation period) averaging BW=642± 23kg, days in milk=55± 11d and MY=32± 0.9kg were randomly assigned in a completely randomized block design that each treatment has 4 cows in second and 4 cows in third lactation period. Cows allocated in individual pens. The characteristics (BW=650kg, days in milk=60d, MY=32kg, Milk fat=3.5% and Milk protein=3.2%) used as same inputs to formulating the rations by NRC 2001 and CPM-dairy softwares with the same feedstuffs. The formulated rations fed to cows for 105 days that two weeks considered as adaptation period in the first of experiment. Milk composition data were collected weekly and cow's body weights were recorded monthly. Individual daily DMI measured by feed intake minus ort. Data were analyzed using GLM procedure of SAS (v8.2).

Results The overall mean of milk yield, milk components and the variables used to evaluate DMI prediction accuracy are given in table 1. DMI for the cows fed formulated rations by NRC 2001 and CPM softwares were 22.90 and 22.32kg/d respectively (P>0.05). Differences for MY, milk composition and milk efficiency (Kg milk yield per Kg DMI) were not significant between two groups. Mean square prediction error was $2.68 kg^2/d$ for NRC2001 and $7.95 kg^2/d$ for CPM, and relative prediction error was 7.16% and 12.63% respectively for NRC2001 and CPM softwares.

Table 1 The overall mean of milk yield, milk components and variables used to evaluate accuracy of DMI prediction[1]

Item	Softwares		SEM	Sig
	NRC2001	CPM-Dairy		
Milk yield (kg/d)	29.80	30.25	1.08	NS
Fat yield (g/d)	950	930	80	NS
Protein yield (g/d)	860	840	70	NS
Efficiency (MY/DMI)	1.30	1.35	0.09	NS
Predicted DMI (kg/d)	21.9	20.6	-	-
Actual DMI (kg/d)	22.90	22.32	-	-
MSPE (kg^2/d)	2.68	7.95	-	-
MPE (kg/d)	1.64	2.82	-	-
RPE%	7.16	12.63	-	-

[1]Definitions: MSPE =mean square prediction error (kg2/d); MPE =mean prediction error (kg/d), defined as the positive square root of the MSPE; RPE =relative prediction error (%), defined as the MPE divided by the mean of the observed intake values.

Conclusions By increasing the RPE rate, accuracy of the DMI prediction decreases. The results of the present study showed that NRC2001 software predicts DMI more accurately than CPM software. Fuentes-pila *et al*., (2003) reported the greater accuracy and robustness of the NRC 2001 equation when predicting average DMI. DMI prediction in NRC2001 is base of empirical equations (NRC2001), so it has more conformity to actual DMI monitored in farm. This investigation showed that predicted DMI of NRC2001 is more reliable to formulating the lactating cows ration in midlactation.

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Prediction of manure nitrogen output of dairy cows from animal and dietary factors

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Introduction In 1991, the European Union introduced the Nitrates Directive which aims to prevent the pollution of groundwater and surface water by nitrates arising from agricultural sources. One mandatory measure stipulated by the Directive involves a limit, set at 170 kg/hectare organic N (manure N), on the amount of livestock manure which may be applied to land each year. This limit will have very significant implications for stocking rates on livestock farms. The objective of the present study was to use total diet digestibility data obtained at this Institute to develop prediction equations for manure N output in lactating dairy cows.

Material and methods The dataset used was obtained from 535 Holstein-Friesian and 29 Norwegian lactating cows in 26 total diet digestibility studies undertaken at this Institute from 1990 to 2002. The animals were of various genetic merits, parity (1-9), live weight (385-781kg) and milk yields (6.1-49.1 kg/d). Forages examined included 71 perennial ryegrass silages, 3 fresh grasses and 1 fodder beet. A total of 49 cows were offered forage as the sole diet, but remaining cattle (n = 515) were offered forages with various proportions of concentrates (211-876, mean 539, s.d. 135.4 g/kg DM). All animals were offered either forage or the complete diet *ad libitum*. Prior to commencing digestibility studies, all cows were loose housed in cubicle accommodation and offered experimental diets for at least 20 days. Animals were then transferred to metabolism units and housed for 8 days with total collection of faeces and urine during the final six days. Linear and multiple regression models were used to develop prediction equations for manure N output. Two sets of variables were used, i.e., animal and dietary variables with or without total N intake, as N intake may not be always available in commercial practice.

Results The mean, s.d., minimum and maximum values (g/d) for N intake were respectively 486, 129.6, 155 and 874; manure N output 351, 97.3, 130 and 679; Milk N output 109, 33.0, 24 and 231 and retained N 26, 35.0, –75 and 150. All relationships in Table 1 are significant ($P < 0.001$) and each predictor had a significant effect on the relationship ($P < 0.001$). Prediction of manure N output using live weight and milk yield, either alone (Figure 1), or combined, produced a low R^2 (0.227 – 0.474) and a high s.e. (70.6 – 85.6) (Eqs. [1] – [3]). When dietary CP concentration was added to equation [3], the R^2 value was considerably increased to 0.754 and s.e. value reduced to 48.2 (Eq. [4]). However, prediction of manure N output using N intake produced a very high R^2 (0.901) and a very low s.e. (30.6) (Figure 1, Eq. [5a]). The omission of the constant had no effect on the R^2 and s.e. (Eq. [5b]). The addition of live weight and milk yield to equation [5a] only resulted in small effects on the R^2 (increased to 0.910) and s.e. (reduced to 29.3) (Eq. [6]).

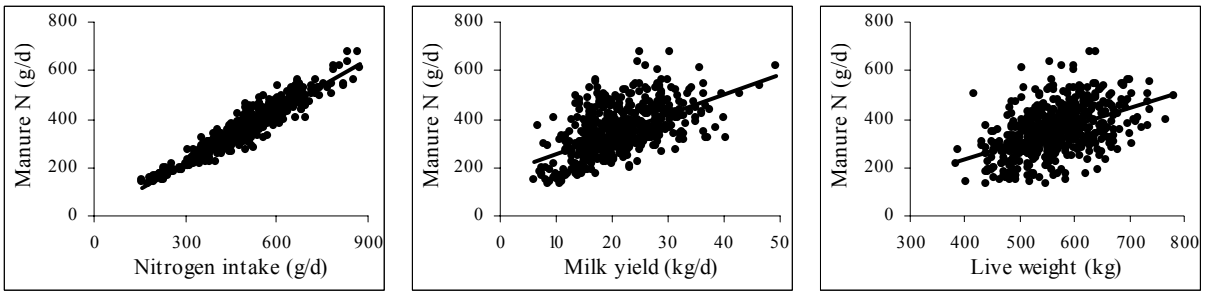

Figure 1 The relationships between manure N output and N intake, milk yield and live weight in dairy cows

Table 1 Prediction equations for manure N output (g/d) using N intake (NI, g/d), live weight (LW, kg), milk yield (MY, kg/d) and dietary CP concentration (CPc, g/kg DM) (values in parentheses are s.e.)

	Equations	R^2	s.e.	Eq. No
Manure N output	$= 0.711_{(0.055)}$ LW $- 51_{(31)}$	0.227	85.6	(1)
	$8.283_{(0.514)}$ MY $+ 174_{(12)}$	0.315	80.5	(2)
	$0.602_{(0.046)}$ LW $+ 7.411_{(0.455)}$ MY $- 147_{(27)}$	0.474	70.6	(3)
	$[0.00287_{(0.00012)}$ LW $+ 0.02429_{(0.00161)}$ MY$]$ CPc $- 44_{(11)}$	0.754	48.2	(4)
	$0.713_{(0.010)}$ NI $+ 5_{(5)}$	0.901	30.6	(5a)
	$0.722_{(0.003)}$ NI	0.901	30.6	(5b)
	$0.749_{(0.014)}$ NI $+ 0.065_{(0.022)}$ LW $- 1.515_{(0.255)}$ MY $- 17_{(11)}$	0.910	29.3	(6)

Conclusion Manure N output can be very accurately predicted from N intake in dairy cows. Prediction of manure N output using live weight and milk yield, either alone, or combined, can cause considerable errors, but the prediction accuracy can be greatly improved when dietary CP concentration is added as a primary predictor

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How milk-fed dairy calves perform in stable verses dynamic groups

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Introduction Conditions concerning group size, composition/homogeneity and stability may have influence on the success of rearing calves in groups. Introduction of calves into groups do determine not only age dispersion, but do also influence hygiene and patterns of disease transfer. Previous results indicate that large groups may increase the morbidity however we do not know the effect of moderate group size (6-8).
The objective of present study was to achieve increased knowledge in how introduction of calves into group housing systems did influence young calf performance. The present study compare data from calves in groups where new calves were continuously introduced (dynamic groups) and from calves in stable groups were all calves were introduced simultaneously ("All in all out"). The following hypothesizes was tested: "All in all out" introduction of calves into group housing systems is expected to have a positive influence on calves health and growth compared to continuous introduction of calves.

Materials and methods The experiment was carried out at six large (230-450 cows) commercial Danish dairy herds in the period from Dec 2003 to June 2004. All calves at all farms were started up in similar single pens. At approximately 3 weeks of age calves were randomly assigned to one of following two treatments. Both treatments were set up on all 6 farms.
Stable groups (All in all out): Six calves in the age of 3-5 weeks were simultaneously introduced to an empty, clean and straw bedded group housing pen. Six weeks later all calves left the pen, which was cleaned and left empty for one week before the next batch entered.
Dynamic groups: At weekly basis, three week old calves were introduced to an already established group where they replaced nine week old calves in order to maintain a group size of six. At any given time age within group varied from 3-9 weeks. Pens were cleaned every third month.
All calves were fed whole milk (waste milk), water and an unmedicated commercial calf-starter ad lib in open troughs. Calves (n=769) were weighed by the herdsman at entrance in groups and again when leaving the groups. From the two registered weights the daily live weight gain was calculated. Calves (n=490) was examined by a researcher on monthly basis. Registrations included diarrhoea (stool consistency), pneumonia, nasal discharge, elevated respiratory rate (>30/min), abnormal respiratory sound (crackles and wheezes), stool contaminated hair coat, otherwise subnormal hair coat finish (dull and dry with loose and/or missing hair), and finally rectal temperature.

Statistical methods Daily weight gain was analysed by means of a linear normal model with treatment and herd as systematic effects and a within-herd calf-group indicator as random effects in order to account for possible correlation between calves within the same housing group. The calculations where performed with Proc Mixed, with a random statement (SAS Institute). The prevalence's of the "illness indicators" are analysed by means of logistic regression modified to allow for a possible correlation between calves within the same housing group within-herd. These models where estimated using Proc Genmod with a repeated statement (SAS Institute).

Results Calves in stable groups had significantly higher daily weight gain than calves in dynamic groups (874 vs. 810 g/d, P=0.0063) and varied also significantly between farms (769-943 g/d, P<0.001).
Data from clinical examinations are presented in table 1. Calves with diarrhoea had significantly higher rectal temperature than healthy calves (39.1 and 38.1 °C, respectively, P<0.001). Rectal temperatures were significantly negatively correlated to daily weight gain (P=0.0147), such that daily weight gain was reduced by 52 gram for each one °C increase.

Table 1 Clinical registrations presented as percent of examined calves showing the respectively symptoms

Variable	Stable groups "All in all out"	Dynamic	*P*
Diarrhoea (% of calves)	21.5	43.1	<0.001
Pneumonia (% of calves)	24.3	44.1	<0.001
Nasal discharge (% of calves)	35.5	69.7	<0.001
Elevated resp. rate (% of calves)	7.9	23.9	<0.001
Abnormal resp. sound (% of calves)	3.7	18.5	<0.001
Stool contam. hair coat (% of calves)	18.2	44.1	<0.001
Subnormal hair coat finish (% of calves)	11.2	30.0	<0.001

Conclusions Our results confirm our hypothesis, and stress the importance of "hygiene units" containing calves of similar age. We assume that the decreased level of respiratory disease among "All in all out" calves is a consequence of lower infection pressure and also due to less diarrhoea. Future research might include relations between stress, group housing management and calf performance.

Performance comparison of lactating Holstein cows fed rations formulated by Spartan dairy ration balancer and Cornell Net Carbohydrate and Protein System

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Introduction The profitability, competitiveness and sustainability of dairy industry depend on minimizing feed costs while reducing nutrient excretion to minimize environmental impacts. A variety of software programs are available that designed to evaluate and formulate rations for dairy cattle. These programs differ greatly in their components and the types of objective functions available for ration formulation and completeness and versatility of report writing (Eastridge *et al*., 1998; Fox *et al*., 2003). The purpose of this study was to compare the performance of lactating dairy cows fed rations formulated with Spartan dairy ration balancer (Spartan) and Cornell Net Carbohydrate and Protein System (CNCPS) software.

Materials and methods 16 Holstein cows (eight cows in second and eight cows in third lactation period) averaging 622 (sd. 42) kg body weight (BW), 55 (sd. 19) days in milk (DIM), and 32 (sd. 0.8) kg of milk yield (MY) were randomly assigned in Two groups of eight cows each (four cows in second and four cows in third lactation period).Two rations using the same feedstuff and similar inputs (DIM 60, MY 32 Kg, BW 650 kg, milk fat 3.5%, milk protein 3.2%) were formulated as mixed by Spartan and CNCPS for each group of cows. Experimental diets fed individually for 90 days three times a day to allow 10% feed refusal. Daily dry matter intake (DMI) and milk yield were recorded. Milk composition data were collected weekly and each cow's body weight was recorded monthly. Data were analyzed as completely randomized block design. A comparison of least square means between each treatment was carried out using GLM procedure of SAS (SAS 1996) and appropriate covariance (DIM) as following model: $Yij = \mu + T_i + R_j + \beta(X_{ij} - X_{..}) + e_{ij}$. That Y_{ij} is observations, μ is the overall mean, T_i is the treatment effect (formulated rations), R_j is the block effect (calving number), β is regression coefficient, X_{ij} is days in milk and $X_{..}$ is the mean of days in milk.

Results The overall means of some of the traits measured are in Table 1. The cows fed ration formulated by Spartan had higher crude protein intake than those fed ration formulated by CNCPS (*P< 0.05*). However, differences for dry matter intake (DMI) and net energy intake were not significant. As well, differences for MY, milk composition and milk efficiency were not significant between two groups. The cost of feed intake per cow/day and per kg of MY, was significantly affected by the different treatments (rations). Comparing Spartan and CNCPS with regard to ration cost, the latter formulated a cheaper ration.

Table 1 Effects of formulated rations by Spartan and CNCPS on cows' performance and feed cost per milk yield.

Item	Formulated rations		s.e.m	*P*
	Spartan	CNCPS		
Dry matter intake kg/d	22.5	22.34	1.28	0.63
Protein intake (crude protein), g /d	3.625	3.470	0.11	0.031
Energy intake (NE_l), Mcal /d	37.8	38.3	1.43	0.24
Milk				
Yield kg/d	30.7	29.5	1.34	0.19
Fat g/d	31.7	31.5	0.6	0.28
Protein g/d	30.2	30.0	0.48	0.22
Efficiency (MY per DMI), kg	1.36	1.32	0.07	0.16
Cost of feed intake per kg milk yield, $	0.154	0.138	0.004	0.007

Conclusion Results of this study demonstrate that CNCPS could decrease the need for protein supplement in lactating dairy cow's diet and feed cost per Kg of milk yield. Based on these results CNCPS could increase profit in dairy farms.

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Effect of sample position within a clamp on the nutritive value of fermented and urea-treated whole crop wheat

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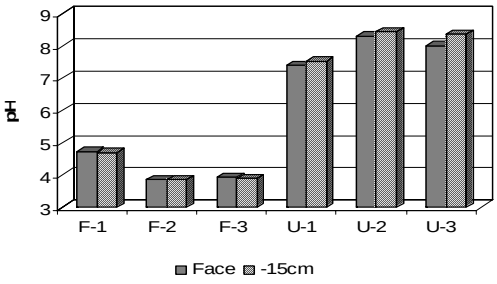
Introduction Much research has been conducted on the effects of whole-crop wheat (WCW) on dairy cow intake, performance and apparent digestibility. Recently attention has focussed on more accurately predicting the nutritive value of WCW. By contrast, relatively little research has been conducted to examine the variation in the chemical composition of WCW within a clamp, and there is little information on the most suitable sampling protocol. The objective of the current experiment was to determine the variation in chemical composition within clamps of fermented WCW and high DM, urea-treated WCW to assist in producing guidelines for representative sampling.

Materials and methods Six clamps on commercial UK dairy farms were used: three that contained fermented WCW and three that contained higher DM, processed and urea-treated WCW. All forages were produced from winter wheat and were ensiled in unroofed clamps. Within each clamp samples were taken at 8 sites and at two positions within each site: from the clamp face and at a depth of 15 cm behind the face. The 8 sites were; at a height of approximately half of the settled clamp, samples were taken at 35, 100 and 200 cm in from the left hand side wall; from the top of the clamp and mid-way along the length of the clamp, samples were collected at 35 and 100 cm down from the surface; from the floor of the clamp and mid-way along the length of the clamp, samples were collected at 35 and 100 cm up from the floor. An additional sample was taken from the middle (height and width) of the clamp. Sub-samples were analysed for DM, pH, N and NDF. The data was analysed as a factorial design with main effects of preservation, clamp, site within clamp, position (face or 15cm in) and their interaction. Analysis was conducted using Genstat 7.1 (VSN Int. Ltd., Oxford, U.K.).

Results The fermented whole crop forages had a lower (P<0.001) DM than the urea-treated clamps (490 *vs*. 830 g/kg respectively). The addition of urea at ensiling was reflected in a higher (P<0.001) pH and crude protein (Fig 1.), whilst NDF content was similar between both types of forage preservation, averaging 417g/kg DM. Within the fermented clamps forage DM was higher (P =0.004) at the face compared with 15cm in from the face (mean of 508 *vs*. 473 g/kg respectively), whilst there was a trend (P=0.066) for DM to be lower at the face in the urea-treated clamps. Within the fermented clamps there was no significant effect of position of sampling on pH (Fig 1a) but was lower at the face in the urea-treated clamps (mean values of pH 7.9 and 8.1 respectively; P < 0.001). Crude protein content (g/kg DM) was not affected by sampling position within the fermented clamps, but there was a trend (P = 0.054) for it to be lower at the face within the urea-treated clamps Fig 1b). Forage DM varied with sampling sites (P<0.001) but there was no clear pattern. Within the fermented clamps forage pH also varied with site (P<0.001) and generally decreased towards the bottom of the clamp. Crude protein content was highly variable and there was no clear effect of site. There was also no clear effect of sampling site on NDF content within the fermented clamps, although the NDF content generally decreased towards the bottom of the urea-treated clamps. A suggested acceptable range of sample analysis and the estimated number of samples at 95% confidence interval to fall within this range is presented in Table 1. Less variable parameters (e.g. pH) would require fewer samples than more variable parameters (e.g. NDF).

a)



b)

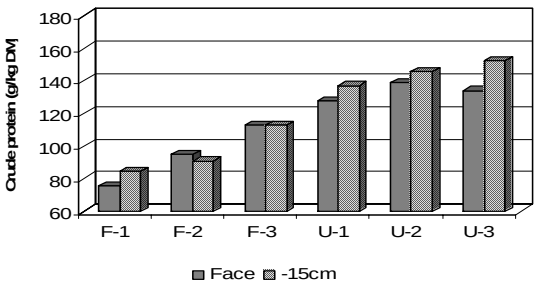


Figure 1 Effect of position (face or 15cm behind the face) on the pH (a) and crude protein content (b) in three fermented (F1, F2, F3) and urea-treated (U1, U2, U3) clamps: s.e.d. = 0.0425 and 8.25 for Figs a and b respectively.

Conclusions For the range of acceptable values outlined, the minimum number of samples required to provide a representative sample would be at least six per clamp. It is recommended that these are taken at 6 vertical and 4-6 horizontal positions in a "W" fashion across the clamp, and at 15cm in from the face. For a narrower range of values more samples would be required.

Table 1 No. samples required per clamp: (95% conf interval)

	Fermented		Urea-treated	
	Range	No. samples	Range	No. samples
DM (g/kg)	460-520	6	810-850	6
pH	4.1-4.3	1	7.9-8.1	5
CP (g/kg DM)	80-110	5	125-155	7
NDF (g/kg DM)	395-455	7	370-450	6

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Polyphenol oxidase activity, protein complexing and lipid profiles in bovine red clover-boluses

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Introduction The enzyme polyphenol oxidase (PPO) requires both plant tissue damage and the presence of oxygen to become activated. It has been shown that with a large window of opportunity for oxygen activation, such as during ensilage of red clover, increases in both dietary-nitrogen utilisation and the concentration of polyunsaturated fatty acids in ruminant products can be achieved. However, there is little evidence as to whether such responses can be achieved during the grazing of fresh red clover, where the window of opportunity for oxygen activation is limited to the mastication period (Lee *et al*. 2006). This experiment investigated whether mastication of fresh red clover would result in the activation of PPO and its potential effects on protein and lipid profiles in the resulting bolus.

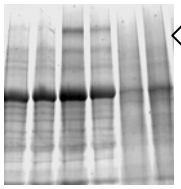
Materials and methods The rumen of a dairy cow fitted with a rumen cannula was emptied. The cow was then offered freshly cut red clover and the ingested boluses caught at the oesophageal orifice in the reticulo-rumen. Six boluses (B) were flash-frozen in liquid N_2 in less than 60 seconds and another six were immediately transferred to air-tight jars containing strained rumen fluid and incubated for 1h at 39^{o}C. These were then removed and the incubated boluses (IB) flash-frozen with liquid N_2. Six replicate samples of the fresh red clover herbage (H) were frozen within 30 minutes of harvesting and all samples stored at -80^{o}C prior to analysis. Lipid was extracted using chloroform : methanol and fractionated using TLC. The separated fractions were converted to methyl esters and analysed by GC. PPO activity, protein and phenol concentration were determined spectrophotometrically as described by Lee *et al*. (2005). Extracted protein was separated by sodium dodecyl sulphate polyacrylamide gel electrophoresis (SDS-PAGE) to visualise protein complexing. Data was analysed using a general ANOVA (Genstat 8.1, Lawes, Agricultural Trust, 2005).

Results There were no significant differences in the lipid profiles of B and H. However IB resulted in a significant drop in the polar and triacylglycerol fractions and a rise in the diacylglycerol and free fatty acid fractions. The concentration of extractable protein and protein-bound phenol was higher in the boluses and in particular IB compared with H. PPO content was higher in H and B than IB but was significantly more activated in both B and IB than H. Figure 1. shows the SDS-PAGE plate for two of each of the three sample types. The B samples show clear protein complexing by the appearance of high-molecular weight bands which are absent in the H and IB samples.

Table 1 Polyphenol oxidase activity, protein and phenol concentrations and lipid profiles in red clover (H), red clover-boluses (B) and red-clover boluses incubated in rumen fluid at 39^{o}C for 1h (IB).

	H	B	IB	s.e.d.	P
Total lipid (mg/g DM)	18.4	17.5	17.6	1.57	NS
Polar fraction (g/100g lipid)	75.0^{a}	78.5^{a}	27.8^{b}	7.78	***
Diacylglycerol (g/100g lipid)	2.77^{b}	3.41^{b}	7.82^{a}	4.31	*
Triacylglycerol (g/100g lipid)	17.0^{a}	15.5^{a}	8.28^{b}	3.95	***
Free Fatty acids (g/100g lipid)	5.19^{b}	2.58^{b}	56.1^{a}	4.95	***
Protein (mg/g DM)	126.6^{c}	167.9^{b}	219.4^{a}	29.48	***
Protein bound Phenol (mg/g DM)	0.54^{b}	3.37^{a}	4.37^{a}	1.02	***
Total PPO activity (ΔOptical density 420nm/g DM/min)	475.0^{a}	328.4^{a}	32.4^{b}	83.47	***
PPO activation (% of total)	41.2^{b}	90.1^{a}	100.0^{a}	22.21	***

^{abc} Different superscripts within rows indicate significant differences.



⇦ Complexed protein bands in lanes 3 and 4

Figure 1 SDS-PAGE showing bands of protein and the formation of complexed protein in the bolus extractions. Lanes: 1 and 2, Herbage; 3 and 4, Bolus; 5 and 6, Incubated Bolus.

Conclusions Mastication resulted in the activation of PPO and the production of phenol/protein complexes as shown by SDS-PAGE and the increase in protein bound phenol concentration between H and B. The changes in the lipid profiles in IB are indicative of the actions of lipolysis and the higher protein content is due to microbial contamination. The PPO in IB appears to have been partly washed out, but that which remained was 100% active.

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The effects of maturity of maize at harvest and level of maize in forage based diets on the performance of beef cattle

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Introduction Major developments, both in agronomic practices and plant breeding, have enabled the consistent production of high yields of forage maize in areas in which it was not possible to grow the crop 20-30 years ago (Keady 2005). Recent studies (Keady *et al* 2002, 2003, Keady and Kilpatrick 2004) have clearly illustrated that the inclusion of maize silage consistently increased the performance of beef and dairy cattle. The objective of the present study was to evaluate the effects of maturity of maize at harvest and level of inclusion in forage based diets on feed intake and animal performance of finishing beef cattle. Furthermore the potential concentrate sparing effect of maize silage inclusion was also determined.

Materials and methods Two maize silages were offered as the sole forage or in addition to grass silage at a ratio of 50:50 maize/grass silage and supplemented with 4 kg concentrates/day. The grass silage was offered as the sole forage supplemented with either 4 or 8 kg concentrates/day. The six treatments formed a 2x2+2 factorial experiment and were offered to 72 continental cross beef cattle (initial liveweight 522kg (s.d. x 23.5)) in a fully balanced completely randomised design and was analysed using Genstat 5. The maize silages were produced from the varieties Nescio which was sown in the open on May 10 (LDM) and Goldcob which was sown under complete cover plastic match on April 27 (HDM). The grass silage (GS) was ensiled on 1 July from the primary regrowth of predominantly perennial ryegrass swards, precision chopped after 24 hours wilting. The forages were offered *ad libitum* following mixing in a diet wagon once per day whilst the concentrate was offered in two equal feeds. The concentrate consisted of 500, 150, 140, 180 and 30 g/kg of barley, maize meal, sugar beet pulp, soyabean and molasses. Each animal received 100 g mineral vitamin mixture per day with the concentrate. Total diet digestibility of the diets was determined using three cattle per treatment.

Results The grass silage was of a medium feed value with DM concentration, intake potential and predicted D-value of 232g/kg, 73g/kg $W^{0.75}$ and 680 g/kgDM respectively. For the LDM and HDM maize silage DM and predicted starch concentrations, were 217 and 304 g/kg and 67 and 269 g/kg DM respectively. The effects of stage of maize maturity at harvest and level of inclusion in the diet on animal performance are presented in Table 1. Relative to grass silage, LDM offered as the sole forage did not alter (P>0.05) food intake or animal performance. Increasing maize maturity at harvest increased feed intake, final liveweight, liveweight gain, carcass gain and carcass weight. Level of inclusion did not alter food intake or animal performance. There was a significant interaction between maize maturity at harvest and level of inclusion for final liveweight, liveweight gain, carcass weight and carcass gain. Final liveweight, liveweight gain, carcass gain and carcass weight did not differ due to stage of maturity at harvest at the low level of inclusion. However at the high level of inclusion HDM silage improved, while the LDM silage reduced final liveweight, liveweight gain, carcass gain and carcass weight. Increasing concentrate supplementation with the grass silage increased food intake and animal performance. Treatment did not alter (P>0.05) carcass conformation classification or kill out proportion. The potential concentrate sparing effect, as determined by carcass gain, was calculated to be +1.3, -0.3, +1.3 and +2.5 kg/concentrate/head/day for the LDM offered as mixed forage or as sole forage and HDM offered as mixed forage and as sole forage based diets respectively.

Table 1 The effect of maize maturity at harvest and level of inclusion in the diet on animal performance

	Forage[1]							
	GS		LDM/GS	HDM/GS	LDM	HDM		
Concentrate (kg/d)	4	8	4	4	4	4	SEM	Sig
Forage intake (kg DM/d)	4.9^{b}	3.1^{a}	5.4^{bc}	5.8^{cd}	5.4^{bc}	6.4^{d}	0.19	***
Total intake (kg DM/d)	8.2^{a}	9.7^{c}	8.7^{ab}	9.1^{bc}	8.7^{ab}	9.7^{c}	0.19	***
Final liveweight (kg)	630^{ab}	692^{d}	650^{abc}	651^{bc}	627^{a}	671^{cd}	8.5	***
Liveweight gain (kg/d)	0.74^{ab}	1.17^{d}	0.86^{b}	0.88^{bc}	0.71^{a}	1.03^{cd}	0.058	***
Carcass gain (kg/d)	0.48^{a}	0.73^{c}	0.56^{ab}	0.56^{ab}	0.47^{a}	0.63^{bc}	0.037	***
Carcass weight (kg)	351^{a}	388^{c}	364^{ab}	364^{ab}	348^{a}	373^{bc}	5.43	***
Fat classification[2]	3.01^{a}	3.73^{c}	3.25^{ab}	3.46^{bc}	3.08^{ab}	3.28^{ab}	0.143	**

[1]GS = Grass silage, LDM = low DM maize silage, HDM = High DM maize silage
[2]EU fat classification, where 5= fat, 1 = lean

Conclusions Increasing the stage of maturity of maize at harvest significantly improved animal performance when offered as the sole forage. However when offered in mixed forage diets, regardless of stage of maturity at harvest, maize silage inclusion tended to improve performance. Feeding maize silage had a potential concentrate sparing effect of up to 2.5 kg/head/day depending on the maturity at harvest and level of inclusion in the diet.

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Selenium enriched winter barley in a diet for fattening bulls: effects on the selenium content in meat and organs

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Introduction Selenium (Se) is a trace element of importance. In plants, Se is present on an organic form which is more efficiently absorbed than the inorganic salts (Fairweather-Tait, 1997). In human diets, meat and meat by-products could provide more than 30% of the daily Se intake. The aim of the present work was to assess the effects of the incorporation of Se enriched barley in a finishing diet for growing fattening bulls in order to improve the Se content in beef meat and organs.

Material and methods The Belgian Blue herd from the Veterinary Faculty of the University of Liege was managed in a feeding system in which grazed grass and feedstuffs for the winter period were produced with or without Se enriched fertilizers (Cabaraux *et al.*, 2006). After the growing period, the male calves were fattened on a finishing diet in which winter barley enriched (Se group) or not (control – C group) in Se was included at a rate of 500 g/kg. The common complementary concentrate was made of soybean meal, sugar beet pulp, spelt, molasses, and mineral mixture without added Se. There were 7 and 4 bulls in the C and Se groups. The animals were slaughtered according to the finishing state.

Results The Se content in the C diet was 67 µg/kg dry matter (DM) while it was 213.5µg/kg DM when the Se enriched barley was offered. There were no effects of the Se content either on the animal performance or on the slaughter characteristics (Table 1). The initial and final live weights were 449.0 and 602.0 kg respectively. The average daily gain was 1.255 kg/d. The DM intake was 9.55 kg/d and the killing-out proportion was 659.5 g/kg. The Se enriched barley largely and significantly increased the Se content in the meat (509.0 *vs* 172.4 µg/kg DM in the *Longissimus thoracis* and 480.5 *vs* 153.1 µg/kg DM in the *Rectus abdominis*). There were also increases in the Se content in the testicles, lungs, kidneys and liver

Table 1 Animal performance, slaughter characteristics and selenium content in muscles and organs in fattening bulls offered a diet based on winter barley grown with fertilizers enriched or not in selenium

	Control	Selenium	SED	Trait effect
Animal performance				
Age at slaughter (d)	552.7	524.5	17.8	NS
Initial weight (kg)	452.5	445.4	48.6	NS
Final weight (kg)	607.4	596.5	23.6	NS
Gain (kg)	154.9	151.1	32.4	NS
Duration (d)	115.0	127.0	17.8	NS
Average daily gain (kg/d)	1.311	1.199	0.105	NS
Daily dry matter intake (kg/d)	9.50	9.59	0.12	NS
Slaughter characteristics				
Slaughter weight (kg)	569.3	561.0	27.0	NS
Warm carcass weight (kg)	372.0	373.5	19.1	NS
Killing-out proportion (g/kg)	653.3	665.7	0.8	NS
Se content (µg/kg dry matter)				
Muscles				
Longissimus thoracis	172.4	509.0	37.9	***
Rectus abdominis	153.1	480.5	39.7	***
Testicles	1949.0	2199.0	120.1	NS
Lungs	428.6	883.3	53.7	***
Kidneys	4799.0	6091.0	444.9	*
Liver	428.7	1149.2	66.8	***

NS: P>0.05; *: P<0.05; ***: P<0.001

Conclusions In the present trial, the improvement in the Se content of beef meat and organs according to the "soil-plant-animal-products axis" was obtained by use of Se enriched fertilizers. This is a method of interest to provide organic Se for human dietetics.

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The effects of grain type, storage and processing method, and feed level on beef cattle performance

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Introduction Recently new techniques have been developed for storing grain which reduce the need to process grain immediately prior to feeding. In a recent study Keady and Kilpatrick (2004) concluded that relative to grain which was treated with propionic acid and rolled prior to feeding, crimping did not alter animal performance while urea treatment tended to decrease carcass gain. Furthermore Keady and Kilpatrick (2004) concluded that there were no interactions between grass silage feed value and grain feed level on animal performance. The objective of the present study was to evaluate the effects of grain storage and processing method on the performance of beef cattle offered grass silage based diets and grain at 3.5kg DM/day or *ad libitum*. Furthermore the feed value of barley and wheat was evaluated at two extreme levels of inclusion in the diet.

Materials and methods Winter wheat (FW) was harvested on 16 August and ensiled either crimped and treated with 4.1 litres/t fresh weight of a proprietary acid based additive or ensiled whole mixed with 21kg urea and 50 litres water/t FW . Representative areas of the crop were harvested conventionally on 2 September and treated with 3.0 litres/t FW of propionic acid prior to storage and rolled immediately prior to feeding. Barley was harvested and treated with 3.0 litres/t FW of propionic acid prior to storage and rolled immediately prior to feeding. Grass silage was harvested from the primary regrowth of predominantly perennial ryegrass based swards on 1 July and ensiled after 24 hours wilting, treated with a bacterial inoculant. Each grain treatment was offered at either 3.5kg DM supplemented with *ad libitum* silage or offered *ad libitum* supplemented with 1.5kg DM of silage. The 8 treatments were offered to 96 continental cross beef cattle (initial liveweight 561 kg (s.d.31.5)) using a randomised block design study for a feeding period of 145 days. Each animal received 100g mineral and vitamin mixture per day with the grain which was offered once daily. Total diet digestibility of the total diets was determined.

Results The silage was of a medium feed value with DM concentration, predicted intake value and D-value of 226g/kg, 69g/$W^{0.75}$ and 680g/kg DM respectively. The DM concentration of the conventional, crimped and urea treated wheat, and barley were 788, 689, 665 and 798g/kg respectively. The effects of grain type and storage and processing method on animal performance are presented in Table 1. Relative to conventional wheat, barley increased silage DM intake ($P<0.01$), otherwise there were no significant effects of grain type on food intake or animal performance. Relative to conventional and crimped wheat, urea treatment increased food intake, had an inferior ($P<0.001$) FCE and tended ($P=0.062$) to decrease carcass gain. Increasing grain feed level increased total DMI, final liveweight, liveweight gain, carcass weight, carcass gain, kill-out proportion, carcass fat classification, carcass conformation and FCE. There was a significant grain type by feed level interaction ($P<0.05$) for grain intake. When offered at the low level of feeding grain intakes were similar. However when offered *ad libitum* the intake of crimped wheat was lower than the urea treated wheat with intakes of 7.4, 7.1, 8.2 and 7.7 kg DM/d for conventional, crimped and urea treated wheat, and barley respectively.

Table 1 Effect of grain type, storage and processing method, and feed level on animal performance

	Grain (G)					Feed level (L)					
	Barley	Wheat			SEM			SEM	(G)	(L)	G x L
		Conv.	Crimp	Urea		Low	*Ad-lib*				
GI[1] (kgDM/d)	5.7	5.5	5.4	5.9	0.13	3.7^{a}	7.6^{b}	0.10	NS	***	*
TDMI[2] (kg/d)	8.9^{b}	8.6^{ab}	8.3^{a}	9.3^{c}	0.15	8.5^{a}	9.1^{b}	0.11	**	***	NS
Final LW (kg)	694	683	680	673	5.9	670^{a}	695^{b}	4.2	0.08	***	NS
LWG (kg/d)	0.91	0.83	0.83	0.77	0.038	0.75^{a}	0.92^{b}	0.027	0.06	***	NS
Carcass wt. (kg)	390	383	385	375	3.9	373^{a}	393^{b}	2.8	0.06	***	NS
Carcass gain (kg/d)	0.59	0.54	0.56	0.49	0.027	0.47^{a}	0.61^{b}	0.019	0.06	***	NS
Kill out (g/kg)	561	560	566	557	3.9	556^{a}	566^{b}	2.8	NS	**	NS
Fat class[3]	3.4	3.5	3.3	3.3	0.11	3.2^{a}	3.5^{b}	0.08	NS	*	NS
Conformation[4]	3.1	3.1	3.2	3.2	0.08	3.0^{a}	3.3^{b}	0.06	NS	**	NS
FCE[5]	16.5^{a}	17.4^{a}	15.3^{a}	21.3^{b}	0.81	19.7^{b}	15.5^{a}	0.58	***	***	NS
DMD[6]	0.760^{b}	0.777^{b}	0.768^{b}	0.702^{a}	0.0853	0.744	0.760	0.0603	***	NS	NS

[1]Grain intake, [2]Total dry matter intake, [3]EU fat classification, where 5 = fat, 1 = lean; [4]EUROP scale: 5, 4, 3, 2, 1 respectively, [5]food conversion efficiency, [6]dry matter digestibility

Conclusions It is concluded that urea treatment of wheat reduced diet digestibility and FCE and tended to decrease carcass gain relative to crimped and conventional treatment. There was no difference in the feed value of wheat and barley when offered at two extreme levels of inclusion in the diet. Increasing grain feed level required an additional 27.9kg grain DM/kg additional carcass gain.

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Effects of selenium enriched fertilizers on selenium content in feedstuffs and on the selenium status in a beef cattle herd

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Introduction Selenium (Se) is a trace element of importance in animals owing to its implication in many biological processes such as antioxidant mechanisms, immune response, reproduction system, thyroid metabolism and anti-cancer processes. However, the Se content in locally produced feedstuffs is low with as result a reduced Se status in the animal. The aim of the study was to assess the effects of Se enriched fertilizers on the Se content of feedstuffs and on the Se status of beef cattle.

Material and methods Since 2002, fertilizers enriched with Se were applied on pastures and arable lands of the Veterinary Faculty of Liege University (Se group). In the pastures for grass silage production 3g Se/ha were applied at the beginning of the season and after each cut. In the grazed pastures, 3g Se/ha were applied at each nitrogen spreading. For winter barley production 6 and 4 g Se were spread with the second and third nitrogen applications while for maize there was only one application of 8g/ha at sowing. There were control plots with no Se addition but with similar management (C group). The Belgian Blue suckling herd (35 cows at the beginning) was divided in two groups, the Se and the C groups. During the grazing season, the animals did not receive any supplement. During the winter period, they were offered a diet composed of 30% dry matter -DM- as grass silage, 30% DM as maize silage and 14% DM as barley, these three feedstuffs being produced with or without Se enriched fertilizer. The Se status of the animals was expressed as whole blood Se content measured by the activity of glutathione peroxidase in red blood cells on blood samples obtained three times over each period.

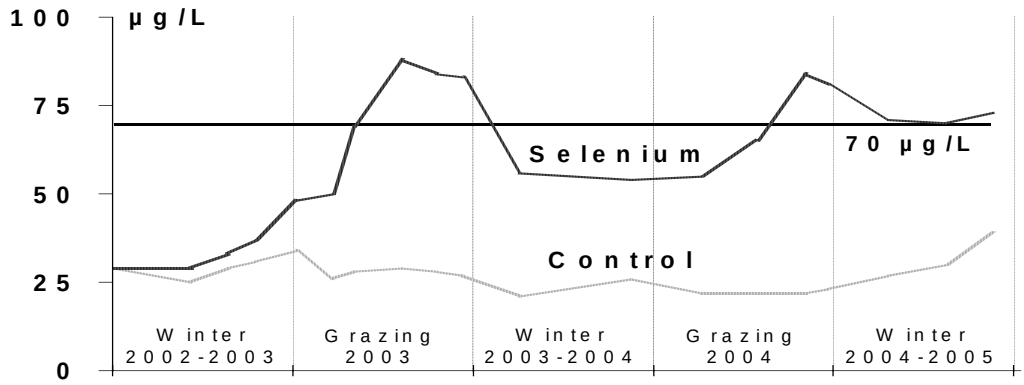
Results The Se supplementation did not affect the chemical composition of the different feedstuffs except the Se content (Table 1). There was an average 4.8 fold increase in the Se content of grazed grass (248.3 *vs* 51.8 µg/kg DM), 3.5 fold increase in grass silage (186.9 *vs* 53.4µg/kg DM), 6.5 fold increase in winter barley (286.3 *vs* 43.7 µg/kg DM) and 3.8 fold increase for maize silage (102.4 *vs* 27.0 µg/kg DM). The changes in the blood Se concentration over the different periods are given in Figure 1. At the beginning of the trial, the blood Se concentration was low at about 25µg/l. The concentration remained low over all the periods for the C group. By contrast, the concentration in Se started to increase during the first winter period with the Se supplemented diet. A further increase with a plateau was observed during the following grazing season. The blood Se concentration was than reduced during the second winter period but was still significantly higher than that in the C group. The pattern of the curve was similar for the two following periods. The slow increase in blood Se during the first winter period was associated to a three months half live of the red cells. The reduction in blood Se content during the winter periods as compared with the grazing periods was due to inclusion of purchased feedstuffs with low selenium content along with the enriched silages and winter barley.

Table 1 Concentration in Se (µg/kg DM) in different feedstuffs grown with (Se) or without (C) Se enriched fertilizer

	Winter 2002-2003		Grazing 2003		Winter 2003-2004		Grazing 2004		Winter 2004-2005		SED	Trait effect
	C	Se	C	Se	C	Se	C	Se	C	Se		
Number cows	17	18	16	16	23	17	16	19	18	20		
Grass grazed			54.0	266.0			49.6	230.6			43.9	***
Grass silage	54.2	165.2			50.1	183.5			56.0	211.9	27.6	***
Maize silage									27.0	102.4	4.1	***
Winter barley	27.0	225.0			27.3	263.9			76.9	369.4	17.2	***

***: P<0.001

Figure 1 Evolution of the whole blood selenium concentration (µg/L) measured by glutathione peroxidase activity in cattle. A concentration of 70 µg/L is considered as normal value.



Conclusion It can be concluded that the use of fertilizer containing Se can improve the Se content of locally produced feedstuffs and the Se status of cattle in areas with low Se availability.

The effect of forage : concentrate ratio on the performance of bulls slaughtered at a range of liveweights

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Introduction Previous studies (Steen 1995) have clearly illustrated that finishing male cattle entire increases animal performance and food conversion efficiency. Post implantation of the Mid Term Review of the Common Agricultural Policy, in the absence of subsidy, there is renewed interest in finishing beef cattle entire. Also with the expected contraction of the suckler herd post MTR of CAP there will be a reduced number of cattle available for finishing. Feed is one of main variable costs of finishing beef cattle. Consequently the objective of the present study was to evaluate the effect of forage:concentrate (F:C) ratio and weight at slaughter on the animal performance of ¾ or more continental bulls from the beef herd.

Materials and methods Grass silage was ensiled on 8 May after a 48 hour wilting period. The concentrate consisted of 400, 230, 170, 170 g/kg of barley, maize, sugar beet pulp, soyabean and molasses. The diets consisted of 50:50:forage concentrate and *ad libitum* concentrate supplemented with 1.2 kg silage DM. Each animals received 100g mineral and vitamin mixture per day daily. The diets were offered to 120, ¾ or greater, continental bull weanling calves (mean initial liveweight of 358 kg (s.d. 47.5). Equal numbers of cattle per treatment were slaughtered at random after either 191, 218, or 254 days on experiment. Total diet digestibility was determined using four animals per treatment. These data were analysed using Genstat regression procedures with a model including the factor treatment and slaughter weight as an independent variable, and their interaction. Predicted values were calculated for each treatment at a range of slaughter weights.

Results The silage had a high feed value with DM concentration and predicted D-value and intake potential of 238g/kg, 775g/kg DM and 84g/kg $W^{0.75}$ respectively. Slaughter weight did not effect (P>0.05) daily liveweight or carcass gains which were 1.60 and 0.86 kg/d respectively. There were no treatment by liveweight at slaughter interactions (P>0.05) on food intake or animal performances. The effect of weight at slaughter on animal performance is presented in Table 1. Increasing the weight at slaughter increased daily food intake, carcass weight, marbling score and food conversion ratio (FCR), and did not alter carcass conformation, food intake and fat classification. The effect of dietary treatment on animal performance is presented in Table 2. Decreasing the F:C ration increased food intake, kill-out proportion, carcass weight, carcass gain and decreased the FCR but did not alter liveweight gain, carcass gain, carcass fat classification or conformation, marbling score or FCR.

Table 1 Effects of slaughter weight on animal performance (prediction*)

	Slaughter weight (kg)								
	500	550	600	650	700	750	800	SEM	Sig
Total intake (kg DM/d)	8.2	8.4	8.6	8.7	8.9	9.1	9.3	0.17	*
Carcass weight (kg)	299	325	352	378	404	431	457	2.8	***
Conformation[1]	3.6	3.7	3.7	3.7	3.8	3.8	3.9	0.092	NS
Fat classification[2]	2.99	3.01	303	3.05	3.07	3.10	3.12	0.081	NS
Marbling[4]	1.30	1.50	1.69	1.89	2.08	2.28	2.47	0.111	***
FCR (kg feed/kg carcass)[3]	9.5	9.7	9.9	10.2	10.4	10.6	10.8	0.193	*

Table 2 Effect of forage: concentrate ratio on animal performance (prediction*)

	Forage : Concentrate ratio		SEM	
	50:50	0:100		Sig
Total intake	8.7	9.0	0.10	*
Kill out (g/kg)	574	584	3.0	*
Carcass weight (kg)	394	401	2.0	*
Liveweight gain (kg/d)	1.60	1.60	0.016	NS
Carcass gain (kg/d)	0.83	0.89	0.013	*
Fat calssification[2]	3.14	3.00	0.058	NS
Conformation[1]	3.8	3.8	0.08	NS
Marbling	2.13	1.94	0.079	NS
FCR (kg feed/kg carcass)[3]	10.5	10.1	0.19	*

*Values predicted from regression analysis [1]EUROP scale : 5,4,3,2,1, respectively. [2]EU Fat classification:3 where 5 = fat, 1 = lean, [3]Food conversion ratio. [4]Eight point scale: 1 = leanest, 5 = fattest

Conclusions Slaughter weight did not alter liveweight gain or carcass gain of young bulls slaughtered within the weight range of 500 to 800 kg liveweight. However food conversion ratio increased with increasing slaughter weight. Replacing 0.50 of the *ad libitum* concentrate diet with high feed value grass silage did not alter daily liveweight gain but decreased carcass gain by 0.06 kg/d and FCR by 0.4 kg feed DMI/kg carcass. Provided there is a market outlet, bulls from the beef herd can be taken to heavy weights effeciently.

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Relationships between manure N output and animal and dietary factors in beef cattle

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Introduction The Nitrates Directive has set a limit of 170 kg/hectare organic N (manure N) as the amount of livestock manure which may be applied to land each year. Consequently there is an increasing interest in finding approaches to mitigate manure N output in animal production. The objective of the present study was to use total diet digestibility data obtained at this Institute to develop prediction equations for manure N output in beef cattle.

Material and methods The dataset used was obtained from 286 beef cattle in 15 total diet digestibility studies undertaken at this Institute from 1984 to 2003. Forages offered were mainly grass silage (n = 278), with the exception of 8 animals that were offered grass silage plus whole crop wheat (n = 4) and grass silage plus maize silage (n = 4). These alternative forages accounted for less than 50% (DM basis) of the total forage consumed. The forages were offered either alone as a sole diet (n = 45), or as mixed diets with concentrates or separately from concentrates (n = 233, dietary forage proportion (DM basis) ranging from proportionately 0.20 to 0.84). Prior to commencing the digestibility studies, all animals were housed in individual stalls and offered experimental diets for at least 20 days. Animals were then transferred to metabolism units and housed for 8 days with total collection of faeces and urine during the final six days. Linear and multiple regression models were used to develop prediction equations for manure N output. Two sets of variables were used, i.e., animal and dietary variables with or without total N intake, as N intake may not always be available in commercial practice.

Results The mean, s.d., minimum and maximum data for live weight were respectively 381, 121.4, 153 and 580 kg; DM intake 6.8, 1.92, 3.2 and 10.9 kg/d; for N intake 167, 49.9, 73 and 316 g/d; manure N output 130, 38.5, 43 and 227 g/d and retained N 37, 18.6, –6 and 101 g/d. All relationships in Table 1 are significant ($P < 0.001$) and each predictor had a significant effect on the relationship ($P < 0.05$). Prediction of manure N output using live weight (Figure 1) produced a relatively low R^2 (0.55) and a high s.e. (26) (Eqs. [1a] – [1b]). The addition of dietary CP concentration considerably increased the R^2 to 0.723 and reduced the s.e. to 20.3 (Eq. [2]). However, the best predictor for manure N output is N intake as the R^2 is very high (0.890) and the s.e. very low (12.8) (Figure 1, Eq. [3a]). The omission of the constant had little effect on the R^2 and s.e. (Eq. [3b]). The addition of live weight and dietary forage proportion to equation [3a] only had small effects on the R^2 (increased to 0.904) and s.e. (reduced to 12.0) (Eqs. [4] – [5]).

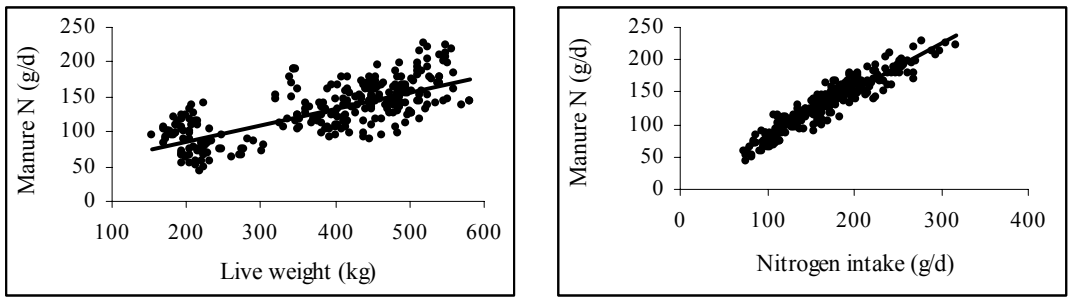

Figure 1 The relationships between manure N output and N intake and live weight in beef cattle

Table 1 Prediction equations for manure N output (g/d) using N intake (NI, g/d), live weight (LW, kg), dietary forage proportion (FP, g/kg DM) and dietary CP concentration (CPc, g/kg DM) (values in parentheses are s.e.)

Equations	R^2	s.e.	Eq. No	
Manure N output = $0.236_{(0.013)}$ LW + $40_{(5)}$		0.550	25.8	(1a)
$1.345_{(0.073)}$ $LW^{0.75}$ + $15_{(6)}$		0.547	25.9	(1b)
[$0.217_{(0.101)}$ + $0.00822_{(0.00061)}$ CPc] $LW^{0.75}$ + $4_{(5)}$		0.723	20.3	(2)
$0.729_{(0.015)}$ NI + $8_{(3)}$		0.890	12.8	(3a)
$0.774_{(0.004)}$ NI		0.887	13.0	(3b)
$0.653_{(0.021)}$ NI + $0.043_{(0.009)}$ LW + $4_{(3)}$		0.899	12.3	(4)
[$0.613_{(0.023)}$ + $0.000096_{(0.000024)}$ FP] NI + $0.050_{(0.009)}$ LW – $2_{(3)}$		0.904	12.0	(5)

Conclusion Manure N output of beef cattle can be very accurately predicted from N intake. Prediction of manure N output using live weight can cause errors, but the prediction accuracy can be greatly improved when dietary CP concentration is added as a primary predictor.

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Effect of sudden short term exercise on hind gut fermentation kinetics in the equine

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Introduction It has been demonstrated by Pagan *et al.* (1998) that horses on a regular extensive exercise regime have an increased whole-tract passage rate and hence a decreased feed degradability. Recent work by Dougal *et al.* (2005) reported that regularly exercised horses had a decreased rate of gas production from faecal slurries compared to non-exercised horses, despite an apparently enhanced bacterial population. Many horses are not exercised during the week but are exercised at the weekend and receive supplementary feed after exercise. This may be fed in vain due to a decrease in feed degradability associated with increased exercise (Pagan *et al.* 1998). The aim of this experiment was to asses the impact of sudden short term exercise on the extent of microbial fermentation within the hind gut of the equine.

Materials and methods Six pony mares, mean live weight (LW) 268 kg (s.d.=45.4) were used in a 2 x 2 latin square design. All animals received a restricted diet of 15 g/kg LW/d haylage fed in 2 equal meals at 08:00 h and 16:00 h. There was a 21 d adaptation period during which animals were walked (1.6 m/s) for 10 min and trotted (2.6 m/s) for 10 min in a horse walker prior to being returned to their individual stables. Each of the experimental periods lasted for 7 d. On the first day of the experimental period only, the animals on the exercised treatment were exercised for 30 min at trot with a 5 min warm up and cool down period at walk, whereas, the non-exercised animals were walked for 30 min in a horse walker prior to being returned to their stables. The second faecal sample passed after exercise was collected into a thermos flask and used as an inoculum source for gas production. During the remainder of the experimental period (days 2-7) all the animals were exercised at 10:00 at walk for 30 min per day and were kept stabled at all other times. Gas production was measured from faecal slurries incubated with 0.3 g DM of barley, maize of haylage as described by Lowman *et al.* (1999). Cumulative gas production curves, corrected to ml/g DM, were fitted to the model $P = a + b (1 - e^{(-ct)})$, where P = volume of gas at time t; a = the intercept of the gas volume curve at t = 0; b = volume of gas produced at asymptote; c = rate of gas production (h^{-1}) (Ørskov and McDonald, 1979). Data were analysed using the ANOVA procedure of Genstat 8.0.

Table 1 Mean cumulative gas production from all feedstuffs of exercised and non-exercised ponies

Incubation time (h)	Exercised GP (ml) n=6	Non-exercised GP (ml) n=6	s.e.d.	*P*
0	0	0	-	-
2	4	4	1.1	0.858
4	9	10	2.2	0.632
6	22	26	2.8	0.232
8	38	44	1.8	0.027
12	69	75	2.4	0.067
16	106	110	3.3	0.248
20	139	144	5.7	0.452
28	171	176	6.3	0.483
40	199	199	6.4	0.942
52	216	214	6.5	0.782
64	227	223	7.0	0.642
76	235	230	6.9	0.501
96	243	235	6.9	0.338
asymptote	260	245	10.3	0.218
rate (h^{-1})	0.043	0.048	0.0022	0.073

Results Mean total gas production was highest ($P<0.050$) in the slurries incubated with maize and lowest for those incubated with haylage with an intermediate value for barley (299, 213 and 245 ml respectively, s.e.d.=7.5). The rate of gas production was highest ($P<0.050$) in the slurries incubated with barley and lowest in those incubated with haylage with an intermediate value for maize (0.069, 0.025 and 0.044 h^{-1} respectively, s.e.d.=0.0030). There was no effect of exercise on total cumulative gas production (Table 1). However, there was a trend ($P=0.073$) suggesting that exercised animals had a decreased rate of gas production compared to non-exercised animals (Table 1). This represented a proportional decrease in rate gas production rate of 0.104 for exercised versus non-exercised ponies. There were no exercise x feedstuff interactions.

Conclusion Despite there being no difference in total gas production of exercised and non-exercised horses the effect of sudden short term exercise tended to reduce the rate of gas production suggesting a possible decrease in feed digestibility.

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The effect of sugar-beet pulp on the nutritive value of ensiled lucerne for ponies

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Introduction Fibre-based diets help to maintain normal fermentation conditions in the large intestine of the horse; conversely concentrate diets high in starch can be detrimental to hindgut function (Mclean *et al*., 2000). Therefore, the ability to reduce the concentrate portion of the diet with a high-energy, fibre-based feed is desirable. Sugar beet pulp (SB) contains high levels of readily fermentable fibre and studies have reported dry matter (DM) digestibility values for SB in excess of 850 g kg^{-1} (Moore-Colyer and Longland, 2001; Hyslop, 2002). Furthermore, the degradability of the fibrous fraction of mature grass hay (Moore-Colyer and Longland, 2001) and high-temperature dried lucerne (Murray *et al*., 2002) fed to ponies has been enhanced by the inclusion of SB in the diet. Consequently, the aim of this study was to evaluate the effect of SB on the nutritive value of ensiled lucerne for ponies.

Materials and methods A 4 X 4 Latin square design experiment evaluated the effect of three levels of SB substitution on the nutritive value of ensiled lucerne for ponies. The silage was prepared from fourth cut lucerne (DM 270 g kg^{-1}) with an inoculant (Powerstart; Genus Plc, Nantwich, UK) applied at 10^6 CFU *Lactobacillus plantarum* g^{-1} FM. Four mature Welsh-cross pony geldings (280 kg LW s.e. 17.6) were fed each diet at 1.75% DM of bodyweight. The experiment consisted of 4 diets; control (WS; lucerne silage only, no SB) and three levels of SB substitution (100 [SB1], 200 [SB2] and 300 [SB3] g SB kg^{-1} DM. Each experimental period consisted of a 16 day adaptation phase and a five day recording phase when *in vivo* apparent digestibilities of DM (DMD), crude protein (CPD), acid detergent fibre (ADFD), neutral detergent fibre (NDFD) and non-starch polysaccharide (NSPD) were determined. Data were analysed using Latin Square ANOVA (Genstat 5, 2000). Values for SB as a sole feed were calculated by difference.

Results *In vivo* apparent digestibility values for each experimental diet and SB as a sole feed are shown in Tables 1 and 2, respectively. The substitution of ensiled lucerne with SB had no effect on DM, CP, ADF or NDF digestibility, but increased NSP digestibility. From Table 2 it appears that the substitution of lucerne with SB increased the digestibility of the lucerne cell wall fraction as the calculated NSP digestibility of SB was greater than unity.

Table 1: *In vivo* apparent digestibility of nutrients (g kg^{-1}) of the experimental diets

	WS	SB1	SB2	SB3	s.e.d.	Sig.
DMD	615	637	636	667	26.2	ns
CPD	757	780	756	787	24.4	ns
ADFD	466	482	463	490	44.0	ns
NDFD	490	517	521	551	39.4	ns
NSPD	550^{a}	618^{ab}	616^{ab}	692^{b}	37.6	P<0.001

Values in rows not sharing common superscripts differ significantly (P<0.05)

Table 2: *In vivo* apparent digestibility values (g kg^{-1}) for sugar-beet pulp calculated by difference

	SB1	SB20	SB30	s.e.d.	Sig.
DMD	857	794	828	99.4	ns
CPD	866	747	885	131.4	ns
ADFD	591	569	554	189.5	ns
NDFD	808	797	717	228.4	ns
NSPD	1139	954	1034	178.7	ns

Values in rows not sharing common superscripts differ significantly (P<0.05)

Conclusion It is clear that both ensiled lucerne and SB were well digested by ponies and may well be effective replacements for grass hay, which typically has a digestibility of between 0.3 to 0.4 in equids. Furthermore, it appears that the inclusion of SB enhanced the digestibility of the cell wall fraction of the lucerne.

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Assessing the structure of equine personality

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Introduction Recently there has been an increased interest in equine personality research. Previous studies have indicated the ability of human raters to assess equine personality (Morris *et al*., 2002). The aim of the present study was to assess reliability of personality assessment using a questionnaire rating method and to correlate personality data with recorded behaviour. Further development of the assessment method, and a greater understanding of equine personality may lead to applications in the equine industry, uses may include the implementation of equine personality during the selection of horses for particular equine disciplines.

Materials and methods The personality of 61 cross-bred horses (39 geldings and 22 mares) age range between three and 27 years (Mean 11.7 yrs) from four yards was assessed. A horse personality questionnaire (HPQ) was developed using 30 behaviourally defined adjectives adapted from the Stevenson-Hinde *et al.,* (1980) Behaviour-Rating Questionnaire. Adjectives were rated on a scale of one to seven: one represented no expression and seven represented extreme expression. Three handlers rated each horse. In addition, a total of two hours observation data were recorded for all 61 horses using continuous focal sampling between 11:00 and 15:00. Husbandry procedures were similar across yards, thus minimising the potential effects on the horses' behaviours. Data were analysed in three stages. Stage 1: Spearman rank order correlation coefficients and Kendall coefficients of concordance were used to assess agreement between raters for horses' scores over all adjectives and for each adjective across all horses. Stage 2: principal component analysis with a varimax rotation was applied to the HPQ data for the reliable horses. Stage 3: component scores for all horses rated reliably were correlated with behaviour variables from field observations using r_s coefficients.

Results Stage 1: Significant agreement was demonstrated for 73.13% of horses and 25 of the adjectives. Unreliable horses and traits were removed from further analyses. Stage 2: PCA extracted six components labelled "dominance", "anxiousness", "excitability", "protective", "sociability" and "inquisitive" and explained 79.3% of the total variance. Stage 3: Significant correlations between components and field behaviour were found as shown in Table 1.

Table 1 Correlation (r_s) between equine personality components and behavioural variables recorded during field observations

Personality component		Behaviour variables					
Component I Dominance		herding	Submissive	kicked	HTG	groom received	
	r_s	0.334*	-0.315*	-0.308*	0.453**	-0.301*	
Component II Anxiousness		passage					
	r_s	0.325*					
Component III Excitability		stand	self groom	nipped			
	r_s	-0.427**	-0.355*	-0.315*			
Component IV Protective		passage	play-fight	browse	greet		
	r_s	-0.361*	-0.363*	0.433**	0.412**		
Component V Sociability		canter	roll	biting	nipping	nipped	kicked
	r_s	0.376*	0.337*	0.437**	0.414**	0.312*	0.396**
Component VI Inquisitive		stand	exploration	graze	wind-suck	greet	groom receive
	r_s	0.313*	0.39**	-0.396**	-0.353*	0.35*	0.301*

HTG = Head threat given. * $P < 0.05$, ** $P < 0.01$

Conclusion This study demonstrated that equine personality can be rated reliably using an adaptation of the Stevenson-Hinde *et al.* (1980) methodology. Equine personality consists of six components that are correlated with behaviour variables from field studies. Further development of the HPQ may enable it to be implemented in the selection of horses for specific equine disciplines. This may reduce costs for trainers and improve horse welfare by avoiding the training of inappropriate horses.

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Some price determinants of sport horse foals at auction

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Introduction Rosen (1974) argued that goods could be treated as packages of characteristics and a comparison of market prices can be made by looking at the composition of the package of characteristics of the goods. Previous research on the variables affecting the price of equines sold at auction (predominantly yearlings) has focused mainly on the Thoroughbred sector. Variables found to be significant included month foaled (Buzby *et al*., 1994), sire progeny performance (O'Dochartaigh, 2004), racing performance of dams, sires and their progeny (Hastings, 1987) and the sex of the animal, (Robbins *et al.,* 2001). Taylor *et al*., (2004) studied show quality Quarter horses and indicated that sale order also affected the price achieved. Only limited research on the variables affecting price at auction has been conducted on the sport horse sector. While Thoroughbreds are typically purchased with racing, breeding, or both in mind, sport horses may be aimed at a much more diverse range of equestrianism. However much less information tends to be available to prospective purchasers with regard to pedigree and performance details of the animals. The purpose of this study was to identify the variables, which might act as salient predictors of sport horse foal prices at auction.

Materials and methods Results of all foals sold through Goresbridge sales complex in Ireland (1999 – 2004) were analysed (1,321 records). Data on each lot, including date of birth, sex, colour, dam, sire, breed of sire and price achieved were obtained from Goresbridge. A multiple regression was used to identify the relative effect of 13 variables on the price achieved.

Results In this study 10 variables were found to have a significant impact on foal price at auction (Table 1). When combined in a multiple regression model, the R square adjusted value for the model was 0.279 hence indicating that 28% of the variability in foal price can be explained by changes in these 10 variables. They including: breed of sire and the number of foals per sire registered that year, the position in the sale (day and portion of the day), the foal's sex, show performance and month born, the performance of its siblings, dam line and her premium status. Coat colour, registration status and the actual breed of the dam, were not significant.

Table 1 Stepwise multiple regression of predictors of price achieved by foals at sport horse auction

Variable	Foals Reg	Day of sale	Lot Pos	Sex	Dam line p	Breed of sire	Foal p	Sibling p	Pre mare	Mth born
R sq adj	.090	.143	.201	.228	.252	.263	.269	.273	.276	.279
B	-238.5	-482.4	-466.1	590.9	790.7	117.5	640.9	447.8	469.5	-165.9
St err of b	26.1	53.1	53.9	86.9	133.5	27.0	238.4	174.2	195.4	72.7
Beta	-.23	-.23	-.21	.167	.144	.105	.07	.06	.06	-.06
T	-9.13	-.91	-8.6	6.8	5.9	4.4	2.7	2.6	2.4	-2.3
Sign of t	.000	.000	.000	.000	.000	.000	.007	.010	.020	.023

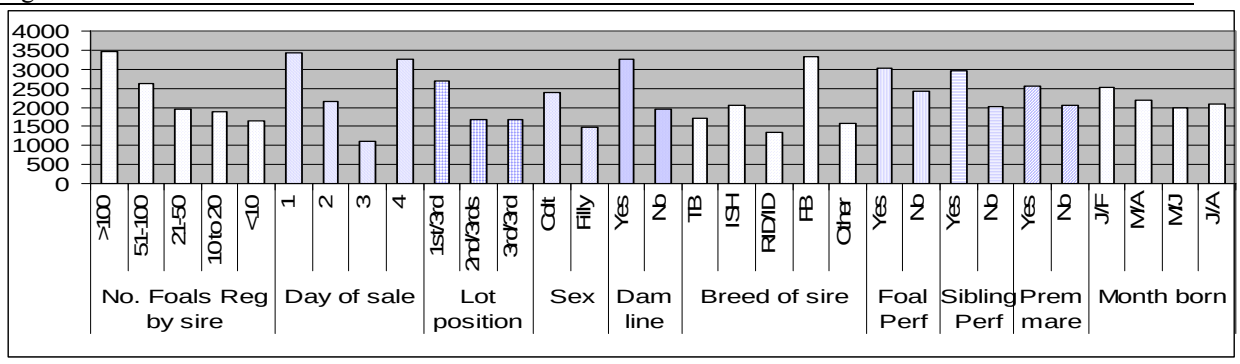

Figure 1 Price averages within price predictor categories

Conclusion. While male foals, with family performance history and by fashionable sires achieved the better prices, the day of sale and position (luck of the draw) were also very influential on the prices achieved by foals. However these variables only account for 28% of foal price, other of foal price variables would include static and dynamic conformation, genetic index of sire and these would warrant further investigation.

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The effect of location of event on scoring patterns at international CCI 4 star eventing competition (2000-2004)

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Introduction Various authors (Deuel and Russek-Cohen, 1995; Whitaker and Hill, 2005) have highlighted that the locations of competitions run at the same competitive level are exerting a non-random effect on scoring patterns within equine eventing competition. Consequently when mean scores are used as comparative measures between competitions some event locations are observed to result in significantly lower mean penalty scores than other event locations. This differentiation in scoring patterns between event locations may be useful from a sporting perspective. Experienced competitors are able to make informed decisions about the relative ease (when using potential penalty score as a measure) of an event location with regard to the development and experience of the horse they will be riding. A problem however is apparent when the subsequently collated competition data is used to make decisions regarding the relative breeding worth of an animal. Some animals may have been exposed to event locations where mean penalty scoring is lower. An inaccurate comparative analysis may therefore be made between the animals. This study investigates whether event location exerts a significant difference in mean penalty scoring patterns at a specific level of eventing - CCI 4 star.

Materials and methods Data were collated from two (Badminton and Burghley) CCI 4 star events over a five year period (2000-2004). In total nine competitions ran through this period (Badminton 2001 was cancelled due to Foot and Mouth). Data from horses completing competitions was used for analysis. In competition phased scores, dressage, cross country (time and jumping penalties) and show jumping (time and jumping) as well as final penalty score were analysed. Descriptive statistics and measures of dispersion were returned. Skewness was observed within the recorded phased scores, consequently data transformation Log (Score+1) (Langlois, 1975) was performed on phased and final penalty scores. Levenes tests for homogeneity of variance were performed, subsequently independent sample student *t* test were applied to the transformed data sets.

Results In total 442 completing horses were used within the analysis. Table one presents descriptive and dispersion statistics for phased penalty scores and final penalty score for the two event locations. All phased scores with the exception of show jumping time showed equality of variance. Additionally final penalty score showed equality of variance. Independent student *t* test highlighted significant differences in mean scores for the dressage phase (t= -3.927, df = 440, P>0.01), cross country (time) phase (t = 6.337, df = 440, P>0.05) and show jumping (time) phase (t = 114.827, df = 302.431, P>0.01) (unequal variances assumed). All other phases of competition and final penalty score displayed no significant differences between the two event locations.

Table 1 Comparative phased and final penalty scores

Phase	Event	Mean	Std. Err	Std. Dev	Min	Max	Skewness
Dressage	Badminton	56.181	0.781	10.710	30.8	106.6	0.504
	Burghley	59.261	0.691	10.040	36	91.2	0.231
X Country	Badminton	23.034	1.491	20.447	0	92	0.962
(time)	Burghley	20.990	1.275	18.518	0	92	1.134
X Country	Badminton	10.027	1.335	18.360	0	100	2.380
(jumping)	Burghley	9.076	1.234	17.930	0	100	2.436
S Jumping	Badminton	5.761	0.664	9.099	0	55	2.337
(time)	Burghley	1.976	0.292	4.245	0	29	3.327
S Jumping	Badminton	5.5	0.473	6.490	0	24	1.042
(Jumping)	Burghley	6.403	0.572	8.305	0	60	2.282
Final Score	Badminton	101.481	2.912	39.926	37.8	225.4	0.909
	Burghley	100.028	2.812	40.852	41.4	249.8	1.225

Conclusions This study demonstrates that event location is exerting a significant effect on scoring patterns within specific phases of competition at CCI 4 star level. The effect is however limited to the dressage and the timed part of the show jumping and cross country phases. The cross country jumping phase is not shown to be subjected to a significant event location effect. Whitaker and Hill (in press) have previously demonstrated this phase to exert the greatest leverage effect over final finishing position at varying levels of eventing competition. Event location needs to be carefully considered when making comparable judgements between certain phase scoring within competition. This is particularly relevant when making judgements as to the potential true worth of an animal to the population. However event location effect does not appear to be significant in determining overall finishing score.

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Analysis of Young Horse Evaluation data for use in the genetic evaluation of sport horses

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Introduction In order to keep on a par with other European horse breeding countries, Britain is now aiming to develop genetic evaluations for British bred sport horses. Data has been collected from the Young Horse Evaluation Series (YHE). The traits measured include a veterinary assessment, an assessment of conformation and assessment for specific traits involved in showjumping (SJ), dressage (D) and eventing (E). For E, paces, loose jumping and ridden jumping are assessed, for SJ only the two jumping traits are assessed and for D only paces are assessed. The qualities looked for in the E traits differ from those measured for SJ and D. A total score is also given for each discipline, which combines the scores from performance with the veterinary and conformation scores. Each trait is scored on a scale of 0-10, in which 0 equates to poor and 10 to excellent. A preliminary investigation was carried out to determine the heritability of each of the traits.

Material and methods The dataset consisted of 294 records for 248 individual horses. The animals were categorised into 3 sexes (gelding, mare or stallion). The records were collected on 30 different evaluation dates. Pedigree information was available for these data with sires (187), dams (235) and dam's sires (191) mainly made up of half sib families. The data was analysed with an animal model using ASReml (Gilmour *et al.,* 2002).

$$y_{ijk} = \mu + \alpha_i + \beta_j + u_{ik} + w_{ik} + e_{ijk}$$

Where y_{ijk} is the trait value for the *k*th animal of sex *i* evaluated on the *j*th date, μ is the overall mean, α_i is the effect of sex, β_j is the effect of the date of evaluation, u_{ik} is the genetic effect of animal *ik,* w_{ik} is the permanent environmental effect of animal *ik* and e_{ijk} is the residual error. u_{ik} were assumed to be normally distributed with a variance/covariance matrix of the form σ^2_a A where A is the relationship matrix, w_{ik} was assumed to be normally distributed with a variance/covariance matrix of the form σ^2_c I and e_{ijk} was assumed to be normally distributed with a variance/covariance matrix of the form σ^2_e I.

Results For the scoring of veterinary, conformation, SJ total, D paces and D total there were significant differences ($p<0.05$) among the sexes with a general pattern of stallions having the highest mean scores and mares having the lowest mean scores. Mean scores for veterinary, conformation, D Paces, D total, E paces and E total differed significantly ($p<0.05$) over the dates of evaluation. Table 1 shows the traits for which the repeatability was significantly different from zero. With the exception of SJ Loose ($h^2 = 0.85 \pm 0.09$) the heritabilities for these traits are not significantly different from zero.

Table 1 Repeatabilities and Heritabilities for YHE Traits

Trait	No. Observations	Phenotypic Variance	Heritability±SE	Repeatability±SE
Veterinary	293	1.060	0.00 (—)	0.56 ± 0.11
Conformation	291	0.609	0.33 ± 0.38	0.66 ± 0.09
SJ Loose	94	1.956	0.85 ± 0.09	0.85 ± 0.09
SJ Total	127	0.729	0.01 ± 1.22	0.75 ± 0.13
D Total	171	0.569	0.05 ± 0.89	0.68 ± 0.12
E Paces	158	0.620	0.36 ± 0.72	0.60 ± 0.14
E Total	159	0.482	0.48 ± 0.69	0.57 ± 0.15

Conclusions The higher scores given to stallions may be explained by pre selection. A stallion may be expected to have sound health and good conformation, because any young male showing slight conformational faults or signs of poor health is likely to be gelded. The significance of the date of evaluation for veterinary, conformation, D paces, D total, E paces and E total may be due to the use of different judges on each evaluation date, establishing differing standards. The small dataset used for this study means that the heritabilities shown have large standard errors, although the large heritability for SJ loose is intriguing. It is plausible that SJ loose has a moderate to high heritability since it is less affected by environmental influences such as training or rider ability, compared to the other traits. This would be consistent with other published information (Olsson *et al.* ,2000). The repeatabilities, given in Table 1, do not exclude the possibility that the heritabilities for the traits given are sufficient in magnitude for developing useful evaluations, since repeatabilities form an upper bound to the heritability.

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Physiology of heat stress in Merino wethers during conditions similar to live export

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Introduction Heat stress of sheep in feedlot systems and within the live export industry is an animal welfare concern and a financial concern due to loss of life and reduced production (Richards *et al.* 1989). Early research has been undertaken in short term studies exposing Merino sheep to excessive heat load (Lee 1950; Ames *et al*. 1971). However, little is known of the physiological changes in animals subject to prolonged periods of excessive heat and humidity as can occur in the live export industry. Knowledge of the physiological responses of Merino sheep to prolonged heat and humidity is needed to correctly manage these sheep during shipment. This experiment investigated the physiological responses of Merino sheep to high heat and humidity similar to that experienced during long haul live export voyages to the Middle-East.

Materials and methods This experiment was designed to investigate the effects of prolonged heat and humidity as can occur on live export vessels. Two climate controlled rooms were used in the experiment, and each housed six Merino wethers. Sheep were held in the rooms for a period of 17 days. During this time they were exposed to two periods of sustained high heat insults extending from days 6 to 8 and days 12 to 14, with a maximum wet bulb temperature of 31.2˚C being achieved during the heat periods. While in the rooms sheep were fed standard shipper pellets that they had previously been adapted to. Water was available *ad libitum*. Measurements were taken of physiological parameters (Table 1).

Results Mean physiological parameters for the Merino wethers are shown in Table 1. The significant rises in core and respiratory rate indicate that Merino wethers were heat stressed. The increase in respiratory rates led to changes in blood gas values, consistent with a compensated respiratory alkalosis. However sheep were able to recover very quickly and by day 17 respiratory rates, blood gas values and urine pH were not different from the beginning of the study (day 2).

Table 1 Means ± SEM for physiological parameters measured on Merino wethers in climate rooms prior to heat insult on day 2 (Mean wet bulb room temperature of 20˚C) compared to during heat insult on day 14 (Mean wet bulb room temperature of 31˚C) and following the heat insults on day 17 (Mean wet bulb room temperature of 16˚C)[#]

Variables	Prior to heat insults	During heat insults	Following heat insults
Feed intake (% body weight)	$1.5^{a} \pm 0.28$	$2.0^{b} \pm 0.13$	$2.1^{b} \pm 0.07$
Water Intake (% bodyweight)	$3.3^{a} \pm 0.5$	$6.2^{b} \pm 0.90$	$5.8^{b} \pm 0.18$
Core Temperature (˚C)	$39.15^{a} \pm 0.12$	$40.7^{b} \pm 0.08$	$38.79^{c} \pm 0.06$
Respiratory rate (breaths/ minute)	$57.4^{a} \pm 8.07$	$179^{b} \pm 11.96$	$48.5^{a} \pm 4.29$
Venous pH	$7.39^{a} \pm 0.02$	$7.46^{b} \pm 0.015$	$7.42^{ab} \pm 0.02$
Venous pCO_2 (mm Hg)	$48.08^{a} \pm 1.99$	$31.42^{b} \pm 1.78$	$46.25^{a} \pm 1.91$
Venous HCO_3 (mmol/l)	$29.6^{a} \pm 0.31$	$21.25^{b} \pm 0.70$	$28.7^{a} \pm 0.37$
Urine pH	$7.05^{a} \pm 0.55$	$8.32^{b} \pm 0.09$	$7.69^{a} \pm 0.104$

[#]Within rows, means with different subscripts differ significantly ($P<0.05$)

Conclusions It appears from this study that sheep are able to quickly recover from the heat stress associated with live shipping, once ambient temperature is again within the animal's thermoneutral zone. The ability of sheep to return physiological values back to normal quickly following the heat insult could be explained by the sheep continuing to eat throughout the study. However, other stressors involved in live shipping may have a deleterious effect on the ability of the sheep to physiologically cope. Further studies will look in other stressors on sheep during live shipping.

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Socio-economic viability of keeping goats on a share basis by resource poor households in Nepal

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Introduction Keeping goats can make an important contribution to the livelihoods of small and landless farmers in Nepal, one of the world's poorest countries. However, the capital cost of buying a goat can be prohibitive for very poor households. An alternative entry point is to keep the goats on a share basis, with a richer neighbour providing a goat or goats to a poorer neighbour to keep. The practice is governed by a complex set of rules and norms which are not easy to set out categorically, but the principle seems to be that equal shares go to owner and keeper. For example, when does that are kept in this way kid, one of the kids is generally given to the goat raiser, while the doe and her other kids may be returned to the goat owner after weaning. The price raised by selling shared goats is generally split equally between the owner and the goat's keeper, after deducting the initial price of the goat if it was bought in by the owner. The objective of this study was to estimate the prevalence of this practice in the southern region of Nepal, and to estimate the contribution that share-keeping goats may make to the livelihoods of households that were unable to buy a goat for themselves.

Materials and methods Twenty households from four different communities (Jamunibas, Kemalipur, Baluwa Bhiman and Birendra Bazaar) in the Dhanusha district on the Gangetic plain of southern Nepal were involved. Each household was visited at fortnightly intervals for one year by a local facilitator and an individual record card maintained for each goat kept by the household was updated at these visits. Information regarding the goat's age, how long it had been in the herd, how it entered the herd and when and how it left the herd (if it did) was collected. In addition, information on the amount and sources of the household's cash income since the last visit was also gathered. A semi structured interview was also conducted with those who share-kept other people's goats in Kemalipur after the study had ended. The nature of the data is such that only qualitative assessments can be made, but these may be useful in indicating the contribution that share raising goats may make to the livelihoods of very resource poor farmers in this area.

Results Records were maintained on 348 goats, 222 of which entered the herds during the study. 51 (15%) of the goats recorded were being share-kept by 25 (32%) of the households involved in the study. In most of the communities, between three and five of the 20 monitored households were share-keeping goats, but in Kemalipur this practice was much more common involving 13 (65%) of the households although this community reported that within a whole community approximately 20% of goat-keepers would be share-keepers. Two of the 25 households involved in shared goat-keeping managed a single shared goat and kept no others, but the usual practice was to manage on average 2.0 shared goats, 3.2 goats that had been born into the herd and 0.36 goats that had been bought. All of the goats that were being used in shared goat-keeping were does, with a mean parity of 0.45. The mean age of entering the herd was 6.6 months and when recording began, their mean duration in the share-keeper's herd was 5.3 months, so that their mean age at the start of the study was 11.9 months. Of the 51 goats that were share-kept, 27 were in the herds at the beginning of the study, and 12 of these 27 does kidded during the study producing a total of 22 kids. The number of goats entering herds on a shared basis during the year was 24 (47% of the total number of share-kept goats). A total of 14 (27%) of share-kept goats left the herds during the year. Most of these goats (11) were returned to their owner (at a mean age of 10.5 months). One died (at six months old) and another two were sold (at four months old) but prior to the sale they had not been sick suggesting the sales were to raise cash rather than to realise some money from goats that would otherwise die.

Shared goat-keepers in Kemalipur estimated that it took 3 h/d to manage five goats, at a cost (for feed) of Rs 1800 pa. They estimated that their income from managing these goats would be Rs 17 000 (Rs 8000 from the sale of finished male goats and Rs 9000 from the sale of kids), yielding a gross margin of Rs 15200 without taking into account the cost of labour. This is equivalent to 58% of the mean annual income of these communities and one of the women interviewed, who was herself landless, pointed out that this return was greater than they could manage from farming 0.67 ha of land (the typical size of landholding in that community).

Conclusions Managing goats on a shared basis is clearly a viable means of starting to keep goats and can make a substantial contribution to the livelihood of the household involved. There is a high turnover of goats between the managing household and the goats' owner but the practice quickly results in the share-keeping household owning goats as they generally keep one of every two kids that are born to a share-kept doe. The prevalence of the practice appeared to vary between the four communities, with the village composed mostly of high caste Brahmins and Chhetris being the one with the greatest proportion of share-keepers. This might point to a greater willingness for high-caste goat owners to enter sharing agreements with those of similar caste. Thus share-keeping may be a means of enabling resource-poor but high-caste households to improve their livelihood.

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Effect of concentrate supplementation and inclusion of condensed tannins on lamb performance and faecal egg counts

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Introduction Increasing resistance to anthelmintic drugs (Waller, 2003) has increased the need for the development of alternative methods of worm control. The antiparasitic properties of condensed tannins have been evaluated in small scale experimental trials (Athanasiadou *et al*. 2000) with positive effects obtained under restricted feeding regimes. However, Dawson *et al*. (2004) did not observe significant effects on strongyle faecal egg counts through the inclusion of 80 g tannins/kg fresh concentrate offered to lambs at grass in the period from weaning to slaughter. The aim of the current study was therefore to evaluate the effect of increased levels of tannin inclusion offered to lambs from six weeks of age on grass-based systems on faecal egg counts.

Material and methods In April 2004, ewes and lambs (4-6 weeks of age) were given a benzimidazole anthelmintic. The lambs were allocated to one of four creep treatments, balanced for lamb breed, sex and numbers of singles/twins/triplets (32 ewes, 47 lambs per treatment), as follows: (1) zero concentrate (NC), (2) concentrate, zero tannins (CT0); (3) concentrate + 80 g tannin/kg fresh (CT1) and (4) concentrate + 100 g tannin/kg fresh (CT2). The concentrate contained 120 g crude protein (CP)/kg fresh and was offered *ad libitum.* The ewes and lambs grazed a perennial ryegrass-based pasture which was divided into four areas. Following weaning, lambs remained in the treatment groups with grazing areas adjusted accordingly. Lambs were weighed fortnightly and live weight gains were determined by linear regression. Individual faecal samples (n=5 per treatment) were collected from each treatment group at the start of the study, prior to dosing and faecal egg counts determined. Faecal egg counts were then used to determine whether subsequent doses were required with faecal samples taken weekly. Faecal egg counts were performed on all samples using the McMaster technique (Urquhart *et al*. 1996). Lamb performance and faecal egg count data were analysed using the Genstat REML procedure. Lamb sex, birth weight, birth type, lamb breed and age were used as covariates for the performance data. Faecal egg counts were log(x+1) transformed prior to analysis, analysed using the Genstat REML repeated measures analysis and the results backtransformed and 95% confidence intervals calculated.

Results Total concentrate consumption throughout the trial was 108 kg (CT0), 89 kg (CT1) and 65 kg (CT2). The lower intake of CT2 treatment was attributed to reduced palatability with the highest level of tannin inclusion. Relative to the zero concentrate treatment, concentrate supplementation increased lamb performance (live weight and live weight gain) in the period from birth to weaning and from weaning to slaughter ($P<0.001$). Increasing level of tannin inclusion had no significant effect on lamb performance in the period from birth to weaning. However lambs offered concentrate containing 100 g tannin/kg fresh had lower live weights at the end of the study ($P<0.05$) and lower live weight gains from weaning to the end of the study ($P<0.001$) relative to lambs offered concentrate containing either zero tannins or 80 g tannin/kg fresh. Increasing level of tannin inclusion had no significant effect on faecal egg counts.

Table 1 Effect of concentrate supplementation and level of tannin inclusion on lamb performance and faecal egg counts

	Zero	Concentrates			sem	sig
Level of tannin inclusion (g/kg fresh)	concentrates	0	50	80		
Live weight at weaning (kg)	35.6^{a}	38.7^{b}	39.4^{b}	37.8^{b}	0.71	***
Live weight gain (birth to weaning g/day)	290^{a}	319^{b}	328^{b}	313^{b}	7.0	***
Live weight at end of study (kg)	41.0^{a}	47.6^{c}	49.2^{c}	45.1^{b}	0.80	***
Live weight gain (weaning to end of study g/day)	180^{a}	304^{c}	312^{c}	230^{b}	13.4	***
Strongyle faecal egg counts (epg)	80.4	20.6	37.7	38.8		NS
(95% confidence intervals)	(34, 186)	(8, 50)	(16, 87)	(30, 160)		

Means within rows with same superscripts are not significantly different ($P>0.05$).

Conclusions Inclusion of condensed tannins in concentrates at levels of up to 100 g/kg fresh and offered to lambs at grass from six weeks of age did not significantly affect faecal egg counts. The results of this work indicate that under commercial conditions tannins have a limited role in worm control.

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Effect of sainfoin (*Onobrychis viciifolia*) hay on parasitism and productivity of lambs infected with *Trichostrongylus colubriformis*

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Introduction Sainfoin is a leguminous, highly nutritive plant with bloat free characteristics, suitable for growing lambs. In addition, sainfoin has been tested for its anthelmintic properties in goats and sheep, because of its high content of plant secondary metabolites (PSM), specifically condensed tannins. However, Athanasiadou *et al.* (2005) compared the effect of sainfoin *vs* lucerne on sheep and the animals showed poor performance, manifested as a lower intake and liveweight gain. But on the other hand, the intake of sainfoin resulted in lower daily parasite egg output. The hypotheses of the present study were: (1) The consumption of sainfoin hay in lambs continuously infected with *T. colubriformis* will result in negative effects on the parasites, measured through levels of faecal egg counts (FEC). (2) The parasite infection will affect grass hay intake more than sainfoin hay intake, because the PSM content will allow animals to cope better with the parasitic infection. (3) The use of an adaptation period to sainfoin hay prior to infection is expected to improve the parasite control of growing lambs continuously infected with *T. colubriformis*, through the establishment of a hostile environment for the incoming larvae and also will ameliorate the possible negative effects of PSM contained in sainfoin on the performance of the sheep.

Materials and methods Forty-eight males and females weaned lambs (initial weight 34.5±0.124 kg), Greyface crosses, were randomly allocated to 6 experimental treatments. The lambs received *ad libitum* either sainfoin (14% crude protein-CP) or grass (9.3% CP) hays as follows: group G and S were fed grass and sainfoin hay, respectively for 8 weeks. A third group GS was fed grass hay for 2 weeks and sainfoin thereafter. Comparisons between groups S and GS aimed to provide evidence on the effect of an adaptation period on sainfoin on parasitism. Half of the sheep within each group were parasitised (+) and half were not (-). The parasite challenge was performed from day 15, using a trickle infection with 12,000 infective larvae (L_3) per week of *T. colubriformis*. The FEC and the weighing of the animals were performed weekly and food intake was measured daily. Dry matter (DM) concentrations of the hays offered and the refusals from each animal were also measured. All the measurements were analysed as a two-way ANOVA for repeated measurements, with two levels of parasite infection (+ and -) and three different forage treatments (G, S and GS).

Results Animals consuming grass hay had significantly higher FEC than those consuming sainfoin ($p=0.021$) from week 5 to week 8 (Fig. 1). There was no evidence of an effect of the adaptation period on sainfoin on the FEC. The intakes were significantly affected by forage treatment, with means of 820, 1211 and 1018 g DM/day for G, S and GS groups, respectively. Intake was also significantly affected by the interactions between time x forage treatment and time x infection, with infected sheep having a reduced food intake during the latter stages of the experiment. GS groups (without adaptation period to sainfoin) increased their DM intake to that of S animals when they were changed from grass to sainfoin hay. Live weight gains from the day of the infection were low and were affected by both factors, the forage treatment ($p=0.005$) and the parasite infection ($p<0.001$), with means being 40.6, 94.0 and 52.6 g/day for G, S and GS, respectively.

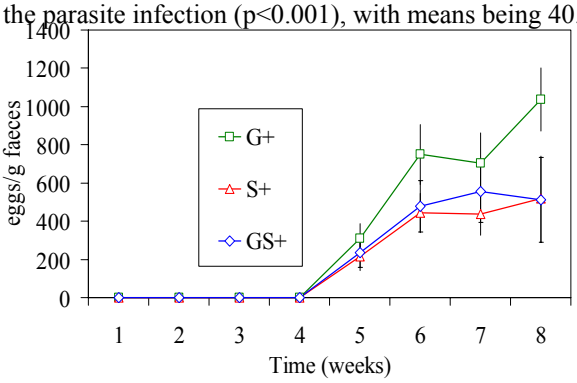

Figure 1 Backtransformed FEC of the parasitised animals, with 95% confidence intervals

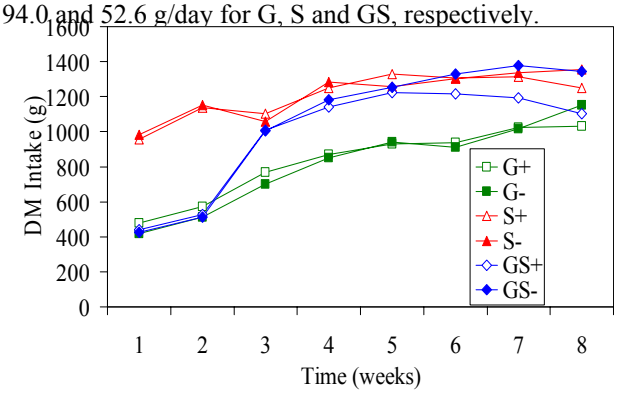

Figure 2 DM Intakes of the animals by treatments

Conclusion Sainfoin intake had an effect on FEC. The results confirm the potential of sainfoin as antiparasitic forage. The intake of sainfoin hay was always higher than the grass hay, but these advantages were not manifested with improvements in the performance of the parasitized animals offered sainfoin. The adaptation period had no clear effect on FEC and also did not improve performance on sainfoin hay. Future studies should focus on how sainfoin could be offered to parasitised sheep to achieve, in addition to the parasite control, an improved animal performance.

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Effects of rumen protected choline chloride on milk yield , milk composition and blood metabolites in dairy Sannan goats

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Introduction Choline is a vitamin like compound that functions in several metabolic ways, mainly as phospholipids. It plays an important role in cell membrane integrity and is involved in lipid digestion and transport (Bindle *et al*., 2000). Much of the choline that reaches the intestines of ruminants is phosphatidil choline contained in protozoa (Broad and Dawson, 1976). Feeding cattle with high concentrate diets cause to decrease in protozoa numbers by decreasing ruminal pH (Towne *et al.,* 1990). Therefore high concentration diets limit the choline supply to the intestinal of cattle. In ruminant diets supplemental choline similar to other compounds of diets affected by rumen micro organism. So if we protect it from these micro organisms, we will protect its vitamin properties. The main objective of this study was to determine the effects of rumen protected choline chloride (RPC) on milk yield, milk composition, and blood metabolites in dairy Sannan goats.

Material and methods Three dairy Sannan goats (DIM = 50 ± 13), arranged in 3×3 Latin square design, were fed a control diet (no supplemental RPC) and diets containing 2, 3 g/d RPC. The diets consisted of 40 % alfalfa and 60 % concentrate. Diets were isonitrogenouse and isocaloric. Each period of the Latin square design consisted of 15 days; 10 days were for adaptation and 5 days were for data collection. Diets were fed twice daily to allow for *ad libitum* intake and orts were weighted once daily at last 5 d of each period. RPC for each treatment was added to morning diet as top dressed. Goats were milked twice a day during each period. Milk yield was measured daily during days 11 to 15 and its samples were taken to analyze its composition. Rumen fluid samples were taken at 2 h post feeding on day 15 of each period via a stomach tube. Blood samples were analyzed for serum TG and cholesterol concentration. Data were analyzed using the general linear model procedure of SAS 2000.Differences were considered significant when $p < 0.05$.

Results The results of this study are shown in Table 1. In this study average dry matter intake of goats was not affected by RPC. Milk production and its composition except solids not fat (SNF) were not affected by RPC. SNF (%) was decreased when dairy goats were fed 2 g/d RPC. RPC had significant effect on serum triglyceride but serum cholesterol concentration didn't affected by RPC. Serum triglyceride increased by supplemental RPC.

