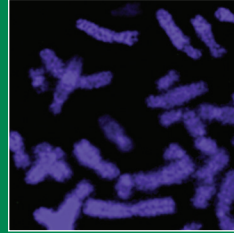


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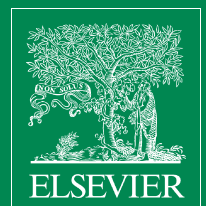
# animal – science proceedings

The Challenge of Change  
The New Normal?

Proceedings of the British Society of Animal Science  
12th – 15th April 2021  
On-line Virtual Conference



animal – science proceedings





## Welcome

The British Society of Animal Science (BSAS) aims to provide an opportunity for those with an interest in animals and animal science to exchange views, ideas and information. It is an energetic and active society with members from countries throughout the world. Today, as ever, the Society is the natural connecting point for all of those with an interest in animal science and related sectors. Its membership is drawn from research, education, advisory work, commerce and practical animal keeping.

At the conference the animal consortium of BSAS, EAAP and INRAE will launch the *animal* family of Gold Open Access journals, published by Elsevier.

- The well-established journal *animal* publishes the best, innovative and cutting-edge science that relates to animals (farmed or managed) used for animal production but now two other linked journals.
- *animal – science proceedings* (formerly Advances in Animal Biosciences) will publish high-quality conference, symposium and workshop proceedings on aspects of the life sciences. The BSAS conference is the first issue.
- *animal – open science* is a new publishing initiative of the consortium. The journal fully embraces Open Science. All relevant activities (research, extension, teaching) in the field of animal science that are well-carried out deserve to be published and contribute to enhanced knowledge. Scientific exchange and interactions with the authors on articles will be through a platform of post-prints commentaries rather than conventional peer review.

Further information can be found on [www.animal-journal.eu](http://www.animal-journal.eu)

BSAS organises major scientific and specialist conferences on key issues facing the science related to animals.

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# Proceedings

of the British Society of Animal Science  
Annual conference 2021

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## Proceedings of the British Society of Animal Science Annual Conference 2021

### Invited Speaker Summaries

#### 1. Opportunities for bioscience research in the 'new normal'

Melanie Welham

BBSRC, UKRI

**Introduction:** The Biotechnology and Biological Sciences Research Council (BBSRC), part of UK Research and Innovation (UKRI), invests in world-class bioscience, pushing back the frontiers of biology to deliver a healthy, prosperous and sustainable future.

BBSRC's strategy for UK bioscience is built around three long-term, high-level objectives: (i) Advancing the frontiers of bioscience discovery; (ii) Tackling strategic challenges, and (iii) Building strong foundations. As evidenced by the COVID19 pandemic, animal studies and models are crucial for our understanding, treatment and prevention of disease, highlighting the importance of One Health approaches. However, the impact of animal science, health and wellbeing is much broader. These are key areas that BBSRC strongly supports, particularly in addressing our strategic challenges 'bioscience for sustainable agriculture and food' and 'biosciences for an integrated understanding of health'.

The past 12 months have been challenging for many and the UK research community's response to the pandemic has been phenomenal. At UKRI we have adapted our processes, practices and ways of working to support the research community. One example being the introduction of a rapid response call to support short-term projects addressing and mitigating the health, social, economic, cultural and environmental impacts of COVID19. However, the economic and social impact of the pandemic will be significant. This is a critical time for research and innovation in the UK as we look to build a resilient, inclusive and sustainable economy in response to the many challenges we are facing, both nationally and globally. This presentation will consider what lessons we can take forward from the pandemic, including the opportunities for animal science specifically, and also reflect on how UKRI can support the development of a more diverse and inclusive research and innovation ecosystem.

#### 2. Plus ça change, plus c'est la même chose

David A. Hume

Mater Research Institute-University of Queensland, Brisbane, Australia

**Introduction:** The more things change, the more they stay the same" is an aphorism that can probably be applied to the challenges associated with leaving the EU. As an Australian/British dual national, I hope Brexit will at least open up markets for Australian agricultural products and lead to the funding and renewal of traditional collaborations. However, the aphorism cannot be applied to the revolutions associated with genomic technologies and big data. One of the many outcomes of these revolutions is the need for new skills in the life sciences, not least much greater inputs from the "hard" sciences. Everything that we eat

is already genetically modified through the combined impacts of mutagenesis and selection. But genomic selection and editing are a genuine step change. Reflecting on 10 years as Director of the Roslin Institute, I will talk about the accelerating pace of change in genomic technologies, how they may impact on livestock agriculture and the policy and management challenges that they present.

#### 3. Facing into the future of livestock production

Nicholas Saphir

Agriculture and Horticulture Development Board (AHDB), UK

**Introduction:** In 2020 the world changed completely and rapidly, but change has been coming for a longer period for agriculture. In this presentation we explore where those changes began, what is driving them over the longer term, and the seismic shift driven by environmental challenges, antibiotic and zoonotic disease scrutiny, changing global trading patterns, upheaval in policy and payments, and of course the new normal post Covid.