Table 1 Effects of rumen protected choline chloride on milk yield , milk composition and blood metabolites in dairy Sannan goats

	RPC level, % of dietary DM			
Item	0	.037	.075	SEM
DMI, g/d	2550	2700	2720	35.21
Milk, g/d	2761	2815	2863	41.43
Milk fat, %	3.30	3.35	3.10	.05
Milk protein,%	2.70	2.78	2.70	.03
Milk lactose,%	4.10	4.10	4.00	
SNF %	7.90^{ab}	8.05^{a}	7.80^{b}	.07
Plasma, mg/dl				
Triglycerides	$.024^{a}$	$.070^{b}$	$.036^{a}$	.001
cholesterol	1.66	1.45	1.94	.04

Differences were considered significant when $p < 0.05$.

Conclusion The results of this study indicated that supplemental RPC has no significant effect on DMI, milk production and its constituents except SNF. The comparison of treatment's SNF means showed that there were significant differences between them. It indicated that 1 g/d RPC was better than other levels. DMI and milk production increased insignificantly when RPC was enhanced in diets. Blood serum analysis indicated that triglycerides were increased by increasing diet RPC that it may be due to attribution of chorine chloride in blood lipoproteins structure. At the end we can say that RPC can improve milk production efficiency in dairy Sannan goat.

Acknowledgements The authors wish to acknowledge for funding and technical supporting from Ferdowsi University of Mashhad and Centre of Excellence for Animal Science

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Effects of adding a 'cocktail' of copper antagonists, molybdenum, sulphur and zinc, on liver copper accumulation in Texel rams given a commercial concentrate

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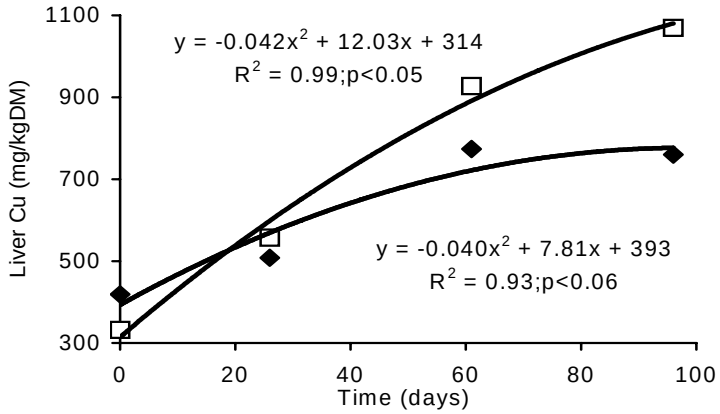
Introduction There is a risk of chronic copper poisoning (CCP) in breeds such as the Texel whenever they are liberally fed pelleted commercial rations. Addition of the Cu- antagonists iron (Fe), zinc (Zn) or a combination of molybdenum and sulphur (Mo + S) to the diet have each significantly reduced liver Cu and risk of CCP in separate experiments but usually in less susceptible breeds. 'Cocktails' of antagonists are routinely added to compound feeds to reduce the risk of CCP but components and inclusion rates vary and the additivity of their protective effects has not been studied. Furthermore, the apparent inability of Texels to limit hepatic Cu retention (HCR) has called into question the reliability of antagonist cocktails for this breed (Suttle *et al* 2002). The objective of this experiment was to test the ability of a cocktail of Mo, S and Zn to restrict HCR by Texel rams, given a pelleted commercial concentrate that raised Cu in the total ration to the EC maximum permitted level of 17mg/kgDM.

Materials and methods Two groups of five, 1-2 year old Texel rams, weighing 40-60kg, were individually penned and offered 1.4kg/d of pelleted concentrate and 0.3kg/d of chopped hay after an adjustment period of 27days: the trial ended after 96days. Feed refusals were recorded but rarely occurred. Individuals were ranked according to initial liver Cu concentrations in biopsy samples and allocated randomly, within strata, to concentrate with (A) or without (0) added Mo (2, as molybdate), S (3000, as 'flowers of S') and Zn (300mg/kgDM, as sulphate). Concentrates O and A contained, respectively (mg/kgDM):Cu, 25.6 and 22.5; Fe, 395 and 405; Zn, 181 and 452; Mo, 0.3 and 2.3: S, 2000 and 5000 (the values for Mo and S were estimated). Plasma was obtained at weekly intervals for analysis of Cu concentration and gamma glutamyl transferase activity, an indicator of liver damage. Further liver biopsy samples were obtained after 26, 61 and 96 days to assess rates of accretion of Cu. Initial and final liveweights were recorded and liver sizes estimated as 5.15g liver DM /kgLW (Suttle *et al*, 2002), allowing HCR to be calculated. Means are given with their s.e. and were compared by Students 't' test, using pairing or log transformation where appropriate.

Results The mean liver Cu concentrations in Group 0 (□) and Group A (♦) rams were fitted better by curvilinear than by linear relationships with time, for which R^2 were 0.93 and 0.86, respectively (Fig 1).

Fig 1 Effect of antagonists on liver Cu accretion



The 'cocktail' of Zn+Mo+S did not prevent liver Cu accretion but it reduced the linear rate of increase by about 1/3rd to give a significantly lower ($p<0.05$, paired test) final liver Cu concentration (760± 63 v 1069±123 mg/kg DM). Mean HCR (% of total Cu intake from concentrate) was reduced from 8.0± 0.88 in Group 0 to 4.3± 0.44 in Group A ($p<0.01$), despite the high level of a fourth antagonist, Fe, in both diets. There were weak signs of adaptation to Cu overload in terms of curvature in the plot for each group. Supplementation did not prevent significant increases in GGT during the trial from initial values on the verge of normality (40 and 45± 2.6U/L) but the final geometric mean for Group A (68.5) was lower than that for Group 0 (114;$p<0.05$).

Conclusions A cocktail of the three antagonists, Mo, S and Zn, reduced but did not eliminate the risk of subclinical copper poisoning in Texel rams given a commercial concentrate.

Acknowledgement
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Effects of dietary protein content and the amino acid profile of metabolisable protein on voluntary intake and milk production of dairy cows fed maize silage-based diets

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Introduction The productive response of dairy cows to relatively high protein diets could be mediated, amongst other things, by provision of essential amino acids. Therefore, it may be possible to reduce the level of dietary protein (main source of N pollution) if the amino acid (AA) profile of metabolisable protein (MP) is considered in formulating diets. The objective of this study was to investigate the effects of both dietary crude protein (CP) content and AA profile of MP on productive responses of dairy cows fed maize silage-based diets.

Materials and methods Sixteen multiparous Holstein cows averaging, at the commencement of the experiment, 619 kg body weight (s.d. 41.6), 134 days in milk (s.d. 45.2) and 38 kg milk yield per day (s.d. 5.5) were used in a cyclical changeover design experiment (four 3-week periods) with eight dietary treatments. Diets contained (dry matter, DM, basis) 450 g/kg maize silage, 50 g/kg wheat straw and 500 g/kg concentrate and were offered as a complete feed for *ad libitum* intake at 0830h and 1630h. Diets were formulated to be isoenergetic and to differ in the dietary CP content (140 g/kg DM and 160 g/kg DM) and in the AA profile of MP achieved by altering the composition of four concentrates (based on soybean meal, SBM, or maize by-products, MBP). The AA profile of MP was also manipulated by adding a mixture of rumen protected lysine (40 g/cow/day; Sil Lysine 50% bypass, microencapsulated SILA, SRL, Italy) and methionine (14 g/cow/day; Smartamine^{TM} M; Adisseo, Alpharetta, GA, USA) on the top of the feed at the time of the morning feeding and mixed in by hand. Jugular blood samples were collected three hours after the morning feeding on one day of the last week of each experimental period and analysed for urea (Bauer, 1982). Data from the last seven days of each experimental period were analysed as a cyclical changeover design using SAS Proc mixed Version 8e (1999-2001). The model included the fixed effects period, dietary CP content, main protein source in the concentrate, addition of rumen protected lysine and methionine (RPLM) and the interactions between the dietary effects, the random effect of cow, and the random residual error. Days in milk were used as covariate. When the interactions were not significant, they were removed from the model.

Results The main effects of dietary treatments on feed intake, milk yield and composition, efficiency of conversion of dietary N into milk N and plasma urea are given in Table 1. Diets with lower dietary CP content significantly promoted lower feed intake, milk yield and plasma urea, and higher efficiency of conversion of dietary N into milk N. Diets based on maize by-products in the concentrate significantly increased milk yield, and decreased milk fat and protein contents. The addition of RPLM only tended to increase milk fat and protein contents.

Table 1 Effects of dietary crude protein (CP; g/kg DM) content, main protein source in the concentrates (soybean meal, SBM, or maize by-products, MBP) and addition of rumen protected lysine and methionine (RPLM) on dry matter intake (DMI), milk yield and composition, efficiency of conversion of dietary-N into milk-N (MP/CPI) and plasma urea.

	CP		Protein source		RPLM		s.e.m.	P		
	140	160	SBM	MBP	-	+		CP	Protein source	RPLM
DMI (kg/day)	21.3	22.4	21.6	22.0	21.9	21.7	0.57	<0.001	<0.10	NS
Milk										
Yield (kg/day)	34.6	35.8	34.5	35.9	35.2	35.2	1.05	<0.01	<0.01	NS
Fat (g/kg)	37.9	38.8	39.1	37.6	37.6	39.0	1.23	NS	<0.05	<0.10
Protein (g/kg)	29.8	29.8	30.2	29.4	29.6	30.0	0.54	NS	<0.01	<0.10
MP/CPI	0.34	0.30	0.32	0.32	0.32	0.32	0.007	<0.001	NS	NS
Urea (mg/dl)	21.8	31.3	27.7	25.4	26.3	26.8	0.90	<0.001	<0.01	NS

Conclusions The results show that a decrease in dietary CP content from 160 g/kg DM to 140 g/kg DM negatively affected the productive response of mid-lactation dairy cows fed maize silage-based diets, increasing the efficiency of dietary N use. Manipulating the AA profile by altering the main protein source in the concentrates, significantly affected milk responses, with maize by-products increasing milk yield and decreasing milk fat and protein contents. The RPLM supplementation tended to increase milk fat and protein contents.

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Effect of source and level of selenium supplementation in the diets of lactating dairy cows on the selenium content of milk and cheese

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Introduction Levels of Se in plant, animal and human food reflect the considerable variations that occur in soil Se throughout the world. In 12 out of 21 countries selenium intake in humans has been found to be below WHO recommended levels (40 male, 30 female ug/h/d). In humans low selenium (Se) intake has been linked to a higher risk of CVD, arthritis, infertility, immunosuppresion, adverse mood states and cancer. As a consequence, the concept of healthy food additives, with the term ”functional foods” has been defined as a food that provides benefits beyond the traditional nutrients it contains. In this case selenium. The purpose of this work was to compare the effect of the source and level of selenium supplementation in the diets of lactating dairy cows on the selenium content of milk and cheese.

Materials and methods A total of 12 multiparous lactating dairy cows were selected at random and assigned to one of three treatments at 40 d pp according to calving date, milk yield, milk composition and somatic cell count (SSC) for a total of 120 days. The cows (n3 per treatment) were offered either 6mgSe/d from sodium selenite (ST), 6mgSe/d of selenium enriched yeast (SeY) (HS) or 3mgSe/d SeY (LS) from Sel-Plex™ offered in 2 kg of dairy compound. The cows were all grazed at pasture and were offered 7kg/h/d of a commercial compound (18 % CP) and *ad libitum* access to Spring grass. Milk samples were collected weekly from each individual cow and were used to manufacture whole milk and unripened soft cheese. Individual milk samples from each cow were collected weekly and the milk samples, whole processed milk and cheese were analysed for Se content. Normally distributed data was analyzed using Mintab (13) GLM ANCOVA using selenium supplementation as a fixed effect.

Table 1 Mean milk, fat and protein yield from cows offered 6 mg sodium selenite (ST), 6 mg (HS) or 3 mg (LS) of Se in the form of SeY/h/d

	ST	HS	LS	SEM
Fat corrected yield	32.0	28.1	29.0	1.43
Milk fat (g/kg)	35.1	32.5	32.0	1.63
Milk protein (g/kg)	31.1 a	30.7 a	29.3 b	0.3
Total milk fat (kg/h/d)	1.27	1.11	1.16	0.057
Total milk protein (kg/h/d)	1.13 a	1.05 b	1.06 b	0.015
Mean milk Se levels(μg/l)	14.7 b	21.6 a	15.5 b	0.65
Total milk Se yield (μg/h/d)	470.4 b	775.6 a	449.5 b	0.93
Mean cheese Se levels (μg/kg)	100 b	160 a	100 b	0.2
Milk per kg cheese produced (kg)	8.4 a	7.3 b	8.1 a	0.21

Results In Table 1 the milk yield and composition are presented. There were no significant differences in fat corrected milk yield, milk fat content, or total milk fat yields between cows offered differing sources and levels of selenium. There was a significantly higher milk protein content and total milk protein yield in the milk from cows offered 6 mg/h/d of sodium in the form of sodium selenite and SeY. Milk from cows offered high level of selenium (6 mg/h/d) from SeY had a greater cheese yield per kg of milk and also had a tendency to have higher selenium content in the cheese. Milk selenium content and total milk selenium yield were significantly higher in milk from cows offered 6 mg/h/d of selenium in the form of SeY compared with cows offered 6mg/h/d of sodium selenite or 3 mg/h/d of selenium in the form of SeY. There was no significant difference in milk selenium content between cows offered either 6mg/h/d of sodium selenite or 3 mg/h/d of selenium in the form of SeY.

Conclusions Selenium from SeY at 6 mg/h/d was transferred into milk and cheese at higher levels than selenium in the form of sodium selenite and was considered to be a more bio-available form of selenium. SeY (6mg/h/d) was effective in increasing the selenium content of milk and cheese and could be significant in increasing the selenium intake in the human diet which includes dairy products such as milk and cheese. However, selenium could not be offered at 3mg/h/d in the form of sodium selenite as this would have been below the recommended levels of intake and as a consequence the effect of these low levels of sodium selenite were not investigated in this experiment. .

Effects of rapeseed silage on dairy performance of Holstein dairy cow in early lactation

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Introduction In recent years increasing milk production in Iran caused to increase nutrient requirement of dairy cattle. In other wise in semiarid countries such as Iran providing forage requirement of cattle is limited by deficiency in forage resources. So it is better to use plants that they have low water requirements, high growth rate and high forage yield per hectare. Rapeseed is reasonably widely adapted and performs well in many areas in Iran. In spite of its low water requirements and high growth rate, its cultivation season is matched with environmental condition in Iran. Rapeseed (*Brassica napus L*.) recently has received consideration as a forage source for livestock. Its use has been restricted by numerous antinutritions such as glucosinolates and Erucic acid (Wilkes *et al*., 1987). Fales *et al.* (1987) reported a 100-fold reduction in Glucosinolate concentration when rapeseed forage underwent ensiling. The main objective of this study was to evaluate effects of rapeseed silage on Holstein dairy cow's performance.

Material and methods Nine multiparious Holstein cows were arranged in 3×3 Latin square design. Each period consisted of 15 days; 10 days were for adaptation and 5 days were for data collection. The treatments were 0, 2.5 and 5 % rapeseed silage in daily TMR that in fact corn silage was substituted by rapeseed silage. TMR consisted of 31% forage and 60% concentrate. Diets fed twice daily to allow for *ad libitum* intake and orts were weighted once daily during day 10 to 15 each period. Milk production was recorded on the last 5 d of each period and its composition data for the last 2 days milking were used for statistical analysis. Also Rumen fluid pH was determined on d 15 each period. Data were analyzed using the general linear model procedure of SAS (2000).

Results The results of this study were shown in Table 1. In this study results indicated that rapeseed silage has no significant effect on DMI, milk yield and milk composition.

Table 1 The effect of rapeseed silage on Holstein dairy cattle performance[1]

	Rapeseed silage % of DM			
Item	0	2.5	5	SEM
DMI kg/d	21	21	20	0.99
Milk yield kg/d	36.48	35.93	35.64	1.8
Milk Fat %	3.21	3.24	3.29	0.036
Milk protein %	3.03	2.97	2.96	0.063
Milk lactose %	4.55	4.57	4.57	0.026
Milk SNF %	8.52	8.52	8.48	0.059
Rumen pH	6.48	6.49	6.56	0.04

[1]None of the responses were significant

Conclusion the results of this study indicated that rapeseed silage didn't have any significant effects on DMI, milk yield and its constituents. In this study results showed that rapeseed can be used as silage especially in dairy cattle nutrition without any adverse effects on the milk production and its constituents.

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The effect of *trans*-10, *cis*-12 CLA on milk fat synthesis, energy metabolism and cheese yield in lactating ewes fed at two levels of energy intake

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Introduction Milk from ewes is characterized by a high fat content and ratio of fat to protein, but for most full fat type cheeses the high fat content is a disadvantage as the excess is lost in the whey. *Trans*-10, *cis*-12 conjugated linoleic acid (CLA) has been shown to be a potent inhibitor of milk fat synthesis in dairy cows and more recently in sheep (Sinclair *et al.,* 2005), and therefore represents a suitable management tool to reduce fat yield. In addition to a reduction in milk fat output, under some circumstances *trans*-10, *cis*-12 CLA results in an increase in milk and milk protein yield, particularly when the animal is under supplied with dietary energy. The objectives of this study were to examine the effects of supplementing dairy ewes with a rumen protected source of *trans*-10, *cis*-12 CLA at a high and low level of energy intake on milk fat synthesis, energy metabolism and cheese yield.

Materials and methods Sixteen multiparous ewes that weighed 59 (± 9.0) kg were individually penned and machine milked at 0800 and 1600h. When the ewes were ~5 weeks postpartum they were randomly allocated to one of four dietary treatments based on their performance in the week prior to allocation. Ewes were fed a complete diet (0.65 concentrates:0.35 hay; DM basis) at one of two restricted levels; High (H; approx. 28MJ ME/d) or low (L; approx. 21MJ ME/d), and supplemented with either Megalac (U: Volac, Royston, UK) or lipid encapsulated (LE) CLA (S: BASF AG, Ludwigshafen, Germany) in each of 4 periods of 21 d duration in a 4 x 4 Latin square design. The Megalac and CLA supplements were fed at 25g/ewe/day, providing 2.4 g/d of *trans*-10, *cis*-12 CLA to ewes on treatment S. The diets were formulated to provide a similar metabolisable protein supply (g/d). During the final 5 d of each period milk yield was recorded at each milking and samples taken for analysis of fat, protein and lactose. Milk was also collected over the final 5 d and made into a cheddar type cheese. Ewes were weighed at the start and end of each period. On day 20 of each period ewes were blood sampled at 0730, 1130 and 1430 h for subsequent analysis. Data was analysed using Genstat 7.2 (VSN Int. Ltd., UK) as a 2 x 2 factorial design with main effects of feeding level (L), fat source (F) and their interaction (Int).

Table 1 Intake and performance of ewes fed at a high (H) or low (L) level of feeding and supplemented with Megalac (U) or lipid encapsulated CLA (S).

	Treatments					Significance		
	HU	HS	LU	LS	s.e.m.	L	F	Int
Intake kg DM/d	2.10	2.08	1.66	1.66	0.018	<0.001	0.463	0.639
Milk yield g/d	1188	1225	1204	1244	40.4	0.664	0.347	0.974
Fat g/kg	6.36	4.87	6.17	4.79	0.093	0.148	<0.001	0.554
Fat g/d	75.2	59.1	73.8	59.9	2.12	0.889	<0.001	0.617
Protein (g/kg)	4.69	4.52	4.80	4.73	0.049	0.003	0.020	0.308
Protein (g/d)	54.8	55.1	57.2	58.6	1.45	0.050	0.575	0.685
Live weight change kg/d	0.134	0.130	0.034	0.066	0.0215	<0.001	0.523	0.415
Avg. plasma 3-OHB (mmol/l)	0.53	0.55	0.56	0.53	0.022	0.991	0.744	0.324
Green curd yield (kg/kg)	0.164	0.156	0.151	0.162	0.0073	0.661	0.851	0.281

Results Intake (kg DM/d) of ewes fed the lower level (L) was 0.79 of those on the high level (H; $P < 0.001$; Table 1). There was no effect ($P > 0.05$) of treatment on milk yield (g/d). By contrast, fat content (g/kg) was proportionally 0.23 lower ($P < 0.001$) and fat yield (g/d) 0.20 lower ($P < 0.001$) in ewes supplemented with CLA *vs.* Megalac. Milk protein content was proportionally 0.035 higher ($P = 0.003$) in ewes fed the low *vs*. the high level of feeding and 0.026 higher ($P = 0.020$) in ewes fed Megalac *vs.* CLA. Milk protein yield was also higher ($P = 0.05$) in ewes fed the low compared with the high level of feeding. Ewes on the high level of energy intake gained 0.083kg/d more ($P < 0.001$) than those on the low level. Plasma ß-hydroxy-butyrate (3-OHB) levels were not affected ($P > 0.05$) by treatment but glucose levels increased after feeding and were lower in animals receiving the high feeding level ($P < 0.001$) or CLA ($P < 0.05$; Fig. 1.). There was no effect of dietary treatment on curd yield (kg/kg; Table 1).

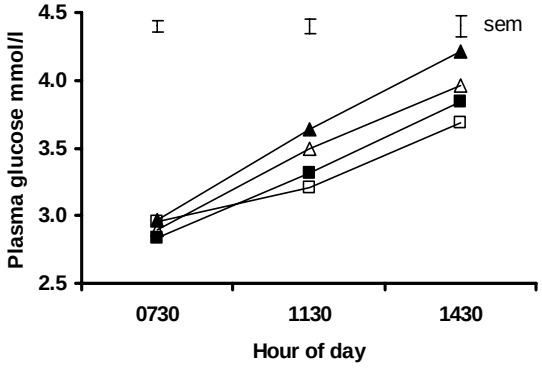

Figure 1 Effect of high feeding level + Megalac (■) or LE-CLA (□), or low feeding level + Megalac (▲) or LE-CLA (Δ) on plasma glucose concentrations (mmol/l).

Conclusions *Trans*-10, *cis*-12 CLA reduced milk fat content and yield similar to that recorded previously in dairy cows and sheep, but there was no effect on curd yield. Ewes responded to the greater level of energy intake by increasing weight gain rather than milk yield, but were in positive energy balance on all treatments.

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Effect of increasing milk replacer crude protein content and feeding level on heifer calf performance pre-weaning

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Introduction Previously reported results (Wicks *et al*., 2005) indicated growth rates from birth to day 28 increased when level of milk replacer feeding was increased from 600 to 1200 g/d, but that there was no increase when milk crude protein content replacer was increased from 230 to 300 g/kg (Wicks *et al*., 2005). Calf starter concentrate intake decreased with increasing milk replacer feeding and calves offered 1200 g milk powder/d at concentrations of 120 g powder/l water did not consume their whole allocation of milk replacer. The objective of the current study was to establish the optimum level of inclusion of milk replacer and to evaluate the effect of an intermediate protein concentration i.e. between 230 and 300 g/kg.

Materials and methods Ninety Holstein Friesian heifer calves were allocated to 8 pre-weaning nutrition treatments. Two milk replacers (230 or 270 g CP/kg DM) were offered at 4 levels of feeding (500, 750, 1000 and 1250 g powder/d). Calves were fed via computerised automatic feeders for both milk replacer and calf starter concentrate (180 g CP/kg). Heifer calves were group housed (maximum number of calves per group = 25) and bedded on straw. *Ad libitum* water and calf concentrate were available at all times. Daily individual intakes of both milk replacer and calf starter concentrate were recorded on 5 days per week. Live weight was recorded weekly from birth to weaning, and skeletal size (withers height, heart girth and body length) and body condition score were recorded fortnightly. Blood samples were taken from the jugular vein (using lithium heparin coated vacutainer sample tubes) at fortnightly intervals and analysed for plasma total protein and urea concentrations. The data were analysed using repeated measures analysis of variance, fitting fixed effects for treatment, sex, age and their interactions with birth weight as a covariate.

Results Increasing level of milk replacer feeding increased growth rates from birth to day 28 ($P<0.001$) but decreased calf starter intake ($P<0.001$) (Table 1). Figure 1 shows the increase in calf starter concentrate intake by age for the four levels of calf milk replacer feeding. The average age of calves consuming 0.5 kg calf starter concentrate was calculated as 38 and 54 days for calves offered 500 or 1250 g milk replacer/d (s.e.d. 3.1, $P<0.001$) respectively. This indicates that increasing milk replacer feeding delays the age at which calves start consuming starter concentrate. Milk replacer crude protein content did not significantly any parameter measured.

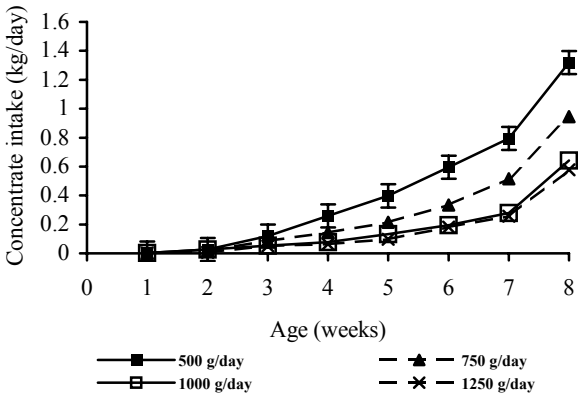

Figure 1 Effect of level of milk replacer on individual daily intake of calf starter concentrate from birth to weaning

Table 1 Milk replacer intakes, live weights, liveweight gains and skeletal size for heifer calves offered 4 levels of milk replacer containing two levels of crude protein (n = 90) from birth to day 28

	Milk replacer offered in 6 l water (g/d)					
	500	750	1000	1250	s.e.d.	Sig.
Milk powder (g/d)	457.7	691.6	921.3	1095.3	15.58	***
Calf starter intake (kg/d)	0.44[c]	0.28[b]	0.18[a]	0.15[a]	0.071	***
Birth weight (kg)	37.0	36.8	35.2	34.1	1.63	NS
Liveweight gain (kg/d)						
0-28 days	0.16[a]	0.25[a]	0.36[b]	0.42[b]	0.050	***
28-56 days	0.58	0.54	0.51	0.54	0.061	NS
0-56 days	0.37	0.40	0.44	0.49	0.047	P=006
Skeletal size at weaning – day 56 (cm)						
Withers height	80.5	81.9	81.4	82.0	1.11	NS
Body length	84.2[a]	86.8[ab]	87.1[ab]	89.1[b]	1.84	**
Heart girth	88.5[a]	90.6[ab]	90.8[ab]	93.3[b]	1.49	*
BCS[†] (day 56)	2.03	2.05	2.11	2.22	0.073	P=0.054

[†] BCS, Body condition score

Conclusion Calves fed higher levels of milk replacer grew significantly faster in the first 28 days of life but started consuming calf starter at a later age. There was no significant difference in growth, development or calf starter intake for calves offered the 230 or the 270 g CP/kg DM milk replacer.

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The effect of dietary fat source (beef tallow or sunflower oil) on fatty acid composition of main tissues and plasma lipid levels in growing – finishing pigs

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Introduction There has been known that body fat of pigs comes from a combination of fats produced endogenously and those from the diet for pigs. Polyunsaturated fatty acid absorbed from the diet specially inhibit endogenous synthesis of fatty acids, inflating the effect of dietary fat composition on body fat composition (Pettigrew and Esnaola, 2001). Futhermore, the supplementation of a highly polyunsaturated oil can decrease total serum cholesterols in pigs compared to those fed the diet supplemented with a highly saturated fat or oil. However, the plasma triacylglycerols concentration can vary greatly, depending in large part on dietary intake, storage in or mobilization from adipose tissues, and synthesis in the liver. Thus, the current study attempt to specify the effect of fat source on fatty acid composition of main tissues and plasma lipid concentration in growing-finishing pigs.

Materials and methods The experiment was approved by the Sakon Nakhon Agricultural Research and Training Center Ethical Committee. Thirty-six castrated-male pigs (Landrace × Large White × Duroc) were used in the study with an average 31.0 kg BW at the start. The pigs were allotted to treatments on the basis of weight in completely randomised design dividing them into 2 groups and then were penned in groups of 3 animals. The pigs were allowed *ad libitum* access to feed and water throughout the experiment. There were two experimental diets with different fat type (beef tallow; BT or sunflower oil; SFO) at 5 % as fed. The pigs were kept in individual metabolism cages at 4 week (50 kg BW) and 10 week (90 kg BW) after the start of the experiment, called 1st period and 2nd period, respectively. At the end of each period, the pigs were deprived of food for 10 hr prior to obtaining each blood sample drawn from the vena cava and collected in heparinized tube. At 12 weeks after the start of the study, all pigs were slaughtered and their samples (liver, subcutaneous adipose tissue, and *m. longissimus dorsi*) were collected for determining fatty acid profile by using gas chromatography. Plasma triacylglycerols and total cholesterol were measured enzymatically using test combinations. The effect of dietary fat type was evaluated for statistical significance by the Student's *t* test. All results are expressed as means ± SD.

Results There were statistically significant difference ($p < 0.05$) for some fatty acid composition in each tissue as shown in Table 1. The percentage of 16:0 and 18:1 in BT diet was higher than that in SFO diet while the percentage of 18:2 (n-6) in BT diet was lower than that in SFO diet (Table 1). The ratios of 16:1/16:0 and 18:2/18:0 in all selected tissues were statistically significant difference between the experimental diets. However, the ratio of 18:1/18:0 in all selected tissues was not significant difference. The concentration of plasma triacylglycerols of both periods in pigs fed SFO diet was significantly lower ($p < 0.05$) than that in pigs fed BT diet. In contrast, the concentration of total plasma cholesterol of both periods in pigs fed SFO and BT diets was not statistically significant difference (Table 2).

Table 1 Percentage fatty acids of selected tissues from pigs fed beef tallow or sunflower oil diet

	Tissue/diet					
	Liver		Adipose tissue		Muscle	
Fatty acid	BT diet	SFO diet	BT diet	SFO diet	BT diet	SFO diet
			Weight percentage			
14:0	0.56 ± 0.27	0.40 ± 0.37	1.56 ± 0.14	1.24 ± 0.21***	1.29 ± 0.23	1.27 ± 0.17
16:0	15.28 ± 2.14	13.41 ± 3.05*	22.97 ± 1.73	20.89 ± 2.56**	23.46 ± 1.90	22.40 ± 1.89
16:1	1.04 ± 0.36	0.58 ± 0.53**	2.13 ± 0.36	1.59 ± 0.35**	3.03 ± 0.38	2.23 ± 0.35***
18:0	21.13 ± 4.43	22.06 ± 4.67	12.80 ± 1.75	11.04 ± 1.75**	12.36 ± 1.15	11.59 ± 1.15
18:1	20.24 ± 4.43	16.34 ± 5.46*	42.49 ± 2.61	38.36 ± 2.26***	42.40 ± 3.25	38.53 ± 2.32***
18:2 (n-6)	15.94 ± 3.41	22.54 ± 2.49***	11.48 ± 3.22	22.29 ± 5.26***	9.37 ± 2.66	17.54 ± 3.97***
18:3 (n-3)	0.93 ± 0.41	0.65 ± 0.42	1.19 ± 0.23	1.01 ± 0.18*	0.65 ± 0.11	0.58 ± 0.17
20:4 (n-6)	13.37 ± 3.00	16.03 ± 4.61	0.15 ± 0.10	0.27 ± 0.06***	1.60 ± 0.91	1.58 ± 0.60
			Ratios			
16:1/16:0	0.07 ± 0.02	0.04 ± 0.02***	0.09 ± 0.02	0.08 ± 0.01***	0.13 ± 0.02	0.10 ± 0.01***
18:1/18:0	1.05 ± 0.49	0.85 ± 0.59	3.41 ± 0.61	3.55 ± 0.56	3.45 ± 0.39	3.35 ± 0.34
18:2/18:0	0.82 ± 0.41	1.08 ± 0.82*	0.92 ± 0.32	2.12 ± 0.74***	0.77 ± 0.27	1.56 ± 0.49***

* $p < 0.05$, ** $p < 0.01$, and *** $p < 0.001$, tallow-fed vs. sunflower oil-fed

Table 2 Plasma triacylglycerols and total cholesterol of pigs fed diets containing BT or SFO

	1st Period		2nd Period		
Fraction	BT diet	SFO diet	BT diet	SFO diet	Significant effect*
Triacylglycerols	4.20 ± 1.00	3.18 ± 0.77	3.30 ± 0.77	2.46 ± 0.74	D , T
Total cholesterol	2.62 ± 0.25	2.51 ± 0.29	2.67 ± 0.29	2.63 ± 0.25	NS

* D, T = significant ($p < 0.05$) diet, sampling time; NS = no significant diet, sampling time or interaction effect

Conclusions The supplementation of sunflower oil in growing-finishing pig diet resulted in higher incorporation of 18:2 (n-6), linoleic fatty acid, in liver, subcutaneous adipose tissue, and *longissimus dorsi* muscle of pigs and lower plasma triacylglycerols compared to the pigs fed the diet supplemented with beef tallow. However, there is no influence of dietary fat source on the plasma cholesterol.

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Influence of access to grass silage on the welfare of sows introduced to a large dynamic group

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Introduction Pregnant sows are commonly fed a restricted diet in order to minimise fat deposition and maximise efficiency of feed utilisation. This restrictive regime can contribute to increased levels of aggression, and also to the development of stereotypic behaviours (Meunier-Salaün *et al.,* 2001). The objective of this study was to assess the effects of providing access to grass silage on the welfare of sows introduced to a large dynamic group.

Materials and methods One hundred and eight Large White x Landrace sows were allocated to one of two treatments over six replicates. Treatments were as follows: (1) access to racks containing grass silage (offering an average of 1.9 kg silage/sow/day), and (2) control treatment with no grass silage racks. Treatments were applied to two separate dynamic groups, each containing 37 (±2) sows. Approximately 9 sows were replaced in each of these groups at 3-week intervals (each replacement constituting a replicate of the study). In a time-based cross-over design, treatments were swapped between the two dynamic groups at the end of the third replicate. Both dynamic groups were housed in identical split-yard systems (18.2 x 7.8 m), with slatted exercise areas and solid-floored kennel areas in both the pre- and post feeding yards. No bedding was provided for the sows. The pre- and post-feeding yards were separated by an electronic feeder which supplied 2.2 kg concentrates/sow/day. In treatment 1, grass silage (223 g/kg dry matter) was provided in a rack (1.2m wide x 0.8m high, with 5.3cm^2 mesh) in the exercise area of both the pre- and post-feeding yards. A collecting mat (1.5 x 0.6m) was placed under the racks to prevent silage falling through the slats. Both dynamic groups were videotaped during two 24-hour periods each week for the first 2 weeks after new sows were introduced to the group. Instantaneous scan samples were made at 1-hour intervals to determine if newly-introduced sows were lying, standing, or performing exploratory behaviour in either kennel or slatted areas. Aggressive behaviour towards newly-introduced sows was observed from video recordings for continuous 5-min periods each hour between 1100 and 1600 hours on the day that sows were added to the group. Observations were made on the basis of zero-one sampling, whereby whether or not aggressive behaviour occurred within each minute of the 5-min observation period was recorded. Aggression-related injury scores were measured from newly-introduced sows 1 week post mixing. Three newly-introduced and three "resident" sows were observed directly for 5-mins during three afternoons in week 1, and two afternoons in week 2, and time spent performing sham chewing was recorded. Aggressive behaviour was analysed by Fisher's exact test, and all other parameters were analysed by analysis of variance using Genstat 5.

Results Selected behavioural results are presented in Table 1. Within kennel areas, newly-introduced sows in the silage treatment spent more time lying down, and less time standing and performing exploratory behaviour than sows in the control treatment ($P<0.05$). Conversely, within slatted areas, newly-introduced sows in the control treatment spent more time lying down and less time standing than sows in the silage treatment ($P<0.01$). Overall, newly-introduced sows in the silage treatment tended to spend more time in kennel areas and less time in slatted areas than sows in the control treatment ($P<0.08$). Time spent standing, lying down or performing exploratory behaviour did not differ significantly between treatments ($P>0.05$).

Table 1 Influence of access to silage on the average proportion of time spent in different behaviours by newly-introduced sows during their first 2 weeks in large dynamic groups

Parameter		Silage	Control	s.e.m.	Significance
Within kennel areas:	Lie	0.89	0.81	0.021	*
	Stand	0.10	0.19	0.021	*
	Explore	0.09	0.18	0.022	*
Within slatted areas:	Lie	0.24	0.54	0.059	**
	Stand	0.75	0.45	0.063	**
	Explore	0.35	0.30	0.042	

Focal observations showed that sows in the silage treatment spent a significantly lower percentage of observation time sham chewing than sows in the control treatment (Silage: 7.6, Control: 18.8, s.e.m. 3.43, $P<0.05$). There was no significant interactive effect between sow type (i.e. newly-introduced and "resident") and treatment on this behaviour ($P>0.05$). Levels of aggression to which newly-introduced sows were exposed were low (average proportion of observations where aggression was observed was 0.01), and did not differ significantly between treatments ($P>0.05$). In addition, injury levels did not differ significantly between treatments ($P>0.05$).

Conclusions These results suggest that the welfare of newly-introduced sows to dynamic groups is improved by provision of access to racks containing silage. This was reflected in reduced sham chewing behaviour and increased use of kennels for lying behaviour. This latter finding suggests improved social integration by newly-introduced sows in the silage treatment. The lack of treatment effect on aggressive behaviour may reflect the low overall levels of aggression that were observed.

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The acute phase protein response during sub-clinical post weaning colibacillosis in pigs depends on the level of dietary protein

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Introduction The concentration of most plasma acute phase proteins (APP) increases at times of clinical infection in pigs (Murata *et al.*, 2004) and may be used as a marker to describe objectively pig health status. However, whether sub-clinical gastrointestinal infections also induce APP responses is less well known. Here, we have assessed whether experimentally induced sub-clinical post-weaning colibacillosis (PWC) affects the concentration of the two APP, i.e. haptoglobin (Hp) and C-reactive protein (CRP), which are two major acute phase proteins in pigs (Murata *et al.*, 2004). Current research aims to reduce sub-clinical PWC through the development of pig feeding strategies that do not rely on in-feed anti-microbial growth promoters (AGP). One such strategy explores the possibility of temporarily lowering dietary protein content (Wellock *et al.*, 2006). Therefore, an additional objective was to assess whether the APP response observed was sensitive to the level of dietary protein.

Materials and methods Fifty-four male Large White x Landrace pigs, weaned at around 3.5 weeks of age, were obtained from a minimal disease herd. They were housed individually, and divided in six groups of nine piglets each, which were balanced for bodyweight (mean 7.2±0.2 kg). They were offered either a high protein feed (H, 23% CP), a low protein feed (L, 13% CP) or H with AGP (HA). Five days post arrival (day 0), pigs were either dosed with 10^8 colony forming units of enterotoxigenic *E. coli* (ETEC) O149, diluted in phosphate-buffered saline, or were shaminfected, i.e. dosed with phosphate-buffered saline only. Feed intake was measured every day until day 3 when a blood sample was taken from three pigs per group for the assessment of HPT and CRP. Post infection feed intake was corrected for pre-infection feed intake to reduce between animal variation, and APP concentrations were log transformed. All data were analyzed through a 2x3 factorial ANOVA, which included pre-infection body weight as covariate. The experiment was approved by the Animal Experiment Committee of the Scottish Agricultural College (ED AE3/2004) and carried out under Home Office regulations for dosing with ETEC.

Results Figure 1 shows least-square mean feed intake post infection, and plasma APP concentrations. Infection and feeding treatment did not interact for feed intake (P=0.48). Across feeding treatments, infection reduced feed intake by 22% (P=0.07). Overall, HA pigs had higher feed intake than L pigs, with H pigs being intermediate (P<0.01). Infection and feeding treatment interacted for both APP (P<0.05). Infection significantly increased Hp in H and HA pigs, but not in L pigs. The concentration of CRP was significantly increased in all pigs due to infection (P<0.001). However, non-infected H pigs had higher CRP concentrations than non-infected L and HA pigs (P<0.05), whilst infected H and HA pigs had higher CRP concentrations than infected L pigs (P<0.05). CRP concentrations were similar in non-infected H pigs and in infected L pigs.

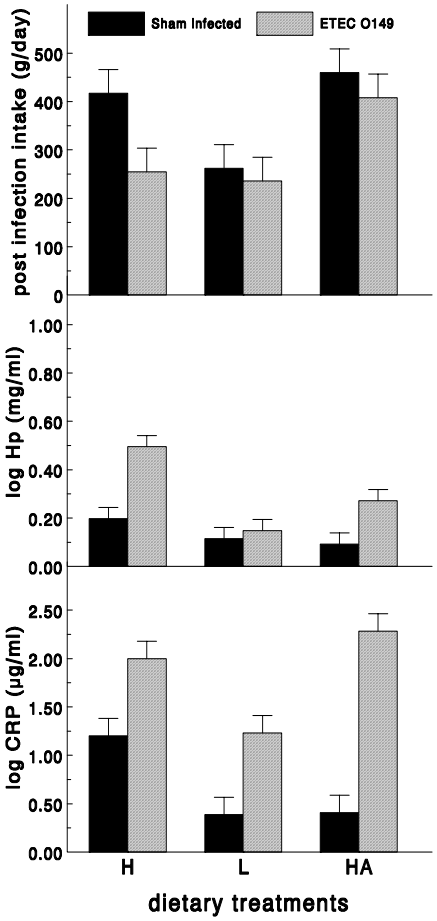

Figure 1 Mean feed intake, and concentration of plasma haptoglobin (Hp) and C-reactive protein (CRP) of pigs, infected with ETEC O149 or sham infected (see text for legend).

Conclusion The results suggest that sub-clinical PWC leads to clear changes in APP profiles. However, the results also indicate that these changes may be sensitive to the level of dietary protein. Hence, the usefulness of APP to describe health status in pigs would need to be considered in relation to their interactions with the nutritional environment. Further investigations into the effect of pig nutritional status on these and other APP are warranted.

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Effects of gender contact on the behaviour and performance of entire boars and gilts from 60-130kg

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Introduction The commercial trend of increasing slaughter weight in finished pigs, combined with the UK practice of using entire boars for production, might have detrimental consequences for behaviour, carcass quality and production efficiency. The associated increase in age is linked to an increased risk of reaching sexual maturity, with endocrine changes that result in behavioural problems and carcass taint. There is limited research examining environmental effects on performance and behaviour of pigs of modern genotypes finished to heavy weights. The sexual behaviour of boars can be influenced by the social environment during rearing (Thomas *et al*, 1979) suggesting that the risk of adverse behaviour around puberty might be modified. This study explored the effect of contact with the opposite gender on behaviour and performance in entire males and gilts in the 60kg-130kg weight range.

Materials and methods 16 pens of 8 (replicates 1-2) or 6 (replicates 3-4) Large White x Landrace finishing pigs were housed from 57kg to slaughter at 125 kg in pens in single sex rooms (SG), or in rooms where they were housed as single sex groups but adjacent to the opposite gender (OG) with sight, sound and touch contact through barred partitions. Pigs were fed a pelleted finisher diet *ad-libitum* with feed intakes and liveweights recorded weekly. Skin lesion scores (total number of lesions) were recorded bi-weekly and rindside damage (0-5 scale) post slaughter. For an assessment of sexual maturity, the weight of the paired ovaries or testicles was recorded at slaughter. Agonistic (biting, head to head knock, parallel pressing) and sexual behaviours (attempted mount, mount, anal nosing) were monitored by weekly behaviour sampling using period occurrence recording methods. Analysis of variance, with pen mean as the statistical unit, was used to compare gender and housing treatment effects and their interaction on performance and behaviour.

Results Over this heavy weight range, boars had increased daily liveweight gain (DLWG) and improved feed conversion ratio (FCR) compared to the gilts. Housing treatment had no significant effect on production (DLWG, P=0.14). There was no effect of gender or treatment on skin lesion scores or rindside damage. Gilts were significantly older at slaughter than boars; 8% had ovaries with corpora lutea and 31% had developing follicles, but this was unaffected by housing treatment. The OG boars exhibited a lower testicle weight at slaughter compared with the SG boars indicating reduced physiological maturity. Boars exhibited increased levels of body nosing (P<0.035) compared to the gilts, but there was no main or interactive effect of housing treatment. Boars exhibited significantly increased levels of aggressive (head to head knock P<0.001, parallel pressing P<0.001) and sexual behaviours (attempted mounting P<0.007, mounting P<0.013). When social contact was restricted to the same gender, the total sexual behaviour levels were significantly increased.

Table 1 Effects of gender and housing (gender contact) on performance, behaviour and physiological maturity

	Boars		Gilts			Sig.		
	SG	OG	SG	OG	SED	Gender	Housing	Interaction
DFI (kg)	2.76	2.97	2.80	2.78	0.093	NS	NS	*
DLWG (kg)	0.90	0.95	0.82	0.85	0.035	**	NS	NS
FCR	3.09	3.12	3.42	3.29	0.130	**	NS	NS
Days to slaughter	78.3	76.0	88.2	87.3	3.05	*	NS	NS
Skin lesions/pig	24.5	23.6	23.4	25.3	4.97	NS	NS	NS
Rindside damage score	1.3	1.3	0.9	1.0	0.45	NS	NS	NS
Testicle weight (g/pair)	635.5	573.5	-	-	31.67	-	*	-
Ovary weight (g/pair)			8.25	8.92	1.009	-	NS	-
Behaviour (no/pig/min): Aggressive behaviours	0.109	0.116	0.061	0.053	0.009	***	NS	NS
Sexual behaviours	0.043	0.023	0.017	0.014	0.005	*	*	NS

Conclusion Boars remained more efficient than gilts at this heavier weight range. Whilst there was no effect of gender contact on gilt physiological maturity, boars with gilt contact were less physiologically mature at slaughter and showed reduced levels of sexual behaviours. Skin damage was not significantly affected by sex or gender contact, despite increased levels of aggression in boars compared to the gilts. Boar aggression was not affected by gender contact. Tissue samples taken at slaughter will enable quantification of carcass taint compounds associated with these treatments.

Acknowledgments ELRS was in receipt of an MLC studentship. We thank staff at Cockle Park farm for technical help.

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Effect of DHA edible algae in maternal and post-weaning diets on piglet health and performance

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Introduction Weaning is a critical process in piglets, with increased risk of health problems and decreased performance. Previous studies have shown that fish oil, rich in long chain omega-3 fatty acids, has an anti-inflammatory effect in pigs, which has led to improved pig performance under infectious challenge conditions (Liu *et al*. 2003). When piglets are weaned onto diets containing plant proteins, local inflammatory responses in the gut can reduce feed intake and performance. This study was designed to investigate the effect of Docosahexaenoic Acid (DHA) edible algae, as an omega-3 fatty acid source, in sow lactation and piglet post-weaning diets on piglet health and performance, when diets of different potential antigenicity were fed after weaning. It was hypothesised that increased omega-3 intake, either in sows' milk before weaning or in the diet at the time of weaning, might facilitate the transition to solid food.

Materials and Methods The pre-weaning phase of the study used 22 litters (247 piglets). The sows were allocated in pairs between treatments providing either 3 g DHA in whole dried algal biomass/ kg or 8 g corn oil/ kg (control of equal added lipid energy) in the feed from farrowing until weaning at 4 weeks. Piglets were weighed weekly from birth to weaning. Post-weaning, the study was a 2x3 factorial arrangement of treatments applied within litter, with either a soya or milk protein based diet and 0, 3 or 5 g DHA/ kg feed. The study included 132 piglets (11 replicates), with six piglets from each litter, selected on the basis of closeness to median weight. Two litters were weaned simultaneously and two piglets, one from each treatment litter, were penned together. All feed was in meal form, and the amounts offered and refused were recorded daily. Piglets were weighed weekly for the following 21 days. Pre-weaning treatments were compared by one way ANOVA with litter as the experimental unit. Post weaning treatments were compared by two-way ANOVA, using pen as the experimental unit, with diet, DHA level and their interaction as treatment factors, time replicate as a blocking factor and mean pen weaning weight as a covariate. The possible residual effect of lactation treatment on piglet adaptation to weaning was examined by within pen comparison of daily liveweight gain.

Results Pre-weaning, there was no significant treatment difference in birth weight, average daily weight gain (ADG) and weaning weight. Post-weaning, there were no significant effects on overall voluntary feed intake (VFI), ADG and feed conversion ratio (FCR) from diet or DHA inclusion level. There was, however, a significant interaction between diet and DHA-level on ADG in the second week post-weaning; increasing DHA-level increased ADG, but only in the soya diet fed piglets. No residual benefit of pre-weaning DHA treatment on post- weaning growth was apparent.

Table 1 Performance data for the piglets in the four week suckling period

	+DHA	–DHA	SEM	Significance
Birth weight (Kg)	1.53	1.66	0.03	NS
ADG (Kg/piglet)	0.22	0.23	0.07	NS
Weaning weight (Kg)	8.00	8.13	0.17	NS

Table 2 Effect of dietary protein source and level of DHA on piglet performance during the three week post-weaning period

Diet	Milk			Soya				
DHA-level (g)	0	3	5	0	3	5	SEM	Sig.
VFI (g/ day)	474	441	464	452	448	473	16.7	NS
ADG (g/day)	322	307	325	303	320	330	12.6	NS
FCR	1.49	1.46	1.44	1.51	1.43	1.45	0.03	NS
ADG week 2	372	336	324^{a}	320^{a}	383	394^{b}	18.6	DietxDHA***

[a,b] Values with different superscripts are significantly different ($p<0.05$)

Conclusions Pre-weaning allocation of DHA to lactating sows resulted in no direct or residual effect on piglet performance. Post-weaning the hypothesised trend for increased performance of piglets given the soya diet, but not the milk diet, when a higher level of DHA was included, was only significant during the second week. Pigs on the soya protein source performed better than anticipated relative to those given milk protein and effects of treatment on feed intake may have been partly obscured by higher wastage with meal diets.

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Behavioural and physiological indicators of piglet survival

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Introduction Pre-weaning piglet mortality is currently 11.8% of piglets born alive in indoor units (MLC, 2005) and is a major welfare concern and a continuing production problem within the pig industry. The farrowing crate was implemented with some success to decrease the amount of crushing of piglets (Edwards & Fraser, 1997). However, this system is restrictive, limits the behaviour and compromises the welfare of the sow (Jarvis *et al.,* 2001). There is growing pressure to abolish this technology in favour of less restrictive systems. It is therefore vital to identify behavioural and physiological characteristics relating to piglet survival, which can then be influential in alternative systems. Important factors in relation to piglet survival include birth weight, birth order, and adequate thermoregulation (Tuchscherer *et al*. 2001). The aim of this study was to identify additional behavioural and physiological indicators, which could predict piglet survival.

Materials and methods Ten sows were moved to standard farrowing crates 5-7 days before they were due to farrow. A commercial lactation diet (18% CP, 13.75 MJ DE kg^{-1}) was offered twice daily. Sows had *ad libitum* access to water and were provided with straw bedding. The piglets had access to a heated and lit creep area, bedded with sawdust. Behavioural and physiological data were collected during the farrowing period and one day post-farrowing. At birth, individual piglet placentae were identified by tagging the umbilical cord. Placental efficiency (Piglet birth weight/Placental weight), placental weight and areolae per cm^2 were analysed. Immediately after birth, the vigour of the piglet was assessed and scored using a categorical scale. A blood sample was taken at birth and at 24h for glucose analysis. Piglet' weight, sex and length were recorded. Rectal temperature was recorded at birth, 1h, and at 24h after birth. Piglets underwent two neurobehavioural tests. The first involved measuring strength and vigour by capturing the rooting response of the piglet soon after birth using an adapted bi-axial potentiometer device with a dummy teat mounted to it. The second test involved timing the piglets' righting response from being placed on its back and scoring the efficiency of righting. Statistical analysis was performed using a Generalised Linear Mixed Model (GLMM), allowing a binomial structure (piglet either died or survived) and using a Logit transformation (Genstat version 7). Spearman's ranked correlations were used to determine the relationships between measures.

Results Ten sows produced 135 piglets (113 survived, 6 stillbirths, 10 crushed, 3 low viability/starved and 3 were euthanased later). The piglets that were euthanased were removed from all the analysis. Stillbirths were included in the analyses relating to pre-natal survival traits and excluded from post-natal analyses. GLMM results for birth weight as a predictor of death were significant (Wald statistic = 13.11, df = 1, P <0.001, effects = –3.508 ± 0.97). With birth weight added as a co-variate other variables significantly predicted death (Table 1).

Table 1 GLMM results using the wald statistic. Significance determined using a X^2 distribution. Birth weight accounted for as a co-variate. Df=1.
Euthanased piglets excluded from all analyses. † Stillbirths not included in analysis

Variable	Wald statistic	P	Effect (SE)
Birth order	11.19	< 0.001	0.20 (±0.06)
Farrowing length	12.10	< 0.001	0.02 (±0.01)
Placental efficiency	6.70	0.010	– 0.50 (±0.19)
Placental weight	7.49	0.006	0.02 (±0.01)
Areolae per cm^2	4.20	0.040	– 1.16 (±0.80)
†Rooting response	23.58	< 0.001	– 9.20 (±1.90)
†Birth rectal temperature	6.15	0.013	– 0.66 (±0.27)
†24h rectal temperature	3.28	0.070	– 0.55 (±0.31)

Placental efficiency was negatively correlated with birth glucose (–0.375, P<0.001) and farrowing length (–0.266, P<0.01). Birth glucose was positively correlated with birth order (0.244, P<0.01) and farrowing length (0.242, P<0.05). Rooting response was positively correlated with birth weight (0.412, P<0.001) and birth rectal temperature (0.313, P<0.01).

Conclusions Birth weight continues to be a critical factor with regard to piglet survival. However, this study implicates additional pre-natal and post-natal measures which predict survival. The sow must provide an adequate uterine environment, with an efficient and sufficient placenta to ensure offspring viability. High glucose at birth, correlating with being born later in the birth order and increasing farrowing length, indicates that piglets are suffering from an inadequate uterine environment or birth stress, which reduces their survival chances. Quantifying a piglet's rooting response provides information on strength and vigour, which is not solely a function of birth weight. This study provides additional behavioural and physiological indicators of piglet survival, which can be used to assess neonates at risk in alternative systems.

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Seasonal changes of lameness and its relation with hoof lesions in dairy cow

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Introduction Lameness of dairy cattle is an economic and welfare issue. This problem is the third most important health related economic loss facing the dairy industry, following fertility and mastitis. Lameness often follows a seasonal trend with an increase in lesions associated with laminitis appearing several weeks after some environmental hardship such as heat stress. Also healthy hooves are an important prerequisite for the welfare and productivity in dairy cow.

Materials and methods This study was conducted at the Ferdowsi University Research Farm with a herd size of 100 milking dairy cows. Prevalence of lameness was evaluated using the modified locomotion scoring system (LS) described by Weary (2004) adapted from Spercher *et al.* (1997). LS was visually assessed with observing cows in a walk distance of 10 m long following a.m. milking. Animals were scored from 1 to 5 with observing various behavioural indications such as back arch, swinging in/out, head bob, tracking up, joint flexion, asymmetric steps and reluctance to bear weight. In this method LS=1 represented normal animal, LS =2 and 3 as cows with sub-clinical lameness and LS=4 and 5 regarded as the cow with clinical lameness. The hoof lesion scoring system was based on the proposed method by Greenough and Vermunt (1991). Description of lesion scores were slight discoloration and localized area for score 1, moderate haemorrhage for score 2, sever haemorrhage for score 3 and exposed corium sole ulcer for score 4. Milk fat depression is commonly used to rule ruminal acidosis as risk factor for laminitis in dairy cow, and because no attempt was done measuring ruminal pH in the experiment the milk fat percentages recorded 8 weeks before and at the time of locomotion scoring were employed as indirect indicator of rumen pH. The data on milk composition mainly fat content were obtained from the Breeding and Improvement Centre of Ministry of Agriculture. The GENMOD Procedure of SAS was employed for analysing the discrete dependent variables in this study.

Result The data in Table 1 show that this herd has had the worst locomotion score in autumn with a significant level of (P<%5) although, the incidence of sub clinical lameness in all seasons was quite high. However, no significant difference between milk fat depression and locomotion scores was found in this experiment. No relation was found between animal locomotion score and hoof lesion in this study. The Pearson correlation between LS and milk fat percentage in the study was 0.14.

Table 1 Herd LS profile of cow at different seasons (%) and its relation with hoof lesions and milk fat

Locomotion Score	Season					Hoof score	Milk Fat (%)	
	summer	Autumn	winter	spring	SEM		8WB[1]	ATS[2]
1	24.8	11.9	20	14.2	0.38	2	3.85	3.34
2	41.6	33.7	37	39.5	0.29	2.3	3.82	3.7
3	28.3	33.7	35	40.3	0.30	2.37	3.65	3.67
4	2.7	11.8	5	3	0.69	1.7	3.01	3.49
5	2.7	9	3	3	0.68	2.5	2.59	3.56
Mean	**2.2**	**2.7**	**2.3**	**2.5**	**-**	**-**	**-**	**-**
Sub clinical lameness	69.9	67.4	72	79.5	0.29	-	-	-
Clinical lameness	5.4	20.8	8	6	0.51	-	-	-

[1]8WB= 8weeks before measuring [2] ATS =At the time of measuring LS

Conclusion The high incidence of sub clinical lameness in this study shows that more attention must be done to diet formulation and preparation as well as improving technical management in the farm. Higher prevalence of clinical lameness in autumn could be due to the effect of heat stress in late summer in the area. Vermunt and Greenough (1996) suggested that haemorrhages in the sole reflect either a feeding change or trauma that occurred 6 to 8 week before observation of the haemorrhage. But in this study no significant correlation between hoof lesion and milk fat percentage as an indirect indicator of rumen pH was found. It was concluded that badly designed housing system like rough concrete floor surface, long and slurry walking distance to milking parlour and crowding more animal in waiting area before milking could be the other reasons for the cow lameness in this farm. However more researches are needed for finding the main reason(s) of seasonal variation and accurately partitioning different risk factors engaged with lameness in this centre.

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Influence of rearing regime on the development of sole lesions in dairy herd replacements

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Introduction The precise aetiology of lameness in dairy cows remains unclear, and it's likely that a number of different factors affect its development. The aim of the current study was to investigate the effects of different management regimes during rearing on the development of sole lesions in dairy herd replacements.

Materials and methods One hundred high genetic merit Holstein-Friesian heifers in an Autumn-calving herd were used in this study. Heifers were reared from 7 weeks of age until calving at 2 years of age on one of four rearing regimes in a 2 x 2 factorial design experiment. Heifers in treatments 1 and 3 were offered grass silage-based diets during winter periods and grass-based diets during summer periods ('Feeding system 1'). Heifers in treatments 2 and 4 were offered straw/concentrate diets during the winter periods ('Feeding system 2'). These heifers remained housed and were offered straw/concentrate diets during their first summer period, and were grazed outdoors during their second summer period. Target calving weight in treatments 1 and 2 was 540 kg ('Low'), and in treatments 3 and 4 was 620 kg ('High'). The higher target calving weight was achieved by additional concentrate supplementation during rearing. Sole lesion and heel erosion scores were recorded from both hind hooves of each heifer at different time points during rearing and first lactation i.e. at approximately 7 weeks of age, at approximately 6, 12, 18 and 24 months of age (Pre-calving), and at approximately 1, 2, 3, 4, 6 and 11 months post calving. The measurements at 6, 12, 18 and 24 months of age corresponded with the end of the 1st winter period, the start of the 2nd winter period, the end of the 2nd winter period and the start of the 3rd winter period, respectively. During each examination, the sole of each claw was functionally trimmed, and sole lesion scores and heel erosion scores recorded using the method of Livesey *et al*. (1998). For reasons not related to the experiment, 19 heifers were excluded from the data set for statistical analyses. Data were analysed using the Genstat 5 repeated measures REML procedure.

Results Sole lesion results are presented in Table 1. There was a significant interaction between target calving weight and time period on sole lesion scores (P<0.001). 'High' heifers had greater lesion scores at 6 and 24 months of age, and at 3 and 4 months post calving than 'Low' heifers (P<0.05). An interactive effect was also shown between feeding system during rearing and time period (P<0.05). At 12 months of age, heifers in 'Feeding system 2' showed greater levels of sole lesions (P<0.05). There was no significant interactive effect between feeding system and target liveweight on sole lesion scores.

Table 1 Effect of feeding system during rearing and target calving weight on sole lesion scores in heifers (Standard error of the difference between means at the same age = 0.065)

	Target calving weight		Significance	Feeding system		Significance
	Low	High		1	2	
7 weeks of age	0.00	0.00		0.00	0.00	
6 months of age	0.25	0.43	***	0.31	0.37	
12 months of age	0.23	0.27		0.17	0.33	*
18 months of age	0.39	0.43		0.36	0.46	
24 months of age	0.27	0.55	***	0.36	0.46	
1 month post calving	0.55	0.54		0.60	0.50	
2 months post calving	0.93	0.94		0.99	0.88	
3 months post calving	0.89	1.01	*	0.95	0.95	
4 months post calving	0.67	0.80	**	0.73	0.74	
6 months post calving	0.64	0.63		0.69	0.58	
11 months post calving	0.41	0.43		0.45	0.39	

Significant interactions were shown between target calving weight and time period on heel erosion scores (P<0.001). 'High' heifers showed greater heel erosion scores than 'Low' heifers at 18 months of age (P<0.001) and at 3 months post calving (P<0.05). Interactions were also shown between feeding system and time period on heel erosion scores (P<0.05). At 12 months of age, heifers in 'Feeding system 2' showed greater heel erosion scores than heifers in 'Feeding system 1' (P<0.001). There were no significant interactions between feeding system and target calving weight on heel erosion scores.

Conclusion Increasing target calving weight appears to contribute to the development of sole lesions in dairy heifers. Increased lesion scores in 'Feeding system 2' heifers at 12 months of age may have reflected their preceding summer housing period. The nature of heel erosion results suggests a relationship between heel erosion and development of sole lesions.

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Differences in defaecation behaviour between dairy cows housed in straw yards or in cubicle systems

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Introduction Very little is known about the eliminative behaviour of dairy cows though housed cattle are traditionally thought to show little or no faeces-avoidance behaviour (Hafez and Schein, 1962). Management practices, such as cubicle housing, are designed to limit contact between cows and their faeces but are often restrictive in nature and can have an adverse effect upon other, unrelated behavioural patterns. For welfare reasons, housing systems for dairy cows should be designed with an aim to reduce the differences in behaviour between outdoor and indoor systems. This study aims to provide a clearer understanding of the eliminative behaviour of dairy cows in the two most commonly used housing systems in the UK, i.e., straw yards and cubicle yards and to investigate whether any difference in behaviour is imposed by housing regime.

Material and methods The eliminative behaviour of two high-yield groups and two low-yield groups housed in cubicles and a straw yard were recorded. The treatment groups were: Straw Low (n =73), mean yield 13.8 $\pm$ 7.8 kg/d; Straw High (n = 72), 40.1 $\pm$ 13.5 kg/d; Low Cubicle (n = 85), 17.8 $\pm$ 6.4 kg/d and High Cubicle (n = 93), 38.7 $\pm$ 7.5 kg/d. The same cubicle yard and straw yard were used for the two yield groups. Both high yield groups were under the same feed and milking regime as were the low yield groups.
Cows were observed by the same observer for a total of 24 hours during their normal daily routine. Any cow that was observed to be about to defaecate had their behaviour classified as Lying (L), standing (S) or walking (W) immediately before, during and after voiding. If the post-defaecation behaviour was only maintained for a short time (up to and including 10 seconds), the subsequent behaviour was also recorded. The total expression of defaecations was recorded and sequences were then classified as showing a) no avoidance of faeces, b) incidental avoidance of faeces and c) intentional avoidance of faeces. Sequences were recorded as incidental avoidance where cows moved away from freshly deposited faeces but were also engaged in a second activity such as changing places at the feed bunk. For goodness of fit, the G-test was used along with its associated William's correction factor.

Results A total of 3438 expressions of defaecation behaviour were recorded for the 323 cows included in the study with group means per cow per 24 hours of Straw Low = 9.41; Straw High = 12.82; Cubicle Low = 10.01; Cubicle High = 10.51. In all, 35 sequences were recorded and these were classified as showing a) no faeces avoidance, b) incidental faeces avoidance, or c) intentional faeces avoidance. Cows housed in cubicles showed the 'no avoidance of faeces' sequences significantly more than did cows housed in straw yards ($P < 0.001$) (Table 1). Conversely, cows in straw yards showed a significantly greater level of both incidental and intentional faeces avoidance ($P < 0.001$ and $P < 0.001$ respectively).

Table 1 Classification of sequences of behaviour indicating intentional, incidental or no avoidance of faeces.

Behaviour category	Straw (N = 145)	Cubicles (N = 178)	G_{adj}	P value
a) No avoidance	19	120.09	81.603	< 0.001
b) Incidental avoidance	1517	1308.17	15.448	< 0.001
c) Intentional avoidance	58	2.32	63.428	< 0.001

Comparing the behaviour of the high and low yield groups in cubicles, the proportion in the high yield group remaining lying throughout (LLL) was significantly greater than in the low yield group ($G_{adj} = 8.209$; $P = 0.004$). As with the cows housed in cubicles, the high yield group on straw showed a significant, but less marked, increase in the expression of LLL ($G_{adj} = 3.889$; $P = 0.049$).

Conclusion Straw housed cows exhibited greater levels of incidental and intentional avoidance of faeces than did cubicle cows, suggesting the existence of an inherent motivation to avoid faecal contamination in dairy cows, which was constrained by movement restrictions in cows in the cubicle system. The levels of incidental and intentional faeces-avoidance behaviour, as well as no avoidance of faeces, were also affected by yield status and the greater incidence of high yield cows remaining lying whilst voiding may be indicative of a greater motivation to lie and rest. The high levels of faeces avoidance recorded for the cows studied challenges the traditional assumption that cows do not avoid contact with freshly deposited faeces.

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Eating and ruminating behaviour of Holstein and Jersey cows

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Introduction Declining longevity, poor fertility and increasing incidence of disease in Holstein cows has led to renewed interest in alternative dairy breeds. The Jersey is the second most popular breed in the UK, and its numbers have increased by 50% since 1999 (The Jersey Cattle Society). Limited research has indicated that there are physiological differences between the Jersey and larger dairy breeds, in addition to those of milk quality. Consequently, current dairy cow rationing systems may be inappropriate for Jerseys because they are based on data from Holsteins. The behaviour data presented here was part of a larger study which also compared the intake capacity, diet digestibility and rate of passage of Jersey and Holstein cows.

Materials and Methods Total time spent eating and ruminating was measured in six Jersey (mean bodyweight (BW) 462 ± 18 kg) and six Holstein (mean BW 678 ± 18 kg) third parity cows for five days during weeks 5 and 2 pre-calving and weeks 2, 6, 10 and 14 of lactation using IGER Behaviour Recorders (Rutter *et al*. 1997). Cows were kept in individual tie-stalls. Restricted amounts of total mixed rations (TMR) based on grass silage, hay and concentrate (660, 220, and 120 g/kg DM respectively; 146 g crude protein (CP) and 523 g neutral detergent fibre (NDF)/kg DM) and grass and maize silages, hay and concentrate (160, 48, 188 and 244 g/kg DM respectively; 166 g CP and 412 g NDF/kg DM) were offered from drying-off until week 3 before calving and during the last two weeks of gestation respectively. After parturition cows were offered *ad libitum* a TMR based on grass and maize silages and concentrates (311, 311 and 378 g/kg DM respectively; 173 g CP and 386 g NDF/kg DM). Feed intake was divided into meals by the method of Sibly *et al*. (1990). The temporal distribution of eating behaviour during the lactation periods was assessed by dividing each day into 4-hour intervals and determining the proportion of total daily eating behaviour that occurred in each interval. Daily means of each variable were calculated for each cow period and the effects of breed on measurements repeated within cows were tested using the Mixed procedure of SAS.

Results Dry matter intake (DMI) was higher in Holsteins compared to Jerseys, but intake capacity (DMI per kg BW) did not differ (P>0.05) between breeds (mean 16.6 ± 0.77 g/kg BW during dry period and 34.1 ± 0.81 g/kg BW during lactation). The number of meals consumed and total daily eating time did not differ between breeds, but Holsteins ruminated for longer (Table 1). Despite their smaller DMI Jerseys spent longer eating and ruminating each unit of DM and total chewing time per kg of BW was also greater (Table 1). Jerseys tended to spend less time eating between 0800 and 1200 h (P<0.093) and more between 2400 and 0400 h (P<0.060) than Holsteins, thus feed intake was more evenly distributed throughout the day in Jerseys (Figure 1).

Table 1 Eating and ruminating behaviour at weeks 5 and 2 pre-calving and weeks 2, 6, 10 and 14 of lactation

	Breed			
	Holstein	Jersey	SEM	P<
DMI (kg/d)	18.1	12.2	0.43	0.001
Eating time				
min/day	310	331	12.0	0.255
min/kg DM	18	29	1.5	0.001
Meals/day	11.1	11.6	0.59	0.611
Ruminating time				
min/day	574	505	13.0	0.004
min/kg DM	35	47	1.3	0.001
Total chewing (eating and ruminating) time				
min/kg BW	1.37	1.86	0.07	0.001

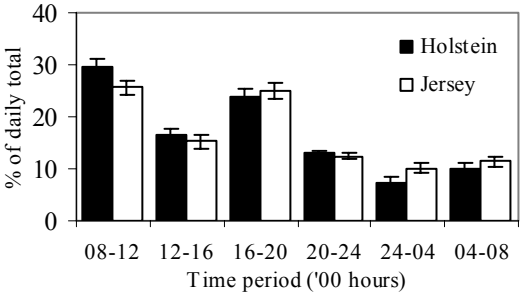

Figure 1. Temporal distribution of eating behaviour during the lactation periods

Conclusion Physical constraints on total chewing time were probably more limiting for Holsteins than for Jerseys in this study, because Holsteins had a larger volume of feed to process. Jerseys were therefore able to spend longer chewing each unit of DM, and it is likely that this was responsible for the increased NDF digestibility and rumen passage rate observed in this study, which was reported previously (Aikman *et al*. 2004). Interactions between cows were limited in this study, but even greater differences between breeds may be observed in grazing or loose-housed animals, due to the influence of environmental factors and competition from herdmates.

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Behavioural responses of dairy cows to novel situations

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Introduction Dairy cattle show individual variation in their behavioural responses to routine practices carried out on farms. These behavioural responses are assumed to reflect underlying temperament traits such as fear. It is widely recognised that individual differences in the behavioural responsiveness of animals to stressful stimuli may be controlled by biological traits rather than simply reflecting random variation. Such individual differences have been interpreted in terms of either coping styles or fearfulness, both of which are assumed to be governed by underlying biological traits including genetic properties of the individual animal (Koolhaas *et al.,* 1999). The aim of this study was to investigate individual cow's level of reactivity and responsiveness to the environment in order to quantify aspects of temperament by devising one novel object (NO) and one human interactive (HI) test practical for use on commercial farms. The tests need to show good repeatability and reliability, be short in duration, non-invasive and not disruptive to the daily farming routine. The long-term objective is to develop suitable temperament scores that can be used in future breeding programmes or as part of welfare assessment schemes.

Material and methods The subjects of the experiment were Holstein-Friesian cows from lactation 1 – 10.
NO Tests: Thirty cows were subjected to 3 novel objects in a latin square design. The novel objects were placed in the exit passageway of the milking parlour. The novel objects were an orange flashing light, a pair of black and yellow striped boards attached to both walls and a spray of water from a water pistol to the hind-quarters. Responses indicative of the cows' fear and curiosity as they approached, interacted and passed the novel situations were recorded and analysed. The cows were allocated a point for each of the following four fear behaviours: Stop, Avoid, Flinch/Jump & Increased pace. The cow's curiosity responses towards the NO was scored on a scale ranging from 1 (no response) to 6 (interacts with object).
HI Tests: Thirty-six cows were tested in 2 different human interaction tests; Flight from Feeder (FF) test and Approach in the Passageway (AP) test in a latin square design. Each test was repeated thrice per individual to assess repeatability. The FF test assessed the animal's reactivity in a conflict situation between feeding motivation and fear of an unfamiliar human by recording the cow's flight distance. The AP test assessed the cow's flight distance and on completion of this test a qualitative assessment of differing aspects of the cow's temperament was scored. The experimenter marked an individual visual analogue scale for six expressive terms (Aggressive, Attentive, At Ease, Nervous, Passive & Social).

Results.
NO: A moderate consistency in frear reactions of cows to all 3 tests was found with level of frear being greatest in the Waterspray test (W=0.226, P=0.02). No level of agreement was found in curiosity responses between tests (P=0.47). A negative correlation exists between fear and curiosity responses (P=<0.001).

NO Test	**Fear**			*Curiosity*		
	Med	Q1	Q3	Med	Q1	Q3
Waterspray	67	50	100			
Board	0	0	25	67	33	67
Light	25	0	25	33	20	60
Kruskal-Wallis Test	*P*<0.001			P<0.05		

Table 1 Fear & Curiosity responses to NO where increasing % indicates an increasing level of response.

HI: Flight distance in the passageway showed high repeatability (0.72) while repeatability of flight distance at the feedface (0.13) was low. Qualitative terms showed good repeatability: Social (0.62), Passive (0.51), Nervous (0.50) and At ease (0.48). Fight distances in the AP and FF tests showed a positive correlation (r_S=0.4833, *P*<0.01). Fear responses in the NO tests showed a strong level of concordance with the flight distance scores for the AP tests (W=0.26, *P*=0.02).

Conclusion The data from the AP test demonstrated high repeatability indicating that it is the most appropriate HI test to be used in commercial farm situations. The strong relationship between the FF and AP flight distances suggest that these tests are indicative of underlying reactivity to humans. The strong level of agreement between fear reaction to novelty in the NO tests and the fear of humans tested in the flight distance from the AP test indicate that both tests are measuring the same underlying trait. The pattern of response shown by cows in the NO tests indicates that more than one trait governs behavioural responsiveness. Behavioural responses in the NO tests may be controlled by underlying fear and/or curiosity responses (van Reenen *et al,* 2004). In summary, after screening 3 potential novel objects the striped board test has been selected for further evaluation. Future work will report on the responses of cows to this stimulus and the AP test in commercial situations.

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Effect of body condition score at calving and diet energy content post calving on behavioural activities of dairy cows during early and mid-lactation.

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Introduction Excessive and prolonged negative energy balance (NEB) has become a major problem in the modern high yielding dairy cow, leading to metabolic disorders, poor fertility and subsequently increased culling rates. Exploring potential indicators and possible strategies to reduce excessive NEB is imperative to improving the sustainability of this system. In this study, treatment effects on animal behaviour patterns (used in assessing animal welfare) were investigated in an attempt to identify possible indicators of compromised welfare. A change in the frequency and duration of behavioural activities may be associated with an excessive NEB and prove useful in early detection of metabolic problems.

Material and methods Forty Holstein cows, between 2nd and 6th parity (live weight 605 ± 68.3kg) were allocated to one of four treatments, based on a 2x2 factorial design: high or low energy density diet pre- and post calving. All diets were fed as a TMR. The pre-calving treatment commenced 100 days prior to the predicted parturition date. All diets were offered *ad lib*, however cows receiving the low energy diet pre-calving were restricted (R) to 6 kg DM complete diet/ day from day 42 pre-calving, while the high energy pre-calving diet continued to be fed *ad lib.* (A). Treatments were balanced for parity, body weight and date of calving. Upon parturition the post calving treatments commenced. The concentrate: forage ratios of the high (H) and low (L) energy density diets post calving were 70:30 and 28:72 respectfully, providing 12.5 and 11.7 ME/ kg DM. Consequently, there were four treatments groups; AH, AL, RH and RL. Cows were fed between 10.00 and 11.00 daily. The behaviour of all animals was observed directly over a 24 hour period once a fortnight, until day 140 post-partum. Each animal was scanned at 15 minute intervals and a number of behaviours recorded. These included; lying, standing, feeding, ruminating, drinking, queuing, walking, mounting, social behaviour (grooming) and aggressive behaviour. Additional measurements included; lying position: stretched (at least one limb fully stretched) or not stretched and abdominal pressing. All the post-partum cows were housed as a single unit in free stalls with concrete flooring and had electronic access to their appropriate diets. As the animals on different treatments were not housed in separate treatment groups, the behaviours observed on the individual treatments may not be totally independent of each other. Data were analysed using REML variance components analysis. The average daily energy balance (ADEB) for each cow was calculated per week of lactation, using 'The Feed into Milk' Equations (Thomas 2004).

Results Selected results are presented in Table 1. The pre-calving treatment (Pre) had no significant effect on any of the 24 hour behavioural measures ($P>0.05$). Post-calving treatment (Post) significantly affected all of the behavioural activities, presented in Table 1, with the exception of feeding and total lying time. A low energy diet post calving significantly increased the proportion of total lying time allocated to ruminating, consequently decreasing the time allocated to lying idle. Ruminating/ unit DMI, the number of rests/ day and the average length of a rest were all significantly affected by post treatment ($P<0.001$). Pre and post treatment had no significant effect on total standing time, standing idle, standing ruminating, or on the percentage of lying time spent in the stretched position. Significant correlations were identified between ADEB and all the behavioural activities shown in Table 1.

Table 1 Effect of dietary treatment on behavioural activities and energy balance of dairy cows in early lactation (day 0-140).

	Dietary Treatments					Significance		
	AH	AL	RH	RL	S.E.M	Pre	Post	Interaction
Feeding (% of time)	14.6	17.6	16.8	16.8	1.56	ns	ns	ns
Ruminating/ unit DMI (hr/kg)	0.40	0.54	0.37	0.56	0.008	ns	***	ns
Total lying (% of time)	54.8	50.4	51.4	52.6	3.62	ns	ns	ns
Lying idle (% of time)	32.2	26.8	30.1	28.3	2.21	ns	*	ns
Lying ruminating (% of time)	22.6	23.6	21.3	24.3	2.21	ns	*	ns
No. of rests/ day	9.7	7.3	9.3	7.5	0.65	ns	***	ns
Average length of rest (hr)	1.2	1.4	1.1	1.4	0.10	ns	***	ns

Conclusions The results indicate a significant effect of post-calving treatment (energy density of the diet) on the frequency and duration of behavioural activities. Additional significant correlations between the average daily energy balance and these behavioural activities may allow the identification and development of indicators of energy imbalance which could be implemented in the development of future technologies.

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Estimate of phenotypic and genetic correlations between milk production and udder type traits in Iranian Holstein cows

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Introduction Correlation among type traits and milk production has been investigated by Meyer (1987), Brotherstone (1994) and Misztal *et al* (1992).One of the primary reasons for collecting and utilizing information on type traits is to aid breeders in selecting profitable functional cows for high production and suitable herd life. The objectives of this study were to estimate phenotypic and genetic correlations among milk production and udder type traits.

Materials and methods Production records on 47615 lactations (adjusted for 305-2X (2X refers to the number of milkings in one day)) of 17946 cows of 8 herds from 1991 to 2004, including milk yield (MY), fat yield (FY), fat contents (FC) and protein content (PC), were obtained by Animal Breeding Center of Isfahan Agricultural Jahad Organisation. A total of 4594 cows received a linear score between 1 and 9 for each trait by using individual scorecards. The type traits considered were rear udder height (RUH), rear udder width (RUW), fore udder attachment (FUA), suspensory ligament (SL), udder depth (UD), fore teat placement (FTP), rear teat placement (RTP) and final score (FS). In order to estimate genetic parameters, the animal model of univariate and multivariate analysis of Derivative Free-Restricted Maximum Likelihood (DF-REML) is used.

Results The means of milk yield, fat yield, fat content and protein content were 6786.80 ± 1327.89 (kg), 195.57 ± 43.61 (kg), % 2.91 ± 0.48 and % 3.06 ± 0.35 respectively. The means of RUH, RUW, FUA, SL, UD, FTP, RTP, and FS, were 27.33 ± 3.17, 17.34 ± 2.74, 5.22 ± 1.90, 5.81 ± 1.30, 5.04 ± 1.13, 5.74 ± 1.44, 4.97 ± 1.29 and 34.16 ± 2.69 respectively. The genetic and phenotypic correlations among MY, FY, FC, PC, and udder type traits are given in Table 1. The phenotypic correlations estimated between MY and udder traits is ranged from -0.12 to 0.26 for SL and RUW respectively, that is whereas genetic correlations ranged -0.33 to 0.76 for FUA and RUW respectively. Genetic and phenotypic correlations between MY and with FS is positive.

Table 1 Heritabilites, genetic and phenotypic correlations among production traits and udder type traits

Traits	MY	FY	FC	PC	RUH	RUW	FUA	SL	UD	FTP	RTP	FS
MY	**0.14**	0.80	-0.57	-0.26	0.04	0.76	-0.33	-0.26	-0.10	0.22	0.09	0.59
FY	0.68	**0.21**	0.65	0.45	0.51	-0.13	0.22	0.01	0.23	0.01	0.04	0.02
FC	-0.42	0.38	**0.39**	0.53	0.16	-0.27	0.12	0.14	0.21	-0.12	-0.03	-0.28
PC	-0.22	0.19	0.31	**0.27**	0.11	-0.78	-0.07	0.16	0.02	0.01	-0.51	-0.05
RUH	-0.07	-0.01	0.09	0.04	**0.27**	-0.22	0.06	0.14	0.37	-0.19	0.14	0.22
RUW	0.26	0.16	-0.14	-0.05	-0.19	**0.24**	-0.24	-0.37	-0.17	0.34	-0.07	0.05
FUA	0.00	0.04	0.04	-0.03	-0.06	-0.04	**0.18**	0.12	0.10	0.34	0.05	0.02
SL	-0.12	-0.07	0.08	-0.05	-0.03	-0.13	0.17	**0.26**	0.41	-0.03	0.33	0.25
UD	-0.05	-0.03	0.05	0.02	0.01	0.00	0.20	0.24	**0.19**	0.08	0.18	0.19
FTP	-0.02	-0.01	0.02	0.07	-0.09	0.07	0.25	0.18	0.39	**0.25**	-0.11	0.31
RTP	0.04	0.20	-0.02	-0.02	0.07	0.01	0.07	0.11	0.06	-0.01	**0.36**	-0.03
FS	0.06	0.07	0.00	0.02	0.14	0.04	0.06	0.00	0.03	0.06	0.08	**0.14**

Phenotypic correlations (lower diagonal), genetic correlations (upper diagonal) and heritabilites (on diagonal)

Conclusion Genetic and phenotypic correlations between milk yield and udder type traits in present study demonstrate that selection for udder type traits especially RUW and FTP could be effective for improving milk yield and lay emphasis on RUW and FTP traits for calculation of FS is recommended. Because the genetic and phenotypic correlations of milk production and most udder type traits with FS are positive, therefore if FS is included in selection index for milk yield selection, could improve the milk yield and udder type traits effectively.

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Application of random regression approach in multivariate genetic analysis of lactation milk yield at different first calving ages for Holstein heifers in Khorasan province of Iran

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Introduction Genetic improvement of farm animals is the process of selecting animals of higher genetic merit than average to be parents of the next generation such that the average genetic merit of their progeny will be higher than the average of the parental generation. In practical dairy cow breeding programmes, many traits such as milk production and fitness traits (consisting of health, fertility, calving ease, temperament and length of herd life) are commonly included in breeding objectives among which milk production is the most important trait and also the main determinant of income to dairy farmers. On the other hand, age at first calving is economically important because it determines when an animal begins its productive life and therefore could influence the lifetime productivity of an animal. Moreover, age at first calving can be considered as a measure of heifer fertility performance associated with reproductive efficiency. The main aim of the present research is multivariate genetic analysis of first lactation milk yield at different calving ages of Iranian Holstein heifers in Khorasan province of Iran.

Material and methods Data comprised 17,454 first lactation milk yields (adjusted for 305-day and 2X), spanning 1988-2003, collected from 17,454 Iranian Holstein heifers in Khorasan province (northern east of Iran) in 132 herds. First calving age was restricted from 20 to 35 months and also categorised to 7 classes (Table 1). To estimate genetic parameters for the trait of lactation milk yield at different first calving ages, a random regression animal model was utilised. In the model, fixed effect of contemporary groups of Herd-Year-Season of Calving $(HYSC)_{it}$ and random effect of direct additive genetic (breeding value) were fitted for the t^{th} trait (milk yield as different trait at each age class) of the j^{th} cow (a_{jRt}) in the i^{th} contemporary group. Orthogonal Legendre polynomials (Kirkpatrick *et al*., 1990) up to cubic order (k=4) was also used to take account of genetic variation between animals over different calving ages. The genetic analysis of the records was carried out using **DXMRR** programme (Meyer, 1998) to obtain REML estimates of variance and covariance components between random regression coefficients. The mathematical model was as follows:

$$(milk)_{ijt} = \mu + (HYSC)_{it} + [\sum_{R=0}^{k-1}(\gamma_R * \phi_R(t)) + \sum_{R=0}^{k-1}(a_{jRt} * \phi_R(t))] + ME_{ijt}$$

Results Heritability of first lactation 305-day, 2X milk yield at different ages of calving along with phenotypic and genetic correlations among seven classes of calving ages are presented in Table 1. As shown, heritability of first lactation milk yield varied between 0.288 and 0.491 over different calving ages with the highest and lowest values for the age groups of 3 (25-26) and 7 (33-35) respectively. The results also showed that genetic correlations were generally higher than the phenotypic correlations and ranged from 0.763 to 0.980 indicating that total lactation milk production could be treated more or less as different trait at different first calving ages. The average heritability of first lactation milk yield was 0.37 which is in the range of heritability estimates reported for Holstein populations (Lobo *et al*., 2000).

Table 1 Heritability (diagonal), genetic (under diagonal) and phenotypic correlations (above diagonal) among lactation milk yield at different first calving ages

Age (month)	20-22	23-24	25-26	27-28	29-30	31-32	33-35
20-22	**0.416**	0.305	0.272	0.308	0.367	0.419	0.441
23-24	0.853	**0.307**	0.292	0.282	0.278	0.295	0.338
25-26	0.786	0.980	**0.288**	0.281	0.270	0.272	0.292
27-28	0.868	0.923	0.950	**0.303**	0.321	0.327	0.309
29-30	0.929	0.820	0.823	0.953	**0.374**	0.396	0.358
31-32	0.979	0.802	0.763	0.894	0.976	**0.440**	0.424
33-35	0.975	0.871	0.775	0.799	0.834	0.911	**0.491**

Conclusion The results of the present research indicated that the heritability of total lactation milk yield changed at different first calving ages suggesting that there is an interrelationship between milk yield and freshening age for Iranian Holstein heifers. The heritability estimate of milk yield was found to be the highest (0.491) for heifers calving at 33-35 months which could be of value if increased accuracy of selection of young bulls has to be considered based on their daughters milk record at first lactation.

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Comparison of heritability estimates for herd life in register and non-register Iranian Holstein heifers

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Introduction The main aim of animal breeding programmes is to increase productivity and profitability of farm livestock through genetically improving the economic merit of farm livestock (Smith, 1998). This can be achieved by increasing the mean value of a population for one or several economically important traits by the genetic improvement of the animals in this population. In dairy cow husbandry, many traits of economic importance such as milk production and reproductive traits has long been of interests for breeders to increase profitability of dairy farm enterprise. Herd life, as a complex trait associated with longevity of animals, has been long considered to be related to profitability of dairy herds due to reduce replacement costs and increase the proportion of farm resources used for the milking herd rather than for replacements (Brotherstone *et al*., 1998). In fact, as pointed out by Burnside *et al*. (1984), lifetime profitability is affected by many factors such as production per lactation, length of productive life, age at first calving, calving interval as well as input and output prices. The main objective of the present study was to estimate heritability of herd life for register and non-register Iranian Holstein heifers.

Material and Methods In this study, a total of 39310 herd life records obtained from 39310 Iranian Holstein heifers calved between 1991 and 1999 and distributed in 260 herds of Iran was used. Data were initially divided into 3 files. File 1 consisted of register cows (100% Holstein gene), file 2 consisted of non-register cows (<100% Holstein genes) and file 3 was total data set for all cows. In the present study, herd life (**HL**) was defined as the interval between first calving date and culling date of individual cows. An animal model was utilized to estimate variance components of additive genetic and residual random effects for the corresponding trait. In the animal model, fixed effect of contemporary groups of Herd-Year-Season Of Calving $(HYSOC)_i$, linear and quadratic (non-orthogonal partial regression coefficients) covariate of age at first calving (A_{ij}) and random effect of additive genetic (a_{jj}) were fitted for the trait (Y_{ij}) of herd life. Univariate analysis was undertaken by **DFREML** programme (Meyer, 1988) to obtain restricted maximum likelihood (**REML**) estimate of variance components. The mathematical model was as follows:

$$Y_{ij} = \mu + (HYSOC)_i + \sum_{R=1}^{2} \beta_R * (A_{ij} - \overline{A})^R + a_j + e_{ij}$$

Results Means, phenotype variances as well as REML estimates of heritability for HL are presented in Table 1. As it is shown in the table, although mean HL for non-register cows was significantly statistically greater than mean HL for register cows and heritability of HL for register cows was greater than heritability HL for non-register cows.

Table 1 Phenotypic variances and heritability estimates of herd life in different data files

Data set	No. records	No. progeny	No. HYS	No. sires	Mean HL	Phenotype variance	h^2(SE)
Reg. cows	8811	8811	1924	297	701.1**a**	139177.8	0.04** (0.016)
Non-Reg. cows	30499	30499	4848	420	729.6**b**	149251.3	0.01* (0.006)
Total data	39310	39310	5603	457	723.2	146035.2	0.01** (0.005)

* significant at P<0.05, ** significant at P<0.01, a,b indicates the means are different from each other statistically

Conclusion The results of the present study indicated that the heritability estimate of herd life for register Iranian Holstein cows was higher than that of non-registers. Moreover, in accordance with other previous research such as Brotherstone *et al*. (1998), the heritability estimate for herd life obtained in the present study was very low. Based upon the definition made for the herd life in this study, low heritability estimate obtained for herd life indicates that there is a greater environmental variation as compared to genetic variation suggesting that a significant genetic improvement in this trait may not be made through selecting animals with higher performance for herd life. The present research also showed that mean herd life for non-register cows was statistically greater than that obtained for register cows.

Acknowledgements The Centre of Animal Breeding, Iran is acknowledged for supplying the data used in this study.

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EM-REML estimation of phenotypic and genetic relationships between 305d-2X-ME milk production traits in Iranian Holstein heifers

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Introduction In animal breeding programmes knowledge of the genetic properties of the traits under consideration is the first prerequisite in establishing a selection criterion. Several methods such as ANOVA, ML and REML have been utilised to estimate variance and covariance components in animal breeding data which are usually originate from selection experiments. Modified ML procedure which is so-called restricted maximum likelihood (REML) has been widely used due to its statistical desirable properties. To apply the REML method, a number of algorithms such as DF-REML, EM-REML and scoring have been developed to maximise likelihood function in order to estimate variance and covariance components. Expectation-maximisation (EM-REML) is an algorithm in which first derivatives of the likelihood needs to be evaluated numerically or analytically. The main objective of the present study was to estimate phenotypic and genetic associations among milk, fat and protein yields as well as fat and protein percentages in Iranian Holstein heifers based upon applying the EM-REML method through using a multiple trait animal model.

Material and methods The data set used in this study comprised a total of 18,989 first lactation records on milk production traits including milk, fat and protein yields as well as fat and protein percentages which were converted to 305-day, 2X (twice a day milking) and mature equivalent by using appropriate adjustment factors developed by Animal Breeding Centre of Iran. These records were collected from 18,989 Iranian Holstein heifers first calved between 1995 and 2001 in 202 herds from 7 different provinces over the country. First calving age was restricted to be between 18 and 40 months with the average 26.22 months. The number of sires and dams were 714 and 16287 respectively. To estimate genetic parameters for the trait of interest, a multiple trait animal model was applied. In the model, fixed effect of contemporary groups of Province-Herd-Year-Season of Calving $(PHYSC)_{it}$, linear covariate of Holstein genes, random effect of direct additive genetic (breeding value) were fitted for the t^{th} trait of the j^{th} cow in the i^{th} contemporary group. Genetic and environmental variance and covariance components were estimated based upon the statistical method of restricted maximum likelihood (REML) through expectation-maximisation (EM) algorithm implemented in **MTC** programme (Misztal, 1994). The mathematical model was as follows:

$$(Trait)_{ijt} = \mu + (PHYSC)_{it} + [b(HF_{ijt})] + (Cow)_{jt} + e_{ijt}$$

Results Heritability, genetic and phenotypic correlations between five traits studied in this research are presented in Table 1. The lowest and highest heritability estimates were found for protein yield and its percentage respectively. As expected, milk yield had positive genetic and phenotypic correlations with other yield traits while it was negatively correlated with fat and protein percentages. Although fat yield was positively correlated with protein yield and fat percentage, it was negatively correlated with protein percentage at phenotypic and genetic levels. Protein yield had genetically and phenotypically antagonist relationships with fat and protein percentages. Milk components were found to be highly positively correlated at both genetic and phenotypic levels. The genetic parameters found in the present study were in the range of those reported by Lobo *et al.* (2000) for Holstein populations.

Table 1 Heritability (diagonal), genetic (above diagonal) and phenotypic (under diagonal) correlations among 305d-2X-ME milk production traits

Trait	Milk yield	Fat yield	Protein yield	Fat %	Protein %
Milk yield	0.22	0.64	0.90	-0.54	-0.59
Fat yield	0.72	0.21	0.77	0.29	-0.01
Protein yield	0.94	0.75	0.19	-0.27	-0.20
Fat %	-0.52	0.18	-0.40	0.26	0.74
Protein %	-0.56	-0.25	-0.27	0.54	0.27

Conclusion The results obtained in the present research revealed that the heritability of mature equivalent protein percentage was generally higher than the other traits under consideration suggesting that there is a meaningful genetic variation between animals to be directly selected based upon this trait. High positive genetic correlation between milk and protein yields reflects the fact that the same genes acting on two traits indicating that when direct selection is practised on milk yield, favourable correlated response to selection is made for protein yield which could be of great nutritional importance. However, since fat and protein percentages are usually of economic value in dairy farm enterprise (Simm, 1998), a selection index could be made to incorporate relative economic values as well as taking account of genetic and phenotypic correlations between the traits to be used in selection programmes.

Acknowledgements The Centre of Animal Breeding of Iran is acknowledged for supplying the data used in this study.

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Association of leptin polymorphisms with production and reproduction traits in Iranian Holstein dairy cows

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Introduction Leptin is a 16-kDa protein that is synthesized by adipose tissue and is involved in regulation of feed intake, energy balance, fertility and immune functions. It has been shown that leptin gene influences milk performance in cattle (Liefers *et al.*, 2002). Leptin is related to both energy metabolism and reproduction and it was shown that leptin polymorphisms had significant effect on calving interval and weight at first calving in beef cows (Almeida *et al.*, 2003). In cattle, the leptin gene is located on chromosome 4 and consists of three exons. The aim of this study was to indicate polymorphism at the bovine leptin gene locus in Iranian Holstein dairy cows and its contribution to the first cumulative 60-d milk yield, 305-d milk yield, days to first breeding (DFB), days open (DO), and days from first breeding to conception (DFBC) in the previous lactations.

Materials and methods In total, blood samples were collected from two hundred and thirty eight Iranian Holstein cows via venipuncture from coccygeal vessels. DNA extraction was done on the blood samples using guanidium thiocyanate-silica gel. PCR-RFLP method was used to detect the polymorphism of a 423 bp fragment from intron 2 of leptin gene. For the genotyped cow, the first cumulative 60-d milk yield, 305-d milk yield, DFB, DO, and DFBC using previous lactation records were also analyzed. For DFB and DO, the follow-up period started at 35 d after calving and ended at 200 and 365 d after calving, respectively. The DFB was defined as the interval from 35 d after calving to first breeding or end of follow-up period, whichever occurred first. The DO was defined as the interval from 35 d after calving to conception or end of follow-up period, whichever occurred first and the DFBC was the interval between DFB and DO. PopGen32 software was used to estimate the allele and genotype frequencies. Data from previous lactations were analyzed using Standard Least Square within mixed models using JMP software (version 4.0.4; SAS Institute Inc, NC. USA). Apart from the genotypic effect, the fixed effects were year, season, parity, and age at calving, and sire. For reproductive traits the cumulative first 60-d milk yield was also added to the model. Animal was fitted as a random effect.

Results Two genotypes, AA and AB, were distinguished which had the frequencies of 0.89 and 0.11, respectively. The genotypes were distributed according to the Hardy and Weinberg equilibrium ($P<0.05$). The results from previous lactations showed that the 305-d milk yield was affected by leptin polymorphism and the heterozygous produced more milk than the homozygous cows ($P<0.05$, Table 1). The 305-d milk yield was also affected by year, season, and parity at calving, and sire ($P<0.01$). The first 60-d cumulative milk yield was similar between two genotypes ($P=0.21$, Table 1) and tended to be higher in the heterozygous cows. The first 60-d cumulative milk yield was also impacted by year and parity at calving, and sire ($P<0.01$).

Table 1 Effect of the RFLP genotypes on milk production using previous lactation records.

	Genotypes		
Item	AA	AB	P
The first cumulative 60-d milk yield, kg	2288	2377	0.21
The 305-d milk yield, kg	9479	9550	0.03

There were no significant effects of genotype on reproductive performance (Table 2). The DFB was affected by sire, season and year of calving and age of cow at calving ($P<0.05$). The DO was influenced by sire, DFB and age at calving ($P<0.05$). Sire and DFB had significant effects on DFBC ($P<0.05$).

Table 2 Effect of the RFLP genotypes on reproduction traits using previous lactation records.

	Genotypes		
Item	AA	AB	P
Days from parturition to first breeding, d	71.04 ± 8.8	67.97 ± 12.2	0.77
Days open, d	126.75 ± 18.7	119.86 ± 28.1	0.79
Days from first breeding to conception, d	50.23 ± 20.7	46.18 ± 28.7	0.49

Conclusion The results of this study showed that leptin polymorphism was associated with 305-d milk production. It seems that breeding programs favouring the B-allele can yield a higher 305-d milk production without negatively affecting fertility.

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Comparison of milk production, mastitis, lameness and days to conception of Brown Swiss, Holstein and Holstein cross Brown Swiss dairy cattle

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Introduction Mastitis is a complex disease causing inflammation of the udder, which has been estimated to cost the dairy farmer between £40-£117/cow per year (Stott *et al.,* 2002). Economic loss occurs as a result of discarded milk, reduced milk yield and milk quality, increased vet costs and an increase in replacement costs. The objective of this study was to examine the effect of breed on the incidence of mastitis and somatic cell counts and milk production capabilities of Holstein Friesian, Brown Swiss and Brown Swiss crossbred cows.

Materials and methods The performance of differing dairy breeds were compared using a total of 41 dairy cattle; 13 Brown Swiss (BS), 14 Holstein Friesian (HF), 14 Holstein Friesian cross Brown Swiss (BSX) dairy cattle. The animals were matched according to calving date and allocated at parturition into groups according to breed. All the experimental animals were housed, milked and fed as a single group and observations were recorded for a total of 122 days. The animals were allowed *ad libitum* access to grass and maize silage (50:50 DM basis) and offered dairy compound during milking at a rate of 0.25kg/l to a maximum of 6kg/h/d. Individual milk yield, milk composition, somatic cell count, locomotion score, lameness and body condition score was measured weekly. The incidence of clinical mastitis was recorded and pathogen(s) present and antimicrobial susceptibility were assessed. Days to conception was measured using milk progesterone levels by using enzyme linked immunosorbant assay (ELISA). The data was found to be normally distributed and was analysed using ANOVA, GLM (Minitab 13) with breed as a fixed effect. The existence of significant differences was assessed using Tukey's test. Discrete data including the counts of days to conception, pathogen incidence and occurrence were analysed by Chi-square and significant differences were determined using Chi-square tables.

Table 1 Mean milk yield, fat corrected milk yield, milk fat, milk protein, total milk fat, total protein and somatic cell count for Brown Swiss, Brown Swiss cross and Holstein Friesian dairy cattle

	Brown Swiss	Brown Swiss cross	Holstein Friesian	SEM	SIG
Mean milk yield (kg)	17.2^{c}	19.0^{b}	23.1^{a}	0.17	**
Fat corrected milk yield (kg) †	19.0^{c}	19.7^{b}	23.1^{a}	0.16	**
Mean milk fat (g/kg)	44.9^{a}	41.8^{b}	40.1^{c}	0.20	**
Mean milk protein (g/kg)	34.3^{a}	32.3^{b}	30.7^{c}	0.10	**
Total milk fat (kg/d)	0.76^{c}	0.79^{b}	0.92^{a}	0.06	**
Total milk protein (kg/d)	0.60^{c}	0.61^{b}	0.70^{a}	0.04	**
Somatic cell count (00,000/ml)	54^{a}	43^{c}	50^{b}	0.1	**
Clinical mastitis (incidence)	2	3	10	-	**
Mean locomotion score (scale 1 to 5)	1.2^{b}	1.3^{a}	1.2^{b}	0.03	**
Body condition (scale 1 to 5)	2.91^{a}	2.44^{b}	2.14^{c}	0.02	**
Days to conception	116	86	114	7.42	NS

** - Row means with different superscripts differ at P<0.05 † - Milk yield corrected to 40.0 g/kg

The mean milk yield and fat corrected milk yields were significantly higher from HF, followed by BSX and BS cows. Milk fat and protein concentration was highest in milk from BS, followed by BSX and HF cows. The total milk fat and protein yields were highest from HF followed by BSX and BS cows. Geometric mean SSC was significantly lower in milk from BS followed by HF and BSX cows. Locomotion was significantly higher in BSX cows, but similar in HF and BS cows. Body condition was significantly higher for BS, followed by BSX and HF. The days to conception was not significantly different between breeds. The incidence of clinical mastitis was significantly higher for HF cows compared with BS and BSX, with no significantly difference between BS and BSX cows. The pathogens isolated were *E. coli* (EC)*, Staph. aureus* (SA) *and Strep. uberis* (SU). BS cows had no SA, BSX cows had no SU or EC, while HF cows had all three pathogens EC, SA and SC.

Conclusions HF cows had significantly higher milk yields, incidence of mastitis and had all three (EC, SA and SU) mastitis pathogens isolated from cows with clinical mastitis. HF cows had the lowest milk fat and protein concentration, but the highest total milk fat and protein production levels. BS cows had significantly lower milk yields, with significantly higher milk fat and protein contents. However, total milk fat and protein production was significantly lower than HF and BSX cows. BS and BSX cows had low incidence rates of clinical mastitis and BSX did not contract SU, while BS did not contact SA. Locomotion score and lameness, days to conception were not significantly different between BS and HF cows. Higher milk and milk solid yield levels from HF cows were associated with higher levels of clinical mastitis. This may reduce milk value due to payment structures, increase the cost of both treatment and culling compared with BS and BSX. The performance of BSX cows tended to be intermediate between HF and BS cows, which is not indicative of the existence of heterosis in HF cross BS cows.

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Genetic polymorphism of the Mongolian cattle kappa casein gene

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Introduction K-Casein (CSN3) is a phosphoglycoprotein that constitutes approximately 12% of the casein complex of bovine milk. The amount and type of CSN3 present in milk vary considerably depending on the individual cow. The use of small quantities of 2-mercaptoethanol in electrophoretic separation led three independent groups to the discovery of two distinct bands of protein denoted as A and B corresponding to CSN3. Later studies showed that the presence of these bands were due to genetic polymophism of this protein. Variant B differs from A by having isoleucine and alanine at positions 136 and 148, respectively, instead of threonine and aspartic acid in the 169 amino acid sequence. In the most cattle populations, there is a predominance of the A over the B type of CSN3 (Ng-Kwai-Hang *et al.* 1991). However, it has been demonstrated that CSN3 B would be more desirable than CSN3 A because this form is associated with higher casein content in milk, higher cheese-yielding capacity, more favorable cheese composition and better coagulating properties in terms of rennet clotting time and curd firmness (Ikonen *et al.* 1999). Therefore, it seems desirable to increase the frequencies of B type of CSN3 through appropriate selection and breeding schemes. This paper reports the genotyping for CSN3 gene and his allele frequencies done for 53 animals of Mongolian breed. Because, heretofore genotyping of CSN3 gene for this breed has not reported, moreover milk of this animals used in cheese industrial.

Materials and methods A total of 53 Mongolian cattle DNA samples were used in this study. DNA was isolated from blood samples using standard protocols. The PCR was used to ampllfy a 99-bp region of the CSN3 gene that contains the nucleotide substitutions diagnostic of the A or B allele as previously described. For PCR we were used GenePak^{TM} PCR Core kit (Moscow) and two primers: SGE, 5'-TATCATTTATGGCCATTCCACCA-3' and SGO, 5'-CTTCTTTGATGTCTCCTTAGAGTT. The PCR was performed in a total reaction valume of 25 µl and about 100-150 ng of DNA. The PCR thermal profile was as follows: the first cycle of denaturation at 92-94˚C for1.5 min, annealing primers at 55-58˚C for1.5 min, and elongation at 70-74˚C for 2 min. in all 30-35 cycles were conducted and using a Go-well thermal cycler (MJ Research). The amplified DNA fragments were digested with HinfI (A-allele specific), TaqI (B-allele specific) under standard conditions. Digestion products were separated by electrophoresis in 6% polyacrylamide gel, stained with ethidium bromide, and analyzed under UV light.

Results We screened 53 Mongolian cattle to detect the polymorphisms at kappa-casein (CSN3) gene using the polymerase chain reaction. CSN3 A and B alleles were identified by PCR-RFLP using the restriction enzymes that detect the underlying nucleotide changes at codon 136 (Taql) and at codon 148 (HindIII or HinfI) in Mongolian cattle. Among the 53 animals that were genotyped directly from the DNA of blood samples using the PCR, 18 (34%), 26 (49%), and 9 (17%) animals were of the AA, AB, and BB types for CSN3, respectively (table 1). Genotypes of animals were assigned based on a chi-square test between the significance of $P < .01$ and obtained $\chi^2_{(2,\,0.01)}$ was 0.0025 that is very lower than table $\chi^2_{(2,\,0.01)} = 9.21$.

Conclusion Data obtained from this study suggest that the frequency of B allele for CSN3 in this breed is high (41.5%). Because of the association of CSN3 B with more favorable milk characteristics, the dairy industry, especially the cheese maker, has shown great interest in increasing the frequency of the B allele in the dairy cattle population. With faster and more direct techniques now available for genotyping of bulls, it is recommended that a larger population of bulls (both proven and unproven) be typed for CSN3. Due to the high demand for CSN3 B milk by the dairy industry, it might be advantageous to increase this allele in the dairy cattle population by identifying sires with homozygous CSN3 B and using them more frequently as service sires.

Table 1 allele and genotype frequencies and chi-square test

genotype	number o	number e	frequency	Allele frequencies
AA	18	18.13	0.34	A: 58.5%
AB	26	25.73	0.49	
BB	9	9.12	0.17	B: 41.5%

$\chi^2_{(2,\,0.01)} = 0.0025 << 9.21$

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Study of some suggested measures of milk yield persistency and their relationships

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Introduction The persistency of dairy milk yield, which refers to the degree to which milk yield is maintained from month to month by the cow during her lactation, is an economic trait. The knowledge of persistency is important for herd management and selection strategy. The benefits of selecting for lactations that are more persistent were only speculative, because very little research has been published. As the time goes by, the more is learned about this trait and its relationship with other important traits. The persistency can be defined in several ways, as either ratios of yield in different parts of lactation period, to be derived from factors in lactation curves models as proposed by Wood (1967) or a simple statistical parameter computed from test-day yield records(3). The incomplete gamma function, which described by Wood is one of the most popular models used to describe the lactation curve: $y_t = at^b e^{-ct}$ where y_t = milk yield on day t, a= a parameter to represent yield at the beginning of lactation, b and c are factors associated with the inclining and declining slope of the lactation curves, respectively. The objectives of this study were to estimate genetic and phenotypic parameters for different criteria of persistency, finding correlation between them and their relationships with 305-days milk yield, by applying REML under animal models in the Holstein dairy cattle of Iran.

Material and methods The data set contained 349668 test-day records of 36487 cows in 157 herds. Five criteria of persistency were defined such as: The maximum production of all test-day records per lactation divided by the related mean, and denoted by MAME, Standard deviation of all test-day records per lactation denoted by STD, The percentage of daily milk retained from the peak to the end of lactation denoted by LAMA, The latest test-day record yield divided by the mean of all test-day records yields and denoted by LAME, The criterion that was proposed by Wood and was calculated from parameters of incomplete gamma function that was fitted to test-day yield records of each cow separately and denoted by WE. In order to estimate variance components by applying animal models, the DFREML software was used (1). In addition, to define applied model, the suggested model in literatures was used (3)

Results Estimates of variance components of 305-days milk yield and persistency criteria by univariate models were given in Table.1. The estimates of heritabilities for persistency criteria were low (0.0469 to 0.0819 to) but in close agreement with previous study using data from Iran (2). Among all persistency criteria, the highest heritability was estimated for STD and the lowest was estimated for LAME. Genetic correlation between persistency and milk yield ranged between 0.6617 to -0.6124 and genetic correlation between persistency traits were ranged between 0.9988 to -0.9265. Phenotypic correlation had similar trend to genetic Correlation for all cases but in some cases, phenotypic correlation is very lower than genetic correlation (result not shown).

Table 1 Estimates variance components and heritabilities along with their standard errors (se (h^2)) for305-days milk yield and persistency criteria.

Parameter	305-days milk yield	STD[1]	MAME[2]	LAMA[3]	LAME[4]	WE[5]
σ^2_p	905922.30	1.96	0.01	143.40	0.012	0.32
σ^2_a	287832.70	0.16	0.0008	9.48	0.0006	.025
σ^2_e	618089.60	1.80	0.01	133.92	0.01	0.30
h^2	0.31	0.08	0.07	0.06	0.04	0.07
se(h^2)	0.01	0.01	0.009	0.008	0.008	0.01

1. Standard deviation of all test-day records per lactation, 2. The maximum production of all test-day records per lactation divided by the related mean, 3. The percentage of daily milk retained from the peak to the end of lactation timed by 100, 4.The latest test-day record yield divided by the mean of all test-day records, 5. The criterion that was proposed by Wood (1976) and was calculated as, s=-(b+1) ln (c) which b and c are parameters of incomplete gamma function.

Conclusions Among all criteria that were considered by this research, WE and MAME had highest absolute genetic correlation with milk yield and seemed to be favourable for breeding programs and selection strategy. Also a similar situation but to a smaller extent power were for LAMA criteria. However, it seemed that STD is not a suitable criterion for persistency of milk yield. However, more research is needed to apply this trait in breeding program.

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Analysis of Kappa casein polymorphism in three Iranian native cattle and Holstein breeds by PCR-RFLP

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Introduction Kappa Casein (□-casein) is a protein in mammalian milk whose two common genetic variants of A and B are well known. The A variant has a Thr and Asp at positions 136 and 148, while the B variant has Ile and Ala in these positions. Difference at position 148 can be detected by PCR-RFLP (Medrano and Agiular-Cordova, 1990). These two alleles determine great differences in milk, casein, protein and fat yields as well as cheese making properties such as coagulation time and curd firmness, with a superiority for cheese production of the κ-casein BB compared to κ-casein AA milk (Lodes *et al.,* 1996). Several internationally studies have been focused on determining the frequency distribution of the various milk protein alleles in different animals. It is important to study not only the major breeds but also those with small animal numbers (Erhardt, 1996). The aim of this study was to estimate the allelic frequency in polymorphic site of exon 4 of κ-casein gene in three Iranian native and Holstein cattle.

Materials and methods A total of 406 genomic DNA was extracted from two Iranian native *Bos indicus* Mazandarani (n = 97) and Golpaygani (n = 112), and one *Bos taurus* Sarabi (n = 87) cattle and Holstein (n = 110). DNA was extracted from whole blood by the procedure of Miller *et al*. (1988). The selected primers (Medrano and Cordova, 1990) were used to amplify a 350 bp fragment of exon 4. DNA was amplified in a total volume of 25 µL containing 50 ng genomic DNA, 0.2 µM of each primer, 0.2 mM dNTP, PCR 1x buffer, 2.5mM $MgCl_2$, and 1 unit Taq DNA polymerase (GENAXXON, Germany). PCR condition was 94 °C for 2 minutes, 56 °C for 30 seconds, 72 °C for 30 seconds and finally 72 °C for 5 minutes. Amplification was done for 35 cycles followed by 72 °C for 5 minutes. PCR product was digested with 5 unit *HinfI* (New England BioLabs, USA) in a total volume of 20 µl. The digested products were loaded on 2% agarose gel containing Ethidium-Bromide and genotyped under UV light.

Results According to κ-casein sequence (GenBank X14908), there are two restriction site for *HinfI* enzyme (GAnTC) in the 350 bp fragment. The permanent site leads to a restriction of the 350 bp product to two fragments with a length of 266 bp and 84 bp. If a polymorphic site exists, the 266 bp can be cut into 132 and 134 bp. Thus, homozygous AA genotype digested with *HinfI* endonuclease enzyme showed 3 fragments with the length of 134, 132, 84 bp, heterozygous AB with 4 fragments of 266, 134, 132, 84 bp and homozygous BB genotype 2 fragments with 266 and 84 bp. The gene frequencies were calculated by counting method as $p = \frac{2(BB) + (AB)}{2N}$, q = 1 - p ; where p= the gene frequency of allele B and q= the gene frequency of allele A. The allele frequencies of B variant were 0.35, 0.57, 0.32 and 0.55 for Mazandarani, Sarabi, Golpaygani and Holstein, respectively. Differences of genotypic frequencies between each two breeds were calculated by Chi-Square test. Results showed significant differences between native indicus breeds, Mazandarani and Golpaygani genotype frequencies compared with Holstein, but there is no significant difference between Sarabi and Holstein (Table 1).

Table 1 Significance levels of the Chi-Square test of genotype frequencies between different breeds

	Holstein	Golpaygani	Sarabi
Mazandarani	0.01	ns	0.01
Sarabi	ns	0.01	****
Golpaygani	0.01	****	
Holstein	****		

Conclusion The B variant of κ-casein showed higher frequency in taurin than in indicus population. The differences in allelic frequencies between native indicus breeds and Holstein might be due to different genetic lineage, selection plans applied to Holstein population for improving milk production traits, low number of samples, or effect of natural selection in native breeds. Regarding to high frequency of B variant in Sarabi breed, it seems this breed has a good potential for cheese making in dairy industry in the Northwest of Iran.

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Polymorphism in gene leptin and its relationship to milk production and reproduction traits in Brown Swiss cows

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Introduction Leptin is 16 KD, a protein that is synthesized by adipose tissue, and it is involved in the regulation of feed intake energy balance, fertility and immune function. In cattle, the leptin gene is located on chromosome 4. It consists of 3 exons and 2 introns and of those only 2 exons translate into protein. Leptin treatment of animals has been shown to cause a decrease in food intake, body weight loss, fat depot weight loss and increase in energy metabolism therefore leptin not only causes a reduction in food intake but the potential body weight losses are enhanced due to an increased metabolic rate. Leptin may be involved in regulating reproduction in that if may also act as the signal to the reproductive system that sufficient body fat exists to support a successful conception and pregnancy. Lindersson *et al* (1998) reported QTL for milk production traits close to the leptin gene (82.8 cm). Liefers *et al*. (2002) reported that heifers with the sau3IA – AB genotype produce 1.32 kg/d more milk and consume 0.73 kg/2 more food compared with the sau3AI - AA genotype. They suggested that RFLP – B alleles could yield a higher milk production without negatively affecting energy balance and fertility. The aim of this study is to estimate the frequency of the leptin gene in Brown Swiss cows and the relationship between polymorphisms at the bovine leptin gene locus with variation in milk production and reproduction traits.

Material and methods Blood samples were collected from one hundred and four Brown Swiss cows. DNA extraction was done on the blood samples using guanidium thiocyanate-silica gel. A strategy employing polymerase chain reaction was used to amplify a 422 bp fragment from blood DNA. PCR conditions were 2.25mm $Mgcl_2$, 200µm dNTP, 1µm of each primer, 50- 100 ng of genomic DNA and 0.2 Taq DNA polymerase. The first cycles were 3 min at $94\,^{\circ}c$ followed by 35 cycles of 45 sec at $94\,^{\circ}c$ 45 sec at $59.5\,^{\circ}c$, 45 sec at $72\,^{\circ}c$ and ending with a 10 min extension phase at $72\,^{\circ}c$. PCR product for each sample was digested with 5 units of *Bst*MB1 at $67\,^{\circ}c$. The data about production and reproduction traits was collected from the Brown Swiss cows and analysed with JAMP 4/0/4. Traits in this study consisted of milk production in 60 and 100 days, days open and days milk.

Results Digestion of amplicons with *Bst*MB1 revealed two alleles: Allele A had 2 fragments 390 and 32bp and allele B had 3 fragments 303, 88 and 32. Frequencies of genotypes were 0.64, 0.36, 0.01 for AA, AB and BB respectively. Allelic frequencies were 0.82 and 0.18 for A and B, respectively. The populations were in Hardy-Weinberg equilibrium. The milk production and reproduction traits were affected by the genotypes. The AB genotype had a significant effect ($P<0.01$) on milk production. Also the AA genotype had a significant effect ($P<0.05$) on reproduction traits than the other genotypes. According to above result, Liefers *et al*. (2002) reported that heifers with the sau3IA – AB genotype produced more milk compared with the sau3AI - AA and BB genotypes. They suggested that RFLP – B alleles could yield a higher milk production without negatively affecting energy balance and fertility.

Table 1 Least sq mean، std error and t student different genotypes leptin gene about traits milk production in 60 and 100 days ،days open and days milk.

Source variation	Days milk	Days open	Milk 100 days	Milk 60 days
AA	370/62±16/88[a]	189/23±18/6[a]	2877/96±65/32[a]	1722/36±24/8[a]
AB	341/53±21/74[a]	160/04±18/14[b]	3008/77±77/91[a]	1724/64±28/44[a]
BB	181/03±45/89[b]	172/44±26/43[b]	2363/36±132/31[b]	1433/52±56/06[b]

Conclusion The AB genotype had high positive significant effect on milk production 60 and 100 days and the AA genotype had a positive significant effect on days open and days milk.

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Reproductive performance of Holstein, Norwegian and crossbred dairy cattle

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Introduction Declining fertility is a major problem on dairy farms throughout the United Kingdom, with a recent farm survey identifying infertility as the primary reason for culling dairy cows in Northern Ireland (Mayne *et al.,* 2002). This decline in fertility is often attributed to the focus on milk production within the Holstein-Friesian breed, and as a consequence, there is a growing interest in alternative dairy cattle breeds and in the use of crossbreeding. Norwegian dairy cattle (NRF) have been evaluated at the Agricultural Research Institute of Northern Ireland since 2000, with a recent on-farm study indicating a significant improvement ($P<0.05$) in conception to 1st service with NRF compared to Holstein dairy cattle (Ferris *et al.,* 2004). This study examines the reproductive performance of Holstein and Norwegian cattle, and of crossbred animals of these two breeds.

Materials and methods The study involved 114 primiparous spring calving animals, 37 in 2003, 42 in 2004 and 35 in 2005. These comprised 38 Holstein (Hol) animals (13, 12 and 13 in 2003, 2004 and 2005 respectively), 23 NRF animals (6, 8 and 9), 33 Hol x NRF crossbreds (9, 13 and 11) and 20 NRF x Hol crossbreds (9, 9 and 2). Animals had mean calving dates of 15 February (s.d., 28.0 days), 4 February (s.d., 22.7 days) and 1 February (s.d., 27.6 days) in 2003, 2004 and 2005 respectively. Post-calving all animals were offered grass silage *ad libitum*, with concentrates offered at 5.0 - 6.5 kg/cow/day, according to silage quality. Animals were turned out to grass on 24 February, 24 March and 18 March in 2003, 2004 and 2005, with full-time grazing beginning on 28 April, 15 April and 4 May in 2003, 2004 and 2005 respectively. Within each grazing season, animals were managed on a study examining various aspects of grazing management, with breeds balanced, as far as possible, across grazing treatments. Concentrate feed levels during full time grazing ranged from 2 – 4 kg/cow/day, across the three years. All animals were bred by artificial insemination (AI), with the breeding season finishing on the 30 June each year. No animal was inseminated before day-42 post-calving, or treated with fertility drugs prior to day-52 post-calving. Conception rates were determined from pregnancy diagnosis at 32 days (minimum) after the last service. Milk progesterone was monitored twice weekly from all animals until day-52 post-calving using an enzyme-linked immuno-sorbent assay (ELISA) kit, with the commencement of luteal activity defined as two consecutive rises above 3ng/ml. The interval from calving until the commencement of luteal activity was determined, as was the proportion of animals commencing luteal activity pre day-42 post-calving. Breed effects on fertility were analysed using a binomial model based on accumulated analysis of variance.

Results Breed had no significant effect on the interval to 1st observed oestrus, the number of animals observed in oestrus pre day-42, the number of services/animal, or the conception rate to 1st service ($P>0.05$). However, conception to 1st and 2nd service was significantly lower for Holstein animals compared to animals of any other genotype ($P<0.05$). All parameters measured were similar for Hol x NRF and NRF x Hol, with all progesterone analysis being similar across all breeds. Animals of the Holstein breed had a significantly lower pregnancy rate at the end of the breeding season compared to NRF x Hol crossbred animals. Breed had no significant effect on the interval to first service or on the numbers of animals that were in-calf by 100 days post-calving ($P>0.05$).

Table 1 Breed effects on fertility parameters (proportion basis, unless stated otherwise)

	Hol	NRF	HolxNRF	NRFxHol	SEM	SIG
Interval to 1st observed oestrus (days)	48	43	49	35	4.1	NS
Animals observed in oestrus pre day 42	0.48	0.58	0.45	0.67	0.096	NS
Number of services/animal	1.76	1.97	1.56	1.35	0.163	NS
Conception to 1st service	0.37	0.39	0.60	0.59	0.094	NS
Conception to 1st and 2nd service	0.50^{a}	0.77^{b}	0.78^{b}	0.87^{b}	0.076	*
Interval to 1st service (days)	73	70	75	74	3.8	NS
Pregnancy rate at end of breeding season	0.66^{a}	0.86^{ab}	0.78^{ab}	0.92^{b}	0.069	*
In-calf rate at 100 days post-calving	0.46	0.61	0.60	0.70	0.092	NS

Means with differing superscripts are significantly different ($P<0.05$)

Conclusions Although 'days to the onset of cyclicity' was unaffected by breed, conception rates were significantly lower with the Holstein breed, with only half of the animals conceiving after two services. The results of this study confirm the improved conception rates with animals of the NRF breed compared to animals of the Holstein breed, in agreement with the observations of Ferris *et al*. (2004). Due to the numbers of animals involved in this study, it is not possible to quantify the extent of heterosis. However, the data does provide some indication that further improvements in reproductive performance can be achieved through crossbreeding.

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Estimation of calving difficulty (Co)variance components of Iranian Holstein heifers using a sire-maternal grandsire threshold model

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Introduction Calving difficulty, biologically and statistically, have special characteristics which require researchers to use more complex models for its analysis. First, it is influenced by maternal effect and therefore the models should take this aspect into account. This is why animal models with maternal effect or sire-maternal grandsire models are employed. Secondly, it is recorded subjectively using usually five scores, one for a normal delivery to five for a Caesarean parturition. In other words, calving difficulty is a discrete trait and is not distributed normally. In theory, methods for analyzing continuous data are not appropriate for categorical data. In the 1980s, researchers developed a standard threshold model (a nonlinear model) for genetic analysis of these traits (Harville *et. al*. 1984). In the threshold model each phenotype in the categorical scale is associated with an unobservable underlying continuous variable with normal distribution. The objective of this study was to estimate (co)variance components of calving difficulty and genetic parameters using a threshold model.

Materials and methods Calving records were obtained for Iranian Holstein calves born between 1990 and 2004 from the Animal Breeding Centre. Records that showed stillborn, twinning, unrecorded sire and calf birth weight were removed along with those relating to sires with less than 10 calves and dams aged more than 30 months. This left 81376 observations. A pedigree file was made using all the available relationship information. Significant factors on calving difficulty were determined by PROC GLM of SAS. A mixed model with the below equation was used to analyze data, containing age of dam by sex of calf(A-S), season of calving(Se) effects as classes, and calf birth weight(Bw) as covariate, and sire of the calf(S), maternal grandsire of the calf(Mgs), and herd by year(H-Y) as random effects:

$$Y_{ijklmno}=S_i+Mgs_j+H\text{-}Y_k+A\text{-}S_l+Se_m+Bw_n+e_{ijklmno}$$

in which $Y_{ijklmno}$ and $e_{ijklmno}$ represent calving score and random residual effect respectively. (Co)variance components for direct(σ^2_d) and maternal(σ^2_m) effect can be obtained from those due to sire(σ^2_S) and maternal grandsire(σ^2_{Mgs}) with the following equation:

$$\begin{pmatrix} \sigma^2_d \\ \sigma_{d-m} \\ \sigma^2_m \end{pmatrix} = \begin{pmatrix} 4 & 0 & 0 \\ -2 & 4 & 0 \\ 1 & -4 & 4 \end{pmatrix} \begin{pmatrix} \sigma^2_S \\ \sigma_{S-Mgs} \\ \sigma^2_{Mgs} \end{pmatrix}$$

Phenotypic variance(σ^2_P) is estimated as: $\sigma^2_p= \sigma^2_S+ \sigma^2_{Mgs}+2\ \sigma_{S\text{-}Mgs}+ \sigma^2_{H\text{-}Y}+ \sigma^2_e$
residual variance(σ^2_e) was fixed to 1 (as is usual in threshold model), and other components of variances estimated proportionally using MATVEC software (Kachman, 2001) with AIREML procedure and choosing Probit function as a link function.

Results The estimated (Co)variance components and genetic parameters of calving difficulty are shown in Table 1. The estimation of heritabilities are in line with results from other studies (Van Tassel *et. al*. 2003).

Table 1 Estimates of (Co)variance components and genetic parameters with the standard errors in the brackets, the marks are introduced in the text.

σ^2_S	$\sigma_{S\text{-}Mgs}$	σ^2_{Mgs}	$\sigma^2_{H\text{-}Y}$	σ^2_e	σ^2_p
0.0284	0.0061	0.0144	0.8090	1	1.8642
(0.0044)	(0.0029)	(0.0027)	(0.0340)		
σ^2_d	$\sigma_{d\text{-}m}$	σ^2_m	h^2_d	h^2_m	$r_{d\text{-}m}$
0.1138	-0.0323	0.0617	0.0611	0.0331	-0.3857

Conclusions Heritabilities obtained from this method are greater than those estimated for this data from a similar model but using a linear method (Abdullahpour, 2005), and indicates that some parts of genetic variance hide in the categorical aspect of calving difficulty scores and will not appear unless using a better model like the threshold model. It seems that the threshold model is a step forward towards an ideal model; however this technique is computationally intensive and has its own problems.

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Comparison analysis and frequency of BoLA-DRB3 alleles in Iranian Sistani and Russian Yaroslavl breeds

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Introduction Major histocompatibility complex genes, also called bovine lymphocyte antigen (BoLA), have received attention because of their association with host immunity. The BoLA gene is located on the short arm of bovine chromosome 23. In the class II region of the BoLA gene, at least 12 loci have been described including in the class IIa region: DRA, DRB1, DRB2, DRB3, DQA, DQA2, DQB1, and DQB2; class IIb region: DYA, DYB, DIB, and DOB. Lewin H. A. (1996) identified 35 DRB3 alleles in exon 2 with a technique described by van Eijk *et al*. (1992) involving polymerase chain reaction (PCR) and endonuclease restriction fragment length polymorphism (RFLP). The BoLA-DRB3.2 locus is highly polymorphic; more than 54 different alleles have been reported. The BoLA-DRB3.2 alleles potentially affect many traits related to immunity, SCC, and mastitis incidence. Associations have been made with some infectious diseases of cattle and BoLA genes. Others indicated that one BoLA-DRB gene pattern was associated with resistance to Staphylococcus aureus mastitis, BoLA-DRB3.2*8, *16, *22, and *28 alleles were associated with elevated SCC and cows with BoLA-DRB3.2*16 and *24 were more susceptible to IMI caused by major mastitis pathogens. Furthermore, cows with BoLA-DRB3.2*ll, *12, and *23 alleles were more resistant to clinical mastitis and to IMI caused by major mastitis pathogens, however BoLA-DRB3.2 *16 was associated with lower SCS in Holsteins and BoLA-DRB3.2 *23 was associated with severe coliform mastitis. Most of the previous studies of the BoLA-DRB3.2 gene have involved Holstein cows or a combination of various breeds. Few studies have analyzed exclusively the BoLA-DRB3.2 gene of Iranian Sistani cattle and has not reported frequency of BoLA-DRB3 alleles in Russian Yaroslavl breed. The purpose of the present study was to determine the BoLA-DRB3.2 allele pattern in Iranian Sistani and Russian Yaroslavl breeds.

Material and methods 120 Russian yaroslavl cattle and 65 Iranian Sistani cattle were used in this study. DNA isolation from 200 ml of blood was perfumed by Diatom^{TM} DNA Prep 200 kit. Oligonucleotide primers used for amplification of the second exon of BoLA-DRB3 were based on previously published sequences of BoLA-DRB3 alleles. Amplification of DRB3 alleles was performed in two stages. For the first round of amplification the primers were: HLO30; 5'-ATCCTCTCTCTGCAGCACATTTCC-3', and HLO31; 5'-TTTAAATTCGCGCTCACCTCGCCGCT-3'. Heminesting PCR was used to increase yield and specificity of the PCR product. In the second round of amplification, employing primers HLO30 (as above) and Hlo32; 5'-TCGCCGCTGCACAGTGAAACTCTC-3'. Reactions were carried out on 30-50 ng of DNA (2-3 µl) in a 25 µl final valume. For PCR we were used GenePak^{TM} PCR Core kit. The thermal cycling profile for the first round of amplification was as described previously. Subseqently, 2 µl of first-round product were transferred to another 500-µl tubecontaining 23 µl PCR buffer for a second round of PCR. The amplified DNA fragments were digested with RsaI, HaeIII or XhoII under standard conditions.

Results Analysis of BoLA-DRB3.2 allele fingerprints of 120 yaroslavl and 65 Sistani cows resulted in identification of 34 and 33 BoLA-DRB3.2 alleles respectively. 13 alleles, representing an allele frequency of 43.06%, observed only in Yaroslavl breed and 12 alleles, representing an allele frequency of 22.12%, observed only in Sistani breed. 21 alleles distinguished in both breeds. The four most frequently isolated alleles in Sistani (BoLA-DRB3.2 *8, *11, *10, and *34) and in Yaroslavl (BoLA-DRB3.2 *12, *13, *24, and *28) accounted for 45.03 % and 45.29% of the alleles in their populations respectively. Significant associations have been made with some infectious diseases of cattle and BoLA genes, particularly diseases that are prevalent during early lactation. Associations between BoLA allele types and persistent lymphocytosis caused by bovine leukosis virus.

Conclusion the results of our study demonstrated that the BoLA-DRB3.2 locus is highly polymorphic in these breeds and indicate that differences exist between breeds of cattle with regard to BoLA-DRB3.2 allelic frequency. More studies need to be conducted on the BoLA-DRB3.2 gene to determine allelic frequencies within two these breeds. Studies are in progress to evaluate the relationship of BoLA-DRB3.2 allele types in two these breeds with SCC, mastitis susceptibility or resistance, and reproductive performance. Further DNA sequence analysis or conventional RFLP analysis with DNA probes needs to be conducted to verify that these are in fact new allele types.

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Within and across breeds differences in fatty acids profiles of milk and milk fat

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Introduction Many studies were already focussing on ways to improve the nutritional quality of milk fat. The most popular way to improve nutritional quality is by feed supplementation; however these methods present certain disadvantages. Most important is that this improvement is not sustainable. If the supplementation stops, the nutritional quality disappears. A genetic improvement however is complementary to feeding and has the additional advantage to create additional value for the animals through selection. However studying the genetics of nutritional quality of milk fat requires a lot of data. The objectives of this study were to use medium infrared spectrometry to get this data and, by using these predicted quantities of fatty acids in milk and milk fat, to study the differences in fatty acids profiles of milk within and across breeds.

Materials and methods After a phase of calibration, the composition of fatty acids in milk and milk fat were estimated by medium infrared spectrometry. A total of 600 milk samples produced at different test-days by 275 cows from 5 different breeds (Dual Purpose Belgian Blue (DPB), Holstein-Friesian (HOL), Jersey (JER), Montbeliarde (MON) and non-Holstein Red and White (RED) were analysed. The differences in fatty acids profiles of milk and milk fat and in delta-9 desaturase activity (C14:1/C14:0, C16:1/C16:0, C18:1/C18:0) within and across breeds were modelled. The following single trait mixed model was used: $\mathbf{y} = \mathbf{X\beta} + \mathbf{Zu} + \mathbf{e}$ where $\mathbf{y}$ is the vector of the analysed trait (fat, fatty acids contents in milk or milk fat or the delta-9 desaturase activity indicator traits), $\boldsymbol{\beta}$ is the vector of fixed herd*test date, number of lactation, stage of lactation and regressions on breed composition effects as a lot of animals were crossbred, when fatty acids contents in milk fat were analyzed a regression on milk fat percentage was added; $\mathbf{u}$ is the vector of random repetition on animal effect; $\mathbf{X}$ and $\mathbf{Z}$ are incidence matrices. Animals were considered unrelated as tests using the relationship matrix and separated genetic and permanent environmental effects did not converge. Computations were done with SAS PROC MIXED, and variance components were estimated using REML. Results for regression on breed composition were reported compared to the reference Holstein breed. In order to allow comparisons across traits, differences were expressed on a standardized scale by dividing them by the total standard deviation obtained as the square root of the sum of the animal and residual variances. Animal repeatabilities reflecting the degree of within breed differences were estimated from the ratio between animal and total variances.

Results By comparing total milk fat with fatty acids on a standardized scale one notices that the values obtained were not constant. For example, DPB had the lowest concentration in milk fat, however results were clearly lowest for saturated and monoinsaturated fatty acids in milk. Also the concentrations in C18:1 and C18:2 9-cis, 12-cis obtained were not the lowest concentrations for these fatty acids (Figure 1). Therefore, the proportion of insaturated fatty acids in milk fat might be high. In fact, the fat of DPB contained the lowest proportion in saturated fatty acids (SAT) (Figure 2). These observations might be explained by the higher activity of delta-9 desaturase for this breed (Figure 3). Similarly, the fat of RED and MON contained a high proportion of SAT. This might be explained by a low delta-9 activity. Few publications have compared the fatty acids compositions for milk produced by different breeds. A publication indicates that the proportion of SAT in JER milk fat was higher than the one produced by the HOL animals (Palmquist *et al*., 1992), a result that our results confirmed.

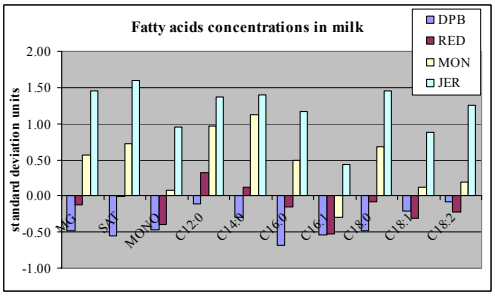

Figure 1 Differences in fatty acids composition in milk (standardized units) compared to Holsteins.

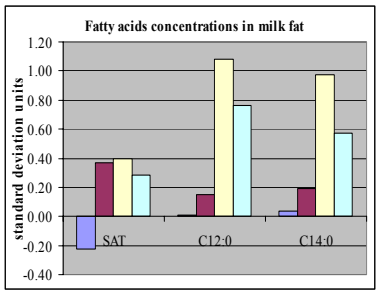

Figure 2 Differences in fatty acids profiles in milk fat (standardized units) compared to Holsteins.

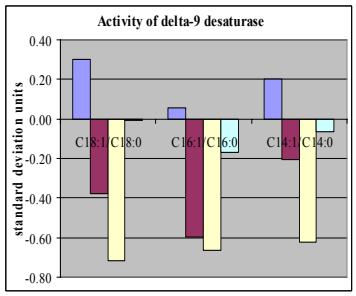

Figure 3 Differences in activity of delta-9 desaturase (standardized units) compared to Holsteins.

To estimate the differences within breed, the repeatabilities for all studied traits were estimated and the obtained results were high with values beween 43 and 61 %. Future studies should show if this is also an indication for reasonable high heritabilities.

Conclusion The differences in fatty acids profiles of milk across the selected breeds have been shown by this research. It might suggest the possibility to obtain milk products with differentiated nutritional quality by the choice of breed. By the estimated repeatabilities, the differences within breed have also been shown. These might suggest a high heritability for each component of fatty acids profile and allow their genetic improvement.

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Effects of sire genotype on lamb performance in extensive sheep systems in Western Patagonia

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Introduction In Western Patagonia (Chile), sheep production is a key contributor to the local economy. Sheep production in Patagonia is largely based on the Corriedale, a dual-purpose breed used for apparel wool and prime lamb production. Over the last decade the relative value of sheep meat compared with wool production has greatly increased. Consequently, there is interest in using terminal sire breeds to improve growth and carcass quality in lambs. In view of this background a study is being undertaken to investigate the relative performance of four sire breeds across a range of sheep systems in Western Patagonia. This paper reports on the results from the first year of this two-year programme.

Material and methods Nine farms, located across the Aisén region of Western Patagonia were involved in the study. During May 2004 on each of the farms, 100-200 purebred Corriedale ewes were allocated to four sire breed treatments, balanced for ewe live weight, condition score and age. A total of 10 Corriedale, 10 Dorset, 9 Suffolk and 7 Texel sires were used in the study, each from different bloodlines. 5 Corriedale, 5 Dorset, 5 Suffolk and 1 Texel sires were selected for use in the trial on the basis of their estimated breeding values (EBV's) to represent the top 10% of recorded animals from flocks in Chile and New Zealand. The remaining sires (control) were selected from flocks not participating in genetic improvement programmes in Chile. Ewes were bred to a synchronised oestrus by artificial insemination. Lambs from each of the crosses (121 Corriedale, 145 Dorset, 157 Suffolk and 120 Texel) were weighed at birth, at 5-10 weeks of age and at weaning. At weaning a representative sample of male lambs were slaughtered and carcass characteristics recorded. The data were analysed using the Genstat REML (Residual Maximum Likelihood) procedure. This fitted fixed effects for sire breed and sire source (non recorded (control) versus recorded (high EBV)).

Results There were no sire breed X source interactions therefore main effects only are presented (Table 1). Sire breed had no significant effect on lamb birth weight. Ewes mated to Suffolk sires required more assistance at lambing ($P < 0.01$). However, overall the number of lambs weaned per ewe was not significantly affected by sire breed. Dorset and Suffolk sired lambs had higher daily liveweight gains (LWG) ($P < 0.05$) compared with purebred Corriedale lambs. Consequently at weaning, Dorset and Suffolk lambs were heavier ($P < 0.01$) compared with Corriedale lambs, with Texel lambs intermediate. Carcass weights of the lambs followed a similar pattern. Carcass conformation was higher in each of the crossbred groups compared with the purebred Corriedale lambs ($P < 0.05$). Similarly, the weight of high value cuts was greater with Dorset and Suffolk sires compared with Corriedale lambs ($P < 0.01$), with Texel intermediate. High EBV sires tended to produce heavier lambs at weaning ($P=0.08$) compared with control (non recorded) sires.

Table 1 The effect of sire breed and source on lamb output at weaning

	Sire breed					Source		
	Corrie	Dorset	Suffolk	Texel	S.E.D.	Control	High EBV	S.E.D.
Performance–weaning								
No. lambs born/ewe	1.14	1.22	1.27	1.18	0.061	1.22	1.19	0.044
Lamb birth wt (kg)	5.3	5.5	5.7	5.6	0.20	5.4	5.6	0.14
Propn assisted lambing	0.01^{a}	0.01^{a}	0.06^{b}	0.02^{a}	0.026**	0.02	0.03	0.018
Propn lamb mortality	0.21	0.27	0.37	0.29	0.076	0.31	0.26	0.054
No. lambs weaned/ewe	0.93	0.93	0.91	0.89	0.073	0.90	0.93	0.052
LWG (birth-wean) (g/day)	247^{a}	283^{b}	276^{b}	266^{ab}	11.5*	262	274	8.1
Weaning wt (kg)	27.4^{a}	30.6^{b}	30.3^{b}	28.9^{ab}	0.92***	28.7	29.9	0.66
Wt lambs weaned/ewe (kg)	29.4^{a}	32.5^{b}	33.0^{b}	29.7^{a}	1.29**	30.5	31.7	0.91
Carcass characteristics								
Carcass weight (kg)	13.2^{a}	15.7^{c}	15.2^{bc}	14.2^{ab}	0.66***	14.4	14.8	0.47
Carcass conformation (1-4)	2.4^{a}	2.9^{b}	2.7^{b}	2.9^{b}	0.14***	2.7	2.8	0.10
Carcass length (cm)	63.0^{a}	66.4^{b}	65.2^{b}	63.4^{a}	0.84***	64.1	64.9	0.60
Carcass fat class (1-3)	1.3	1.4	1.5	1.3	0.17	1.4	1.4	0.12
Grade rule (mm)	6.3	6.6	6.7	7.3	1.22	6.7	6.8	0.87
Kidney, Knob & Channel fat (1-3)	1.3	1.5	1.4	1.4	0.17	1.5	1.32	0.12
Wt high value cuts (kg)	3.37^{a}	4.02^{b}	3.93^{b}	3.73^{b}	0.176***	3.69	3.84	0.125
Wt lower value cuts (kg)	5.18^{a}	6.31^{b}	5.89^{b}	5.82^{b}	0.261***	5.72	5.88	0.184

Conclusions

Compared with breeding pure, crossbreeding Corriedale ewes with Dorset and Suffolk sires increased lamb weights at weaning by 10-11% and carcass weights by 15-19%. Carcass characteristics were also improved by crossbreeding. Using high EBV sires tended to increase the live weight of lambs at weaning.

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Genetic parameters of growth, ultrasonic tissue depths and slaughter traits for sheep reared in a hill environment

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Introduction The construction of selection indices for sheep improvement programmes requires reliable estimates of the genetic and phenotypic correlations among traits in the selection objective and the criteria on which selection is based. Parameter estimates for growth, ultrasonic predictors of body composition and slaughter traits are scarce for sheep reared in a hill environment in comparison to the information available for terminal sire breeds (Conington *et al.*, 1998). The aim of this study was to estimate the genetic parameters of growth and carcass characteristics of lambs reared in a hill environment when slaughtered at a constant level of finish.

Materials and methods Cheviot, Dorset, Lleyn and Texel rams were mated to Welsh Mountain ewes, using 3 rams per year for 3 years. Records included birth weight (BWT) and weights at 5 (5WW), 10 (10WW) and 16 (16WW) weeks of age for all lambs (n=1793). Female lambs were retained for breeding and males (n=800) were slaughtered at a standard level of finish. Prior to slaughter, ultrasonic muscle and fat depths were measured at the third lumbar vertebra and carcass weight, fat class (2-4L) and conformation score (EUROP recoded 1-5) were recorded post-slaughter. The fixed effects of sire breed, year, age of dam (2- 6 years), litter size (single, twin and triplet), sex (entire male or female, growth data only), fat class (slaughter data only) and grazing type (rough grazing, semi-improved and improved) and all two-way interactions were tested using the REML procedure of Genstat with sire fitted as a random effect. Genetic parameters were estimated using ASREML, fitting all significant fixed effects and interactions, with sire and dam fitted as random terms.

Results Estimates of the genetic correlations between adjacent live weights were high (Table 1) and declined with increased time between measurements. Birth weight was strongly correlated to early growth rate, but relationships with average daily gains (ADG) after five weeks of age were poor. Live weights were strongly correlated to previous, and poorly correlated to subsequent average daily gains. Genetic correlations among slaughter traits were generally not significantly different from zero (Table 2). However, ultrasonic fat depth was strongly correlated genetically with increased killing-out proportion and improved conformation score.

Table 1 Genetic parameters for live weights and average daily gains: heritability on, phenotypic correlations above, and genetic correlations below the diagonal

	BWT	5WW	10WW	16WW	ADGB-5	ADG5-10	ADG10-16
BWT	**0.16**†	0.62	0.52	0.43	0.34	0.16	0.03
5WW	0.91	**0.39**	0.79	0.71	0.91	0.15	0.13
10WW	0.88	0.93	**0.24**	0.81	0.71	0.72	0.01
16WW	0.68	0.77	0.95	**0.17**	0.67	0.50	0.60
ADGB-5	0.63	0.76	0.86	0.80	**0.11**	0.13	0.16
ADG5-10	0.33	0.12	0.48	0.75	0.45	**0.10**	-0.13
ADG10-16	-0.35	-0.34	0.05	0.36	0.00	0.84	**0.06**

† Approximate standard errors were 0.02 for phenotypic and 0.30 for genetic correlations and 0.05 for heritability

Table 2 Genetic parameters of slaughter traits: heritability on, phenotypic correlations above, and genetic correlations below the diagonal

	Slaughter weight	Muscle depth	Fat depth	Carcass weight	Killing-out %	Conformation score
Slaughter weight	**0.09**†	0.20	0.06	0.75	-0.17	-0.16
Muscle depth	-0.45	**0.20**	0.17	0.33	0.24	-0.27
Fat depth	-0.11	-0.42	**0.10**	0.20	0.23	-0.13
Carcass weight	0.78	-0.47	0.61	**0.10**	0.52	-0.33
Killing-out %	-0.56	0.05	0.87	0.10	**0.10**	-0.27
Conformation	0.43	0.14	-0.78	0.32	-0.16	**0.15**

† Approximate standard errors were 0.04 for phenotypic and 0.40 for genetic correlations and 0.07 for heritability

Conclusions Estimates of heritability were more variable than, and genetic correlations were comparable to those of Conington *et al.* (1998). Antagonistic relationships between fat depth, carcass weight and conformation need to be considered when constructing selection indices.

Acknowledgements The financial support of Hybu Cig Cymru – Meat Promotion Wales is acknowledged.

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Dynamic computer models to identify the optimal system and management of population structure for self-contained meat sheep crossbreeding enterprises

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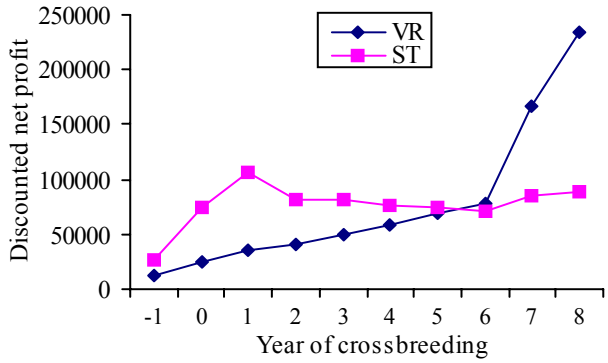
Introduction Systems analysis enables us as to know how the genetic, management, or environmental changes affect the system or how the system should be used for testing new hypothesis (Cartwright, 1979). Modelling is a major tool in this respect. The objective of this study was to develop two computer programs with year-by-year flow chart presentation to adopt the optimum-design, two-breed crossbreeding systems for self-contained meat sheep enterprises. The economic consequences based on the user-input data envisaged for a 10-year time period.

Materials and methods Two deterministic computer models called Stable (ST) and Variable (VR) models were developed applying the standard cash flow discounting (CFD) procedure (optional). The former, has stable- and the latter, variable flocks' size. In the ST, the flocks' size is stable in all years and in the VR, it is allowed to vary from one year to another (Fig. 2). There are two breeds of sheep and up to three flocks for each model with 27 input parameters for the breeds including inflation rate and total number of the ewes available at start. For any given set of the input data, the computer models examine all options to find the optimal crossing system and structure of the flocks under a detailed year-by-year scheme for 10 years. There are two primary years; "Year -1" and "Year 0" during which the system is established, with eight more years thereafter. Each model predicts the relevant parameters and proposes one of the rotational, rota-terminal and two-way crossing systems, or a combination of these with optimal number of the ewes for all of the flocks concerned. The flocks' structure and most of the important phenotypic, genotypic and management parameters as well as the economic consequences are displayed diagrammatically for a clear understanding of the simulation modelling. The used data relate to the Australian Merino and the Border Leicester. The effect of the CFD on profitability and flocks structure examined.

Results With same sets of the inputs for both models, the ST model proposed a rota-terminal and the VR model a combination rotational crossing system, the latter including some sub-systems. Using same numbers of the ewes at start (500) in both models, a very large amount of profit per year gained for the VR model in the final two years with little profit in previous years (Fig.1). The CFD had no effect on the flocks' structure but changed the general trend of the annual net profit to some extent. Next, the initial number of the ewes just in the VR model was held constant, and that in the ST model was changed until the models' cumulative net profits (CNP's) in with-CFD procedure equalised. As a result, the whole flocks' structure had some changes i.e., the ST model started with 916 ewes predicting 20.22% more CNP per ewe owing to 16.85% fewer ewes raised and 6.57% fewer lambs sold, all compared to the VR model. These figures for without-CFD procedure were 985 (13.8% larger), 8.38% smaller, 6.27% larger and, 7.03% larger respectively. In addition, the initial number of the ewes was always higher for the ST model; 83.2% higher in with- and 97% higher in without-CFD method, both with equal CNP's.



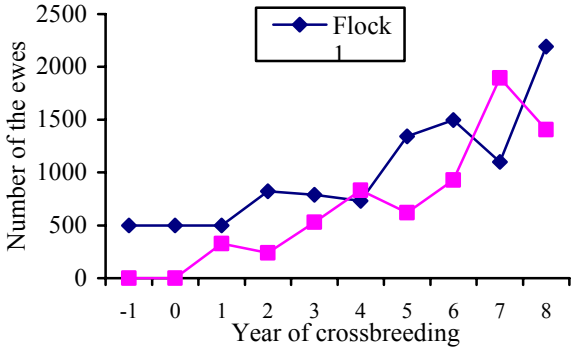


Figure 1 Trends of profit in the ST and the VR models **Figure 2** Variability of No. of the ewes in the VR model

Conclusions The ST model requires more ewes at the start and results in a higher CNP per ewe. It has less variation in the meat quality by use of generation preference and better utilization of the breed effects. This is due to less variation in the meatier breed's gene contribution and to the stable flocks' size. The VR model needs more ewes to be as profitable as the ST model in the end. Nonetheless, with same initial numbers of ewes for both models, the VR model gains more end profit due to a larger cumulative number of the ewes raised. In addition, in the VR model, the meat quality is unstable and there is too much delay in gaining a notable annual net profit. However, where there are low initial investments and limited resources available at the commencement of crossbreeding, the VR model would be more suitable.

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Genetic analysis on fleece traits of colour coated Markhoz goats in Kurdistan

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Introduction Many studies have been carried out to determine the quantity and quality of mohair production, and high variation reported by some workers for fleece trait in Angora goats (1, 2, 3, and 4). But very little is known about mohair production in colour coated Markhoz goats. The objective of this study was to determine the fleece characteristics, analyse the effects of some environmental factors on fleece traits and estimate the heritability for the average fibre diameter and yearling fleece weight in Markhoz goats.

Material and methods 775 mohair samples were taken from the mid sides of Markhoz goats and analysed for the evaluation of fleece traits. The variables were average fibre diameter (AFD), staple length (SL), true length (TL), percentage of continues medullated fibres (CMF), interrupted medullated fibres (IMF), kemp (K), clean yield (CY) and wax (W). Measurement of AFD, CMF, IMF and K were made with a projection microscope, from 300 fibres per sample. SL and TL of fleece samples were determined as average 10 randomly selected staple and 200 fibres per sample respectively. CY of the samples were determined by washing with SedoxNFK detergent and air drying to a constant weight at 20^0 C in the laboratory. Wax of the samples was determined by washing with Dicloromethan and air dried to constant weight. The environmental factors effects (herd, sex, age and colour) also were studied. Heritability were estimated with single trait analysis using restricted maximum likelihood (REML) with the animal model for AFD (n = 1234 records collected at 2000 - 2003) and YFW (n = 3213 records collected at 1990-2004) respectively.

Results The least squares means and standard errors for AFD, SL, TL, CMF, IMF, K, CY and W are given in Table 1. The effect of sex on SL, IMF and CY were significant (P<0.01), but had no effects on other traits. The female goats have longer SL (14.96±.21 vs 13.71±.40 cm), higher CY (82.16±1.07 vs 75.80±1.07 %) and lower IMF (0.20±0.06 vs 0.79±0.12 %) than the males. The effect of age on AFD, IMF, CY (P<0.01) and W (P<0.05) were significant. The least squares means of ADF, IMF, and CY increased from 27.92±0.65 micron, 0.24±0.09 % and 76.37±1.01 % at one year of age to 33.99±1.16 micron, 0.89±0.15 % and 80.99±1.37 % at six years of age, respectively. But least squares means of W decreased from 2.15±1.14 % at one year of age to 1.48±1.68 % at six years of age. The effect of herd on AFD, SL, CMF, CY, W (P<0.01), K and W (P<0.05) were significant. The ranges of AFD, SL, CMF, CY, K and W between herds were 6.69 cm, 1.92, 15.64, 2.37 and 0.5% respectively. Effect of colour on AFD, CMF, IMF (P<0.01) and SL (P<0.05) were significant. The highest and lowest least square means in colour type of goats were 29.29±0.71 micron in sandy brown vs 33.10±0.87 in white for ADF, 0.30±0.19 % in black vs 1.08±0.16 in saddle brown for CMF, 0.28±0.11 % in black vs 0.66±0.09 in white for IMF and 13.75±0.33 cm in sandy brown vs 14.70±0.04 in white for SL respectively. Direct heritability estimates for AFD and YFW were 0.12 ± 0.06 and 0.18±0.04 respectively.

Table 1 Environmental effects and least square means (±SE) of fleece traits in Markhoz goats

Traits	n	Mean±SE	Environmental factors			
			Sex	Age	Heard	Colour
AFD(µm)	731	30.96±1.76	ns	**	**	**
SL(cm)	571	14.51±1.03	**	ns	**	*
TL(cm)	50	11.72±2.13	ns	ns	ns	ns
CMF(%)	731	0.8±0.76	ns	ns	**	**
IMF(%)	731	0.49±0.17	**	**	ns	**
K(%)	731	1.44±0.47	ns	ns	*	ns
CY(%)	545	79.02±0.82	**	**	**	ns
W(%)	41	1.80±0.98	ns	*	*	ns

ns: P> 0.5 *:P< 0.05 **:P< 0.01

Conclusion Fleece characteristic of Markhoz goats show high variation according to colour, sex, herd and age. AFD, IMF and CY increases at 2 year of age to maximum at 6 years of age, but W decrease thereafter. These results indicated that the level of K, CMF and IMF in mohair production of Markhoz goats were low. Also it appears that the AFD and YFW in Markhoz goats are low heritability.

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Using indices of skeletal maturity to better understand musculoskeletal development in sheep

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Introduction Animal maturity is an important concept in the meat industry, with emphasis on producing less mature (*i.e.* "physiologically younger") animals with superior carcase characteristics such as greater tenderness and lower fatness. Unfortunately the concept of 'maturity' remains poorly defined. Where specified, 'maturity' usually refers to some proportion or index expressed relative to the mature state, in which the animal is in anatomical and/or compositional equilibrium. However indices of maturity referenced to body weight or composition (*e.g.* muscle:bone ratio) are problematic for assessing genotypic effects in modern prime lamb production, where terminal sires may be selected for specific compositional traits such as rapid muscle growth or low body fat. In such cases it may be preferable to define other indices such as *skeletal* maturity, by staging development in relation to longitudinal bone growth and mineral maturation and thus lead to a better understanding of musculoskeletal development in sheep.

Materials and methods Data for this experiment were derived from biometric measurement (total hindlimb length- femur to proximal phalanx) and mineral analysis (Ca, P, Mg) of the metacarpal (mid shaft) from Sheep CRC Resource Flock 1 serial slaughter (n=609) (age range 4 to 22 months). This flock consisted of five divergent crosses of Merino (M), Border Leicester (BL) and Poll Dorset (PD), including PD sires selected for high growth or eye muscle depth EBVs (MPDg and MPDm). Growth functions and mineral declines were fitted using Brody's function $y = a(1\text{-}e^{-k(t-t^*)})$. All animals were maintained on the same nutritional plane throughout the experiment.

Results Initial results indicate significant differences in bone growth, in particular the pronounced bone hypotrophy associated with sire EBVs for high muscling compared to more general selection for growth (Figure 1). The shorter mature bone length of the MPDm group dictates that these animals are earlier maturing in terms of skeletal growth (*i.e.* limb length relative to mature length). However, MPDm lambs retained a bone mineral profile (especially Mg^{2+} content[1]) more consistent with that of physiologically less mature animals (Figure 2), a result consistent with other indices of physiological maturity such as tooth eruption and eye lens weight. By contrast, the bigger MBL and MBLPD crosses show relative skeletal *immaturity* which contrasts with their obviously rapid physiological maturation.

Figure 1. Proportions of total hindlimb length as predicted from growth curves modeled by Brodies equation

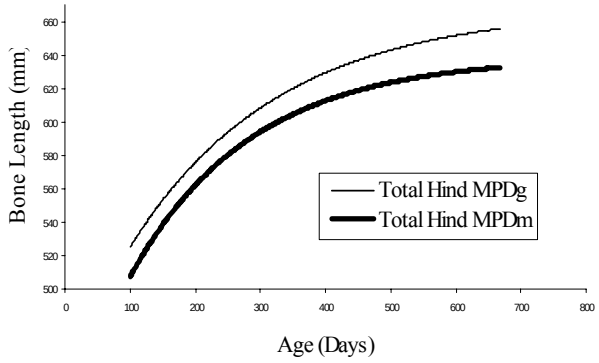
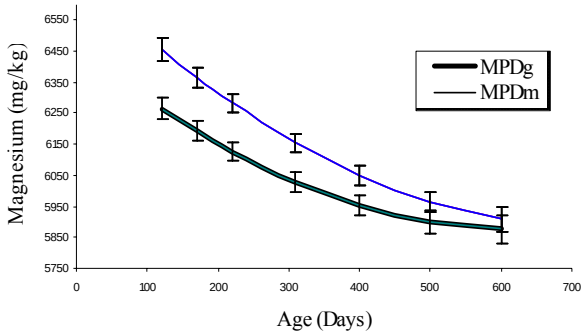


Figure 2. Age-related decline in metacarpal cortical bone Mg++ as predicted from growth curves modeled by Brodies equation



Conclusions We propose that estimates of maturity proportion (M) based on relative limb bone length or limb proportions may present significant advantages over weight- or composition-based maturity indices, or qualitative variables such as dental eruption or USDA-type maturity scores.
Future analysis will be aimed at describing more complex indices of skeletal growth (*e.g* growth plate analysis, limb bone proportions[2]) to further test whether genetic influences on skeletal maturation may in fact be independent of physiological maturation rate.

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Heterosis, general combining ability (GCA) and special combining ability (SCA) for resistance traits against disease in some lines of silkworm *Bombyx mori L.*

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Introduction The final goal in the most animal breeding programs is increasing of producing ability of the animals. Measuring the GCA and SCA for selecting the superior parents is important. 3-7 additive genes play more important role in the inheritance of some economical characters. Investigation on GCA and SCA of production and reproduction parameters has been showed that amount of SCA is more than that of GCA. Parents' possessing high GCA are generally considered for population development and for initiation of pedigree breeding as it is heritable and can be fixed. SCA consists of non-additive, dominant effects and other interactions (Ravindra *et al*, 2003). Since these value are not constant in different regions, thus must be estimated in different countries.

Materials and methods Some bivoltine pure lines of silkworm (Bombyx mori) and their hybrids including Japanese 31, 103 and 107 as well as Chinese 32, 104 and 110 were reared under standard conditions. Heterosis percentage was calculated by means of following formula:

$$\text{Heterosis} = [(\text{Hybrid Average} - \text{Parent Average})/ \text{Parent Average}] \times 100$$

SCA and GCA were estimated using Griffing (1950) model by means of following statistical model:

$$y_{ijkl} = \mu_i + jGCA_{ij} + cGCA_{ik} + SCA_{ijk} + e_{ijkl}$$

Where: y_{ijkl} = the record of l^{th} replication obtained from trait from cross between j^{th} Japanese line and k^{th} Chinese line for i^{th} character, μ_i = the mean of i^{th} character, $jGCA_{ij}$ = the effect of GCA of j^{th} Japanese line for i^{th} character, $cGCA_{ik}$ = the effect of GCA of k^{th} Chinese line for i^{th} character, SCA_{ijk} = the effect of SCA of hybrid obtained from cross between j^{th} Japanese line with k^{th} Chinese line for i^{th} character, e_{ijkl} = the effects of residue factors. In the above model all the effects were random and to estimate the variance parameters the model followed VARCOMP procedure (REML) of SAS software. Larval mortality calculated from hatching until end of larval duration. Larval vitality calculated only during 4^{th} and 5^{th} instar. Pupae vitality also calculated from end of 5^{th} instar until beginning of butterfly emerge.

Results A portion of obtained results from this study is summarized in table 1. The highest SCA of larval mortality, alive pupae and pupae vitality belonged to 107×110, 104×103 and 104×103 respectively. The highest heterosis of larval vitality, larval mortality and pupae vitality belonged to 110×107, 107×110 and 107×110 respectively. 31×32 showed the lowest larval mortality SCA. 110×107 showed the lowest alive pupae and pupae vitality SCA. 32×31 showed the lowest larval vitality heterosis. 110×107 showed the lowest larval mortality heterosis. 103×104 showed the lowest alive pupae and pupae vitality heterosis.

Table 1 Special combining ability (SCA) and heterosis percentage in six studied silkworm hybrids

Hybrid	Special combining ability (SCA)			Heterosis percentage (%)			
	Larval mortality	Alive pupae	Pupae vitality	Larval vitality	Larval mortality	Alive pupae	Pupae vitality
31× 32	-7.08	22.04	1.07	9.84	-31.03	-24.69	-1.63
32×31	-6.38	18.79	0.24	8.91	-28.13	-25.98	-2.54
103×104	-0.68	12.04	3.04	11.48	-40.81	-30.44	-5.54
104×103	-2.78	22.54	5.53	14.17	-50.37	-26.50	-2.78
107×110	8.51	-36.95	-4.78	23.18	-91.12	-8.30	0.47
110×107	8.41	-38.45	-5.10	23.30	-91.61	-8.86	0.14

Conclusions Significance of GCA at the larval and pupal resistance characters in Japanese lines (even though the mean of these characters are low) indicated the additive effects of genetical control on these characters. Therefore it could be expected that with the selection of Japanese lines with the better resistance characters as a maternal breeds for combination, resistance of the hybrids will considerably increase. In contrast, in Chinese lines (although they have higher resistance characters) the resistance characters have lower additive genetical variance and it is expected that in the resulting hybrids considerable improvement would not occur. Parents possessing high GCA are generally considered for population development and for initiation of pedigree breeding, as it is heritable and can be fixed. Parents with high GCA produce high heterosis as GCA consists of non-additive effects, dominant effects and other interactions. SCA is not heritable and therefore it cannot be utilized in pure line breeding. Hybrids with high SCA are useful for commercial exploitation.

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Analysis of combining ability for double cocoon characters in silkworm

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Introduction Silkworm is an important industrial insect with many unique characters. Combining ability of parents at animals and silkworm is dependent on different genetic effects that cannot be analyzed completely with phenotype performance. Many reports have showed that many good characters at silkworm have heterotic effects (Rajanna and Puttaraju, 1998). In Sericulture, the parents with high general combining ability are used generally for improvement of silkworm population performance. These parents considering additive effects and additive×additive effects produce high heterosis. Meanwhile the special combining ability includes non-additive effects, dominant effects and other genetic effects (Singh *et. al.* 1990). Double cocoons have two pupae together in one cocoon. These cocoons are an important section of production by silkworm. Therefore, analysis of combining ability for double cocoon character is beneficial for understanding of its genetic specifications and improvement of production level.

Materials and methods At this experiment, heterosis, general combining ability and special combining ability were estimated for double cocoon characters including number of double cocoons (NDC), double cocoon percentage (DCP) and weight of double cocoon (WDC) in silkworm (*Bombyx mori*) lines including Japanese lines of 31, 103 and 107 and Chinese lines of 32, 104 and 110 and their hybrids. For each line and hybrid, eight replications and 250 larvae for each replication were used. Therefore, a total number of 2000 larvae were reared under standard conditions for each line and hybrid. Statistical model for heterosis trial was as follows:

$$H_{ijk} = \mu_i + V_{ij} + e_{ijk}$$

Where H_{ijk} = percentage of heterosis of k^{th} replication of j^{th} hybrid for i^{th} characteristic, μ_i = the mean of heterosis percentage for i^{th} characteristic, V_{ij} = the effect of j^{th} hybrid on the percentage of heterosis of i^{th} characteristic, e_{ijk}= the effects of residual factors. To estimate the parameters of specific combining ability (SCA_{ij}), general combining ability (GCA_i) and heterosis (H_{ij}), the following equations were used:

$$GCA_i = M_i - \mu$$

$$SCA_{ij} = M_{ij} - (GCA_i + GCA_j + \mu)$$

$$H_{ij} = \frac{M_{ij} - (\frac{M_i + M_j}{2})}{(\frac{M_i + M_j}{2})} \times 100$$

Where μ = the mean of all hybrids, M_i = the mean of hybrids which obtained from i^{th} line, M_{ij} = the mean of hybrid which obtained from cross between i and j lines.

Results Estimated genetic parameters showed that heterosis effects are significant for all characters ($P<0.001$). Obtained results from this study are summarized in table 1. The highest general combining ability for number of double cocoons and double cocoon percentage belonged to 107 (among Japanese lines) and 110 (among Chinese lines) lines. Furthermore, the highest general combining ability for weight of double cocoon belonged to 103 (among Japanese lines) and 104 (among Chinese lines) lines.

Table 1 Heterosis percentage and special combining ability (SCA) in six studied silkworm hybrids

Hybrids	Heterosis %			Special combining ability (SCA)		
	NDC	DCP	WDC	NDC (No.)	DCP (%)	WDC (g)
31× 32	-32.67	-54.03	67.96	1.58	0.39	-0.26
32×31	-16.83	-42.48	73.05	2.58	0.92	-0.12
103×104	-60.00	39.16	104.07	1.83	0.72	-0.06
104×103	-55.55	-33.66	75.37	2.33	0.91	-0.82
107×110	24.06	32.45	45.38	-2.41	-0.74	0.68
110×107	-9.99	-4.85	42.19	-5.91	-2.21	0.59

Conclusions From obtained results, it was distinguished that general combining ability for hybrids is accorded with their heterosis. Then it is concluded that additional effects affect studied lines and application of these lines resulted to production improvement. In crossbreeding programs, combining ability and heterosis must be pointed under attention jointly together. The obtained results of this study indicated that the traits related to double cocoon production are strongly under heterotic effects. This criterion can be applied to improve the efficiency of silkworm egg and cocoon production. Furthermore, the results revealed high and positive amounts of average heterosis in all hybrids for double cocoon character. Other researchers (Singh et. al. 1990) also confirmed these results. The studied lines showed high combining ability which along with the high heterosis can provide valuable genetic sources for further breeding programs.

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Performance of one Iranian commercial silkworm hybrid at different rearing temperature and humidity

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Introduction The silkworm is an important industrial insect and like the other species needs proper adopted rearing conditions according to its physiological requirements to fulfil desirable performance. Since most of the farmers at different provinces of Iran use traditional rearing systems which will be affected by their regional climate conditions, it is necessary to evaluate the performances of Iranian commercial silkworm hybrids at different rearing conditions. Elena (2002) studied and confirmed the effects of different microclimate conditions on the depression of silkworm performance. Guar and Upadhyay (2003) demonstrated the effects of ecological changes on the amino acid content of the silk gland in silkworm larvae. The silkworm hybrid of 151×154 is the most recently introduced commercial hybrid in Iran and the objective of this study is to evaluate its performance at different microclimate conditions which will help to identify the proper regions for this hybrid.

Materials and methods From the first day of 4^{th} instars the larvae of 151×154 silkworm hybrid were reared under 4 different microclimate treatments (each treatment in 3 replications) including: 1-high temperature (28-29$^{\circ c}$) and high humidity (85-90%), 2-high temperature (28-29$^{\circ c}$) and low humidity (60-70%), 3-fluctuation of temperature (27$^{\circ c}$ at daytime and 18-20$^{\circ c}$ at night time) with 70% of relative humidity, 4-control treatment (temp. of 23-24$^{\circ c}$ and R.H. of 70-75%).Generated linear model (GLM) of SAS was used for statistical analysis while mean comparison within different groups accomplished by means of DUNCAN method at $P<0.05$.

Results A portion of obtained results from this study are summarized in Table 1. High temperature and humidity treatments showed the highest larval mortality, pupae mortality, total mortality, the number of cocoon per liter. This treatment also showed the lowest weight of cocoon per 10000 larvae, number of cocoon per liter and single cocoon shell weight and cocoon shell rate. Control treatment showed the highest good cocoon percentage, weight of cocoon per 10000 larvae, single cocoon weight and single cocoon shell weight and the lowest number of cocoon per liter.

Table 1 The mean value of traits at different studying treatments

Traits / Treatments	Good cocoon (%)	Larval mortality (%)	Pupae mortality (%)	Total mortality (%)	Weight of cocoon per 10000 larvae (kg)	Number of cocoon per liter	Single cocoon weight (gr)	Single cocoon shell weight(gr)	Cocoon shell rate(%)
High temp. & humidity	70.45^{b}	5.39^{a}	6.63^{a}	11.29^{a}	14.17^{c}	109.50^{a}	1.656^{c}	0.322^{d}	19.49^{d}
High temp. & low humidity	76.4^{a}	3.67^{b}	4.14^{ab}	7.54^{b}	14.88^{b}	105.33^{b}	1.632^{d}	0.337^{c}	20.79^{a}
Temp. fluctuation	74.25^{ab}	2.06^{c}	1.86^{b}	3.81^{c}	16.95^{a}	105.5^{b}	1.84^{b}	0.365^{b}	20.07^{c}
Control	78.1^{a}	2.97^{c}	3.03^{b}	5.73^{bc}	17.09^{a}	99.17^{c}	1.872^{a}	0.379^{a}	20.56^{b}

Means followed by different letters in the same column are significantly different ($P<0.001$).

Conclusions There was no significant difference for good cocoon percentage between control, temperature fluctuation and high dry temperature treatments. The highest value for larval mortality was observed under high temperature and high humidity treatment while high dry and humid temperature resulted to the highest pupae mortality. Indeed the interaction of hybrid × treatment affects the mortality or resistance traits at a lower extent. The highest productivity performances (cocoon weight per 10000 4^{th} molted larvae) were obtained under control and temp fluctuating treatments. Increasing of the humidity on the other hand decreased the weight of cocoon shell. Therefore it can be concluded that this hybrid is more suitable for rearing under relatively low humid conditions and the environmental parameters such as temperature and humidity leave very great effects on silkworm performances.

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Derivation of economic values of important traits for minimizing production system costs in silkworm (*Bombyx mori* L.)

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Introduction The general aim of selection in animal breeding is to acquire new generation of animals, which under future economic conditions of production system are more efficient than the present generation (Groen, 1990). Economic values are functions of incomes and expenses of a production system and changes in these factors may result in changing economic values. Estimates for the economic values depend on the definition of the production system, the goals to be optimized and particular production circumstances. Production systems can be optimized to different goals, e.g. maximum profit, minimal costs of product or maximum return on investment. Circumstances that limit input or output, e.g. a quota system for production or environmental legislation, might affect the economic value of a trait. This study focuses on the derivation of economic values based on a cost to return function for Iranian commercial silkworm lines and effect of different perspectives on economic values.

Materials and methods The present investigation has been carried out on commercial varieties which were produced in Iran's Silkworm Research Center. In order to derive economic values (E.V.s) of single cocoon weight, shell weight, shell percentage, fecundity, fertility and hatchability using data simulation (system analysis) method, a deterministic model was defined as follows:

$$Q = NC/NR = C/R = (nfhvxm + t)/(nfhvxr)$$

where Q is minimizing cost function, N is breeding moth number, R and C are annual production system return and costs per moth, respectively and n, f, h, v, x, r, m, t are averages of fecundity, fertility, hatchability, cocooning percentage, cocoon weight, cocoon price per gram, cocoon cost per gram and fixed costs per breeding moth, respectively. Also, potential magnitude of variation in economic value equations was estimated when calculated for some alternative perspectives including animal unit, product unit and return unit.

Results The absolute economic value equations of production and reproduction traits were different in three scenarios while relative E.V.s of traits were stable against change in perspective (Table 1). Economic values of cocoon weight and reproduction characters are related to fixed costs per moth. Also they have inverse relation with the trait mean. On the other hand, profitability due to increased trait mean will be resulted by decreased fixed costs. E.V.s of these traits have inverse relation with total cocoon produced per moth (X) in product unit perspective and R in return unit one. The absolute and relative E.V.s of cocoon shell weight and cocoon shell percentage are related to best cocoon percentage, shell price per gram and costs per moth. Relative E.V.s of reproduction traits have a direct relation with cocoon weight mean and inverse relation with trait mean. In the lines with higher cocoon weight mean, the breeding objective is focused on improving cocoon shell percentage and reproductive traits and selection emphasis on cocoon weight decreases.

Table 1 Economic value equations in different scenarios as well as relative E.V. of traits to cocoon weight E.V.[#]

	Perspective	Trait					
		cocoon weight	shell weight	shell percentage	fecundity	fertility	hatchability
Economic value	Return unit	-t/xR	$-P_bsQ/rx$	$-P_bsQ/r$	-t/nR	-t/fR	-t/hR
	Product unit	-t/xX	$-P_bsQ/x$	$-P_bsQ$	-t/nX	-t/fX	-t/hX
	Moth unit	-t/x	$-P_bsC/rx$	$-P_bsC/r$	-t/n	-t/f	-t/h
Relative E.V.		-	P_bsC/rt	xP_bsC/rt	x/n	x/f	x/h
Relative E.V.		-	129.70	207.96	0.0034	1.70	1.69

[#] P_b and s are best cocoon percentage and shell price per gram, respectively and X=nfhvx

Conclusions Different perspectives are appropriate for different sectors of the society. Return unit and product unit perspectives cause product price to decrease, therefore they provide for consumer interests. Cocoon producers are interested in moth unit perspective because it decrease costs per investment unit and cause farm profit to increase. Defining future scenarios for agricultural production and deriving economic values of genetic improvement for these scenarios is a useful tool in developing breeding strategies that are robust to changes in markets and politics.

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Study on genetic-economic responses to index selection in commercial silkworm lines

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Introduction The primary goal of silkworm breeding programs is gathering the desirable genes in appropriate combinations for improving genetic performance to maximize the yield and productivity per animal (Kang *et al.*, 2003). The general aim of selection in animal breeding is to acquire new generation of animals, which under future economic conditions of production system are more efficient than the present generation. In multiple-trait selection by means of selection index method, it is necessary to evaluate economic values of traits. In this method, genetic trend of important traits is directed based on their weights in economic efficiency of production system to acquire maximum improvement in economic merit (Mirhosseini *et al.*, 2005). The objective of this study was to estimate genetic parameters, additive genetic values and economic values of cocoon traits to design selection index program and evaluate genetic-economic responses to index selection.

Materials and methods The present investigation has been carried out on six commercial silkworm lines produced in Iran's Silkworm Research Center including 110 (L1), 107 (L2), 101433 (L3), Xinhang1 (L4), Koming1 (L5) and Y (L6). The data used were approximately 8800 records for cocoon characteristics including cocoon weight (CW), cocoon shell weight (CSW) and cocoon shell percentage (CSP) which obtained from six generations (24 full sib families reared in each generation) in each line. After estimating (co)variance components by REML method, additive genetic values of animals were predicted using Best Linear Unbiased Prediction (BLUP). Economic values of traits were derived using a deterministic model developed by data simulation method. Breeding objectives (H) in six lines were defined based on estimated economic values (v) and additive genetic values (g) for CW, CSW and CSP traits as follows: $H = v_{cw}g_{cw} + v_{csw}g_{csw} + v_{csp}g_{csp}$. Then, three-trait selection indices were constructed on the basis of defined breeding objectives to estimate economic merit of animals. The selection index program was used for selecting valuable animals and producing next generation.

Results Total additive genetic gains of the traits resulted from index selection in five generations are given in table 1. The lines Xinhang1, 101433 and Y revealed the highest genetic improvement for CW, CSW and CSP, respectively. The line 107 showed the lowest genetic gain for all the traits indicating low heritability for cocoon characters (0.374, 0.366 and 0.192 for CW, CSW and CSP heritability, respectively). Xinhang1, 101433 and Y showed higher heritability for CW (0.50), CSW (0.752) and CSP (0.337), respectively. The maximum and minimum genetic gain percentage over mean belonged to CSW and CSP, respectively, which is in accordance with their heritability. CSW showed high level of genetic advance and heritability values implying that this may be predominantly under the control of additive gene action. Total economic gain obtained in five generation was greater in Xinhang1 and lower in 107 lines (figure 1). It is expected that Xinhang1×Koming1 hybrid could play an important role in Iran's commercial egg production.

Table 1 Total additive genetic gains of the traits

Line	Trait		
	CW (g)	CSW (g)	CSP (%)
110	0.260	0.0920	1.98
107	0.176	0.0494	0.88
101433	0.341	0.1319	1.80
Xinhang1	0.427	0.1151	1.33
Koming1	0.188	0.0698	2.06
Y	0.244	0.0909	2.25

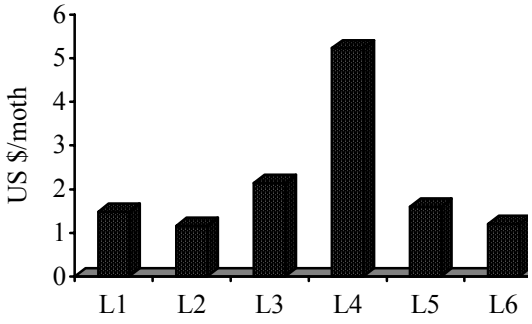

Figure 1 Total economic response to selection

Conclusions The results obtained demonstrated that genetic parameters (because of close mating system in the lines) and economic values of cocoon traits are different in commercial lines. Therefore, genetic and economic responses to selection vary among them. Silkworm lines have high heritability for economic cocoon characters. Therefore, selection index programme could be efficiently performed to improve genetic-economic performance of the lines. Knowledge of actual economic response to selection can help to silkworm breeder to use high economically efficient lines in hybridization program to maximize cocoon producers' profitability in the future economic conditions.

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Assesment of tree fodder preference, intake and digestibility by white tailed deer (*Odocoileus virginianus yucatanensis*)

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Introduction White tailed deer is distributed along America. In Yucatán, México, the sub specie *O. virginianus yucatanenses* was been hunted for food, leather and ritual purposes in prehispanic times. The yucatecan native white tailed deer (*O. virginianus yucatanensis*) is considered an endangered sub specie due to severe hunting practices. The establishment of farms (governmentally approved) has been encouraged in recent years in order to alleviate the pressure upon its numbers in the wild. However, most of the incipient farms are been set without adequate knowledge of deer biology. In order to establish proper management practices it is necessary to gather information of deer biology including the forage feed preference, intake and digestibility. Thus, the objective of the present work was to evaluate white tailed deer preference, intake and digestibility of four forage trees.

Material and methods A group of four deer males with an average LW of 43 ±1.6 kg (16.8 ± 0.62 kgPV$^{0.75}$) were used. Four tree fodders were evaluated; *Brosimun alicastrum, Leucaena leucocephala, Bursera simaruba, Guazuma ulmifolia*. Stage 1: Fresh tree fodder was offered separately on plastic containers. A 4x4 latin square design was used, where columns=position of the tree fodder in the pen, row=day of measurement and treatments the four tree fodders under evaluation (*Brosimun alicastrum, Leucaena leucocephala, Bursera simaruba, Guazuma ulmifolia*). Forage was offered for a 4-h period and intake measured by difference. Allocation of tree fodder within the pen (in the container) was changed every day to avoid the animal associate position with any particular forage. Stage 2: In order to know if the deers will increase its intake of forages with previous low intakes, the most preferred feed was eliminated from stage 2 and the remaining forages tested on a similar design in a 3x3 latin square design. The trial was repeated twice. Stage 3. Each fodder was evaluated separately during two weeks. Forage was offered *ad libitum* and intake measured every day. Animals were observed during the day in order to take fecal ground samples associated with the each animal. Digestibility was hence estimated using the Acid Insoluble Ash (Owens and Hanson, 1991) technique. Feed Samples were taken for chemical analysis (AOAC, 1980), polyphenols and condensed tannins (Price and Butler 1977, Price *et al*, 1978).

Results Chemical composition of the feeds is presented in table 1. During stage 1, *B. alicastrum* was the most preferred tree (P<0.0001) followed by *L. leucocephala*, *G.ulmifolia* and *B. simaruba* eaten in similar amounts. When *B. alicastrum* was removed the intake of the remaining trees increased but w/o difference amongst them (P>0.05) (Table 2). Similar to previous report with cattle (Sandoval *et al*., 2005), preference seems associated with lignin. The relationship was described by the following equation: Intake g DM/Kg LW$^{0.75}$ = -3.6486 (%lignin) + 51.95. To our knowledge this is the first report of forage tree *in vivo* DMD in *Odocoileus virginianus yucatanensis* (Table 3). Highest intake was obtained with *B. alicastrum*, although it was not associated with the highest digestibility.

Table 1 Chemical composition of fodder trees (% DM, except DM)

	DM	CP	Lignin	ADF	NDF	TP	CT
B. alicastrum	56.60	16.44	5.55	27.28	45.32	1.28	0.84
B. simaruba	44.00	12.64	12.18	26.73	44.84	2.54	28.72
L. leucocephala	45.40	24.63	10.23	19.34	43.95	1.68	2.09
G. ulmifolia	70.60	12.09	11.78	31.77	50.8	1.16	4.59

CT condensed tannins, TP, total polyohenols

Table 2 White tailed deer forage tree intake (g DM/Kg LW$^{0.75}$) on a two stage cafeteria trial.

	BS	*GU*	*LL*	*BA*	SEM	P
1st stage	7.86 **b**	8.42 **b**	11.66 **b**	28.65 **a**	2.310	0.0001
2nd stage	14.41 **a**	9.46 **a**	13.52 **a**	------	2.061	0.242

BS, B. simaruba, GU, G. ulmifolia, LL, L. leucocephala, BA, B. alicastrum

Table 3 Dry matter (DM), digestible DM intake (g DM/KgLW$^{-0.75}$) and digestibility of tree fodder by white tailed deer.

	B. simaruba	*G. ulmifolia*	*L. leucocephala*	*B. alicastrum*
DM intake	154.2 ± 19.96	224.3 ± 33.56	166.3 ± 32.36	265.3 ± 17.48
DMD (%)	80	60	81	61
DDM intake	123 ± 16.0	134.6 ± 20.1	134.7 ± 26.2	161 ± 10.7

Conclusion White tailed deer tree fodder preference seems to be associated with lignin, however further research is needed.

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Comparative digestive physiology of capybara (*Hydrochoerus hydrochaeris*) and collared peccary (*Tayassu tajacu*) and rabbit (*Oryctolagus cuniculus*)

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Introduction The continuing need of protein sources for the human population aside of the economic world is of considerable social interest, so the rational use of the local fauna has both economic and social advantages, while protecting wild species from extinction through irrational hunting and habitat destruction. Among Brazilian mammals the capybara is a big rodent, with high reproductive potential, are herbivorous, they increase their digestive efficiency through extensive mastication followed by fermentation by cellulolitic bacteria and protozoa in the cecum (González-Jiménez, 1995). Furthermore, capybaras are able to use microbial protein from the cecum after fermentation through the cecotrophy, or 'reingestion' of the cecal content (Mendes *et al*., 2000). In function of the controversial findings regarding cecotrophy in capybaras, this study was conducted to compare the faecal excretion curves of capybara, collared peccary (*Tayassu tajacu*) - animal that does not accomplish the cecotrophy, and rabbit (*Oryctolagus cuniculus*) - animal that accomplishes the cecotrophy.

Materials and methods Five females of each studied species: capybara, collared peccary, and rabbit, captivity born and reared, were introduced in individual pens (capybaras and collared peccaries) or metabolic cages (rabbits). This work was accomplished in three stages, being the first with capybaras, followed for collared peccaries, and finally, with rabbits, having an interval of five days among a stage and other. After 10 days of adaptation period to the new environmental conditions and feeding, all the animals were fed with corn, coast cross (*Cynodon spp*.) hay, soybean meal, calcium carbonate (experimental ration) and grass (*Pennisetum purpureum*) and were given one dose of chromium mordant mixed in the ration as faecal marker. Total collection of faeces was carried out in intervals of 4 hours during 96 hours. The t-Student test and a multivariate analysis profile were used to compare the similarity among the marker faecal excretion curves for the three species.

Results The t-Student test showed that there was no significant difference (t = 1.5; D.F. = 177; $p>0.05$) among the faecal excretion curves of chromium for capybara and rabbit. For capybara and collared peccary there was significant difference (t = -65.7; D.F. = 162; $p<0.05$), as well as for rabbit and collared peccary (t = -66.5; D.F. = 147; $p<0.05$). The multivariate analysis profile also allowed the comparison of the faecal excretion curves of accumulated chromium among the animal species, in times of collections intervals. The initial phase of excretion analyzed was from 0 to 16 hours (Figure 1), where it was verified differentiates among the faecal excretion curves for capybara and collared peccary (Contrasts for the F test, $p=0.0014$), and for rabbit and collared peccary (Contrasts for the F test, $p=0.0001$). Otherwise, there was no difference among the faecal excretion curves for capybara and rabbit (Contrasts for the F test, $p=0.0772$). The ascending phase of excretion analyzed, from 16 to 32 hours (Figure 1), also showed no differences among the faecal excretion curves for capybara and rabbit (Contrasts for the F test, $p=0.1029$). In this interval of time the faecal excretion curves for capybara and collared peccary (Contrasts for the F test, $p=0.0003$), and for rabbit and collared peccary (Contrasts for the F test, $p=0.0001$) differed too. The final phase of excretion analyzed was from 32 to 72 hours (Figure 1), where the faecal excretion curves between capybara and collared peccary (Contrasts for the F test, $p=0.9774$), capybara and rabbit (Contrasts for the F test, $p=0.4239$), and for rabbit and collared peccary (Contrasts for the F test, $p=0.4667$), they did not show differences.

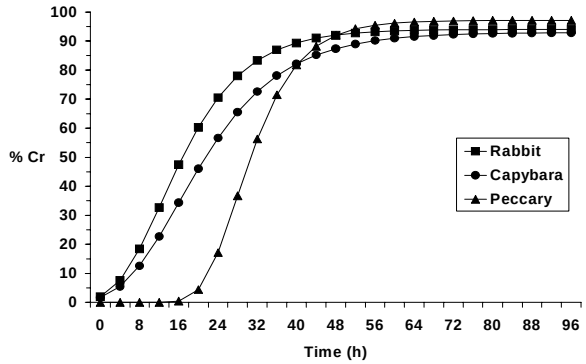

Figure 1 Comparison of the accumulated fecal excretion curves of mordent chromium among capybara, collared peccary, and rabbit.

Conclusions This work showed a similarity of the excretion curve of capybara in relation to rabbit, supporting previous evidences of cecotrophy in capybara. Peccary may show longer retention time because the presence of the fore-stomach with active fermentation. The cecotrophy is important because it suggests that maintenance expenses with protein and vitamins supplements for capybara in captivity can be reduced, making it possible to reduce production costs.

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The effects of changes in protein supply and demand on gastrointestinal parasitism in lactating rats at a constant level of energy intake

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Introduction The breakdown of immunity to parasites during the periparturient period plays an important role in parasite epidemiology and it is believed that it has a nutritional basis. A lactating rat model has been used to address this issue, as their nutritional status can be easily manipulated through altering greatly supply and demand and they exhibit a breakdown of immunity to the gastrointestinal nematode, *Nippostrongylus brasiliensis.* Previous experiments have shown that an increasing protein supply and reducing protein demand improves resistance (Houdijk *et al*., 2005; Normanton *et al*., 2005). However, in these studies, an increase in protein contents was associated with an increase in feed intake *per se*. Therefore, effects of protein supply could be confounded with effects of any nutrient or energy intake. The objective of the current experiment was to test the effect of increased protein supply or reduced protein demand on the resistance to parasites in lactating rats whilst energy intake was kept constant.

Materials and methods 32 second parity female Sprague-Dawley rats were given a single dose of 1600 third-stage infective *N. brasiliensis* larvae after arrival in the animal house. Post primary infection, females were mated and during pregnancy, all rats were fed similarly. Parturition was considered as day 0, and from then onwards rats received at 7.5% of their parturition body weight either a low protein diet (LP, 100g CP/kg DM) or a high protein diet (HP, 300g CP/kg DM). Both diets contained 18.0 MJ gross energy / kg DM. Litter sizes for the LP groups were standardised to 9 or 3 pups, while HP groups had only 9 pups. LP3 was included to show the effects of manipulating protein demand whilst keeping the low protein supply constant. The rats were re-infected with a single dose of 1600 *N. brasiliensis* on day 2. They were slaughtered on day 12 for the collection of colon contents and mucosal scrapings. Sections of small intestine were also removed for analysis of mucosal mast cells, eosinophils and globule leukocytes. ANOVA was used to assess the treatment effects on colon egg content and immunological cell analysis.