Balancing the nutritional needs of a growing population with the drive towards sustainability, whilst not forgetting the absolute necessity of productivity and profitability to create a robust and stable livestock farming industry for the future are key factors in preparing for change. This presentation by the Chair of AHDB Nicholas Saphir, tasked with creating a levy body fit for the future, and creating a long term vision for livestock farming, will give a broader perspective of the challenges and opportunities ahead, and how science will be the driver and supporter of this change.

#### 4. Net zero carbon & UK livestock

Elizabeth Magowan

Agri-Food and Biosciences Institute (AFBI)  
Director, Sustainable Agri-Food Sciences Division

**Application:** The UK livestock industry has a complex carbon footprint, but due to its important role in the UK economy and fabric of rural life, as well as what it provides to sustain human and environmental wellbeing in many respects, efforts must be urgently applied to progressively and aggressively deliver livestock's contribution to the UK's net zero carbon goal. The UK has made significant progress in understanding and reporting the emissions associated with livestock production in recent years. There have also been major advances in our capacity to mitigate emissions through a range of innovative management interventions targeted at the core livestock industries.

**Introduction:** However, reducing the carbon footprint of livestock farming enough to achieve net zero in the UK will be a major challenge. The report highlighted that using all known cost effective greenhouse gas mitigation options, at a high rate of adoption, the UK farming sector could achieve approximately 19% of the greenhouse gas reductions

required by the agriculture and land use sector by 2035, against the 2050 target of 64% reduction within the agricultural sector.

**Materials and methods:** The report highlighted eight key areas where focus needed to be applied, namely improved efficiency, design and adoption of novel and alternative feeds, reducing and redefining the use of nitrogen fertilizer, development and adoption of smart technology and precision livestock farming techniques, enabling and enhancing knowledge with regard to carbon sequestration and accounting for it, developing decision support tools to aid the understanding of whole systems, enhancing our calculators and finally filling data gaps to improve the reporting of emissions and addressing the uncertainties. The report recommended significant investment in time, strategizing and finances are required, as well as a renewed focus on carbon accounting tools and data sets and finally accelerated and increased effective methods to enable education, knowledge exchange and adoption were urgently required.

**Conclusion:** Overall, it is vital to remember that a single focus on carbon may result in a system which is skewed, under-delivering in other vital sustainability metrics such as nitrogen and phosphorus pollution, animal behaviour (positive welfare), and rural communities; social wellbeing and the economy. These aspects must be considered in concert, through a multi actor approach, to achieve a truly sustainable livestock industry which delivers to both human and planetary health.

## 5. Pig amino acid metabolism in immune regulation

Yulong Yin

Institute of Subtropical Agriculture, The Chinese Academy of Sciences, China

**Introduction:** Amino acids (AAs) are protein's building blocks, which could help body produce energy and form tissues. There are 20 different AAs, nine of them are called "essential" and 11 of them are labelled as "non-essential". Nonessential AAs can be made by the body, while essential AAs cannot be made by the body. AAs deficiency or disorder cause a number of disease states, nutritional deficiencies, fatigue, accelerated aging, and even premature death. In recent years, the role of AAs has been highlighted in immune regulation. Findings from our previous studies indicated certain functional AAs and their metabolites could involve in the control of immune response. Firstly, glutamine (Gln) could promote intestinal sIgA production and reduce proinflammatory cytokines secretion. In pig model, Gln enhances antiviral innate immunity through mTOR-type I IFN in small intestinal epithelial cell. It has been demonstrated that Gln affect Th17 cells differentiation through remodelling intracellular metabolic pathways. Accordingly, alpha-ketoglutarate (AKG) involved in the regulation of macrophage polarization. Secondly, GABA plays an important role in maintaining intestinal immune barrier. GABA could regulate Th17 cells function and macrophage polarization, as well as the sIgA secretion through mTOR pathway. Thirdly, aromatic AAs and their metabolites are key regulators of intestinal inflammatory response. For example, tryptophan (Trp), which mediates the host-microbial immune crosstalk, can affect the differentiation of T cells by degrading to 5-hydroxytryptamine (5-HT). Last but not least, branched-chain AAs can also improve the mucosal surface defense and inhibit pathogen introgession into the lamina propria by stimulating the secretion of sIgA. Meanwhile, leucine (Leu) metabolite HMB regulates muscle related inflammatory responses via activating Akt-FoxO3a signalling, and promotes type I IFN expression through sensing protein Sestrin 2 - mTOR signaling. Now, AAs have become cost-effective nutraceuticals for improving health and preventing infectious disease in both humans and animals. Although great advances have been made in the rapidly growing field of nutritional immunology, there is a paucity of information about the molecular mechanisms that regulate the actions of amino acids on the immune system.

## 6. Carbon net zero approaches for South American livestock

Alexandre Berndt

Brazilian Agricultural Research Corporation – EMBRAPA, R&D, EMBRAPA Southeast Livestock

**Introduction:** The social isolation and the greater permanence of families in the homes, caused by the pandemic, strengthened feelings of self-preservation and a greater search for quality of life. Physical and mental health, social life and preservation of the environment were valued in the "new normal". In this context, the concern with climate change gained prominence, as well as initiatives in the sense of decarbonizing the various human activities, including food production.