Results There was no significant difference between feed and hence energy intakes. Figure 1 shows the number of back transformed eggs in the colon contents taken on day 12 of lactation. HP9 and LP3 rats had significantly lower egg counts (P=0.03) than LP9 rats. Figure 2 shows the significantly different litter weights for the feeding treatments. HP9 final litter weight reached 141g while LP9 reached 108g, and LP3 61 g (s.e.d. 11.1g; P= <0.05) over the whole lactation. For the immunological analysis, globule leukocytes were absent in all tested sections. Feeding treatments had no significant effect (P=0.85) on mucosal mast cell numbers (mean 33 cells/0.45mm^2, 24-47 95% CI) or eosinophils (mean 115 cells/0.45mm^2, 93-145 95% CI).

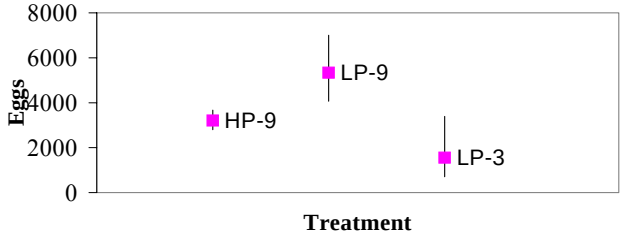

Figure 1 Colon egg count on day 12 of lactation, for for rats receiving high or low protein diets.

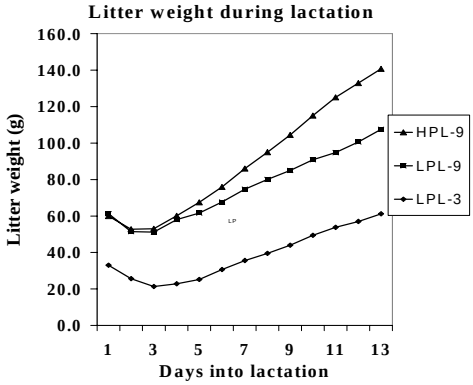

Figure 2 Litter weight of the pups, from rats receiving high or low protein diets, during lactation during lactation.

Conclusion The results support the view that under a restricted feeding regime, the periparturient breakdown of immunity to *N. brasiliensis* (measured by a reduced number of eggs in the colon content) is sensitive to changes in protein scarcity. Rats given the same amount of protein but having a different protein demand (litter size) show a different degree of breakdown of immunity. These responses are independent of energy intake. The immune responses measured were not effected by changes in nutrition. Hence, further analysis is now required to understand the underlying immunological basis of relaxation in immunity during the periparturient period and its sensitivity to nutrient scarcity.

Acknowledgements This work was supported by SEERAD

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A study on the integrated farming of Beijing duck (Anas platyrhynchos) and Silver Carp (Hypophthalmichthys molitrix)

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Introduction The grazing of ducks in carp feeding ponds is a profitable proposition. Ducks droppings are valuable organic manure; 100 kg of duck excreta contains 0.8 kg nitrogen, 1.5 kg phosphorus, and 0.4 kg potassium (Martyshev 1983). Woynarovich (1979) reviewed the practice of growing ducks and fish together. He calculated that one duck produces 6 kg of droppings during a 30-40 d period. At least 500 ducks can be kept on 1 ha of pond for a year, which corresponds to 36000 $kgha^{-1}$ of duck manure. Also, he estimated that each 100 kg of duck manure, distributed continuously in pond water, might result in an increase of 4 to 6 kg of fish flesh. The aim of this study was to assess the fish production performance between two ponds (with and without duck).

Materials and Methods The experiment was conducted in two ponds one hectare each in Guilan province, Iran from April to August for duration of 150 days. Each pond was stocked with 2000 Silver carp (*Hypophthalmichthys molitrix*), fingerlings. A total of 900 pieces of Beijing ducks (300/interval) were reared on the banks of a pond in three intervals (7 weeks/interval), whereas the other pond remained void of ducks. After stocking of the pond with equal number of fingerlings, only the duck free pond was supplemented with additional cow manure during the experiment and that of the pond with duck remained intact. In order to establish natural food resources prior the fish stocking, the ponds were enriched through addition of cow manure, urea and ammonium phosphate. Feeding of duckling during the first two weeks was carried out through administration of a ration containing 21% crude protein and 2800 $kcalkg^{-1}$ ME. During the last five weeks period of duck rearing a ration composed of 15% crude protein and 2800 ME (kcal/kg) was administered. The duck's ration consisted of corn, wheat, and barley, rice bran, soybean meal, and fishmeal, bone meal, and mineral and vitamin supplements. Two aerators were used in order to inject $1.7m^3min^{-1}$ of air with one atmospheric pressure/hectare in each pond. In this research, growth of silver carp in two ponds was investigated for five months. Then data were analyzed using Completely Randomized Design with 60 replicates (hunted fish) and mean values were tested using T- test.

Results In total, 2142 kg duck meat (live) was produced from three 50-d periods on one-hectare pond. The average weight of silver carp during the five months rearing period indicated that the growth rate of the fish in the integrated pond was higher than that of carp alone, since phytoplankton is the main diet to silver carp. The higher growth rate of the fish in the pond with the poly culture of fish and duck indicated remarkable positive effect of duck excreta on the growth and propagation of phytoplankton. The average weight and growth rates of silver carp in both of the ponds are presented in Table1.

Table 1 Average weights of Silver carp

	April	May	June	July	August
Fish pond	150.7 ^{j}	279.3 ^{h}	457.0 ^{f}	702.7 ^{d}	1036 ^{b}
s.e	4.06	1.86	2.52	1.33	15.68
Fish and Duck pond	173.7 ^{i}	358.0 ^{g}	565.7 ^{e}	847.3 ^{c}	1422 ^{a}
s.e.	1.33	1.67	1.20	1.33	1.86

Means within columns with different superscripts differ ($P \leq 0.01$).

Conclusions As a result of conducting this experimented design after 150 days of rearing a total of 1967 kg of fish from the fish pond and a total of 2698 kg of fish and a total of 2142 kg of live duck in the fish-duck pond was harvested, which means a total 4840 kg production of meat from Fish & duck pond. Sharma *et al*. (1998) reported that annual fish production was 4098.72 kg ha^{-1} from duck-fish integrated pond compared with 3844.92 kg ha^{-1} from the control pond where no ducks were released. An additional 258.03 kg of duck live weight and an annual production of 16625 duck eggs ha^{-1} were also obtained, which is comparable with the production obtained from this trail.

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Farmers knowledge in the utilisation of indigenous browse species for feeding livestock in Kweneng district of Botswana

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Introduction Farming systems in Botswana can be divided into two, subsistence and commercial. Most livestock producers are smallholder farmers, who do not have concentrate feedstuffs that could be used for supplementation during the extended dry seasons. Browse plants in genera *Acacia, Boscia, Combretum, Grewia* are valuable resources for smallholder farmers because they are rich in proteins and minerals (Aganga *et al*., 2001). However there are some browse plants that have anti nutritional chemicals e.g. tannins, which decrease availability of protein. Some are known to have medicinal properties for example Acacia trees. Farmer's knowledge has been there for a long time, and this research aims at finding out what oral tradition knows that has never been documented. The bank of indigenous knowledge in livestock and local feed resources such as browses when harnessed, adapted and utilized can improve the lives of local communities. The objectives of this study is to document Kweneng farmer's knowledge of browses species fed to livestock.

Materials And Methods The data and browse samples used in the study were obtained from four villages in Kweneng District, which were Kgope, Dikgatlhong, Ramankhung and Medie. Kgope is situated 20km west of Lentsweletau. Dikgatlhong and Ramankhung are on the northern part of Lentsweletau whereas Medie is located about 22 km northeastern of Lentsweletau.The vegetation is generally a hardveld. It comprises of mixed Acacia/ Combretum Tree Savannah. Among the common tree and shrubs are Acacia tortilis, A.fleckii, A.Erusbences and Combretum appiculatum. The soils are intrazonals of calcimorphic nature interspersed with sparsely distributed azonal litho soils. The average rainfall ranges from 450 to 500mm. The survey was conducted by means of a structured questionnaire, relating personal characteristics of farmers e.g. name, gender, marital status and level of education. The structured questionnaire focused on awareness, use and management of locally available trees and shrubs in feeding local livestock mainly small stock. One hundred farmers were interviewed. The system of livestock production was noted during the survey.

Results

Table 1 Kweneng farmers knowledge of browse species, and the parts of tree used (N=100)

Browse species	No. of farmers Mentioning trees	Parts of tree used			
		Leaves	Fruit	Leaves + fruit	Pods
Grewia flavansens	12	4			8
Acacia mellifera	51	51			
Terminalia sericea	15	15			
Acacia tortilis	53	28			25
Boscia albitrunca	23	17	6		
Dichrostacys cinerea	20	6	1		13
Grewia flava	21			21	
Grewia retinervis	16			16	
Grewia bicolor	17			17	
Acacia erubescens	15	15			
Acacia fleckii	17	17			
Sclerocarya caffra	17	7	10		

Conclusion It can be concluded that some of the farmers in Medie, Ramankhung, Dikgatlhong and Kgope are knowledgeable about the browse species that their livestock feed on especially goats. This knowledge includes knowing their names, parts preferred by livestock and which species are most preferred to others.
Age and gender of respondent had an effect on the farmers' knowledge since the older the respondent is the more browse species they know. Male respondents know more browse trees compared to females, because in most cases traditionally men associate with livestock while women take care of the field crops and their families. The bank of indigenous knowledge in livestock and local feed resources (e.g. browses) when harnessed, adapted and utilized can improve the lives of local communities.

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Variation in the growth potential of pigs sourced from different farms and managed in a common environment

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Introduction A study reported by Magowan *et al*. (2006) determined a large variation in growth rate, between herds of pigs in Northern Ireland reaching a live weight of 100 kg, equating to an average difference of 18 days. All pigs used in the study were of the Landrace x Large White breed. It was considered that the large variation in growth rate was mainly a result of different management or disease levels. It was therefore hypothesised that, if a representative sample of pigs from the various sources were housed in a common environment, performance of the pigs would be similar. The aim of this work was to investigate the variation in the performance of pigs sourced from herds with varying growth performance and housed in a common environment.

Materials and methods Three non-sibling boars, typical of pigs being weaned, were selected at 4 weeks of age from 3 of the 5 litters in each of the 8 herds used in the study of Magowan *et al*. (2006). At 4 weeks of age, pigs were transferred to a common environment. In total, 24 boars were mixed at 4 weeks of age in the common environment and group housed to 6 weeks of age, after which they were individually housed until slaughter (115 kg live weight). All Pigs were offered the same commercial diets *ad libitum* in the following sequence: Diet 1, CP 200 g/kg, Lys 15 g/kg, DE 17.5 MJ/kg (3 kg/pig); Diet 2, CP 200 g/kg, Lys 13 g/kg DE 16.9 MJ/kg (7 kg/pig); Diet 3, DE 15.1 MJ/kg, CP 200 g/kg (offered until pigs were 20 kg); Diet 4, DE 14.8 MJ/kg, CP 200 g/kg (offered from 20 – 40 kg live weight) and Diet 5, DE 14 MJ/kg, CP 200 g/kg (offered from 40 kg live weight to slaughter). All pigs received in-feed medication through Diets 1, 2 and 3 (3.1 kg/tonne Zn (Pigzin), 2 kg/tonne Stabox, 2 kg/tonne Pulmotil G1 in each diet) to minimize disease challenge. Pigs were weighed individually and feed intakes calculated twice weekly until 20 weeks of age. The average daily gain (ADG), average daily feed intake (ADFI) and feed conversion ratio (FCR) were calculated. Individual pig data were used for statistical analysis and results were analysed by ANOVA using Genstat 6.

Results Significant differences were observed between the production performance of pigs from different farms. For example, a significant difference of 143 g/day and 243 g/day was observed between pigs from the top and bottom performing herds in the common environment from 8-12 and 12–20 weeks of age respectively (Table 1). This difference equated to an average difference of 19 days between pigs from the top and bottom 25% of herds in reaching a live weight of 100 kg. There was no significant difference in feed conversion efficiency between the top and bottom 25% of herds. However the average daily feed intake of pigs in the top 25% of herds was significantly higher than that of pigs in the bottom 25% of herds, from 8 weeks through to 20 weeks of age.

Table 1 Performance of pigs in the top and bottom 25% of herds in the common environment

Age (weeks)	Top 25%	Bottom 25%	SEM	Sig
Average Daily Gain (g/day)				
8-12	831	688	16.9	**
12-20	1064	821	41.2	***
Est days to 100 kg	139	158	5.1	***
Average Daily Feed Intake (g/day)				
8-12	2628	1973	52.8	***
12-20	2625	2110	78.0	***
Feed Conversion Ratio				
8-12	2.47	2.40	0.035	NS
12-20	2.49	2.44	0.042	NS

Conclusions Although individually housed pigs performed better than those on farm, the variation in growth rate between herds was similar. In this study the difference in average time required to attain a live weight of 100 kg, was found to equal 19 days similar to that observed by Magowan *et al.* (2006). Pigs from different farms converted feed equally efficiently, however pigs in the top 25% had significantly higher feed intakes suggesting greater appetites and therefore promoted higher growth rates. It is difficult to assess the extent to which the pigs genetics or pre-weaning environment contributed to these differences. Further work is required to investigate the individual contribution and interaction of genetics, management, nutrition and disease status on the variable growth potential of pigs.

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Variation in on-farm growth performance of pigs

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Introduction Variation in pig performance is common within and between herds. Between herds, this variation may occur as a result of different management and nutritional regimes (Bruininx *et al.*, 2002), disease levels or genetics (Kennedy, 1984). However, the extent of this variation and its subsequent effect on the profitability of a pig unit has not yet been quantified. The aim of this work was to investigate the variation in growth performance of pigs from different herds within Northern Ireland and estimate its impact on profitability.

Materials and methods Eight herds (four with perceived poor performance and four with perceived good performance) offered the same nutritional regime were selected from the pig herds of Northern Ireland. Five litters, born within 3 days of each other, were randomly selected from each of the eight herds. On each litter, five pigs (3 boars and 2 gilts), representative of the litter, were selected. On each herd, 22 pigs (12 boars and 10 gilts) were weighed individually, every 4 weeks, from 4 to 20 weeks of age. The average daily gain (ADG) and coefficient of variation for pigs in each herd was calculated. Results were analysed by ANOVA using Genstat 6. The average growth rate of the top and bottom 25% of herds was inputted to an economic model (Devenish Nutrition Ltd) based on 1100 finishing places, to establish differences in profitability between herds. Correlations were established between the weights of pigs at various ages on the top and bottom 25% of herds using Genstat 6, and taking into consideration farm effects.

Results A growth rate difference of 61 g/d, 112 g/d and 170 g/d was observed between the top and bottom 25% of herds during 4-8, 8-12 and 12-20 weeks of age respectively (Table 1). As a result of this variation, the average time taken for pigs to reach a live weight of 100 kg differed by 18 days between the top and bottom 25% of herds. This difference in growth rate equates to an average difference of 9 p/kg per carcass, on a birth to bacon pig herd capable of housing 1100 finishing pigs. A higher coefficient of variation for weight of pigs at any age was found in poorer performing herds (bottom 25%) (Table 2). When the weights of pigs at various stages of growth were correlated against each other, weaker correlations were attained on good performing herds (Top 25%) (Table 3).

Table 1 The average growth rate (g/d) of pigs in the top and bottom 25% of herds

Age (weeks)	Top 25%	Bottom 25%	SEM	Sig
4-8	404	343	12.5	***
8-12	593	481	15.4	**
12-20	810	640	19.9	***
Est days to 100 kg	162	180	4.6	***

** P<0.01; *** P<0.001

Table 2 The coefficient of variation for weight of pigs from 4 to 20 weeks of age in the top and bottom 25%

Age	Top 25%	Bottom 25%
4	0.164	0.253
8	0.136	0.207
12	0.122	0.201
16	0.130	0.198
20	0.109	0.205

Table 3 Correlations between the weight of pigs at various ages in the top and bottom 25% of herds (n=43 each).

Age (weeks)	4	8	12	16	20
4	-	0.863***	0.733***	0.812***	0.800***
8	0.565***	-	0.884***	0.844***	0.836***
12	0.461**	0.816***	-	0.886***	0.874***
16	0.278 NS	0.610***	0.663***	-	0.917***
20	0.077 NS	0.447**	0.463**	0.861***	-

NS Not Significant, * P<0.05; ** P<0.01; *** P<0.001

Values below the diagonal report correlations between the weights of pigs in the top 25% of herds whereas values above the diagonal report correlations between the weights of pigs in the bottom 25% of herds

Conclusions Large variations in growth rate were observed between the top and bottom 25% of herds in this study equating to an average difference of 18 days to attain an average live weight of 100 kg. This growth rate difference equated to a difference of 9 p/kg saved per carcass or £32,000 on a herd capable of finishing 1100 pigs per year. Greater variation in growth rate within herd was common in poorer performing herds, this may have contributed to the higher correlations on poorer performing herds observed between the weights of pigs at various stages. It is difficult to assess separately, the effects of management, disease or genetics as contributors to this variation and a more comprehensive investigation would be required.

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Use of near infrared spectroscopy to trace autochthonous breed of pig from Uruguay

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Introduction During the last 15 years, analytical methods (inmunological and enzymatic procedures) have been focused on the identification of meat species in raw, cooked and processed products. Near infrared reflectance (NIR) spectroscopy methods are attractive options due to the speed of analysis and minimal sample preparation. NIR spectroscopy has emerged in the last 30 years as a rapid method for testing the quality of intact samples from the light they reflect. One of the advantages of NIR technology is not only to assess chemical structures through the analysis of the molecular bonds in the near infrared spectrum, but also to build a characteristic spectrum that represents the "finger print" of the sample. The Pampa-Rocha breed of pig is originated from Uruguay and it has been a zootechnic resource related to medium to small farm production systems. The objective this study was to evaluate the potential use of visible (Vis) and near infrared reflectance (NIR) spectroscopy combined with chemometrics to identify and trace muscles from autochthonous breed of pig (Pampa-Rocha) and commercial cross (Pampa x Duroc) from Uruguay.

Materials and methods Forty-four (n= 44) pork muscle (*longissimus thoracis*) samples were obtained after a feeding trial which compared the use of both commercial feeds and pastures to finishing pigs (T1 = cross Duroc x Pampa 100% commercial feed; T2 and T3 cross Duroc x Pampa feed with pastures and two levels of commercial feed and T4 pure Pampa and feed as in T3). Pork muscles were taken from the 10th rib, wrapped in aluminium foil and kept on freezer (two weeks) before analysis. About 100 to 200 g of muscle was thawing at room temperature (20 - 22 ºC), homogenised during one to two minutes with a food multiprocessor blender (Philips RI - 3142, Brazil) and scanned both intact and minced in the reflectance mode (400 – 2500 nm) in a scanning monochromator NIRS 6500 (NIRSystems, Silver Spring, MD, USA). Spectral data was transformed into a NSAS format and exported into *The Unscrambler* (CAMO ASA, version 6.0, Norway) software for multivariate analysis. Principal component analysis (PCA), linear discriminant analysis (LDA) and soft independent modelling of class analogy (SIMCA) were used to attempt the classification of the muscles related with different breeds.

Results and discussion Figure 1 showed the PCA score plots for the two types of muscles analysed using the first two PC's. The score plots showed clusters of samples related with the two breed of pig used, namely Pampa and Duroc x Pampa, although some muscles overlap. PC1 explains 79% of the variation in the muscles analysed. Table 1 showed the number of both correctly and incorrectly classified muscles using LDA based on the Vis and NIR data. LDA models based on PCA scores from the Vis and NIR data, correctly classified 87% of the muscles belonging to the Pampa-Rocha breed of pig, and 68% of the Pampa x Duroc muscles.

Conclusions In this study it was showed that the use of Vis and NIR spectroscopy combined with chemometrics analysis (PCA, and LDA) might be used as a tool to trace muscles from an autochthonous breed of pig in Uruguay. The application of discriminant techniques (LDA) have shown potential to discriminate between muscles based on NIR spectra.

Table 1 Linear discriminate analysis (LDA) of Pampa-Rocha and Pampa x Duroc cross based on visible and near infrared data (CC= number of samples correctly classified, IC= number of samples incorrectly classified).

	CC	IC
Pampa -Rocha (n= 20)	2	14
Pampa x Duroc (n = 24)	16	8

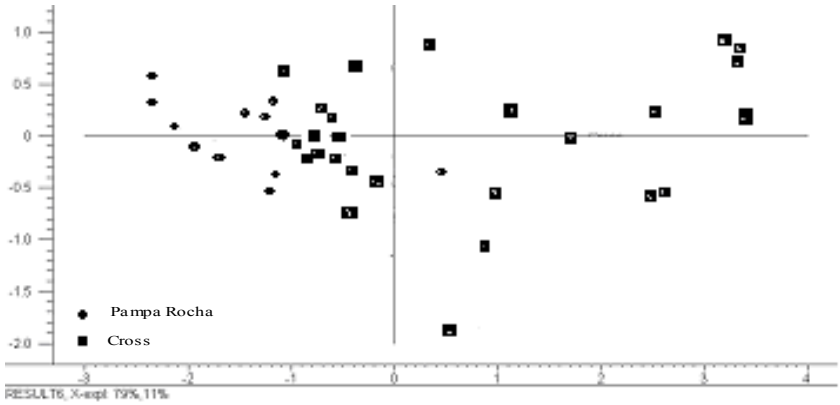

Figure 1. Principal component score plot of Pampa-Rocha and Pampa x Duroc cross based on visible and near infrared data.

The effects of lactation feeding strategies on the feed intake and litter performance of gilts

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Introduction In sows, excessive loss of live weight and body tissue reserves during lactation is associated with reduced subsequent reproductive performance. This is a particular problem in the gilt where the combination of low feed intake potential and relatively low body fat reserves often results in large amounts of body tissue catabolism during lactation to support milk production. The key to minimizing weight loss during lactation is to maximize feed intake of the gilt during lactation. Although there has been a substantial amount of research in this area, most of this has been carried out in research facilities. The objective of this research was to investigate the impact of feeding strategy (i.e., method and frequency of feeding) on feed intake in gilts during lactation under commercial conditions.

Materials and Methods Two experiments were conducted to evaluate the effects of lactation feeding strategy on gilt feed intake and piglet performance. Experiment 1 was conducted as a randomized complete block design and utilized 90 PIC C23 gilts allotted to one of three treatments: 1) Fixed amount of feed fed four times daily according to an increasing scale, 2) Feeding to appetite four times daily, and 3) Increasing restricted feeding scale for the first 10 days of lactation followed by to appetite feeding. Experiment 2 was conducted as a randomized complete block design and utilized 96 PIC C23 gilts allotted to one of two treatments. The treatments were 1) Hand fed to appetite and 2) *Ad libitum* access to feed via a self-feeder. The self feeder consisted of a feed dispenser with a gilt-operated valve that allowed the animal to dispense feed into the feeding trough in small amounts. In both experiments, gilts were weighed and backfat depth was measured at the P2 location upon entry to the farrowing house and at weaning. Cross-fostering was performed to equalize litter size and weight across treatments, and individual piglet weights were recorded at birth, 10 days, and weaning. Daily feed additions and refusals were recorded for individual gilts. Data were analyzed using the PROC MIXED procedure of SAS (SAS Inst. Inc., Cary, NC), and the model included the effects of replicate, farrowing room, and treatment.

Results In Experiment 1 (Table 1), gilts on Treatment 3 had lower ($P < 0.05$) average daily feed intake on days 1 through 7 (3.2, 2.8, and 2.1 ± 0.19 kg for Treatments 1, 2, and 3, respectively), however, average daily feed intake was similar ($P > 0.05$) on all other days, and for the overall lactation period (Table 1). There were no differences observed in any other gilt or litter performance measures. In Experiment 2, gilts on self feeders had higher ($P < 0.05$) average daily feed intake (4.4 vs. 4.1 ± 0.11 kg) and lost significantly less ($P < 0.05$) backfat (1.8 vs. 3.1 ± 0.43 mm) than hand fed gilts. All other gilt and litter performance measures were similar ($P > 0.05$) for the two feeding strategies.

Table 1 The effects of lactation feeding strategy on gilt and litter performance (Experiment 1).

	Treatment				
Item	1	2	3	SEM	*P* value
Overall average daily feed intake (kg/day)	3.9	3.7	3.5	0.13	0.215
Gilt body weight loss during lactation (kg)	42.1	39.5	40.4	2.12	0.667
Gilt backfat loss during lactation (mm)	2.1	3.7	2.9	0.60	0.150
Total litter weight gain (kg)	37.6	36.9	38.5	1.58	0.744

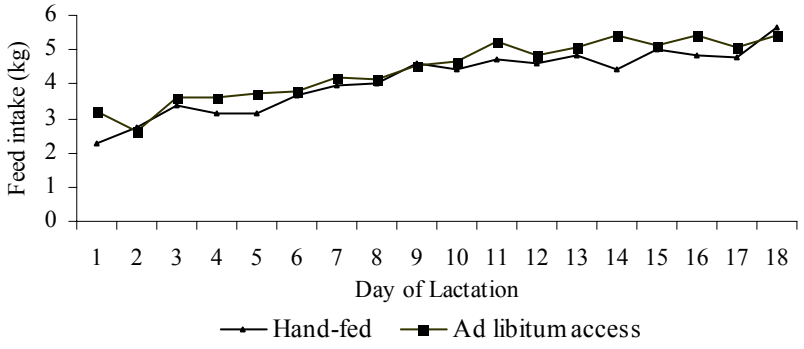

Figure 1. Average daily feed intake (Experiment 2)

Conclusions The results of this research suggest that allowing the gilt to determine the amount of feed dispensed into the feeder (via the self feeder evaluated) resulted in higher feed intakes and would obviously reduce the labour input into the feeding process. The use of self feeders therefore offers a practical approach to increasing feed intake under commercial conditions.

The effect of dietary protein supply on the health and performance of newly weaned pigs experimentally challenged with enterotoxigenic *Escherichia coli*

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Introduction The routine inclusion of in-feed antimicrobial growth promoters (AGPs) have long been used to improve performance and to protect against enteric disorders in newly weaned pigs (Hampson, 1994). However, due to increasing concerns about their use in pig production, they are being phased out within the EU. This will put the weaner pig at greater risk to post weaning enteric disorders such as post weaning colibacillosis (PWC), and accentuate the need for alternative, non-pharmaceutical, strategies for disease prevention. These strategies may include the manipulation of dietary protein supply, in order to minimise protein availability to intestinal microflora including enterotoxigenic *E. coli* (ETEC). The objective of the current experiment was to investigate the effects of dietary protein availability to intestinal microflora through manipulating protein quantity and/or quality on the health and performance of newly weaned pigs in the absence of AGPs when experimentally challenged with ETEC.

Materials and methods Thirty two pigs were weaned at 28.2 ± 1.18 days of age (±S.D) and 8.0 ± 0.98 kg bodyweight. They were balanced for initial weight, sex and litter and assigned to one of the four dietary treatments; a high protein (H, 230g CP/kg) and a low protein (L, 130g CP/kg) both with dried skimmed milk powder (DSMP) (H-DSMP and L-DSMP respectively), a high protein with DSMP replaced with soya meal (H-SOYA) and H-DSMP with added AGPs: ZnO (3100 mg/kg), $CuSO_4$ (170 mg/kg) and avilamycin (Maxus G200, Elanco; 40mg/kg) (H-AGP). All diets were formulated to contain 16.0 MJ DE/kg and were balanced for amino acid composition as a proportion of total protein, sodium and lactose content and were fed for 14 days. DSMP accounted for 0.25 of CP in the DSMP diets. From day 14 until day 42 a standard ration was fed to all animals. Piglets were challenged *per os* with 10^9 ETEC (*E. coli* O149: K91) on day 3 post weaning. Individual feed intakes, weight gain and faecal consistency (assessed through subjective scoring) were recorded. Fresh faecal samples were collected regularly to detect the presence and concentration of ETEC. On day 6 post weaning 4 pigs per treatment were slaughtered in order to assess gut health through digesta pH and lactobacilli to coliform ratio (L:C). All pigs were housed individually. Data were analysed using the REML procedure of Genstat. The animal experiment committee of SAC approved this work.

Results An increase in CP supply increased (P = 0.093) the number of ETEC shed (Figure 1), an increase in faecal score and a decrease in gut health as assessed by the L:C ratio of contents from the proximal colon on day 6 (H = 1.09 vs L = 1.17; P = 0.077). There was no effect of protein source on ETEC shedding (Figure 1), faecal score or gut health. Increasing CP supply and the inclusion of DSMP both led to increases in performance in the 3 day post challenge period (Table 1). The inclusion of AGPs led to a decrease (P<0.001) in the number of ETEC shed (Figure 1) and an increase in both gut health and performance. There was no significant difference in body weight between treatments at 10 weeks of age with an. overall mean (± SD) body weight of 34.8 (± 3.53) kg

Table 1. Effect of dietary protein supply on pig performance post weaning (days)

	ADFI (g/d)			ADG (g/d)		
Day	3-6	0-14	14-42	3-6	0-14	14-42
H-DSMP	280[a]	373	1125	238[a]	324	791
L-DSMP	239[b]	351	1055	213[b]	262	772
H-SOYA	233[c]	346	1214	173[a]	316	829
H-AGP	248[a]	330	1081	311[b]	335	745
S.E.D	26.8	26.4	83.2	25.6	47.5	69.7
Response	L*			L*		
	S**			A*		

Main effects * P < 0.05, *** P<0.001; where A = AGPs, L = Protein Level and S = Protein Source

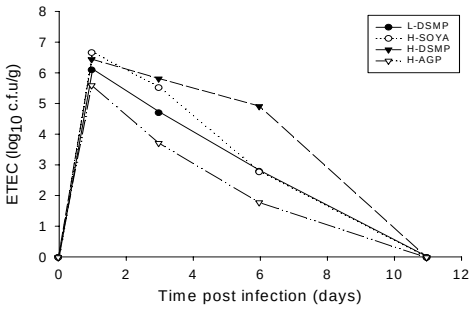

Figure 1 Effect of protein supply on the number of ETEC shed in faeces

Conclusion Feeding a diet high in CP increased the number of ETEC shed, reduced gut health and increased faecal score of pigs weaned at 4 weeks. Consequently, in order to apply the most suitable feeding regime to newly weaned pigs it is important to balance the trade off between the temporary detrimental effect on performance of feeding low CP levels and the positive effects of reducing occurrence and severity of PWC. This will depend upon the management system and stressors to which pigs may be exposed and is particularly important in an environment where AGPs are prohibited.

Acknowledgements This research was financially supported by ABNA Ltd, Frank Wright Ltd, Home-Grown Cereals Authority, Meat and Livestock Commission/British Pig Executive, Primary Diets Ltd and Provimi Ltd with match funding from Defra, through the Sustainable Livestock Production LINK programme.

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Effect of removing different proportions of pigs from pens at slaughter weight on the growth performance of the remaining animals

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Introduction When emptying finishing barns, it is common practice on U.S. operations to dispatch pigs over a two- to three-week period with the heaviest animals being selected first. However, there has been little research carried out under commercial conditions on the effect of removing different proportion of slaughter weight pigs from a pen on the subsequent growth performance of the remaining pigs. This study was carried out to investigate the effect of removing 0 to 45% of the heaviest animals from a pen on subsequent pig performance at slaughter.

Materials and methods Two studies were carried out in two different wean-to-finish facilities. In Study 1, crossbred pigs (n = 651) were used to evaluate removing three different proportions of the heaviest pigs from a pen as follows: 1) 0% - no pigs removed, 31 pigs/pen; 2) 10% - 3 pigs removed, 28 pigs/pen; 3) 20% - 6 pigs removed, 25 pigs/pen). Pig performance was measured from wk 22 (time of removal) to day 14 post-removal. Floor and feeder spaces per pig were 0.69 m^2 and 5.3 cm, 0.76 m^2 and 5.8 cm, and 0.85 m^2 and 6.5 cm for the 0, 10, and 20% removal treatments, respectively. In Study 2, crossbred pigs (n = 1,040) were used to evaluate removing four different proportions of the heaviest pigs from a pen as follows: 1) 0% - no pigs removed, 26 pigs/pen; 2) 15% - 4 pigs removed, 22 pigs/pen; 3) 30% - 8 pigs removed, 18 pigs/pen; 4) 45% - 12 pigs removed, 14 pigs/pen. Pig performance was measured from wk 20 (time of removal) to day 21 post-removal. Floor and feeder spaces per pig were 0.66 m^2 and 2.3 cm, 0.78 m^2 and 2.8 cm, 0.95 m^2 and 3.4 cm, and 1.22 m^2 and 4.4 cm for the 0, 15, 30, and 45% removal treatments, respectively. Mixed-sex pens (equal ratio of barrows to gilts) were used in Study 1 (n = 21) and single-sex pens were used in Study 2 (n = 40). In both studies replicates were formed from pens of pigs which were grouped on the basis of similar body weight, group size, and sex ratio. In both studies, pigs had free access to feed and water; feed intake data were only collected in Study 1. Data were analyzed as a randomized complete block design with pen considered the experimental unit. All data met criteria for normality and were analyzed using the PROC GLM procedure of SAS. Least squares means were evaluated using the PDIFF and STDERR options of SAS. The model used for analysis of data included effects of removal rate, barn and replicate nested within barn; the residual error was used to test main effects.

Results Growth performance data for the two studies are given in Table 1. Two analysis were conducted. The first compared growth performances between the entire group of pigs after removal (31 vs 28 vs 25 pigs/pen in Study 1 and 26 vs 22 vs 18 vs 14 pigs/pen in Study 2). The second analysis compared performance of the lightest 25 pigs in each treatment in Study 1 and the lightest 14 pigs in each treatment in Study 2. In Study 1, for the entire group of pigs daily gain (16%; $P < 0.05$) and feed intake (9%; $P < 0.05$) increased linearly with the proportion of pigs removed. Feed efficiency was not affected ($P = 0.11$) by removal treatment. In Study 2, growth rates increased (11%; $P < 0.001$) linearly with the proportion of pigs removed. No treatment differences in mortalities were observed in either study.

Table 1 Effect of pig removal treatment on pig performance for Studies 1 and 2

	Study 1				Study 2				
Item	0%	10%	20%	s.e.m	0%	15%	30%	45%	s.e.m.
Daily gain, g[a]	872	988	1,048	46.2*	861	942	956	976	19.5*
Daily feed intake, g[a]	2,848	3,064	3,137	53.7*	-	-	-	-	-
Gain:feed[a]	0.31	0.32	0.33	0.012	-	-	-	-	-
Daily gain, g[b]	847	970	1,034	45.8*	850	927	940	979	20.4*

* Data increased linearly as proportion of pigs removed increased, $P < 0.05$.
[a] Data represents the analysis of the entire group of each treatment.
[b] Data represents the analysis of the lightest 25 pigs in Study 1 and lightest 14 pigs/pen in Study 2.

Conclusions These results suggest that growth performance increases linearly as the proportion of pigs removed increases from 0 to 45%. Further research is needed to establish the independent effects of increased floor space, feeder space and pig removal on subsequent pig performance.

Metabolisable energy and digestible lysine levels for piglets – effects in performance and nitrogen retention during nursery initial-1 phase

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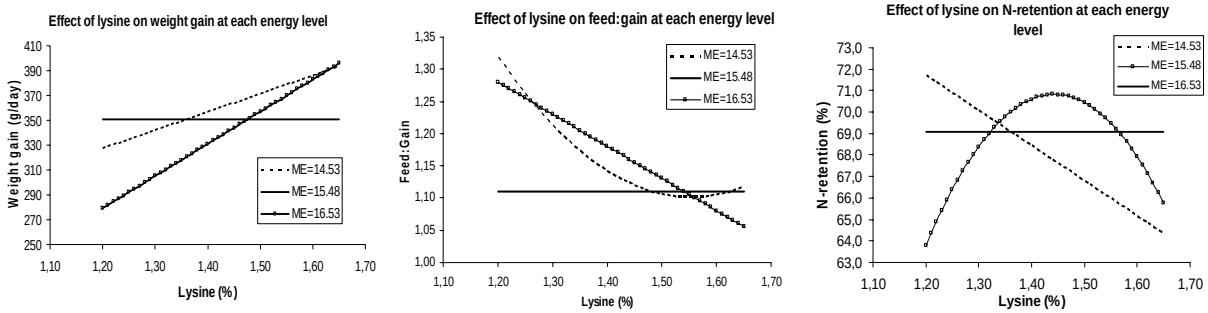
Introduction Low feed intake after weanimg is common, in which case it is recommended that concentrations of nutrients, such as lysine and energy by increased. Lysine is the first limithing aminoacid in most diets used for maximizing protein accretion during pig growth. A fundamental factor that determines protein depositionis the content of metabolizable energy (ME) which is need for protein anabolism. It is convenient that the dietetic energy and lysine (LYS) contents supply the requirements for maintenance and body mass accretion, according to specificities involved in modern pig production. The purpose of the present study was to investigate the effect of ME and digestible LYS levels on the performance and nitrogen (N) retention of piglets during the initial-1 phase and subsequent effects during the initial-2 phase.

Materials and methods Two hundred and sixteen piglets weighing an average of 6.98 ± 1.16 kg at weaning were alloted to a ramdomised block design with 12 treatments, 6 replicats and 3 animals per experimental unit. Tree ME levels (14.43, 15.48 or 16.53 MJ/kg) and 4 digestible LYS levels (1.20, 1.35, 1.50 or 1.65%) were usedin a 3 X 4 factorial. The diets contained corn, soybean meal (48% CP), dairy products, soybean protein concentrate (90% CP), corn gluten meal (60%CP), spray-dried plasma, synthetic aminoacids (L-lysine, L-threonine, D L-methionine, L-tryptophan and L-isoleucine), organic acids, zinc oxide and vitamin and mineral supplements. Performance was evaluated at the end of the initial-1 phase, when the piglets reached an average weight of 12 kg. Subsequent effects were evaluated at the end of the initial-2 phase, during which the piglets consumed a standard diet until they reached an average weight of 22 kg. A metabolism assay was conducted using 48 piglets (average weight = 9.31± 2.49 kg) in individual cage, obeying the same factorial plan. For the regression analyses of the performance ad metabolism data, only linear and quadratic effects were considered.

Results and discussion There was an interaction (P<0.01) of LYS and ME for weight gain, feed:gain and N retention. A linear (P<0.01) effect of LYS on weight gain was observed at ME= 14.43 and ME= 16.53. For feed:gain, a quadratic (P<0.05) and a linear (P<0.05) effect of LYS was observed at ME= 14.43 and ME= 16.53, respectively. For N retention, a negative linear (P<0.05) and a quadratic (P<0.05) effect of LYS was observed at ME= 14.43 and ME= 15.48, respectively. The quadratic effect of LYS at 14.43 ME diet in feed: gain ($\hat{Y}$ = 5,301501 – 5,4077786X + 1,740741X^2, R^2= 0.99) indicated 1.55% of digestible amino acid as optimum level or 1.07 g LYS/MJ ME intake. The linear improvement in feed: gain at ME= 16.53 indicate that piglets responded efficiently to LYS increase when the energy level is high. The dates indicate the necessary relation energy: lysine on nutrient utilization efficiency. The improvement of feed: gain in response to increase LYS in the present study confirms observations of De Rouchey *et al*. (2003) and Tokach *et al*. (2003). In ME= 15.48 diet the quadratic N retention increase ($\hat{Y}$ = – 176,0111 + 341,97019X – 118,44451X^2, R^2=0,98) indicated optimum level 1.44% LYS or 0.93 g LYS/MJ; intake above the optimal level failed to increase daily N retention, which can be explained by the reduction of net energy and high protein diet due excess amino acids deamination and N elimination. Similar interaction and N retention increase was observed by Urinec & Buraczewska (2003). There was no residual effect of treatments applied in initial-1 phase on initial-2 phase.



Conclusions The digestible lysine: metabolizable relation depends on diet energetic level and considered characteristic.

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Influence of wheat endosperm texture and degree of cook on digestibility of starch in the small intestine of the weaned piglet

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Introduction The piglet is susceptible to digestive upsets associated with the post-weaning growth check. To minimise this, heat treatment of cereals is used to cereal starch by reducing crystalinity, thereby increasing digestibility and dietary energy-yielding value (Lawrence, 1973a). However, benefits from heat treating are difficult to compare due to the number of processing variables used and, often, a failure to declare variables. Two trials were designed using controlled variables to assess the effect of heat processing (through micronisation and extrusion) on wheat nutritional value, and to determine whether responses to processing are affected by endosperm texture.

Materials and Methods The same 2 batches of wheat were used in each trial; trial comparisons are thus valid**.** Trial 1 examined wheats micronised at 250°C for 10 (Low cook) or 30 seconds (High cook). The trial was a 2 (Hard vs. Soft endosperm) x 2 (degree of micronising, low vs. high cook) factorial. Trial 2 examined endosperm texture and degree of extrusion (high SME, specific mechanical energy; 570 kJ kg^{-1} vs. low SME; 370 kJ kg^{-1}). Raw soft wheat was a control. Hard/Low SME wheat was not examined as rheology tests had not indicated much 'cook'. Wheats were ground through a 1.5mm screen and incorporated into diets at 580g/kg. Experimental period was for 14 days, with digesta samples taken at slaughter (day 0, n=2, and on days 2, 4, 6, 10 and& 14 post weaning; n=4/day; 2 per diet) at 0.25, 0.5 and& 0.75 proportionally along the small intestine from the gastric pylorus to the ileocaecal valve and freeze dried immediately on collection. Piglets slaughtered at Faecal samples were collected for 2 time periods (days 1-5 & 9-13 post weaning; 6 piglets/diet/period) analysed for acid insoluble ash (inert marker) and starch; coefficients of apparent starch digestibility were calculated. Data were subjected to analysis of variance using Genstat 7.

Results Extruded wheat diets had greater coefficients of starch digestion than the micronised diets (Table 1); digestibility was significantly affected by region of the small intestine, with coefficients increasing significantly from the proximal to distal end ($P = <0.001$). For micronised wheat, degree of cook proved significant with high cook resulting in greater digestibility coefficients than low cook ($P = 0.047$). There was no observed effect of endosperm texture, but a significant interaction was found between endosperm texture and cook level ($P = 0.004$).

For extruded wheat, significant differences were observed for endosperm texture (greater coefficients for soft wheat than for hard; $P = <0.001$) and degree of cook (high SME greater coefficients than low SME; $P = <0.001$). A significant interaction was observed between endosperm texture and day ($P <0.001$) but not between cook level and day ($P >0.05$). In both trials, starch coefficients in the proximal region of the small intestine (0.25) were negligible. Faecal apparent starch digestibilities were all greater than 0.970 for the micronised diets and 0.990 for extruded.

Table 1. Coefficients of starch digestion in small intestine

Trial	Diet	0.5 region	0.75 region
1. Degree of Micronising	Hard Wheat / Low Cook	0.451	0.722
	Soft Wheat / Low Cook	0.610	0.779
	Hard Wheat / High Cook	0.656	0.938
	Soft Wheat / High Cook	0.615	0.688
2. Degree of extrusion as measured by Specific Mechanical Energy (SME) expended	Soft Wheat / Raw	0.392	0.809
	Soft Wheat / Low SME	0.855	0.973
	Hard Wheat / High SME	0.803	0.968
	Soft Wheat / High SME	0.959	0.973

Conclusions These results demonstrate differences in digestibility of processed wheats within the small intestine of the young piglet; the lower the digestibility in this region the greater the amounts entering the large intestine. Degree of cook appears an important factor with a higher cook level yielding more beneficial results. Response to micronising was not affected by endosperm texture, but soft endosperm wheat responded better to extrusion than hard wheat under the conditions of the trial described. The high coefficient of the soft wheat / high SME diet within the 0.5 region suggests enhanced digestion and more rapid release of starch within this region of the tract. This could be an area of interest with regard to rate of starch breakdown within the weaned piglet.

Acknowledgements This research was financially supported by ABNA Ltd, Frank Wright Ltd, Home-Grown Cereals Authority, Meat and Livestock Commission/British Pig Executive, Primary Diets Ltd and Provimi Ltd with match funding from Defra, through the Sustainable Livestock Production LINK programme.

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The effect of the inclusion of formic acid and phytase in weaner pig diets upon feed intake, piglet performance and gut health

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Introduction The newly weaned pig has a limited capacity to secrete acid, which acts as a natural barrier against bacteria and activates pepsinogens (Kidder and Manners, 1978). We hypothesised that more acidic stomach conditions could be created by adding organic acid to the diet or by replacing inorganic phosphate, in the form of dicalcium phosphate, with phytase. The addition of formic acid to the diets of growing pigs has been shown to increase growth rate and feed conversion ratio. This effect was greater when supplementary phytase was also added to the diets, Jongbloed *et.al* (2000).

Materials and methods Two-hundred and fifty six pigs (Large White x Landrace) were weaned at 27.5 ± 2.2 (±standard deviation) doa and 7.9kg ±1.6 liveweight into 32 flat deck pens each of 8 pigs balanced for liveweight, sex and litter across treatments. Piglets were offered *ad libitum* access to diets (16 MJ DE/kg, 1.5g/kg lysine) containing either 0 or 6g/kg formic acid and supplemented with either 0 FTU/kg phytase and 10.0g/kg dicalcium phosphate or 500 FTU/kg phytase and 3.7 g/kg dicalcium phosphate for 18d following weaning. All diets were calculated to have similar levels of available phosphorus. Average Daily Gain (ADG), Average Daily Intake (ADI) and Feed Conversion Efficiency (FCE) were calculated on a pen basis for days 1 to 18. On day 6 post weaning 8 piglets from each treatment were slaughtered to measure intestinal morphology at 0.25, 0.50 and 0.75 along the small intestine. Data were analysed as a 2x2 factorial using theGLM procedures of Minitab 13.1.

Results
The overall performance of the pigs for days 1 to 18 post weaning was as expected for diets containing no growth promoters; ADG, ADI and FCE were 219 g/d, 291 g/d and 0.759 respectively. There was no effect of either formic acid or phytase inclusion upon any of the parameters measured (Table 1) nor were there any interactions between them. Gut parameters were within the range expected for day 6 post weaning. There was no effect of formic acid inclusion and/or phytase inclusion upon the small intestine structure at the 0.25, 0.50 and 0.75 sites measured. The inclusion of formic acid and/or phytase had no effect upon stomach pH.

Table 1 Effect of inclusion of formic acid and/or phytase upon ADG, ADI, FCE, stomach pH, villus height and crypt depth post weaning.

N=16	Acid		Phytase			Significance		
	No	Yes	No	Yes	s.e.m	Acid Effect	Phytase Effect	Interaction
Weaning age (d)	27.5	27.5	27.4	27.5	0.37	NS	NS	NS
Weaning Weight (kg)	7.9	7.9	7.9	7.9	0.29	NS	NS	NS
Day 18 Weight (kg)	11.8	11.8	11.8	11.8	0.35	NS	NS	NS
ADG 1-18 (g/d)	217	222	217	222	7.4	NS	NS	NS
ADI 1-18 (g/d)	285	297	292	291	9.2	NS	NS	NS
FCE 1-18	0.768	0.750	0.750	0.767	0.0238	NS	NS	NS
Stomach pH	3.33	2.77	2.93	3.18	0.270	NS	NS	NS
Villus height 0.25 (μm)	234	236	235	235	14.1	NS	NS	NS
Crypt depth 0.25 (μm)	245	261	256	250	14.2	NS	NS	NS
Villus height 0.50 (μm)	257	236	247	247	17.0	NS	NS	NS
Crypt depth 0.50 (μm)	230	231	230	232	11.1	NS	NS	NS
Villus height 0.75 (μm)	211	211	211	211	10.4	NS	NS	NS
Crypt depth 0.75 (μm)	201	218	211	208	10.5	NS	NS	NS

Conclusions In this study inclusion of formic acid and/or phytase in weaner pig diets for the first 18 days post weaning, had no effect upon feed intake, piglet performance or gut structure. This result is perhaps unsurprising given that these additives did not reduce stomach pH as was hypothesised. Inclusion of phytase to replace 63% dicalcium phosphate did not adversely affect piglet performance, feed intake or gut structure thus indicating that phytase inclusion can adequately compensate for reduced inorganic phosphorus in the diets of newly weaned piglets.

Acknowledgements This research was financially supported by ABNA Ltd, Frank Wright Ltd, Home-Grown Cereals Authority, Meat and Livestock Commission/British Pig Executive, Primary Diets Ltd and Provimi Ltd with match funding from Defra, through the Sustainable Livestock Production LINK programme.

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Performance effects of graded levels of wheat inclusion in diets for growing and finishing pigs

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Introduction Wheat and barley are common ingredients in diets for growing and finishing pigs. Traditionally, barley has been included at higher inclusion levels due to a lower purchase cost and also because of reports that high levels of wheat inclusions (700g/kg) can cause stomach ulceration (Nielson and Ingvartsen 2000) and pig grading problems (Hanrahan and O'Grady 1984). However, wheat contains a higher digestible energy (DE) content than barley (15.2 vs. 13.9 MJ/kg DM) and therefore substituting wheat for barley should result in better pig performance (Van Barneveld 1999). The aim of the study was to investigate the effect of increasing the level of wheat in growing/finishing pig rations on performance and stomach ulceration.

Materials and methods Four experimental diets were formulated to contain wheat: barley at 400:300, 500:200, 600:100 and 700:0g/kg. The diets also contained (g/kg): soyabean meal 192, soya oil 30, moalferm 30, water 20, lysine 3 and minerals and vitamins 25. The formulated DE contents were (MJ/kg) 14.2, 14.0, 13.9 and 13.8 for the 700, 600, 500 and 400 g/kg wheat diets respectively. In the first study, the diets were offered *ad libitum* to a total of 480 crossbred (Large White x Landrace) pigs over six replicates. The pigs were housed in groups of 20, which were balanced for weight and gender. Average daily gain (ADG), feed intake and feed conversion ratio (FCR) were determined between 28kg and 100kg (finish age was 21 weeks plus 5 days). At finish, one pig from each pen was slaughtered, the gastrointestinal tract eviscerated and the stomach dissected and scored for ulceration on a scale of 0-10, according to the method of Nielsen and Ingvartsen (2000). In the second study, the diets were offered *ad libitum* to 72 individually housed pigs from 10 weeks of age until finish (100kg). ADG, feed intake and FCR were determined between 10 weeks of age (27kg) and finish. The results were analyzed by ANOVA using Genstat 6.

Results There was no significant difference in the performance of pigs offered the diets with different wheat and barley inclusion rates (Table 1). The level of wheat inclusion had no effect on backfat depth at the P_2 position or on killing out percentage. There was little evidence of stomach ulceration across the treatments with scores of 1.0 or less on the scale of 0-10. Furthermore, the level of wheat inclusion did not significantly effect the degree of stomach ulceration. The performance of individually housed pigs was superior to that of group housed pigs.

Table 1 Effect of graded levels of wheat inclusion on performance and incidence of stomach ulceration in pigs

	400g/kg wheat	500g/kg wheat	600g/kg wheat	700g/kg wheat	SEM	P
Group performance						
Average daily gain (g/d)*	879	850	897	857	14.4	NS
Feed intake (g/d)	2114	2040	2133	2011	41.3	NS
FCR	2.40	2.40	2.38	2.35	0.354	NS
P_2 (mm)	12.9	12.1	13.7	13.5	0.56	NS
Kill out %	76.2	76.5	75.8	76.5	0.78	NS
Stomach ulceration[+]	0.64	0.43	0.08	1.01	0.484	NS
Individual performance						
Average daily gain (g/d)*	1002	1012	1027	970	21.3	NS
Feed intake (g/d)	2351	2387	2411	2301	43.1	NS
FCR	2.35	2.37	2.36	2.38	0.034	NS

*Pigs housed individually reached the weight of 100kg approximately 12 days earlier than those housed in groups
[+] Scale of 0-10 (Nielsen and Ingvartsen 2000)

Conclusions There was no significant effect of increasing wheat inclusion on pig performance, carcass quality or on the occurrence of stomach ulceration. There was a tendency for feed intake to decrease as wheat level increased, which may be attributed to the pigs eating to satisfy energy requirements as wheat contains a higher level of DE (Weatherup *et al* 2002). Pigs consumed more feed when housed individually which is in keeping with that reported by Weatherup *et al* (2002).

Acknowledgements This work was funded by the Department of Agriculture and Rural Development for Northern Ireland.

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Level of parasitism in organic and non-organic dairy farms in Scotland

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Introduction

The health and welfare of organically-farmed livestock are of concern due to restrictions on the use of veterinary medicines. Non-organic systems control nematodes using prophylactic treatment with anthelmintics. Routine drenching is banned within organic agriculture, and farmers are encouraged to find alternative methods of control, for example grazing management practices. This study investigated the level of nematode parasitism in organic and non-organic dairy herds around Scotland.

Materials and Methods

Eleven organic dairy farms were matched with non-organic dairy farms in a similar location, in order for them to be subjected to similar environmental conditions, such as average rainfall, daily temperature, and length of grazing season. These farms were visited three times during the summer grazing season for determination of gastrointestinal nematodes. Six organic and six non-organic farms were picked at random from the 11 pairs and visited once during the winter for determination of liver fluke. Faecal samples were taken from the number of youngstock that was equal to 10% of the total herd size (between 18 and 20 animals on each farm). Samples were taken at random as they were produced meaning that they were fresh and there was less chance that an animal would be sampled more than once. Young cattle in their first grazing season were faecal sampled for this purpose and coat condition was also recorded on a scale of one to three on the same number of animals.

Mean FEC for each farm was log-transformed and a repeated measures ANOVA was used to test for the effect of farm type (organic vs non-organic) and time. Although different animals were sampled at each timepoint, a repeated measures model was used as the experimental unit was the farm. The analysis was blocked for pair within sample period. A Mann-Whitney U Test was performed to compare the variation between FEC organic and non-organic farms at the three time points.

Coat condition score was analysed based on the proportion of 1 scores (flat, glossy coat) recorded per farm. These proportions were then given an arcsine transformation and analysed to identify differences between organic and non-organic farms using an ANOVA. Different individuals were sampled between the first, second and third time points, therefore analysis of the proportions gave results at farm level.

Presence or absence of liver fluke in the faeces of every animal sampled on each farm was analysed using a Generalised Linear Mixed Model (GLMM) (Breslow and Clayton, 1993). A binomial error distribution was assumed and a logistic link function used. The identity number of the farm was fitted as a random effect.

Results

No significant difference was found between the mean faecal egg counts (FEC) of organic and non-organic farms ($P > 0.05$) (figure 1) indicating that control strategies implemented by organic farmers (mostly evasive grazing management) seem to be working. There was, however, a significant difference between the three time points ($P < 0.05$) with an increase in FEC over the grazing season. This was expected due to the lifecycle of gastrointestinal parasites. Additionally, no significant difference was found between the variation of FECs on organic and non-organic farms at the three time points ($P>0.05$). No significant difference was found between the coat conditions of organic and non-organic farms ($P > 0.05$). Similarly, no significant difference was found between the proportion of animals positive for liver fluke on the two farm types ($P > 0.05$) (figure 2).

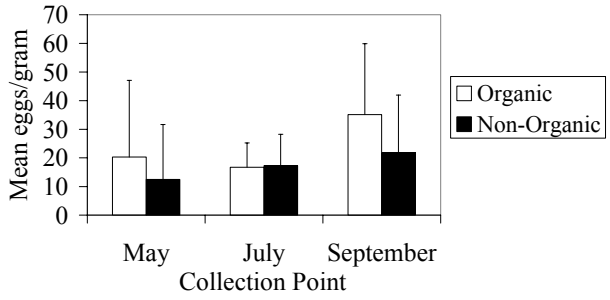

Fig 1: Levels of Parasitism

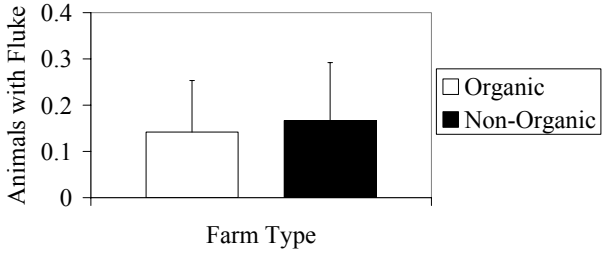

Fig 2: Mean Proportion of Animals with Fluke

Conclusion

The results of this study suggest that concerns about restrictions on prophylactics to treat internal parasites adversely influencing welfare may be unfounded in Scottish dairy calves. Furthermore, this study implies that levels of parasitism in Scottish dairy calves are low. However, as levels of parasitism vary with annual weather conditions and the between-farm variation was large, further study may be required.

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Effect of dietary phosphorus content on the fertility of dairy cows over four successive lactations

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Introduction Excessive phosphorus (P) inputs to agricultural systems contribute to the problem of eutrophication of water courses, with dairy farms often implicated as potential sources of P pollution. To help address this issue, there is currently considerable interest in reducing the P content of dairy cow diets. For example, recent research has indicated that the P content of dairy cow rations can be reduced without any negative effect on animal performance (Wu and Satter, 2000). The objective of the current study was to examine the effect of reducing dietary P levels over four successive lactations, on dairy cow fertility.

Materials and methods One hundred winter calving, primiparous, Holstein Friesian dairy cows (80 in Year 1, and 20 in Year 2) were allocated to diets containing either 'normal' or 'reduced' contents of dietary P (50 animals to each P treatment) post-calving. Animals remained on their specified P level for either four (Year 1 animals) or three (Year 2 animals) complete lactations. During the winter period, animals were offered diets containing grass silage and maize silage (approximately 70 : 30 DM basis), supplemented with between 10.0 – 13.0 kg concentrate/cow/day (according to silage quality). During the summer period (Years 1 and 2) half of the animals grazed both by day and by night, while the remaining animals grazed by day, and were housed by night and offered grass silage. During Years 3 and 4, all animals grazed by both day and night. Concentrate feed levels during the summer periods were either 3.0 or 4.0 kg/day, according to grazing conditions. During both the winter and summer periods, total ration phosphorus content was adjusted by modifying the amount of P in the concentrate component of the diet offered. During the winter period, concentrates were formulated to contain either 4.4 or 7.2 g P/kg DM (approx.), while summer rations were formulated to contain either 3.5 or 6.7 g P/kg DM (approx.). The concentrate containing the lower P level was formulated without additional mineral P being added, while di-calcium P was added to this concentrate to produce the higher P concentrate. Throughout the study, all animals were bred by artificial insemination, with AI commencing early December, and continuing until late June the following year. No animal was inseminated or treated with fertility drugs before day 42 post-calving. Milk progesterone concentrations were measured twice weekly until day 52 post-calving, with Commencement of Luteal Activity (CLA) defined as two consecutive progesterone rises above 3ng/ml. Data were analysed by ANOVA as a 2 x 4 factorial design, the model including 'Year', lactation number and P level.

Results Animal performance data (milk yields and dry matter intakes) from this study have been presented at this conference. Dietary phosphorus level had no significant effect on any of the fertility parameters presented ($P>0.05$). Lactation number had a significant effect on days to CLA, conception rates and the proportion of cows in-calf at the end of the breeding season ($P\leq0.05$). A significant P level x lactation number interaction was observed for conception rates to first plus second AI.

Table 1 Effect of dietary P level over four successive lactations on cow fertility

	'Normal P ration'	'Reduced P ration'	SEM	Sig		
				P	Lact	P x Lact
Pre-day 42						
Proportion of cows showing luteal activity	0.59	0.62	0.42	NS	NS	NS
Days to CLA	22.8	24.9	0.87	NS	**	NS
Proportion of cows with observed heat	0.26	0.22	0.037	NS	NS	NS
Conception						
First AI (proportion)	0.37	0.34	0.042	NS	*	NS
First + second AI (proportion)	0.67	0.60	0.042	NS	***	*
100 day in-calf rate (proportion)	0.50	0.42	0.044	NS	NS	NS
Mean number of services per cow	2.0	2.1	0.10	NS	NS	NS
Proportion of cows in calf at end of breeding season	0.89	0.82	0.030	NS	***	NS

Conclusions While dietary phosphorus level had no effect on any of the fertility parameters measured, a number of fertility parameters became significantly poorer with increasing lactation number. The results of this study are in line with the findings of a review of 13 studies involving normal and reduced P diets (approximately 400 animals/diet), which indicated that fertility was unaffected by level of phosphorus in the diet (Satter and Wu, 1999).

Acknowledgements Funded by DARDNI, AgriSearch, John Thompsons and Sons Ltd and Devenish Nutrition Ltd.

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Effect of dietary phosphorus content on the performance of dairy cows over two successive lactations

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Introduction Phosphates represent one of the primary sources of water pollution in Northern Ireland, an issue of particular concern in view of existing EU legislation, such as the Water Framework Directive. Inputs of phosphorus (P) to agricultural systems contribute to this problem, with dairy farms often implicated as a potential source of P pollution. Part of this problem can be attributed to the fact that large quantities of P are imported onto dairy farms in concentrate feeds which tend to be high in P (7.3 g P/kg DM in a survey of 35 winter concentrates for dairy cows). However, recent research has indicated that the P content of dairy cow rations can be reduced without any negative effect on animal performance (Wu and Satter, 2000). Nevertheless, the rations offered in this study were very different from those commonly offered within grassland based dairy systems in the UK. Consequently a study was established to examine the effect of reducing dietary P levels on cow performance, with the study being conducted over two successive lactations. The long-term nature of the study was deemed necessary as cows can mobilise bone phosphorus reserves for milk production when dietary phosphorus levels are inadequate.

Materials and methods One hundred primiparous winter calving Holstein Friesian dairy cows (80 in Year 1, and 20 in Year 2) were allocated to diets containing either 'normal' or 'reduced' contents of dietary P (50 animals to each P treatment) post-calving. Animals remained on their specified P treatment for two complete lactations. During the winter period, animals were offered diets containing grass silage and maize silage (approximately 70 : 30 DM basis), supplemented with between 10.0 – 13.0 kg concentrate/cow/day (according to silage quality). During the summer periods half of the animals grazed both by day and by night, while the remaining animals grazed by day, and were housed by night and offered grass silage. Concentrate feed levels during the summer periods were either 3.0 or 4.0 kg/cow/day, according to grazing conditions. During both the winter and summer periods, total ration phosphorus content was adjusted by modifying the amount of P in the concentrate component of the diet offered. During the winter period, concentrates were formulated to contain either 4.4 or 7.2 g P/kg DM (approx.), while summer concentrates were formulated to contain either 3.5 or 6.7 g P/kg DM (approx.). The concentrate containing the lower P level was formulated using feed ingredients low in phosphorus and without additional mineral P being added, while di-calcium P was added to this concentrate to produce the higher P concentrate.

Results Ninety-four animals completed lactation 1, while 70 animals completed lactation 2. Dietary P level had no significant effect on mean daily DM intake during the winter period in either lactation. Similarly, dietary P level had no effect on total lactation milk yield, total lactation milk fat and protein concentrations, or end of lactation live-weights in either lactation.

Table 1 Effect of dietary P level on animal performance

		'Normal P ration'	'Reduced P ration'	SEM	Sig.
Total DM intake (kg/day, winter)	Lactation 1	17.6	17.4	0.18	NS
	Lactation 2	19.9	19.6	0.17	NS
Total lactation milk yield (kg)	Lactation 1	7520	7480	135.2	NS
	Lactation 2	8246	8419	148.6	NS
Milk fat (g/kg)	Lactation 1	40.6	41.2	0.59	NS
	Lactation 2	39.9	40.4	0.61	NS
Milk protein (g/kg)	Lactation 1	34.6	34.6	0.25	NS
	Lactation 2	34.1	34.2	0.29	NS
Live-weight at drying off (kg)	Lactation 1	546	541	6.4	NS
	Lactation 2	625	614	6.8	NS

Conclusions The results of this study indicate that animals managed on diets containing reduced levels of dietary P over a two years period demonstrated no adverse effects in terms of DM intake, milk yield, milk composition or body tissue reserves. This suggests that dietary phosphorus levels for dairy cows could be reduced, with this having the potential to reduce phosphorus losses to the environment.

Acknowledgements Funded by DARDNI, AgriSearch, John Thompsons and Sons Ltd and Devenish Nutrition Ltd.

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Effect of additional concentrate supplementation for dairy cows at three time points in lactation

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Introduction Optimisation of supplementation of forage with concentrates is essential to improve efficiency and economic sustainability of dairy cow production systems, as well as reducing the environmental impact of intensive dairy farming. However, continuing fertility and health problems indicate that we are not responding effectively to the needs of high genetic merit dairy cows in terms of current feeding and management strategies. A programme was initiated to examine the consequences of nutritional manipulation of dairy cow diets on animal performance in order to attempt to re-programme the nutrient partitioning of the animal towards maintenance of body reserves rather than excessive milk yield and to potentially alter the lactation curve to give a flatter curve of milk production.

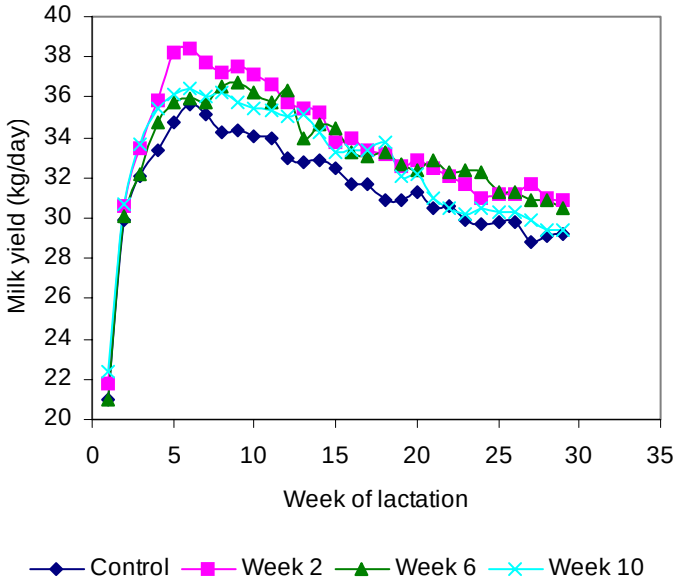
Materials and Methods Eighty four Holstein Friesian cows (42 primiparous, 42 multiparous) were allocated to four treatment groups according to parity, liveweight and for multiparous animals, lactation number and milk yield from the previous lactation, in order to measure the response to feeding additional concentrate (+4kg/day) at three different stages of lactation. Groups consisted of a control treatment (C) in which animals received a forage and concentrate TMR mixture (50:50 DM basis) and three treatments (2, 6, and 10) in which animals received an additional 4kg of concentrate from week two, six or ten of lactation onwards. The animals remained on the trial until week 29 of lactation. Individual daily milk yields and dry matter (DM) intakes were recorded for all cows while milk composition, liveweight and condition score were recorded on a weekly basis (Table 1). Data were analysed by ANOVA (Genstat) and the effects of time, treatment and parity were assessed.

Results

Table 1 Average performance of cows and heifers from calving to 29 weeks of lactation

	Treatment					
	C	2	6	10	SEM	P
Total DM intake (kgDM/day)	17.0	19.2	18.7	18.7	0.622	<0.05
Milk yield (kg/day)	31.5	33.5	33.1	32.0	1.258	NS
Fat + Protein corrected yield (kg/day)	30.7	32.6	32.1	32.0	1.110	NS
CS change (scale 1-5)	-0.10	0.00	+0.04	+0.05	0.112	NS

Figure 1 Average milk yield of cows and heifers



Performance data are presented in Table 1. Supplemented treatment groups had increased total DM intake compared to control animals ($P<0.05$). Concentrate supplementation had no significant effect on mean milk or fat plus protein corrected milk yield. However, weekly milk yield data presented in Figure 1 suggest positive responses in mean weekly peak milk yield by offering additional concentrates from either week 2 or week 6, and similar sustained post intervention yield responses. Introduction of the concentrate supplement from week 10 only produced a transient positive response in yield. Cows on treatments with additional concentrate, particularly treatments 6 and 10, tended to gain condition relative to the control treatment.

Conclusions Additional concentrate supplementation tended to increase milk production, however the milk yield increase tended to be lower when supplementation was introduced later in lactation. Additional concentrate supplementation also tended to prevent loss of body condition and may reduce the extent of negative energy balance in early lactation.

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Dietary fatty acid supplementation and immune response in cattle

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Introduction The imminent ban on the use of antibiotics and antibiotic growth promoters in animal feed in EU member states from 1st January 2006 provides a new focus for the development of alternative strategies to augment immune function in farm animals. In recent years there has been an increased interest in the ability of certain nutraceuticals to enhance immune response and consequently improve health status. For example, there is now some evidence for non-ruminants that consumption of polyunsaturated fatty acids (PUFA) including eicosapentaenoic acid (EPA; 20:5*n*-3), docosapentaenoic acid (DPA; 22:5 *n*-3*)* and docosahexaenoic acid (DHA; 22:6*n*-3), belonging to the *n*-3 fatty acid series, can affect immune function. However, both positive and negative effects have been reported depending on the specific fatty acid and amount used (see review by Calder, 1998). There is a dearth of information, however, on the possible effects of PUFA on immune status in ruminants. One recent study (Wistuba *et al*., 2005) suggests that fishoil may act as an immune stimulant in grazing beef calves. The objective of the current study was to examine the effect of level of dietary omega-3 fatty acid supplementation on the haematological profile and indicators of immune function in cattle.

Materials and methods Heifers (n=31) with a mean ± s.e. live weight of 441.71 ± 5.58 kg were blocked on liveweight and BCS and randomly allocated within block to one of four dietary treatments. All animals were individually fed a concentrate (6 kg DM) and straw (1.5 kg DM) based ration and supplemented with one of four levels of a fishoil supplement to provide (1) 0g (Control; n=6); (2) 59g (n=6); (3) 118g (n=8) or (4) 236 g (n=11) of dietary EPA plus DHA combined on a DM basis. All diets were isolipid and isonitrogenous (14% CP). Animals were blood sampled prior to allocation to diet (Day – 34), and again on days 30 and 43 to establish the haematological variables, including white blood cell counts (WBC), lymphocyte (Ly), granulocyte (Gr) and monocyte (Mo) numbers, haemoglobin (HgB), red blood cell numbers (RBC), hematocrit (%) mean corpuscular volume (MCV), mean corpuscular haemoglobin (MCH), and red blood cell distribution width (RDW). Haematology variables were analysed using an automatic Celltac haematology analyser. Normally distributed data were analysed using repeated measures ANOVA (PROC MIXED, SAS, v. 8.2) and nonparametric variables were analysed using the Kruskall-Wallis test following a rank transformation.

Results The effect of dietary treatment on normally distributed haematological variables is presented in Table 1. There was no effect on any of the red blood cell related variables ($P > 0.05$) though most of these variables were affected by day of sampling. There was no effect of treatment or day of sampling on platelet numbers ($P > 0.05$). There was no difference ($P > 0.05$) between treatment groups in baseline WBC, GR, Mo or Ly. After 30 days on treatment, WBC, GR, Mo and Ly were higher on the fishoil fed animals than the control but there was no difference within fishoil-supplemented treatments ($P > 0.05$). There was no difference between treatments ($P > 0.05$) in any of the WBC related variables at day 43 of the experimental period.

Table 1 Effect of dietary treatment on normally distributed haematological variables

	0g	59g	118g	236g	Trt	Day	Trt x Day
HgB (g/dl)	12.43±0.375	13.30±0.372	12.79±0.326	12.06±0.273	NS	*	NS
RBC ($x10^{12}$/l)	9.58±0.290	9.06±0.288	9.06±0.253	8.90±0.211	NS	*	NS
HCT (%)	34.51±1.032	36.53±1.024	35.29±0.898	33.86±0.753	NS	*	NS
MCV (fl)	36.20±1.185	40.39±1.181	39.08±1.028	39.11±0.871	NS	NS	*
MCH (pg)	13.08±0.429	14.68±0.428	14.12±0.372	13.59±0.316	NS	**	NS
RDW (%)	16.97±0.335	16.50±0.331	17.21±0.292	16.62±0.242	NS	**	NS

Conclusion Blood concentrations of all the variables were within normal ranges across all four treatments. The results of this study suggest that supplementation of cattle with omega-3 fatty acids is unlikely to affect immunologically related blood variables in the absence of an immunological challenge.

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The effect of including a yeast culture on the intake and performance of high yielding dairy cows fed a diet high in starch

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Introduction Benefits when yeast cultures are added to ruminant diets may include increased intake, ruminal fibre digestion, reduced ruminal accumulation of lactic acid, increased growth efficiency in beef cattle and increased milk yield in dairy cows. However, the effects on milk performance can be variable, with small or negative responses being reported, as well as positive effects. Part of this variation may be attributable to differences in animal potential and basal diet, and it has been suggested that the greatest benefit may occur in high yielding cows when fed diets high in high starch. The objective of the current experiment was to determine the effect of a yeast culture on the intake and performance of high yielding dairy cows fed a complete diet that was high in starch.

Materials and methods Forty Holstein-Friesian dairy cows that were 69 d into lactation were allocated to one of two treatments: Control and Treatment, for 12 weeks. All cows received a total mixed ration containing (proportion of DM): 0.36 maize silage, 0.17 grass silage, 0.15 caustic wheat, 0.12 rapeseed meal, 0.12 soyabean meal, 0.04 standard concentrate, 0.04 molasses, and 0.007 minerals and vitamins, that was predicted to contain 220g/kg DM starch. In addition, treatment cows received Yea-Sacc®1026 (Alltech (UK) Ltd., Stamford, Lincs) at the rate of 10g/cow/day and mixed in with the diet. Feeds were offered fresh daily at 1.05 of *ad libitum* intake with refusals collected twice weekly. The diets were fed through Insentec roughage intake feeders fitted with an automatic animal identification and electronic forage weighing system. Cows were milked twice daily at 06.00 and 17.00 h. Milk yield was recorded at each milking and samples taken weekly for subsequent analysis of fat and protein. The cows were weighed and condition scored after the evening milking during the week prior to the start of the experiment and then fortnightly. Rumen fluid samples were taken by rumenocentesis from the ventral sac at 1100 h during weeks 6 and 12 of the experiment from 8 animals per treatment. Data was analysed by ANOVA as a randomised block design using Genstat 7.1 (VSN Int. Ltd., Oxford, U.K.).

Table 1 Intake and milk performance in dairy cows fed diets without (Control) or with (Treatment) Yea-Sacc®1026 included at 50g/cow/day.

	Control	Treatment	s.e.d.	P-Value
Intake (kg DM/d)	21.9	21.8	0.62	0.933
Yield (kg/d)	35.3	36.2	0.71	0.215
Fat (40g/kg) corrected yield	33.5	34.5	1.50	0.541
Fat (g/kg)	38.1	38.1	0.15	0.992
Protein (g/kg)	32.1	31.6	0.05	0.268
Fat (kg/d)	1.34	1.38	0.060	0.570
Protein (kg/d)	1.13	1.14	0.028	0.714
Food efficiency (kg milk/kg feed DM)				
Average	1.61	1.68	0.052	0.351
Weeks 1-4	1.66	1.76	0.039	0.040
Weeks 5-8	1.69	1.70	0.055	0.811
Weeks 9-12	1.52	1.61	0.061	0.162
Live weight change (kg: wks 0-12)	12.1	14.2	14.2	0.887
Condition score change (wks 0-12)	0.32	0.29	0.073	0.678
Ruminal pH	6.3	6.4	0.43	0.825

Results There was no effect (P>0.05) of treatment on daily DM intake, which averaged 21.9 kg/d (Table 1). There was also no effect (P>0.05) of treatment on average milk yield, although yield was higher (P<0.05) during week 5 of treatment in cows receiving Yea-Sacc®1026 (Fig. 1). Milk fat content (g/kg), fat yield (kg/d) and milk fat adjusted (40 g/kg) yield were similar between treatments. Similarly, milk protein content and yield were not (P>0.05) affected by treatment. The efficiency of food use for milk production (kg milk yield per kg DM intake) was improved (P<0.05) during weeks 1 to 4 in cows receiving the yeast supplement, but not during any of the other time periods. Neither live weight nor body condition score change over the 12 week period were affect by treatment (P>0.05). Ruminal pH was similar between treatments, averaging pH 6.35.

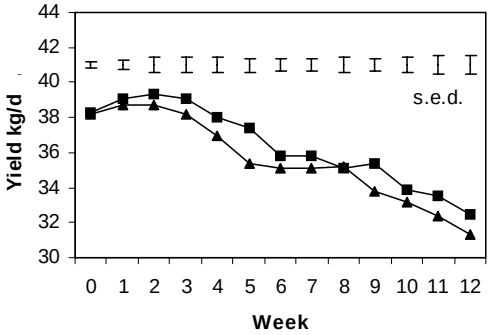

Figure 1 Weekly milk yield in cows fed diets without (Control: ▲) or with (Treatment: ■) Yea-Sacc®1026 included at 50g/cow/day.

Conclusions. Average milk yield was not significantly affected by treatment, although cows fed Yea-Sacc®1026 had a higher milk yield during week 5 of the experiment. There was no effect of treatment on milk composition or component yield, although milk efficiency (kg milk/kg DM feed intake) was improved in cows receiving a yeast supplement in earlier lactation when milk yield was highest.

The association between postpartum mastitis and days open in Iranian Holstein cows using survival analysis methodology

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Introduction Reproduction and health in addition to milk production are the principal factors that are influencing dairy farm profitability. The dairy industry in Iran has changed dramatically in the last decade. The shift toward more productive cows and larger herds is associated with a decrease reproductive efficiency (Heravi Moussavi *et al.*, 2004). Increased knowledge about the principal causes of reduced fertility is essential. It was shown that clinical mastitis during early lactation markedly influenced reproductive performance of Jersey cows (Barker *et al*., 1998). Recently, survival analysis was used to assess the effects of diseases on days from calving to conception. Survival analysis allows for a more appropriate management of censored data and time-dependent covariates, which are very common in the study of variables such as days open, days to first breeding, and days from first breeding to conception (Maizon *et al.,* 2004). The objective of this study was to evaluate the effect of postpartum mastitis on days open in Iranian Holstein dairy cows.

Materials and methods A data set of 13,000 lactations during 1996 to 2003 from five large dairy herds were used for this study. Mastitis definitions included mastitis treated by systemic therapy or mastitis treated by local therapy, acute mastitis or chronic mastitis, all cases of clinical mastitis, treatment for mastitis, and clinical mastitis diagnosed by a veterinarian. According to the parturition date, days open and mastitis data were analyzed by survival analysis. Survival statistical analysis was performed using the statistical software package JMP (SAS Institute Inc., NC, USA). Mastitis was considered if it occurred after calving and 210 d after calving (uncensored data) or censored time (greater than 210 d). The days open was defined for an uncensored cow as the interval from 35 d after calving to conception and for a censored cow as the interval from 35 d after calving to either culling or 350 d after calving, whichever came first.

Results The descriptive statistics of days open in healthy and mastitic cows are shown in Table 1. Figure 1 shows the survival analysis curves for days open in healthy and mastitic cows. The number of days to conception for cows with mastitis was greater than that for the healthy cows. The Log-Rank and Wilcoxon tests showed significant difference between the two groups (P<0.0001).

Table 1 Descriptive statistics of days open in healthy and mastitic cows

Item	Mean	Median	Lower 95%	Upper 95%	25% failure	75% failure	SE
Healthy	124.32	101	98	103	69	154	0.02
Mastitic	143.65	116	106	127	75	195	0.27

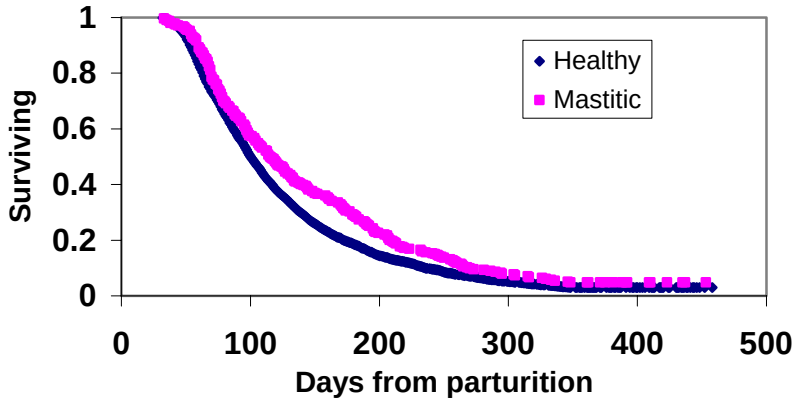

Figure 1 The survival analysis curves for days open in healthy (♦) and mastitic (■)cows. The number of days to conception for cows with mastitis was greater than that for the healthy cows.

Conclusions The results of the present study demonstrate that mastitic cows had more days open compare with the healthy cow. Therefore in practical terms the incidence of mastitis in a herd reduces reproductive performance.

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Chemical composition, and ruminal and post-ruminal protein disappearance of lucerne silage treated with formic acid

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Introduction Greater feeding value of lucerne silage than lucerne hay has been a common finding of ruminant feeding studies in tropical countries. However, during ensiling, a large proportion of the crude protein (CP) in lucerne is broken down to non protein nitrogen (NPN), (Broderick *et al*., 1995). Additives such as formic acid may reduce proteolysis during lucerne ensiling. The objective of the present experiment was to compare the nutrient content, and ruminal and post-ruminal dry (DM) matter and protein disappearance of lucerne silage treated with urea and formic acid.

Materials and methods Lucerne (30% DM) was harvested, chopped and mixed with different levels of formic acid and urea, then ensiled for 40 days (four replications per each treatment). The treatments were lucerne silage (LS), LS treated with urea (4 g/kg DM) + formic acid (8 ml/kg DM), (LSu+f), and LS treated with formic acid (8 ml/kg DM), (LSf). Formic acid was carried on and used under the safety protocol of Ferdowsi University of Mashad, using special instruments. The acid was diluted with water (acid:water 1:4, vol/vol), then mixed with the forage. Chemical composition (CP, NPN, and DM) was determined (AOAC, 1980). pH was measured directly in silage extract. Silage DM was determined using air-forced oven (60 °C, 48 h). The ruminal and post-ruminal disappearance of dry matter and protein of samples were determined using the mobile nylon bag procedure (Danesh Mesgaran., 2005). Four Holstein steers (400 kg) fitted with rumen fistula and T- shaped cannulae were used. For the ruminal disappearance studies, the experimental samples were milled (2 mm) and weighed (1.2 g DM) into bags (3 × 6 cm) made of polyester cloth with 50 µm pore size (12 bags per each sample). The bags were incubated in the rumen for 12 hours. After removal From the rumen, six bags were washed using cold water and dried in a forced- air oven (58 °C, 48 h) for determining rumen disappearance and other bags were inserted into the small intestine via the cannulae at the rate of one bag every 30 min and removed from the voided faeces, rinsed in cold ruining water. Finally, the bags were dried in a forced air oven (58 °C, 24 h), then weighted to determine the dry matter disappearance. The kjeldhal technique was used for nitrogen analysis. Data were analysed using the GLM procedure of SAS.

Results Chemical composition of LS and LS treated with urea and formic acid is shown in Table 1. formic acid and urea caused to reduce pH, NPN and increased CP ($P<0.05$). Ruminal and post-ruminal disappearance of CP and DM are summarized in Table 2.

Table 1 Chemical composition (g/kg DM) of lucerne silage treated with urea and formic acid

	Treatments				
Item	LS	LSu+f	LSf	SEM	P-value
pH	4.78	3.93	3.75	0.5	0.2
CP	177	196	169	14.0	0.1
NPN	18	15	16	1.5	0.3
DM	292[a]	395[b]	349[ab]	51.6	0.03

[a,b] Means in the same row for each variable with different superscript letters differ ($p < 0.05$)

Table 2 Ruminal and post-ruminal CP and DM disappearance (g/kg) of lucerne silage treated with formic acid and urea

	Treatment				
Item	LS	LSu+f	LSf	SEM	P-value
Ruminal DM disappearance	536[a]	524[ab]	512[b]	12.0	0.01
Ruminal protein disappearance	767	763	767	23.0	0.9
Post- ruminal disappearance of ruminal-undegraded DM	432[a]	472[ab]	482[b]	26.5	0.01
Post- ruminal disappearance of ruminal-undegraded protein	868	875	881	65.0	0.8

[a,b] Means in the same row for each variable with different superscript letters differ ($p < 0.05$)

Conclusions The results of the present study demonstrated that formic acid improved the nutritional value of the LS, however, the difference between the treatments was not statistically significant. formic acid might reduce proteolysis during ensiling by either reduction of pH or by providing additional substrate to enhance the reduction of pH. Variation in ruminal and post-ruminal dry matter disappearance of treatments maybe attributed to differences in DM and neutral detergent fiber (NDF) of these silages.

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A comparison of feeding whole milk versus a milk replacer on the performance of artificially reared calves

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Introduction Rearing pre-weaned calves is one of the most challenging tasks on a dairy farm. Intensively reared calves are typically separated from dams within 24 hours of birth and fed restricted quantities of either whole milk or milk replacer until weaning. In contrast, a calf left with its dam will consume significantly more milk which leads to increased weight gain and improved health (Albright and Arave, 1997). Milk can be provided through *ad libitum* feeding systems with artificial teats allowing calves to consume more milk than with the traditional, bucket fed system. Milk replacer is a commonly used substitute feedstuff for whole milk on many calf rearing units in the UK. Milk replacers are formulated to meet the nutritional requirements of the pre-weaned calf but typically contain significantly less fat, on a DM basis, than whole milk. The aim of this trial was to investigate the effects of feeding whole milk versus a milk replacer to calves on a cold *ad libitum* basis on the milk and concentrate feed intakes and liveweights between the groups.

Materials and methods Eighteen Holstein Friesian calves (eleven heifer and seven bull calves split equally between groups) were randomly assigned into one of two treatment groups at two days old and housed in individual pens for twelve weeks. Group one calves were fed whole milk on a cold *ad libitum* basis and Group two calves received a milk replacer also on a cold *ad libitum* basis. The milk replacer (Heiferlac, Volac International) contained 27% protein and 16.6% fat in the DM. This was reconstituted at a rate of 150g powder made up to a litre with water. The whole milk and milk replacer was offered in fifteen litre buckets fitted with artificial teats for six weeks, at which point the calves were weaned abruptly. Buckets were replenished with fresh milk daily with any remaining liquid measured and replaced. Both treatment groups received Pye Nustart Calf Sprinter pellets (Pye Bibby Agriculture, Lancaster) from day one. Milk and concentrate intakes were recorded daily and calves were weighed at birth, 3, 6, 9 and 12 weeks. Data was then assessed for normality and analysed with ANOVA (Minitab v14). Milk intake data was not normally distributed and therefore log transformed.

Results Milk intakes between the two treatment groups differed, with calves in Group two consuming significantly more milk ($P<0.05$) than calves in Group one (Table 1). On average, milk replacer fed calves consumed 1.7 litres more per day during the six week period than the whole milk fed group. There was no significant difference, $P>0.05$, in the concentrate intakes between the groups over the twelve week period. There was no significant difference, $P>0.05$, in the birth weights of the calves in the two groups at birth however there was a significant difference between the overall weight gains of the two groups ($P<0.05$), with Group two calves gaining significantly more weight. At twelve weeks of age, milk replacer fed calves weighed on average 17kg more than whole milk fed calves (Figure 1).

Table 1 Average milk and concentrate intakes in the two groups of calves

	Group 1	Group 2	s.e.	*P*
Milk intake (l)	6.41	8.16	0.101	0.030
Concentrate intake (kg)	1.66	1.70	1.737	0.660

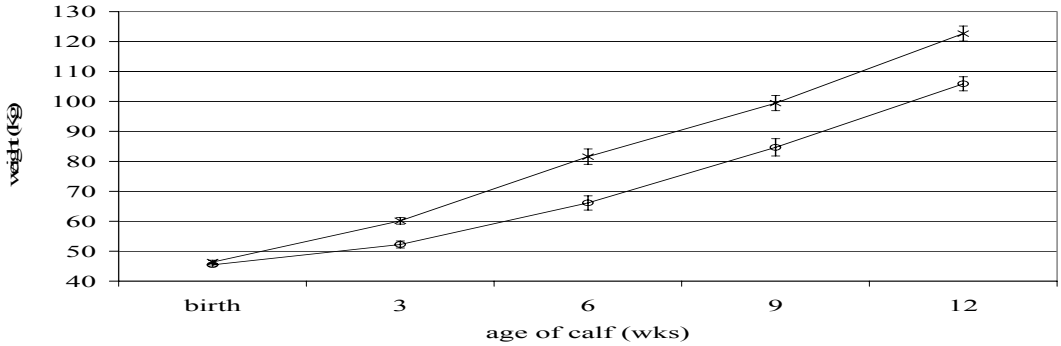

Figure 1 Average weight of calves (±s.e.) at different stages fed whole milk (o) or milk replacer (x)

Conclusions Even the lowest intake recorded for teat fed *ad-libitum* calves was greater than the milk intake of traditional bucket fed calves (Jasper & Weary, 2002). Increased calf growth rates reflected increased intakes. Calves fed on *ad-libitum* milk replacer consumed significantly more and therefore gained more weight than those fed whole milk. The significantly greater liveweight of calves fed milk replacer compared to whole milk at weaning was still evident at 12 weeks of age although there was no significant difference in concentrate consumption between the groups, suggesting that differences in early growth influences overall efficiency to 12 weeks.

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Effects of ethanol processing of soya-bean meal on protein degradation in the rumen

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Introduction Various ethanol solutions have been used to alter ruminal degradation of soya-bean meal (SBM) protein, thus increasing its post-ruminal availability. The best ethanol solutions for this purpose in wide range of ethanol concentration were reported to be 600 and 700 ml/l by Van der Aar *et al.* (1984), and Lynch *et al.* (1987), respectively. However in the literature, there is no degradability data concerning comparison of narrow range of ethanol solutions. Therefore our objective was to compare crude protein (CP) and true protein degradation characteristic of 500, 600 and 700 ml/l ethanol treated SBM.

Materials and methods Three SBM samples (500 g each) were soaked 2:1 (v/w) in 500, 600 and 700 ml/l (w/w) ethanol solutions for 1 h at 25˚C. *In sacco* degradation of CP was measured in the rumen of three Holstein steers according to Ørskov and McDonald (1979) and *in vitro* CP digestibility as described by Sadeghi *et al.* (2005). The degradability parameters for the nylon bags (Two bags for 2, 4, 6, 8, 12, 16, 24 and 48-h incubation time in each steer) and *in vitro* data analysed as a randomised complete block design, using steers as blocks. Analyses were performed according to GLM of SAS (1996). Differences among treatments were separated using polynomial orthogonal contrasts to determine linear, quadratic, and cubic responses. SDS-PAGE technique was used to monitor the degradation of true protein subunits in the rumen as described by Sadeghi *et al.* (2005).

Results and discussion There was a linear (P<0.001) decrease in the water soluble fraction and a linear (P<0.001) increase in the potentially degradable fraction of CP (Table 1). Processing of SBM with 500, 600 and 700 ml/l ethanol solutions decreased effective degradation of CP at rumen outflow rate of 0.05/h by proportionately 0.27, 0.27 and 0.33 compared with untreated SBM, respectively. Although there were differences for effective rumen degradation of CP between treatments, there were not, in general, explained by polynomial orthogonal contrasts. Ethanol processing has neither linear nor quadratic effect (P>0.05), but has cubic effect (P<0.001) on *in vitro* digestibility of ruminally undegraded CP. We could not find differences between 500 ml/l and 600 ml/l ethanol solutions for CP degradability, but ethanol solution of 600 ml/l resulted in a 0.07 proportional increase in CP digestibility compared with 500 ml/l ethanol solution. Ethanol solution of 700 ml/l decreased *in vitro* digestibility of CP compared to other solutions. From the SDS-PAGE pattern, β-conglycinin ά and α subunits of untreated, 500, 600 and 700 ml/l ethanol treated SBM were degraded completely within 4, 6, 8 and 12 h of incubation, respectively. Whereas the β subunit of β-conglycinin as well as the basic and acidic polypeptide components of glycinin of untreated and treated SBM were more resistant to degradation. About 350, 540, 570 and 620 g/kg of untreated, 500, 600 and 700 ml/l ethanol treated SBM proteins were not degraded after 12 h of incubation in the rumen, respectively.

Table 1. Degradability and digestibility of soya-bean meal treated with ethanol solutions

Parameters	Untreated	Ethanol solution (ml/l)			SEM	L	Q	C
		500	600	700				
Degradation characteristic								
Rapidly degradable fraction	177	74	61	29	7.8	***	*	*
Potentially degradable fraction	802	886	909	956	16.5	***	NS	NS
Degradation rate	0.085	0.047	0.047	0.042	0.0051	***	***	***
Effective degradability at rumen outflow rate of:								
0.02/h	827	696	698	678	21.2	***	**	NS
0.05/h	683	504	501	466	22.6	***	**	**
0.08/h	591	402	397	359	20.5	***	**	**
In vitro crude protein digestibility								
	806	902	961	930	26.8	NS	NS	***

Contrast: L, Linear; Q, quadratic; C, cubic contrast; *** P<0.001, ** P<0.01,* P<0.05, NS; not significant.

Conclusion In this study, processing of SBM with 600 ml/l ethanol for 1 h had the greatest potential to increase digestible undegraded protein. SDS-PAGE indicated that the basic subunit of glycinin when untreated SBM, but both glycinin and β-conglycinin when ethanol treated SBM are fed to ruminants, make an appreciable contribution to metabolisable protein.

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Effect of a rumen buffer (Acid Buf) on the performance of cereal fed beef cattle

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Introduction The recent change in beef cattle support payments from headage to area based payment and relatively low cereal prices could see an increased interest in earlier finishing of beef cattle. An increased number of producers are therefore likely to adopt intensive cereal finishing systems for beef cattle. Antibiotic based feed additives e.g. monensin sodium, have been successfully used for over 40 years to manipulate microbial activity and improve beef cattle performance especially in cereal beef systems. The use of monensin sodium was banned from January 2006 and therefore there is a requirement to find alternative 'natural' products that can improve the efficiency of beef production with cereal based rations. Acid Buf is a natural rumen buffer derived from calcified seaweed *(Lithothamnium calcareum)* ground to a very fine particle size. It contains calcium and magnesium as well as a range of trace minerals. The claimed characteristics of Acid Buf are: excellent acid absorbency, long term buffering activity and fine particle size for optimum availability to help overcome problems with acidosis in high starch based rations. The objective of this experiment was to determine the effect of feeding Acid Buf on the performance of intensively finished dairy-bred beef cattle fed *ad libitum* cereals.

Materials and methods Thirty six Belgian Blue cross Holstein bulls and heifers with a mean live weight of 280kg were randomly assigned in a 2 x 2 factorial designed experiment to either a control ration fed *ad libitum* or control including 5kg/t Acid Buf. The control ration contained the following ingredients (g/kg): rolled barley 730, soyabean meal 100, molassed sugar beet pulp 100, molasses 50, mineral 20. The cattle were housed in straw bedded yards with nine bulls and nine heifers per pen and per treatment. Straw was available *ad libitum* from racks. The bulls were selected for slaughter at MLC fat class 3 and heifers at fat class 4L. The data was analysed using ANOVA. Carcass weight at the beginning of the experiment was estimated by assuming a dressing proportion of 0.47 (Patterson *et al.,* 1995).

Results The mean chemical composition of the cereal based concentrates was: DM 889 and 879g/kg; crude protein 154 and 156, ether extract 19 and 21, neutral detergent fibre 228 and 223, starch 408 and 410 and ash 69 and 71 g/kg DM for the control and Acid Buf rations respectively. Data on animal performance and feed intakes are shown in tables 1 and 2. The bulls recorded significantly higher daily liveweight gains (DLWG), slaughter weights, carcass weights, killing out percentage and carcass grades compared to the heifers ($P<0.001$). Feeding Acid Buf resulted in a significant increase in DLWG ($P<0.05$) and a reduction in the number of days to reach slaughter ($P<0.073$). There was a reduction in total feed intake with Acid Buf with a marked improvement in feed conversion ratio (FCR).

Table 1 Animal Performance (kg)

	Acid Buf	Control	s.e.d	Sign
Start wt	278.8	282.1	12.39	NS
Slaughter wt	511.4	520.1	8.78	NS
Days	174.0	190.3	8.77	0.073
DLWG	1.34	1.24	0.048	*
Carcass wt	285.1	291.3	5.26	NS
Carcass DWG	0.89	0.82	0.039	NS
KO %	55.7	55.8	0.51	NS
Conformation[1]	4.33	4.44	0.171	NS
Fat score[2]	4.27	4.22	0.186	NS

[1] 7 point scale: 1=-P (worst), 7 = E (best); [2] 7 point scale 1= 1 (leanest), 7 = 5H (fattest)

Table 2 Feed intakes (kg/head)

	Acid Buf	Control
Total Feed intake	1443	1662
FCR[1]	6.20	6.98
Carcass FCR[2]	8.23	9.30

[1] Feed Conversion Ratio (kg concentrates/kg weight gain); [2] FCR (kg concentrate DM/kg carcass gain)

Based on the prices prevailing at the time of the study, feeding Acid Buf increased the gross margin by £20 per head (£118 versus £98) and reduced the concentrate feed cost per kg liveweight gain from 78p to 69p.

Conclusions The results indicate that feeding Acid Buf to cereal fed beef cattle can improve production efficiency by increasing daily liveweight gain, reducing the number of days to reach slaughter condition and improving financial performance.

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Ruminal degradation rates of soya-bean meal and rapeseed meal protein subunits using densitometrical scanning

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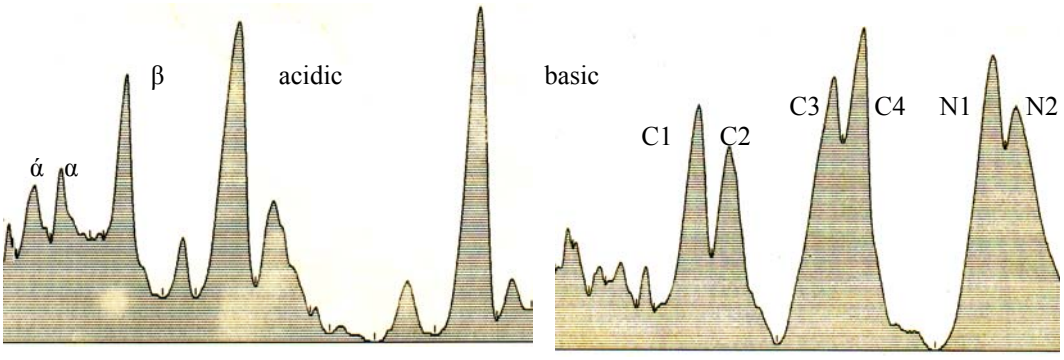
Introduction We have reported previously (Sadeghi *et al.*, 2005; Sadeghi and Shawrang, 2005) on the use of sodium dodecyl sulfate-polyacrylamide gel electrophoresis technique (SDS-PAGE) to study subunits degradation of soya-bean meal (SBM) and rapeseed meal (RSM) proteins in the rumen. As far as we are aware, there is no published information concerning degradation rates of protein subunits of feedstuffs. The objective of this study was to investigate degradation rates of SBM and RSM protein subunits in the rumen.

Materials and methods *In situ* degradation of SBM and RSM true proteins measured at 0, 2, 4, 6, 8, 12, 24 and 48 h in the rumen of three Holstein steers as described by Sadeghi and Shawrang (2005). All the ruminal undegradable fractions from each incubation period were freeze-dried, well ground (0·25-mm particle size) and replicate samples pooled together. SDS-PAGE technique was used to monitor the rates of ruminal degradation of protein subunits as described by Sadeghi *et al.* (2005). Subunits changes with time in the rumen were quantified using densitometric analysis of Coomassie blue stained protein bands at 580 nm. Degradation rates of individual protein subunits were estimated according to Ørskov and McDonald model (1979).

Results Densitometric analysis of SBM protein residues revealed that ά and α and β subunits of β-conglycinin were degraded over 0.50/h., β-conglycinin subunits were degraded completely after 2 h of ruminal incubation, but degradation of glycinin subunits became evident after 8 h. At 24 h, the main bands on the gel represented the basic subunit of glycinin. Acidic and basic subunits of glycinin were degraded at 0.143 and 0.106/h. Overall protein degradability of SBM measured by summation of degradability of subunits components, was 723 g/kg. In RSM, relative rates of degradation for napin subunits (N1 and N2) were over 0.80/h, and for cruciferin subunits (C1, C2, C3 and C4) were 0.180, 0.090, 0.150 and 0.112/h, respectively. Total protein degradability of rapeseed meal was 756 g/kg.

Table 1 Molecular weight and degradation rate of protein subunits of soya-bean meal and rapeseed meal

Soya-bean meal			Rapeseed meal		
Subunits	Molecular weight	Degradation rate (/h)	Subunits	Molecular weight	Degradation rate (/h)
ά	90.5	> 0.50	C1	31.2	0.180
α	71.5	> 0.50	C2	26.8	0.090
β	55.2	> 0.50	C3	21.1	0.150
Acidic	37.6	0.143	C4	20.5	0.112
Basic	19.8	0.106	N1	10.3	>0.80
Molecular weight: kDa			N2	8.2	>0.80



Soya-bean meal Rapeseed meal

Figure 1 Densitometrical scaning analysis of soya bean meal and rapeseed meal individual proteins in the rumen.

Conclusion This study provides further evidence that SDS-PAGE and densitometric analysis of protein subunits can be used to predict degradation rates of dietary protein in the rumen. This methodology will combine with digestibility studies of the ruminally undegradable fractions and qualitative and quantitative identification of the individual amino acids contributing to these fractions to strengthen our ability to predict the contribution of ruminally undegradable protein to the amino acids profile delivered to the small intestine.

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Evaluation of cracked head-cut urea treated whole crop wheat and barley and conventionally combined wheat and barley for finishing beef cattle

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Introduction A preliminary investigation by Marsh and Gibson (2004) indicated that cracked head-cut urea treated whole crop wheat could offer beef producers the opportunity to achieve high levels of animal performance and reduce feed costs per kg gain when compared to diets based on rolled barley for intensively fed beef cattle. The objective of this experiment was therefore to evaluate crop yield and beef cattle performance from winter wheat and winter barley crops harvested both conventionally and at the end of physiological grain fill (approx 70% dry matter), the latter being made into cracked head-cut urea treated whole crop.

Materials and methods Seventy-two Belgian Blue x Holstein bulls and heifers with a mean live weight of 265kg were allocated by weight to the following dietary treatments in a randomised block design. B: *Ad lib* 130g crude protein/kg concentrates (840g/kg rolled barley, 80g/kg soya-bean meal, 50g/kg molasses, 20g/kg minerals, 10g/kg sodium bicarbonate) plus *ad lib* barley straw. HWB: *Ad lib* cracked head-cut urea treated whole crop barley (DM 776 g/kg: 138 g CP/kg DM, 470 g NDF/kg DM, 390 g starch/kg DM) plus 150g minerals per head per day. W: *Ad lib* 130g CP/kg concentrates (840g/kg rolled wheat, 80g/kg soya-bean meal, 50g/kg molasses, 20g/kg minerals, 10g/kg sodium bicarbonate) plus *ad lib* barley straw. HWW: *Ad lib* cracked head-cut urea treated whole crop wheat (DM 710 g/kg: 146 g CP/kg DM, 355 g NDF/kg DM, 430 g starch/kg DM) plus 150g minerals per head per day. The whole crop barley was made from the variety Pearl with a standing height of 87cm, harvesting the top 35cm. It was cut at growth stage 92 with a harvester fitted with a grain processor and ensiled with 35kg/t Home 'N' Dry (a mixture of urea and urease, Volac International Ltd.). The whole crop wheat was made from the variety Solstice with a standing height of 76cm, harvesting the top 25cm. It was cut at growth stage 90-91 with a harvester fitted with a grain processor and ensiled with 35kg/t Home 'N' Dry. Cereal grain and straw yields were 7.31t and 5.33t, and 9.75t and 4.95t/ha for the barley and wheat respectively. Whole crop and straw yields were 10.08t DM and 2.63t, and 11.46t DM and 3.46t/ha for the barley and wheat respectively. The total DM yields per hectare were 10.92, 12.96, 12.60 and 14.71t for B, HWB, W and HWW treatments respectively. The data was analysed using ANOVA.

Results There were no significant differences in animal performance between the barley and wheat grain fed beef cattle. Compared to the barley and wheat grain treatments the cattle fed the cracked head-cut urea treated whole crop wheat recorded lower daily live and carcass weight gains ($p<0.05$), took longer to reach slaughter ($P<0.001$) and recorded lower fat scores ($P<0.05$). The bulls recorded significantly heavier slaughter and carcass weights ($P<0.05$). The cattle fed cracked head-cut urea treated whole crop barley recorded lower daily and carcass weight gains ($p<0.001$), took longer to reach slaughter ($P<0.001$) and recorded lower fat scores and killing out percentage ($P<0.05$). Feed intakes and costings are shown in table 2 which were calculated based on the feed prices prevailing at the time of the study.

Table 1 Animal performance

	B	HWB	W	HWW	s.e.d	Sig
Slaughter weight (kg)	498.2	516.4	501.8	524.7	9.12	N.S
Days to slaughter	181.2^{a}	242.4^{b}	189.8^{a}	229.9^{b}	11.10	***
DLWG (kg)	1.29^{a}	1.01^{c}	1.26^{a}	1.12^{b}	0.038	***
Carcass weight (kg)	274.5	275.6	276	285	5.51	N.S
Killing out %	55.0^{a}	53.3^{b}	54.9^{a}	54.2^{a}	0.40	*
Carcass DG (kg)	0.84^{a}	0.62^{c}	0.81^{a}	0.70^{b}	0.03	***
Conformation*	4.28	3.94	4.17	4.22	0.176	N.S
Fat classification*	3.44^{a}	2.83^{b}	3.38^{a}	2.83^{b}	0.166	*

Within row, means with the same superscripts are not significantly different (*$p>0.05$*)
* EUROP carcass classification: Conformation: P+=1 and E=7. Fat class: 1=1 and 5H=7.

Table 2 Feed intakes (kg/head) and costs

	B	HWB	W	HWW
Total Intake (kg DM)	1488	2139	1486	1962
FCR (kg DM feed/kg carcass gain)	9.76	14.25	9.64	12.18
Feed costs (p/kg carcass gain)	94.0	95.5	89.6	80.7
Stocking rate (cattle/ha)	5.57	4.79	7.53	5.94
Gross Margin/ha (£)	168	122	287	343

Conclusions Rolled wheat can replace rolled barley in cereal beef diets. It must however be lightly rolled and fed with a rumen buffer and *ad lib* straw to minimise problems with acidosis. Feeding cracked head-cut urea treated whole crop wheat offers beef producers the opportunity to increase slaughter weights or produce leaner carcasses, reduce feed costs per kg gain and increase margins per hectare. Cracked head-cut urea treated whole crop barley is more suitable for semi-intensive beef production.

Acknowledgement Financial support from the English Beef & Lamb Executive (EBLEX) is gratefully acknowledged.

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Intake, rumen fermentation and plasma metabolites in beef cattle offered grass silage, maize silage, fermented whole crop wheat and alkalage

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Introduction Grass silage can be less attractive as a ruminant feedstuff when compared to alternative forages due to relatively modest yields obtained in a single harvest, variability in digestibility and ensilability, coupled with ever increasing costs of production and effluent management. The composition of feeds consumed differs markedly from the nutrients available for metabolism due to the modifications caused by microbial fermentation in the rumen. Volatile fatty acids produced in the rumen as a result of the fermentation of feed are the major source of energy supplied to the ruminant. The objectives of this experiment were to determine the effect of alternative forages on intake, rumen fermentation and plasma metabolites in beef cattle.

Materials and methods Four ruminally fistulated Holstein-Friesian steers with a mean liveweight of 440 kg were used in a 4 (21 day period) x 4 (diet) Latin Square experiment. The four experimental forages offered were grass silage (GS), maize silage (MS), fermented whole crop wheat silage (FWCW) and alkalage (ALK) (urea-treated whole crop wheat silage). In addition each diet was supplemented with 3 kg of the same cereal-based concentrate consisting of 650g rolled barley, 280g soya-bean meal, 50g molasses and 20g minerals and vitamins per kg. Forage and concentrates were offered *ad libitum* in two equal daily feeds at 0830 h and 2030 h until day 13 of each period and then at 0.95 *ad libitum* until the end of each period. Rumen fermentation characteristics were determined on rumen fluid samples collected at 0830, 1030, 1230, 1430, 1630 and 1830 h on day 14 of each period. On day 21 of each period, blood samples were obtained by jugular venipuncture from each animal at 0830 and 3 and 6 hours late. Data were analysed using analysis of variance in accordance with a Latin Square design experiment.

Results The composition of the GS, MS, FWCW and ALK respectively was; dry matter (DM) 190, 317, 410 and 725 g/kg, *in vitro* DM digestibility 712, 765, 689 and 712 g/kg DM, pH 4.2, 4.0, 4.1 and 4.3, lactic acid 92, 65, 27 and 3 g/kg DM and acetic acid 32, 15, 13 and 8 g/kg DM. The effect of dietary treatment on intake, rumen pH, ammonia N, lactic acid, volatile fatty acid concentrations and plasma metabolites are shown in Table 1. Alternative forages resulted in higher forage and total DM intakes compared to grass silage ($P<0.01$). There was a significant effect of forage on rumen ammonia N, propionic acid, butyric acid, valeric acid and total VFA concentrations and, acetate to propionate ratio. Maize silage produced significantly lower concentrations of ammonia N and higher concentrations of propionic and butyric acids and, total VFA than the other forages. Rumen pH, lactic acid and acetic acid concentrations were not significantly affected by treatment. There was a significant effect of forage on plasma β hydroxybutyrate and urea concentrations but not on glucose concentrations.

Table 1 The effect of dietary treatment on feed intake, rumen fermentation and plasma metabolites

	GS	MS	FWCW	ALK	s.e.m.	Sig.
DM Intake (kg/day)						
Forage	5.55^{a}	8.84^{b}	8.51^{b}	8.99^{b}	0.631	**
Total	7.98^{a}	11.27^{b}	10.94^{b}	11.42^{b}	0.631	**
Rumen parameters						
pH	6.57	6.31	6.43	6.45	0.055	NS
Lactic acid (mg/l)	85.5	120.5	36.0	37.5	24.06	NS
Ammonia N (mg/l)	103.3^{a}	28.1^{b}	87.5^{a}	160.4^{c}	8.24	***
Acetic acid (mmol/l)	41.7	47.4	43.2	42.9	2.04	NS
Propionic acid (mmol/l)	11.5^{a}	14.7^{b}	11.7^{a}	9.7^{a}	0.72	*
Butyric acid (mmol/l)	7.4^{a}	11.2^{c}	9.3^{b}	8.8^{b}	0.26	***
Valeric acid (mmol/l)	1.8^{a}	2.9^{b}	2.6^{bc}	2.2^{ac}	0.14	**
Total VFA (mmol/l)	62.4^{a}	76.2^{b}	66.8^{a}	63.6^{a}	2.72	**
Acetate:Propionate ratio	3.7^{a}	3.3^{a}	3.8^{a}	4.4^{b}	0.16	*
Plasma metabolites (mmol/l)						
Beta hydroxybutyrate	0.33^{a}	0.51^{c}	0.43^{b}	0.46^{b}	0.01	***
Glucose	4.13	4.25	4.26	4.27	0.071	NS
Urea	3.97^{a}	2.43^{b}	3.63^{a}	5.82^{c}	0.155	***

Within row, means with the same superscripts are not significantly different ($p>0.05$)

Conclusions Dry matter intake was lower with grass silage than the alternative forages. The feeding of alternative forages significantly altered rumen fermentation parameters and plasma β hydroxybutyrate and urea concentration.

Comparison of commercial DNA extraction kits and sample preparation for extraction of bacterial DNA from different fractions of whole rumen fluid

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Introduction Previous work has shown that the biohydrogenation of fatty acids is different in solid and liquid fractions prepared from rumen fluid (Dorel *et al*., 2005). This study aimed to relate this observation to differences in the microbial population in the different fractions. Molecular techniques allow the rapid investigation of microbial numbers and diversity in environmental samples. However, description of the microbial diversity can be affected by the efficiency of DNA extraction from samples as it is known that it is harder to quantitatively extract DNA from some microbes than others (Clement and Kitts, 2000). The aim of this study was to investigate different protocols for preparing samples and extracting DNA from different fractions of rumen fluid, in terms of both the numbers and diversity of bacteria that could be recovered.

Materials and methods Rumen fluid was withdrawn 2 h after feeding from four rumen cannulated Hereford X Friesian steers that had been fed a diet of grass and legume silages for 14 weeks prior to sampling. Rumen fluid was separated into strained rumen fluid (SRF), large particles (LP) and small particles (SP) as described in Dorel *et al*. (2005). Sub-samples were freeze-dried and ground under liquid nitrogen (FD&G) or processed directly from storage at -80^{o}C. The samples were extracted with the Qbiogene FastDNA® SPIN kit for soil (Qbiogene, UK, Kit A) or the Qiagen QlAamp® DNA stool mini kit (Qiagen Ltd, UK, Kit B). Extracted DNA was quantified by fluorescence using BisBenziminde (Sigma, UK). PCR was performed with 1ng DNA using primers described by Nubel *et al*. (1996) to amplify the V6-V8 region of bacterial 16S rRNA. Denaturing Gradient Gel Electrophoresis (DGGE) was performed using an equal loading of PCR products on 6% polyacrylamide gel with a 35-60% gradient, run for 16 h at 85V at 60^{o}C. DGGE gels were visualised by silver staining. Gels were analysed using Bio-Rad Fingerprinting II software to obtain Unweighted Pair Group Method using Arithmetic averages dendograms. Species richness was calculated from the number of bands in each lane while diversity was estimated using the Shannon's diversity index. Bacterial DNA concentrations where determined by quantitative PCR (QPCR) as described by Maeda *et al*. (2003). Results were analysed by ANOVA. Total and bacterial DNA data were transformed prior to analysis and are reported as back-transformed means with transformed SEDs.

Results Significantly ($P<0.001$) more total DNA and bacterial DNA was extracted with Kit A than Kit B with more DNA extracted from the SRF fraction ($P<0.05$) compared to SP and LP (2.1, 1.7, 1.0 *v* 0.2, 0.1 & 0.1 µg total DNA/mg DM extracted, SED 0.08 and 10.1, 7.6, 7.5 *v* 1.0, 0.6 & 0.6 µg bacterial DNA/mg DM extracted for A *v* B from fractions SRF, SP and LP respectively SED 0.09). The method of preparation frozen *v* FD&G did not have an effect on the amount of total DNA extracted, but did have an effect on the amount of bacterial DNA extracted, with significantly more ($P=0.01$) bacterial DNA being extracted with FD&G samples. However, there was a significant ($P<0.001$) kit by preparation interaction in the bacterial DNA extracted in that Kit B was significantly more effective with FD&G samples (8.2, 8.4 v 1.1, 0.4 µg bacterial DNA/mg DM extracted for Kit A v Kit B for FD&G and frozen samples respectively SED 0.073). Species richness, estimated from the number of bands recovered by DGGE was significantly higher ($P<0.01$) with Kit B compared to Kit A with FD&G (32.3, 27.8, 31.5 *v* 35.5, 35.0 & 33.0 A *v* B from fractions SRF, SP and LP respectively SED 1.0) and frozen samples (32.5, 34.2, 39.2, *v* 50.2 47.8 & 55.0 from fractions SRF, SP and LP respectively SED 3.4). Diversity estimated from the Shannon's index showed a similar trend with a significantly higher diversity ($P<0.01$ except SRF with FD&G samples) with Kit B than Kit A (3.87 *v* 3.84 SED 0.030, 3.74 *v* 3.90 SED 0.036, 3.81 *v* 3.90 SED 0.031, A *v* B from fractions SRF, SP and LP respectively with FD&G samples, and 3.97 *v* 4.27 SED 0.028, 4.01 *v* 4.27 SED 0.029, 4.04 *v* 4.29 SED 0.025 A *v* B respectively with frozen samples). No grouping based on extraction method or sample preparation was discernible in the dendograms with lanes tending to cluster based on animal.

Conclusions Although sample preparation had little effect on the diversity of bacteria that could be recovered from fractions of rumen fluid, FD&G extracted significantly more bacterial DNA. There are also obvious advantages in terms of ease of representative sampling with FD&G, in particular with LP, which would make it the method of choice. Although Kit A extracted more DNA from samples, Kit B gave a higher proportion of bacteria to total DNA with FD&G samples, and generated the greatest species richness and diversity.

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Effects of using dry sugar beet molasses on dairy cattle performance

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Introduction Sugarcane and sugar beet molasses are dark brown juices which are remained after crystallization or concentration of sugar syrup. Problematic nature of molasses prompted sugar producers to solve difficulty of preservations, transportation, mixing and the problem of sticky concentrate mixtures or its hard handling in cold weather. Thus, in recent years, Iran sugar industry made a shift to develop dehydration technology of molasses, but it was not clear weather the applied technology could be efficient employed and if such a product could replaced by sugar beet pulp which is in rare in cold seasons. This trial performed to study if Dry Sugar beet Molasses (DSBM) could be efficiently replaced with sugar beet pulp with no side effects.

Materials and methods In a 56-d trial, 12 second parous Holstein (mid-lactation) cows which their average milk yield were 32.7 ±3.41 kgd^{-1} and weighed 581±18 kg were fed two times a day as *ad libitum* on total mixed rations with the same net energies for lactation and crude protein content (NRC, 2001) and residues were weighed after oven drying for 48h (60 ^{o}C). Table1 illustrates composition of rations as they contained 0, 3 and 6% DSBM replacing with sugar beet pulp. Cows underwent 2 weeks as adaptation period and in the following 6 weeks, milk yield records were taken 3 times a day, every week. On each recording time 3 cows were random selected to get samples for milk analysis and samples were mixed with potassium dichromate to inhibit bacteria growth. After finalization of the experiment, frozen samples were analyzed for fat, protein, lactose, solids non fat (SNF) and total solids percents. Results were analyzed in a completely randomized experimental design by using SAS 6.12 statistical software.

Table1 Composition of experimental rations

	Daily Intake			Percent in Diet		
Feed ingredient	Control	Group1	Group2	Control	Group1	Group2
Fresh red clover	2.8	2.8	2.8	12.03	12.03	12.07
Corn silage	3.63	3.63	3.63	15.59	15.59	15.65
Lucerne hay	3.4	3.4	3.4	14.6	14.63	14.66
Sugar beet pulp	1.82	1.12	0.42	7.82	4.8	1.81
Ground Barley	4.4	4.4	4.4	18.9	18.9	18.99
Cottonseed meal	2.9	2.9	2.9	12.46	12.46	12.5
Wheat bran	4.04	4.04	4.04	17.36	17.36	17.42
DSBM	0	0.7	1.4	0	3.0	6.04
Mineral mix.	0.18	0.18	0.1	0.77	0.77	0.43
Salt	0.11	0.11	0.11	0.47	0.47	0.43
Total	23.28	23.28	23.20	100	100	100

Results Results showed no significant difference for milk yield and FCM for 3.5% fat and also, fat, CP, lactose, SNF and total solids percents among control and treatments (Table 2). FCM for 3.5% fat was used according to Scalan *et al.* (1989) because milk yield regardless of its fat content is not enough of value. The used formula for this calculation is as following:

%3.5 FCM = milk yield x (0.44 + 0.16 x %Fat)

Table 2. Effects of replacing DSBM with sugar beet pulp on milk production and composition

Parameter	DMI	Milk yield	%3.5FCM	%Fat	CP	Lactose	SNF	Total Solids
Control	23.00	29.99	28.42	3.48	3.02	4.91	8.73	11.00
Group1	23.18	29.71	28.17	3.47	2.89	5.02	8.56	11.02
Group2	23.15	30.56	28.04	3.32	2.83	5.00	8.45	10.55
s.e.m.	------	0.74	0.86	0.1	0.6	0.7	0.12	0.21

Conclusions Observed an insignificant improvement for voluntary feed intake; also, results of milk yield and milk fat percent was consistent with ones reported by Khan and Chaudrhy (2000) which they observed an increase in %3.5 FCM production and a decline in milk fat percent with Holstein x Sahiwal hybrids, how ever these changes were not statistically significant. Therefore, DSBM could be replaced efficiently wish sugar beet pulp up to 6% of ration.

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Liveweight and condition score in Aberdeen Angus cross and Limousin cross suckler cows either housed or outwintered on alternative forage crops from November to March

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Introduction Lowering the costs of suckler cow production systems is one of the key challenges facing the beef industry as a result of recent changes in the Common Agricultural Policy which has resulted in farm based subsidies being de-coupled from food production. One of the largest costs associated with production in spring calving suckler cow systems is the costs of over-wintering the non-lactating cow. The capital tied up in buildings is a major component of these costs in countries such as the UK where the relatively harsh winter environments have traditionally led to many cows being housed and fed conserved forages over the winter months to ensure excessive losses in cow liveweight (LW) and condition score (CS) do not occur. Applied studies are required to develop systems which allow non-lactating suckler cows to be out-wintered under UK conditions without excessive LW and CS loss as a means of reducing production costs. The objective of the current study was to determine LW and CS changes in non-lactating suckler cows when managed under a range of outdoor conditions from November to March.

Materials and methods After their calves had been weaned in late October 2004, a total of 56 spring-calving suckler cows were allocated to one of 4 winter feeding groups on the basis of cow breed type, LW and CS. Cow breed types used were either Aberdeen Angus cross (AAx) or Limousin cross (LIMx) and the 4 winter feeding groups were as follows:- deferred grazing (GRAZ), kale grazing, (KALE), yellow turnip grazing (TURNIP) and a housed control group where diets were based on *ad libitum* spring barley straw and 15 kg/h/day of grass silage (HOUSED). Cows on the deferred grazing group (n=9) were outwintered and given access to long grass which had not been cut or grazed since mid summer 2004 using a strip grazing system controlled by an electric fence. These cows were also given access to grass silage bales that had been placed throughout the field, as the electric fence moved over the field area such that cows could eat either deferred grazing or grass silage each day. Both the KALE (n=14) and TURNIP (n=14) groups were also outwintered and given access to a grass run-back area and their respective forage crops using a strip grazing system. They also had access to spring barley straw bales which had been placed along the edges of the forage crop areas prior to the start of the feeding period. Cows in the HOUSED group (n=19) were kept indoors in a straw bedded yard and fed the grass silage and spring barley straw diets on a daily basis. Both outwintered and housed cows had free access to a mineral supplement. Individual cow LW and CS (Lowman *et al*, 1976) measurements were determined at the start (15/11/04) and end (22/02/05) of the winter feeding period respectively. LW and CS changes over the winter feeding period were determined by difference and all data were statistically analysed using the residual maximum likelihood procedure (REML) in Genstat 5 according to a 4 x 2 randomised continous block design with 4 treatment groups (GRAZ, KALE, TURNIP and HOUSED) and 2 cow breed types (AAx and LIMx).

Results No significant differences between cow breed types were found so only feeding group averages are given for cow LW, LW change, daily liveweight gain (DLWG), CS and CS change in Table 1. The HOUSED group of cows gained weight (34 kg) whilst all out-wintered cow groups lost weight over the winter feeding period. At an average weight loss of 49 kg the cow group on deferred grazing lost significantly ($P<0.05$) more weight than all other groups. All groups lost CS over the trial period (0.10 – 0.31units) except the KALE group who gained 0.05 CS units.

Table 1 Cow LW (kg), DLWG (kg/d), CS (1 – 5 scale) and CS gain in outwintered spring-calving suckler cows.

	GRAZ	KALE	TURNIP	HOUSED	s.e.d.	Sig.
LW @ start (kg)	660	683	702	698	21.6	
LW @ end (kg)	611[a]	681[b]	684[b]	732[c]	19.8	*
LW change (kg)	-49[a]	-2[b]	-18[c]	+34[d]	7.8	*
DLWG (kg/d)	-0.50[a]	-0.02[b]	-0.08[c]	0.35[d]	0.079	*
CS @ start	2.52	2.64	2.63	2.57	0.120	
CS @ end	2.12[a]	2.70[b]	2.38[a]	2.47[ab]	0.146	*
CS change (units)	-0.31[a]	+0.05[b]	-0.25[a]	-0.10[ab]	0.127	*

Conclusions Under appropriate field conditions, non-lactating, spring calving suckler cows can be out-wintered on both KALE and TURNIP systems with an acceptable degree of LW and CS loss. Further work is required to establish if cows can be out-wintered on deferred grazing for a shorter part of the winter period and to determine the performance levels of other classes of cattle when out-wintered on a range of feeding systems.

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Effect of increasing milk replacer crude protein content and feeding level on nitrogen output by bull calves pre-weaning

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Introduction Previously reported results indicated that feeding heifer calves more milk replacer increased growth rates in the pre-weaning period, but had little or no effect on plasma protein concentration. However, increasing the crude protein content of milk replacer from 230 to 300 g/kg, had no effect on growth rates but plasma total protein and plasma urea concentrations increased (Wicks *et al*., 2005a, b). Kohn *et al*. (2005) showed that plasma urea was an indicator of nitrogen excretion. An experiment was therefore initiated to investigate the influence of level of milk replacer feeding and the crude protein content of the milk replacer on the nitrogen excretion of calves.

Materials and methods Forty-four autumn-born Holstein-Friesian bull calves were allocated to 8 treatments at 5 days of age. Birth weight was recorded and calves received colostrum for the first 5 days of life. From day 5 until weaning at 56 days, milk replacer (MR) was fed via teated bucket in two 3 l feeds/d. Calves were housed in pairs by treatment and given access to water and calf starter (180 g CP/kg) *ad libitum*. A 4x2 factorial design was used which involved four levels of feeding: 500, 750, 1000 and 1250 g/d; and two MR with crude protein (CP) contents of 230 and 270 g CP/kg fresh powder. Live weight was recorded at weekly intervals throughout the study, and fortnightly blood samples were taken. Calves were on balance between weeks 3-4 (Period 1) and 7-8 (Period 2) of life. Total output of faeces and urine was recorded and samples were taken for nitrogen and dry matter analysis (faeces only). Repeated measures and regression statistical analysis were applied, where appropriate, fitting fixed effects for level of milk replacer feeding, milk replacer crude protein content and their interactions. There were no significant interactions between period and level of milk replacer feeding, period and milk replacer crude protein content, or level of milk replacer feeding and milk replacer crude protein, therefore only the main effects of level of feeding and crude protein content are presented.

Results Liveweight gain from birth to day 28 was significantly (P<0.05) higher for calves offered 1000 or 1250 g milk powder/d compared with calves offered 500 or 750 g/d. However, from day 28 to weaning (day 56) calves offered 500 g milk powder/d grew significantly faster (0.75 kg/d, P<0.05) than calves offered 1250 g/d (0.63 kg/d; s.e.d. 0.045) (Table 1). Calves offered the standard protein (230 g CP/kg) milk powder had significantly higher liveweight gains from day 28 until weaning (P<0.01) compared with calves offered the 270 g CP/kg milk powder. There was no significant difference in blood protein levels between calves offered the 4 levels of milk replacer. Calves offered the 270 g CP/kg milk replacer had higher plasma urea concentrations (2.98, P<0.001) compared with calves offered the 230 g CP/kg milk powder (2.57, s.e.d. 0.114). Nitrogen output in urine was significantly higher for calves offered the 270 g CP/kg milk powder compared with calves offered the 230 g CP/kg milk powder (P<0.01).

Table 1 Milk replacer intakes, live weights, linear measurements, plasma parameters and nitrogen output for calves offered 4 levels of milk replacer feeding/d and either a powder containing 230 or 270 g CP/kg fresh.

	Milk replacer offered (g/d)						CP content of milk replacer (g/kg)			
	500	750	1000	1250	s.e.d.	Sig.	230	270	s.e.d.	Sig.
Birth weight (kg)	47.4	46.8	47.5	46.9	1.84		47.2	49.1	1.30	NS
Liveweight gain (kg/d)										
Day 0-28	0.30[a]	0.36[a]	0.46[b]	0.51[b]	0.045	*	0.40	0.42	0.032	NS
Day 28-56	0.75[b]	0.67[ab]	0.67[ab]	0.63[a]	0.045	*	0.73	0.63	0.032	*
Day 0-56	0.47[a]	0.51[ab]	0.54[b]	0.54[b]	0.031	*	0.54	0.49	0.022	NS
Plasma parameters										
Total protein (g/l)	54.28	52.64	55.27	53.03	1.971	NS	52.54	55.07	1.394	NS
Urea (mmol/l)	2.93	2.78	2.70	2.69	0.161	NS	2.57	2.98	0.114	***
Nitrogen output										
N faeces (g/d)	8.8	10.1	9.9	9.5	1.36	NS	8.9	10.2	0.97	NS
N urine (g/d)	22.9	25.1	26.2	26.6	2.40	NS	23.2	27.1	1.70	*

* = P<0.5; ** = P<0.01; *** = P<0.001

Conclusion Calves offered higher levels of milk replacer grew faster in the first 28 days of life. In view of the lack of an effect of the higher protein milk replacer on animal performance and the increased excretion of nitrogen which has potential environmental costs, the results of this study do not support increasing protein content of milk replacer above current recommended levels of 230 g/kg DM.

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Effect of Megalac supplementation on the total lipid content, moisture content and bound water content of sheep claw horn

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Introduction Lameness in adult ewes can result in reduced prolificacy, lower milk yields and a reduced wool yield (Anon, 1992) and a report by DEFRA (2003) showed 90% of sheep farms had lameness problems. The profiles of fatty acids found in claw lipids from lame cattle have been shown to be different to those in sound animals (Offer *et al*. 2000). Although the underlying mechanism behind this remains unclear, this offers the possibility of influencing the degree of lameness by manipulating claw lipid composition (Offer *et al*. 2000). Inoue *et al*. (1986) have shown that human epidermis with higher levels of lipid also has higher moisture content and bound water content. As water is known to modulate the mechanical properties of claw horn (Baillie *et al*. 2000), it may be postulated that a change in lipid content through supplementation may allow manipulation of the biomechanical properties of the claw. This may be important in lameness where brittle claw horn is manifest. This study investigates whether a dietary lipid supplement, protected against rumen fermentation, may be a useful tool in controlling the properties of the sheep claw.

Materials andmMethods Ten female Mule x Charollais lambs were fed a basal diet containing 550g/kg barley, 350g/kg oats, 50g/kg molassed feed meal, 25g/kg extracted soybean meal and 25g/kg mineral and vitamin mix (Frank Wright Ltd., U.K.), to achieve a growth rate of 180g/d and assigned to control group (n=6) and treated group supplemented with 86.6g/kg diet DM Megalac (Volac Ltd., U.K.) (n = 4). All diets were fed for 10wks. At slaughter the lateral digit of the right forefeet of each animal was taken. The wall and heel horn were sampled 2.5cm distal to the coronary band to ensure the material taken was that produced during the 10wk feeding period. Total lipid was extracted using 2:1 chloroform and methanol mixture. The moisture content for fully hydrated claw horn was measured by drying over P_2O_5 after immersion in distilled water for seven days. Bound water content was measured by differential scanning calorimetry. The data was analysed by two-sample t-test.

Results The mean values for total lipid content, moisture content and bound water content are presented in Table 1. There was no significant difference in total lipid content, moisture content or bound water content of the wall horn between the control and treated animals. The heel horn of the treated group had significantly higher total lipid content ($P<0.05$), moisture content ($P<0.01$) and bound water content ($P<0.05$) when compared to the controls.

Table 1 Total lipid content, moisture content and bound water content of sheep claw horn for control and treated animals

Parameter	Control	Treated	s.e.d	*p*
Wall				
Total Lipid (g/100g DM)	4.1	3.9	0.43	0.60
Moisture content (g H_2O/100g DM)	76.9	74.5	3.51	0.51
Bound Water (g H_2O/100g DM)	48.4	55.8	7.12	0.33
Heel				
Total Lipid (g/100g DM)	4.9	6.2	0.51	0.05
Moisture content (g H_2O /100g DM)	108.9	133.2	7.18	0.01
Bound Water (g H_2O/100g DM)	75.7	108.1	11.07	0.05

Conclusions Dietary supplementation with Megalac had no effect on the composition of sheep claw wall horn; however the heel horn was much more responsive to dietary supplementation. The increase in lipid content was associated with an increase in gross moisture within the horn and this is reflected by increasing the bound water within the claw. The removal of bound water from the claw is less affected by the environment; therefore the lipid supplementation may result in permanent changes to the horn. It could be suggested that this change in lipid content and moisture content could increase the flexibility of claw horn, which may allow for increased absorption of loads imposed through locomotion. However, further work is needed to assess directly the impact of these changes in lipid and moisture on the mechanical properties of sheep claw horn.

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Relationship between digital dermatitis and sole lesions, heel erosion, and locomotion score in dairy herd replacements

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Introduction Digital dermatitis is a major challenge to the UK dairy industry, affecting more than 70% of farms (Dawson, 1998). It is a highly contagious condition that affects the skin next to the claws and may even extend to the claws themselves. The extent to which digital dermatitis is affected by different management systems, and its relationship with other hoof health issues remains unclear. The aim of the current study was to assess the effects of different rearing regimes on the occurrence of digital dermatitis in dairy herd replacements. In addition, relationships between digital dermatitis and the severity of other hoof health parameters, including sole lesions, level of heel erosion and locomotion score, were also examined.

Materials and methods One hundred high genetic merit Holstein-Friesian heifers in an Autumn-calving herd were used in this study. Heifers were reared from 7 weeks of age until calving at 2 years of age on one of four rearing regimes in a 2 x 2 factorial design experiment. Heifers in treatments 1 and 3 were offered grass silage-based diets during winter periods and grass-based diets (grazing) during summer periods (Feeding system 1 ('FS1')). Heifers in treatments 2 and 4 were offered straw/concentrate diets during the winter periods (Feeding system 2 ('FS2')). These heifers remained housed and were offered straw/concentrate diets during their first summer period, and were grazed outdoors during their second summer period. Target calving weight in treatments 1 and 2 was 540 kg ('Low'), and in treatments 3 and 4 was 620 kg ('High'). The higher target calving weight was achieved by additional concentrate supplementation during rearing. Sole lesion and heel erosion scores, and presence or absence of digital dermatitis lesions, were recorded from both hind hooves of each heifer at different time points during rearing and first lactation i.e. at approximately 7 weeks of age, at approximately 6, 12, 18 and 24 months of age (Pre-calving), and at approximately 1, 2, 3, 4, 6 and 11 months post calving. The measurements at 6, 12, 18 and 24 months of age corresponded with the end of the 1st winter period, the start of the 2nd winter period, the end of the 2nd winter period and the start of the 3rd winter period, respectively. During each examination, the sole of each claw was functionally trimmed, and sole lesion scores and heel erosion scores recorded using the method of Livesey *et al*. (1998). Each heifer was also locomotion scored at each sampling interval using the method of Manson and Leaver (1988). For reasons not related to the experiment, 19 heifers were excluded from the data set for statistical analyses. Binomial regression was used to assess treatment affects on digital dermatitis, and relationships between dermatitis and the maximum sole lesion, heel erosion and locomotion score measured were analysed by REML analysis using Genstat 5.

Results There were no significant treatment effects on the percentage of heifers with digital dermatitis (FS1: 'Low' 81%, 'High' 84%; FS2: 'Low' 68%, 'High' 95%; $P>0.05$). On average, 82% of heifers showed signs of digital dermatitis at some stage during the study. Dermatitis was most prevalent at 18 months of age when 42% of heifers showed evidence of infection. The relationship between digital dermatitis and severity of other hoof health parameters is shown in Table 1. Heifers that showed clinical signs of digital dermatitis during the study showed greater maximum heel erosion scores and greater maximum locomotion scores ($P<0.001$).

Table 1 Relationship between presence or absence of digital dermatitis over experimental period and maximum score measured for other hoof health parameters

	Dermatitis	No dermatitis	s.e.d.	Significance
Sole lesion score	1.19	1.12	0.112	
Heel erosion score	2.90	1.77	0.375	***
Locomotion score	2.29	1.80	0.123	***

The majority of heifers (64%) showed their maximum sole lesion and locomotion scores during the lactation period. A smaller proportion (54%) of heifers showed maximum heel erosion scores during the lactation period.

Conclusions These results suggest that the occurrence of digital dermatitis is related to the severity of other hoof pathologies and locomotion problems. The fact that treatment group did not affect digital dermatitis reflects the infectious nature of the disease.

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Influence of management factors on the performance and behaviour of weaned pigs

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Introduction Commercial pig producers may wish to house more pigs than normal in a pen in the immediate post weaning period to reduce heating requirements. However, this may necessitate groups being split or pens being expanded at a later stage. This study investigated the effects of a number of different management regimes on the performance and behaviour of weaned pigs.

Material and methods A total of 1440 pigs were assigned in a randomised block design to one of six treatments over eight replicates. Treatments are outlined in Table 1, and were applied from weaning at 4 weeks of age until 10 weeks of age. At 7 weeks of age pigs were either split into two groups (Treatments 3, 4 and 5), pens were expanded to give additional space (Treatment 1), or groups remained as they were (Treatments 2 and 6).

Table 1 Description of treatments used in the study

Treatment	4 to 7 weeks of age		7 to 10 weeks of age	
	Group size	Space allowance (m^2)/pig	Group size	Space allowance (m^2)/pig
1	20	0.2	20	0.4
2	20	0.4	20	0.4
3	20	0.4	10	0.4
4	40	0.2	20	0.4
5	40	0.4	20	0.4
6	40	0.4	40	0.4

The pigs were housed in fully-slatted, combined stage 1/stage 2 accommodation throughout the study. All pigs were weighed individually at 4, 7 and 10 weeks of age. Group feed intakes were recorded weekly, and food conversion ratios (FCR) calculated. Behavioural observations were made each hour between 1300 and 1600 hours on 5 separate days when pigs were 4 and 7 weeks of age, and on 1 day a week at 5, 6, 8 and 9 weeks of age. Each group was observed directly for 2-mins, and all aggressive behaviours (fighting, headthrusting, chasing or displacing from feeder or drinker) were recorded. The group was then scanned and the number of pigs lying or standing was recorded. All groups were allowed at 30-second 'settling period' prior to commencement of behavioural observations. Production data (using weaning weight as a covariate) and behaviour data were analysed by ANOVA using Genstat 5. In addition to treatment comparisons, effects of moving or staying in the same pen after groups were split were also assessed.

Results Between 4 and 7 weeks of age, feed intake was significantly lower in groups of 20 and 40 pigs when space allowances of 0.2m^2/pig rather than at 0.4m^2/pig were used (*40 pigs* (*0.2m^2*): 347.5^{a}, *40 pigs* (*0.4m^2*): 391.4^{bc}, *20 pigs* (*0.2m^2*): 361.3^{ab}; *20 pigs* (*0.4m^2*) 414.2^{c}; s.e.m. 14.47 g/day; P<0.05)). There were no significant treatment effects on growth rate or FCR, or on behaviour measures during this period (P>0.05). In addition, body weight at 7 weeks of age did not differ significantly between treatments (P>0.05).

Selected production performance results from the period between 7 and 10 weeks of age are presented in Table 2. When considering Treatments 4, 5 and 6, growth rate was significantly lower in Treatment 4 than in Treatments 5 and 6 (P<0.05). Body weight at 10 weeks of age was also lower in Treatment 4 than in Treatment 5 (P<0.01). No significant differences in production performance parameters were shown between Treatments 1, 2 or 3 during this period (P>0.05).

Table 2 Influence of treatment on production performance between 7 and 10 weeks of age

Parameter	Treatment						s.e.m.	Significance
	1	2	3	4	5	6		
Feed intake (kg/d)	1.12	1.10	1.14	1.06	1.13	1.12	0.027	
FCR	1.67	1.60	1.66	1.63	1.58	1.64	0.038	
DLWG (g/d)	669.2^{ab}	688.9^{bc}	685.5^{bc}	645.3^{a}	715.7^{c}	681.0^{bc}	12.56	***
10 week weight (kg)	29.3^{ab}	30.0^{b}	30.1^{b}	28.6^{a}	30.3^{b}	29.5^{ab}	0.41	**

No significant effects of either moving to a new pen or remaining in the same pen on production performance or behaviour between 7 and 10 weeks of age were recorded (P>0.05).

Conclusions Reducing space allowance for newly-weaned pigs adversely affected feed intake. A combination of increased group size and reduced space allowance during the immediate post-weaning period appeared to adversely affect performance in the second half of the post-weaning period. These effects did not appear to be accompanied by differences in aggressive behaviour or general activity levels.

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Effect of floor space during transport of market weight pigs on incidence of transport losses (dead and non-ambulatory pigs) at the packing plant

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Introduction Losses of pigs (dead and non-ambulatory) during transport are of great concern from both animal welfare and also economic perspectives. Based on several field studies, we have reported the incidence of transport losses in market weight pigs to be ~1% (Ellis *et al*., 2003). Transport losses can be influenced by numerous factors including genetics, carcass muscling, health status, structural soundness, live weight, nutrition, handling, facility design, and conditions during transport to the plant. Few, if any, of these factors have been examined under typical commercial conditions. Floor space on the trailer during transport is a potential contributing factor that can be easily changed by varying the number of pigs placed on the truck and represents a simple approach to managing transport losses. The objective of this study was to investigate effects of two floor spaces (0.39 and 0.48 m^2/pig) during transport, that represent the range currently being used in commercial practice in the U.S., on the incidence of dead and non-ambulatory pigs and to evaluate relationships between transport conditions and transport losses.

Materials and methods A total of 74 loads of market weight (mean BW 129.0, SEM 0.63 kg) pigs were taken from 2 commercial sites with 2 replicates per site. Loads from the first site were taken in April (n = 18) and October/November (n = 24) and loads from the second site were taken in July (n = 14) and January (n = 18). Standard commercial procedures were used for pig handling and transportation. Two different designs of flat-deck trailers were used with floor space treatments being compared in two similar sized compartments on each deck of each trailer type. Pigs were transported ~3 h to a commercial packing plant. The incidence of non-ambulatory pigs at the farm during loading and at the plant after unloading, average load weight, load number within each day, event times, and temperature and relative humidity in the trailer from loading to unloading were recorded. Also, non-ambulatory pigs at the plant were classified as non-ambulatory, injured (structure/injury related) or as non-ambulatory, non-injured (stress/fatigue related). Data for transport losses were subjected to a Chi-square rank-based transformation using PROC RANK procedure of SAS and were analyzed as a split-plot design using PROC MIXED procedure of SAS; the main plot was transport date, the subplot was load number within day, and the experimental unit was trailer compartment. The model included fixed effects of trailer design, trailer deck, trailer compartment, floor space, and all possible interactions, and random effects of transport day nested within farm and the load number within day by transport day interaction. Relationships between transport conditions and losses were evaluated using Pearson correlations derived using the PROC CORR procedure of SAS.

Results Increasing floor space during transport from 0.39 to 0.48 m^2/pig reduced ($P < 0.05$) the incidence of total non-ambulatory pigs, non-ambulatory, non-injured pigs, and total losses at the plant (Table 1). There was an interaction ($P < 0.01$) between floor space and trailer compartment for deads on arrival (Table 1). At the lower floor space, incidence of deads was higher in compartment 1 than in compartment 2, however trailer compartment did not affect the percentage of deads on arrival when pigs were provided with 0.48 m^2/pig during transport. The percentage of total losses on arrival at the plant was positively related to waiting time at the plant, unloading time, and total time from loading to unloading ($r = 0.24$, 0.51, and 0.36, respectively; $P < 0.05$). Average temperature in the trailer at various times during the journey (loading, waiting at the farm, transport, waiting at the plant, and unloading), and average pig weight on the trailer were not related to losses.

Table 1. Effects of transport floor space during transport on losses at the plant

Plant Losses	Floor space			
	0.39 m^2/pig	**0.48 m^2/pig**	**SEM**	**P value**
Non-ambulatory, %	0.62	0.27	0.13	0.04
Injured, %	0.09	0.12	0.06	0.98
Non-injured, %	0.52	0.15	0.11	0.01
Deads on arrival, %	-	-	-	-
Compartment 1	0.48^a	0.00^b	0.09	0.01
Compartment 2	0.06^b	0.17^b	-	-
Total losses, %	0.88	0.36	0.16	0.01

[a,b]Transport floor space by trailer compartment interaction, means with different superscripts differ ($P<0.05$).

Conclusions Floor space on the trailer had a substantial effect on transport losses and providing a higher level (0.48 m^2/pig) reduced transport losses and, consequently, improved welfare of pigs during transportation. In addition, transport times may impact losses at the plant.

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Influence of access to straw provided in racks on the welfare of sows in large dynamic groups

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Introduction Pregnant sows are traditionally fed a restricted diet which leaves sows unsatisfied and with a high motivation to forage (Meunier-Salaün, *et al.,* 2001). Recent amendments to European Union pig welfare legislation state that pregnant sows and gilts must be given a sufficient quantity of bulky or high-fibre food, as well as high-energy food, in order to satisfy hunger and the need to chew (Council Directive 2001/88/EC). The aim of this study was to assess the effect of providing access to straw in racks on the welfare of sows in a large dynamic group.

Materials and methods One hundred and twenty-two Large White x Landrace sows were allocated to one of two treatments over seven replicates. Treatments were as follows: (1) Access to racks containing chopped barley straw (offering an average of 0.2kg straw/sow/day), (2) Control, with no straw racks. Treatments were applied to two separate dynamic groups each containing 35 (±3) sows. Approximately 9 sows were replaced in each of these groups at 3-week intervals (each replacement constituting a replicate of the study). Treatments were swapped between the two dynamic groups after three replicates, with a 3-week adjustment period before observations recommenced. Both dynamic groups were housed in identical split-yard systems (18.2 x 7.8 m) with slatted exercise and drinking areas and solid-floored kennel areas in both the pre- and post-feeding yards. The pre-feeding yard was separated from the post-feeding yard by an electronic feed station, which supplied 2.2 kg concentrates/sow/day. In treatment 1, straw was provided in a rack (0.8m high x 1.2m wide, with 5.3cm^2 mesh) which was located in the slatted exercise area of the pre- and post-feeding yards. The rack was attached to railings and was suspended 0.3m above a collecting mat (0.6 x 1.5m). Resident pen behaviour was observed directly from three newly-introduced and three resident sows during a 5 min period on three afternoons in the first week, and two afternoons in the second week, after sows were replaced in the group. Sows were observed consecutively in a predetermined randomised order. A full ethogram of social, exploratory and aggressive behaviours was used. Each sow was also scanned instantaneously three afternoons during week 1, and on two afternoons during week 2, to determine their location and whether or not they were performing sham chewing behaviour. Aggression-related injury scores were measured from all newly-introduced sows at 1 week post mixing. Resident pen behaviour and injury scores were analysed by ANOVA, and sham chew scans were analysed by Binomial Regression using Genstat 5.

Results Selected behavioural results are presented in Table 1. Observations of resident pen behaviour showed that sows with access to straw spent approximately 9% of their time exploring the racks/consuming straw. Sows in the straw treatment spent a significantly lower percentage of time exploring the floor ($P<0.05$). No significant treatment effects were shown in overall exploratory behaviour ($P>0.05$). Overall levels of aggressive behaviour were low, however significantly greater levels were shown in the straw treatment ($P<0.05$). Injury scores did not differ significantly between treatments ($P>0.05$).

Table 1 The effect of access to straw on the duration of exploratory behaviours and frequency of aggressive behaviours

Parameter	Control	Straw	SEM	Significance
Exploratory behaviours (% of time)				
Explore straw rack/consume straw	-	9.21	-	-
Explore floor	12.4	4.8	1.76	*
Explore fixtures	7.1	7.0	1.72	
Overall exploration	19.6	21.0	1.75	
Aggressive Behaviours (min^{-1})				
Aggressive biting	0.01	0.02	0.005	
Fighting	0.00	0.00	0.001	
Headthrusting	0.01	0.03	0.006	
Overall aggression	0.00	0.01	0.001	*

A greater proportion of sows performed sham chewing behaviour in the post- rather than the pre-feeding yard ($P<0.01$), however, average levels of sham chewing behaviour did not differ significantly between treatments ($P>0.05$).

Conclusions The fact that sows with access to straw racks diverted almost half of their exploratory behaviour towards the racks, showing a preference for them over the floor/fixtures, suggests that the racks improved welfare. However, the lack of effect on sham chewing behaviour, and the slight increase shown in aggressive behaviour in the straw treatment, suggests that welfare benefits associated with this system are limited.

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Environmental conditions and changes in body temperature of sheep in a Western Australian summer feedlot

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Introduction Feedlots are important in both fattening of lambs prior to slaughter and familiarizing sheep with a pellet diet prior to live export. Feedlotting is prominent in Western Australia, with 19% of prime lamb producers identifying feedlotting as their main finishing method (Connell *et al*., 2002). Anecdotal reports are that temperatures within a West Australian feedlot during summer can often be in excess of 40˚C. Past research has shown that excessive heat load is a problem with feedlotted cattle (Hahn and Nienaber, 1993) and could be of particular importance when animals are being fed high energy rations; however, the extent of the problem in sheep is unknown. Previous work has shown that high temperatures lead to a decrease in feed intake and therefore productivity of the animal (Baile and Forbes, 1974). The aim of this study was to determine environmental conditions in a Western Australian feedlot during summer and to determine the core temperature of both wethers and ewes within this feedlot. The hypothesis was that sheep exposed to feedlot conditions during summer would have an increased core body temperature above normal.

Materials and methods Three Suffolk cross Merino ewes (35.2 kg ± 0.93) and three wethers (36.0 kg ± 1.26) were surgically implanted with core temperature loggers (Maximum Dallas). Sheep were held in a feedlot for a period of 40 days during February and March. Normal core temperature (NCT) was determined while sheep were in thermoneutral conditions indoors, prior to being taken to feedlot. While in the feedlot sheep were fed pellets and hay *adlib* and had water available *adlib*. Sheep had access to shaded areas while in the feedlot. Individual behaviour of the six sheep was monitored from 0900 to 1600 hours on four of the days that sheep were in the feedlot. Climatic conditions within the feedlot were monitored during the study using temperature data loggers (T-TEC Datalogger, South Australia) and a weather station (Onset computer corporation, USA). A Dunnett t-test was used to compare the mean core temperature of each day with the normal core temperature. A two sample t-test was used to test for differences in time that sheep spent doing particular activities while in the feedlot.

Results Both the ewes and the wethers had increased core body temperatures above normal during their time in the feedlot ($p < 0.05$). During the times behaviour was monitored, the sheep spent significantly more time resting in the shade then resting in the sun or eating ($p < 0.01$). The dry bulb temperature reached a maximum at or above 35˚C on 20 out of the 40 days that sheep were in the feedlot.

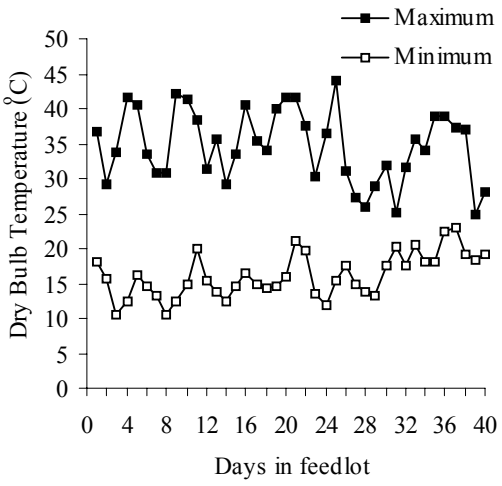

Figure 1 Daily minimum and maximum environmental dry bulb temperature within the feedlot

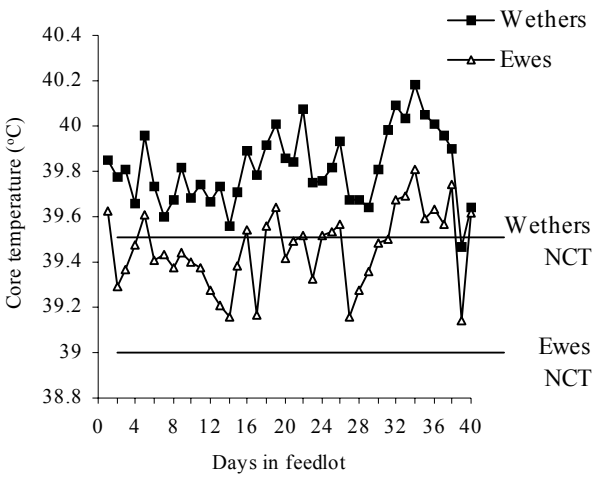

Figure 2 Mean daily core temperature of ewes and wethers while in the feedlot, compared to normal core temperature (NCT) at thermoneutral conditions

Conclusions The results of this study demonstrate that both ewes and wethers had increased core body temperatures above normal while in the feedlot. Other studies have shown that increased core temperature is associated with a decrease in feed intake (Baile and Forbes, 1974; Hahn and Nienaber, 1993). More intensive studies will use the data gathered here to test if there are deleterious effects of heat stress on feed intake and production of sheep under conditions experienced in summer feedlots.

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Comparative analysis of wither height, body depth and ground clearance between elite, potential elite and non-achieving event horses

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Introduction The relative diversity in body shape and type between breeds indicates a relationship between body shape and performance. Numerous studies on the relationship between body shape and performance are reviewed by Sasstamoinen and Barrey (2000). A limited number of studies (Langlois *et al,* 1978) have looked at the relationship between horse height at the wither and performance. Holmstrom *et al,* (1990) found elite dressage and show jumping horses had a higher mean wither height than four year old quality assessed riding horses and riding school horses. This study aimed to investigate the relationship between wither height and performance in three groups of event horses, elite, potential and non-achieving. Additionally wither height is analysed via body depth and ground clearance and performance level within the three groups. The UK a world leader within the equestrian sport of eventing, currently the Olympic individual and team silver medallist winning nation.

Materials and methods Three groups of event horses were used for the analysis. The groups were defined by competitive level. Elite performers n=12, consisting of horses that were competing at international advanced eventing level (CCI 3 star). Potential elite (PE) young horses n=11, consisting of horses that were competing in the British Equestrian Federation young Horse Evaluation Series – event horse section. Non-achieving (NA) performers n=14, consisting of horses that had competed at affiliated eventing for a minimum of two years but had failed to progress beyond pre-novice level. PE horses were all over 4 years of age so maturity was not deemed to be a factor in the analysis. Each individual horse was stood square on a level piece of ground and had a digital image taken from a 3m distance. The recorded digital image was analysed via a computer aided design programme. Measurements were recorded for three height dimensions (Figure 1). Wither height (distance from ground to top of wither), body depth (distance from wither to brisket) and ground clearance (distance from brisket to ground). Normality of data distribution was undertaken via skewness test. Descriptive statistics and measures of dispersion were produced. One way Analysis of variance was performed on the data sets.

Results Table 1 present's measures of central tendency (mean) and dispersion (standard deviation, minimum, maximum, skewness) for the three groups. Table 2 returns the Analysis of variance for the studied groups, highlighting a significant difference between the groups with regard to ground clearance. Although not significant a trend was apparent in relation to overall wither height (Table 2).

Table 1 Descriptive statistics, wither height, body depth and ground clearance between groups

Measurement	Group	Mean	Std.Err	Std.Dev	CL, P=0.05
Wither Height (WH)	NA	158.61	1.653	6.183	3.638
	PE	162.51	2.069	6.861	4.610
	Elite	164.87	2.008	6.954	4.340
Body Depth (BD)	NA	77.06	0.859	3.216	1.891
	PE	75.58	1.802	5.978	4.109
	Elite	79.32	0.835	2.892	1.804
Ground Clearance (GC)	NA	81.56	0.981	3.671	2.159
	PE	86.90	1.872	6.208	4.171
	Elite	85.55	1.425	4.936	3.078

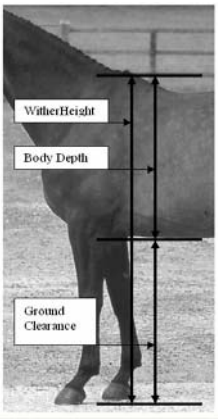

Figure 1 Measured heights.

Table 2. ANOVA, wither height between groups.

Measurement	SS Between	SS Within	F value	P value
WH	260.467	1499.753	2.956	0.066
BD	82.005	583.767	2.388	0.107
GC	197.705	828.644	4.056	0.026

df = 2,34, SS= sums of squared.

Conclusion. This study highlights that elite and potential elite horses have greater wither height than non-achieving horses. On average elite horses are 3.8% and potential elite horses 2.4% taller than non-achievers. This difference is observed to be most notable and significant when assessing the distance between the ground and brisket of the horse. The difference equates to 4.7% for elite horses and 6.1% for potential elite horse. The sample studied is small and any conclusions have to be carefully considered.

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Comparative analysis of shoulder angles between selected groups of elite and non-achieving event horses

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Introduction The relationship between conformational structure and function in the equine is thought to be important in the judgement of the future soundness and performance of the horse (Holmstrom *et al* 1990). Holmstrom *et al* (*ibid*) demonstrated that elite dressage and show jump horses possess a more horizontal shoulder enabling the horse to demonstrate an improved forelimb technique. Holmstom *et al* (*ibid*) further related this difference as a contributory factor to the higher levels of performance observed in the elite groups he studied. Furthermore Holmstrom and Philipsson (1993) showed a positive correlation between 'good' gaits and a sloping shoulder. Additionally Holmstrom and Philipsson (1993) found that those horses which had a more vertical position of the scapula and a straighter shoulder joint suffered more concussion and are, therefore, at a greater risk of developing lameness. This study investigated the relationship between shoulder angle and performance within three groups of event horses.

Materials and methods Two groups of event horses had two shoulder angle measurements taken. The groups were defined by competitive level. Elite performers n=32, consisting of horses that were competing at international advanced eventing level (CCI 3 star). Non-achieving (NA) performers n=30, consisting of horses that had competed at affiliated eventing for a minimum of two years but had failed to progress beyond pre-novice level. Each individual horse was measure at a competitive event before the cross country phase of competition. Horses were stood square on a level piece of ground. Three reference points were marked on the horses shoulder using 2cm diameter square fluorescent stickers. The reference points used were the proximal end of the scapula (reference point 1), the cranial part of the greater tubercle of humerous (reference point 2) and the olecranon process of ulna (reference point 3) These points were established through palpation in accordance with the protocol of other studies (Holmstrom *et al,* 1990). A digital image was then taken of the horse from a 3 metres distance. Analysis of the digital image was via a computer aided design programme (AutoCad 2003). Two angles of shoulder were analysed 'closed' shoulder angle (CS) and 'open' shoulder angle (OS) (Figure 1). Normality of data distribution was undertaken via skewness test. Descriptive statistics and measures of dispersion were produced for each data set. Independent-sample *t* tests were returned for both normal and open shoulder angles between the groups. Confidence intervals were returned at 95% for all tests. Pearsons product moment correlations were returned between the two shoulder variables within each studied group.

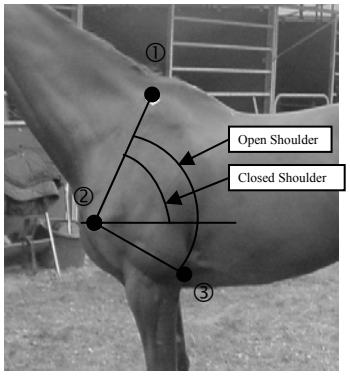

Figure 1 Measured shoulder angles

Table 1 Comparative measures of shoulder angle between elite and non-achieving groups

Angle	Group	Mean angle °	Err Std.	Std.Dev	CL, P=0.05
CS	Elite	58.766	0.709	4.010	1.450
	NA	62.033	0.705	3.862	1.442
OS	Elite	88.609	0.949	5.367	1.938
	NA	91.183	1.051	5.757	2.149

Results Table 1 returns descriptive statistics and measures of dispersion for each data set. Levenes test established equality of variance for both data sets. Independent sample *t* test results for the normal shoulder angle highlighted a significance $P<0.01$, $t=-3.264$, $df=60$. Open shoulder angle highlighted a significant difference $P<0.01$, $t=-1.822$, $df=60$. Pearsons product moment correlations for the elite group returned a value of 0.400 ($P<0.05$), the non-achieving group returned a value of 0.629 ($P<0.01$)

Conclusion The study highlighted elite horses as having a significantly ($P<0.01$) smaller angle of shoulder with respect to both the closed and open angle. This means that the elite horses having a less 'upright' shoulder angle. This finding concurs with the work of other authors, (Holmstrom *et al,* 1990; Holmstrom and Philipsson, 1993). These differences may be useful as quantifiable aids to selection of potential elite horses at a young age. It however should be noted that the sample size is relatively small and further work on a larger population is required. Additionally the differences highlighted in this study amount to a 3.267° difference in normal shoulder angle and 2.574° difference in open shoulder angle. These differences are relatively small and maybe difficult to apply in practise to a selection programme. Correlation coefficiencies showed a reasonably strong relationship between the two angles within both groups studied.

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Relationship between Third Metacarpal length and lower forelimb unsoundness in National Hunt racehorses

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Introduction Unsoundness amongst racehorses has considerable financial and welfare implications; Sasstamoinen and Barrey (2002) state the value of an athletic horse is largely determined by its ability to exercise and the health of its limbs. The forelimb of the horse is more likely to be predisposed to injury as a consequence of it supporting more than 65% of the horse's bodyweight (Hodgson and Rose, 1994). Unsoundness associated to the third metacarpal (MC3) and associated soft tissues has been documented within the equine (Walter and Davies, 2001). Davies (2001) established that MC3 size and shape were significantly related to strain loadings within the equine forelimb. He concluded that unsoundness may be closely related to bone size and shape. Speirs (1994) states that the ability of bone to resist imposed loads is related to many factors – including bone geometry. Additional work (Davis, 2001) has concluded that bone strength and stiffness depend on the physical properties of the bone, overall anatomical shape as well as loading effects. This study aimed to investigate whether there is any link between MC3 length and recorded lower forelimb unsoundness in National Hunt racehorses.

Materials and methods Data were collected from a sample of 100 National Hunt racehorses. Horses were defined as; Group one, having no previous history of lower forelimb unsoundness n=63; or Group two, having a previous history of lower forelimb unsoundness n=37. Allocation of horses to groups was established via the previously recorded veterinary history for each animal. All horses measured were at least four years of age, therefore maturity was not deemed to be a contributor factor. Data were collected from the near fore leg of each animal. Length of MC3 was obtained from measuring from the bottom of the fourth carpal bone to the top of the lateral proximal sesamoid bone. Minimal soft tissue structures surrounding the MC3 make the MC3 a good site for measurement and assessment (Davis, 2001). Measurements were taken with accuracy to the nearest millimetre. Data were tested for normality of distribution via the skewness test. Descriptive statistics were established from each group. Levenes test of equality of variance were performed on the data sets. Additionally data was analysed via independent sample student *t* test (two tailed). Confidence intervals were returned at 95% for all tests.

Results Descriptive statistics for each group are given, table 1. Skewness test demonstrated normality in distribution within the two groups. The Levenes test established equality of variance between the two groups.
Independent sample *t* test established a significant difference in the measured MC3 length (t =4.149, df = 98, P<0.001) between those horses that had no history (Group One) and those that had a history of lower limb unsoundness (Group Two)

Table 1 Lower forelimb descriptive statistics of two measured groups.

	Mean	Std.Err	Std.De	CL, P=0.05
Group One	18.76	0.899	0.714	1.798
Group Two	19.4	0.131	0.794	0.266

Conclusions This study demonstrates that there is a significant difference in MC3 length between those horses that have a previous history of lower forelimb unsoundness and those that have no previous history. Such information may aid selectors in assessing the potential suitability of horses for National Hunt racing. Additionally this information may aid trainers in formulating appropriate training regimes for National Hunt horses in training. However it should be noted that the difference although significant is small, 0.64cm between groups. This difference equates to 3.3% of the total length of the measured MC3. It may therefore be argued that such a difference may be very difficult to accurately quantify in a practical situation. It should furthermore be noted that within this study due to confidentiality agreements a number of confounding factors are clearly evident. There is no linkage of horses to specific trainers, no analysis of training regimes, number of times a horse has previously run or distance performed over. Such factors may to play a significant role in the occurrence of lower forelimb unsoundness in the national hunt racehorse.

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Forage plant preference of work donkeys in the Kweneng Sandveld, Botswana

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Introduction Donkeys have historically been the least valued farm animal in Botswana. However, following commercialization of the beef cattle industry coupled with escalating oil prices, donkeys have gained popularity as both draught and transport animals in most remotely located rural areas of the country. Further, due to their resistance to drought, the population of donkey in the country has substantially increased while those of cattle, sheep and goats have decreased. Without exception, all donkeys in Botswana derive their food from the open free-range. Despite their growing popularity as draught animals, very little attention has gone into research on foraging tactics of donkeys. This study was undertaken to determine forage preference of free-ranging work donkeys utilizing an *Acacia* dominated sandveld in Botswana. The expectation of the researchers was that due to limited grazing time, work donkeys grazed all available grasses without regard to choice in which case the quality of their diet from the range, compared to that of their non-work donkey compatriots would be suspect.

Materials and methods The study was conducted during the peak agricultural season when donkeys were working the most. Faecal materials were collected from kraals in homesteads known to use donkeys for draught power. Forage availability samples were collected in the surrounding grazing areas. Both faecal and forge samples were collected monthly from December 2003 to March 2004. Faecal samples were analyzed michrohistologically (Storr 1961) to determine diet botanical composition (Sparks and Malechek 1968) while plants from the range provided data on forage availability as well as reference material for identification of diet botanical composition material. Forage preference was determined via the Ivlev's Elasticity Index (Ivlev 1961): $E_i = r_i - n_i / r_i + n_i$ where: E_i = Inlev's elasticity index measure of species i; r_i = % of species in the diet; n_i = % of species in the range (Ivlev, 1961). The index has values from –1 to 1. Species with values close to 0 are consumed without regard to choice while those with values close to –1 and 1 are rejected and preferred, respectively. Availability of forage on the range was determined via the point intercept procedure (Bonham 1989). Available forage was classified as being of good, intermediate and poor forage value based on their crude protein content and digestibility

Results Of the 25 grasses species isolated in the botanical diet composition of donkeys (Table 1), 36% were of good forage value while the intermediate and the poor forage value each comprised 32% each. All the good forage value grasses were highly preferred while the intermediate ones were either slightly preferred or consumed without regard to choice. All poor forage value grasses were rejected. These results show that work donkeys are sensitive to the forage value of the plant they consume. They prefer grass plants high in both crude protein content and digestibility. These results agree with Van Soest (1987) that survival, growth and productivity of free-ranging animals depend on forage quality.

Table 1. Forage preference (Pref) rating of the good, intermediate and poor grasses by donkeys in the Kweneng sandveld, Botswana

Good value species	Pref	Intermediate value species	Pref	Poor value species	Pref
Anthephora pubescens	0.75	Cynadon dactylon	0.15	Aristida congesta	-0.45
Bothriochloa insulpta	0.65	Chloris virgata	0.05	Eleusine africana	-0.40
Brachiaria nigropedata	0.35	Eleusine africana	0.10	Enneapogon scoparium	-1.00
Cenchrus ciliaris	0.55	Eragrostis lemanniana	0.30	Eragrostis pallens	-0.20
Digitaria milanjiana	0.70	Eragrostis rigidior	0.20	Parotis patens	-0.30
Eragrostis superba	0.60	Heteropogon contortus	0.00	Pogonathria squarrosa	-1.00
Panicum maximum	0.75	Stipagrostis uniplumis	0.25	Rhynceletrum repens	-0.40
Schmidtia pappophoroides	0.80	Setaria sphacelata	0.0	Tragus racemosus	-0.20
Orochloa trichopus	0.55				

Conclusion Work donkeys in the study area highly preferred grasses of good forage value, grazed the intermediate forage value species without regard to choice and rejected all poor forage value grasses during the study period. These results suggest that despite time spent working, work donkeys, like other animals preferred those forage grasses that helped ensure survival, growth and productivity of the animal implying that they acquire through grazing just as much nutrients as their none work donkey compatriots.

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Embryo production in defined sequential media and survival following vitrification in a sealed system

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Introduction Conventional *in vitro* embryo production and cryopreservation strategies applicable to livestock could benefit from further enhancement of biosecurity features. In particular, there is scope for a new generation of sequential culture media that avoid ill-defined constituents but closely match changing embryo needs. Likewise, complementary cryopreservation protocols using sealed containers would be beneficial, if they emulate or surpass existing embryo storage and subsequent survival standards. This study compared blastocyst yields from zygotes incubated in a conventional monoculture medium or in an alternative defined 2-stage sequential media system, with or without Trolox, a water-soluble antioxidant, and subsequently investigated blastocyst survival following vitrification in sealed CryoTips.

Materials and methods Good quality presumptive zygotes, generated via conventional in vitro maturation and fertilization (n = 7 replicates; IVF = Day 0) were allocated randomly for culture from Day 1 in Synthetic Oviduct Fluid medium supplemented with BSA (4 g/l), in the absence (SBSA) or presence of Trolox (0.1 mmol/l; SBSAT), or in a two-stage culture regimen with chemically defined formulations, V1 (up to Day 4) and V2a (Day 4 onwards), also in the absence (V1/V2a) or presence (V1T/V2aT) of Trolox. V1 and V2a were formulated to meet the needs of early and later stage embryos, respectively; of these, only V1 contained polyvinyl alcohol (3 g/l), EDTA (0.05 mmol/l), citrate (0.5 mmol/l) and taurine (0.1 mmol/l) while only V2a included hyaluronic acid (0.5 g/l) and essential amino acids (BME, 50x; 20 ml/l). Incidence of egg cleavage was recorded on Day 2 and blastocyst yields, zona-inclusive diameters and (in 3 replicates) cell numbers were recorded on Days 7 to 9. In addition, blastocysts from 4 subsequent replicates, produced in either SBSA (n=25) or V1/V2a (n=28), were vitrified in biosecure vials (CryoTip; Irvine Scientific) using a minor modification of the method of Moreira *et al.* (2005) and then stored in liquid nitrogen. Incidences of egg cleavage, of blastocyst yields as proportions of eggs cleaved, and of blastocyst survival post-vitrification were compared (following arcsin transformation) using ANOVA. Where two-way ANOVA was precluded, ANOVA with Bonferroni test was used. Blastocyst cell count (log-transformed) and diameter data were tested using REML.

Results Incidence of egg cleavage revealed significant interaction ($P<0.05$), with Trolox apparently of benefit only when SBSA was used. The V1/V2a system yielded blastocysts only when Trolox was excluded (Table 1) and then blastocyst yields were equivalent to those from SBSA and SBSAT, with corresponding blastocyst cell numbers and diameters also not differing significantly. Post-vitrification blastocyst survival data are presented in Figure 1; survival of Day 8 blastocysts from V1/V2a was superior to that of counterparts from SBSA ($P<0.05$; *).

Table 1 Yields of cleaved eggs and blastocysts. Data are untransformed mean±s.e.m. values.

	n	Cleaved zygotes (proportions)	Blastocysts on Day 7 (proportions)	Blastocysts on Days 7, 8 & 9 (proportions)	Blastocyst cell numbers (n)	Blastocyst diameters (μm; n)
SBSA	123	0.83±0.04	0.09 ± 0.04^{a}	0.29 ± 0.08^{d}	85.9±9.5 (12)	173.0±4.1 (28)
SBSAT	123	0.90±0.04	0.09 ± 0.02^{b}	0.31 ± 0.04^{e}	91.0±6.3 (12)	171.6±2.7 (31)
V1/V2a	173	0.92±0.04	0.03±0.02	0.14 ± 0.05^{c}	94.0±8.4 (5)	168.1±4.6 (17)
V1T/V2aT	161	0.87±0.04	$0^{a,b}$	$0^{c,d,e}$	-	-

Within columns, values with a common superscript differ as follows: $^{a,b}P<0.05$, $^{c}P<0.01$, $^{d,e}P<0.001$

Figure 1 (above) CryoTips used in the present study were sealed at positions indicated by the vertical lines.

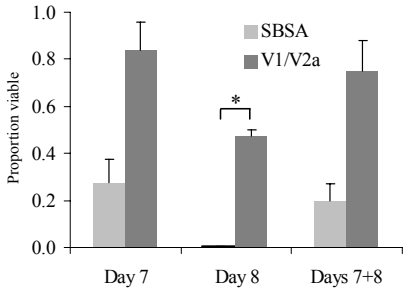

Figure 2 (right) Incidence of vitrified blastocyst survival after re-warming and short-term culture.

Conclusions Embryos cultured in defined conditions in the absence of Trolox achieved developmental rates equivalent to counterparts cultured in conventional medium. Blastocyst survival following vitrification in sealed CryoTips was good and the protocol was applicable to blastocysts produced in the defined *in vitro* culture system.

Acknowledgements SAC is funded by SEERAD; CryoTips were donated by Irvine Scientific; VCM is supported by the Meat and Livestock Commission and by Britbreed Ltd.

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Changes in testicular size, body weight and plasma testosterone concentration in Iranian Holstein bull during the growth period

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Introduction Testicular development of bulls, during the post-weaning period, is associated to the age and breed of the animals, environmental conditions, and nutritional regime. A positive correlation between testicular development and fertility has been documented through numerous studies (Bailey *et al.* 1996). Scrotal and testicular measurements, which are simple and inexpensive to obtain, have been used to predict sperm production and semen quality. However, because SC is an indirect measurement of testicular mass which discounts variation in scrotal wall thickness and individual testis shape, the length and width measurements could provide a useful tool for predicting bull fertility. Therefore we conducted a study to evaluate testicular growth during puberty and to determine 1) appropriate models for analysis of phenotypic relationships of testicular dimension and growth traits to age during puberty; and 2) correlations of testis width and length to plasma testosterone and body weight in pubertal Holstein bulls under environmental conditions of Iran.

Materials and methods Eleven Holstein bull calves with approximately six months of age were selected from the herd of department of animal science, university of Tehran in Karaj. Calves were fed twice a day with the ration balanced on the basis of NRC 1996. Body weight (BW), testicular width (TW) and testicular length (TL) were measured at three week intervals from six through twelve months of age. A calibrated caliper was applied to determine TW and TL. TL was measured in the left testis, while maintaining the testicles toward the bottom of the scrotum. In order to determine the plasma concentration of testosterone, blood samples of six calves were collected from the jugular vein in 7 (peripuberty), 8.5 (puberty) and 10 (post puberty) months of age and plasma testosterone were measured using TESTO-RIA-CT kit (Bio source Europe S.A. Belgium).
Regression analysis was performed using the Statistical Analysis System (SAS 2001) in which age was included as independent variable and each trait was analyzed as dependent variable; followed by scatter graphs were generated and curves were fitted based on goodness of fit through the R-squared (R^2) method (Aponte *et al*. 2005). Moreover, Pearson correlation analyses between variables were used to measure relationships among age, testicular measurements and plasma testosterone using the proc corr of SAS.

Results Data from BW, TW, and TL fitted linear curves from which interpolations could be made for analysis of individual cases (figures 1, 2 and 3). The general equation for the model is shown in each figure. Pearson correlation coefficients between BW, testicular measurements and plasma testosterone are shown in Table 1.

Figure 1.

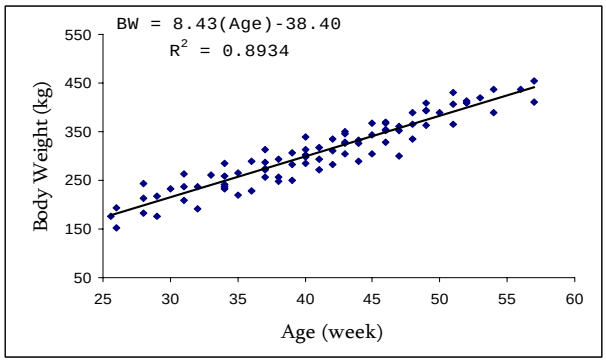

Figure 2.

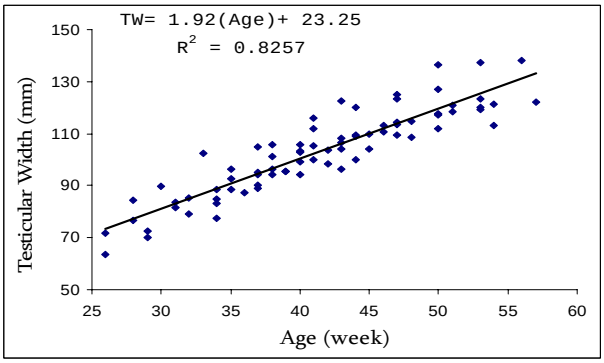

Figure 3.

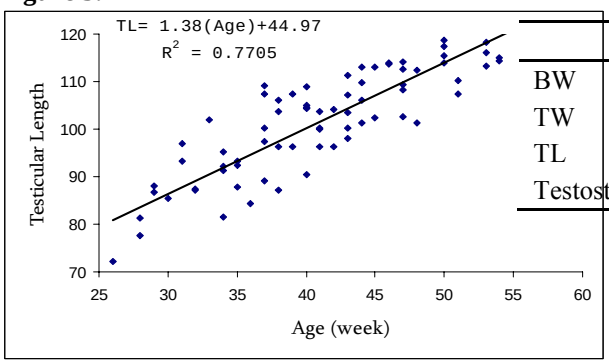

Table 1

	BW	TW	TL	Testosterone
BW	1.00	0.92**	0.95**	0.42ns
TW		1.00	0.86**	0.46ns
TL			1.00	0.51*
Testosterone				1.00

* Significant ($p<0.05$)
** Significant ($p<0.01$)
ns Non-significant

Conclusion Results from the present study indicated that as the bulls reached the age of puberty, testicular dimensions and body weight increased rapidly; however the increase in plasma concentration of testosterone was not highly correlated with these traits.

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Systemic progesterone and retinol binding-protein expression in the bovine uterus

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Introduction Early embryo loss is a major cause of reproductive wastage in the dairy cow. Stronge *et al.* (2005) reported that low concentrations of progesterone around days 5 to 7 was associated with a low probability of embryo survival. McNeill *et al.* (2005) have shown that the gene encoding retinol binding-protein (RBP) is sensitive to changes in progesterone around day 7 of the oestrous cycle. However, changes in gene expression are not always translated into corresponding changes in protein expression. The objective of this study was to investigate the relationship between systemic progesterone and retinol binding protein expression in the uterus of cyclic cows on days 3, 7, 11 and 15 of the oestrous cycle.

Materials and methods Spontaneously cycling, lactating Holstein-Friesian dairy cows at least 50 days post-partum were used. In Experiment 1 flushings were collected non-surgically from the uterine horns ipsilateral and contralateral to the corpus luteum (CL) on days 3, 7, 11 and 15 of the oestrous cycle (n=6 per day) by flushing with 50ml saline into a tube containing protease inhibitors. In Experiment 2 flushings were similarly collected from 26 cows on day 7 only. All flushings were centrifuged and stored at -80°C until analysis. Plasma samples were taken before and after uterine collections for progesterone and RBP determinations. Before assay for RBP uterine flushings were concentrated using centrifugal filter devices with a cut-off of 10kDa. Retinol binding protein was determined in uterine flushings and plasma by an ELISA using retinol-binding protein purified from bovine plasma as a standard. Protein was determined using the Bradford assay. Progesterone concentration was determined using the Coat A Count (DPC) direct RIA. Statistical analysis was carried out using Proc GLM (SAS v.8.02, Cary, NC). Data was transformed to approximate normal variates where appropriate. Significant differences were compared using Tukey's option. Data are presented as backtransformed means and standard errors. A probability of $p<0.05$ was considered significant.

Results In Experiment 1, uterine total protein was lower on day 11 than days 3 or 15 ($p<0.01$). There was no difference in uterine total protein between any of the other days ($p>0.05$). There was a significant day and day by side interaction on uterine total RBP with the ipsilateral side over 3-fold higher ($p<0.05$) than the contralateral side on Day 15 (Fig. 1). There was no difference between sides on any other day ($p>0.05$). Day 7 and 11 uterine total RBP was lower than Day 3 ($p<0.05$) and Day 15 total RBP content was 6- to 13-fold higher ($p<0.001$) than that on all other days. There was a significant day and day by side interaction on uterine RBP concentration when expressed per unit of uterine total protein, with the ipsilateral side over 2-fold higher ($p<0.001$) than the contralateral side on Day 15. RBP concentrations were lower on day 11 compared to day 3 ($p<0.05$) and 5-15-fold higher on day 15 compared to all other days ($p<0.001$). In Experiment 2, uterine RBP concentrations were numerically higher in the uterine horn ipsilateral to the CL, however, these differences were not significant ($p>0.05$). There was a significant correlation in RBP concentrations between both uterine horns ($R^2=0.86$, $p<0.001$). There was no significant relationship between RBP content or concentration and the concentration of systemic progesterone ($p>0.05$). In both Experiments RBP concentrations were not different to that of blood plasma concentrations on day 3 to 11 but were 6-15 fold higher on day 15.

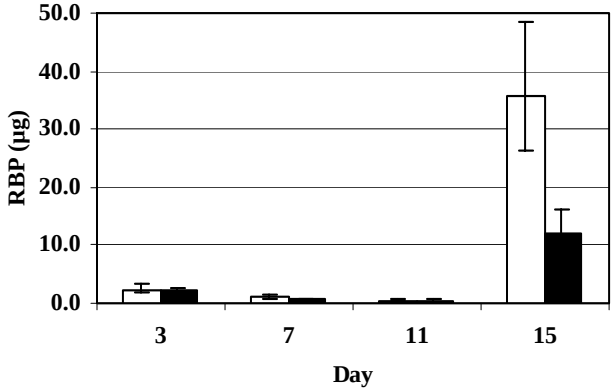

Figure 1 Uterine total RBP content of ipsilateral and contralateral uterine horns on day 3, 7, 11 and 15 of the oestrous cycle (open bars = ipsi; closed bars = contra).

Conclusions This study shows that both uterine RBP content and concentration when expressed per unit of total uterine protein declines from Day 3 to Day 11 but increases again dramatically by Day 15. It also shows that the RBP content and concentration of the uterine horn ipsilateral to the corpus luteum is over 3-fold and 2-fold higher respectively than the contralateral side on Day 15, suggestive of an effect of the local hormone environment on uterine RBP. Uterine and plasma RBP concentrations are similar on Days 3 to 11 but uterine values are 6- to 15-fold higher on day 15, a further indication of a local controlling mechanism. This study shows that the increase in uterine RBP is consistent with the observed increase in uterine RBP gene expression recorded in previous studies.

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The relationship of plasma leptin concentration and puberty in the Holstein bull calves

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Introduction Puberty is the phase of development during which the potential to reproduce is first realized. Puberty is associated with a variety of physiological and behavioural changes brought by testicular maturation and the production of adult levels of testosterone. These events are the result of re-activation of the gonadotropin releasing hormone (GnRH) neuronal system, which after a brief prenatal period of activation is quiescent throughout early postnatal life (Romeo *et al*., 2002).
The hypothesis that leptin plays an important role in regulating GnRH secretion, and ultimately in reproduction, stems from several studies. However, both sexual maturation and nutritional status are important determinants of how leptin affects the H-P-G axis in ruminants. Leptin is one of several permissive factors, whose presence is necessary but alone is not sufficient to initiate sexual maturation in rodents (Williams *et al*., 2002; Spicer *et al*., 2002). Furthermore, Garcia *et al*. (2002) reported that serum leptin increases linearly as puberty approaches in heifers, hence it is plausible to hypothesize that leptin may play a functional role in regulation of the central reproductive axis. The objective of this experiment was to study the changes of plasma leptin during puberty and its relationship with testosterone and testis dimensions in bull Holstein calves.

Materials and methods Six Holstein bull calves with approximately 7 months of age were selected from the herd of department of animal science, university of Tehran in Karaj. Calves were fed twice a day with the ration balanced on the basis of NRC 1996. In order to determine the plasma leptin concentration, blood samples were collected from the jugular vein in 7 (peripuberty), 8.5 (puberty) and 10 (postpuberty) months of age before feeding. Moreover, blood samples were collected at 20 minutes intervals for four hours to determine plasma testosterone secretion pattern on the same day. Plasma obtained after centrifugation was stored at -20°C. Plasma leptin and testosterone were measured using leptin RIA kit (Tabeshyarnoor Ltd, Iran) and TESTO-RIA-CT kit (Biosource Euroupe S.A. Belgium) respectively. In addition, body weight (BW), body condition score (BCS) and testicular width and length were determined. Circulating concentrations of leptin and testosterone, frequency and amplitude of testosterone pulses, BW, BCS and testicular dimensions were analysed by ANOVA for repeated measures using the Mixed Procedure of SAS (Garcia *et al*., 2002). Bull was used as the subject for the MIXED procedure to account for correlated variation within animals. The least squares means procedure was utilized to compare means when a significant difference was detected in the MIXED analyses.

Results Mean plasma leptin concentrations during the period of peripuberty was significantly higher than those during puberty ($P<0.01$) and postpuberty ($P<0.01$). Mean plasma testosterone concentrations during peripuberty and puberty was significantly different ($P<0.05$), however during the peripuberty and postpuberty no significant differences were observed. Testosterone pulses frequencies and amplitudes during peripuberty, puberty and postpuberty was also different ($P<0.01$). Mean plasma concentrations of leptin and testosterone and pulse frequency and amplitude of testosterone are shown in Table1.

Table 1 Mean hormone concentrations and testosterone pulse frequency and amplitude during three stages of blood sampling

	peripuberty	puberty	postpuberty	S.E.M
**Leptin concentration (ng/ml)	3.40^{a}	2.34^{b}	1.95^{c}	0.220
*Testosterone concentration (ng/ml)	0.49^{a}	0.74^{b}	0.66^{ab}	0.073
**Testosterone pulse frequency (per 4 hours)	2.00^{a}	1.00^{b}	1.20^{b}	0.116
**Testosterone pulse amplitude (ng/ml)	0.99^{a}	1.81^{b}	1.92^{b}	0.128

$^{a\,b\,c}$ Means with different superscript are significantly different (* $P<0.05$; ** $P<0.01$).

Conclusions Results indicate that in growing bull calves, plasma concentrations of leptin decreased during puberty while, circulating testosterone increased. It seems that decrease of plasma leptin is due to the negative effect of testosterone on production of leptin (Spicer *et al*., 2001). On the contrary, serum leptin level increased linearly in growing heifers (Garcia *et al*., 2002). Hence, it can be concluded that role of leptin in the neuroendocrinology of sexual maturation is different in bulls and heifers.

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Dose-response effects of exogenous letrozole on Day 11.5 porcine uterine and conceptus oestradiol concentrations

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Introduction Previous studies from our group suggest that the synchronised onset of oestradiol secretion by pre-implantation conceptuses from Meishan pigs may contribute to the enhanced prenatal survival characteristic of this breed (Ashworth and Pickard, 1998). We have recently reported that *in vitro,* the onset of oestradiol secretion by conceptuses can be regulated in a dose-dependent manner by an aromatase inhibitor (letrozole; Ashworth *et al*., 2005). Before commencing *in vivo* trials to assess the efficacy of letrozole to promote prenatal survival, it is important to determine whether exogenous letrozole reaches the uterine lumen and to establish the relationship between the dose administered and the impact on oestradiol secretion at the conceptus-uterine interface.

Materials and Methods Oestrous cycles of 13 Large White crossbred gilts were synchronised using Regumate during their second post-pubertal cycle. Gilts were naturally mated at their next oestrus and received either 0 (n=4), 1 (n=3), 10(n=3) or 100 (n=3) mg letrozole dissolved in 3.5% w/v polyethylene glycol 300 as 2 ml intra-muscular injections at the base of the ear at -26 and -2 hours before slaughter on Day 11.5 (Day 0 = first day of standing oestrus). Each uterine horn was flushed with 20ml Minimum Essential Medium to recover conceptuses. Conceptuses and uterine flushes were separated by centrifugation and frozen at -20^{0}C. Blood samples were collected immediately prior to the first letrozole injection and at slaughter. In addition, samples of liver and kidney were obtained at slaughter to assess the distribution of letrozole. Concentrations of oestradiol-17β in plasma, uterine flushes and conceptus homogenates were determined by radioimmunoassay. Concentrations of letrozole in the liver, kidney, uterine flush and conceptus homogenates were determined by HPLC. The effects of letrozole dose on actual and log-transformed concentrations of letrozole and oestradiol-17β were analysed by regression.