The challenge of reaching the goal of carbon neutrality is international, but the solutions to achieve it will be local and distinct, considering the regional characteristics of climate, soil, producers' aptitude, public policies and available technologies. This challenge will be overcome with the development of science and technology, raising the awareness of rural producers, agricultural leaders, private companies, governmental and non-governmental organizations, with training and rural extension efforts. South America has important agricultural production and many countries have included mitigation actions in this sector in their NAMAs. Livestock specifically adopts technologies to mitigate and fully offset emissions, especially enteric methane, the main greenhouse gas emitted "on farm". In Brazil, for example, a large meat processing company launched in August 2020 a line of cuts with the "Neutral Carbon Meat" label, initially intended for the national market. The concept of Neutral Carbon Meat is based on the total compensation of enteric methane emissions by removing carbon in tree trunks planted in crop-livestock-forest integration systems, known as ILPF. This compensation considers the stages of rearing and finishing beef cattle in tropical pastures, receiving or not supplementation.

To achieve the neutralization of emissions, a gradual path of compensation can be drawn, reaching different levels that can be considered low carbon production levels. Local productive arrangements, integrated with national and international markets, will guide actions and define the scope of these initiatives on a global scale.

## 7. Why should African livestock production have to be climate "neutral"?

Polly Ericksen

International Livestock Research Institute (ILRI), Sustainable Livestock Systems

**Introduction:** Proponents of livestock production in Africa have struggled for more than a decade to combat the narrative that all livestock production is bad for the environment and is a major contributor to greenhouse gas emissions (GHGe). Undoubtedly industrial and large scale livestock production is a significant driver of deforestation and a major source of GHGe. However, in most African low and middle income countries, where small scale production is the norm and the majority of livestock are reared in dryland agro-ecologies, livestock production must be understood in a different context and using different data. Overall emissions from African systems are much lower than in OECD systems. Their GHGe intensities are high, but this is due to the low productivity, low input nature of these systems. Hence intensities can be reduced as productivity increases. But livestock serve multiple functions in African agriculture. They are assets that buffer against shocks. They are the best adapted commodity to be produced in the extensive dryland rangelands. They are integrators that tighten nutrient cycles in mixed crop livestock systems. They maintain biodiversity in grasslands. Furthermore, livestock in Africa are more vulnerable to climate change than those in OECD countries, yet to date most of the attention has been on reducing GHGe from livestock, rather than recognizing their role in sustainable and income generating

agricultural systems. Based on a decade of research at ILRI, I will present data showing how the narrative in SSA about “climate neutrality” needs to change for livestock production so that these systems, which are essential for not only social and economic growth but also agricultural sustainability, can continue to flourish.

## 8. Towards net zero: CSIRO mission to agricultural opportunity

Michael Battaglia

CSIRO (Commonwealth Science and Industrial Research Organisation), Sustainability, CSIRO Agriculture and Food

**Introduction:** Governments and businesses around the world have set net zero attainment dates. Need and aspiration are high but the pathway is unclear, with both significant technological and economic and socio-political implementation barriers. CSIRO as Australia’s national science agency has catalysed a Toward Net Zero mission that looks at creating supply chain and precinct/place based exemplars of net zero that will assist in technology implementation and building the institutional coupling required to create economic and social and environmental value from such a transition. Agricultural and the land sector is critically involved in these aspirations and can be a significant net beneficiary. And for many nations, Australia included, the livestock represents a key source of emissions that must be considered to enable this transition. This presentation will finish with a description of three emerging technologies to address livestock sector emissions and will particularly focus on the use of *Asparagopsis* as a livestock feed supplement.

## 9. UKRI approaches to international research

Tim Willis

UKRI-BBSRC, UK

**Introduction:** Tim will cover current Government and UKRI approaches to international engagement and activities in research and in partnership within Europe, globally and research for development.

## 10. Innovation in the livestock sector – The role of stakeholders

Cledwyn Thomas

EAAP Rome, Italy

**Introduction:** The paper will examine the role of users/stakeholders in the process of innovation in the livestock sector. Science-driven research identifies emerging science that can potentially solve a problem and involves stakeholders mainly in dissemination/demonstration. It measures success in terms of scientific quality primarily through output of peer reviewed publications. On the other hand innovation driven research defines a problem that is emerging in practice (technical/financial/societal). Stakeholders are actively involved in defining the problem and are active during the process in determining research priorities. Success is judged as relevance to the sector and the extent of implementation of the project outputs. The involvement of users can take the form of stakeholder advisory groups/platforms to guide the work and aid dissemination. However, in most part, this results in token representation and in reality the project continues to be science driven with the research groups involved seeing success as numbers of publications in high impact factor journals. To foster an innovation rather than science driven approach instruments such as the European Innovation Partnerships (EIP) were created to ‘facilitate the information flow between research and practice’. Partnerships were formed for the Beef and Dairy sectors but it is still not clear if they are meeting the long-term EIP goals.

The paper will examine projects that have specifically adopted an innovation strategy and created a participatory approach in the research

programme. For example, iSAGE used a multi-actor approach in which 18 of the 34 project partners represented sheep and goat producer groups that are closely linked with processors and retailers. The practical knowledge of these groups informed the research programme in evaluating and developing new practices that are relevant to the whole sheep and goat sector.