Results 3/4, 1/3, 3/3 and 3/3 gilts from the 0, 1, 10 and 100mg letrozole treatment groups respectively were pregnant at slaughter. Positive dose-dependent relationships were detected between the dose of letrozole injected and its abundance in liver, kidney, uterine flush (P<0.001) and conceptuses (P< 0.01). There were no differences between treatments in pre-injection plasma concentrations of oestradiol-17β, although both plasma and uterine flush samples collected at slaughter showed a dose-related reduction in oestradiol-17β concentrations. Conceptuses from the control and 1 mg letrozole treated gilts contained most oestradiol-17β. A dose-related reduction in conceptus oestradiol-17β content was observed following administration of the higher doses of letrozole (Table 1).

Table 1 Effect of different doses of letrozole on oestradiol-17β levels in plasma, uterine flushes and conceptuses

	Dose of Letrozole (mg)				Effect of dose *P*
	0	1	10	100	
Oestradiol-17β					
Plasma prior to treatment (pg/ml)	1.55±0.25	1.23±0.21	1.44±0.45	1.11±0.18	0.354 (ns)
Plasma at slaughter (pg/ml)	1.60±0.2	0.70±0.14	0.42±0.06	0.49±0.09	0.16 (ns)
Uterine flush at slaughter (pg/ml)	38.32±26.32	12.87±12.58	12.67±7.06	3.33±2.03	0.33 (ns)
Conceptus (pg)	260.7±142.0	697.5	123.7±103.5	42.8±21.5	0.09 (ns)

ns: Not significant

Conclusion This study demonstrated that letrozole, administered as an intramuscular injection (a) appears to be distributed throughout the body in a dose dependent manner and (b) reduces *in vivo* oestradiol-17β production by the Day 11.5 porcine conceptus. Further studies will be required to assess the effects of transient letrozole-induced changes in the synthesis of oestradiol by the conceptus on prenatal survival.

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The importance of body composition in explaining variation in feed conversion efficiency and residual feed intake between meat sheep at two different ages

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Introduction Residual feed intake (RFI) is a measure of feed efficiency calculated as the difference between an animal's actual intake and its expected intake based on its liveweight and growth rate over a specified period of time (Richardson, Herd *et al.* 2002). The efficiency with which an animal utilises energy during growth, is dependent on a number of physiological factors including body composition and the relative proportions of lean tissue mass (LTM) and fat tissue mass (FTM), due to differences in the energy cost of depositing and maintaining these tissues. Protein or lean tissue is continually degraded and resynthesised and thus the rate at which this occurs may contribute to the variation in energy required by an animal for maintenance and growth (Archer, Richardson *et al.* 1999). The objective of this work was to determine the relationship between feed conversion ratio (FCR, kg feed:kg gain) and residual feed intake (RFI) with body composition in rams at six months of age (6mo), and then again in the same rams at thirteen months of age (13mo).

Materials and methods Two experiments were conducted on fifty-two (52) cross-bred rams at 6mo (36kg liveweight) and then at 13mo (64kg liveweight). Whole animal body composition, using dual energy X-ray absorptiometry (DXA) was measured in all rams at the start and the end of each experiment. The individually-housed rams were fed *ad libitum* pellets (12 MJ/kg DM, 16% CP) with dry matter intake (DMI) and liveweights recorded for 49 d (6mo) and for 62 d (13mo). Feed conversion efficiency was calculated as feed eaten:liveweight gain (FCR). To calculate RFI, feed intake was regressed against mean metabolic liveweight ($W^{0.75}$) and average daily gain for each individual animal, with the residual portion, used as the measure of efficiency. The more negative the RFI value, the more efficient the animal is. Restricted maximum likelihood (REML) analysis was used to develop models relating body composition to either FCR or RFI.

Results Mean feed intake, average daily gain (ADG), and efficiency data are given in Table 1. There was no significant difference in growth rate between the two age groups, however both DMI and FCR were significantly different ($P<0.001$). Mean DXA body composition measures for lean tissue mass (LTM), fat tissue mass (FTM) and inorganic mass (ASH) for the start and end of each experiment at 6mo and 13mo is given in Table 2.

Table 1 Mean (± s.d) values for growth, intake and efficiency in rams

Age	ADG (g/d)	DMI (kg DM/d)	FCR	RFI
6mo	407.3± 9.62	1.75±0.285	4.6±0.83	-0.01±1.023
13mo	417.1±55.50	2.36±0.187	5.9±0.63	0.00±1.009

Table 2 Mean (± s.d) start and end DXA body composition measures for LTM, FTM and ASH in rams

	6mo		13mo	
	Start	End	Start	End
LTM (kg)	28.1 ± 3.52	41.3 ± 4.65	49.2 ± 3.36	62.9 ± 4.65
FTM (kg)	3.6 ± 0.76	7.6 ± 1.62	6.48 ± 1.04	16.4 ± 2.78
ASH (kg)	0.8 ± 0.14	1.2 ± 0.18	1.5 ± 0.15	2.26 ± 0.25

At 6mo, the change in LTM was a key driver for differences in FCR (R^2=0.61), however change in FTM was not significant (R^2=0.0). LTM at the start and the end was significant (R^2=0.66) in predicting FCR_{6mo}. The DXA body composition parameters displayed no significant associations with RFI_{6mo}. Similarly, at 13mo, change in LTM was significant for FCR (R^2=0.62) but there were no significant relationships with RFI_{13mo} and composition.

Conclusions LTM is a major determinant of FCR in young sheep at both 6mo and 13mo. Given that FCR is primarily a function of growth, and sheep at this age typically deposit more lean tissue over fat tissue, this result is not surprising. However, the non-significant relationship between RFI and body composition in sheep at the two different ages, suggests that efficiency of energy use is independent of composition and is driven by other physiological functions.

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Phosphorus metabolism in Santa Inês sheep fed four levels of mineral supplementation

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Introduction The isotope dilution technique is the most important way to study the metabolism of minerals. With the use of radiophosphorus (^{32}P), it is possible to describe the kinetics of P including the endogenous fraction. The aim of this work was to study the metabolism of supplemented P using the isotope dilution technique and to apply bio mathematical model to investigate its kinetics.

Material and methods During a 28 day period (21 for adaptation and 7 for collection), 12 Santa Inês male sheep (LW=22.6±2.21 kg) received a basic diet (75% forage + 25% concentrate) supplemented with mineral mixture at maintenance level (except for P). Dicalcium phosphate was added to each treatment (T0: 0; T1: 1.5; T2: 3.0; and T3: 4.5 g P day^{-1} per animal). At day 22, 0.5 ml of ^{32}P solution was intravenously injected in each animal, corresponding to 7.4.MBq per animal. Blood, faeces and urine samples were taken at 24-h intervals. Total P and radioactivity in all the samples were measured. After the end of collection period, tissues samples were taken *post mortem* (liver, heart, kidney, semitendinosus muscle and 12th rib) for analysis. Inputs experimentally measured and model outputs were statistically analyzed as a completely randomized design. Treatment means were compared by Duncan test. The whole body P metabolism model by Vitti *et al*. (2000) (Figure 1) was adopted to represent P flows in sheep.

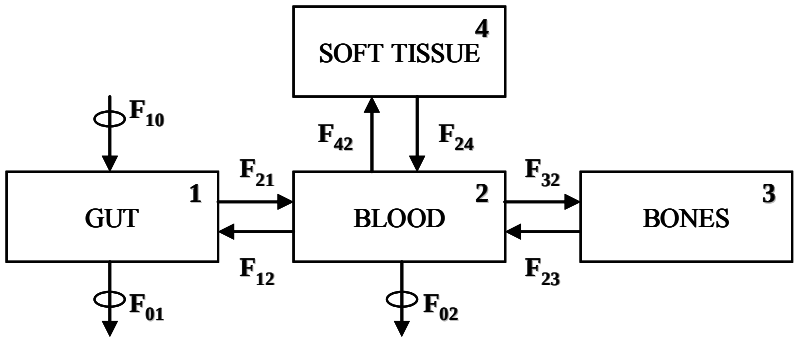

Figure 1. *Schematic model of P metabolism (Vitti et al., 2000)*

Results Results are summarized on Table 1. As expected, total P intake (F_{10}) was affected by increasing levels of P in diets (P<0.001). The total P excreted in faeces (F_{01}) also was affected by treatments (P<0.05) and lower values were observed for animals with no P supplementation. Losses of P in the urine (F_{02}) were similar for all treatments (P>0.05). Also no differences (P>0.05) were observed for fluxes F_{32}, F_{42}, F_{24} and F_{23}. The fluxes F_{21} (P absorption) and F_{12} (from blood to gut) were smaller for animals with no P supplementation (treatment T0) (P<0.05) but there were no differences (P>0.05) between those data for animals with any P supplemented (treatments T1, T2 and T3). Treatments T0 and T1 did not supply enough P to animal metabolism and balance values highlight that animals supplemented with less than 3 g P day^{-1} (T2) were in mineral deficit.

Table 1 Phosphorus metabolism and kinetics for Santa Inês sheep fed mineral supplement with 0, 1.5, 3.0 or 4.5 g P day^{-1} (respectively, treatments T0, T1, T2 and T3)

Parameters (g day^{-1})		Treatments				S.E.	P value
		T0	T1	T2	T3		
Input and outputs							
intake	F_{10}	0.910 d	2.550 c	4.433 b	6.590 a	0.0751	< 0.0001
faeces	F_{01}	1.390 c	2.853 b	4.090 b	5.507 a	0.3823	0.0004
urine	F_{02}	0.003	0.004	0.006	0.005	0.0021	0.7291
Model fluxes							
blood to gut	F_{12}	0.768 b	5.696 a	5.756 a	5.683 a	1.2056	0.0463
P absorption	F_{21}	0.288 b	5.393 a	6.100 a	6.767 a	1.3392	0.0328
blood to bone	F_{32}	0.819	1.625	1.344	2.185	0.4634	0.2848
blood to tissue	F_{42}	0.308	0.499	0.410	0.656	0.1959	0.6519
tissue to blood	F_{24}	0.173	0.252	0.144	0.194	0.0517	0.5360
bone to blood	F_{23}	1.437	2.181	1.273	1.569	0.4246	0.4949
Balance							
gut ↔ blood		- 0.480 b	- 0.303 b	0.343 ab	1.083 a	0.3947	0.0488
blood ↔ bone		- 0.617 b	- 0.556 ab	0.071 a	0.616 a	0.3780	0.1467
blood ↔ tissue		0.135	0.247	0.266	0.462	0.1837	0.6632

a,b means followed by different letters, within rows, are significantly different

Conclusions. By the bio mathematical model results, supplementation of 3 g P day^{-1} (T3) for this breed sheep is enough to meet animal requirements and above this (T4) losses are increased causing pollution.

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Effect of calcium sources on calcium flows in sheep: comparison of two models

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Introduction Calcium is needed for bone formation, muscle contraction, nerve transmission and a variety of other functions in the organism. Experiments has been carried out to study phosphorus metabolism in sheep and cattle, by using isotope dilution techniques however, there is very little information on Ca metabolism in sheep. The objective of this experiment was to study the effects of various Ca sources on the Ca flows in sheep by two models.

Material and methods Twenty Brazilian breed male sheep received during a period of 28 days, a basic diet supplemented with different sources of Ca: limestone (LM), alfalfa hay (AH), dicalcium phosphate (DP), shell meal (SM) and citrus pulp (CP). After 21 d each animal was intravenously injected with 7.4 MBq of radio-calcium (^{45}Ca). Blood samples, feces and urine were taken at 24-h intervals. Total Ca and radioactivity in all the samples were measured. After the end of collection period tissues samples were taken (liver, heart, kidney, muscles and 12th rib) for analysis. Primary data used in this experiment are presented in Roque (2004). Model inputs experimentally measured and model output was statistically analyzed as a completely randomized design. A comparison of means from each category was carried out using the GLMP (SAS, 1991). Treatment means were assessed for significant differences at $P<0.05$. Calcium metabolism was evaluated by model by Vitti *et al*. (2000) and Fernández (1995).

Result Calcium flows calculated using the model of Vitti *et al*. (2000) are shown in Table 1 and flows calculated using the model of Fernández (1995) are shown in Table 2. There were no differences ($P>0.05$) between Ca intake (5.20; 6.20; 6.33; 6.77 and 6.26 g/day, respectively for treatments LM, AH, CP, DF and SM). The total Ca excreted in faeces was affected by treatments ($P<0.05$) and lower values were observed for animals fed limestone (2.88; 6.24; 6.26; 5.20 and 5.03 g/day, respectively for treatments LM, AH, CP, DF and SM). There were no significant differences in losses of Ca in urine between the treatments (0.06; 0.04; 0.12; 0.15 and 0.08 g/day, respectively for treatments LM, AH, CP, DF and SM). Ca balance was negative for AH and CP ($P<0.05$). The presence of pectin in the CP and oxalate in AH could affect the Ca balance. Comparison between the two models showed an agreement for flows between gut and blood in both directions. Total Ca absorption and Ca flow from the central pool to gut were higher for LM ($P<0.05$) for the two models. Ca flow between bone and blood in both directions was higher for Vitti model ($P<0.01$) and Fernandez model showed higher ($P<0.01$) Ca flux from and to blood and tissues. Ca flux from blood to soft tissue and vice-versa was different for the treatments in the Vitti model, but they were different ($P<0.05$) in the Fernández model. Ca recycling from blood to bone were lower ($P<0.05$) for AH and CP for both models.

Table 1 Flows calculated using the model of Vitti *et al.* (2000) for sheep fed different Ca sources

	Calcium sources				
	LM	AH	CP	DF	SM
Gut to blood	5,51^{a}	0,90^{b}	1,5^{b}	2,82^{ab}	2,49^{b}
Blood to gut	3,18^{a}	0,94^{b}	1,49^{ab}	1,25^{b}	1,26^{b}
Blood to bone	1,85^{a}	0,03^{c}	1,14^{c}	1,26^{ab}	0,98^{b}
Bone to blood	0,01^{a}	0,16^{a}	0,24^{a}	0,01^{a}	0,01^{a}
Blood to tissue	0,41^{a}	0,07^{a}	0,06^{a}	0,15^{a}	0,18^{a}
Tissue to blood	0,01^{a}	0,02^{a}	0,03^{a}	0,01^{a}	0,01^{a}

Table 2 Flows (g per day) calculated using the model of Fernández (1995) for sheep fed different Ca sources

	Calcium sources				
	LM	AH	CP	DF	SM
Gut to blood	5,40^{a}	0,85^{b}	1,60^{b}	2,77^{b}	2,46^{b}
Blood to gut	3,08^{a}	0,89^{b}	1,53^{b}	1,20^{b}	1,23^{b}
Blood to bone	0,35^{ab}	0,04^{b}	0,08^{b}	0,43^{a}	0,28^{ab}
Bone to blood	0,01^{a}	0,05^{a}	0,09^{a}	0,01^{a}	0,01^{a}
Blood to ti ssue	1,92^{a}	0,20^{b}	0,13^{b}	0,99^{ab}	0,85^{ab}
Tissue to blood	0,01^{b}	0,26^{a}	0,19^{ab}	0,01^{b}	0,01^{b}

Conclusions The chemical form of the calcium present in the different sources affected calcium metabolism. Inorganic sources had higher Ca absorption. The kinetics model could be used to illustrate the different processes that occur in sheep fed various Ca sources. Although flows in bone and tissue differed the models found similar values for Ca net retention in bone and tissue.

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Effect of fat sources and sodium fumarate supplementation on nitrogen utilization in goats

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Introduction Supplemental fat is commonly incorporate in diet of ruminant as to elevation of energy intake of the animals (Palmquist and Jenkins, 1980). It has been known that added fat was interfered rumen fermentation. Vegetable oils were pronounced in alteration of rumen fermentation than animal fats. Fumarate is a promising substance to reduce methanogenesis and fumarate is an intermediate of propionate production (Asanuma *et al*., 1999). Yuangklang *et al.* (2005) found that added fumarate in tallow diet lowered gas production in vitro study and enhanced fat digestibility in beef cattle fed tallow containing sodium fumarate. Thus, the present experiment was to study the effect of fat source and sodium fumarate supplementation on nitrogen utilization.

Materials and methods Four crossbred goats were used in a 4x4 Latin square experiment with 4 21days periods. Treatments consisted of two sources of fat (tallow and sunflower oil) and two levels of sodium fumarate (NaF) supplementation (0 and 0.5%). Treatments were; A = tallow, B = sunflower oil, C = tallow + 0.5% NaF and D = sunflower oil + 0.5 %NaF. Fresh Nepeir grass was offered ad lib. Concentrate was offered at 1.5%BW. At the last 5 days of each period, total collection was performed to determine nitrogen utilization. Nitrogen in feed, feces and urine were analyzed and calculated for nitrogen utilization according to standard procedure.

Results Fat sources did not influence any parameters. While goats fed added NaF were higher in concentrate and total intake than goats fed non-supplemented NaF ($P<0.05$). N intake, N balance, N retention and N absorption (% of intake) were significantly different ($P<0.05$) by supplemental NaF (Table 1). These finding might be explained that NaF is used as feed additive such as monensin and lasalocid. Ruiz *et al*. (2001) found that addition of monensin decreased fecal nitrogen and increased nitrogen digestibility in dairy cows.

Table 1 Feed intake and nitrogen utilization in goats fed different treatments

	Treatments					P-Value		
Items	**A**	**B**	**C**	**D**	**SEM**	**Fat**	**NaF**	**X**
Intake, gDM/d								
Roughage	498.0	507.0	523.5	540.0	11.1	0.58	0.21	0.87
Concentrate	306.3	331.6	369.8	407.9	11.5	0.19	0.01	0.79
Total	804.3	838.6	893.4	947.9	21.7	0.33	0.04	0.82
N intake, g/d	15.1	15.5	16.3	18.3	0.40	0.16	0.03	0.34
N in feces, g/d	2.99	2.79	3.27	2.12	0.18	0.09	0.61	0.22
N in urine, g/d	0.45	0.54	0.55	0.36	0.11	0.84	0.86	0.53
N balance, g/d	11.6	12.1	12.5	15.8	0.37	0.22	0.01	0.08
N retention, g/d	12.1	12.7	13.0	16.2	0.40	0.41	0.02	0.14
N absorption, % of intake	80.1	81.9	79.9	88.3	0.17	0.44	0.02	0.16

Conclusions The present study showed that sodium fumarate supplementation improved nitrogen utilization in goats fed fresh Nepeir grass.

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The effect of formaldehyde and urea on rumen and blood metabolites of sheep

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Introduction Formaldehyde reduces protein degradability by forming crosslink between protein chains and has antimicrobial properties that may alter the bacterial population and fermentation pattern of silage (Nagel and Broderick, 1992). Urea is man made feed ingredient containing 46 g/Kg nitrogen or 287 g/Kg crud protein equivalents. Ingested urea is degraded to ammonia, and the ruminal bacteria in corporate the ammonia into bacterial protein (Haffman, 1999). The objective of this study was to determine of effect of formaldehyde and urea on rumen and blood metabolites of sheep.

Materials and methods 16 sheep (41.85±4.6 kg) are randomly received one of 4 diets in a completely randomized design. The composing of diet based on NRC (1985), consisting of 430 g/Kg DM of alfalfa, 350 g/Kg DM of corn silage, 100 g/Kg DM of barley grain and 120 g/Kg DM wheat bran containing predicted metabolizable energy 2.98 Mcal/Kg DM and containing crude protein 140 g/Kg DM. The treatments contain CS: treatment untreated corn silage, CSF: CS + 4 g/Kg DM formaldehyde, CSU: CS + 10 g/Kg DM urea, and CSFU: CS + 4 g/Kg DM formaldehyde + 10 g/Kg DM urea. The period of present study was 21 days. The rumen fluid of each treatment was obtained by stomach tube. The effect of treatments was determined on rumen pH, ammonia-N, total volatile fatty acids (VFA) (Stuchbury and Sake, 2001), methylene blue reduction time, the sedimentation and flotation period, glucose and urea of blood. The sedimentation and flotation time was determined using filtration of collected rumen liquor from cheese cloth and then collected in experimental tubes, as the small partial precipitated, while large partial was suspended in surface of liquor and the spending time was recorded (Dirksen and Smith, 1987). The methylene blue reduction time was measured using combining of 20 ml rumen liquor and 0.3 % methylene blue and recording of reduction time (Dirksen and Smith, 1987). The data was analyzed using the GLM procedure of SAS (SAS Institute, 1987).

Results Results are shown in Table 1. Results showed that sedimentation and flotation time, methylene blue reduction time, total VFA, blood glucose and blood urea after feeding had no significant effects between treatments. Ammonia-N in CSU and CSFU was rather than the other treatments ($p<0.05$). Blood glucose after 2 h feeding is lower than of 4 h. Blood urea in 4h in CSU and CSFU was higher than the other treatments. The higher blood urea in CSU and CSFU can be resulted from high ruminal ammonia-N concentration of the mentioned treatments.

Table 1 Effect of formaldehyde and urea on rumen and blood metabolites*

Item	CS	CSF	CSU	CSFU	SEM
pH	6.19^{b}	6.4^{ab}	6.51^{a}	6.185^{b}	0.16
Sedimentation and flotation time (sec)	361.25	387.5	369.0	370.0	21.81
Methylene blue reduction time (sec)	188.75	201.25	175.0	186.25	18.49
Total VFA mM/L	101.0	100.75	111.5	107.0	11.2
Ammonia-N mg/L	127.75^{ab}	91.0^{b}	136.5^{a}	136.5^{a}	24.3
Blood Glucose 2h	52.0	57.0	52.0	51.0	4.76
Blood Glucose 4h	58.5	59.0	61.25	64.75	3.16
Blood urea 2h	17.5	18.0	20.25	21.0	2.15
Blood urea 4h	17.5^{ab}	14.0^{b}	19.5^{a}	19.5^{a}	2.7

* The means within a row without common letter differ ($p<0.05$)

Conclusions Using urea treatment in silage increased ammonia-N of rumen fluid and blood urea resulting high ruminal nitrogen source for microbial protein synthesis. Formaldehyde and urea in sheep diet had no negative effect on rumen metabolites.

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Effect of supplemental feed in early lactation of Arabi ewes on wool and lamb growth

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Introduction The growth rate of Arabi lamb during the suckling period (from birth to weaning at about day 100) is very slow particularly for lambs born in the autumn (Dabiri, 1999). Poor quality and limited quantity of feed for lactating ewes in free grazing system was shown to limit milk production, which was the main reason for slow lamb growth (Dabiri and Mosavi 2000). On the other hand, in conventional Iranian sheep production systems, offering a limited quantity of low quality supplemental (Sup) feed (or even no supplemental feed) to the lactating ewes was an important factor for severe reduction of the growth rate of the wool of lactating ewes during the autumn season (Dabiri, 2002). It was hypothesized that offering a high quality supplemental feed to ewes for a short period during early lactation would improve the growth rate of lambs and may alleviate the poor wool growth rate of lactating ewes during the autumn season. Therefore, the aims of this experiment were to 1) compare the growth of lambs suckled from experimental supplemented ewes with the growth of lambs suckled from conventionally ewes, and 2) compare the wool growth of lactating (wet) ewes with non-lactating (dry) ewes during autumn season

Materials and methods Thirty six ewes with similar conditions from a flock of autumn lambing of Arabi sheep of the Ramin Agricultural complex of Shahid Chamran University were included in this experiment. The ewes were divided into 4 groups of 9 sheep (with 3 groups of ewes were wet (pregnant/lactating) and 1 group were dry). The wet ewes allocated into 3 different levels of supplemental feed (0 (as conventionally similar to dry ewes), 350 and 700 grams/ ewe) for a period of one month after lambing in a completely randomized design using a GLM. The diets were formulated according to NRC (1985) and had similar quality for protein (160 g/Kg DM CP), energy (790 g TDN/Kg DM) and high concentrations of other nutrients. Ewe liveweight and body condition score were measured at lambing and 2 weeks interval after that until weaning. Lamb liveweight were measured at birth and 2 weeks interval until weaning. Wool growth rate measured for a period of 14 weeks (2 weeks before lambing until weaning at week 12 after lambing). Wool growth rate determined from the weight of wool clipped from the mid side (mg wool/cm^2/d).

Results Despite the similar live weight and body condition score of 4 ewe groups at lambing time, the dry ewes were heavier ($P<0.05$) than wet ewes during whole period of 2 weeks interval from lambing to weaning . The same trend was found for body condition score. Live weight and body condition score of 3 groups of wet ewes were not affected by levels of supplemental feed. At weaning (weeks 12 of lactation) the dry ewes had about 10 Kg greater live weight than comparative wet ewes (52 VS 42-43.6 ±1.64). The body condition score had a similar trend with ewe live weight (4.6 VS 1.9 – 2.2 ± 0.12). With the exception of those wet ewes offered 700 g/day supplemental feed, the 2 other wet groups of ewes had significantly ($P<0.05$) slower wool growth rate than dry ewes during a 14 period weeks (from 2 weeks before lambing until weaning). The growth rate (g/d) of lambs reared by 2 offered supplemental feed group ewes were greater ($P<0.05$) than lambs reared by conventionally (165 ± 9.1 g/d) ewes group during the whole period (10 weeks), but this superiority was accord during the first 6 weeks age of lambs not later 4 weeks. The differences of lamb growth rate between ewes offered 350 (198 ± 9.1g/d) or 700 (204 ± 9.1 g/d) g/d supplemental feed were not significant ($P>0.05$).

Table 1 Effects of different levels of supplemental feed in early lactation of ewes

Treatments Traits	Dry ewes	wet ewes Non-Sup	wet ewes 350g Sup	wet ewes 700g Sup
Ewe liveweight (kg)				
2 weeks pre-lambing	45.5^{b}±2.11	55.9^{a}±2.11	56.1^{a}±2.11	53^{ab}±2.11
Lambing time	50.0^{a}±2.09	50.8^{a}±2.09	51.5^{a}±2.09	48.0^{a}±2.09
4 weeks post-lambing	53.0^{a}±1.98	47.2^{b}±1.98	47.1^{b}±1.98	48.0^{ab}±1.98
12 weeks post-lambing	52.0^{a}±1.64	43.1^{b}±1.64	43.6^{b}±1.64	42.0^{b}±1.64
Lamb growth rate (g/d)	----	165^{b}±9.1	198^{a}±9.1	204^{a}±9.1
Wool growth rate (mg wool/cm^2/d)	0.65^{a}±0.45	0.44^{b}±0.045	0.389^{b}±0.045	0.514^{ab}±0.045

Conclusions These results are shown that by offering 350 g/d supplemental feed can improve only lamb growth rate, but using 700 g/d supplemental feed can improve both lamb and wool growth rate. Thus, it is recommended that, for similar conventional system of Iranian sheep production, like this experiment a high quality supplemental feed at least 350 g/d should be offered to the early lactation period of the autumn lambing of ewes.

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In vitro effect of condensed tannin extract from *Acacia molissima* on gastrointestinal nematodes of ovine

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Introduction Although some argument is still ongoing about the resistance to different anthelmintics, there is a general agreement that reversion to susceptibility is rare once drug resistance (DR) has developed in livestock helminths, even when other drugs with completely different working mechanisms are used for prolonged periods. Some *in vitro* methods have been used to investigate the efficacy of towards nematode parasite: larval development (LD), larval migration inhibition (LMI), larval feeding inhibition (LFI) and egg hatch (EH) assays (Amarante *et al*., 1997; Coles *et al.,* 1988). The aim of this work was to determine the potential inhibitory effect of condensed tannins or other bioactive compounds from *Acacia molissima* extract on the feeding of first stage larvae of *Haemonchus contortus*, *Trichostrongylus vitrinus* and *Teladorsagia circumcincta*, using the LFI test.

Material and methods A commercially condensed tannin extract from Brazil (CTE), made from bark of *Acacia molissima,* was used as the condensed tannin source. Several dilutions were made with this extract from 0.02 to 2.50 mg CTE/ml of distilled water and a blank control (distilled water) was used in each assay (in duplicate). The effect of polyethylene glycol (PEG) on the anthelmintic activity of the CTE was also tested by adding 10 µl of PEG (10 mg/ml) per flask with the CTE in all concentrations. The addition of PEG was supposed to enable the determination of the anthelmintic activity and if it was attributable to condensed tannins or other polyphenols. The study was carried out using anthelmintic susceptible ovine isolates of *Haemonchus contortus*, *Trichostrongylus vitrinus* and *Teladorsagia circumcincta*. All strains are maintained at Moredun Research Institute, Scotland, UK. For LFI assay, the first stage larvae were incubated for 2 h with CTE, after this time the labeled *E. coli* (fluorescein isothiocyanate-SIGMA) were addicted to all flasks and incubated for 18 h. After the incubation the percentages of larval feeding were determined at each concentration and the LD_{50} values were calculated, after logarithmic transformation of dilution values. The statistical design was a complete factorial 3x8x2 (three nematode species, eight dilutions of CTE and two treatments - with and without PEG).

Results The CTE had affected severely the feeding ability of first stage larvae (L1) of all tested species (Table 1). There were significant differences ($P < 0.01$) between control (distilled water) and the CTE in concentrations from 0.04 to 1.25 mg CTE/ml. The LD_{50} (mg CTE/ml) for *H. contortus*, *T. vitrinus* and *T. circumcincta* were 0.043; 0.038 and 0.050 (SE: 0.0024), respectively. The addition of PEG increased the LD_{50} ($P < 0.01$), to 0.096; 0.100 and 0.250, respectively for *H. contortus*, *T. vitrinus* and *T. circumcincta*, showing the effect of CT.

Table 1 Means of viability of nematode larvae based on the feeding ability of L1 larvae of *Haemonchus contortus*, *Trichostrongylus vitrinus* e *Teladorsagia circumcincta* using condensed tannin extract (CTE) as anthelmintic.

Dilutions mg/ml	*Haemonchus contortus*		*Trichostrongylus vitrinus*		*Teladorsagia circumcincta*	
	CTE	CTE + PEG	CTE	CTE + PEG	CTE	CTE + PEG
Control *	0.73 (0.71; 0.75)	0.75 (0.71; 0.79)	0.75 (0.75; 0.75)	0.75 (0.73; 0.77)	0.74 (0.73; 0.75)	0.75 (0.72; 0.78)
0.625	0.00	0.30	0.00	0.00	0.00	0.20
0.31	0.00	0.60	0.00	0.30	0.00	0.38
0.155	0.00	0.80	0.00	0.20	0.20	0.44
0.08	0.11	0.38	0.04	0.51	0.10	0.71
0.04	0.42	0.60	0.26	0.65	0.40	0.71
0.02	0.71	0.71	0.65	0.74	0.71	0.72

* mean (minimum; maximum)
PEG: polyethylene glycol

Conclusions CTE showed high anthelmintic activity. This effect was demonstrated via inhibition of the feeding of immature nematodes, but the rumen concentration of CTE was not determined, and this result would be necessary to relate in vitro and *in vivo* studies. In future studies, it can be suggested to use rumen material of sheep drenched with CTE, in order to know the real activity the CTE in gastrointestinal tract.

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Effects of low concentrations of aflatoxin B1 in diet on performance, blood enzymes and organs weight in broiler chickens

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Introduction Aflatoxins (AF) are secondary fungal metabolites mainly produced by *Aspergillus flavus* and *Aspergillus parasiticus*. Aflatoxin contaminated feedstuffs (ACF) are a major problem in developing countries, where that most of poultry feedstuffs contain low levels of different kinds of AF; particularly due to inappropriate conditions of transportation and storage. This could cause silent economical losses in poultry production. The number of studies who have investigated the effects of ACF in broiler nutrition is much; however, little of them focused on low concentrations of AF. Moreover, it has been suggested that negative effects of AF on broiler performance are both dose- and time- dependent (Leeson *et al*., 1995). In this study, the effects of long-term low level administration of AFB1 into diet (0 to 28 days of age), on performance, blood enzymes and some organs weight of male broiler chickens have been investigated.

Materials and methods One hundred and twelve day-old male broiler chicks were divided into 16 groups and allotted to four dietary treatments (levels of 0.0, 0.4, 0.8, and 1.2 ppm AFB1 in starter diet for 28 days) in a randomized complete-block design. AFB1 required for the experiment was produced using *Aspergillus parasiticus* (Shotwell *et al*., 1966) and its concentration was measured by HPLC (AOAC, 2000). Feed intake and body weight gain were recorded weekly. On days 7, 14, 21, and 28, one bird from each replicate was weighed and humanly killed to collect blood samples and to weigh organs. Concentrations of serum enzymes were measured based on routine laboratory methods (Darman Kave Res. Lab., Isfahan, Iran, 2001). This research was preaproved by the animal care and use committee of Excellence Center in Animal Science, Ferdowsi University of Mashhad. Data were analyzed by analysis of variance and comparisons between treatment means were performed using Duncan's Multiple Range Test (SAS Inst., 2001).

Results Feeding AFB1 at 1.2 ppm significantly ($p<0.05$) decreased feed intake and body weight gain and increased relative weights of liver and brain at 28 days of age (table 1). On day 21, serum concentrations of alanine amino transferase (ALT) and aspartate amino transferase (AST) significantly increased ($p<0.05$) at 1.2 ppm AFB1 (table 1). Serum concentration of lactate dehydrogenase (LDH) showed a significant reduction ($p<0.05$) by 1.2 ppm AFB1 on day 28 (Table 1).

Table 1 Relative organs weight, performance and Serum enzymes concentrations of broiler chickens fed diets containing different levels of AFB1

AFB1[1]	28 d		0-28 d			ALT(U/l)		AST (U/l)		LDH (U/l)	
ppm	Liver (%)	Brain (%)	FI (g)	BWG (g)	F/G	7 d	21d	7 d	21 d	7 d	28 d
0.0	2.54^{b}	0.25^{b}	1564^{a}	814^{ab}	1.92^{a}	63	57^{b}	162	142^{b}	1770^{a}	1328^{a}
0.4	2.54^{b}	0.28^{ab}	1510^{a}	834^{a}	1.84^{a}	65	59^{ab}	151	145^{ab}	1647^{ab}	986^{b}
0.8	3.20^{a}	0.29^{ab}	1543^{a}	787^{ab}	1.96^{a}	68	61^{ab}	159	159^{ab}	1321^{ab}	1241^{ab}
1.2	3.65^{a}	0.30^{a}	1323^{b}	680^{b}	1.95^{a}	66	64^{a}	169	179^{a}	1142^{b}	949^{b}
±SEM	0.172	0.012	48.9	44.6	0.056	3.0	1.6	8.7	10.7	161.4	99.1

In each column, means with different superscripts are significantly different ($p<0.05$)
[1]AFB1, aflatoxin B1; d, days of age; U/l, units per litre; FI, feed intake; BWG, body weight gain; F/G, feed to gain ratio; ALT, alanine amino transferase; AST, aspartate amino transferase; LDH, lactate dehydrogenase

Conclusions In this study, administration of AFB1 into starter diet at the level of more than 1 ppm, adversely influenced performance of broilers and caused significant increase in liver and brain relative weights. Increase in serum concentrations of AST and ALT in this experiment, may be indicative of injury to hepatocytes. This is inconsistent with data reported by Tedesco *et al*., (2004). Reduction in serum concentration of LDH, also has been reported by Huff *et al*., (1986). Generally this study showed that, along side other negative effects, AFB1 could also have adverse effect on broilers' brain. However, liver is the first organ that is affected by AFB1.

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The effect of xylanase enzyme and fat type on growth performance of broilers fed wheat-based diets

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Introduction Wheat grain is a major ingredient in broiler diets in many countries, and due to its high protein and starch content, it is considered to be a good source of nutrients for broilers. But wheat contains 5 to 8 percent non starch polysaccharides (NSP), pentosans, which may be impair nutrient digestibility and growth performance (Carre and Brillouet, 1986). Several studies have shown that, among the nutrients, fat digestion suffers the most pronounced impairment due to high digesta viscosity (Choct and Annison, 1992). It is suggested that to reduce adverse effects of anti nutritional factors content in wheat, diet supplements with enzyme. The effect of dietary supplementation of xylanse enzyme on growth performance is depended upon the amount and fat type (Langhout, *et al.* 1997). Different fat sources are usually added to broiler diets to increase energy concentration and to improve productivity. This study was carried out to determine effect of xylanase enzyme and fat type on growth performance and intestinal viscosity of broiler fed wheat based diet.

Materials and methods An experiment was carried out using 384 seven old day Arian 110 chickens in a 4×2 factorial arrangement based on randomized complete design with 5% different fat sources [Beef tallow, soybean oil, blend fat [soybean oil (37.5 gr/kg diet) + Beef tallow (12.5gr/kg diet)] or other blend fat [soybean oil (12.5 gr/kg diet) + Beef tallow (37.5gr/kg diet)], which were termed T, S, ST1 and ST2, respectively and Nutrex® enzyme addition [none or enzyme (0.5 gr/kg diet)]] to a wheat (60%) based broiler diet. All of the diets were isocaloric and isonitrogenous with considering of the difference between fat sources and enzyme supplementation (NRC, 1994). At 42 d of age, one bird per replicate was killed and carcass of them was analyzed. Body weight (BW), feed intake (FI) and coefficient feed ratio (FCR) were recorded weekly with pen as the experiment unit. Obtained data were analyzed by the GLM procedure of SAS (2001) program.

Results The results are shown in Table 1. Results showed that enzyme supplementation improved body weight, body weight gain and feed/gain ratio of broiler fed diet contains enzyme ($p<0.01$). Where as the chickens fed diet no supplemented with enzyme have lower feed intake but showed not significant different. Although no significantly different was found between performance of broilers fed different fat source, however performance of broiler fed soybean oil was better than the other groups. Except of feed intake no interaction was observed between fat type and enzyme supplementation. In addition, supplementation of diets with enzyme reduced the intestinal viscosity of broilers.

Table1 Effects of treatments on performance of broiler chickens

Effects	BW (kg)	BW gain(kg)	FI (kg)	FCR	Weight of carcass (kg)	Thigh muscle(g)	Breast muscle(g)	Viscos-ity (cp)
Enzyme								
+E	2.26[a]	1.99	3.37	1.68[b]	1.537	450[a]	474	4.766[b]
-E	2.12[b]	1.8[b]	3.28	1.82[a]	1.473[b]	395[b]	444	6.653[a]
s.e.m.	0.007	0.013	0.016	0.006	0.016	9.22	7.96	0.304
Fat type								
S	2.223	1.928	3.31	1.72	1.537	443	486.6	5.46
ST1	2.172	1.875	3.27	1.75	1.511	431	452.6	5.37
ST2	2.199	1.91	3.33	1.74	1.477	417	450	5.47
T	2.18	1.88	3.38	1.8	1.495	398.1	447.3	6.52
s.e.m	0.007	0.013	0.016	0.006	0.016	9.22	7.96	0.304

a, b Values in the same column with no common letter differ ($P<0.05$).

Conclusions Using xylanase enzyme in broiler improved growth performance and weight of carcass, it is evident from the present studies that the addition of xylanase could eliminate negative effect of non starch polysaccharide content in wheat grain however it's application with different fat sources required further research.

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The effects of different fat sources on the growth performance, blood metabolites and abdominal fat of broiler chickens

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Introduction The most practical method for increasing the energy concentration in poultry diets has been by the addition of fats and oils (Hill and Dansky, 1954). Now a days use of fats because of increasing of the energy, improving growth and physical quality of diet have special importance. Fats rich in unsaturated fatty acids are better digested and absorbed than saturated fats (Danicke 2001). Studies with rats and broilers have reported that unsaturated vegetable oils produce lower fecal energy losses and, consequently higher metabolism energy that animal fats (Zollitsch *et al*, 1997). This higher metabolism energy of unsaturated fat could be expected to cause higher fat deposition because the additional energy could be stored as triglycerides (TG) in adipose tissue of fat depots. Opposite of this results Crespo and Steve (2002) observed that among broilers fed diets with added fat (tallow, olive, sunflower, and linseed), linseed oil had less abdominal fat (AF) than those fed tallow. The objective of this study was determination of effects of different fat sources on performance, blood metabolites and abdominal fat of broiler chickens.

Materials and methods An experiment was conducted to investigate the effect of different fat sources on the performance, blood metabolites (BM) and abdominal fat of broiler chicks at 7-42 days of age. One hundred ninety two day-old broiler chicks (Arian 110) were used in a completely randomized design in 4 treatments with 3 replicates of 16 birds each. Experimental treatments were include basal diet addition tallow, soybean oil, blend fat [37.5 gr/kg diet soybean oil + 12.5gr/kg diet Beef tallow] and other blend fat [12.5 gr/kg diet soybean oil1 + 37.5 gr/kg diet Beef tallow], which were termed T, S, ST1 and ST2, respectively. All of the soybean meal-wheat based diets were isocaloric and isonitrogenous with considering of difference between fat sources (NRC, 1994). At the end of 6 weeks of experiment, one bird of every experimental unit was selected and before killing was weighted and that's blood was collected to analysis. Data were analyzed by the GLM procedure of SAS (2000) program. Significant differences among treatment means were measured by Duncan's new multiple range test with a 5% level of probability.

Results The different fat sources had no significant effects on feed intake, body weight, feed/gain ratio, weight gain (WG) weekly, carcass efficiency and AF at the end of the experimental period. Replacement of soybean oil with tallow fat had significant effects on triglycerides of blood's broiler and it decreased triglycerides of that broilers ($P<0.01$), probably because of high clearance of TG in soybean oil treatment. Also tallow treatment was increased the amount of blood's glucose ($P<0.05$) compared soybean oil. Tallow fat may because the changing fatty acids profile of cell membrane so confusing insulin action and finally increase glucose serum of broiler. Bloods cholesterols (Ch) and AF not affected by experimental treatments.

Table1. Effects of treatments on performance and blood metabolites #

Measurements	*Unite*	S	ST1	ST2	T	*s.e.m*	*P*
Feed Intake	gr	3227	3356	3301	3244	0.0436	0.77
Body Weight	gr	2184	2084	2136	2097	0.008	0.56
weight Gain	gr	1826	1785	1850	1753	0.038	0.85
FCR	gr/ gr	1.79	1.89	1.78	1.85	0.027	0.36
Carcass Weight	kg	1.52	1.48	1.37	1.5	0.024	0.09
Breast Weight	gr	480	433	425	436	11.25	0.34
Thigh Weight	gr	435	408	365	368	11.95	0.08
Triglycerides	mg/dl	76.6^{b}	78^{b}	80^{b}	125^{a}	7.007	0.007
Cholesterol	mg/dl	123.3	124.7	126	129	4.01	0.97
Glucose	mg/dl	232.3^{b}	230^{b}	237^{b}	269^{a}	5.99	0.041
Abdominal Fat	gr	46.6	38.67	39.68	30	2.44	0.17

#a, b means within a row without a common letter differ ($P<0.05$).

Conclusions The result of this experiment indicated that energy of used soybean oil almost is equal with tallow fat in practical condition and tallow fat probably changes the abdominal fat with effect on blood TG and glucose.

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Effect of Virginiamycin and *Saccharomyces cerevisiae* on broiler performance and carcass yield

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Introduction Feed-borne antibiotics have been used since early of 1950's in feed as a first primary to control of disease and more recently as a growth promoter and improve feed conversion efficiency in the poultry industry. Two of the more popular feed grade, spectrum antibiotics utilized within the poultry industry includes Virginiamycin (VIR) and Bambermycin (BAM) (Parks *et al,* 2001). Studies with broiler were indicated that probiotic bacteria are an alternative to antibiotics to be used extensively as a growth promoter in form animals (Green and Sainsbury, 2001). The aim of this study was to determine the effects of antibiotic (Virginiamycin) and probiotic (*Saccharomyces cerevisiae*) and their interaction on broiler performance and carcass yield.

Materials and methods In this study totally on hundred eighty-one day-old of commercial broiler strain (Arian 110) chickens were obtained from local hatchery. The chicks were randomly assigned on each of four treatments Experimental diets were four dietary treatments with three replicates (each replicate contained fifteen birds allocate to each experiment unit). Experimental design was based on completely randomized design (CRD). The treatments were four diets 1- control feed without feed additive (C) 2- control feed supplemented by 250 g/ton Virginiamycin (VIR) 3-control fed supplemented by 0.1% *Saccharomyces cerevisiae* (Sc) 4- control fed supplemented by combination of 250g/ton Virginiamycin plus 0.1% *Saccharomyces cerevisiae* (VIR+Sc). Formulated diets for each period were based on broiler nutrient requirement (NRC 1994). Feed and water were offered *ad libitum* during experiment and lighting was containing continuous through the experimental period (42d). Live weight, feed intake were recorded weekly and then feed conversion ratio (g feed/ g gain, FCR) were calculated. At the end of 6 weeks two birds in each replicate were slaughtered to measure carcass yield (% of live weight), abdominal fat (% of carcass yield) and liver weight. Data were analyzed by using the GLM procedure of SAS (1996).

Results Growth performance and body composition of broiler chicks fed a various growth promoters are shown in Table 1. As in shown in this table the Analysis of variance revealed that body weight, feed intake, FCR, carcass yield (% of cold carcass/Liveweight) and abdominal fat weight were significantly ($P<0.05$) affected by Virginiamycin, *Saccharomyces Cerevisiae* and their combination (VIR+Sc). However carcass efficiency and liver weight were not affected by treatment groups ($P>0.05$). The result obtained in the experiment showed that the broiler chicks which received 250 g/ton VIR + 0.1% SC was exhibited higher body weight, feed intake and carcass yield and lower abdominal fat weight than the control and other groups. In this experiment, however FCR was not significant affected by growth promoters.

Table 1 Effect of various growth promoter (VIR, SC and VIR + SC) on broiler performance and carcass yield from 7to 42 days of age[1]

Variable	BWG (g) (7-42 days)	Feed Intake (g) (7-42 days)	FCR	Carcass yield (% Of live weight)	Liver (g)	Abdominal fat (% of carcass)
C	2068^{c}	4041^{b}	1.89^{a}	67.16^{b}	57.33^{a}	2.15^{b}
VIR	2260^{a}	4145^{ab}	1.81^{a}	71.62^{a}	55.89^{a}	2.22^{b}
Sc	2169^{b}	4203^{a}	1.92^{a}	69.09^{ab}	56.67^{a}	2.76^{a}
VIR+Sc	2308^{a}	4278^{a}	1.88^{a}	71.71^{a}	51.67^{a}	2.01^{b}
SE	47.03	73.52	0.057	1.67	3.39	0.26

[1]Different superscripts within the same column are significantly different ($P<0.05$).

Conclusion the results presented here demonstrated that supplementation of broiler diets with probiotic (SC) or in combination is a effective management tool to aid in improving the production efficiency, carcass yield and composition of broiler chickens.

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Effect of wheat source and wet feeding on AME, protein and phosphorus digestibility of wheat-based diets for broiler chicks

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Introduction The literature and recent research (Scott 2002) demonstrated that wet feeding may overcome limitation in feed intake of wheat-based diets, thereby increasing growth rate. However, there have been inconsistent effects on feed conversion ratio (FCR). Thus it is important to differentiate whether differences in feed efficiency can be explained by monitoring nutrient retention. This experiment was conducted to study the effect of two types of wheat ((Durum, HRS (Hard Red Spring)) on broiler performance and nutrient digestibility, when diet were offered in a wet or dry form.

Materials and methods Day-old male broiler chicks (96 Hubbard × Hubbard) were fed one of 4 dietary treatments from 1 to 17 d. The diets were based on two wheat sources ((Durum, HRS (Hard Red Spring)) and two form of feeding (wet/dry).The two wheat sources were included at 80% of a standard bioassay diet (with enzyme) (Table 1). The diet were fed as is or with 1.2 g water per g feed. The wet diets were prepared daily and all diets were fed to four groups of six male broilers from 1 to 17 d of age. The measurements of body weight and feed intake were determined to 17 d of age and the feed conversion ratio was determined after correcting for mortality. Excreta was collected for eight-h from each cage at 16-d of age. The excreta and respective diets were ground and analyzed for acid insoluble ash and gross energy to determine AME level of diets. Phosphorus concentration in feed and excreta were determined by colorimetry. The protein content (N×6.25) of diets and individual samples of excreta was determined by the combustion nitrogen analyzer (Leco). Data were analyzed with the fixed main effects of wheat source and feed form and all the two-way interactions, using the ANOVA procedure of the SAS statistical package, and Duncan multiple range test was used to identify significant differences between mean values of the treatment effects.

Results Body weight was not affected by wheat type. The broilers fed HRS-based diets consumed more feed than broilers fed Durum-based diets. The FCR for HRS-based diets (1.25) was greater than for Durum-based diets (1.19) Mixing wheat-based diets with water was shown to be advantageous with respect to broiler growth and feed intake but it produced no change in feed conversion efficiency (Table 1). With respect to wheat type, the protein (54%) and phosphorus (76%) digestibility and AME (3560kcal/kg) of Durum wheat was significantly higher than HRS based diets (46 and 68%, and 3280, respectively). There were no differences in AME and protein digestibility of wet or dry diets (3410 vs. 3430 kcal/kg and 71 vs. 72%, respectively). In the present experiment, the reduction in phosphorus digestibility was caused by wet feeding as compared to dry feeding (52 vs.57%, respectively). There were no significant interactions for in protein, phosphorus digestibility and AME between wheat sources and wet feeding.

Table 1. The effect of wheat type (Durum vs Hard Red Spring (HRS),and feed form (wet vs dry) on 17 d body weight, feed intake, and feed conversion ratio

	Body weight 17 d (g)			Feed Intake 0 to 17 d (g b^{-1} d^{-1})			Feed Conversion Ratio (g:g)		
Wheat Type	Total	Durum	HRS	Total	Durum	HRS	Total	Durum	HRS
Total	507	507	508	38.4	37.5*b*	39.2*a*	1.22	1.19*b*	1.25*a*
Feed form									
Wet	543*a*	538	548	40.9*a*	39.9	41.8	1.21	1.18	1.24
Dry	472*b*	476	469	35.8*b*	35.2	36.5	1.23	1.21	1.25

Conclusions In conclusion, The broiler performance, AME, protein and phosphorus digestibility for Durum was higher than for HRS. Therefore, there were significant differences in nutritional value of different sources of wheat-based diet. The improvement in growth rate of broilers fed the wet diets can be explained by an increase of feed intake rather than changes in ME availability and nutrient digestibility. However, the shorter time that the digesta spends in the gastrointestinal tract provides less opportunity for digestion and absorption, which may explain the absence of any effect on nutrient retention.

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Use of broken rice in broiler diet

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Introduction Maize is the principal energy source in most poultry diet in developing countries. As the cultivating of maize is limited in Iran, annually a large amount of maize is imported. Broken rice (BR) is a major cereal by-product for animal feeding in rice-growing countries. The annual production of rice paddy in Iran is about 2 million tonnes. After milling of paddy, broken rice is available for utilization in animal diet. BR consists of 40 to 80 g/kg of rice paddy. It contains good sources of protein (100-110 g/kg), fat (60-110 g/kg), carbohydrate, fibre (20-50 g/kg). Due to mixing with bran and hull the chemical composition of BR varies in different samples. In some part of the country the price of this ingredient is lower than maize. The objective of the present experiment was to evaluate the effect diets containing different levels of BR on performance of broiler chicks from 1 to 8 weeks of age.

Materials and methods After determining the chemical composition of BR (Table 1) six diets were formulated with 0.00, 100, 200, 300, 400, and 500 kg/t of BR by using of UFFDA software. These diets were isocaloric and isonitrogenous. The main ingredients in control diet (1) were about maize (600-700 kg/t), soybean meal (200-300kg/t) and fish meal (30-60 kg/t). In the other diets (2, 3, 4, 5, 6), 100, 200, 300, 400, 500 kg/t BR were replaced instead of maize respectively. Feed and water were offered *ad libitum*. In this experiment chicks were reared on floor pens and 40 chicks were placed in each pen. During the experiment, feed intake, body weight gains, feed conversion ratio (FCR) were measured weekly. Mortality was measured throughout the experiment. Data from this experiment were subjected to one-way analysis of variance by using Mstatc software (Mstat directory, 1990). Means differences were separated by Duncan's range test. The level of significance was set at $P<0.05$. There were 3 replicates of 40 chicks in each treatment.

Results Data are given in Table 2. Use of BR up to 500 kg/t of broiler diets had not statistically significant effects on feed intake, body weight gain, and mortality in whole period (1-8 weeks) of the experiment, but improved FCR ($P<0.05$). Diets 5 and 6 (400 and 500kg/t BR) had the best FCR. These data are in agreement with findings of other studies ((Armas and Chicco, 1970), Smith (1984), Treat and Stephenson (1959)).

Table 1 Chemical composition of BR

Nutrients	g/kg
DM	917.2
CP	106.9
EE	69.7
CF	24.5
ASH	99.1
NFE	699.8
P	4.2

Table 2 Effect of diets on performance of chicks

Traits / Diets	Feed intake (g)	Body weight gain (g)	FCR	Mortality
1	4955	2160	2.30[a]	3.33
2	4789	2180	2.19[b]	1.67
3	4651	2160	2.16[bc]	2.50
4	4721	2210	2.14[bc]	1.67
5	4734	2230	2.12[c]	2.50
6	4705	2220	2.12[c]	1.67
SEM	76.90	31.86	0.001	-

Conclusion The results of the present study indicated that use of BR up to 500 kg/t of broiler diets had not adverse effect on performance. Due to lower price of BR, inclusion of this feed in broiler diets decreases the cost of production. Furthermore use of BR improved FCR in comparison with control diet.

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Slaughter results and chemical composition of various muscles of Holstein-Friesian bulls at different ages

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Introduction Holstein-Friesian is dominant cattle breed in Hungary, so it plays an important role in beef production. That's why it is important to know more about slaughter results and meat quality of this breed. As only little information is available in this topic Szabó *et al.* (2002) and Lengyel *et al.* (2003), the purpose of the study was to investigate some slaughter parameters and composition of various muscles of young bulls slaughtered at different ages.

Materials and methods For the study thirty half sib Holstein-Friesian bulls were fattened from 2 months of age on the same diet consisting of maize silage, beet pulp, alfalfa hay and concentrate mixture. According to the requirement 30-40% of daily dry matter ration was concentrate consisting of yellow corn, sunflower seed meal solvent extracted (36%CP), soybean meal (46% CP), mineral and vitamin premix. Ten of the animals were slaughtered approximately at 7 months of age, ten at 14 months, ten bulls at 19 months of age. Carcass composition was measured by dissecting the left site of the carcass. Muscle samples were excised from *psoas major*, *longissimus dorsi* and *semitendinosus* muscle. Chemical analyses was done using Kjeldahl, Soxhlet methods, fatty acids were determined according to Husvéth, Karsai and Gaál (1982), using an automated gas liquid chromatograph. Statistical analyses were performed using SPSS 8.0 for Windows.

Results Average daily weight gain and carcass weight of the 7, 14 and 19 month age slaughtered groups was 1042±14.7g, 830±4.3g, 746±10.8g and 117.3±20.6kg, 202.4±6.6kg, 245.3±11.6kg, respectively. Carcass percentage (killing out %) (50.91±1.35%, 54.57±0.76%, 5746±1.43%), and separable lean content (67.4±2.0%, 70.1±2.4%, 72.5±1.7%) significantly increased, that of bone (25.5±1.1%, 21.4±1, 5%, 19.0±0.8%). significantly decreased by age. Fat content of the calves slaughtered at 14 months of age shoved significantly higher values comparing with calves slaughtered at 7 months of age. However, no significant differences were obtained between the fat content in the carcass of bulls slaughtered at 14 or 17 month of ages. Age had an only a slight influence on protein and lipid content of the muscles tested. Some fatty acid components of different muscles are summarized in Table 1.

Table 1 Fatty acid content of the muscles tested

Items/Slaughter age groups	7 months	14 months	19 months
Muscle semitendinosus			
saturated fatty acids,%[1]	39.2±0.8 [a]	42.1±0.4 [a]	42.0±0.5
monounsaturated fatty acids,% [1]	26.1±0.9	26.5±0.9	26.5±1.1
polyunsaturated fatty acids, %[1]	32.1±1.4 [a]	29.4±1.1 [a]	29.5±1.1
Muscle *longissimus dorsi*			
saturated fatty acids,% [1]	41.5±0.6 [a]	45.2±0.6 [a]	47.2±0.8 [a]
monounsaturated fatty acids,% [1]	30.8±1.3 [a]	34.5±1.1 [a]	37.2±1.5 [a]
polyunsaturated fatty acids, %[1]	25.5±1.6 [a]	18.4±1.3 [a]	13.6±1.8 [a]
Muscle psoas major			
saturated fatty acids,% [1]	39.5±0.6 [a]	47.2±1.3	47.9±0.9 [a]
monounsaturated fatty acids,% [1]	30.2±1.3 [a]	33.1±1.1	33.5±1.5 [a]
polyunsaturated fatty acids, %[1]	27,8±1.8 [a]	16.3±2.4 [a]	15.8±2.2 [a]

Values are means ±SD. Same letters ([a]) show significant ($P < 0.05$) differences. [1] = in the intramuscular lipids.

As seen in the table ratio of *saturated fatty acids* (SFA) at 14 and 19 months ages shows significantly higher values in each type of muscle than 7 months of age. However, no significant changes could be detected in this group of fatty acids between the two higher age groups. Significant changes in the amount of *monounsaturated fatty acids* (MUFA) can be detected only in the longissimus dorsi. This muscle showed significantly higher levels of this fatty acid group at age 14 and 19 months than at 7 month age. As young bulls became older the percentage of *polyunsaturated fatty acids* (PUFA) in the mentioned muscle markedly decreased, except in the semitendinosus, mostly from 7 to 14 months of age.

Conclusions The study indicates clearly that age and anatomical location of muscle samples are important sources of variation in the intramuscular lipid content and fatty acid profile of muscles in Holstein-Friesian bulls. Consequently, these two factors should be taken into consideration when evaluating the nutritive value of the meat of Holstein Friesian bulls for human consumption.

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The effect of absence of protozoa on ruminal biohydrogenation and the fatty acid composition of lamb muscle

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Introduction It has been shown that rumen protozoa have a higher content of conjugated linoleic acid (CLA) and trans-vaccenic acid (TVA) than rumen bacteria, suggesting that protozoa may make a significant contribution to TVA and CLA flow from the rumen (Devillard *et al.,* 2004). As part of a project studying the role of protozoa in rumen bio hydrogenation, this study was carried out to determine the effect of the absence of protozoa in the on ruminal biohydrogenation, microbial diversity and the fatty acid composition of lamb muscle.

Material and methods Twenty crossbred lambs were used. Ten of them were taken from the ewes within the first 24 h after birth and kept isolated from adult animals (protozoa-free group, PF) while the other ten were left with the ewes (control group, C). Both groups were weaned at six weeks. Once weaned the lambs grazed on two different pastures (predominantly perennial ryegrass) supplemented with a concentrate supplement (Wynstay Lamb Finisher, 250 g/head/d) for 4 months prior to slaughter. Protozoa-free lambs remained isolated from other ruminants during this phase. At slaughter samples from the rumen, abomasum and tail muscle were collected and stored at -20°C until analyses were performed. Lipid extraction and subsequent GC analysis on abomasal content and muscle samples were, respectively, performed as described by Lee *et al.* (2003) and Scollan *et al.* (2003). Bacterial diversity in rumen samples was assessed by polymerase chain reaction and denaturing gradient gel electrophoresis (PCR-DGGE) using 16S ribosomal DNA specific primers (Yáñez Ruiz *et al*., 2004). Total bacterial DNA in the rumen was estimated by real-time PCR (Maeda *et al*., 2003). Statistical analysis of fatty acid profiles was undertaken by ANOVA.

Results Protozoa-free lambs remained defaunated during the trail and no ciliates could be recovered from their rumen at slaughter. Protozoa free lambs had lower ($P<0.05$) abomasal concentrations (Table 1) of C18:0 but C18:1 *trans* and C18:2 *cis*9, *cis 1*2 concentrations were higher. The fatty acids of muscle in protozoa free animals had lower concentrations of C18:0 and higher levels of C18:2 *cis* 9 *cis* 12 and CLA (C18 *cis* 9, *trans* 11 and C18:2 *trans* 10, *cis* 12). Total bacterial DNA did not differ ($P>0.05$) between experimental groups, however banding DGGE profiles showed a markedly different bacterial population in protozoa-free and control animals.

Table 1 Fatty acid composition (moles/100 moles fatty acids) of abomasal contents and muscle.

	Abomasum				Muscle			
	C	PF	s.e.d.	P value	C	PF	s.e.d.	P value
C16:0	16.8	17.9	0.775	0.367	18.9	18.2	0.69	0.473
C16:1	1.61	1.76	0.173	0.55	1.36	1.42	0.091	0.631
C18:0	41.2	32.8	2.04	0.02	23.6	19.7	0.76	0.008
C18:1 t	5.81	7.47	0.484	0.042	5.31	4.94	0.465	0.6
C18:1 c	10.6	10.1	0.89	0.699	32.0	30.8	1.22	0.526
C18:2 c9 c12	10.3	14.0	1.07	0.042	8.65	11.53	0.89	0.052
C18:3	5.82	6.14	0.728	0.765	1.32	1.98	0.118	0.004
C18:2 c9, t11	0.05	0.04	0.006	0.34	1.34	1.85	0.111	0.043
C18:2 t10, c12	0.02	0.02	0.004	0.955	0.11	0.42	0.04	0.007
C20:4n3	3.73	4.67	0.64	0.329	3.14	3.89	0.53	0.344

Conclusions Despite the high CLA and TVA content of rumen protozoa the present study found an increase in TVA in the abomasum and CLA in muscle of lambs when protozoa were not present in the rumen. It seems that hydrogenation of linoleic to stearic acid is lower in the rumen of protozoa-free lambs, leading to a higher CLA concentrations in the muscle and suggesting a shift in the processes involved in CLA and TVA formation in the rumen. More studies are required to fully elucidate the role of rumen protozoa in ruminal lipid biohydrogenation and TVA and CLA production.

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Relationships between carcase characteristics in beef cattle

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Introduction Information from cattle passports and abattoirs now make it possible to add breed type, sex category and age at slaughter to records of carcase weight, fatness and conformation. The objective of this study was to investigate relationships between slaughter age, carcase weight, fatness and conformation in crossbred beef cattle using Limousin X steers as an example.

Materials and methods A combined cattle passport and beef carcase database of 156,646 cattle slaughtered between early December 2003 and mid February 2005 was dominated by continental crosses sired by Limousin, Charolais, Simmental and Belgian Blue bulls, in that order. The database was analysed to examine relationships between slaughter age and carcase characteristics.

Results There was large variability in slaughter age and carcase weight at a given slaughter age, even for a single breed type and sex category slaughtered in the same fat grade (Figure 1). Slaughter was mainly confined to the period after 22 months of age, when steers qualified for the second Beef Special Premium, and before the Over Thirty Month Scheme limit at 30 months of age – a reflection of the prevailing subsidy system. There was little or no increase in average carcase weight over the main slaughter age range. Table 1 shows a relationship between fat and conformation grade. As carcases increased in fatness up to over fat grade 4H, percentages of average conformation grade R and better –U grades increased, while poorer than average O+ and –O grades declined. The improved conformation profile benefited price per kg but at fat grade 4H this is more than offset by the usual penalty for over fatness. The increase in fat grade was associated with reduced age at slaughter but greater carcase weight, i.e. higher daily liveweight gain. It seems that, at least in part, cattle that gain weight more slowly are more likely to remain in leaner fat grades. A similar general relationship existed for all breeds and sex categories.

Figure 1 Slaughter age and carcase weight in Limousin cross steers slaughtered at fat grade 4L

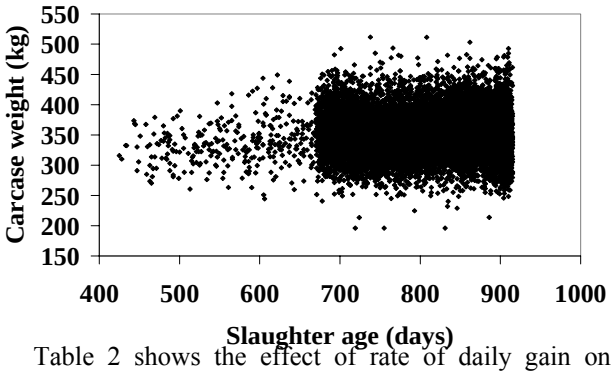

Table 2 shows the effect of rate of daily gain on carcase weight at an example slaughter age of 23 months and the percentages of carcases that achieved target fat grade 4L and their conformation profiles. At a weight of 325 – 375 kg (daily gain 0.82 kg) almost two thirds of carcases were in fat grade 4L and almost 90% in conformation grades R or better, whereas carcases less than 275 kg (daily gain 0.58 kg) had over 50% in discount grades O+ and –O. The conformation profile was even better in heavy carcases over 375 kg but these are penalised as being overweight by some processors. The effects of daily gain were more pronounced at 23 months than at older slaughter ages.

Table 1 Relationships between fat grade and carcase quality in Limousin X steers

Fat grade	2	3	4L	4H
No of carcases	842	6680	13355	1750
% of total	3.7	29.5	59.0	7.8
Carcase weight (kg)	311	336	352	362
Age (days)	836	823	809	789
Calculated daily liveweight gain (kg) *	0.62	0.69	0.74	0.78
Conformation (% in each fat grade)				
E	1.1	1.5	1.0	0.2
U+	3.5	4.9	3.1	1.5
-U	19.2	24.0	31.2	31.9
R	37.8	45.8	51.0	57.0
O+	31.0	21.9	13.3	9.0
-O	7.4	1.9	0.4	0.3

* Killing out 55%, birth weight 45 kg

Table 2 Effects of carcase weight on conformation in Limousin X steers slaughtered at 23 months

Carcase weight (kg)	<275	275-325	325-375	>375
Number	42	499	1002	458
Fat 4L (%)	35.7	57.1	64.3	61.1
Gain (kg/day)	0.58	0.72	0.82	0.95
Conformation grade (%)				
E	0	0	0.4	7.4
U+	0	0.4	2.8	14.4
-U	7.1	18.4	36.5	48.7
R	38.2	57.1	49.9	26.0
O+	47.6	22.9	10.1	3.5
-O	7.1	1.2	0.3	0

Conclusion At a given slaughter age increased growth rates and therefore carcase weights were associated with improved carcase conformation. Knowledge of the relationship between fat and conformation grades and the way it and carcase weight are driven by growth rate allows minimum lifetime rates of daily gain targets to be set for each cattle type in beef systems with a given slaughter age that allow the target carcase weight to be achieved at fat grade 4L with a favourable conformation profile.

Acknowledgement The study was funded by the English Beef & Lamb Executive (EBLEX).

Relationships between liveweight and condition score in Aberdeen Angus crossbred and Limousin crossbred commercial suckler cows

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Introduction Since the announcement that mature cull cows can re-enter the human food chain, there is increasing interest from commercial producers in devising systems that finish suckler cows to an acceptable degree of fatness and conformation score. A detailed understanding of the relationships between cow liveweight (LW) and condition score (CS) is necessary if cows are to be grown and finished to achieve optimum carcass weights and marketed at optimum commercial returns. Previous work conducted over 20 years ago with traditional UK suckler cow types at that time, showed that for every one unit change in suckler cow CS the corresponding change in cow LW ranged from 61 – 110 kg with an average value of 95.6 kg (Wright and Russel, 1984) for cows that ranged in average LW from 434 – 560 kg. However, little information is available on the relationships between suckler cow LW and CS in suckler cow genotypes present in the UK today. The objective of this study was to estimate the relationship between suckler cow LW and CS in modern Aberdeen Angus cross (AAx) and Limousin cross (LIMx) suckler cows.

Materials and methods Both LW and CS data were gathered over two years (2003-2004) from a two-way rotational crossbred suckler cow herd maintained at SAC and managed under commercial conditions. Individual cow LW and CS (Lowman *et al*, 1976) measurements were determined at various times between weaning in late October and immediately pre-calving in April from mature spring calving suckler cows of either AAx or LIMx genotypes that were 2nd calvers or older. 1st calved heifers were excluded from the analysis since they were considered to be immature and still growing without neccessarily laying down body condition (i.e. subcutanous fat). A total of 118 and 104 data sets where both LW and CS had been determined simultaneously were available for the AAx and LIMx genotypes respectively. Average cow age was 5.3 (s.e. 0.81) and 5.8 (s.e. 1.40) years for the AAx and LIMx cows. Linear regression analysis was used to establish the relationships between suckler cow LW and CS for both the AAx and LIMx genotypes using Genstat 5.

Results Average cow LW (kg) and CS (1 – 5 scale) measures for the AAx suckler cows were 682 (s.e. 34.6) and 2.51 (s.e. 0.358) whilst the corresponding average LW and CS measures for the LIMx suckler cows were 688 (s.e. 40.0) and 2.62 (s.e. 0.283) respectively. The linear regression relationships between LW and CS for the AAx and LIMx cow genotypes are also shown in Figures 1 and 2 respectively. These linear relationships show that for AAx cows, a one unit change in CS is associated with a LW change of 47.0 kg over the CS range from 1.75 to 3.75. Similarly, a one unit change in CS for LIMx cows is associated with a LW change of 75.3 kg over the CS range from 1.75 to 3.5.

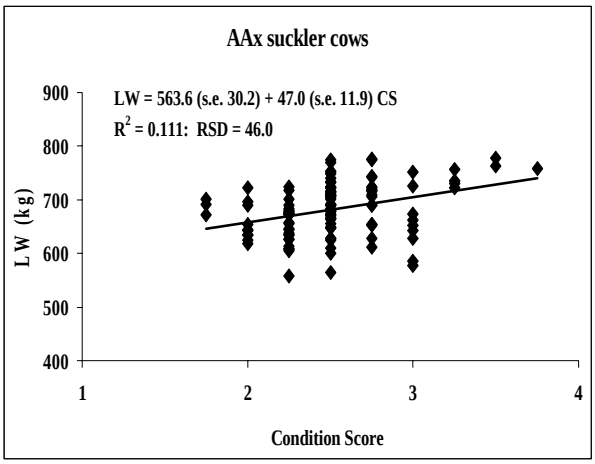

Figure 1 LW *vs* CS in AAx suckler cows.

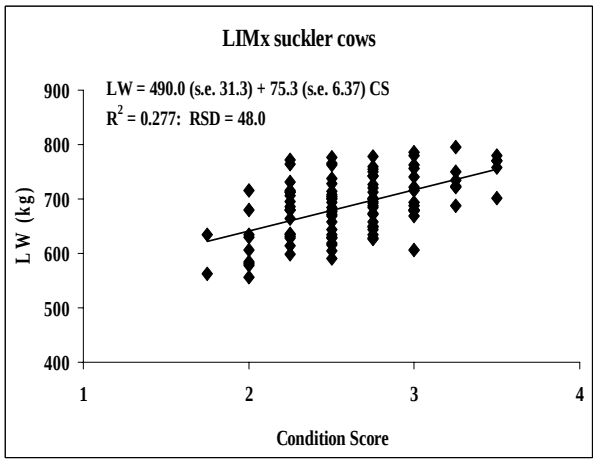

Figure 2 LW *vs* CS in LIMx suckler cows.

Conclusions Results from this study indicate that average cow LW at approximately 680 kg is considerably higher than the average cow LW reported by Wright and Russel (1984), probably as a result of genetic selection for higher body weight over the last 20 years. However, a one unit change in cow CS in this study is generally associated with a smaller range of corresponding change in cow LW than previously. Commercial producers should apply these updated relationships between LW and CS when finishing modern suckler cull cow genotypes.

Acknowledgements SAC receives financial support from SEERAD.

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Commercial beef production from Holstein Friesian bulls

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Introduction Information from cattle passports and abattoirs now make it possible to add breed type, sex category and age at slaughter to records of carcase weight, fatness and conformation. The objective of this study was to examine the performance of Holstein Friesian bulls in commercial beef production.

Material and methods Records from 7524 bulls registered with BCMS as Holstein Friesian and slaughtered between early December 2003 and mid February 2005 were analysed to characterise commercial bull beef production from this type of cattle. Lifetime daily liveweight gain was calculated assuming a killing out percentage of 53 and a birth weight of 45 kg. The final farm holding number was used to identify producers that slaughtered at least 20 bulls for a study of farm effects.

Results The variability of slaughter age and carcase weight at a given slaughter age was far greater than expected (Figure 1). At the average slaughter age of 470 days calculated daily liveweight gains ranged from 0.9 kg per day at the bottom of the main carcase weight range (about 120 kg) to 1.4 kg/day at the top. Average carcase weight increased from approximately 250 kg at 300 days to 300 kg at 550 days, but started to decline after 600 days. Few bulls were slaughtered by the 380-day standard that is widely accepted for an all concentrate Holstein Friesian bull beef system. Indeed, more than 25% of bulls were over 16 months of age at slaughter and more than 40% of carcases were below 275 kg, which are penalised severely in some price schedules. It seems probable that many producers restricted concentrate feeds for at least part of the feeding period and included forage, including grazing in some cases.

Results from 91 farm groups of Holstein Friesian bulls are presented in Table 1 ranked in order of calculated lifetime daily liveweight gain. Top third producers had daily gains more than 0.2 kg LW per day greater than the bottom third. This enabled them to slaughter cattle seven weeks sooner yet with carcases more than 30 kg heavier. Nevertheless, the top third slaughter age of 446 days was considerably older than the 12 month standard usually set for intensive dairy bull beef. Not only were top third carcases heavier but more were in target fat grades 3 and 4L and there were fewer highly undesirable conformation grade P carcases and more acceptable conformation grade O+. Any effects on cost of production are not known. Individual top third producers were generally adept at selecting bulls in target fat grades 3 and 4L. However, whilst one individual producer selected 95% of carcases in these fat grades, another was less certain and had over 20% of carcases in under finished fat grade 2.

Figure 1 Age at slaughter and carcase weight

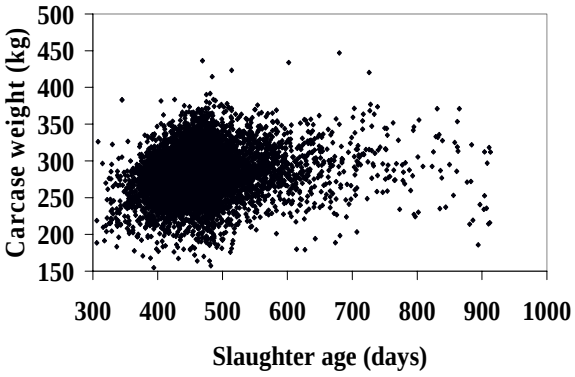

Table 1 Results from 91 farms

	Bottom 1/3	Average	Top 1/3
Cattle per farm	84	59	46
Age (days)	482	462	446
Carcase (kg)	266	282	299
Daily gain (kg)	0.95	1.06	**1.16**
Fat (% in each grade)			
1	4.1	2.3	0.9
2	42.5	25.7	9.6
3	44.4	47.9	44.3
4L	9.0	23.1	**42.7**
Conformation (% in each grade)			
O+	19.7	23.9	26.5
-O	55.9	56.6	59.2
P+	21.8	16.9	12.8
-P	1.5	1.2	**0.8**

Conclusions Performance of Holstein Friesian bull beef on commercial farms was highly variable and, on average, fell well short of the generally accepted cereal beef target of a minimum 260 kg carcase at around a year of age from cattle gaining at least 1.15 kg per day from birth to slaughter. Producers with the top third of calculated daily liveweight gains achieved the target gain for cereal beef but retained bulls for almost 10 weeks longer to a carcase weight almost 40 kg heavier. An advantage of the greater carcase weight was a better conformation profile that would benefit sale price per kg carcase. It is not known whether this was economically advantageous taking the cost of extended feeding into account. With financial margins for this type of beef production under pressure, more information is required from commercial farms to identify methods and costs of production.

Acknowledgement This study was funded by the English Beef & Lamb Executive (EBLEX).

The determination of *in situ* protein degradability characteristics of some feedstuffs and comparing to AFRC Standard Tables in the feeding of lactating cows

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Introduction Rumen microorganisms change feed protein characteristics and convert some feed protein to microbial protein. Determination of feed protein degradability coefficients and acid detergent insoluble nitrogen (ADIN) content is necessary for calculation of metabolisable protein (MP) content of feedstuffs. Absence of true information about these coefficients of native feedstuffs is a prime problem for diet formulation in Iran. The objective of this study was to determine the protein degradability coefficients of some native feedstuffs and compare them with AFRC (1995) data (the system now being used in Iran) in the feeding of lactating cows in terms of intake, milk yield and milk composition.

Materials and methods This study was fulfilled in two stages. Experiment I) 2 fistulated Blouchi castrated rams (wether) were used for determination of protein degradability coefficients for five feedstuffs (barley grain, wheat bran, soybean meal, beet pulp and cottonseed meal) with *in situ* technique. Rams were fed a mixed diet of forage and concentrate (50:50) (880 g DM /day) in TMR. Experiment II) 16 lactating Holstein cows (milk yield 29.5 ± 3 l/d) used in a complete block design (2 block, 2 treatment, 4 repeat). Animals spent 10 days for adaptation with experimental diets. Experimental period was 35 days. The diets were formulated to meet the nutritional requirements of cows according to either AFRC (1995) or the experimental data. Animals were fed diets and water without any restriction (ad-lib). Two diets (1 and 2) were formulated according to AFRC coefficients. Diet 1 contained forage (lucerne hay and maize silage 20.33 and 37.76% of DM respectively) and concentrate (barley, wheat bran, beet pulp, cottonseed meal, soybean meal and premix 18.88, 6.39, 5.23, 7.46, 4.36 and 0.64% of DM respectively). Diet 2 contained forage (lucerne silage, lucerne hay and maize silage 10.17, 10.17 and 37.76% of DM respectively) and concentrate similar to diet 1. Diets 3 and 4 were formulated according to experimental data and had a similar composition to diets 1 and 2 respectively except for cottonseed meal and soybean meal (8.13 and 2.61% of DM respectively). Feed intake and milk yield were measured daily. Milk samples were taken from the a.m and p.m milking in mid and end of experimental period. Milk samples were analyzed for fat, crude protein, NPN and DM. Data were analyzed by analysis of covariance using the GLM procedure of SAS.

Results Protein degradability coefficients of feedstuffs in experiment I are presented in Table 1.DMI, milk yield and milk composition is given in Table 2. The results indicated that the cows fed with diets based on the degradability coefficients of experiment I had higher milk yield (kg/d), milk protein (g/d), milk NPN(g/l) and milk dry matter (g/d) ($p<0.05$), when compared to the other groups.

Table 1 Protein degradability coefficients of feedstuffs in stage 1

Feedstuffs	Degradability coeff		
	a	b	c
Soybean meal	0.49	0.39	0.14
Barley grain	0.62	0.2	0.06
Beet pulp	0.59	0.33	0.1
Wheat bran	0.72	0.24	0.2
Cottonseed meal	0.58	0.34	0.03

Table 2 Effects of degradability coefficients on DMI, Milk yield and Milk composition[1](end period collection)

Item	Degradability coeff		SEM
	Current coeff	AFRC coeff	
DMI(kg/d)	22.48[a]	21.68[a]	0.78
Milk yield(kg/d)	32[b]	27.97[a]	0.57
Milk composition			
fat(g/l)	32.33[a]	34.83[a]	3.84
protein(g/l)	31.58[a]	30.07[a]	0.96
NPN(g/l)	0.353[a]	0.344[a]	0.009
DM(g/l)	111.73[a]	117.1[a]	2.98

1.Values with unlike letters were significantly different :$P<0.05$

Conclusion: The results showed that current coefficients are more capable to supply requirements of animals for milk yield persistency than AFRC coefficients. Use of diets formulated with experimental data did not have significant effect on milk composition. However additional researches are need.

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An alternative test protein for the determination of protein precipitation capacity of tannins

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Introduction Tannins are secondary plant compounds frequently encountered in tropical feeds. They are known to complex with proteins, thus reducing the availability of protein to ruminants. Tannin determination is thus a crucial factor when estimating the feed quality of tropical plants. Bovine serum albumin (BSA), which is commonly used in tannin binding assays has the drawbacks of being a non-plant protein and requires a rather low pH (4.9, iso-electric point of BSA) for complex formation with tannins. Papain is a commercially available pure protein from *Carica papaya* that may be a suitable alternative. This study evaluates papain as a test protein in a tannin binding assay in comparison to BSA by determining the specific protein precipitation capacity (sPPC) of the tannin rich plants, *Salix alba* (salix) and *Rhus typhina* (sumach). Salix and sumach leaves contain exclusively condensed tannins and hydrolysable tannins respectively.

Materials and methods Leaves of salix and sumach were freeze-dried and ground to pass through a 1 mm sieve. Tannins were extracted from the leaves using 70% aqueous acetone. The ability of tannins to precipitate BSA and papain was assayed as described by Hagerman and Butler (1980). A series of crude tannin extract containing 10 - 50 mg/ml tannin were each added to a series of protein concentrations (0.5, 1, 2, 4, 6, 8, 10 mg/ml) in 0.2M acetate buffer (pH 4.9) and 0.1M phosphate buffer (pH 6.8). Tannin and total protein content of the isolated tannin-protein complex were determined using ferric chloride (Makkar *et al*., 1988) and dot blot (Hoffmann *et al*., 2002) assays respectively.

Results BSA was precipitated by the tannins more effectively compared to papain at pH 4.9 (salix: 67 g protein/g tannin; sumach 63 g/g) but papain more effectively at pH 6.8 (salix: 56 g/g; sumach 42 g/g) as shown in Table 1. These results confirmed previous findings that tannins do not precipitate BSA at pH 6.8, which is the pH encountered in the rumen. Optimal protein concentration was 4 mg protein/g plant material for both BSA and papain. At this concentration, titration curves showed statistically linear ($p<0.05$) correlations for increasing quantities of tannins for BSA (salix: up to 2.6 mg tannins/g plant material; sumach: up to 12.7 mg/g) as well as for papain (salix: up to 7.1 mg/g; sumach: up to 5.9 mg/g). These values indicate differences in the affinities of the tannins for the proteins.

Table 1 Specific protein precipation capacities of tannins using BSA and Papain

Tannin	Protein	sPPC (g protein/ g tannin)	
		pH 4.9	pH 6.8
Salix	BSA	65.2A[a]	7.0B[b]
Salix	Papain	28.0B[b]	55.0A[a]
Sumach	BSA	55.7A[a]	0.9B[b]
Sumach	Papain	18.2B[b]	38.9A[a]

* Means are compared only within tannins
* Means in the same column with different capital letters are significantly different ($p<0.001$)
* Means in each row for each protein with different small letters are significantly different ($p<0.001$)

Conclusion This study avails a new test protein for protein precipitation assays that may be used under physiological conditions. Further studies are required to test the correlations of the PPC obtained using BSA and papain to other rumen fermentation parameters to compare the potential of the test proteins to predict the feeding value of tanniniferous plants.

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An *in vitro* methodology to estimate ruminal starch degradation kinetics

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Introduction Problems with current ruminal starch degradation estimates (e.g. *in sacco* technique - excessive initial losses; rate of passage studies - expense and time consuming) render them unsuitable for routine use. These together with inaccurate starch assays (Krystallidou and Mould, 2004) and inappropriate rumen outflow kinetics, combine to produce a questionable data. Considering the role of starch in the nutrition of high producing dairy cows and its impact on the rumen fermentative environment, a system that accurately predicts both the extent and site of starch degradation would be of strategic importance. It was hypothesised that starch degradation *in vitro* is likely to be similar if not identical to that of the rumen. Thus starch outflow kinetics can be calculated using the difference between expected (*in vitro*) and assayed rumen levels following a discrete starch meal. Further, the administration of an appropriate marker, with outflow kinetics similar to starch, will allow effective ruminal starch degradation to be estimated from *in vitro* data, using intact animals offered different feeding regimes

Materials and methods Two rumen fistulated Holstein-Friesian dairy cows were used. The animals were 225 days in milk (mean), produced 25 kg day^{-1} and consumed about 20 kg DM of a grass silage/ hay-based TMR. The experiment examined two different starches in consecutive studies. Days 1 to 7 was an adaptation phase with either 3.5 kg cracked maize or wheat grain included in the ration. Days 8 to 11 no supplemental starch was offered with the exception of a single feed on Day 10 when the animals were also dosed with two solid-phase markers, namely; chromium-mordanted wheat straw (Cr) and ytterbium acetate (Yb) and a liquid-phase marker cobalt-EDTA (Co). Thereafter the same diet as days 1 to 7 was offered. On Days 10 and 11, rumen grab samples (5 l) were taken at intervals to estimate rumen volume (initial marker concentration) and starch disappearance. Total faecal collections (Days 10 to 15) were used to determine marker outflow kinetics. Starch degradation kinetics were assessed using *in vitro* (Days 13-14) and *in sacco* (Day 15) methodologies. Theoretical ruminal starch degradation was estimated by applying *in vitro* values to the initial rumen starch content assayed post-feeding. Marker passage rates were calculated using a multi-compartmental model (Dhanoa *et al*., 1985).

Results The *in sacco* technique provided poor estimates of ruminal starch degradation. Excessive initial losses of wheat occurred, while maize was poorly degraded due to the lack of physical disruption normally associated with mastication (Table 1). *In vitro* data showed maize to be more slowly fermented than wheat and to a lesser extent. A similar effect was identified with the gas release profiles with wheat peaking earlier and higher than maize. In addition highly significant (P>0.001) relationships between cumulative gas production and starch degradation were obtained suggesting that gas release may offer a predictive estimate of starch degradation kinetics.

Table 1 Starch degradation and outflow

		Wheat	Maize
in sacco	Washing losses (g g^{-1})	0.485	0.131
	Effective degradation (g g^{-1})	0.874	0.551
in vitro	Cumulative gas (ml) - 12h	166	44
	- 24h	305	246
	DM degradation (g g^{-1}, 24h)	0.970	0.692
in vivo	Ruminal starch loss (g) 4h	1800	1491
	Residual starch (g) 18h	406	923
	Starch outflow (h^{-1})	0.102	0.110

Table 2 Marker outflow data

	Co	Yb	Cr
Rumen outflow (h^{-1})	0.105	0.043	0.048
Transit time (h)	11.9	15.9	24.0
Retention time (h)	25.3	44.6	98.4

Passage rates differed between markers (Table 2) with Co providing much higher values than either Yb or Cr. Transit and retention times also varied with Yb producing more consistent results than Cr. No effect of starch source on marker outflow was observed. Although assayed rumen starch contents of 3.0 kg were identified immediately post-feeding with either wheat or maize, this had almost completely disappeared after 18 h. Combining starch contents and estimated degradation, identified a substantial quantity of starch left the rumen in the period immediately post-ingestion. Rumen starch outflow estimates obtained using 2 to 18 h data strongly suggest that cereal starch particles leave the rumen in the liquid, rather than the solid, phase.

Conclusions These data indicate that cereal starch particles appear to leave the rumen rapidly in the liquid phase. It is suggested that the extent of ruminal starch degradation may be predicted from gas release kinetics, modified by an appropriate rate of outflow obtained from liquid-phase marker studies. Such a methodology will allow a more accurate estimation of the partitioning of starch degradation products into propionic acid (rumen) and glucose (post-rumen), so leading to improved diet formulation and nutrient utilisation.

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Cultivation of oyster mushrooms (*Pleurotus* species) to improve the *in vitro* dry matter digestibility of wheat straw for feeding to ruminants

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Introduction Wheat is grown because it provides grains for human food and straw that can be fed to ruminants. In recent years, wheat varieties that have shorter, thicker stems for supporting a higher grain yield have been a popular choice by farmers. Straw derived from these wheat varieties has a higher lignin content. Several methods to attempt to improve the nutritional value of wheat straw have been tested without much success, due mainly to the reduced palatability of chemically treated straw, the high cost of the treatments or to the increased labour required to perform the treatment. It has already been shown that some Pleurotus species (white rot fungi of the Basidiomycota) can selectively degrade lignin in wheat straw, making the cellulose and hemi-cellulose move available for digestion by ruminants. Selecting a Pleurotus variety which could be induced to produce edible fruit bodies could result in a profitable enterprise and thus offset the increased costs of a biological treatment. The ability of five strains of Pleurotus to increase the digestibility of wheat straw has been investigated in this study.

Materials and methods Wheat straw used in the study was grown in Eastern England. All of the straw was derived from the same Heston bale. The straw was chopped into 10 cm lengths to improve the fungal colonisation. Straw was soaked overnight to reduce the free phenolics content, which may retard the growth of the fungi on the straw substrate. Chopped straw (200 g) was placed into 36 autoclavable polypropylene bags (30 * 60 cm). Prior to fungal inoculation all bags were autoclaved at 121ºC, for 20 minutes. All fungi (Table 1) were maintained on 4% malt agar plates, in the dark at 28ºC and were precultured on sweetcorn kernels for 6 days before inoculation into straw containing bags, after Hodgson (2004). After inoculation the bags were shaken to ensure even distribution of the kernels throughout the bags and after 5 days to improve aeration and distribution of the inoculum. Each fungus was grown in six straw filled bags whilst in a further three bags a similar quantity of sterile kernels were introduced (i.e. controls without fungus). The study involved 36 straw bags in total. Bags were incubated in the dark at 28ºC for 18 days then placed into a refrigerator at below 9°C for 24 hours. Subsequently all of the bags were incubated at 20ºC, 90% relative humidity with light for 8 hours per day. Pinholes were made in bags to ensure adequate ventilation and allow emergence of developing fruit bodies. Eighteen of the bags were incubated for 6 weeks and subjected to chemical analysis whilst the remaining 18 were incubated for a further 12 weeks to see if they would produce fruiting bodies. Chemical analyses were performed on the 6 week incubated straw samples after milling to pass a 1mm mesh. Crude protein levels of the treated straw were determined by the Kjeldahl method, lignin was determined by the Effland (1977) method and the in vitro dry matter digestibility was determined by the Modified Faeces Liquor method (Omed *et al*., 1989).

Results Mean values for crude protein, in vitro dry matter digestibility and lignin are shown (Table 1).

Table 1 Mean values for results (SD in italics, *statistically different from the control (P<0.05))

Fungus (strain)	Crude protein (%)	In vitro DM digestibility (%)	Lignin (%)
Pleurotus ostreatus (40C)	2.56 (0.04)	28.45* (1.84)	8.90 (0.05)
Pleurotus ostreatus (027)	2.56 (0.04)	26.65* (1.22)	8.86 (0.21)
Pleurotus ostreatus (136)	2.50 (0.18)	31.47 (3.04)	7.91 (0.62)
P. eryngii (DSM8264)	2.47 (0.10)	34.83 (1.08)	7.96 (0.21)
P. ostreatus (commercial)	2.91* (0.02)	28.24 (1.32)	9.11 (0.19)
Control	2.26 (0.26)	33.56 (3.52)	8.70 (0.24)

Discussion Most of the fungal strains studied in this experiment did not develop fruiting bodies within 6 weeks. However, Pleurotus ostreatus (027) and P. ostreatus (136) did produce fruiting bodies under these experimental conditions and the second set of replicate bags of the commercial strain fruited at 10 weeks. Most of the fungi significantly reduced the in vitro digestibility or showed no difference but P. eryngii (DSM8264) showed a slight increase (ns) as compared to the control samples. Crude protein levels were increased with all fungal treatments; with the P. ostreatus (commercial) the increase was statistically significant. Pleurotus strains DSM8264 and 136 reduced the lignin content (ns). High variability between replicates obscured the results probably due to the heterogeneous nature of the substrate (e.g. leaves and straw were indistinguishable after fungal treatment).

Conclusion Pleurotus eryngii (DSM8264) is a potential candidate for improving the digestibility of wheat straw as the straw treated with this fungus had a higher digestibility value and lower lignin content than most of the other fungi. However, in this experiment these values weren't considered to be significant ($p<0.05$). Future work should involve this fungus in particular, have greater sample numbers and should look at modifications to the culture conditions.

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Effect of steam-pressure treatment on physico-chemical properties and bio-utilization of sugarcane bagasse

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Introduction Agricultural by-products like cereal straws and sugarcane bagasse are high in lignocellulose. Sugarcane bagasse contains more than 60% of its dry matter in the form of cellulose and hemicellulose but its degradability is very low. One of the main reasons for this low degradability is the presence of lignin which protects carbohydrates from attack by rumen microbes (De La Cruz, 1990). The main effect of steam treatment is to increase feed intake, overall digestibility and live-weight gain which is achieved because of the extensive destruction of cell wall and hemicellulose hydrolysis which increases soluble sugars and makes cell wall more degradable. The aim of this study was to assess the effect of steam pressure and reaction time on physico-chemical and bio-utilization of sugarcane bagasse.

Material and Methods Samples containing 50% moisture were put in stainless steel baskets and placed in a reaction chamber. Steam explosion of samples was carried out by direct injection of steam into the chamber and the samples were kept under a specified pressure and period of time (reaction time) followed by depressurization. A 3×3 factorial arrangement with three replicates was used to assess the effect of pressure and reaction time on hemicellulose (Ternud *et al*, 1989) content. After characterization of optimum enzyme/substrate ratio, samples were incubated with 128 IU Cellulase Onozuka (EC: 3.2.1.4) for 48 h and then reducing sugar content determined (Nelson, 1944). Accessible pore volumes (APV) for 8, 12, 51, 110, 270 and 550 A^o probe molecules (Stone and Scallan, 1968) were measured according to unabsorbed sugar concentration of samples. Data from three pressures 14, 17 and 20 atm and 3 reaction times 120, 180 and 240 seconds were analyzed (ANOVA) and means were tested according to Duncans's least range test while all means were compared with untreated bagasse (control) by Dunnett's test.

Results Results are presented in Table 1 which showed that steam-pressure treatment significantly decreased hemicellulose and increased reducing sugar concentrations after enzymic hydrolysis and also, all values for APV improved in comparison with untreated sugarcane Bagasse ($P<0.05$). Increasing pressure resulted in considerable improvement of all parameters except for APV_{12} ($P<0.05$), However increasing the reaction time did not significantly ($P<0.05$) influence sugar and APV values. The only observed significant interaction between pressure and temperature was for hemicellulose.

Table 1 Hemicellulose content, reducing sugars and APV of steam treated sugarcane bagasse.

Factor		Pressure (atm)			Reaction Time (s)						
Parameter	Control	14	17	20	120	180	240	s.e.m	P	T	P*T
Hem[1]	23.95	21.48[a2]	18.59[b]	15.17[c]	20.58[a]	18.12[b]	16.53[c]	0.639	**[3]	**	**
R.Sug	6.48	9.25[a]	10.62[ab]	11.57[b]	10.13	10.47	10.84	0.318	*	NS	NS
APV_8	0.70	0.86[b]	0.92[ab]	1.00[a]	0.89	0.96	0.93	0.021	*	NS	NS
APV_{12}	0.32	0.76	0.80	0.84	0.77	0.81	0.82	0.017	NS	NS	NS
APV_{51}	0.23	0.49[b]	0.56[ab]	0.60[a]	0.51	0.56	0.58	0.017	*	NS	NS
APV_{110}	0.15	0.25[b]	0.33[a]	0.39[a]	0.28	0.34	0.35	0.016	*	NS	NS
APV_{270}	0.07	0.10[c]	0.14[b]	0.19[a]	0.13	0.15	0.15	0.009	*	NS	NS

[1]Hem: Hemicellulose%, R.Sug: Reducing Sugar%, APV_8, APV_{12}, APV51, APV_{110} and APV_{270} accessible pore volumes for 8, 12, 51, 110 and 270 A^o probe molecules (ml/g).
[2]Means with different letters in the same row are significantly different ($P<0.05$).
[3] ****** , $P<0.01$, ***,** $P<0.05$ **NS**, non significant

Conclusion Steam treatment of sugarcane bagasse significantly decreased its hemicellulose content and thus its bio-utilization because of the more cell wall destruction and extended accessible pore volumes which resulted in better availability of cell wall for cell free enzymes.

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Use of gamma irradiation to increase digestible undegraded protein of soya-bean meal

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Introduction Soya-bean meal (SBM) proteins are extensively degraded in the rumen, and various physical and chemical treatments have been used to decreased its extent of ruminal degradation (Sadeghi *et al.,* 2005), thereby increasing its content of metabolizable protein. As reported by Urbain (1986), gamma irradiation can affect proteins by causing conformational changes, formation of protein free radicals, and recombination and polymerization reactions. To our knowledge, no information is available concerning effects of gamma irradiation on ruminal protein degradation and type of SBM true proteins that leave the rumen undegraded. The main objectives of present study were to monitor fate of gamma irradiated SBM true proteins in the rumen by using sodium dodecyl sulphate-polyacrylamide gel electrophoresis (SDS-PAGE) methodology.

Materials and Methods Gamma irradiation was carried out in a cobalt-60 irradiator. Four polyethylene packages of SBM samples were irradiated in a gamma cell at doses of 0, 25, 50 and 75 kGy at room temperature. The dose rate was 2.0 kGy/h (Holm and Berry, 1970). Four 500-kg Holstein cows fitted with rumen fistulas were used for *in situ* incubations. Ruminal disappearance of CP (two bags for each cow at each incubation time) was measured at 0, 2, 4, 6, 8, 10, 12, 24 and 48 h. Digestibility was measured by nylon bag method of Subuh *et al.* (1996). Fifteen mg of well-ground dried untreated or treated SBM was placed into 750-µl SDS-PAGE sample buffers. After 30 min of thorough mixing, samples were immersed at 90°C for 3 min, and then centrifuged at 10000g for 1 min. A 30-µl of each protein sample was then loaded into the sample well. Samples were fractionated by a SDS-PAGE discontinuous system as described by Sadeghi *et al.* (2005).

Results and discussion In soya-bean, the two major protein observed were β-conglycinin and glycinin. The approximate molecular weights of the β-conglycinin ά, α and β subunits were 90.5, 71.5 and 55.2 kDa and glycinin acidic and basic subunits were 37.6 and 19.8 kDa, respectively. Electrophoretic analysis of untreated SBM proteins (Figure 1, A) incubated in the rumen revealed that β-conglycinin α and ά subunits were degraded completely within 2 h, whereas the β subunit of β-conglycinin as well as the acidic and basic polypeptide components of glycinin were more resistant to degradation, until 8, 12 and 48 h, respectively. Electrophoretic profiles showed that the β-conglycinin α and ά subunits of 25 (Figure 1, B), 50 (Figure 1, C) and 75 kGy gamma irradiated SBM were degraded after 4, 8 and 12 h of incubation respectively, and the β subunit of this protein was resistant until 24 h of incubation. The basic and acidic subunits of glycinin of were not completely degraded after 48 h of incubation. Gamma irradiation decreased protein degradability through denaturing, increasing surface hydrophobicity, aggregation and cross linking of proteins. Gamma irradiation of 25 and 50 kGy increased, but 75 kGy decreased, mobile nylon bag digestibility of CP.

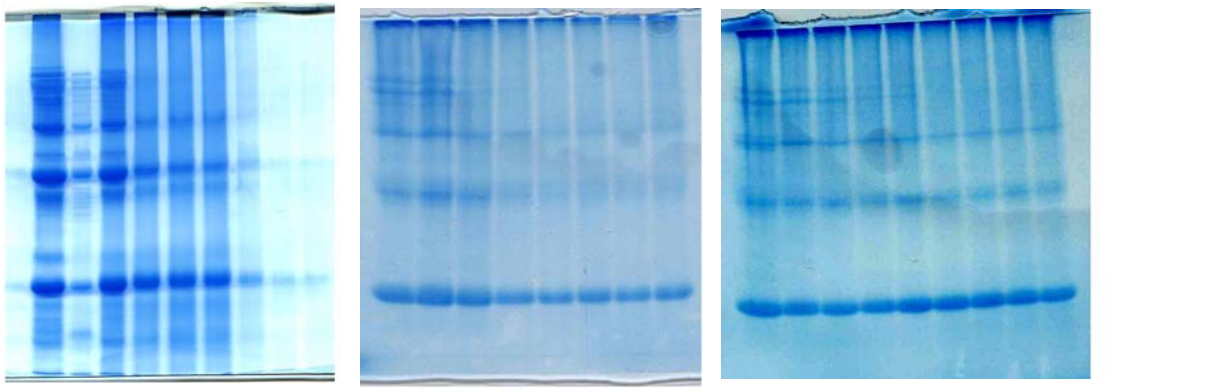

Figure 1 SDS-PAGE analysis of untreated (A), 25 kGy (B) and 50 kGy (C) gamma irradiated SBM incubated in the rumen (W is water soluble protein fraction; Each line was for the hours of incubation).

Conclusion Results of this study showed that gamma irradiation could decrease protein degradability in the rumen. Dose level of 50 kGy had greater potential to increase rumen undegradable protein, with positive effect on CP digestibility, than 25 and 75 kGy.

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Biological activity of tannins extracts from four tropical forages

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Introduction Sheep and goats browse c. 260 of leguminous species in Yucatán, Mexico (Flores Guido *et al*., 2003). Forage trees are a feed resource around the world. However its use is currently limited due to their content of antinutritional factors such as tannins and scarce data on their biological activity. The objective of the present work was to determine the biological activity (protein binding capacity) of condensed tannins extract from *Leucaena leucocephala*, *Acacia pennatula*, *Lysiloma latisiliquum* and *Piscidia piscipula*.

Material and methods Foliage (500g FM) from each specie was collected during the summer of 2005. Tannins were extracted 1 L acetone:water (70:30) adding 1 g/L ascorbic acid to avoid oxidation. Acetone was evaporated at 60 ºC and the remaining extract filtered (Filter paper) and mixed with methilene chloride (1:1) to take away fat and pigments. The remaining aqueous fraction was used in the biological activity studies. Tannins content of extracts was quantify as described by Price *et al*. (1978). Biological activity was determined by a radial diffusion method; briefly; agar was prepared with 1% agarose in acetate buffer, bovine haemoglobine, and chloramphenicol, pH was adjusted to 5.0. Twenty five ml were placed on Petri dishes (10 cm diameter). On each Petri dish 5 wells (4mm diameter each) were done in the agar. The outer wells were used to place 15 ul of each individual aqueous extract and the centre well to place resorcinol as standard. Samples were incubated 24 h at 25 ºC. The diameter of radial diffusion was measured with a digital caliper and activities expressed as area relative to the standard (Reyes, 1993). Each extract was assayed with 12 replicate. Data was analized as a complete randomized design.

Results Table 1 shows the concentration of tannins in both fresh and dry matter basis. Due to the large variation of tannin extracted (from 2.94 to 16.62 % DM) the biological activity was converted to activity/mg extracted tannins prior to analysis. Biological activity is expressed relative to resorcinol.

Table 1 Condensed tannins (CT) and biological activity of aqueous extract of four forage tree species.

	CT (%)		Biological
	FM	DM	Activity*
L. leucocephala	4.29	13.55	3.078[a]
A. pennatula	7.52	16.62	3.264[a]
L. latisiliquum	2.32	5.13	2.504b
P. piscipula	0.74	1.94	2.622b
SEM			0.860

* Activity per mg CT unit and expressed relative to resorcinol,
Values with different literal differ at 0.05. FM: Fresh matter, DM: Dry matter

According to Perevolotsky *et al*., (2003) classification *P. piscipula* can be considered a plant with low tannin content (<5% DM), *L. leucocephala* and *L. latisiliquum* have moderate tannin content (5-10% DM) and *A. pennatula* has high tannin content (15-20% DM). However, tannin concentration can not be consider an indicator of their activity as *L. leucocephala* and *A. pennatula* tannins had similar high activity (Table 1) while *L. latisiliquum* and *P. piscipula* were also similar. Paolini and Hoste (2003) have reviewed the potential of tannins to control internal parasites in ruminants and found that CT extract can inhibit motility and migration of larvaes. There are limited information on the relationship among biological activity of CT and its potential to control internal parasites, while considering at the same time the antinutritional effects of their tannins. This is currently being investigated in our lab.

Conclusión Both *L. leucocephala* and *A. pennatula* have high biological activity and are suitable candidates to further study mode of action of its tannins.

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Application of tracer methodology to measure nitrogen kinetics in the rumen in cattle

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Introduction The efficiency of use of dietary nitrogen (N) in cattle is influenced by the rate of degradation of dietary protein to peptides, amino acids and ammonia and the concomitant rate of assimilation of these nitrogenous products by rumen micro-organisms for their protein synthesis. The latter depends heavily on fermentable energy availability (FME). Ammonia that is not assimilated is absorbed across the rumen wall and is mostly excreted. This excretion represents inefficient use of dietary N. The stable isotope ^{15}N provides a useful means of making quantitative estimates of N utilisation in ruminants. However, owing to technical challenges associated with implementation of tracer dilution methods, measurement of ^{15}N content in biological materials and cost of ^{15}N-labelled materials, there have been relatively few studies using ^{15}N in larger animals. Our objective was to develop and evaluate tracer dilution methods using ^{15}N that would be appropriate for studying the efficiency of use of the N in perennial ryegrass silages when used for feeding cattle.

Materials and methods Within a larger digestion study, one Holstein-Friesian steer (live weight 444 kg) equipped with rumen and duodenal cannulae was used to evaluate the methodology. A first-cut perennial silage supplemented with sucrose (40 g/kg dry matter intake) was offered *ad libitum* but given in equal portions each hour to promote metabolic steady state. A solution containing 1.91 g/L of $(^{15}NH_4)_2SO_4$ (99.3 mol %, CK Gas products Ltd., UK) in distilled water was administered into the rumen in two 500 ml portions (i.e. 20 mmol ^{15}N) over about 3 min, after which filtered rumen fluid (RF) was collected at increasing intervals for about 24 h. RF samples were centrifuged at 20,000 g (25 min, 4°C) to give pure fluid-phase bacteria which were washed free of ammonia by re-suspension in water and re-centrifugation. A sub-sample of the supernatant (8 ml) was mixed with 0.8 ml 65 % (w/v) TCA to precipitate soluble protein. Ammonia-N was isolated from a second sub-sample of supernatant by diffusion at room temperature. ^{15}N enrichments of RF ammonia, bacteria and soluble protein determined by combustion (Dumas) and mass spectrometry were modelled relative to time after ^{15}N administration, using a double exponential model. Flux rates, compartment sizes and fractions of N in secondary compartments arising from ammonia-N were determined as described by Nolan and Leng (1974).

Results The silage contained 228 g DM/kg and 17.9 g total-N/ kg DM. Intake of silage DM was 7.51 kg and that of dietary FME was estimated as 64.2 MJ/d. Mean RF ammonia concentration was 69 mg N/L and the compartment size, estimated by ^{15}N dilution was 5.47 g N; RF volume was therefore 79 L. The estimated total flux of ammonia through the rumen fluid was 200 g N/d whereas net flux (irreversible loss) was 127 g N/d, indicating that there was recycling of ammonia-N through non-ammonia-N compartments before its return to RF ammonia. 0.87 of the N in fluid phase bacteria was derived from RF ammonia and the remainder (0.13) was apparently derived from peptides or amino acids, or both. Labelled protein was synthesised, presumably by rumen micro-organisms, and secreted into RF; some of this may have been microbial enzymes. Only 0.45 of the N in the soluble protein in RF, however, was derived from ammonia, indicating soluble protein from dietary or endogenous sources was also present. If it is assumed (AFRC, 1993) that the bacterial-N enrichment was representative of all rumen micro-organisms and that 1.57 g microbial N (=9.8 g crude protein) was formed per MJ of FME (i.e. total 101 g microbial N/d of which 0.87 was derived from ammonia), then 88 g N/d of RF-ammonia was assimilated by rumen micro-organisms for this synthesis. The majority of the remaining ammonia irreversibly lost from the RF compartment (39 g N/d) was most likely absorbed across the rumen wall, converted to urea and excreted in urine.

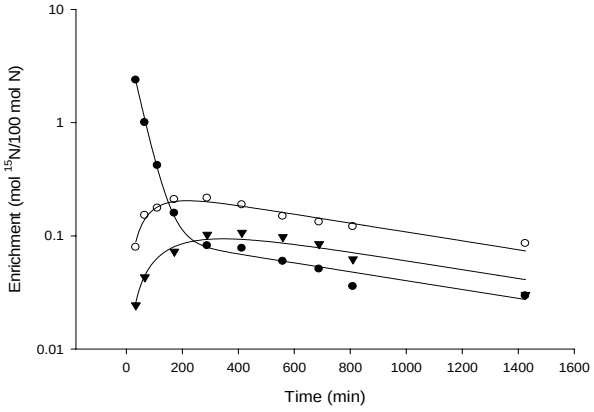

Figure 1 ^{15}N enrichment after a single intraruminal injection of 20 mmol $(^{15}NH_4)_2SO_4$. Terminal slopes constrained to fitted bacterial slope, i.e. 0.0009/min.

● Ammonia
$y = 5.390e^{-0.0259x} + 0.0990e^{-0.0009x}$, $r^2=0.996$, rsd=0.0474

○ Fluid-phase bacteria
$y = -0.2665e^{-0.0132x} + 0.2665e^{-0.0009x}$, $r^2=0.949$, rsd=0.0361

▼Soluble protein
$y = -0.1484e^{-0.0067x} + 0.1484e^{-0.0009x}$, $r^2=0.927$, rsd=0.0458

Conclusions The techniques used in this study can potentially be used to make quantitative measurements of N transactions in cattle given silage-based diets. In the steer on the diet used in this study, most dietary protein passed through the RF ammonia pool and, probably because of the additional FME from sucrose, the majority of this ammonia was used for rumen microbial protein synthesis rather than being absorbed and excreted.

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Liquid, concentrate and forage total tract mean retention times estimated using two algebraic methods in ponies given either oat straw or grass haylage as a basal forage

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Introduction Previous work (Hyslop, 2005) has shown that alternative algebraic calculation methods give similar estimates of total tract mean retention times (TMRT) in ponies given complete pelleted diets. This similarity in TMRT estimates across calculation methods remains to be confirmed with other dietary constituents. This study's objective was to compare liquid, concentrate and forage TMRT estimates using alternative algebraic calculation methods.

Materials and methods 6 mature Welsh-cross pony geldings (mean LW 298 kg) were individually housed and used in an eight treatment, 6 x 4 partially balanced incomplete block design experiment consisting of four 21 day periods. Either oat straw (DM: 831 g/kg, CP: 22 g/kg DM, NDF: 884 g/kg DM - treatments 1-4) or grass haylage (DM: 583 g/kg, CP: 67 g/kg DM, NDF: 732 g/kg DM - treatments 5-8) were offered at either 3 kg dry matter (DM) per day (treatements 1 and 5) or 1.5 kg DM/day (treatments 2-4 and 6-8) as the basal forages. The concentrate portion of the diets (DM: 879 g/kg, CP: 160 g/kg DM, NDF: 127 g/kg DM) comprised 0.8 micronised wheat and 0.2 mineralised, high protein supplement. Concentrate was either not fed (O) or offered at 1.5 kg DM/day 2 hours before (B), with (W) or 2 hours after (A) the forage portion of diets 2-4 and 6-8 respectively. Meals of either concentrate or forage were offered at either 08:00 or 10:00 hours and at either 15:00 or 17:00 hours. Each 21 day period consisted of a 16 day adaptation and a 5 day recording phase with dietary liquid, concentrate and forage digesta TMRTs determined for each pony during the 5-day recording period following a pulse dose of each marker. 100 ml of Cobalt EDTA (Co) was used as a liquid passage marker whilst Dysprosium (Dy) labelled samples of concentrate (75 g) and Ytterbium (Yb) labelled samples of forage (100 g) were used as either concentrate or forage particulate digesta passage markers respectively. Faceal samples were collected throughout the 5 day recording period for subsequent marker analysis by either atomic absorption spectrophotometry (Co) or atomic emission spectrophotometry (Dy and Yb). TMRT (hours) was calculated using 2 algebraic methods as follows:- (1) TMRT = ΣtiMi where ti = time post dose (mid-point between faecal samples) and Mi = amount of marker excreted as a proportion of total marker excreted (Faichney, 1975): (2) TMRT = ΣtiCiΔti / ΣCiΔti where ti is as above, Δti is the difference in time between collection of successive samples and Ci = marker concentration in each sample (Theilmans *et al*, 1978). Regression analysis was used to estimate relationships between the two algebraic TMRT calculation methods using Genstat 5.

Results Average TMRT (h) – Faichney and TMRT (h) – Theilmans across all diets were 35.2 and 35.5 (s.e.d. 7.31), 43.0 and 43.0 (s.e.d. 7.74) and 40.4 and 40.8 (s.e.d. 8.74) for the liquid (Co), concentrate (Dy) and forage (Yb) markers respectively. The regression relationships between the 2 algebraic calculation methods for the liquid (Co), concentrate (Dy) and forage (Yb) markers are also shown in Figure 1.

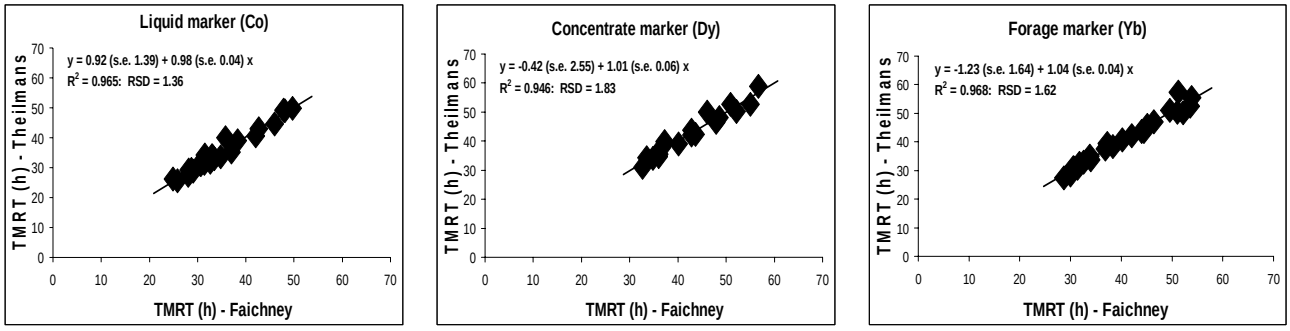

Figure 1. TMRT comparisons across calculation methods for liquid, concentrate and forage dietary constituents.

Conclusions Results confirm earlier work (Hyslop, 2005) and illustrate that both algebraic calculation methods yield similar estimates of TMRT with liquid, concentrate and forage dietary constituents. The Theilmans calculation method allows only spot feacal samples, rather than total faecal collections to be taken where estimates of TMRT are required.

Acknowledgements This work was funded by Dodson & Horrell Ltd.

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Chemical composition, dry matter and crude protein degradability coefficients of various whole crop cereals

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Introduction There is an increased range of alternative forage available to Iranian dairy farmers. Improvements in breading, growing, harvesting and storage technology in this area have helped the expansion of alternative to whole crop cereals. Whole crop cereal silages are very flexible, allowing the decision to have the crop ensiled as fermented whole crop. Incubation of feeds in nylon bags in the rumen of fistulated ruminants have been used to evaluate the extent of digestion. The objective of the present study was the determine chemical composition, dry matter and crude protein degradability coefficients of whole barley, triticale and maize silages treated with urea.

Materials and methods Triticale, barley and maize whole crop were harvested at late growing (about 33% DM triticale and barley, and 25% DM of maize whole crop) chopped, then ensiled with urea (5 g/kg DM) for 40 days. Standard procedures were used to determine the chemical composition of the samples. PH was determined in the silage extraction. CP was determined in dry samples. The ruminal disappearances of dry matter and crud protein were determined using the nylon bag technique. Tow Holstein steer fitted with the rumen fistula were used in the present study. The bags (10×12 cm) were made of artificial silk cloth with a pore size of 46 µm. About 5 g DM of the samples was placed in each bag, and four bags for each treatment were incubated for each time (0.0, 2, 4, 8, 16, 24, 48, 72, 96 h). Bags were washed in cold running water and dried in oven (60 °C, for 48 h), then weighted to determine dry matter disappearance. The equation developed by Ørskov and MacDonald (1979) was used; $P=a+b(1-e^{-ct})$ where p is the potential of degradability of DM and CP at time t, a is the quickly degradable fraction of DM and CP, b is the slowly degradable fraction of DM and CP, c is the constant fractional rate of degradation and t is the time of incubation (h). Data were analyzed using the GLM procedure of SAS. Statistical significance effects were determined at $P < 0.05$.

Results Chemical composition of whole crop triticale, barley and maize silage are shown in Table 1. The data related to the degradability coefficients of DM and protein using *in situ* procedure are shown in Table 2. The concentration of CP in whole cop triticale was significantly higher than the other silages ($P < 0.05$). Significant differences in concentration of NDF were observed among the silages ($P < 0.05$).

Table 1 Chemical composition of whole crop triticale, barley and maize silages treated with urea.

Item	Triticale silage	Barley silage	Maize silage	SEM	*P* - value
PH	5.54^{a}	5.58^{a}	4.29^{b}	0.068	*
CP (g/kg DM)	112.4^{a}	96.4^{b}	67.2^{c}	0.054	*
N-NH_3 (ml/dl)	16.85^{a}	15.04^{b}	6.48^{c}	0.310	*
NDF (g/kg DM)	498^{a}	470^{b}	436^{c}	5.40	*

^{a,b,C}Means with different superscript letters in the same row differ ($p < 0.001$)

Table 2 Dry matter and protein disappearance of whole crop triticale, barley and maize silages treated with urea from nylon bags during incubation in the rumen.

Item		Triticale Silage	Barley Silage	Maize silage
DM (Fraction %)*				
	a	0.37 ± 0.01	0.33 ± 0.01	0.24 ± 0.01
	b	0.44 ± 0.02	0.52 ± 0.04	0.54 ± 0.02
	c	0.03 ± 0.01	0.02 ± 0.05	0.03 ± 0.01
CP (Fraction %)*				
	a	0.82 ± 0.01	0.80 ± 0.01	0.65 ± 0.01
	b	0.09 ± 0.01	0.11 ± 0.02	0.19 ± 0.04
	c	0.02 ± 0.01	0.02 ± 0.01	0.02 ± 0.03

*parameters ± SE

Conclusions PH of triticale and barley whole crop silages were higher than that of the maize silages ($P < 0.05$). This finding is supported by the study of Golmahi *et al.* (2005), which reported that the pH of triticale and barley silages were higher than that of maize silage. The triticale silage contained the higher soluble Dry matter and crude protein, barley silage was intermediate for DM and CP and maize silage contained the lowest soluble DM and CP contents. This result are in agreements with those of Khorasani *et al.* (1993), who reported that barley and triticale whole crop silage usually contains more soluble DM and CP compared with the other cereal silages.

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The concentration of oxygen and its depletion in bovine grass-boluses

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Introduction The enzyme polyphenol oxidase (PPO) may be responsible for increases in both dietary-nitrogen utilisation and the concentration of polyunsaturated fatty acids in ruminant products when animals eat red clover (Lee *et al*. 2004). The enzymatic reaction requires the presence of oxygen and therefore is assumed to be inhibited in the anaerobic rumen. However, little information exists as to the concentration of oxygen in boluses and whether this could sustain the aerobic enzymes activity in the reticulo-rumen. This study investigated the concentration of oxygen in grass-boluses measured *in vitro* or *in vivo* in the rumen of two rumen fistulated cows.

Materials and methods The rumen of a Holstein × Friesian dairy cow fitted with a rumen cannula was emptied. The cow was then offered freshly cut perennial ryegrass (PRG) and the freshly ingested bolus caught at the oesophageal orifice in the reticulo-rumen. This was immediately transferred to an air-tight jar filled with de-oxygenated water maintained at 39^{o}C. A dissolved oxygen probe (Orion Research Inc, MA, USA, calibrated according to the manufacturer's instructions) was inserted into the bolus and the oxygen concentration measured at 30 second intervals over a time course of ten minutes. This procedure was repeated twice more with PRG and the procedure repeated with freshly cut timothy (n=3) these were then analysed using a repeated measurement ANOVA (Genstat 8.1, Lawes, Agricultural Trust, 2005). A second set of PRG boluses (n=3) were then incubated in an air-tight jar filled with strained rumen fluid and the oxygen concentration measured as before. *In vivo* measurements were then taken using two rumen fistulated dairy cows. The oxygen probe was positioned inside the reticulo-rumen by the oesophageal orifice and a reading taken as the initial oxygen concentration. The animals were then allowed to eat for one mouthful before the grass was removed. The time was recorded between swallowing and observing the oxygen peak as the bolus entered the rumen. When a stable oxygen reading was observed this was recorded as the initial value and the animal offered the grass once again for one more mouthful. This was repeated several times (cow 1, n=4; cow 2, n=10).

Results Fig. 1. shows the oxygen depletion from the two grass boluses incubated in de-oxygenated water. Timothy contained significantly ($P<0.05$) more oxygen than PRG although both still contained trace levels after 10 minutes. When the grass bolus was immersed in strained rumen fluid the oxygen disappeared within 2 to 3 minutes (Fig. 2). In the *in vivo* study there were no significant differences between cows, the initial concentration of oxygen in the reticulo-rumen was 0.01 (±0.008) mg O_2/l. The entrance of a bolus into the rumen caused a peak in oxygen concentration of 0.95 (±0.600) mg O_2/l, 6.4 (±1.53) seconds after swallowing, returning to the initial oxygen concentration within ten seconds.

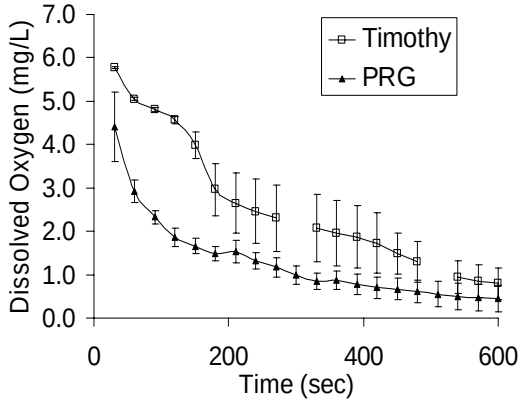

Figure 1 Dissolved oxygen content of grass boluses incubated in water at 39^{o}C n=3 (± SEM)

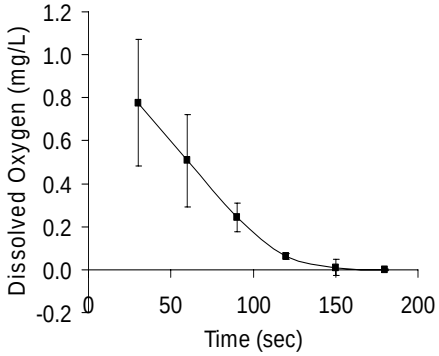

Figure 2 Dissolved oxygen content of a grass bolus incubated in rumen liquor at 39^{o}C n=3 (± SEM)

Conclusions The higher oxygen content of the timothy boluses may be due to the coarser nature of the grass possibly resulting in greater mastication. Although the bolus alone resulted in oxygen depletion, it appears to be largely due to the oxygen scavenging nature of the microbes in the rumen fluid as oxygen disappeared from the boluses immersed in rumen fluid within 2-3 min but some oxygen was still present after 10 min when immersed in de-oxygenated water. Given these results it is apparent that any occurrence of PPO activity during grazing of red clover would be largely confined to the period of mastication and that the level of oxygen brought in from the boluses would be rapidly scavenged.

Acknowledgement Department for Environment Food and Rural Affairs for funding the research.

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Chemical composition and *in situ* dry matter degradability of Sugar cane pith treated with steam at high pressures

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Introduction The use of by-product in animal nutrition is necessity since it may increase the availability of feed for animal and avoid accumulation that contributes to environmental problems. Sugar cane bagasse and pith, by- product which leave the last processing stage of the sugar mill (bagasse) and passed through rotary sieves to separate fine particle (pith), are the most abundant by-product in Iran. The low digestibility, high lignin and very low content, are considered the main reasons for unsatisfactory performance of animals fed this roughage. Steam-pressure treatment cleavage the bounds between lignin and the other component of the cell wall, in order to improve it's degradability by enzymes of the rumen microbial ecosystem. The objective of this work was to evaluate the chemical composition and *in situ* degradability of sugar cane pith treated by steam at high pressures.

Materials and methods The sugar cane pith, untreated (UTP) and steam treated (STP) were prepared at 20 bar for 3 min. STP dry matter (DM) was determined using air-forced oven (60°C,48h) and chemical composition including: crud protein (CP), neutral detergent fibre (NDF), acid detergent fibre (ADF), Ash and total soluble sugar were determined using standard procedures (AOAC.1990). DM degradability of the silages was measured by *in situ* technique using two fistulated Holstein steers (400±12 kg). The animals fed a 40:60 concentrate: forage diet. The experimental samples were milled (2 mm screen) and weighed (5 g DM) into four bags (12x19 cm, 52µm pore size), made of polyester cloth, for each incubation time. The bags were incubated in the rumen for 2, 4, 8, 16, 24, 48, 72 and 96 h. Four bags also were washed with cold tap water to estimate the wash-out at zero time. After each incubation time, the removal bags were hand-washed with cold tap water, and then dried in a forced-air oven (60° C, 48 h). The degradable coefficients of DM was determined using the equation of $P=a + b (1 - e^{-ct})$.

Results Chemical composition of STP and UTP is shown in table 1. Ruminal degradation coefficients (a, b, c) of dry matter of both STP and UTP are summarized in Table 2. Treatment with high pressure-steam significantly reduced NDF of sugarcane pith and increased total soluble sugar (TSS), indicative of hemicellulose hydrolysis during steam treatment, but not affected ADF. Others indicated that steam-pressure treatment reduced NDF, but no ADF (Pate, 1982). Ruminal DM degradable coefficients were influenced by the steam treatment.

Table 1 Chemical composition of sugar cane pith treated by steam at high pressures

Chemical composition	Treatment		SEM
	STP	UTP	
NDF(g/kg)	617.5	770	0.88
ADF(g/kg)	505	525	1.76
CP(g/kg)	23	21	0.66
Ash(g/kg)	145	110	1.76
TSS	123.7	20	1.75

Table 2 I*n situ* DM (mean) rumen degradation coefficients (a, b and c) of sugar cane pith treated by steam at high pressures± SE

Treatments	DM			PD	ERD		
	a	b	c		k=0.08	k=0.05	k=0.02
STP	0.174± 0.02^{a}	0.712±0.21^{a}	0.012±0.006^{a}	88.6	0.271	0.315	0.444
UTP	0.05±0.04^{b}	0.542±0.06^{a}	0.030±0.01^{b}	59.2	0.199	0.255	0.377

Values with different superscripts within columns differ at P<0.05

a is the rapidly degradable fraction, *b* is the slowly degradable fraction and c is the fractional digestion rate constant (h^{-1}) of the fraction *b***,** PD is the potential degradability, ERD is the effective degradability, *k* is out flow rate

Conclusions The results of the present study demonstrate that steam treatment had significant effect on reduction of NDF, and increased rapidly (a) and decreased fractional digestion rate (c). But had no significant effect the potentially (b) degradable fraction of DM and ADF of sugar cane pith. Therefore steam-pressure treatment improved degradation and nutritional value of pith for ruminants.

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The nitrogen fractions of selected feeds in ruminant nutrition

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Introduction There is increasing interest in the Cornell Net Carbohydrate and Protein System (CNCPS) for rationing cattle. Crude and digestible protein determinations do not completely account for the dynamics of ruminal fermentation and potential loss of nitrogen as ammonia (Sniffen *et al* 1992). When measured data on the protein and carbohydrate contents of feeds were used, the predictions of the performance of dairy, beef and dual-purpose cattle were more accurate than when tabular values were used (Fox *et al* 2004). To obtain the best predictions, the CNCPS model requires an accurate description of the carbohydrate and protein fractions and the unique rates of digestion and passage of individual feed fractions that are being fed. This information can be used as a basis for amino acids absorption. The CNCPS use in Iran is limited because the inputs data is not determined. The present work was carried out to determine nitrogen fractions of some feeds are using in ruminant nutrition.

Materials and methods In this work, some of ruminant feeds were selected. These were alfalfa (AL), corn silage (CS), wheat straw (WS), wheat bran (WB), barley (B), soybean meal (SBM), beet pulp (BP), commercial fish meal (FM), whole cottonseed (WCS), and mechanical cottonseed meals. The feed samples were collected during the harvesting period. All samples were analysed at the animal nutrition laboratory of Zanjan University. Analysis were carried out by standardized procedures for nitrogen fractionation of ruminant feeds (Licitra *et al*. 1996). Crude protein is partitioned into five fractions. Fraction A is non protein nitrogen (NPN), which is trichloroacetic (TCA) acid soluble nitrogen. Unavailable or protein bound to cell wall (fraction C) is acid detergent insoluble nitrogen (ADIP), and slowly degraded true protein (fraction B3) is neutral detergent insoluble nitrogen (NDIP) minus fraction C. Rapidly degraded true protein (fraction B1) is tungstic acid or TCA percipitable protein from the buffer-soluble protein minus NPN. True protein with an intermediate degradation rate (fraction B2) is the remaining nitrogen. The sequential analysis of neutral detergent fibre and acid detergent fibre used to determine NDIP and ADIP. Fraction B1 is precipitated with tungstic acid.

Results The protein fractions of selected feeds are shown in table 1. These fractions were A, B1, B2, B3, and C. All fractions expressed as proportion of CP. The AL and CS have half of their protein as NPN, with the majority of the remainder associated with the neutral detergent soluble protein fraction. The fraction A in by product feeds were more than 300g/kgDM. The fraction C in AL, CS, BP, and CSMs were more than 100g/kgDM.

Table 1 Nitrogen fractions of selected feeds (g/kgCP)

Fraction	AL	CS	WS	WB	B	SBM	BP	FM	WCS	CSM1	CSM2	CSM3
CP(g/kgDM)	150	75.0	41.0	145	125	450	90.0	650	220	285	310	254
A	550	472	320	350	99.0	40.0	350	140	15.0	170	154	119
B1	35.0	24.0	70.0	66.0	148	175	30.0	160	410	200	220	180
B2	171	255	430	463	653	727	100	280	495	402	388	402
B3	100	110	100	150	60.0	80.0	350	400	5.0	100	120	170
C	140	112	80.0	20.0	40.0	50.0	150	20.0	90.0	127	117	127

Conclusions The protein fractions in analysed feeds revealed that AL, CS, WB, and BP had a high NPN. The whole cottonseed had large amounts of the soluble protein (B1). The cottonseed meals, soybean meal, barley and wheat bran had higher proportions of the fraction B3. The cottonseed meals analysis indicated that these feeds had a higher proportion of neutral detergent fibre (310-400g/kgDM) than the CNCPS tabular values and were affected by variable mechanical and heat processing. Then these feeds have a higher proportion of acid detergent insoluble protein. The AL and CS may be affected by dehydration or drying and then these feed had a higher proportion of fraction C. It should be noticed that this research has done on few feeds, so for comparing with CNCPS, it should be collected a wider range of samples in several seasons, years and different area to establish a feed composition database as inputs the CNCPS model.

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The influence of pH on the degradation characteristics of forages with different structural composition assessed *in vitro*

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Introduction In addition to environmental and management factors, the degradation characteristics of forages are greatly influenced by both the structure and composition of cells within that structure. In particular, and even when grown under the same environmental conditions, marked difference exists between the tissue composition of C_3 (temperate) and C_4 (tropical) grasses. This differentiation, a reflection of their origin, is based on the photosynthetic pathways with C_3 and C_4 grasses using phosphoglycerate and oxaloacetate, respectively as intermediates. C_3 grasses tend to have a lower cell wall content which is usually restricted to the stem component and a greater proportion of highly degradable mesophyll tissue. The possibility that C_3 and C_4 grasses exhibit different degradation characteristics depending on the rumen fermentative environment therefore exists. This is of major importance when it is considered that about 0.15m ha forage maize, a C_4 grass, are grown annually in the UK. This study compared the chemical composition and *in vitro* degradative characteristics of three C_3 forages, viz; grass hay (GH), wheat straw (WS) and rice straw (RS) and maize stover (MS) as influenced by incubation medium pH using a 4 x 3 factorial design. .

Materials and methods All four substrates were pre-dried (60°C) then milled to pass a 2 mm screen. Organic matter content was calculated by difference following ashing at 500°C overnight and NDF, ADF and lignin contents were estimated using procedures based on Goering and Van Soest (1970). The *in vitro* methodology (Mauricio *et al*., 1999) was used to determine both degradation and fermentation kinetics of each substrate as influenced by pH of the incubation medium. This was altered by the addition of a 1:1:1 mixture of 1M hydrochloric, phosphoric and sulphuric acids, to produce initial pH levels of 7.2 (untreated), 6.6 and 6.4. Fermentation headspace gas pressure was measured at intervals over a 48 h period while sets of flasks were withdrawn at 12, 24 and 48 h post-inoculation and fermentation medium pH measured. Residues were recovered by filtration through Gooch crucibles and analysed to estimate dry matter and NDF degradation.

Results RS had the highest ash content (132 g kg^{-1} DM) more than twice that of the other substrates, while RS and WS had the greatest lignin contents (119 and 116 g kg^{-1} DM, respectively). Although cell wall content (as estimated by NDF) of MS was higher than GH (706 and 646 g kg^{-1} DM), respectively) both were lower than either RS or WS (743 and 806g kg^{-1} DM, respectively). The hemicellulose to cellulose ratio showed considerable variation with values of 1.35, 1.55, 1.63 and 1.72 for MS, GH, WS and RS, respectively. Although initial pH values were insufficient to depress degradation, the subsequent accumulation of fermentation acids as a result of degradation caused pH to decline over time, with fermentation medium pH depressed to the greatest extent with MS and least with GH. Overall RS was the most poorly degraded substrate (419 g kg^{-1} DM after 48 h incubation) and this was depressed to 343 g kg^{-1} with the most acidic incubation medium, although actual pH only decreased to 6.20. In contrast DM degradability of MS (552 g kg^{-1}, untreated medium) was only depressed to 497 g kg^{-1} despite pH declining to 6.01 as a result of fermentation acid accumulation.

Table 1 *In vitro* 48 h degradation characteristics as influenced by initial fermentation medium pH

Initial	Medium pH				Gas release (ml)				NDF degradability (g kg^{-1})			
pH	GH	MS	RS	WS	GH	MS	RS	WS	GH	MS	RS	WS
7.2	6.78^{a}	6.61^{a}	6.66^{a}	6.59^{a}	179^{a}	197^{a}	146^{a}	173^{a}	465^{a}	576^{a}	521^{a}	572^{a}
6.6	6.41^{b}	6.21^{b}	6.39^{b}	6.30^{b}	167^{b}	173^{b}	132^{b}	151^{b}	407^{b}	546^{ab}	443^{b}	478^{b}
6.4	6.27^{b}	6.01^{b}	6.20^{b}	6.12^{c}	155^{c}	161^{c}	121^{c}	134^{c}	338^{c}	533^{b}	377^{c}	457^{b}

Values in columns without common superscripts are significantly different (P>0.05)

Conclusions These results suggest that even a minor decline in pH will inhibit cell wall degradation. However it has to be remembered that this depression is continuous, in contrast to the marked diurnal variation normally associated with rumen fermentation. The results also suggest that with poor quality substrates, degradation is inhibited at a higher incubation pH than with more readily degraded material (pH x NDF degradation interaction, P>0.001). Thus it would appear that MS degradation can occur at a lower pH which may, in part, explain the observation that intakes of high-producing dairy cows offered maize silage-based TMRs are maintained even when rumen fluid pH is depressed below 6.0 for considerable periods each day.

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Biological activity of tannins of Brazilian browses using semi-automated gas production technique. 1. Plants from Bahia state

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Introduction Brazil has arid regions where livestock production is limited by forage source. However, some native herbaceous browses have a dry tolerance and have been used as animal feeds. Some of those plants have anti nutritional compounds such as tannins that can interfere on intake and digestibility. Tannins have a high affinity to proteins and could make these molecules unavailable for animal. Compounds as polyethylene glycol (PEG) has been used on tannin studies, because it has more affinity with tannins than proteins. Based on that, it is possible to evaluate the nutritive potential of tanniniferous plants, using PEG in gas based techniques for assessing anti nutritional factors in tanniniferous plants for ruminants. The aim of this work was to investigate the biological activity of tannins using the *in vitro* gas method with the addition of polyethylene glycol (PEG).

Material and methods Five browse species with potential as forage were selected: Catingueira (*Caesalpinia bracteosa*), Jurema preta (*Mimosa hostilis*), Leucaena (*Leucaena leucocephala*), Malva branca (*Sida cordifolia*.) and Maniçoba (*Manihot pseudoglaziovii*). Samples (leaves and branches with 5 mm or less of diameter) were collected during the wet season (April-2003) with three replicates for each species. Total phenols (TP) were determined using Folin-Ciocalteu reagents, total tannins (TT) as the difference of phenolics before and after tannin removal using the insoluble polyvinyl polypyrrolidone (PVPP) and condensed tannins (CT) were analyzed as described by Makkar (2000). The *in vitro* gas production assay was conducted according to Mauricio *et al*. (1999). Four sheep with rumen cannulas were used as inoculum donors. Inoculum was prepared with 50% liquid and 50% particulate ruminal liquor (Bueno *et al*., 2005). About 1 g of each sample was used in duplicate, with or without PEG (1:1). Gas production was recorded at 3, 6, 9, 12, 16 and 24 h., using a pressure transducer. Dry matter degradability was measured by filtering bottles content at 24 h. Means were compared by Tukey test and correlations were analysed (SAS, 2000).

Results The gas production (GP) from treated browses, after incubation with or without PEG for 24 h and chemical compositions of tested plants are presented on Table 1. Correlations between tannins and parameters studied are showed in Table 2. Although Malva and Jurema presented no differences for ADF content, Malva was more degradable than Jurema. This variation can be attributed the high values of tannins observed for Jurema. On the other hand, plants like Maniçoba, and Leucaena presented expressive concentration of condensed tannins, but had superior values of degradability and crude protein content.

Table 1 Means of gas production (ml/g DM), after incubation with (+) or (-) PEG for 24h and chemical analysis (g.kg^{-1} DM), total phenol (TP),total tannins (TT) (eq-g TA kg^{-1} DM) and condensed tannins (CT) (eq-g leucocyanidin kg^{-1} DM) of tested plants

Plants	CP	ADF	TP	TT	CT	Degrad	-PEG	+PEG	P^1
Catingueira	138.7^{c}	277.3^{a}	129.7^{a}	125.0^{a}	0.8^{c}	382.7^{b}	42.1	46.5	0.614
Jurema	142.0^{bc}	289.7^{a}	174.0^{a}	148.3^{a}	60.3^{a}	278.7^{c}	14.3	41.1	0.005
Leucaena	197.3^{a}	293.3^{a}	110.7^{a}	95.0^{a}	66.5^{a}	431.7^{b}	41.2	62.7	0.021
Malva	115.0^{c}	405.7^{a}	15.3^{c}	10.7^{c}	0.3^{c}	448.7^{ab}	65.8	68.2	0.786
Maniçoba	172.7^{ab}	356.3^{a}	50.3^{b}	40.3^{b}	20.1^{b}	521.7^{a}	71.7	89.9	0.046
SEM	6.6	28.8	11.4	10.8	7.9	16.8			

^{a,b} means with different superscripts, within column, are different (Tukey test, P<0.05)

[1] probability of the differences between values of gas volume with and without PEG (in the line).

Table 2 Correlations between tannins content and gas volume, increase of gas volume an chemical composition

Tannins	Gas 24h	Increase	Degrad	CP	NDF	ADF
Total phenol	-0.832 ***	0.635 *	-0.754 **	0.135 ns	-0.741 **	-0.695 **
Total tannins	-0.809 ***	0.594 *	-0.735 **	0.113 ns	-0.742 **	-0.725 **
Condensed tannins	-0.628 *	0.560 *	-0.409 ns	0.504 ns	-0.443 ns	-0.252 ns

ns: non significative (P>0.05); * P<0.05, ** P<0.01 ; *** P<0.001

Conclusions Browses investigated in this study presented low values of degradability due to the tannins or fiber content. More studies are important to investigate the action of tannins and the possibilities to diminish its effects.

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Biological activity of tannins of Brazilian browses using semi-automated gas production technique. 2. Plants from Ceará state ecosystem

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Introduction. Animal production in Brazilian semi-arid north-east is limited by dry-season feed shortage leading to high mortality and low production of the herds. Most of vegetation consists of forage plants with high protein content but low digestibility and high levels of tannins (10%). The aim of this work was to investigate the biological activity of tannins using the *in vitro* gas method with the addition of polyethylene glycol (PEG).

Material and methods Four browse species with potential for forage production were selected: Catingueira (*Caesalpinia bracteosa*), Jucazeiro (*Caesalpinia ferrea*), Jurema preta (*Mimosa hostilis*) and Sabia (*Mimosa caesalpinifolia*). Samples (leaves and branches with 5 mm or less of diameter) were collected during the wet season (April-2004) with three replicates for each species. The data were collected at the Centro Nacional de Pesquisa em Caprinos (Embrapa-CNPC), Sobral, Ceará, Brazil, in a representative area of the ecosystem of Caatinga. The climate of the area is characterized by a rainy period that extends from January to June (95% of the annual precipitation) and a dry period the rest of the year. The mean annual precipitation is of 790 mm. The temperature varies from 22ºC to 35ºC with an average of 28ºC. Total phenols (TP) were determined using Folin-Ciocalteu reagents, total tannins (TT) as the difference of phenolics before and after tannin removal using the insoluble polyvinyl polypyrrolidone (PVPP) and condensed tannins (CT) were analyzed as described by Makkar (2000). The *in vitro* gas production assay was conducted according to Mauricio *et al*. (1999). Four sheep with rumen cannulas were used as inoculum donors. Inoculum was prepared with 50% liquid and 50% particulate ruminal liquor (Bueno *et al.,* 2005). About 1 g of each sample was disposed in duplicate, with or without PEG (1:1). Gas production was recorded at 3, 6, 9, 12, 16 and 24 h, using a pressure transducer. Degradability was measured by filtering bottles content after 24 h of incubation. Means were compared by Tukey test and correlations were analysed using SAS system (SAS, 2000).

Results Table 1 shows chemical compositions of tested plants, the gas production (GP) and percent increase in GP from treated browses, after incubation with or without PEG for 24 h. Correlations between tannins concentration and gas volume, increase of gas volume, degradability, crude protein and fibre content are demonstrated in Table 2. Jucazeiro presented lower CT concentration and high value of DM degradability. Sabia presented high CT concentration and its protein content probability is not available, causing low values of DM degradability. Catingueira was the browse that produced highest microbial inhibition. Correlations between tannins, gas production and DM degradability had a significance results only for condensed tannins.

Table 1 Means of gas production (ml/g DM), after incubation with (+) or (-) PEG for 24h and chemical analysis (g.kg^{-1} DM), total phenol (TP), tannins (TT) (eq-g TA kg^{-1} DM) and condensed tannins (CT) (eq-g leucocyanidin kg^{-1} DM) of tested plants

Plants	CP	ADF	TP	TT	CT	Degrad	Gas volume		P^1
							-PEG	+PEG	
Catingueira	142.1^{a}	298.2^{c}	154.0^{b}	149.5^{ab}	4.2^{b}	387.5^{a}	45.9	55.0	0.056
Jucazeiro	152.0^{a}	352.5^{bc}	130.1^{b}	125.0^{ab}	1.3^{b}	445.0^{a}	24.0	37.4	0.008
Jurema	148.0^{a}	440.5^{ab}	283.5^{a}	217.7^{a}	94.5^{a}	301.5^{b}	14.0	55.1	<0.001
Sabiá	162.3^{a}	565.5^{a}	155.2^{b}	110.5^{b}	121.8^{a}	236.5^{b}	6.1	33.3	<0.001
SEM	11.4	29.6	21.9	19.2	16.3	16.1			

^{a,b} means with different superscripts, within column, are different (Tukey test, P<0.05)
^{1} probability of the differences between values of gas volume with and without PEG (in the line)

Table 2 Relationship (Pearson´s coefficient) between phenolic compounds and gas volume, increase of gas volume an chemical composition

Plants	Gas 24h	Increase	Degrad	CP	NDF	ADF
Total phenol	-0.306 ns	0.191 ns	-0.352 ns	-0.243 ns	0.034 ns	0.079 ns
Total tannins	-0.026 ns	-0.098 ns	-0.059 ns	-0.344 ns	-0.260 ns	-0.244 ns
Condensed tannins	-0.731 ns	0.676 *	-0.837 ***	0.027 ns	0.600 *	0.695 *

ns: non significative (P>0.05); * P<0.05, *** P<0.001

Conclusions Studied browses showed potential as ruminant feed although some of their fermentation characteristics were affected by tannins content.

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In vitro fermentation kinetics of a range of fresh leguminous forages, measured using equine faecal innocula

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Introduction Evolutionary diets composed solely of grass & hay are rarely able to provide domesticated equidae with the energy required for the athletic performance. Modern feeding practices have dictated that horses with a high-energy demand are fed cereal-based concentrate foods, often with minimal forage inclusion within the diet. Such high starch, low-fibre diets, when fed to excess, are recognised to induce metabolic disorders such as laminitis. Alternative forage sources capable of providing higher amounts of energy would be preferable in some cases, allowing the reliance on cereal diets to be reduced. It has been found that when fed to ruminant animals, ensiled red clover has a higher D-value and leads to an increase in both voluntary feed intake (VFI) & animal production, when compared to grass silage, (Freudenberger *et al*, 1994; Rutter *et al*, 1998). Recent studies have shown that when fed to ponies, red clover silage had a significantly higher apparent digestibility & VFI than either grass silage or hay (Hale and Moore-Colyer, 2001). The aim of this study was to investigate the potential benefits of several leguminous forages in their fresh, rather than conserved form, as suitable feeds for horses. Degradation of the fresh products was estimated using *in vitro* gas production techniques.

Materials and methods The extent & rate of degradation of the forages was measured using the gas production technique as described by Theodorou *et al* (1994). Substrates were tested either with, or without pepsin pre-treatment (substrates incubated at 40 g DM/l in pepsin HCL solution, 2 mg/ml pepsin in 0.075 M HCL, for 2 h at 39C & extensively rinsed prior to use). Substrates (400mg ground and freeze dried) were incubated in a faecal slurry (50 ml) at 39°C & gas production measured over a 96 h period using a gas transducer (Theodorou *et al*, 1994). Faecal slurries were prepared from faeces collected prior to the morning feed from five horses fed a mixed concentrated forage diet. The faeces were mixed with anaerobic buffer, as described by Theodorou *et al* (1994) (1:2 v/v) & strained through 4 layers of muslin prior to use. Each substrate was incubated, in duplicate, with the slurry from each horse (n=5). The resultant data were fitted to the model $p = a + b\ (1 - e^{-ct})$, where p = volume of gas after time t; a = the intercept of gas volume curve at t = 0; b = volume of gas produced at asymptote; c= rate constant of gas production (h^{-1}).

Results The extent of gas production was lower following pre-treatment with pepsin HCL in all cases except with white clover (Figure 1). The initial rate of gas production (c) was significantly higher for red clover, white clover and vetch ($P<0.05$) than all other substrates. However, the extent of gas production was significantly higher for pepsin treated white clover and perennial rye grass mix than all other substrates ($P<0.01$).

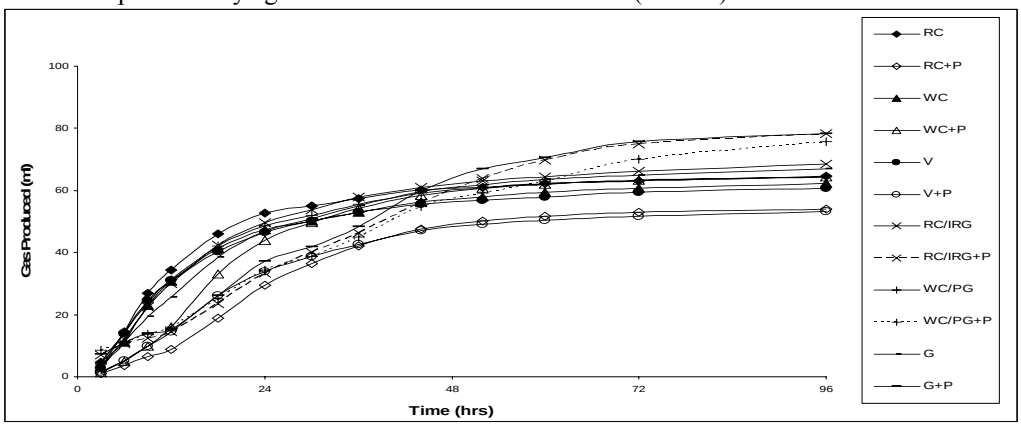

Figure 1 Gas production form red clover (RC), white clover (WC) vetch (V) red clover/Italian rye grass (RC/IRG), white clover/ perennial rye grass (WC/PRG) & perennial rye grass (G) both with & without pepsin treatment. (* $P<0.05$; ** $P<0.01$

Conclusions Although pre-treated white clover/perennial rye grass had an overall higher extent of gas production over 96 hours, it is more appropriate to consider rate of gas production as the mean retention time in the horse is around 36 hours. The rate of gas produced showed that RC, along with WC & V were significantly higher, thus indicating that RC is more easily degraded by horses than grass. If the amount of gas production at 36h is considered, both RC & RC/IRG are significantly higher than G. It is therefore possible to conclude that horses given access to either a ley with red clover inclusion, would benefit from a higher nutrient intake, than those grazing grass only, as red clover is degraded at a significantly higher rate. This is in keeping with published work considering *in vivo* digestibility of red clover silage.

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Methanogenesis and microbial variation evaluation by qPCR in *in vitro* gas production test of Lucerne or Tifton hays incubated with BES

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Introduction Ruminal fermentation produces methane (CH_4), which represents a loss of feed energy and a significant source of greenhouse gas. Researches have been searching for ways of inhibiting ruminal CH_4 yield and promising results have been achieved in *in vitro* assays (Hu *et al*., 2005). The *in vitro* gas production technique can predict the *in vivo* enteric CH_4 production from ruminants. Our research focused on the effect of 2-Bromoethanesulfonic acid (BES) on gas and CH_4 production and microbial variation of Lucerne and Tifton-85 hays fermented *in vitro*.

Materials and methods Commercial Lucerne (LUC) and Tifton-85 (TIF) hays were used as substrate in a complete factorial design (79.1 mg of 5 mM BES were added per 75 mL of fermentation fluid in each sample treated with BES). The *in vitro* gas production (GP) assay was carried out using a pressure transducer for measuring the gas produced in 160 mL serum bottles incubated at 39 ºC, using rumen fluid of five sheep as inoculum. *In vitro* gas production (GP) and methane (CH4) output were determined in each bottle incubated with 0.5 g of substrate plus 50 mL of Menke's buffered medium and 25 mL of inoculum (Longo *et al*., 2005). CH4 production was estimated (CH4 = [GP + HS] x Conc) following Tavendale *et al*. (2005). Incubation was stopped at 24 h incubation (cold water), and aliquots of the contents were collected for microbial evaluation by Real Time Polymerase Chain Reaction. The DNA was purified with glassmilk and adjusted to a concentration of 10 ng/µL (templates). 16S rDNA gene was amplified using primer pairs for broad rumen bacterial (Bacteria), broad rumen fungi (Fungi), broad rumen methanogen (Methanogen), Ruminococcus flavefaciens (Ruminococcus) and Fibrobacter succinogenes (Fibrobacter) in a real-time thermal cycler. Four templates were analyzed (triplicates) for each primer pair and in each run, non-template and inter run controls were analyzed. The thermal cycler was programmed with an initial denaturation step (3 min at 50 ºC) followed by 40 cycles of 15 s at 95 ºC (step 1) and 60 s at 60 ºC (step 2), with a final dissociation analysis (2 min at 95 ºC; 15 s at 60 ºC increasing to 95 ºC). The threshold cycles (Ct) was set to 0.300 and the comparative Delta-Delta Ct method for relative template quantification was used to quantify the percentage of the target population (Fungi, Methanogen, Ruminococcus and Fibrobacter) relative to the normalizer (Bacteria). Data were statistically compared by ANOVA.

Results Gas and methane were significantly ($P < 0.01$) lower in both substrates incubated in the presence of BES. Irrespectively to the BES treatment, LUC produced higher ($P < 0.05$) amount of total gas than TIF (183 and 100 mL/g DM; SE = 5.7). Total methane production was significantly different ($P < 0.05$) between substrates (14.5 and 7.4 mL/g DM; SE = 0.67, respectively for LUC and TIF) and BES reduced significantly ($P < 0.05$) the methane production (0.4 and 21.6 mL/g DM; SE = 0.67). Methane efficiency was significantly different for the BES treated substrate (13.40 and 0.30 mL/100 mL GP for LUC with and without BES and 12.85 and 0.26 mL/100 mL GP for TIF with and without BES; SE = 0.349). Quantification of microbial population is shown in Table 1. At 24 h incubation, LUC $^{+}$BES showed methanogenic microbial population 1.80 fold lower ($P < 0.08$) than the LUC without BES, but did not affect the other studied methanogenic microbial population. The data suggest that BES reduce the CH_4 production

Table 1 Microbial population (Relative abundance expressed as proportion of Rumen broad Bacteria) measured by quantitative PCR (qPCR) using primer pairs for Broad rumen bacterial (Bacteria), Broad rumen fungi (Fungi), Broad rumen methanogen (Methanogen), *Ruminococcus flavefaciens* (Ruminococcus) and *Fibrobacter succinogenes* (Fibrobacter)

Microbial population	Lucerne		Tifton-85		SED	Probability	
	+ BES	- BES	+ BES	- BES		Hay	BES
Fungi	0.101	0.276	0.269	0.168	0.1682	ns	ns
Methanogens	0.064	0.115	0.030	0.039	0.0255	0.02	ns
Ruminococcus	0.030	0.019	0.043	0.043	0.0065	0.01	ns
Fibrobacter	0.381	0.387	1.00	0.631	0.2319	ns	ns

Conclusion BES can reduce CH_4 production on *in vitro* assay, but at 24 h incubation, it did not affect the relative abundance of Broad rumen fungi, Broad rumen methanogen, *Ruminococcus flavefaciens* and *Fibrobacter succinogenes* as proportion of Rumen broad Bacteria.

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The effect of ferulic acid esterase on the *in vitro* degradability of wheat straw

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Introduction The complete hydrolysis of complex structures, such as plant cell walls, requires the application of a range of multifunction enzymes in appropriate quantities. However while xylanase and cellulase activities in the rumen are adequate, such that additional (exogenous) enzymes are not required, their action is restricted due to the close association between cellulose and hemicellulose and other polysaccharides and to extensive substitution. In particular the arabinoifuranosyl residues in the hemicellulose chain form ferulic acid cross-links with lignin, so limiting biodegradability. Thus the so-called minor enzymic activities may be critical in making additional cell wall sites available for microbial attachment and subsequent digestion. To examine this effect the ability of ferulic acid esterase (FAE), which releases ferulic acid, to enhance fibrolytic activity by rumen microorganisms was evaluated *in vitro*.

Materials and methods An FAE, purified from Depol 740L, was identified as free from xylanase, β-xylosidase, endoglucanase, β-glucosidase and arabinofurinosidase activities. The steps involved included $(NH_4)_2SO_4$ precipitation, desalting, anion-/cation-exchange and hydrophobic interaction chromatographic procedures and gel filtration. The volume of purified FAE available (only a few ml, despite 1L Depol 740L being used) limited the number of replicates available and only a single application rate, based on earlier studies (0.2 µl g^{-1} dry matter) could be evaluated. The *in vitro* methodology (Mauricio *et al.*, 1999) was used to examine the effect of FAE treatment on the *in vitro* degradability of wheat straw (WS). In each flask, 1.0g WS was combined with 89 ml reduced buffer and stored overnight. FAE was added, diluted in 1 ml distilled water, immediately prior to inoculation with 10 ml prepared rumen fluid (hand-squeezed samples obtained pre-feeding from two lactating cows offered a grass hay/silage based dry cow diet). The flasks were incubated at 39°C for 48 h. Headspace gas pressure readings were taken at standard intervals and sets of flasks withdrawn 4, 8, 12, 19, 24 and 48 h post-inoculation to estimate *in vitro* degradation of dry matter (iDMD), neutral and acid detergent fibre (iNDFD and iADFD, respectively) and both lignin (ADL) and ferulic acid release. Gas production and iDMD data were statistically compared using a one-way ANOVA (SPSS Version 11.0), with significance was declared at $P<0.05$. Since bulked samples were used to assay fibre and lignin residues these data were not statistically analysed.

Results The addition of purified FAE significantly improved ($P<0.05$) iDMD during the later stages of the incubation period (+24 h post-inoculation). In addition enzyme treatment enhanced iNDFD and iADFD and increased ADL disappearance (see Table 1). It is worth noting that this latter component is the most recalcitrant plant cell polymer and is very poorly degraded by rumen microorganisms. As such the increased loss recorded is a significant effect. With respect to fermentation gas release, the treatment of straw with purified FAE resulted in similar increases to those seen with fibre degradation, especially at incubation periods in excess of 10 h post-inoculation. At 48h post-inoculation the fermentation of FAE treated straw was found to have liberated significantly ($P<0.05$) more gas (173 and 156 ml g^{-1} organic matter, for the treated and untreated material respectively). Although fermentation dynamics of the treated straw were enhanced throughout the entire incubation period, due to the activity of the enzyme applied, it was unlikely that the initial difference was a treatment effect. However the increased gas release kinetics observed from 12 h post-inoculation clearly demonstrated the highly significant effect which this enzyme has with respect to the enhancement of fibre degradation. The qualitative determination of ferulic acid further confirmed its release into the fermentation medium with a tendency for higher peaks to be identified with samples obtained 19 and 24h post-inoculation. With respect to mode of action of the purified FAE, chemical analysis of both the feed and residues identified an increase in NDF degradability. This indicates that not only did the esterase cleave lignin but that it appeared to alter either the structure of the various feed fractions so making them more amenable to rumen microbial enzymes or to enhance attachment and therefore access to the cell wall polymers.

Table 1 Degradation characteristics (48 h post-inoculation)

Treatment	Degradation (g kg^{-1})			Disappearance	Gas
	DM	NDF	ADF	ADL (g kg^{-1})	(ml)
Control	470[b]	566	555	206	156[b]
FAE	497[a]	596	579	252	173[a]
s.e.	7.3	-	-	-	6.1

Values in columns without common superscripts are significantly different ($P>0.05$)

Conclusions The application of a purified FAE significantly improved the fermentation and degradation dynamics of a poor quality feedstuff (wheat straw) widely used in ruminant nutrition. The esterase was able to cleave the cross-links which closely bind lignin to the main plant polysaccharides, as identified by the appearance of lignin in the fermentation medium. This allowed fibrolytic enzymes increased access with the end result being an increase in iDMD and more specifically enhanced NDF and ADF degradation.

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Methane production from tannin rich plants incubated *in vitro*

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Introduction Researches worldwide have been studying ruminal methane production *in vitro* and the *in vitro* gas production technique has demonstrated an ability to predict the *in vivo* enteric methane production from ruminants (Getachew *et al*., 2005; Tavendale *et al*., 2005). Studies of rumen microbial ecology in animals fed with tannin rich plants have demonstrated that tannins can affect rumen ecology. The objective of this study was to measure methane (CH_4) production of Brazilian tannin rich plants incubated *in vitro*.

Materials and methods Ten plant species were used as substrate: aerial parts (leaves and fine branches) of Arachis pintoi (ARA), Crotalarea juncea (CRT), Cajanus cajan (GND), Dolichos lablab (LAB), Leucaena leucocephala (LEU), Mucuna pluriens (MCZ), Mucuna aterrimum (MPR), Mimosa caesalpiniaefolia (SAN), Tephrosia candida (TFR) and Tifton-85 (Cynodon sp) hay (TIF), as control feed, which also is representative of the grasses Brazilian ruminants have available for pasture. Chemical composition and tannin assays were determined on oven-dried samples. Increase in gas production due to the presence of PEG was measured as tannin bioactivity (PEG effect). Two *in vitro* gas production (GP) assays were carried out using a pressure transducer for measuring the gas produced in 160 mL serum bottles incubated at 39 ºC (Bueno *et al*., 2005). Five adult rumen cannulated sheep were used as inoculum donor. *In vitro* gas production (GP) and methane (CH_4) output were determined in each bottle (triplicates for each substrate) incubated with 0.5 g of substrate plus 50 mL of Menke's buffered medium and 25 mL of inoculum. The amount of gas production (GP) was estimated (GP (mL) = 0.046 psi^2 + 6.45 psi). CH_4 determination was done in a Shimatzu 14A gas chromatographer and CH_4 production was estimated (CH_4 = [GP + HS] x Conc) following Tavendale *et al*. (2005).

Results The crude protein (CP) and condensed tannin (CT) contents of the substrates are shown in Table 1. Except for TIF (78 g/kg DM), all substrata had CP content greater than 200 g/kg DM and the fibre content present a wide variation (means of 608 ± 86.9 and 359 ± 61.3 g/kg DM, respectively for NDF and ADF). The extractable tannin content presented a coefficient of variation greater than 75 %. The increase in gas production due to the presence of PEG (PEG effect) showed significant ($P < 0.01$) correlation with the extractable tannin content of the substrates, with the Pearson correlation coefficient of 0.798, 0.749 and 0.809 respectively for TT, TP and TC.

Table 1 Chemical composition of tanniniferous plants used as substrates in *in vitro* gas and methane production

Chemical composition	Substrates										Mean	SD[1]
	ARA	CRT	GND	LAB	LEU	MCZ	MPR	SAN	TFR	TIF		
Crude protein[2]	218	245	253	250	246	236	241	191	225	78	218	52.7
Condensed tannin[2]	21	0.3	29	0.5	56	34	20	105	0.3	0.2	30	35.1
PEG effect[3]	12	0.7	27	2	18	19	4	215	7	5	31	65.1

[1] standard deviation; [2] in g/kg DM, 3 in %

There were differences ($P < 0.05$) among the substrates in GP and CH4 production (Table 2). Both substrates with lower CP and higher ADF and NDF contents (SAN and TIF) produced less GP and CH4. Methane efficiency was significantly ($P < 0.01$) different among substrates. The studied phenolic parameters were not significantly ($P > 0.05$) correlated with CH4. The CT content and PEG effect were significantly ($P < 0.01$) correlated with methane efficiency ($r = -0.662$ and -0.808 respectively).

Table 2 Total gas and methane production (ml/g DM) and methane efficiency (ml CH4 / 100 ml GP) with incubation of different substrates

	Substrates										SED[1]
	ARA	CRT	GND	LAB	LEU	MCZ	MPR	SAN	TFR	TIF	
Total gas	270	263	227	271	259	227	231	127	230	171	29.3
Total methane	37	37	32	39	33	30	30	12	35	22	5.2
Methane efficiency	13.5	13.9	13.5	14.2	12.6	12.9	12.7	9.1	15.2	12.1	1.02

[1] standard error of the difference

Conclusions Methane production *in vitro* can be affected by the tannin content in the substrate, but information on microbial mass production would be needed for complementing selection of plants for methane mitigation studies.

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The determination of fermentation characteristics of some Iranian feedstuffs using gas production technique

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Introduction The gas production technique has been widely used for evaluation of nutritive value of feeds (Pell *et al.*, 1998) and gas production profile has been shown to have good relationships with VFA produced in the rumen (Blummel and Ørskov, 1993; Brown *et al*., 2002), as well as neutral detergent fiber (NDF) (R^2 = 0.98) (Herrero and Jessop, 1996) and dry matter (DM) disappearance (R^2 = 0.95) (Prasad *et al*., 1994). Carbohydrate portion of each feed is divided into three digestible fractions: The A fraction, containing sugars, short oligosaccharides and organic acids; the B1 fraction, containing starch and pectin; and B2, the digestible fiber fraction (Doane *et al*., 1998). The objective of this study was to evaluate of nutritive value of some Iranian feedstuffs by using gas production technique.

Materials and methods Test feeds including vetch (V), bitter vetch (BV), sunflower head (SH), barley grain (BG), flaked barley (FB), canola meal (CM) and canola straw (CS) are evaluated using gas production technique. Three sheep (40±2 kg) used as donors of ruminal fluid for preparation of inoculums. The sheep were fed a diet consisting of 600 g kg-1 concentrate and 400 g kg-1 forage containing DE (3.35 Mcal/kg DM) and CP (160 g/kg DM). Gas production was measured by Fedorake and Hrudey (1983) method. The data were analyzed using the GLM procedure of SAS institute Inc (2000). The gas production profiles were in triplicate fitted with equation of $P = a + b\ (1\text{-}e^{-ct})$ that p is gas production in time t, a + b is potential of gas production, c is rate of gas production of insoluble fraction and t is time of incubation.

Results The results are shown in Table 1. Results indicated that gas production of feedstuffs at all time of incubated differed significantly ($p<0.01$). The fractional rate (c) and gas production volume of SH at 2 to 16 h of incubation rather than the other feeds ($p<0.05$). The potential of gas production fraction and fractional rate of gas production of insoluble fraction had significant difference between experimental feeds ($p<0.01$) and Potential fraction for BV and CM is maximums and minimum, respectively. These results may be due to the difference between chemical compositions specially carbohydrate contents.

Table 1 Gas production volume of feedstuffs (ml/gr) #

	Time of incubation (h)									
Feedstuffs	2	4	8	12	16	24	36	48	(a+b)	c
V	17.7^{b}	43.1^{b}	78^{b}	114.6^{b}	155.1^{dc}	210.4^{bc}	252.4^{a}	278.6^{a}	299.3^{b}	0.048^{c}
BV	17.7^{b}	42^{b}	77.1^{b}	107.3^{b}	141.3^{d}	189.3^{c}	234.6^{a}	254.6^{a}	305.8^{a}	0.046^{c}
SH	68.8^{a}	119.7^{a}	166^{a}	199.7^{a}	219.3^{a}	236.2^{ab}	249.5^{a}	253.7^{a}	250.92^{b}	0.120^{a}
CM	20.4^{b}	40.4^{b}	52.8^{c}	61.1^{c}	65.4^{c}	70.7^{d}	73.4^{b}	74.3^{b}	109.9^{m}	0.026^{g}
CS	23.3^{b}	43.1^{b}	59.66^{c}	67.2^{c}	70.7^{c}	74.7^{d}	77.8^{b}	80.5^{b}	118.5^{k}	0.050^{d}
BG	23.1^{b}	39.7^{b}	2.4^{bc}	100.8^{b}	179.3^{bc}	254.6^{a}	276.8^{a}	284.6^{a}	290.4^{c}	0.057^{b}
FB	18.5^{b}	36.7^{b}	56.36^{c}	107.6^{b}	202.1^{ab}	265.6^{a}	287.6^{a}	295.2^{a}	290.6^{c}	0.060^{b}
s.e.m.	4.44	65.43	8.47	9.7	12.8	17.3	19.8	20.9	0.22	0.00
Sig.	**	**	**	**	**	**	**	**	**	**

a, b means without a common letter differ at $p<0.05$.

Conclusions The results showed that the differences between chemical compositions of feedstuffs caused to change fermentation parameters determined by *in vitro* gas production technique. These results indicated that difference characteristics of degradability of feedstuffs and must be that considered in diet formulation in ruminant.

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The *in-vitro* study effect of dietary whole cottonseed and protein level on volatile fatty acids and gas production

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Introduction Whole cottonseed (WCS) is a by-product of the cotton-gin industry. This feedstuff is of significant feeding value for animals. WCS dry matter is high in fat, crude protein and neutral detergent fiber (NRC). The effect of fat on rumen-fermentation characteristics is explained by its detrimental effect on rumen microorganisms (Arieli, 1998). Chalupa *et al* (1984) found in an *in-vitro* study that addition of free C18:1, C18:2 and C18:3 to a hay-grain diet decreased the volatile fatty acids (VFA's) concentrations and acetate-to-propionate ratio. In some experiments, WCS had no effect on rumen fermentation. Sklan *et al*. (1992) found that the acetate:propionate ratio increased with WCS. The aim of this study was to survey the effect of addition of WCS and decreasing dietary CP level on rumen parameters.

Materials and methods Treatment diets were different in crude protein (CP) level and amount of WCS. The composition of diets given to the different treatments were as follow: 1) control (without WCS), 16% CP, 2) 20% WCS, 16% CP, 6.5% ether extract (E.E.), 3) 20% WCS, 13% CP, 6.4% E.E. *In-vitro* DM disappearance of the dried treatment diets was determined in five replications after incubation with ruminal fluid for 24 and 72 h using the Menke's gas test apparatus. *In vitro* gas production was determined at 0.5, 2, 4, 8, 12, 24, 48 and 72 h after incubation in rumen fluid in triplicate. Gas production data were fitted with equation using non-linear regression: $P = a + b\ (1- e^{-c(t-lt)})$, where *p* represent the *in-vitro* gas production (ml) at time *t*, *(a+b)* is the potential gas production, *c* the fractional rate of gas production per hour and l_t represents the lag phase before gas production commenced. VFA's and lactic acid concentrations were determined in the supernatant from samples incubated with rumen fluid for 24 and 72 hours. A single factor ANOVA was used to determine statistical differences between means for derived parameters (*a+b*), *c* and l_t.The GLM procedure was used to determine statistical differences between treatment diets for VFA's.

Results The addition of WCS did not decrease dry matter disappearance (DMD) 24 h incubation in diet 2 compared to control diet, but decreased for diet 3 with lower crude protein level. After 72 h incubation, DMD decreased by WCS supplement and protein level. The lag time prior to incubation of gas production from the treatment diets was zero, indication that there was no lag time. The protein level decreased fractional rate of gas production (c), but the differences between diets were not significant for potential gas production (a+b) (Table 1). The total VFA's concentration (mM) at 24 h after incubation was similar for diets, but the total VFA concentration (mM) at 72 h incubation was decreased with WCS. After 24 h incubation, WCS caused reduction of molar proportion of acetate and propionate and low crude protein percentage increased the molar proportion of butyrate.

Table 1 Dry matter disappearance (DMD) and the kinetics of *in vitro* gas production.

	Diet				
	1	2	3	SE	P value
DMD, % / 24 h after incubation	56.36[a]	57.62[a]	45.50[b]	2.62	0.0020
DMD, % / 72 h after incubation	76.63[a]	71.80[b]	68.64[c]	1.48	0.0013
In vitro gas production [a]					
a + b	22.99	21.48	21.27	0.90	0.7910
c	8.55[a]	8.34[a]	7.15[b]	0.26	0.0470
l	0.00	0.00	0.00	------	------

[a,b,c] Means denoted with different superscript in a row differ (P value as indicated).

Table 2 Volatile fatty acid content (% of molar proportions) and total VFA's (mM) for 24 and 72 h incubation of diets

		Diet				
Incubation Time (h)	VFA	1	2	3	SE (n=5)	P value
24	Total VFA	121.93	120.66	118.41	0.711	0.3099
	Acetate	56.36[a]	56.16[b]	56.15[b]	0.078	0.0021
	Propionate	24.36[a]	23.12[b]	22.60[b]	0.289	0.0206
	Butyrate	12.77[b]	12.41[b]	13.83[a]	0.285	0.0063
	Acetate/propionate	2.32	2.43	2.44	0.027	0.1618
72	Total VFA	150.05[a]	134.05[b]	142.25[a b]	2.340	0.0405
	Acetate	55.96	56.79	56.47	0.210	0.2452
	Propionate	22.23	22.87	22.57	0.140	0.1872
	Butyrate	11.86[ab]	11.58[b]	12.33[a]	0.122	0.0497
	Acetate/propionate	2.51[a]	2.48[a]	2.50[a]	0.011	0.0228

[a,b,c] Means denoted with different superscript in a row differ (P value as indicated).

Conclusions The results of the present study demonstrate that it's advisable to use 20% WCS in diets without any change in rumen parameters, because in the *in-vitro* system the addition of 20% WCS have not effect on total VFA concentration, DM disappearance and gas production. The lower CP percentage decrease DM disappearance and increase the molar proportion of butyrate.

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The effects of cadmium on parameters of fermentation by rumen micro-organisms *in-vitro*

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Introduction Animals grazing on land contaminated with cadmium due to pollution such as sewage sludge application and traffic emissions ingest cadmium adhered to plants and in soil. The effects of cadmium on ruminal enzymes have been investigated (Faixová and Faix, 2002) as have the effects on the bacterial and protozoal fractions of rumen fluid (Forsberg, 1978). These studies used a single cadmium salt, usually for ease of solubility, however, differences in toxicity have been found between cadmium salts (Barrouillet *et al.*, 2001). The aims of this study were to investigate the effects of cadmium salts and of differing concentrations of cadmium on fermentation by rumen microbes *in-vitro*.

Materials and methods Solutions of cadmium acetate, chloride, nitrate, oxide and sulphate at 0, 3, 10, 30 and 90 mg Cd/kg DM concentrations were added to 1g of ground hay (*Lolium perenne*) and 90 mls of cysteine free anaerobic buffer (Morgan *et al*, 2003) in 125 ml fermentation flasks and warmed (38^{o}C). Rumen fluid collected post slaughter from three Suffolk Cross lambs, fed a 75:25 forage:concentrate diet, was strained through 4 layers of muslin under a constant flow of CO_2 and 10 ml added to the flasks and sealed. Three replicates of each treatment were included along with buffer and rumen fluid controls. Gas pressure readings were taken at intervals up to 96 hrs using a pressure transducer (Mauricio *et al.,* 1999). Samples were analysed at 96 hrs for pH, volatile fatty acids (VFA) and neutral detergent fibre digestibility (NDFd). Gas production data was modeled for lag phase and fibre degradation using the method of Orskov and McDonald (1979) and all data was analysed using analysis of variance using SPSS, 13th Edition.

Results The rate of fibre degradation (c) varied significantly between Cd salt and concentration (table 1), with an interaction between salt and concentration ($p<0.001$). Increased concentrations of Cd sulphate (30 and 90mg Cd/kg DM) produced a higher rate of degradation than the control group ($p<0.05$). The presence of Cd acetate or nitrate did not have an effect on the rate of fibre degradation, neither did any Cd salt present at 10 mg Cd/kg DM. No significant differences or interactions were observed between Cd salts for the other constants of the modelled data, a - lag phase and b - extent of degradation insoluble but potentially degraded fraction. No significant differences were observed in total volatile fatty acid production or neutral detergent fibre degradation (table 2).

Table 1 Rate of fibre degradation (g/hr). Differing superscripts within columns are significantly different.

mg Pb/kg DM	Ac	Cl	Ni	Ox	Su	S.E.D.	Significance
0	0.0365	0.0365^{ab}	0.0365	0.0365^{ab}	0.0365^{a}	0.0008	ns
3	0.0386	0.0379^{b}	0.0368	0.0354^{ab}	0.0400^{ab}	0.0011	*
10	0.0335	0.0372^{b}	0.0363	0.0344^{a}	0.0394^{ab}	0.0019	ns
30	0.0375	0.0318^{a}	0.0360	0.0362^{ab}	0.0404^{b}	0.0016	**
90	0.0360	0.0380^{b}	0.0366	0.0379^{b}	0.0402^{b}	0.0011	*
S.E.D.	0.0020	0.0015	0.0010	0.0014	0.0011		Interaction
Significance	ns	*	ns	*	*		***

Table 2 Total VFA (mmol/l) and NDFd (g/g DM)

Salt	Total VFA	NDFd
Con.	46	0.71
Ac	45	0.72
Cl	46	0.73
Ni	45	0.72
Ox	45	0.71
Su	47	0.70
S.E.D.	1.499	0.015
	ns	ns

mg Cd/ kg DM	Total VFA	NDFd
0	46	0.71
3	46	0.72
10	46	0.71
30	46	0.71
90	45	0.72
S.E.D.	1.325	0.015
	ns	ns

Conclusion The presence of Cd showed no inhibitory effects on rumen fermentation. The increase in the rate of fibre degradation in the presence of Cd sulphate at high concentrations would suggest that it is the increase in sulphur, above that provided in the buffer, rather than the increase in Cd which acts to stimulate rumen fermentation. The interaction between concentration and salt implies that the salt species is responsible for the effects rather than the presence of Cd, although the lack of any consistent effect of Cd concentration on the rate of fibre degradation is difficult to explain.

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Evaluation of nutritive value of treated and untreated corn silage by formaldehyde and urea using gas production technique

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Introduction Gas measurement provides a useful data on digestion kinetics of both soluble and insoluble fractions of feedstuffs (Getachew *et al.* 1998). Carbohydrate portion of each feed is divided into three digestible fraction: the A fraction, containing sugars, short oligosaccharides, and organic acids; the B_1 fraction, containing starch and pectin; and B_2, the digestible fiber fraction (Doane *et al*. 1998). The objective of this study was to measure of fermentation parameters of corn silage treated by urea and formaldehyde using gas production technique.

Materials and methods The corn silage samples were chopped to 2 cm length. Treatments contain CS: untreated corn silage, CSF: CS + 4 g/Kg DM formaldehyde, CSU: CS + 10 g/Kg DM urea, and CSFU: CS + 4 g/Kg DM formaldehyde + 10 g/Kg DM urea. The isolated NDF was prepared by Doane *et al.* (1998) method. Three steers were used as donors of ruminal fluid for preparation of inoculum. Gas production was measured by Fedorak and Hrudey (1983) method. Since the accumulation of gas production was measured, so for determining of the effects of urea and formaldehyde supplementation on digestion kinetics, the data at the different times was analyzed using completely randomized design by the GLM procedure of SAS Institute Inc (1987). The gas production profile fitted with equation of $p=a+b\ (1-e^{-ct})$ that p is gas production in time t, a + b is potential of gas production, c is rate of gas production of insoluble fraction and t is time.

Results The results are shown in Table 1. Results indicated that gas production of isolated NDF was rather than the other treatments ($p<0.05$). Isolated NDF at 2 hour after incubation gas production equal zero. Corn silage + formaldehyde (CSF) treatment gas volume production at 96 h had different significantly from CSU ($p<0.05$). The rate of gas production of CS was higher than the other treatments ($p<0.05$). The potential of gas production had significant differences between treatments ($p<0.05$). Regarding to the differences in chemical composition of the treatments, the obtained results can be predicted.

Table 1 Gas volume production of treatments at the times of incubation (ml/g)*

	Time of incubation (h)									
	2	8	12	24	36	48	72	96	(a +b)[1]	c[2]
CS	7.11^{a}	38.2^{a}	57.1^{a}	111^{a}	161.7^{a}	186.2^{ab}	206.0^{b}	236.7^{b}	241.8^{a}	0.028^{a}
CSF	6.66^{a}	36.8^{ab}	54.6^{ab}	97.7^{a}	144.6^{ab}	168.7^{b}	197.6^{dc}	232.1^{b}	240.0^{b}	0.023^{c}
CSU	6.66^{a}	33.5^{b}	52.2^{ab}	99.3^{a}	142.6^{b}	167.7^{b}	193.1^{d}	217.6^{c}	225.0^{e}	0.025^{bc}
CSFU	7.11^{a}	36.0^{ab}	53.1^{ab}	105^{a}	151.3^{ab}	178.2^{ab}	202.9^{c}	232.6^{b}	238.1^{d}	0.027^{ab}
NDF	0.0^{b}	13.5^{c}	21.5^{c}	63.1^{b}	158.8^{b}	197.3^{a}	234.4^{a}	254.0^{a}	239.3^{c}	0.026^{ab}
SEM	0.73	1.83	3.74	8.7	8.8	9.72	5.07	5.6	0.0001	0.0009

1- The potential of gas production
2- The rate of gas production
* The means within a column without common letter differ ($p<0.05$)

Conclusions Using formaldehyde and urea in corn silage changed gas production parameters and fermentation characteristics.

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In vitro DMD and gas production of *Leucaena leucocephala, Manihot esculenta* (cassava) root and *Pennisetum purpureum* (Taiwan) grass mixtures

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Introduction Tropical fodder trees are commonly used as supplement to grass feeding. Usually supplemental foliage is used without any additional energy source. Therefore, the potential of fodder trees to increase microbial biomass production (due to foliage N capture for microbial growth) might not be achieved. In addition, it has been suggested that forage digestibility might be stimulated if an energy source, ideally, locally available, is provided with the foliage. Thus, the objective of the present experiment was to assess the DMD of *Leucaena leucocephala* and *Manihot esculenta* (cassava) mixtures, and the *in vitro* gas production of Leucaena-cassava-grass mixtures.

Material and methods *L. leucocephala* foliage, cassava root and *Pennisetum purpureum* (Taiwan) grass were harvested in summer 2004. The elaboration of the *in vitro* medium was carried out following the Menke and Staingass (1988) methods. Gas production technique as Theodorou *et al*. (1994) was used. Rumen content was collected from two canulated heifers, which were fed *ad libitum* with *Pennisetum purpureum* grass and supplemented with 5 kg commercial concentrate (16 % of Crude Protein (CP)) and 5 kg fresh leucaena fodder (aprox. 1.5 kg Dry Matter (DM)). Two different incubation sets were used. A) 24 h incubation for DM Digestibility (DMD) and B) 96 h incubation for gas production; in both cases gas volume and pressure were recorded at fixed time intervals. For both sets Leucaena and cassava were mixed at 100:0, 75:25, 50:50, 25:75 and 0:100 ratios and 100% grass. In all mixture treatments, each bottle contained 0.5 g Taiwan grass and the remaining 0.5 g was made of leucaena:cassava mixture. Each mixture was placed in a 100 ml bottle and added 6 ml of inoculate and 54 ml of *in vitro* medium. All treatment mixtures have a total weigh of 1 g of DM and four replicates. Gas profiles were fitted using the monophasic model of Groot *et al*. (1996), 24h DMD was analysed using anova (SAS, 2000).

Results Chemical composition of the feeds evaluated is presented in Table 1. In leucaena- cassava mixtures no difference was found (P>0.05) between observed and predicted DMD (based on each ingredient DMD), it increased proportionally to cassava in the mixture. DMD on 100:0, 75:25, 50:50, 75:25, 0:100 mixtures and 100% Taiwan were 41.5, 47.8, 52.8, 57.7, 61 and 41.1±0.41 % respectively. Gas production followed a similar pattern between observed and predicted values (Table 2). *In vitro* gas production was related to the DMD and described by the following relationship: ml gas/g DM= 2.45(DMD%) + 25.53, R^2= 0.983

Table 1 Chemical composition of feed (g/kg DM)

	CP	OM	CF	NDF	ADF	L	ADF-N	EE	TP	CT
Leucaena	260.6	921.3	ND	353.8	168.8	72.0	3.6	44.7	32.2	100
Taiwan	81.0	910.0	ND	712.6	410.7	46.8	2.2	25.3	ND	ND
Cassava	30.1	926.9	68.5	118.8	ND	ND	ND	1.2	ND	ND

OM, organic matter, CF, crude fibre, NDF, neutral detergent fibre, ADF, acid detergent Fibre, L, ligning ADF-N, Nitrogen bound to ADF, EE, ether extract, TP, total polyphenols, CT, Condensed tannins

Table 2 *In vitro* gas production profiles of leucaena, cassava and Taiwan mixtures (ml gas/g DM).

	100:0	75:25	50:50	25:75	0:100	Taiwan
VT	167.3±2.8	181.4±2.1	192.5±0.065	201.5±4.1	212.7±5.3	182.4±1.9
B	30.1±0.7	24.5±0.4	21.5±0.5	20.4±0.6	20.7±0.8	30.1±0.4
C	1.9±0.052	2.0±0.049	2.0±0.994	2.0±0.096	1.9±0.104	2.3±0.0
Rm	0.028	0.036	0.040	0.042	0.039	0.032
t_{RM}	28.2	24.7	21.1	20.1	19.0	33.4
Sy.x	3.1	3.3	4.8	7.5	8.9	2.8
IVDMD	57.6±0.99	62.8±0.99	68.3±0.99	72.6±0.99	75.4±0.99	65.2±0.99

VT, total gas volumen, B, time to half gas production, C, parameter to define curve profile w/o biological value, Rm, fractional rate of sustrate degradation t_{RM} time at which the fractional rate occurs, (Rm and t_{RM} were calculated from the means thus, no statistical analysis is reported), IVDMD, *in vitro* DMD at 24h, Sy.x residuals from curve fitting

Conclusion DMD and *in vitro* gas production of grass and leucaena:cassava mixtures can be described as additive result of the individual ingredients. No associative effects were found.

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Nutrient composition and *in vitro* organic matter degradability of desert plants from Iran

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Introduction A vast area in central Iran is desert in which the soil salinity is high. Therefore, plant varieties capable of growing in this area are very limited, the most typical ones which can be used by animals, mainly camel, are *Alhogi spp*. (AL), *Salsola soda spp*. (SS) and Shooreh (SH). However, the nutritional value of these plants in terms of their chemical compositions and degradability characteristics is not available. The objective of this study was to determine the chemical composition and extent organic matter (OM) digestion of these plants compared with grass nuts (GN) as a reference feed by using an *in vitro* method (Rezaeian and Chaudhry 2005).

Materials and methods The plant samples were harvested in April 2004, dried at 60°C for 72 h in a fan-assisted oven and their dry matter (DM) contents determined. Dried samples were then milled (<1 mm) and analyzed for CP, NDF, ADF, EE and ash contents using standard laboratory procedures. Organic matter degradability (OMD) of each plant was determined by using an *in vitro* fermentation method as described by Rezaeian and Chaudhry (2005). Rumen fluid was collected from two rumen fistulated sheep consuming fixed ratios (65:35) of medium quality grass hay and a concentrate with 160g CP/kg DM. Rumen fluid from each sheep was strained through four layers of muslin, purged with CO_2 and stored in a screw capped bottle at 39°C until its use as the inoculum. About 0.5 g dried sample of each plant was weighed into a 50 ml plastic test tube to which 10 ml of the inoculum and 40 ml of a buffer (pH = 7) were added. The tubes were flushed with CO_2 and capped with Bunsen valves. Duplicate tubes were incubated at 39°C for each inoculum from each sheep for 0, 4, 8, 16, 24, 48 and 72 h. After each incubation time, the tubes were centrifuged to collect residues which were washed with water and then first dried and then ashed to calculate *in vitro* OMD of of each sample. Degradability parameters (a, b and c) were calculated by using an exponential equation in FigP programme. Degradability data were analyzed using GLM model in Minitab to determine the LS values for comparing means. The differences between means were declared significant if P <0.05.

Table 1 Nutrient composition of feeds

Feeds	AH	SH	SS	GN
OM, *g/kg DM*	798	844	860	922
NDF //	470	406	371	547
ADF //	281	213	175	306
CP //	76	137	193	160
Ash //	202	156	140	78
EE //	9	26	16	19

AH: Alhogi, SH: Shooreh, SS: Salsola soda, GN: grass nuts

Table 2 *In vitro* organic matter degradation (g/kg) of various feeds during different incubation times

Times	AH	SH	SS	GN	SEM
0	175	232	235	194	9.7
4	216	309	285	227	11.5
8	268	368	354	254	24.2
16	316	458	407	425	28.5
24	414	566	469	519	27.2
48	546	674	586	679	8.6
72	586	718	630	710	4.4

Results Table 1 shows the chemical composition whereas Table 2 presents the OM degradation of different desert plants over various times. The ash contents of all three tested plants were considerably higher than that of GN. AH had relatively low protein content whereas the protein contents of SS and SH were comparable to that of GN. There was a significantly greater ($p<0.05$) immediately soluble fraction (a) of OM from SH and SS (230 and 237 respectively) compared to AH and GN (171 and 161 respectively). The OMD of SH was significantly greater than the other two plants but it was nearly the same to that of GN at 48 and 72 of incubation. This was despite the fact that SH contained twice more ash than the GN. Alhogi showed lower OMD at each time and so a lower rate of degradation (c= 0.026) compared with other feeds (0.040, 0.032 and 0.033 for SH, SS and GN respectively). The slowly degradable fraction (b) also differed ($P<0.05$) for different feeds (50.6, 52.1, 43.6 and 62.6 for AH, SH, SS and GN respectively).

Conclusions Results from both chemical composition and the degradability data indicated that the nutritional value of Shooreh is comparable even to grass nuts despite its very high ash content. The degradability profiles of the other two plants were also acceptable. The lower OMD of *Alhogi* compared with other feeds could be due to its lowest CP and highest ADF contents. It appears that these desert plants could be used as potential feed ingredients to formulate diets for ruminant animals in Iran. However, further studies are necessary to determine the nutritional value of these plants at various maturities and to investigate the probable effect of their relatively high ash content on the health and performance of ruminant livestock in Iran.

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The effects of lead salt speciation and concentration on rumen microbial activity, *in-vitro*

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Introduction Forage pollution by lead has been caused by mining, sewage sludge application, nuclear plants and traffic emissions. Animals grazing on contaminated land have a higher exposure to the toxicity posed by contamination adhered to plants and through soil ingestion. Strojan and Phillips (2002) found a decrease in gas production in the presence of lead although other parameters were not measured. The objectives of this study were to investigate the effects of different lead salts and concentrations on microbial activity of rumen micro-organisms *in-vitro*.

Materials and methods Ground hay (*Lolium perenne)* was contaminated with lead acetate, chloride, nitrate, oxide and sulphate at concentrations of 50, 100, 200, 400 and 800 mg Pb/kg DM. Hay samples (1 g) were incubated in serum flasks containing 90 ml of a cysteine free anaerobic buffer (Morgan *et al*, 2003) containing a microbial marker ($[^{15}NH_4]_2SO_4$) plus 10 ml of rumen inocula. Rumen fluid, collected post slaughter from five Suffolk Cross lambs fed a 75:25 forage:concentrate diet, was strained through four layers of muslin under a constant stream of CO_2. Six replicates of each treatment were included along with buffer and rumen fluid controls. Gas pressure readings were taken at intervals up to 96 hrs using a pressure transducer (Mauricio *et al.,* 1999). Samples were analysed at 96 hrs for pH, volatile fatty acids (VFA), microbial growth by ^{15}N uptake using mass spectrometry and neutral detergent fibre digestibility (NDFd). Data was analysed by analysis of variance using SPSS 13th Edition.