## 11. Science into practise

Sara Place

Elanco Animal Health, United States

**Introduction:** Science is the ability to know. In the past year, what we know and the most appropriate way to react to the pandemic and economic challenges has required agility. In livestock agriculture, disruption of supply chains necessitated new solutions to lessen stressors on human and animal welfare and a shift to new realities of point-of-sale (e.g., shift from food service to retail). Additionally, while the pandemic has put a renewed interest on the basic, day-to-day necessities, focus on larger, longer-term challenges has not wavered. In the past year, companies and supply chains have continued their focus on sustainability, the creation of goals (especially as they related to greenhouse gas emissions), and advancement of data-driven strategies to track progress in livestock supply chains against sustainability goals. This presentation will cover some of major trends in innovation that are affecting the livestock industries.

## 12. From concept to reality

Anne Richmond

Moy Park, Research and Development, UK

**Introduction:** This presentation will give an overview of Moy Park and their uniquely integrated poultry production system. As well as giving an overview of R&D activity, it will detail their work regarding the implementation of windowed poultry housing and platform perching. It will also provide some insight into future research challenges and priorities.

## 13. Animal agriculture and alternative meats: learning from past science communication failures

Alison Van Eenennaam

Department of Animal Science, University of California, Davis, USA

**Introduction:** The new normal for scientists globally is how to best respond to misinformation. Some in the alternative meat industry are promising to “end world hunger”, and eliminate animal-sourced foods. This dichotomous framing leaves animal scientists in an awkward position. Much of the rationale behind alternative meats invokes an overly simplistic narrative, especially as it relates to greenhouse gas (GHG) emissions, the nutritional attributes and dietary importance of animal-sourced foods, and the ecosystem services of grazing ruminant production systems. Not to mention the interests of the one billion people involved in the rearing, processing, distribution and sale of animal products, with half being smallholder farmers reliant on livestock for their livelihood. Perhaps now is an opportune time to communicate how producers of both alternative and conventional meats are using science and innovation to implement more sustainable approaches to produce protein and essential dietary micronutrients. And that jointly, we need to tell compelling stories to counteract the possibility that misinformation, fear, and propaganda will inhibit the adoption of safe innovations in culturally-appropriate food production systems worldwide, to the detriment of global food security.

#### 14. Rescaling genetic expression of carcass traits in cattle by environment using herd best linear unbiased estimates (BLUES)

David Kenny<sup>a,b</sup>, Roy D. Sleator<sup>b</sup>, Craig P. Murphy<sup>b</sup>, Ross D. Evans<sup>c</sup>, Donagh P. Berry<sup>a</sup>

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<sup>b</sup>Munster Technological University, Bishopstown Campus, Co. Cork, Ireland

<sup>c</sup>Irish Cattle Breeding Federation, Bandon, Co. Cork, Ireland

**Application:** Herd BLUES are generated from routine genetic evaluations and represent the contribution of herd environment to animal performance. The impact of herd BLUE on the response to selection for carcass traits could be used to determine the herd genetic merit required to generate carcasses with desired attributes.

**Introduction:** Genetic evaluations separate phenotypes into their additive genetic components (i.e., estimated breeding values (EBVs)), and their environmental and non-additive components (i.e., BLUES). To date, only the EBVs generated from genetic evaluations have been retained and published. Nonetheless, the often discarded BLUE solutions could have tangible applications in herd management. For instance, the BLUE solutions for herd-year contemporary groups could be useful as they represent the environmental contribution, independent of the genetic contribution, to animal performance. The objective of the present study was to determine if the response to selection for carcass traits in beef cattle varied by herd BLUE.

**Materials and methods:** After edits, carcass weight, conformation, and fat score information was available for 4,616,761 animals from the Irish Cattle Breeding Federation database. The dataset was split into a

validation population (229,773 cattle slaughtered in 2019) and a calibration population (the 4,386,988 remaining animals). Univariate national genetic evaluations were conducted for each carcass trait in which the phenotypes of the validation animals were masked. The EBVs of the validation animals, and the contemporary group BLUES for the validation herds, were estimated from the univariate genetic evaluations. Herd BLUES were generated as the weighted average of all contemporary groups BLUES from a single herd, with each herd stratified separately for each trait into one of five groups based on the percentile rank of the herd BLUES. Multivariable linear regression models were used to quantify the response to selection for the carcass traits in the different BLUE strata.

**Results:** The response to selection for carcass weight, conformation and fat varied by herd BLUE (Fig. 1). For example, the response to selection for carcass weight in herds ranked in the lowest, average and highest 20% of herd BLUES for carcass weight was 0.73 kg, 0.79 kg and 0.96 kg, respectively, per unit increase in the corresponding EBV.

**Conclusion:** Differences in the association between genetic merit and animal performance by herd environment represent deviations in animal performance for the same genetic merit. In the case of carcass traits, such deviations can have economic implications due to the desired thresholds imposed on carcass traits by many meat processors. Should the herd BLUES and the response to selection estimates corresponding to the BLUES be deployed, perhaps through a decision support tool, producers could make better informed breeding decisions that are bespoke to their herd.

**Acknowledgement:** Funding from the Department of Agriculture, Food and the Marine (RSF 17/S/235; GreenBreed) is gratefully acknowledged.

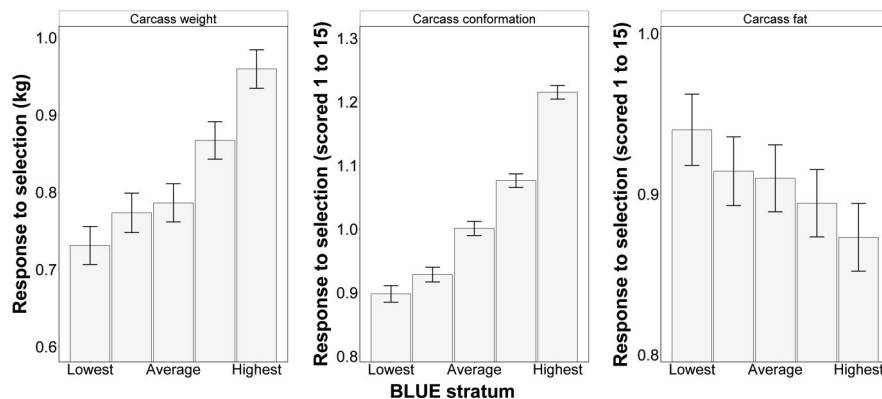


Fig. 1. Response to selection (95% confidence interval as error bars) for carcass weight, conformation and fat in the various corresponding BLUE stratum.

## 15. Breed composition in cattle estimated using SNP-BLUP and admixture

Cliona Ryan<sup>a,b</sup>, Donagh Berry<sup>a</sup>, Aine O'Brien<sup>a</sup>, Thierry Pabiou<sup>c</sup>,  
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<sup>c</sup>Irish Cattle Breeding Federation, Bandon, Cork, Ireland

**Introduction:** Genotypes are now widely available, and while they have predominately been used for genomic evaluations, genotypes also have other potential applications such as parentage verification and discovery, and breed evaluation. While software exists to estimate breed composition, the ability to exploit existing pipelines used in genomic evaluations for breed prediction could be advantageous and less computationally intensive. The objective, therefore, of this study was to use a large database of purebred and crossbred cattle genotype to compare breed composition predictions using a freely available software, Admixture, with those from Mix99 SNP-BLUP.

**Materials and Methods:** Genotypes from 52,655 single nucleotide polymorphisms (SNPs) were available for 703,078 dairy and beef cattle. Following quality control edits, 49,221 SNPs remained across 703,078 animals; sporadically missing genotypes were imputed. Purebred animals for 13 breeds were verified using principal component analyses as well as with Admixture. Two-way and three-way crossbred animals identified using admixture were retained for further analysis. The allele substitution effect per SNP for each breed was estimated using SNP-BLUP and summed to generate an estimate of the proportion of each breed in each animal. A total of 6,000 previously verified purebred animals across 13 breeds were included in the training population and with a further 8,000 identified crossbred and purebred animals included in the validation population. Breed predictions were compared using a Bland-Altman plot, which highlights the difference in the mean predictions of both methods.

**Results:** The majority of the variability between the two methods in predicting breed composition was observed in the crossbred animals;

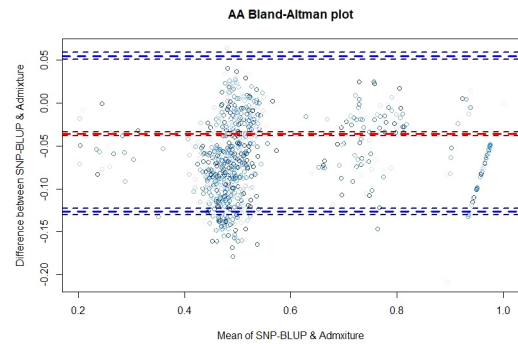


Fig. 1. Bland-Altman plot demonstrating the difference between SNP-BLUP and Admixture for Angus breed predictions.

overall both SNP-BLUP and admixture gave similar breed proportion estimates, with a mean difference of 0.0358 between both methods for predicting Angus proportion, for example. The majority of the predictions resided within the limits of agreements, calculated as the mean standard deviation of the difference (Fig. 1).

**Conclusions:** There was a strong similarity in predicted breed composition per animal between both approaches. This suggests that breed composition estimations could be readily integrated into the SNP-BLUP pipelines used for genomic evaluations thus replacing stand-alone software.

**Acknowledgement:** Funding from the Department of Agriculture, Food and the Marine (Dairy for Beef) is gratefully acknowledged.



## 16. Evaluation of UHF technology in transportation of cattle

Jenna Bowen<sup>a</sup>, David Kerr<sup>b</sup>, Alistair Robson<sup>b</sup>, Lewis Mackenzie<sup>b</sup>, Carol-Anne Duthie<sup>a</sup>

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<sup>b</sup> ScotEID, Huntly, UK

**Introduction:** Compulsory electronic tagging of cattle using ultra-high frequency (UHF) technology will be coming into force in Scotland in the near future. Ultra-high frequency technology offers many benefits over “traditional” low frequency including speed, accuracy, cost, read distance and the ability for multiple simultaneous readings. The aims of this study were to (i) determine the best combination/position of UHF antennae within a commercially relevant transportation vehicle and (ii) evaluate accuracy of UHF transponders during transportation from farm to abattoir.