Results Total VFA production was affected by both concentration and salt, with an interaction occurring between the two (table 1). The presence of lead oxide, except at 400 mg Pb/kg DM, caused decrease in microbial growth, which was not dependent on concentration (table 2). There was no clear pattern between the other salts. At any given concentration, except 400mg Pb/kg DM, lead oxide and sulphate caused an inhibition of microbial growth. No differences in NDFd were observed in the presence of lead and in the control group.

Table 1 Total VFA produced (mmol/l), I interaction of salt and concentration

mg Pb/ kg DM	Control	Ac	Cl	Ni	Ox	Su	S.E.D	Signif.
0	63	63	63	63	63	63	0.813	ns
50	63	60	64	63	63	62	0.452	ns
100	63	68	62	66	60	65	0.853	***
200	63	62	63	61	61	66	0.570	**
400	63	61	64	66	66	66	0.607	***
800	63	60	64	67	65	65	0.740	***
S.E.D.	0.813	0.874	0.312	0.712	0.649	0.541		I.
Signif.	ns	***	ns	***	***	ns		***

Table 2 Total microbial N (mg). Differing superscripts within columns are significantly different.

mg Pb/ kg DM	Control	Ac	Cl	Ni	Ox	Su	S.E.D	Signif.
0	9.65	9.65	9.65	9.65	9.65	9.65	0.140	ns
50	9.65	8.89	9.87	9.44	8.71	7.82	0.266	**
100	9.65	9.54	9.63	10.31	8.73	8.97	0.179	***
200	9.65	9.24	9.89	9.78	8.25	8.18	0.129	***
400	9.65	9.73	9.61	10.11	9.79	9.07	0.316	ns
800	9.65	9.16	9.60	9.30	8.60	8.63	0.110	***
S.E.D.	0.140	0.148	0.233	0.244	0.153	0.268		I
Signif.	ns	**	ns	*	***	**		***

Conclusion Solubility of lead salts affects microbial growth, with the insoluble lead salts (oxide and sulphate) inhibiting growth. The effects of the soluble salts on microbial growth varied from the control slightly but were not related to concentration. The effect of the presence of lead on total VFA production did not follow a consistent pattern, although statistically significant, numerical differences were small and would be negligible in practice. This investigation shows that the species of lead salt does have an effect on rumen microbial activity which may not be apparent from gas production data alone.

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Determination the *in vivo* and *in vitro* digestibilities and *in situ* degradability of some Iranian local feedstuffs

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Introduction Determining the feed value of local feedstuffs is necessary for the optimum utilization of resources in animal nutrition. Considering climate, plant variety and animal breed, there are many variations among the nutritional value of feedstuffs in any region of the world. The objective of this experiment was to determine the *in vivo* and *in vitro* digestibilities and *in situ* degradability of some local feedstuffs in Tehran province of Iran.

Materials and methods Six local feeds including alfalfa hay (1st, 2nd and 3rd cut of Hamedani variety), wheat straw (Qhods), barley straw (Valfajr) and corn silage (Single cross 702) were used in this study. Three fistulated Afshari wethers (mean BW 39 ± 2.3kg) were used for *in vivo* and *in situ* measurements. Animals were fed with a maintenance diet (40 g feed/kg$BW^{0.75}$). Alfalfa hay was fed as the sole diet for *in vivo* measurements. The *in vivo* digestibilities of the other feedstuffs were determined by the difference method. The experiment lasted for 21 days including 14 days adaptation and 7 days of collection period. Tilley and Terry method (1963) was undertaken for the *in vitro* measurements. For i*n situ* degradability, nylon bags (21×10 cm) containing 5g samples were incubated in the rumen for 0, 2, 4, 8, 16, 24, 36, 48, 72 and 96 hours. Naway program was employed for determination the soluble (a), slowly degradable (b) fractions, constant fractional rate (c), effective degradability (P) and residual standard deviations (RDS). Outflow rate of %.0.02 was used to calculating (P). All the feed and fecal samples were chemically analysed according to AOAC procedures (1990). MSTATC package was used for statistical analyses.

Results CP content of feedstuffs were 13.5, 13.2, 18.0, 4.6, 2.4 and 6.7 for 1st , 2nd and 3rd cut alfalfa hay, wheat straw, barley straw and corn silage respectively. Low level of CP for 1st and 2nd cut of alfalfa was probably due to the pest damage. *In vivo* and *in vitro* digestibility data for DM, OM and D-value are shown in Table 1. For all feedstuffs, *in vivo* digestibilities were higher than *in vitro* values ($p < 0.05$). There was a significant difference between the third cut and other cuts of alfalfa ($p < 0.05$). The correlation equations between *in vivo* and *in vitro* data were as follows: $Y_1=1.092X_1+2.65$ ($r = 0.99$) for different cut's of alfalfa and $Y_2 = 0.985X_2+10.12$ ($r = 0.94$) for all of feedstuffs where Y_1 and Y_2 were *in vivo* digestibility and $X_1$and X_2 as *in vitro* digestibility of feedstuffs respectively.
Undegradable protein content in the 3rd cut of alfalfa hay was higher than other cuts. It means more UDP can be provided by feeding this hay to dairy cows.

Table 1 *in vivo* and *in vitro* digestibility values for DM, OM and D-Value (g kg^{-1}) and protein degradability parameters for feedstuffs[1]

Feedstuffs	digestibility						degradability					
	DM		OM		D-Value		a	b	100-a-b	c	P	RSD
	in vivo	*in vitro*	*in vivo*	*in vitro*	*in vivo*	*in vitro*	%	%	%		%	
Alfalfa(1stcut)	610.2[a]	533.6[b]	626.4[a]	539.1[b]	557.6[a]	479.3[b]	35.34	54.26	10.2	0.021	63.3	1.49
Alfalfa(2ndcut)	607.5[a]	532.9[b]	619[a]	544.4[b]	575.5[a]	477.5[b]	19.27	63.96	16.77	0.034	59.5	2.97
Alfalfa(3rdcut)	512.6	445.1	530	483	464.1	435.5	39.69	39.69	20.62	0.048	67.6	1.65
mean	576.8	503.9	591.8	522.2	532.4	464.1	31.76	51.23	17.01	0.032	63.4	1.21
Wheat straw	319.3[a]	201.4[b]	322.5	211.4	299.7[a]	199.0[b]	35.07	25.76	39.17	0.025	49.3	1.65
Barley straw	355.2	302.1	392.2	326.2	394.4	310.3	17.83	21.08	61.09	0.038	31.7	0.41
Corn silage	644.2[a]	464.7[b]	687.9[a]	501.2[b]	620.7[a]	452[b]	40.93	50.83	8.24	0.006	52.3	1.40

[1]Data are the mean of 3 replicates.

Conclusion According to the significant positive correlation between *in vivo* and *in vitro* digestibility values, the *in vitro* data can be used for prediction the *in vivo* digestibility as it has been reported elsewhere. According to the results of this study it can be concluded that feeding the 3rd cut of alfalfa hay to dairy cow could lead to better performance due to its higher content of UDP which was come out from the higher content of CP.

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Ruminal retention time and indigestible dry matter fraction of various Iranian forages measured by *in situ* technique

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Introduction The extent of ruminal forage dry matter digestion is a function of rate of digestion and rate of passage. It is a complex process that is affected not only by the forage composition, but also by the retention time of forage particles in the different segments of the gastrointestinal tract (Noziere & Micalet-Doreau, 2000). A major factor affecting ruminal NDF digestibility of forage is the rate at which the forage passes from the rumen. This is a unique characteristic of individual forages or fibre sources. Daily intake of digestible organic matter is also closely correlated with ruminal retention time for forages and this relation applied to both temperate and tropical feeds. The objective of the present study was to determine the ruminal retention time (RRT) and indigestible dry matter fraction (IF) of various Iranian forages using *in situ* nylon bag procedure.

Materials and methods Samples of various forages including lucerne hay and silage, maize silage, whole barley hay and silage, wheat straw and halophytes (*Atriplex sp., Sauda sp,*and *Kochia sp.*) were incubated in four Holstein steers (330±15 kg body weight) with ruminal fistula. Animals were fed to maintenance body weight with a medium quality lucerne hay (35%), maize silage (15%), wheat straw (15%) and concentrate (25%) twice a day, at 9:00 and 16:00 h, individually. Ground (2 mm) samples (5 g DM) were incubated in the rumen for 0.0, 2, 4, 8, 16, 24, 48, 72 and 96 h (n=8). Samples were placed in artificial silk bags (10×20 cm, 50µm pore size) and previous to incubation they were hydrated for 15 minutes (37 ºC). Additional bags per animal at 96 h were used (n=12) to estimate indigestible fraction. After extraction, bags were rinsed thoroughly with cold tap water until the rinse water was clear. Then, they were dried (65 ºC) until constant weight and weighed. Data of DM degradation beyond the lag-time were further adjusted to the model: $P=a+b(1-e^{-ct})$, where P= fraction degraded in the time t, a= rapidly degradable fraction, b= slowly degradable fraction, c= fractional degradation rate and t= incubation time. Ruminal retention time (day) calculated by the following equation: RRT=(((1-a-b)/k)+(b/(c+k)))/24; where k= ruminal outflow rate (0.06 and 0.08 h^{-1}). Data were analysed using the general linear model procedure of SAS.

Results The data of ruminal retention time and indigestible dry matter fraction are shown in Table 1.

Table 1 Ruminal retention time (day) and indigestible dry matter fraction (g g^{-1}) of various Iranian forages using *in situ* nylon bag procedure.

Forages	Ruminal retention time*		Indigestible dry matter*
	K=0.06	K=0.08	
Lucerne hay	0.241	0.203	0.180
Lucerne silage	0.290	0.223	0.210
Maize silage	0.370	0.295	0.280
Whole barley hay	0.261	0.208	0.200
Whole barley silage	0.376	0.287	0.250
Wheat straw	0.451	0.402	0.370
Atriplex sp.	0.310	0.243	0.320
Sauda sp.	0.276	0.213	0.270
Kochia sp.	0.325	0.257	0.360
SEM	0.021	0.018	0.019
P-value	< 0.05	< 0.05	< 0.05

* When the difference between means is greater than two times the SEM, it is considered as significant (P < 0.05).

Conclusion Nylon bag technique has been used as a method for prediction of the RRT (Noziere & Micalet-Doreau, 2000). It has been demonstrated that the RRT is a good estimate of both fill index and indigestibility (Noziere & Micalet-Doreau, 2000). In the present study, there was a large variation in RRT and IF of the evaluated forages by *in situ* method. Lucerne and whole barely hays had lower RRT and IF compared with the other forages. Therefore, these forages are more susceptible for digestion in the ruminant gastrointestinal tract than their silages. This study showed that in general, *Sauda sp.* may be better than the other species of halophytes observed because this plant had a lower RRT and IF.

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Bringing scrapie under control

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Scrapie is an invariably fatal, infectious disease of adult sheep and goats characterised by degeneration of the central nervous system. It is one of the Transmissible Spongiform Encephalopathies (TSE), a disease family which includes Bovine Spongiform Encephalopathy (BSE) in cattle, Chronic Wasting Disease (CWD) in deer, and Kuru and variant Creutzfeldt-Jakob Disease (vCJD) in humans. Scrapie was first discovered in European sheep in the early 18th Century and is now present in many sheep populations worldwide.

Key epidemiological features of scrapie are its incubation period, its transmission routes, and the genetic control of susceptibility.

In common with other TSEs, the incubation period is long. The mean age at which sheep die from scrapie in Great Britain is 44 months, with the majority believed to be infected at or soon after birth.

Scrapie appears to be transmitted between sheep by several routes: (1) there may be some vertical transmission (direct transmission *in utero*) although this is not yet proven; (2) there is good evidence for horizontal (direct lateral) transmission of scrapie between adult sheep housed together, although the mechanism is unknown; (3) in natural flocks, transmission may often be mediated via contact with infectious birth tissues; infectivity and PrP^{Sc} can be readily detected in the placenta and foetal membranes of infected ewes; (4) finally, sheep can somehow acquire scrapie from 'infectivity' that persists - for up to several years - in the environment.

Whether a sheep can acquire scrapie infection or, following infection, proceed to clinical disease, is under the control of the gene that encodes its PrP protein, called the *PrP* gene. Five haplotypes of the *PrP* gene that exhibit this linkage to scrapie are commonly found in sheep. The haplotypes encode the following amino acid sequences in PrP: alanine (A) at codon 136, arginine (R) at codon 154 and arginine (R) at codon 171 ($A_{136}R_{154}R_{171}$or ARR for short) and, continuing this notation, ARQ, AHQ, ARH and VRQ (Q is glutamine, H is histidine and V is valine).

Each sheep inherits one haplotype from each of its parents, giving it one of fifteen possible *PrP* genotypes often written as, for example, ARQ/VRQ or ARR/AHQ. The relationship between the haplotypes/genotypes on the one hand, and susceptibility to scrapie on the other, is complex. Briefly, ARR and AHQ confer resistance to classical scrapie, while ARQ and VRQ confer susceptibility. The ARR/ARR genotype of sheep is the most resistant to classical scrapie, while the VRQ/VRQ genotype is the most susceptible. The effect of the ARH haplotype varies, depending on the other haplotype present. Small changes in genotype can have a remarkably large effect on susceptibility to scrapie. For example, British sheep of the ARQ/VRQ genotype are 300 times more likely to be reported with scrapie than sheep of the AHQ/VRQ genotype, despite differing only in arginine or histidine being encoded by codon 154 of one of the two haplotypes. The molecular mechanism that links *PrP* genotype to resistance/susceptibility is not understood.

Many approaches to scrapie control have been tried in the past, such as cleaning and disinfection of premises, culling of affected animals, culling of entire age-cohorts, culling of genetically-related animals and even whole-flock slaughter. Such methods have generally not brought long-term success. The recent discovery of the strong association between classical scrapie and *PrP* genotype has, however, raised the possibility of controlling the disease at a national level by selective breeding. Genetic control programmes are now underway in many countries, including all member states of the European Union. These programmes are based on large-scale genetic testing, followed by culling of the most genetically susceptible sheep (those that encode VRQ) and promoting the use of ARR/ARR sheep for breeding.

The logic underpinning these programmes is now being challenged following the discovery of 'atypical' scrapie. Scrapie has long been known to exist as different strains. Atypical scrapie appears to be caused by one (or more) newly discovered strains; key differences from classical scrapie are an older age at death, different distribution of PrP^{Sc} in the brain and a low within-flock incidence. The genotype also tends to differ. In one form of atypical scrapie, designated Nor98, affected sheep most frequently encode the AHQ allele while VRQ appears to encode resistance. Most atypical scrapie has been identified through a recent, large-scale, and Europe-wide initiative of active surveillance, based mostly on the testing of either fallen stock or animals at abattoirs. As with Nor98, atypical cases identified by active surveillance tend to encode ARR, AHQ or ARQ, while VRQ is exceptionally rare. Importantly, a significant proportion is of the ARR/ARR genotype, resistant to classical scrapie and the genotype most favoured by current breeding programmes.

As well as challenging current control programmes, the rise of atypical scrapie raises fundamental questions about the origins of the different PrP alleles. Has each allele evolved to confer resistance to different scrapie strains that occurred in the past? If so, a new approach to (long-term) control may be preferable: breeding out susceptibility in individual affected flocks while preserving genetic diversity in the national flock.

The role of meat lipids in human health

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Diet related chronic conditions such as cardiovascular disease (CVD), cancers and diabetes are the main sources of morbidity and mortality in westernized societies, being responsible for approximately 64% of deaths in men and 73% of deaths of women within the EU (Peterson *et al.,* 2005). The amount and composition of dietary fat is recognised as an important determinant of the initiation and progression of these chronic conditions.

With respect to CVD numerous epidemiological and intervention trials have conclusively demonstrated that a diet high in total fat, saturated fat (SFA) and *trans* fatty acids (TFA) is associated with increased risk of disease, whereas diets with an increased polyunsaturated fatty acid (PUFA) or monounsaturated fat (MUFA): SFA ratio, and increased dietary intakes of the long chain n-3 PUFA are associated with reduced disease risk. The data accumulated thus far has indicated that every 1% reduction in dietary SFA intake is associated with an approximate 3% decrease in CVD risk, based on its impact on LDL cholesterol alone. It is likely that dietary fatty composition is a less important in the etiology of cancer. However, positive associations between total fat and n-3 PUFA intake and cancer development at certain sites, in particular the breast has been reported. Also recently there has been much research interest in the protective influence of conjugated linoleic acid (CLA), in particular the *c9 t11* isomer, against the development of breast cancer, with rodent carcinogenic models and some epidemiological evidence demonstrating a protective effect (Aro *et al.,* 2000). However a number of recent population studies have reported no association (Voorrips *et al.,* 2002). Ruminant products represent the almost exclusive dietary source of these particular fatty acids.

Currently in the UK adult diet, meat and meat products contribute 23%, 22% and 21% of total fat, SFA, and TFAs, with equivalent figures in children being 20%, 18% and 15% respectively. Therefore reducing the SFA content of animal products (including milk and mil products) represents an obvious target in order to reduce population SFA and TFA intake and CVD risk, with even modest alterations in meat fatty acid composition having the potential to result in a significant reduction in the population disease incidence.

In response over the last decade there has an explosion of research activity investigating the ability to positively influence meat fatty acid composition through altered dietary exposure of the animal/fowl, primarily with the aim of increasing the polyunsaturated fatty acid (PUFA): SFA ratio, lowering the n-6: n-3 ratio, and increased long chain n-3 PUFA and CLA content (Dewhurst *et al*., 2003). Such dietary changes have been brought about by increasing either the monounsaturated fatty acid (MUFA) or PUFA composition of the animals' diet, and the role of grass-based forages for supplying dietary C18:3 n-3 has been of particular interest. In non-ruminant animals and fowl, decreases in the SFA:PUFA ratio of up to 3-fold have been achieved. However, a key difficulty in using dietary unsaturated lipid to alter the fatty acid content of ruminant meat, results from the extensive biohydrogenation of dietary fatty acids by rumen microbes. Another less studied consequence of rumen microbial action on PUFA is the enhanced production of deleterious trans fatty acids (TFA) resulting in substantially increased TFA in meat, in some cases by a factor of two. Ruminant tissues possess substantial desaturase activity with the result that a proportion of the TFA are converted to the potentially beneficial conjugated linoleic acid (CLA) thus enhancing CLA concentrations in meat. Thus the use of protected lipid sources, which are resistant to biohydrogenation, is thought to result in greater meat enrichment efficiency following PUFA/MUFA ingestion by the animal and also reduces TFA/CLA synthesis in the rumen. Currently some refinement is necessary to produce a meat product with 'maximised' fat composition with respect to health, and it remains to be demonstrated that such meat products with 'enhanced' fat profiles can positively influence the development of disease such as CVD. Also careful consideration needs to be given to the impact such dietary and meat fatty acid changes will have on the welfare of the animal.

In summary, meat fats have traditionally been considered a source of 'negative fats' in the diet. However, emerging research is demonstrating the potential to produce meats with an enhanced fat profile, whose incorporation into the diet in place of currently consumed products, holds the potential to positively influence population dietary fat composition and reduce the burden of chronic disease.

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Bioavailability of haem-iron in relation to pregnancy anaemia and the down-stream consequences to the offspring

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Introduction During pregnancy, the developing fetus obtains all its iron from the mother. At birth in humans, the neonate has a total of about 1g of iron. The mother can provide up to 600 mg of iron from cessation of menstruation and from increased absorption, with about 300 to 400 mg coming from maternal stores. It is clear, therefore, that maternal absorption, both before and after pregnancy, to build up and replenish iron stores respectively, and to provide iron directly for the fetus, is critical for normal growth and development.

Iron deficiency during pregnancy has consequences for the mother and her offspring, both in the short and long term. Several studies in humans have identified an increased risk of haemorrhage in an anaemic mother, and other studies have shown that deficiency in the neonate results in developmental delay that can be reversed by supplementation in the neonatal period. In epidemiological studies, it has been suggested that maternal iron deficiency results in an increased risk of cardiovascular disease and stroke in the offspring. More recently, it has been demonstrated that supplementation of women of apparently normal iron status during the first half of pregnancy results in a decrease in percentage or small babies, an increase in the size of the babies at birth and a decreased risk of prematurity. None of these effects were seen when iron was given in the second, rather than the first, half of pregnancy. All these data, together with supplementation studies in Nepal, point to the critical importance of having enough iron stores and iron absorption in the early stages of pregnancy. Disconcertingly, however, the percentage of women who are anaemic, over and above the normal anaemia of pregnancy, is remarkably high. WHO estimates for the Europe are about 15 to 20 %, and in one study we performed in Aberdeen, a relatively wealthy part of Scotland, the fraction was as high as 30 %. In this presentation, therefore, we discuss the way that iron is presented to the developing fetus, how the fetus, the placenta and the maternal liver act in concert to ensure that the fetus has adequate iron, even at the expense of its mother, and what the short and long term consequences may be.

Results and Discussion During pregnancy, the iron is transferred from the mother to the fetus through the placenta. Serum iron, bound to the carrier protein, transferrin, attaches to the transferrin receptor on the surface of the placenta. The complex is internalised into endosomes and the pH decreased to about pH6.0 by a H-ATPase. This results in release of Fe from the Tf and it is transferred into the cytoplasm of the cell through a divalent iron channel – DMT1. The iron (as Fe(II)) exits the trophoblast cell through another channel, Ireg1, and is oxidised to Fe(III) by a Cu-oxidase called eleutherin. This Fe(III) will bind to fetal transferrin and is carried to the fetal liver. The whole process is under very tight homeostatic control. Changes in Fe levels are detected and the effect minimised by changes in expression of the transporter proteins and in the stability of the mRNAs for the transporters. In addition, a systemic regulator, called hepcidin, is inversely expressed as a consequence of the Fe levels in maternal and fetal livers. Our data show that the maternal levels of iron are regulated by the fetus, and not by the mother, and this poses some very intriguing questions about the way that the fetus and the mother communicate nutritional status.

If the baby is born deficient, their problems can be severe and we have developed a rat model to study the long term consequences of maternal Fe deficiency. The pups are born smaller, with increased heart and decreased kidney. As adults they develop high blood pressure, despite a normal iron intake throughout their post-natal life. They also develop dyslipidaemia and obesity. We are currently using molecular biological, genomica nd proteomic techniques to try and elucidate the mechanisms underpinning these observations.

In summary, iron deficiency during pregnancy results in many and various complications. How iron is supplied to the mother and her fetus is very important, but the period when iron is given is also critical. Our data will demonstrate how these problems may arise, how the developing fetus tries to cope with the nutritional stress and what the consequences may be for itself, both as a child and later, as a full grown adult.

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Meat as an essential source of micronutrients

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Meat is frequently associated with a “negative” health image due to its “high” fat content and in the case of red meat is seen as a cancer-promoting food. Therefore a low meat intake, especially red meat is recommended to avoid the risk of cancer, obesity and metabolic syndrome. However, this discussion overlooks the fact, that meat is an important source for some of micronutrients such as iron, selenium, vitamins A, B12 and folic acid. These micronutrients are either not present in plant derived food or have poor bioavailability. In addition, meat as a protein rich and carbohydrate “low” product contributes to a low glycemic index which is assumed to be “beneficial” with respect to overweight, the development of diabetes and cancer (insulin resistance hypothesis). Taken together meat is an important nutrient for human health and development. As an essential part of a mixed diet, meat ensures adequate delivery of essential micronutrients and amino acids and is involved in regulatory processes of energy metabolism.

Genomic tools, what does the industry gain – examples from pigs

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Introduction Pig breeding companies have been using DNA markers for selection for nearly 15 years, since the introduction of the test for the "Halothane gene". Other tests have been developed and are used by the industry, although the total number in use was probably less than 100 at the end of 2005. However, large numbers of SNPs have now been identified in the pig and these are being used for relatively high density genome scans (using 1,000s of markers). It is anticipated that significantly more use will be made of DNA markers in the next decade. This paper will provide specific examples of the impact made by markers on the pork chain.

Results The identification of a single nucleotide polymorphism in the ryanodine receptor gene (*RYR1* or *CRC1*) associated with porcine stress syndrome (PSS) and pale soft exudative (PSE) meat (Fujii *et al*. 1991) provided the pork industry with a solution to a significant problem. This major gene (the "Halothane gene") had reached a relatively high frequency in several populations and breeds around the world, presumably because it positively impacted desirable conformation in an additive way, while the unfavorable effect on mortality was recessive. The negative impact on meat quality (PSE) was not initially observed or valued. However, by the time that the test was available the industry was well aware of the negative impact of the recessive allele and programmes were quickly introduced to eliminate the "Halothane gene". Indeed, "Halothane free" became a specific requirement to market pigs in many markets. This meant that the much of the value derived from use of the test was realised downstream in the chain and it did not create gain at the breeding company or producer level (although mortality due to PSS was reduced). Other major gene effects that have been elucidated at the molecular level include the Rendement Napole (RN^-) defect associated with reduced yield of ham, dominant white and resistance to *E. coli* F18 and more recently *E. coli* K88. The RN test has enabled some breeding companies to eliminate the gene without the use of invasive biopsy methods. This means that Hampshire based lines continue to have value as long as they are RN-free (a similar situation to "Halothane", although in this case the unfavourable allele was really only segregating in the Hampshire breed), that new white dam lines could be more easily developed (e.g. white Duroc-based lines) and that farmers and producers could be offered genetically-resistant stock where *E. coli* disease was a problem. In these cases breeding companies provide greater product choice which translates into market share. In the case of *E. coli* F18 resistance the breeder is also able to devise payment schemes that lead to increased margins on these products. The producer benefits from "insurance" against losses due to the disease (mortality is typically 20% in affected herds). Understanding the molecular basis of coat colour provided an interesting model for the study of gene interactions (e.g. epistasis) as well as marker discovery. In addition, analysis of *MC1R* and *cKIT* variants provides the basis for breed identity tests where increased value is ascribed to specific breeds e.g. Berkshire in Japan (Alderson and Plastow 2004). Such tests help the consumer be sure of the provenance of the meat product being purchased. The most widespread application of DNA markers will be for those markers that explain a proportion of the quantitative variation that is the raw material for genetic improvement. The mutation identified in the porcine *MC4R* gene is a good example. This marker explains variation in appetite and as a consequence growth and leanness. Boars can be selected to be homozygous for the "lean" allele for a market such as the UK where lean carcasses are essential in order to obtain full value for a pig. Semen from boars selected in this way was sold at a premium. The producer expected to see a saving in feed efficiency as well as 0.5mm less backfat in progeny of these boars which equates to a return of approx. £3.50 per dose (i.e. additional value for £0.80 investment). Backfat results actually realised 0.75-1mm less backfat giving an even better return than expected. Similar results have been obtained for markers explaining variation in ultimate pH of pork. This measurement correlates well with water holding capacity and is therefore of interest to packers and retailers in terms of yield and visual appearance respectively. Meat quality traits such as pH are very suitable for a marker assisted selection approach to improvement as the trait can only be measured post mortem. Once an association has been identified marker information can be routinely incorporated into the selection index. As shown here DNA markers are already available that add value at different parts of the pork chain. General health and robustness as well as other difficult to measure traits are expected to be amenable to these genomic approaches as long as sufficient phenotypic information has been collected.

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Strategies for the use of molecular information in breeding programmes

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Since the 1970's, several developments have raised high expectations for the use of molecular genetic technology to enhance selection in livestock through application of marker-assisted selection (MAS). These include the discovery of technologies that enabled identification and genotyping of large numbers of genetic markers, and research and statistical methods that demonstrated how these technologies can be used to identify genomic regions that control quantitative traits and how the resulting quantitative trait loci (QTL) can be used in MAS. Yet, to date, the application of MAS in livestock has been limited (see e.g. review in Dekkers 2004). Recent further advances in technology, including genome sequencing and the associated discovery and identification of large numbers of single nucleotide polymorphisms (SNPs) that can be used as genetic markers, combined with the development of high-throughput SNP genotyping that allows the genotyping of large numbers of individuals for large numbers of SNPs at substantially reduced costs, have however stimulated a renewed interest in the large-scale application of MAS in livestock.

The purpose of this paper will be to review strategies for QTL detection in livestock and to discuss opportunities and limitations of their use in commercial breeding programs through MAS. For these purposes, an important distinction will be made between the following three classes of markers,

1) Direct markers: loci for which the functional polymorphism can be genotyped.
2) LD-markers: loci in population-wide linkage disequilibrium with the functional mutation.
3) LE-markers: loci in population-wide linkage equilibrium with the functional mutation but which can be used for QTL detection and MAS based on within-family LD.

Strategies for detection of these alternate types of markers and their advantages and disadvantages in QTL detection and for MAS will be discussed. While direct markers are easiest to implement in MAS because of their consistent association with the trait across the population, they are also most difficult to detect. On the other hand, although using within-family or breed-cross QTL mapping has provided ample results on the identification of QTL, because of their loose association, the application of the resulting LE-markers for MAS within outbred populations has proven difficult and limited because of the need to estimate QTL associations and effects on a within-family basis. LD-markers offer an important middle ground, allowing much of the benefit of direct markers in their implementation in MAS but with much greater opportunities for detecting LD-markers that are associated with QTL.

To be associated with a QTL, LD-markers must be close enough to the QTL that sufficient LD exists across the population. Apart from random change, the relationship between LD and distance is a function of population history, including effective population size, breed composition, and selection, but has been shown to be substantial within 2 cM in livestock populations. The extent of LD will be much greater in recent crosses, which allows LD-MAS with a limited number of markers across the genome for marker-assisted development of synthetic lines.

In outbred populations, LD-markers associated with QTL can be detected on a targeted basis using candidate gene approaches and several examples of the commercial application of resulting LD-marker tests are available. The feasibility of high-density SNP genotyping, however, offers the possibility to identify LD-markers that are associated with QTL on a genome-wide basis. Opportunities and challenges for the application of these technologies for QTL detection and MAS in commercial breeding programs will be discussed.

In contrast to LE-markers, direct or LD-markers also provide opportunities to select for traits that are expressed at the commercial production level. Although they represent the ultimate goal of any commercial breeding program, these traits are difficult to improve using conventional selection because of the difficulty of obtaining pedigreed phenotypic data from commercial production facilities

In conclusion, although current applications of MAS are limited and several challenges for its successful implementation remain, advances in molecular genetic technology now provide prime opportunities to reap its benefits in commercial livestock breeding programs.

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New Molecular Tools for Dog Breeders

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The last ten years or so have seen a major, concerted international research efforts to produce genome maps for the dog. One of the major driving forces behind the research was to provide the tools required to track down and identify mutant gene alleles that are responsible for canine inherited disease. Early phases of map development involved the cloning and characterisation of 1000s of canine microsatellite sequences and the production of microsatellite-specific primer pairs that could be used to amplify specific microsatellite loci. Microsatellites were then mapped to their individual chromosomal loci using a combination of meiotic linkage analysis in standard canine reference families and radiation hybrid mapping. Molecular cytogenetics has also been used to gain a clearer picture of the canine karyotype and how it relates to the human karyotype.

The success of these studies can be partly measured in the steady increase, since around 2000, in the number of single-gene mutations that have been characterised as the cause of canine inherited disease. Today we know the precise mutation for around 50 different inherited single-gene disorders in the dog. This knowledge has immense significance for the dog breeder; knowing the precise mutation responsible for an inherited disease means that a very simple DNA test can usually be developed to detect the particular mutation in an individual dog. Most of the single-gene disorders in the dog are recessive, which means that traditionally breeders have had great difficulty identifying the clinically normal carrier dogs and taking account of them in their breeding programmes. Disease-specific DNA tests are an invaluable new tool in the breeders' arsenal for tackling inherited disease through selective breeding. Individual dogs can be tested easily, using simple buccal swabbing, and before they are mated, to determine their genotype with respect to a particular inherited disease. Breeders can then decide how best to use the identified carrier and affected dogs. For example, an identified carrier could be mated to a DNA tested normal dog and, moreover, the subsequent puppies can also be DNA tested early in their lives to identify the normal and carrier progeny.

Other valuable bonuses from these genome studies include the establishment of canine DNA profiling using a subset of the microsatellites developed for genome mapping. This provides for individual canine DNA identification and provides the basis for a reliable and robust method for verifying biological parentage, something that can add great integrity to the many purebred canine registries, like the ones that we maintain at the kennel Club. A more recent advance in this area is the use microsatellite loci to provide a means of determining individual breed-specific DNA signatures. This means for the very first time, an unknown canine DNA sample can, after analysis, be attributed to a particular breed, if it is from a purebred dog. If the sample came from a crossbred dog, the analysis will identify the ancestral breeds that lie behind the dog.

Finally, 2005 saw the publication of a high quality canine DNA sequence with 99% coverage of the genome. Funded entirely by the National Institutes of Health in the USA, and completed in just over 12 months, this resource will take canine genome studies to a whole new level of exploration. The information will certainly accelerate the identification of the mutations behind other canine single-gene disorders, but the new information opens up the possibility of ever more sophisticated association studies to identify mutations that are responsible for canine inherited disease and, for the first time, we can think of identifying the multiple genetic mutations involved in things like canine hip dysplasia, epilepsy, inherited heart disease, autoimmune diseases and cancer. Over 2 million individual single nucleotide polymorphisms have been identified during the DNA analyses and these will form a fundamental part of future disease/mutation association studies. The availability of such a detailed and comprehensive genome sequence will certainly enhance comparative genetics in man, mouse and dog. I have no doubt that human geneticists will be looking far more seriously now at those canine inherited diseases that are homologues of human inherited conditions that are proving hard to solve, and using them as models to give them new insights into the human condition.

Animal products and human health: present situation and future potential
Sir John Hammond Memorial Lecture

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Over the last century, research in food-animal agriculture has focused predominantly on improving productive efficiency through gains in yield and animal well-being. Impressive gains have been and will continue to be achieved, and Sir John Hammond has been among the leading contributors through his own work and his influence on scientists that followed, including myself. Nutritional quality is an important consideration in food choices and the contribution of animal-derived foods in supplying essential nutrients has been recognized for some time. However, the last decade has seen increased consumer interest in the link between diet and health, and as a result scientists are increasingly asked to clarify the role of specific foods and food components in health maintenance and disease prevention. The term 'functional foods' has been adopted to describe foods or food components that have beneficial effects on human health beyond that expected on the basis of nutritive value. Traditionally, functional food components in fruits and vegetables have been featured for their health promoting properties, but recent investigations have identified an impressive list of bioactive components present in animal-derived foods that have beneficial effects on health-related variables in studies with biomedical models. A recent National Academy of Sciences report titled "Frontiers in Agriculture Research" identified research on bioactive food components as a key area for future research to enhance human health through nutrition.

Advances in our understanding of the benefits of specific fatty acids in ruminant fat are of special significance due to the generally negative public perception that a food containing saturated fat is unlikely to be beneficial to human health. Investigations over the last decade have clearly established that generalizations about fat and fatty acids are of little value and often led to public confusion and misunderstanding. One of the most exciting advances in this area has been the discovery that conjugated linoleic acids (CLA) may have beneficial effects on human health and disease prevention. Ruminant meat and milk fat account for over 90% of our dietary intake of CLA and their presence in ruminant fat relates to rumen biohydrogenation of dietary PUFA. Several CLA isomers are present, but *cis*-9, *trans*-11 (rumenic acid; RA) is the major isomer and both diet and variation among individuals are predominant factors affecting the RA content of fat. Although RA is an intermediate in the biohydrogenation of linoleic acid, the principle source in ruminant fat is via endogenous synthesis by Δ^9-desaturase. Vaccenic acid (VA), another biohydrogenation intermediate produced in the rumen, is the substrate and because of the precursor-product relationship between VA and RA, their concentrations in ruminant fat always change in parallel. RA is among the most potent naturally occurring anticarcinogens that have been identified in foods and its anti-cancer effects have been consistently demonstrated across a range of cancer types and biomedical models, with particularly impressive results in models of breast cancer. Antiatherogenic properties have also been observed for RA using classical animal models, and these effects relate to improvements in both plasma lipoprotein-cholesterol profile and inflammatory biomarkers. Anticarcinogenic and antiatherogenic effects have also been demonstrated for VA and these effects appear to be mainly, perhaps exclusively, due to its conversion to RA. Of special importance, the beneficial effects in cancer and atherosclerosis models have been observed using a natural form of VA/CLA (esterified in triglycerides) in a naturally-enriched food (dairy products).

Examinations of human applications in disease prevention are limited and problematic because the development of cancer and atherosclerosis has long latency periods and for cancer there is a lack of consensus biomarkers. Further, there have been significant advances in the analysis of CLA and *trans* fatty acids so that values in food tables used for health-related studies are generally inadequate. Successful application of functional food components found in animal fats will also have to include the education of the public that fatty acids differ in their effects. This is of special importance with the public perception that all *trans* fatty acids are undesirable and the fact that both VA and RA are *trans* fatty acids, although their biological effects are in marked contrast to *trans* fatty acids derived from partially hydrogenated vegetable oils. The range of functional food components in animal products is impressive and defining their role in health maintenance and the prevention of chronic diseases is progressing at a rapid rate. The identification and characterization of bioactive components will not only aid in the development of new products to benefit human health but also help improve the public perception of animal products in general.

Red meat and green farming: Can we improve product quality, enhance farm income and deliver environmental goods on the same farm?

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This paper will review the factors that will, over the next 10-15 years, determine the sustainability of UK livestock farms. Based on an analysis of the industry as it is today and an assessment of the likely impacts of CAP reform, I will argue that there is likely to be a greater plurality of enterprise. For those farms wishing to compete in a global market at commodity prices, cost efficiency will remain paramount, but there will also be incentives driving towards greater vertical and horizontal integration. Such farms are likely to comply with best practice, but would not see the delivery of environmental goods as an essential part of the farm's outputs. I will consider for such producers what advantages may accrue from the rapid deployment of new technology. Under these circumstances, quality considerations will emphasise reliability and safety over taste and (possibly) health benefits, with the need to remain competitive on price remaining paramount. Efficiency gains will only bring a brief benefit to producers, with competition rapidly forcing the benefits further down the food chain.

For both niche producers and for producers such as the National Trust where outputs from land are diverse, improved quality offers a potential financial reward whilst helping to support a less intensive production system. In such systems, the delivery of environmental goods may be a primary objective (e.g. where it is supported by higher-level agri-environmental payments) and might also contribute to the marketing image of the farm or the ownership organisation. Under such circumstances, quality-added value goes hand-in-hand with things like local marketing, agri-tourism, farm shops and local produce being served in hotels and restaurants. Product quality considerations will emphasise taste, taking safety for granted, and consumers may accept some fluctuations in supply if the quality is high enough. Success in this market depends upon consistently high standards throughout the production, processing and marketing chain, with cost minimisation being important but not the major driver.

The technology challenges for niche producers are, therefore, different to those facing commodity producers. These challenges will vary even more where there are multiple outputs from the farm system. Here there will be the need to balance costs and impacts in one area with benefits and markets in another. I will argue that the current way we do research on land use systems does not support such an approach. It is also difficult within this framework to estimate the value-added of potential health benefits associated with modified product composition. However, the biggest issue here is one of orderly market growth. The problems of oversupply in the organic milk sector illustrate how vulnerable niche added value systems can be if the enterprise is linked to a single output.

In the final part of the talk, I will consider the possible future impact of global drivers on UK livestock production systems. Growth in the market for animal products will, in principle help the industry, but pulling higher-cost producers into commodity trading could also have an adverse influence on the ability of these systems to deliver environmental goods. Higher prices also encourage the exploitation of marginal land, particularly where delivery of ecosystem services is not valued. I will suggest that many more of the externalities of farming need to be internalised before it can be described as genuinely sustainable, and capable of supporting a transparent market in the delivery of a range of economic, social and environmental goods.

Potentials for linking food and health strategies

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Climate change and environmental issues have emerged as major present drivers of UK animal agriculture. However, the longer-term vision for a profitable, sustainable future is for the food industry (food production, processing, retailing catering and marketing) to become more integrated in terms of providing good food for consumers who place more emphasis on healthy diets (Curry Report 2002). BSAS has seen considerable change in the types of animal science that have been reported at its meetings over the last 50 years, from the emphasis on greater production in the 1950s and 60s to greater efficiency in the 70s and 80s to better quality products in more recent times. However, whilst the animal industry has come a long way in terms of the quality of its products, it has had to face a series of 'scares' over recent years that have eroded consumer confidence in these products and the farmer's share of a basket of food staples (down 25% from 1988). Nowadays total farm income in the UK (£3 billion per year) is tiny when considered in terms of consumer expenditure on food, drink and catering (£152 billion per year).

It is also tiny when compared to the rising cost of health care in the UK (total managed NHS spending in 2005/06 of £76 billion) and so, perhaps not surprisingly, there have been major efforts made over recent years to raise the public's (consumer's) awareness of 'Health and Quality of Life' issues. Indeed, the Government is committed to reducing CVD, strokes and related diseases in people under the age of 75 by 40% by 2010. Deaths from cancer and cardiovascular disease have fallen over recent years, however CVD still costs the NHS £7 billion per year and accounts for some 125,000 deaths and 275,000 heart attacks each year and with over 1.4 million suffering from Angina. Similarly, Cancer is reported to claim 155,000 deaths each year while 1.5 million suffer from Diabetes.

Over recent years better diet (as well as increased exercise and reduced smoking) has become increasingly recognised as an important potential contributor to disease prevention. It is perhaps now time therefore for those of us in animal science to consider what our colleagues in clinical medicine and human nutrition can tell us about which nutrients are health-enhancing and then explore how animal products might be promoted as contributing to a balanced diet for the health-conscious consumer. This paper will develop messages delivered in the 'Meat as a Functional Food' session and in the Hammond Lecture to explore ways in which animal products could be promoted as good sources of 'better quality lipids' and 'extra micronutrients'; two areas that are seen as alleviating important risk factors in poorer quality diets. In terms of 'the Curry Report' this might also be one way of planning for the time beyond agricultural subsidies when the perception of consumers may contribute significantly to the economics of farming.

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Factors affecting muscle growth and meat quality in pigs

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Introduction The understanding of growth and development of skeletal muscle is one of the most important goals in animal and meat science. The major component of a given muscle is the constituent muscle fibres. Muscle mass is therefore largely determined by the number of muscle fibres and the size of those fibres.

Principles of myogenesis and postnatal muscle growth Prenatal myogenesis is considered as one of the most important events that determine postnatal growth, lean accretion and meat quality. During myogenesis stem cell commitment and the extent of muscle cell multiplication largely determine how many muscle fibres are formed. Myoblasts develop from mesenchymal precursor cells by proliferation and myogenic commitment and subsequently fuse to form multinucleated myofibres. Muscle fibre formation in the pig is ceased by day 90 of gestation. In late gestation and postnatal, muscle fibres grow in length and size and develop contractile and metabolic diversity, which is associated with accumulation of myonuclei (satellite cell proliferation) and muscle-specific proteins. It is of significance that postnatal fibre hypertrophy inversely correlates with the number of prenatally formed muscle fibres.

Correlations of muscle fibre traits with lean accretion and meat quality Both fibre number and fibre size correlate positively with lean mass. However, extreme fibre hypertrophy, as observed in some meat-type pigs, may be associated with problems in stress adaptability and ultimate meat quality. Likewise, high percentages of fast-twitch glycolytic muscle fibres or of abnormal giant fibres may indicate poor pork quality in terms of pale, soft, exudative (PSE) meat.

Significance of prenatal nutrition Low birth weight in piglets is mainly a result of prenatal undernutrition as littermates in this polytocous species highly compete for nutrients *in utero*. In the majority of low birth weight piglets low numbers of muscle fibres differentiate during prenatal myogenesis, and those piglets with reduced fibre numbers are unable to exhibit postnatal catch-up growth. Pigs of low birth weight show the lowest growth performance and the lowest lean percentage at market weight. In addition, they tend to develop extremely large muscle fibres (giant fibres) and poor meat quality.

Improvement of lean growth and meat quality Prenatal growth/myogenesis is under the control of various genetic and environmental factors, which can be targeted for growth manipulation. Increased nutrient availability during embryonic/foetal development by maternal nutrition above request or by growth hormone action could stimulate prenatal myofibre formation, in particular of small littermates that are severely disadvantaged by insufficient nutrient supply. On the other hand, genetic selection is considered to be a suitable tool to improve foetal growth and myogenesis. First, selection for an optimum balance of litter size and birth weight could reduce the consequences of malnutrition *in utero*. Second, genetic variability and heritability of muscle fibre number and size are sufficiently high to include these traits in farm animal selection in addition to commonly used selection criteria for simultaneously improving lean meat content and meat quality.

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Molecular basis of skeletal muscle phenotype

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Muscle phenotype is central to the quantity and quality of meat production. Quantity is a function of muscle hyperplasia and hypertrophy. Hyperplastic growth *in utero* is a function of myocyte proliferation and differentiation, and post-natal growth is primarily the result of hypertrophy of existing and replaced muscle fibres. Quality is a more complex trait and is assessed by a variety of objective and subjective parameters, such as colour, pH, tenderness, odour and juiciness. It is increasingly evident that fibre type composition is a major determinant of meat quality. Favourable meat traits, such as tenderness, have been found to associate with the greater abundance of oxidative fibres. Therefore, knowledge of the molecular events that affect hypertrophy (or atrophy), hyperplasia (or hypoplasia), and fibre type-specific expression is of fundamental agricultural importance.

This presentation examines the molecular events (including the IGF-1 signalling pathway) and factors (such as β2 adrenergic agonists and nutrition) that govern the processes of muscle hyperplasia, hypertrophy, and fibre-type specific gene expression. Calcineurin signalling is arguably one of the most significant pathways responsible for muscle fibre type determination. It has been shown to enhance muscle cell differentiation and up-regulate the expression of type I/slow myosin heavy chain (MyHC) gene in the promotion of an oxidative phenotype. Additionally, we show that calcineurin signalling facilitates an oxidative fibre type switch by the differential regulation of the 3 fast post-natal MyHC genes. Specifically calcinuerin up-regulates the expression of fast oxidative-glycolytic MyHC 2a gene, but down-regulates the expression of the fast oxidative-glycolytic MyHC 2x gene and the fast glycolytic MyHC 2b gene. Underpinning knowledge of the regulation of fibre growth and fibre type, in particular the effector targets of calcineurin signalling, could enable the manipulation of muscle phenotype through marker-assisted selection or by pharmacological targeting of novel effector genes in muscle.

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Improving muscle quality during processing

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Introduction Meat quality, including eating quality, can be modified by production factors such as breed and diet and by various processing factors considered here. Tenderness, the most important and variable aspect of meat quality, is more affected during processing and, in the UK, the major retailers normally specify the post-slaughter conditions that are applied.

Tenderness Various post-slaughter interventions are employed to improve tenderness: slow or delayed chilling, electrical stimulation (ES), hip suspension and meat ageing. The first stage of processing, chilling, is critical. If chilling is too fast, then it can lead to 'cold shortening' and toughness (Table 1). It occurs if muscle is cooled below about 10^{o}C before the onset of rigor. Lamb is especially prone to cold shortening because the small carcasses cool rapidly and they do not enter rigor quickly. Cold shortening can be prevented by cooling more slowly, or by using ES or pelvic suspension. ES was developed to prevent cold-induced shortening in New Zealand lamb. By promoting a rapid pH fall it prevents cold shortening when carcasses are cooled quickly; it also improves muscle tenderness in carcasses when cooling is slow enough to avoid cold shortening (Table 1). A major advantage of ES is that it can afford a way to promote tender meat without the need for extended post-mortem ageing (Table 1). In hip suspension, the carcass is hung from the pelvis instead of the Achilles tendon. This stretches certain muscles as they enter rigor, resulting in a tenderisation of the valuable *longissimus* muscle and other muscles on the outside of the hip. A disadvantage of hip suspension is the extra labour required to re-hang carcasses in the chiller. Nonetheless, ES and hip suspension are now widely accepted practices in the UK, especially for producing tender beef. Extending the period during which meat is held at 1^{o}C post-slaughter, termed ageing or conditioning, is one of the most important ways to increase tenderness (Table 1). The rate of tenderization differs widely between species and this has led to different recommended ageing times for pork (4-10 days), lamb (7-14 days) and beef (10-21 days). Keeping meat in refrigerated storage is expensive and long ageing times can adversely affect the colour shelf life. There is debate about whether meat should be aged in carcass form (dry ageing) or in vacuum pack (wet ageing).

Juiciness Modern meat is sometimes criticised for being dry and lacking succulence and this is often attributed to poor water holding capacity (WHC) or low levels of intramuscular fat (IMF). The pH of meat at butchery (ultimate pH) has a marked influence on the ability of fresh meat to retain natural or added water, a property usually referred to as the WHC. Meat of lower WHC is associated with higher drip loss, higher cooking loss, lower juiciness and lower tenderness values. Retention of water within the muscle structure seems to be vital for optimal tenderness which could explain why meat cooked to a high internal temperature is less tender and juicy (Table 2). Adding a marinade (usually based on salt or polyphosphate) to increase water retention during cooking has a large and positive effect on tenderness and juiciness (Table 2). The level of IMF in pork loin muscle can vary from <1% to >5% but typical values in the UK are about 0.8%. Lower IMF levels are usually associated with lower tenderness and juiciness scores, but reported correlations are usually no higher than about r=0.3. However, the higher quality of Duroc pork seems to be linked with higher marbling fat.

Flavour Raw meat has little flavour and it is only during cooking that meaty flavours and aromas are developed. These are generated from precursors in the meat: sugars, peptides and lipids. Most processing factors are aimed at improving tenderness and have relatively subtle effects on flavour. However, longer ageing times combined with exposure to oxygen and light during retail display can lead to lipid oxidation and flavour alteration, usually described as undesirable. Meats with high levels of polyunsaturated fatty acids and low levels of vitamin E are especially prone to lipid oxidation. Marination with polyphosphate can have an adverse effect on flavour (Table 2).

Table 1 Mean shear force values (kg) for cooked lamb loin samples at 3 and 10 days post-slaughter (Taylor, 1996)

Chill	ES	3 day	10 day
quick	No ES	8.0	5.9
quick	ES	5.4	4.2
slow	No ES	5.4	3.2
slow	ES	4.1	2.8

Table 2 Influence of polyphosphate concentration (0 and 5%) on eating quality of grilled pork loin steaks cooked to an internal temperature of 72.5 or 80^{o}C (Sheard *et al*., 1999)

	72.5	72.5		80	80	
	0	5	Sig.	0	5	Sig.
Tenderness	4.5	5.7	***	4.2	5.3	***
Juiciness	4.0	4.4	ns	3.4	3.9	*
Pork flavour	3.3	3.1	ns	3.8	3.2	**
Abnormal flavour	3.4	3.5	ns	2.9	3.4	ns

1-8 scale

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Nutritional approaches to improving pork quality

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Introduction Producing high quality pork has become a major goal of swine industries worldwide. Pork quality is a very broad term that encompasses many aspects including hygienic, technological, and organoleptic attributes. From an economic perspective, two critical components of pork quality are muscle colour and water-holding capacity. There is evidence that the incidence of pork colour and water-holding capacity defects are increasing in many countries. For example, slaughter plant surveys carried out in the US suggested that between 1993 to 2003 the incidence of Pale Soft Exudative (PSE) pigmeat increased from less than 10% to above 15% of carcasses, despite a major effort by all sectors of the industry to reduce this problem. This review will focus on nutritional approaches to enhancing the colour and water-holding capacity of pork.

Causes of reduced pork colour and water-holding capacity Detrimental changes in pork colour, and possibly water-holding capacity, can occur due to oxidation in the muscle. There is an exhaustive literature on the feeding of antioxidants, particularly vitamin E, prior to slaughter to address this problem and this area will not be considered in this review. Most of the variation in pork colour and water-holding capacity results from aberrations in post-mortem muscle glycogen metabolism. Particularly, rapid post-mortem glycolysis results in a relatively low muscle pH early post-mortem when muscle temperature is still high and produces the PSE condition. Also, extensive glycolysis is associated with a low ultimate muscle pH post-mortem (typically observed in animals that have the mutation of the Rendement Napole gene) and pale muscle colour and reduced water-holding capacity. In contrast, a more limited post-mortem glycolysis results in a higher than normal ultimate muscle pH and improved muscle colour (darker) and water-holding capacity. Obviously, approaches to improve pork colour and water-holding capacity, be they nutritional or other methods, should target reducing the rate and/or extent of post-mortem muscle glycolysis.

Nutritional approaches to manipulating post-mortem muscle metabolism Nutritional approaches to reduce the rate of postmortem muscle glycolysis that have been evaluated include compounds that inhibit glycolytic enzymes such as **Quercetin, Sodium oxalate,** and **Vitamin C. Magnesium** is a mild sedative and also antagonizes the action of calcium to stimulate muscle activity which could slow glycolysis. **Tryptophan** is a precursor of serotonin, a brain neurotransmitter that could reduce the pig's response to pre-slaughter stress. Compounds that could buffer post-mortem pH changes in muscle include **Creatine,** which binds phosphate within the muscle, and **Electrolytes** such as sodium bicarbonate. Other compounds that have been evaluated include **Betaine**, a methyl donor that could also maintain osmotic balance within the muscle cells and reduce moisture loss and **Trimethylamine oxide**, another methyl donor. Various **Chromium** compounds have been studied for effects on growth, carcass, and meat quality. Feeding high levels of **Vitamin D_3** to finishing pigs improves muscle colour and water-holding capacity but also reduces growth performance. In general, the responses observed in pork colour and water-holding capacity to the administration of all of the compounds listed above have been extremely variable and inconsistent. In addition, a number of these compounds (e.g., vitamin D_3) have significantly reduced the growth performance and/or increased production costs.

Reducing muscle glycogen content at slaughter Another potential approach to improving pork colour and water-holding capacity is to reduce muscle glycogen content at slaughter, thus, lowering the amount of energy available for glycolysis and, in theory, limiting the extent of pH decline and increasing the ultimate pH in the muscle. The most practical approach to achieve this is to withdraw feed from pigs prior to slaughter. In addition, changes in dietary energy substrates offer potential to impact muscle glycogen levels.

Feed withdrawal Feed withdrawal prior to slaughter has a number of potential benefits including reducing deaths in transport, reducing gut fill and the incidence of punctured intestines during evisceration, and improving pork colour and water-holding capacity. This later effect is generally considered to result from a reduction in muscle glycogen content at slaughter, however, feed withdrawal also results in a reduction in body temperature; reduced muscle temperature at slaughter would be potentially favourable for pork quality. Although there has been a considerable volume of research carried out in this area, the results have generally been very variable and inconclusive and it is still difficult to give precise recommendations on the optimum time of feed withdrawal to improve pork quality.

Dietary energy source Approaches to reduce muscle glycogen levels by manipulation of the energy content of the diet have included feeding diets low in digestible carbohydrate and/or high in fat for periods of around 3 to 5 weeks prior to slaughter. This approach has been successful in reducing muscle glycogen levels, and in changing post-mortem muscle pH. However, responses in pork colour and water-holding capacity have been variable and inconsistent and the low carbohydrate diets have generally resulted in substantial reductions in the growth performance of the pigs. In a recent study carried out at the University of Illinois, feeding diets containing 10% supplemented fat for 4 weeks prior to slaughter reduced muscle glycogen levels, without negatively affecting growth rates. Although further research is needed in this area, it is possible that the use of high fat diets in late finishing could be part of a strategy to improve pork colour and water-holding capacity without negatively affecting growth performance or costs of production.

Dairy breeding for grazing systems

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Introduction In seasonal production systems feed requirements for production are matched to pasture supply, thus feed efficiency and achieving a single concentrated seasonal calving pattern are likely to be more important than in non-seasonal systems. Breeding objectives and choice of breeds are discussed, using New Zealand (NZ) as an example.

Breeds, strains and crossbreeding In countries where grazed pasture does not form the major part of cow diets, the predominant breed is generally Holstein. In NZ, Jerseys are comparable to Holsteins for overall genetic merit. In recent years crossbreeding has become popular, 28% of cows are currently identified as being crosses between Holsteins and Jerseys (an animal with 14/16ths or less of a single breed; LIC, 2005). To cope with multi-breeds and crosses, genetic evaluation has been multi-breed trait animal evaluation since 1996. The Production Worth index is designed to compare cows for their expected ability to convert feed to profit over a lifetime and includes heterosis. The Production Worth index was highest for Holstein x Jersey cows over the period 1985 to 2005 (LIC, 2005).

Breeding objectives The NZ dairy industry's breeding objective is to identify animals whose progeny will be the most efficient converters of feed into farmer profit. Traits included in this objective to form the Breeding Worth (BW) index are milkfat yield, protein yield, milk volume yield, liveweight, cow fertility, somatic cells and residual survival.

Liveweight Liveweight has been negatively weighted in sire selection indices in NZ since 1988. Increased liveweight has a major effect on feed demands for body maintenance. Consequently, increased liveweight entails a lower stocking rate of milking cows per hectare – and lower milksolids production per hectare for cows with the same genetic merit for per-cow milksolids performance. NZ cows have increased in body size by only a small amount since 1988, while maintaining near optimal rates of gain for the yield traits.

Fertility Maintaining a 365-day calving interval is an important management objective in grazing systems. The breeding objective for fertility in NZ is 42-day calving rate, chosen because it identifies animals that become pregnant early in the mating period. In addition, several other selection criteria are included in the analysis to improve the prediction of fertility breeding values; these include 2-year-old milk yield and body condition score in addition to submission rate, which is the probability of being presented for mating in the first 21 days of the mating season. Genetic trends for fertility show a decline in fertility in the Holstein-Friesian and Ayrshire breeds, with a small improvement in the Jersey breed. Despite the negative genetic trend in the predominant breed (Holstein-Friesian), phenotypic indicators of national fertility, such as calving interval, show that New Zealand farmers operate at close to optimal levels in calving interval. However, some management intervention is necessary to achieve this.

Mobilisation of body tissue Lactation length is determined by pasture availability, milk production of the cow and her body condition score (BCS). Cows of low body condition are often dried off earlier than contemporaries of higher BCS. Genetic correlation estimates between BCS and 270-day milkfat and protein yields are positive in early lactation and negative in late lactation (Pryce and Harris, 2006). Cows of high BCS in early lactation (spring) are more likely to have higher total yields of fat and protein because they have more reserves available for production in the autumn, when pasture availability is often limited. This contradicts the moderate negative genetic correlation estimates between production traits and BCS reported in studies from countries where non-grazing systems dominate. In grazing systems cows need reserves in later lactation to buffer milk production when pasture may be limited.

Genotype by environment interactions Interbull genetic correlations between countries (http://www-interbull.slu.se/eval/framesida-prod.htm) are weaker between countries where the main production systems differ, for example, the genetic correlation for protein yield between NZ and the UK is 0.75, between NZ and Ireland the genetic correlation is 0.85; which reflects the greater emphasis on pasture in NZ and Irish production systems, although some re-ranking sires would still be expected. The practical implication of substantial genotype by environment interactions is that sire choice would differ in each environment. Within NZ, clusters of systems defined using temperature-humidity index, altitude, herd-size and average herd milk production revealed no major re-ranking of sires (Bryant *et al*., 2006).

Conclusions Interbull genetic correlations imply that the choice of sires in systems where grazing is the main feed source differs from countries where greater feed supplementation is offered. Accounting for maintenance costs is important in breeding decisions for grazing systems. In NZ the Jersey is comparable to Holstein in overall genetic merit and when heterosis is included, the crossbred is most profitable. Maintaining 365-day calving intervals is a key management objective, and selection criteria differ from non-grazing systems. Feed constraints in the autumn period means that cows that can maintain body condition score in early lactation tend to have higher total yields.

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Genes and the environment

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The ability of an individual to change the phenotype in response to changes in the environment is called ***phenotypic plasticity*** or ***environmental sensitivity***. Plasticity has a genetic basis can be observed at the biochemical, physiological, behavioural, and other levels of the organism. Plasticity is often described by reaction norms, which describe the phenotypic expression of a genotype as a function of the environment Differences in plasticity between individuals (genotypes) will result in ***genotype by environment interaction (GxE)***, i.e. the difference between the expected phenotypic values of two genotypes is not the same in two environments. If the difference changes sign, we have re-ranking of genotypes. In Figure 1 four different situations of plasticity are described.

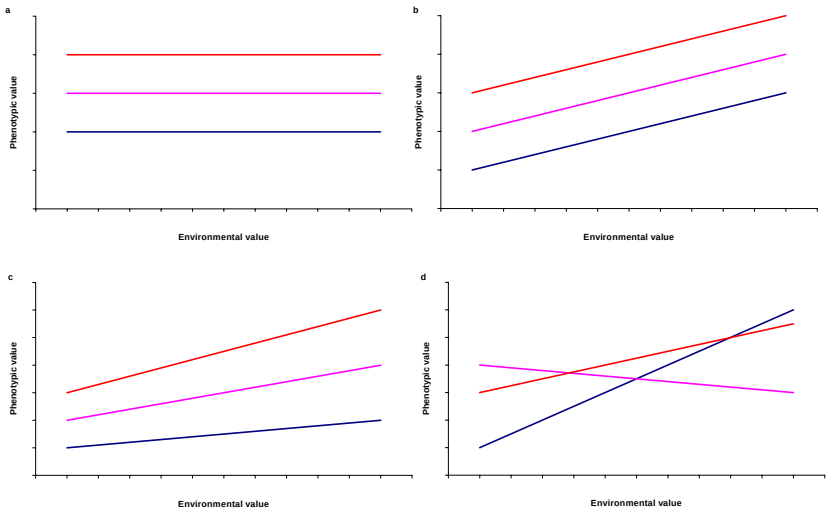

Figure 1. Description of the reaction norms for three genotypes. a) No plasticity but genetic variation in level. b) Plasticity but no variation in plasticity. c) Variation in plasticity but no re-ranking of genotypes. d) Variation in plasticity and re-ranking of genotypes.

There are basically three different methods to describe the extent of GxE. For all methods, observations on the same or related individuals in two or more different environments are needed. 1) The ***interaction term model*** is a simple extension of the usual genetic model to incorporate also the interaction between some classifications of genotypes and environments, e.g. breeds and feeding systems, respectively. The model is $P = G + E + GxE + e$ and if var(GxE) is different from zero we say that there is GxE. We do not directly know whether this GxE is of the type in Figure 1c or d. 2) In the ***multiple trait method*** we consider the phenotypic expression in two environments as two separate traits and if the genetic correlation between these two "traits" is different from unity, we have GxE (as in Figure 1d). We can also use the estimates of genetic variances to indicate GxE as in Figure 1c. 3) When the production environment can be described as a continuous variable, a third method called the ***reaction norm model*** is possible to use. In practice this means using a random regression approach where a reaction norm is estimated for each individual (or sire). The model can be described as: $P = G(E) + e$, where $G(\ )$ is the reaction norm function, which in its simplest case consists of an intercept ("level", corresponds to the usual breeding value) and a linear regression coefficient ("slope", a measure of plasticity). If we find genetic variation for the slope (or higher terms) we have GxE, as seen in Figure 1c-d. To distinguish between the two types we can calculate the genetic correlation between environments using a covariance function.

For dairy cattle, GxE has been studied for various environmental factors. Between herd production levels, feeding regimes or management systems within a country, or a group of neighbouring countries, there is seldom re-ranking of genotypes. Between countries or regions that differ considerably, *e.g.* in climate or management system, re-ranking of genotypes is more common. For example, the genetic correlation between the same milk production trait evaluated in any country in Western Europe, the USA, or Canada (the northern hemisphere group) is high (0.85-0.9), whereas the correlation between the northern hemisphere group and New Zealand and Australia is lower (0.75-0.84), indicating that re-ranking occurs. Low genetic correlations have also been estimated between milk yield evaluated in Mexico and the US (0.63), milk yield in the UK and Kenya (0.49), and longevity in Canada and New Zealand (-0.07—0.21). It was also found that there was GxE between environments with different heat-humidity indexes, with a lowest correlation of around 0.6 between low and high stress environments.

Up to now, few if any estimates of GxE between organic and conventional production systems have been published, but several studies are under way, both for dairy cattle but also for other species. Given reasonably large differences in conditions (feeding intensity, allowed substances etc.) one can expect some GxE to be found for several traits. At the total breeding goal level, there may also be some GxE, owing to different weightings of the breeding goal traits, whether there is GxE or not for the traits included in the breeding goal. This may lead to different selection decisions for the various production systems, and potentially separate breeding programs.

Predicting nutrient responses in poultry: future challenges

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Introduction The responses of poultry (broilers, laying hens and broiler breeders) to nutrients, discussed here, are in economically important outputs such as body weight (or protein) gain, breast meat yield, egg output, food intake and conversion efficiency, numbers of chicks produced per hen, etc. Of interest are the (usually) curvilinear responses of populations (flocks), which are the result of integrating the responses of the individuals making up that population. Populations of birds therefore do not have 'requirements' for nutrients: what nutritionists seek are the optimum economic dietary contents of each nutrient, and for this they need to know how populations respond to increasing dietary contents of the essential nutrients. Descriptions of such responses, whilst taking account of the marginal costs and revenues, are therefore invaluable in determining how to maximise or minimise the objective function chosen for any given commercial operation. Clearly, being able to predict these nutrient responses may be seen as the foundation of a successful nutritionist.

Measuring and interpreting responses It is intriguing to note that controversy still exists in the methodologies used to measure and interpret nutrient responses. For example, there are two general methods used to measure responses to individual amino acids; the graded supplementation technique and the summit dilution technique. Issues have been, and are still being raised regarding these techniques, which would be resolved by measuring instead the response to balanced protein, thereby eliminating issues of unbalanced or imbalanced amino acid mixtures. Various methods of interpretation of responses are still used in the literature in spite of the compelling review by Morris (19xx) that concludes by pointing out the value of including marginal costs and revenues in such calculations.

Responses of interest Recent evidence suggests that it is not only the conventional responses in broilers that need to be considered when maximising profitability, but that mortality and flock uniformity, food intake and breast meat yield appear to be influenced by the level of feeding and by the strain of broiler. Whereas some of these strain differences, such as their response in food intake to dietary protein content (Kemp *et al*., 2005), may be accounted for mechanistically in simulation models, others, such as the effect on uniformity and mortality, lack a mechanistic explanation, and can only be accounted for empirically. Perhaps the underlying mechanisms for these responses will eventually become known, at which time such models can be improved. It is clear that responses need to be strain-specific if these are to be of value when optimising poultry feeds.

Constraints to potential responses Social interactions and disease influence the response of poultry to nutrient supply. Whether such constraining factors can be modelled mechanistically is debatable, but the overall constraining effect of, for example, disease, stocking density and population size, on the population response may be subject to some general analysis. Reducing the rate of maturing parameter (B) in the Gompertz growth equation successfully simulates the resultant down-regulation of lean tissue growth (Wellock *et al*., 2005). This implies that stressors do not reduce the efficiency of utilisation of amino acids, and corroborative evidence (Gous, unpublished) indicates that this is indeed the case with stocking density and group size. However, certain diseases may cause a reduction in utilisation efficiency and this would be worth exploring further.

Whereas birds benefit in cold weather from the insulative properties of their feather cover, this thermal barrier constrains the amount of heat that may be lost to the environment in hot weather. As the potential growth rate of broilers is increased by genetic selection, their inability to lose sufficient heat to the environment is becoming a major constraint in commercial broiler operations worldwide. Accounting for all the factors that contribute to the environmental heat demand placed on the birds, such as temperature, humidity, wind speed and thermal radiation, and then accounting for the response of the bird to this 'effective' temperature, is a major challenge when modelling the response of broilers to nutrient supply. There is much still to be done in explaining these relationships.

Responses to minerals and vitamins Responses to major and minor minerals and vitamins have not received as much attention as have responses to amino acids, the philosophy being instead to promote the idea of 'recommendations'. This has limited the progress made in understanding the impact of these nutrients on performance. There may be some point in investigating the use of ratios (e.g. calcium to ME or to lysine) when measuring the responses to higher nutrient densities or higher amino acid: ME ratios in the feeding of broilers, where these nutrients may limit the maximal response in, for example, breast meat yield.

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The prediction of intake and performance of pigs: remaining challenges

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The Past

Since the early attempts of Whittemore and Fawcett (1974) a lot of progress has been made in our ability to predict the responses of pigs to nutrients. There are several simulation models that predict the performance of pigs under ideal environmental conditions when they are given access to a defined amount of food of a certain composition. Some of these models form the basis of a number of decision support systems used by the industry. A smaller number of models are able to predict the food intake of pigs, as well as their performance on foods of different compositions. These models agree that the pig needs to be described by two dimensions, at least, that relate to its maximum capacity to deposit lean tissue and its genetically determined level of fatness. The issue of whether pigs differ genetically in the manner that they partition limiting nutrients is a more contentious one.

The Present

The environmental components that are usually addressed in the above models relate to the physical dimensions, such as ambient temperature and air humidity. However, there are other aspects of the environment, which may influence significantly pig performance and in order to make better predictions these need to be accounted for. The focus of this talk will be on the problem of accounting for two of the most important groups of environmental stressors that can influence pig performance: the social and infectious environments. As far as social stressors are concerned, we have recently completed a model that includes the effects of the main social stressors; space allowance, group size, feeder space allowance and mixing. We have assumed that the effects of these stressors are mediated through (biochemical) factors that down regulate lean tissue growth, thus decreasing the capacity of the animal to attain its genetic maximum for lean tissue. In order to better predict the effects of such stressors on pig performance, we needed to describe the pig by an additional dimension: its ability to cope with social stressors (i.e. excitability). This description was achieved through close collaboration with animal breeders.

The Future

Accounting for the effects of infectious stressors on pig performance represents a significant challenge. The challenge arises from: (i) the need to describe the infectious environment adequately; this is more difficult than describing say, the physical or social environment, and it is likely that we would need to resort to using a proxy for its description, (ii) the description of the ability of pigs to cope with pathogens (resistance) and performance whilst challenged by them (resilience), and (iii) fully account for the direct (e.g. damage) and indirect (e.g. immune response) effects of the infectious stressors on nutrient requirements. If food intake is also to be predicted then the mechanism by which infectious stressors directly affect food intake (pathogen-induced anorexia) needs to be accounted for. A first modelling attempt that incorporates the effects of infectious stressors on pig performance will be developed in the presentation. The value of the attempt is mainly heuristic, as it will point towards the issues that need to be addressed, if we are to succeed in this task. It is obvious that progress can only be made by a close collaboration with the people who are responsible for the characterisation of the infectious environment and pig genotype.

Finally, the vast majority of models have been developed to predict the performance of an individual 'average', pig. In most instances, however, we are interested in the performance of a population, in terms of its mean and its variation. Accounting for between animal variation may be important when predicting nutrient requirements, optimising pig production systems (e.g. minimising variation in carcass weight) and devising breeding strategies. In this presentation we will develop an approach that is able to explore, and at least in principle, predict the performance of both individuals and populations differing in growth potential, initial state and ability to cope when raised under given dietary, physical and social environmental conditions.

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From nutrient requirement to animal response: predicting the profile of absorbed nutrients in dairy cattle

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Introduction Current feed evaluation systems for dairy cattle aim to match nutrient requirements with nutrient intake at pre-defined production levels. However, these systems were not developed to address, and are not suitable to predict, the responses to dietary changes in terms of production level and product composition, excretion of nutrients to the environment, and nutrition related disorders. Quantitative research into ruminant digestion and metabolism has expanded greatly (Dijkstra *et al.,* 2005) but has not (yet) given rise to practical, easy-to-use response systems. The change from a requirement to a response system to meet the needs of various stakeholders requires prediction of the profile of absorbed nutrients and its subsequent utilisation for various purposes. This contribution will examine the challenges to predicting the profile of nutrients available for absorption in dairy cattle and will provide guidelines for further improved prediction with regard to animal production responses, environmental pollution and animal health.

Representation of fermentation processes The level of detail adopted in predicting the profile of available nutrients depends on the objective. As a minimum, the availability of the major VFA, long chain fatty acids, amino acids and glucose has to be predicted. Thus the importance of processes in the reticulo-rumen is obvious. Much research into rumen fermentation is aimed at the determination of substrate degradation rates. Technical and methodological aspects of various *in vitro* and *in situ* systems for estimating substrate degradation have been studied extensively. However, data on ruminal retention times are essential to predict effective degradation in several situations. Yet quantitative knowledge on passage rates of nutrients out of the rumen is only a fraction of that on degradation rates, and thus should be an important theme in future research.

Equally, current systems largely ignore microbial metabolic aspects, whereas extant mechanistic models of rumen fermentation give only limited attention to explicit representation of microbial metabolic activity. Recent molecular techniques indicate that our knowledge on the presence of various microbial species is far from complete. Such techniques may give a wealth of information, particularly qualitative, on rumen micro-organisms. However, to include such findings in systems predicting the nutrient profile requires close collaboration between molecular scientists and mathematical modellers on interpreting and evaluating quantitative data. Protozoal metabolism is of particular interest here given the paucity of quantitative data.

Finally, processing of feeds, such as toasting, extrusion and expansion, can be used to manipulate the dynamics of nutrients in the rumen in order to optimise ruminant diets with respect to production, environmental and health issues. The effects of various feed processing methods are difficult to predict. Elucidation of the mechanisms underlying such treatments on molecular structures of nutrients by employing new techniques, especially proteomics, and the quantitative description of these mechanisms is another important research aspect in predicting substrate fermentation.

Environmental pollution and animal health Empirical models lack the biological basis necessary to evaluate mitigation strategies to reduce excretion of waste, including CH_4, N or P. Such models may have little predictive value when comparing various feeding strategies. For example, the Tier II models recommended by IPCC to quantify methane emissions do not represent differences in VFA profiles, and consequently methane production, that result from various feeding strategies. Similarly, the inadequate representation of N recycling to the rumen in current protein evaluation systems hampers the evaluation of feeding strategies of low protein diets to reduce N losses to the environment. Nutrient based mechanistic models can address such issues. Since environmental issues generally attract more funding from governmental bodies, further development of nutrient based models may well take place within an environmental framework.

To avoid metabolic disorders commonly related to high production levels, feeding cows correct types and levels of nutrients is important. Further development of nutrient based models offers good prospects for the direct prevention of conditions such as ruminal acidosis or ketosis. As the liver has a major role in uptake, metabolism and release of nutrients to other organs and tissues, with subsequent effects on the immune system and the reproductive physiology of cattle, prediction of the profile of absorbed nutrients at the portal vein is a key issue in addressing many metabolic disorders in dairy cattle.

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Towards a biological basis for predicting nutrient partitioning: the dairy cow as an example

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Introduction Prediction of animal responses to supplementary nutrients has long been recognized as a problem in nutritional science (Kellner, 1926) that has still not been solved (Hanigan *et al*.,2005). Another substantial problem is how to incorporate genetic differences into nutritional models (McNamara and Baldwin, 2000). These two problems are linked as their biological basis lies in the relative priorities of different life functions (growth, reproduction, health, etc.) and how they change both through time and under genetic selection. The purpose of this paper is to present recent developments in describing this biological basis and evidence in support of the concepts involved, using the cow as an example species.

The Problem We have tended to see partition as a mechanism for accommodating discrepancies between the composition of the feed and the composition of products (milk) with "overflow" being dumped in body reserves, i.e. partition as a homeostatic mechanism. Whilst this aspect of partition undoubtedly exists, there is another aspect of partition that is usually overlooked: partition that is due to the changing priorities of the animal through time, i.e. partition as a genetically driven, homeorhetic, mechanism. It is becoming increasingly clear that lack of progress in predicting nutrient partitioning is in large part due to the failure to consider this aspect of partitioning. Partition cannot be predicted from feed properties alone.

A Solution The cow has drives that relate to functions other than milk production, functions such as safeguarding reproduction, maintaining disease resistance etc. Recognizing that these drives and thus their relative importance are affected by genotype, and change with physiological state/age, is a key step. Evidence for genetically driven body lipid mobilization and a method to predict this are used to exemplify this concept. Given a biologically meaningful description of these genetic drives through time, it becomes possible to predict nutrient partition under non-limiting conditions. This can be built into a nutritional model as an average effect but how do we adjust this for different cows? The extent to which this is currently feasible, and important gaps in our current knowledge are highlighted. One substantial challenge is to create a genetic description of animals relevant to partitioning. Traditional index values are currently not so useful, in part because they ignore the time component. This is getting better with genetic parameters for shapes of e.g. lactation curves but there is still a gap and two major challenges: the problem of translating high-level descriptions of genetic drives to metabolic level descriptions, and the treatment of genotype-environment interactions. Substantial progress is now being made on both of these through studies of: gene expression, proteomics etc for the first challenge, and reaction norms and genetics of plasticity for the second.

Perspectives The genotype-environment problem can be looked at in terms of trade-offs. This places nutrient partitioning at the centre of the ways in which animals cope with competing demands. If we accept that there are a number of life functions, all with a genetic basis, we can look at them explicitly in terms of the partition of resources between them. We can choose to view the key genetic parameters as not being the total amount of milk but the proportion of resources directed towards milk, etc. This has a number of advantages. It would provide a clear way to characterize genotypes in models predicting partition. It will allow us to predict consequences of selection. It also provides the correct framework for including environmental effects. Under conditions where nutritional availability becomes a limiting factor, cows that differ in their genetic propensity to apportion, or trade-off, resources between e.g. current milk production and future reproduction will experience different degrees of depression in production. Genetic selection for production in such an environment can only be achieved by trading-off against reproduction. This can explain why we have seen a dramatic worsening of reproductive performance in cows in recent years. Some animals are better able to cope with changes in environmental quality, they have a greater ability to modulate the trade-off at the phenotypic level. Characterizing this plasticity in nutrient partitioning can only be done in the context of trade-offs. Modelling these trade-offs would be a powerful tool for progressing our understanding of these interactions and predicting the consequences of future selection strategies for increasing animal production.

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Effects of feed additives on the development on the ileal bacterial community of the broiler chicken

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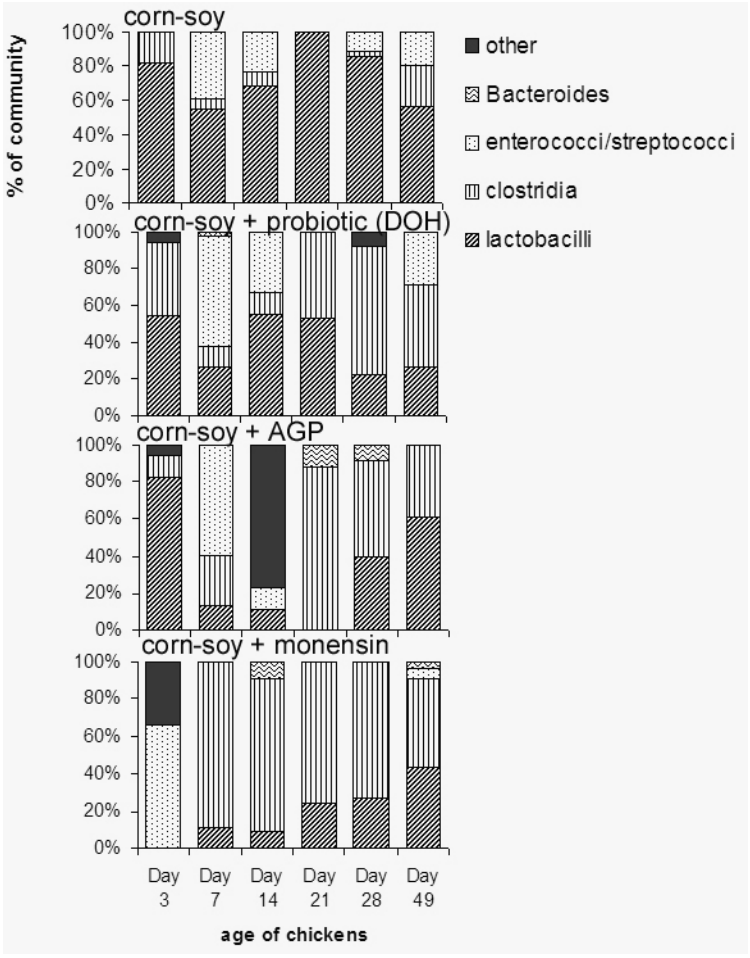
Introduction Intensifying concerns about the use of antimicrobials in meat and poultry production has enhanced interest in the application of prebiotics, probiotics, and enzymes to enhance growth and prevent disease in food animals. Growth-promoting antibiotics enhance growth of animals either by reducing the load of bacteria in the intestine, reducing colonization by intestinal pathogens, or by enhancing the growth and/or metabolism of beneficial bacteria in the intestine. Therefore other types of treatments could theoretically elicit similar responses as the use of antimicrobials.

Materials and mthods Much is known about the cultivable bacteria present in the intestinal tracts of food animals. Recently molecular ecology has been applied to study the noncultivable members of the animal intestine and has revealed a surprising diversity of uncharacterized bacteria inhabiting this ecosystem. We have been using DNA sequence polymorphisms of the small subunit RNA gene to better understand the microbial ecology of the chicken intestine and a detailed description of our studies can be found in published literature (Lu 2003a,b). This approach was used to determine whether feed additives would affect the development and composition of the ileal bacterial community. We evaluated the effects of an antibiotic growth promotion formulation (bacitracin-virginiamycin), monensin, and a commercial probiotic.



Results Our initial studies revealed that the bacterial community of the ileum of broiler chickens is primarily composed of gram-positive bacteria such as lactobacilli, streptococci, and enterococci. However, clostridia also comprised a large portion of the bacterial population at various times during the life of the broiler. These broiler clostridia are a relatively unknown group of anaerobes. While we detected known species of *Clostridia*, such as *Eubacterium*, *Weisella*, *C. perfringens*, *C. irregularis,* and *C. lituseburense,* many of the DNA sequences showed low similarity to clostridial species that have been cultured indicating that these organisms represent new members of the family. Figure 1 shows the composition of the ileal microbiota of broiler chickens over a 49 day grow-out. Other bacteria detected included *E. coli* and *Bacillus*.

Conclusions Both monensin and AGP have been shown to produce some growth-enhancing effects. In this study, these feed additives also enhanced the levels of ileal clostridia. Similar effects were seen in the probiotic group; this probiotic has also been shown to enhance growth and intestinal health. Strategies that modulate intestinal communities have experienced a resurgence in interest because of the desire to reduce the use of nontherapeutic antibiotics. Future studies should focus on characterizing the important bacterial species needed to stabilize the intestinal microbiota and identifying those commensals that stimulate and enhance development of intestinal function. New tools that allow detection of intestinal responses to the presence of symbiotic bacterial will allow rational design of antibiotic-free animal production strategies.

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Protozoal evolution in animal guts: from elephants to cockroaches

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Introduction Large herbivorous mammals such as cattle, sheep, goat or deer host "symbiotic" protozoa that are believed to be of importance for the digestion of the plant-based diets of their hosts. However, at least in under laboratory conditions, these mammalian hosts exhibit a rather normal digestive performance if the protozoa in the gastro-intestinal tract were removed by defaunation. A similar "commensalistic" relationship has been found between intestinal ciliates and arthropod hosts such as millipedes and cockroaches. In contrast, certain termites cannot survive without their symbiotic flagellates. Moreover, throughout the animal kingdom there is no simple correlation between the feeding habits of the hosts and the presence or absence of symbiotic/commensalic protozoa. Rather the taxonomic position of the host appears to be crucial for the evolution of the protozoal communities in the gastro-intestinal tract.

Materials and methods Arthropod and vertebrate hosts were analysed for the anatomy of their gastro-intestinal tract, for the presence of protozoa, and for methane/hydrogen production under taxonomic aspects. DNA-libraries of the 18S rRNA genes of the gut protozoa were generated and analysed phylogenetically.

Results As a rule, symbiotic protozoa can be found in the intestinal tracts of animals, which also host methanogenic archaea in their guts. Neither the size of the animal host nor its feeding habits are limiting their presence in more or less specialized compartments of the gastro-intestinal tract. Taxonomy and evolution of the hosts, on the other hand, are of crucial importance. For example, termites host peculiar flagellates in their microliter-sized guts, whereas cockroaches, which possess guts of a comparable size, host ciliates. Related ciliates are found in millipedes, frogs and reptiles, but not in warm-blooded animals. Marsupials and mammals host a different kind of ciliates, which co-evolved with their hosts – regardless whether the hosts possess differentiated foregut- or hindgut compartments. Molecular techniques allow analysing the composition of such gut communities, e.g. by generating PCR-based libraries of ciliate 18S rRNA genes. Phylogenetic analysis of the 18S rRNA genes of "type-strain" ciliates and of hundreds of such genes from community libraries revealed that all ciliates from elephants, horses and ruminants are monophyletic. Since also ciliates from marsupials belong to this group, a single evolutionary origin of the gut ciliates is likely, which can be dated to more than 100 million years ago.

Conclusions The ciliates living in the gastro-intestinal tract of warm-blooded mammals and marsupials are monophyletic. The ciliates living in cold-blooded vertebrates or arthropods belong to a different order of ciliates**.**

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Rumen bacterial diversity and its consequences in livestock agriculture

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Reducing the environmental footprint, allied with enhancing food quality and safety are key elements of the new agenda to improve the sustainability of livestock farming. Improving nutrient use efficiency is a key challenge in ruminant production systems with improvements in dietary forage utilisation, enabling reductions in use of cereal and/or protein rich supplements, being one of the primary means to achieve this goal (Kingston-Smith & Thomas, 2003). Consumers are also becoming increasingly aware of the relationships between diet and health, in particular food quality and safety. Research into functional food components has focused on increasing the quantities of those nutrients present naturally in ruminant products which are considered to play important roles in health promotion and disease prevention. Food safety on the other hand has focused on the ability to eliminate or decrease food-chain pathogens that can be harboured in the gut of ruminants in order to improve both human and animal health. Microbial transformations in the digestive tract (in particular the rumen) have a major impact on our ability to improve nutrient use efficiency, food quality and safety. The rumen microflora is very diverse making it a challenging ecosystem to manipulate. Application of cultivation independent methodologies has also revealed that only as little as 11% of the bacterial diversity has been cultivated to date (Edwards *et al*., 2004). This paper reviews how increasing our understanding of microbially mediated processes in the rumen is central to achieving improvements in nutrient use efficiency, food quality and safety.

Ruminants typically convert only ~ 20% of dietary protein into saleable product (i.e. meat and/or milk). A large part of this inefficiency relates to rumen processes and in particular the efficiency of microbial growth. Cultivable rumen bacteria are considered to play a major role, with specific bacterial species known to contribute to the inefficiencies associated with the protein degradation pathway. It is now also established that in fresh forage systems plant-mediated proteolysis can play a substantial role in initial protein breakdown in the rumen (Kingston-Smith *et al.* 2005). This plant-based activity generates amino acids and short chain peptides which can be utilised by the proteolytic rumen microorganisms, potentially giving a very different nitrogen fingerprint for input into the rumen proteolytic pathway compared with that derived from conserved forages. Hence, differences in ruminal microbial colonisation and ecology are likely to occur depending on how diets are presented (i.e. fresh, conserved or concentrate-based), so contributing to observed differences in efficiency of protein utilisation by the whole animal.

Studies in relation to enhancing the nutritional value of ruminant products have focused most attention on lipids, particularly in relation to reducing the content of less desirable saturated fatty acids (SFA), and increasing beneficial omega-3 polyunsaturated fatty acids (PUFA) and conjugated linoleic acid (CLA). Dietary PUFA are rapidly hydrogenated in the rumen by microbes, resulting in the production of SFA (principally 18:0) but it also results in the formation of CLA and trans monoene intermediates (Palmquist *et al*., 2005). It is now recognised that this desaturation pathway is carried out almost exclusively by specific rumen bacteria. Understanding of the pathways and the key microbial species involved in biohydrogenation has increased in recent years with the objective of evolving strategies of manipulating the balance of the biohydrogenation pathway to enhance beneficial fatty acids in ruminant products.
Research on the incidence and control of food-chain pathogens in ruminants (i.e. *Escherichia coli*, *Salmonella* spp. and *Listeria monocytogenes)* has focused on factors that affect their prevalence and survival, or on 'hygiene' measures to limit their impact later in the food chain (Leitch *et al*. 2001). Little emphasis has been placed on developing strategies for tackling the problem at the source of contamination on farm. Understanding the relationship between faecal shedding of pathogens, their source (soil, feed, farm waste) and presence in the ruminant gut relative to feed type and composition is crucial and yet basic knowledge of the ecology of food chain pathogens in the gut and the effects that both diet and symbiotic gut microbial communities have on the survival and pathogenicity of these microorganisms is currently unavailable.

In conclusion, characterization of the activity and function of the rumen microflora will aid in the development of novel strategies to increase process efficiency and improve food quality and safety in ruminants.

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Influence of secondary plant metabolites on intestinal microflora in livestock

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Introduction Secondary metabolites represent a diverse group of natural products. Although precise numbers are, at best, an estimate, at least 100,000 different compounds of natural origin have been described, of which at least 80,000 of those are derived from plants. It is likely that many more than these will be isolated in the future, as increasing numbers of novel plant species are being identified and studied for their potential use in the pharmacological, medical and agricultural industries. The increased interest in phytochemicals in animal diets has been spurred on by the EU ban on the use of 'in feed' antibiotics, the removal of animal proteins from the diet and thus the increased variety and inclusion levels of vegetable protein sources.

The most frequently encountered secondary metabolites include alkaloids, phenolics from the group classified as tannins, terpenes from essential oils, saponins, and some non-protein amino acids, mostly at concentrations <10 g/kg; their effects can range from influences on growth promotion, antimicrobial or antifungal effects, feed intake, defaunation, antinutritional effects or even toxicity when ingested by animals (Acamovic *et al*, 2004). Of this vast range of potential feed additives or supplements, perhaps the most interesting are the tannins and the essential oils, and many of these have been tested widely in a number of livestock species. Condensed tannins (proanthocyanidins), at concentrations greater than 50 g/kg of feed, are known for their antinutritional effects, by complexing with protein, carbohydrates and minerals to reduce feed intake and restrict nutrient utilisation. At lower concentrations however, they can have protein-sparing effects in ruminants, and in poultry, may reduce the colonisation of the gut with pathogenic microorganisms. Sesquiterpene-containing essential oils have a wide range of effects depending on concentration, composition and animal species. In poultry, a growth enhancing effect of essential oils has been found (Denli *et al*, 2004), while in other studies, essential oils appear to stimulate digestion (Lee *et al*, 2004). Some essential oils appear to have selective antimicrobial effects against poultry pathogens such as *Clostridium perfringens*, or *Eimeria sp*. Antioxidant activity (resulting in reduced lipid oxidation in long-term frozen stored chicken meat) and hypocholesterolemic effects have also been reported in chickens (Botsouglou *et al*, 2003). Individual components of an essential oil, of which there may be up to 100 in any particular oil, have a wide range of activities and may act in additive, synergistic or antagonistic fashions. At high concentrations, essential oils can be toxic. Although essential oils have been studied most for their antimicrobial activities, their effects in livestock may not be confined to the microflora, but may extend to animal metabolism, and some blends of essential oils are commercially available for livestock use. An essential oil blend inhibited ruminal degradation of soyabean meal *in situ*, and reduced amino acid deamination *in vitro*. Both effects are probably by inhibition of rumen microbial activity. Screening of various plants species for bioactive extracts has also demonstrated that the essential oils can modify ruminal fermentation characteristics. Therefore although dietary essential oils may be used as alternatives to antibiotics, they may also have positive effects on growth performance; whether this is a consequence of anti-microbial activity or direct effects on other metabolic functions is not known.

This review will summarise the evidence for antimicrobial effects of secondary plant metabolites, including essential oils and tannins, on intestinal microorganisms, as well as positive effects on animal growth and performance, and will discuss current research aimed at elucidating their mechanisms of action.

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Gut microbiology - broad genetic diversity, yet specific metabolic niches

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Latest estimates based on comparisons of 16S rRNA-encoding gene sequences from gut microbial ecosystems reveal a daunting genetic diversity, with at least 341 distinct phylotypes evident in the ruminal bacteria (Edwards *et al*. 2004) and 395 in the human intestine (Eckburg *et al*. 2005). Some metabolic functions are fairly widespread throughout the genetic spectrum, such as glucose utilization, for example. Others, however, are not. In spite of so many phylotypes being present, single species or perhaps only two or three species often carry out key functions. Among ruminal bacteria, only three species can break down highly structured cellulose, despite the prevalence and importance of cellulose in ruminant diets, and one of those species, *Fibrobacter succinogenes*, is distantly related to the most abundant ruminal species. Fatty acid biohydrogenation in the rumen, which has so many health implications in terms of the fatty acid composition of ruminant-derived foods, is effectively carried out only by the *Butyrivibrio* group. Furthermore, the final step of biohydrogenation of C18 fatty acids, stearate formation, can be achieved only by a tiny sub-group of the butyrivibrios. Single bacterial species can also have very distinctive effects on the health of the host animal or human. *Oxalobacter formigenes* is a member of the ruminal and human intestinal microflora. It converts oxalic acid to formic acid, thus removing the potentially toxic oxalic acid, which is present at high concentrations in some foods such as rhubarb and spinach. In some individuals, however, *O. formigenes* cannot be detected, perhaps having been eliminated by antibiotic therapy. The resulting inability of the intestine to metabolise oxalic acid can cause kidney stones composed of calcium oxalate. Re-inoculation of these individuals with *O. formigenes* results in relief from disease. In ruminants, the conversion of tryptophan to indoleacetic acid and then to 3-methylindole causes bovine pulmonory disease. Elimination of the *Lactobacillus* species responsible for the indoleacetic acid to 3-methylindole conversion corrects the problem. Finally, perhaps the most celebrated example of the difference a single species can make is the 'mimosine story' in ruminants. Mimosine is an amino acid that is found in some plants, including *Leucaena leucocephala*. *Leucaena* had been identified as a potentially useful protein-rich feedstuff for Australian livestock. However, the mimosine caused thyroid problems by being converted to the goitrogen, 3-hydroxy-4(1H)-pyridone, in the rumen. Observations that mimosine-containing plants were not toxic to ruminants in other continents led to the discovery of *Synergistes jonesii*, which, when inoculated into Australian cattle, metabolised 3-hydroxy-4(1H)-pyridone and protected the animals from toxicity. Thus, in the almost overwhelming flood of information that emanates from molecular microbial ecology and genomics of gut bacteria, it should never be forgotten that this vast community consists of many important metabolic niches inhabited by species each with specific metabolic capability.

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