**Materials and Methods:** In total six UHF antennae (HD7-868 UHF RFID antenna, MobileMark, Staffs, UK) were integrated into a newly fabricated livestock box. The livestock box consisted of three compartments, within the two front compartments UHF antennae were mounted to the ceiling (1 per compartment; Fig. 1A). Antennae were mounted on the side wall in the rear compartment due to a retractable sheep deck, with three possible orientations (Fig. 1B; mirrored on either wall). Antennae were connected to a CAEN RFID Proton Reader (CAEN, Viareggio, Italy) and a Raspberry Pi 3, data (tag reads) were uploaded to the ScotEID database.

A UHF transponder was attached to the ear of each animal. Antenna logged transponders ~ once per minute. This study was split into two

phases (i) Animal Loadings (n = 40) and (ii) Transportation to Abattoir (n = 49). Animal Loadings – Combinations of antenna were assessed during 10 loadings and unloadings of cattle at each orientation (n = 40 in total), location was also assessed using GPS. Note: initial issues with the technology meant accurate data was not recorded for all loadings (inaccurate/inconsistent date/time). Each loading consisted of a batch of 16 animals loaded onto the livestock box (5 or 6 animals in each compartment), cattle were held for ~ 5 minutes before releasing. Loadings were spread over a 3-month period to minimise stress. Transportation to Abattoir – Animals were monitored during transportation from SRUC’s Easter Howgate Farm to 2 abattoirs (3 transportations in total). Note: GPS was not installed during this stage. Read rates and accuracy of GPS was assessed where accurate data was recorded (e.g. accurate date/times), retention of tags were also assessed.

**Results:** Animal Loadings. Twenty-six loadings had accurate data. All tags were continuously read whilst animals were loaded giving a read rate of 100%. Accurate locations were recorded (GPS). All tags were retained and fully functional after 3-months. Transportation to Abattoir. Two loadings had accurate data, both had a read rate of 100% and were read continuously throughout the journey.

**Conclusions:** Exceptionally high read rates (100%) were noted regardless of antenna orientation, GPS also accurately recorded location of the livestock box.

**Acknowledgement:** This work was funded by the Scottish Government’s Rural and Environmental Science and Analytical Services Division.



Fig. 1. (A; LEFT) ceiling mounted (1) and (2) in front compartments and wall antennae (3) and (4). (B; RIGHT) wall antennae (1) fixed and (2) adjustable into 3 positions (a, b or c).

## 17. Host genomics affects the abundances of several uncultured *Methanobrevibacter* species genomically associated with methane emissions in beef cattle

Marina Martinez-Alvaro<sup>a</sup>, Marc Auffret<sup>b</sup>, Carol-Anne Duthie<sup>a</sup>, Richard Dewhurst<sup>a</sup>, Larissa Zetouni<sup>c</sup>, Matthew Cleveland<sup>d</sup>, Mick Watson<sup>e</sup>, Rainer Roehe<sup>a</sup>

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**Introduction:** The variability amongst hosts on the abundance of the most ubiquitous ruminal methanogenic archaea genus *Methanobrevibacter* is expected to be at least partially responsible for CH<sub>4</sub> production in cattle. However, several studies have failed to show this relationship suggesting more complex mechanisms. A host genetic control of the ruminal abundance of *Methanobrevibacter* has been recently reported [1], but further knowledge at a lower taxonomic level is required as different species may exhibit different functions [2]. The aim of this study was to estimate the host genomic effect on the ruminal abundances of uncultured *Methanobrevibacter* sp. and their genomic correlations with CH<sub>4</sub> in beef cattle.

**Materials and methods:** This study used phenotypic, genomic and metagenomic data from 359 steers from different breeds (Aberdeen Angus, Limousin, Charolais and Luings) offered *ad libitum* forage or concentrate-based diets. Methane data and ruminal samples were collected as described in Roehe *et al.* 2016. Ruminal uncultured microbial genomes (RUGs) were *de novo* metagenome-assembled as explained in Stewart *et al.*, 2019. We selected 10 uncultured *Methanobrevibacter* sp. (defined by their RUG numbers) with a high prevalence (present in at least 200 animals), relative abundance (>0.21%) and variability amongst animals (coefficient of variation 80–93%) for genomic analysis. Their heritabilities ( $h^2$ ) and host-genomic correlations with CH<sub>4</sub> were estimated by fitting univariate/bivariate animal genomic models. We tested the significance of  $h^2$  by conducting a likelihood-ratio test and of host-genomic correlations by estimating the probability of their distribution being different from 0 ( $P_0$ ).

**Results:** The host genome influenced the abundance of the 10 *Methanobrevibacter* RUGs ( $h^2$ - $P$ -values  $\leq 1.98 \times 10^{-3}$ ). Estimates ranged from 0.12–0.48 with RUG12025 and RUG11042 being the most heritable ( $h^2$ [HPD<sub>95%</sub>] 0.47[0.08,0.85] and 0.48[0.10,0.90]). These  $h^2$  are above previous estimates reported for *Methanobrevibacter* genus

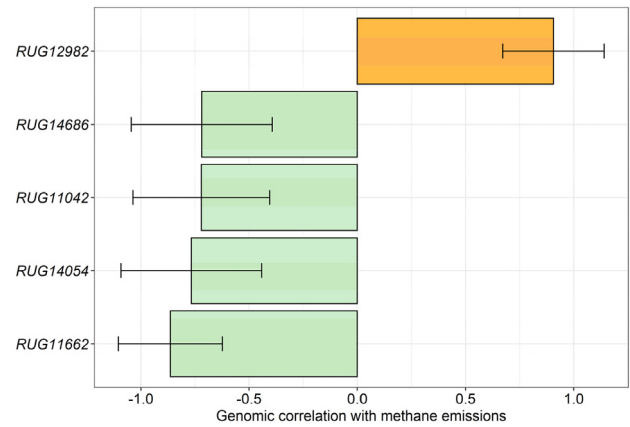


Fig. 1. Host-genomic correlations (medians and s.e.) amongst CH<sub>4</sub> and the abundances of different uncultured *Methanobrevibacter* sp. ( $P_0 > 95\%$ ).

( $h^2 \sim 0.2$ , Zhang *et al.*, 2020). Methane showed significant ( $P_0 > 95\%$ ) and strong ( $> |0.72|$ ) host-genomic correlations with 5 *Methanobrevibacter* RUGs (Fig. 1). Different species displayed positive or negative values highlighting their functional versatility. These results indicate that *Methanobrevibacter* identification at a species level is fundamental to understand the complexity of their host-genomic effect on CH<sub>4</sub> production.

**Conclusion:** Our results show a significant host genomic effect on the abundance of uncultured *Methanobrevibacter* species identified by *de novo* metagenome-assembly, suggesting new opportunities to integrate microbial information into breeding programs. Strong but opposite-directed host-genomic correlations with CH<sub>4</sub> highlight the necessity of classifying microbes at low taxonomic levels (at least species) for breeding purposes as complex relationships amongst the trait (i.e., CH<sub>4</sub>) and different microbial species within a genus may exist.

**Acknowledgement:** BBSRC, Genus plc and the Scottish Government.

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## 18. Effect of contrasting grazing management methods on the behaviour and activity levels of dairy × beef steers at pasture

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**Application:** This study reveals that grazing management alters the behaviour and activity levels of grazing animals, highlighting the need to consider impacts relating to animal behaviour and welfare when implementing management intensive, rotational grazing systems.

**Introduction:** Grazing livestock have traditionally been managed using set-stocking (SS) or continuous stocking, where animals have access to relatively large grazing areas for prolonged period(s) of time at low to medium stocking rates. Cell grazing (CG) is a planned, rotational, time-controlled method which uses high stocking densities and frequent movements of animals to mimic the natural grazing behaviours of large wild herds of grazing ungulates (Voisin, 1959; Savory, 1969). Accordingly, animals are confined to relatively small grazing areas day-to-day, which raises concerns regarding the impact of such grazing systems on animal behaviour and welfare. This study assessed the impact of grazing management on the behaviour and activity levels of dairy × beef steers at pasture. [1,2].

**Materials and methods:** Dairy × beef steers were assigned to either SS or CG in April 2019. Plots were setup in triplicate with a fixed size of 1.75 ha for SS and 1.0 ha for CG, the latter of which was sub-divided into 42 evenly-sized cells. SS plots were continually grazed for the whole grazing season. CG cells were rotationally grazed with a daily area allocation determined by average pasture cover and ad-lib feed demand. Data were collected using leg-mounted IceQube pedometers and a mobile trailer mounted CowAlert system (IceRobotics Ltd, Edinburgh, UK) from 9 steers per treatment (3 per replicate plot) between 29 May and 21 October. Days where only partial data (<24hrs) had been captured and where animals had been released from their grazing plots (e.g. for weighing) were excluded. Data were analysed in GenStat 21 (VSNi, Hemel Hempstead, UK) using a linear mixed model with grazing management as fixed and animal, experimental block and day as random model components.

**Results:** Daily step count for CG was lower than SS (Table 1). Grazing management did not affect daily total standing time ( $P > 0.05$ ) but did affect number of lying bouts, with SS lower than CG (Table 1). Diurnal step count pattern is presented in Fig. 1. Both groups had a similar pattern in the morning, albeit slightly delayed for CG animals due to waiting to be moved, however there was a marked difference in step count in the afternoon.

Table 1

Average daily activity of steers managed by either cell grazing (CG) or set-stocking (SS).

	CG (n = 9)	SS (n = 9)	s.e.d	p
Total daily steps	927	1644	148	<0.001
Standing (mins)	698	687	21.5	0.614
No. lying bouts	12.0	10.1	0.812	0.041

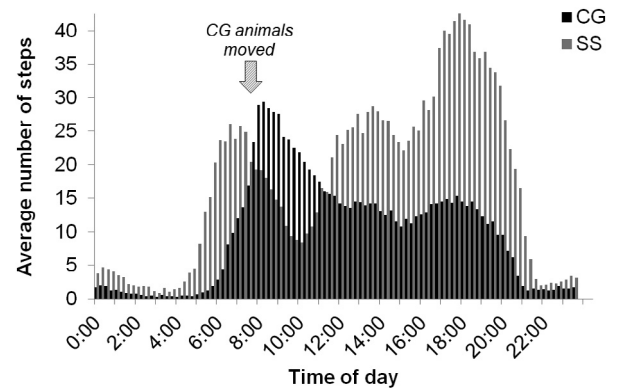


Fig. 1. Influence of grazing management on average diurnal step count pattern

**Conclusion:** These results show that grazing management alters certain behaviours and activity. Further investigation is required to determine the extent to which this altered behaviour and activity impacts overall animal welfare and health.

**Acknowledgement:** Agri-tech Cornwall is a 3 year £10 m initiative, part-funded by the European Regional Development Fund, to increase Research Development and Innovation in the Agri-tech sector across Cornwall and Isles of Scilly ([www.agritechcornwall.co.uk](http://www.agritechcornwall.co.uk)).

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### 19. Comparison between qualitative behavioural assessment (QBA) on winter housed beef cattle scored by video and live

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**Application:** The validity of V-QBA reinforces the opportunity for assessments to be conducted at greater temporal and spatial scales than live assessments otherwise could be.

**Introduction:** Animal welfare is an essential part of livestock production and sustainability. Not only is it of moral importance, but it is also intertwined with animal health, productivity, economics, and environmental impacts. Qualitative behavioural assessment (QBA) [1] is a widely used method for assessing the welfare and emotional state of animals including livestock. Numerous studies have utilised video as a medium by which to perform QBA (V-QBA). The benefit of V-QBA is primarily the ability to monitor sites at scale, distance, and convenience. The Covid-19 pandemic has highlighted the value of being able to perform tasks remotely, without the need for travel and social interaction. However, the validity of V-QBA has not been tested. The objective of this study was to compare V-QBA to live QBA.

**Materials and methods:** This work fell within part of an on-going welfare program comparing housed and grazing beef cattle by the use of a variety of indicators. Qualitative Behaviour Assessments (QBA) are taken weekly throughout the year – a systematic scoring of twenty behaviours/descriptors for the assessment of welfare and emotional state. Forty live QBAs were conducted by two assessors, previously trained by experienced researchers, across two herds of thirty housed beef cattle. During each 10-minute assessment period, cattle were recorded by four cameras. At least one month later, the same two different assessors

watched the footage of cattle for the same session they had conducted live and performed a new assessment by the same criteria. Results from the live QBA and V-QBA were compared with paired T-tests (differences between pairs were normally distributed) for each of the 20 descriptors and by determining correlation coefficients to assess if there were any significant differences or correlations between results derived from these two approaches.

**Results:** Of the 20 descriptors, three ('Distressed', 'Fearful', 'Frustrated'), were discounted from statistical analysis as all or an overwhelming majority of values were zero. For the remaining 17 descriptors, a statistically significant difference between QBA and V-QBA was only found for one descriptor, 'Inquisitive' ( $T = 3.16$ ,  $p = 0.003$ ). For 12 descriptors (including 'Inquisitive'), significant medium to high correlation coefficients were found between QBA and V-QBA, with  $r$  ranging from 0.57 (for 'Content') to 0.91 (for 'Positively occupied').

**Conclusion:** Results provide evidence that assessments in-person or live do not generate significantly differing results. Therefore, the selection of which approach should be used will depend on the circumstances, resources and/or infrastructure of the farm. Furthermore, the study broadly supports that V-QBA results are reflective of live QBA. Whilst additional work is necessary, across a wider variety of scenarios, to conclusively determine this, the substantial practical benefits of V-QBA are considered to outweigh that uncertainty.

**Acknowledgements:** This work was funded by BBSRC grant number: BBS/E/C/00010320.

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## 20. Site use preference of temperate pastures by beef cattle in spring-summer grazing

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**Application:** Cattle show a clear preference for utilising particular areas of the grazing areas. This may impact soil compaction, sward condition and soil greenhouse gas emissions in those hotspots. Knowing these patterns can be crucial to develop management strategies to increase the sustainability of grazing systems.

**Introduction:** Understanding the behaviour of grazing animals at pasture, i.e., how grazing animals distribute themselves and move across pasture and what activities they perform in each area, is crucial to develop management strategies that will increase the potential productivity of grazing systems and also decrease the negative impact on the environment (nutrient losses to water and gaseous emissions) [1]. Therefore, it is necessary to record and study their movements in space and time to reveal the motivation for their site use preferences. This would enable the design and management of more sustainable grazing systems. The aim of this study was to map out the distribution of grazing beef cattle during the daylight hours in order to identify variables associated with their site use preference.

**Materials and methods:** The location of cattle in two fields of the North Wyke Farm Platform (field #5: permanent pasture, 15/05/2018–22/05/2018; and #9: white clover/perennial ryegrass, 21/06/2018–27/06/2018) was recorded every 1 s by attaching GPS tags during spring-summer 2018. Then, the extent to which cattle preferentially use particular

areas of each field was determined. Monitoring in field #9 involved 24 cattle (out of 30 animals grazing in the field in that period) and in field #5 (bottom half of the field) for 18 cattle (out of 30 animals). Point density was calculated, using R software, for each cell in a 100x100 grid overlaid over each field to calculate the percentage of time that the cattle spent in each part of the field during daylight hours.

**Results:** The location of cattle tracked showed uneven use of the respective field areas (Fig. 1). For field #5, 50% of cattle time was spent in just 11% of the field area whereas in field #9, 50% of time was spent in 14% of the field area. A clear tendency for the cattle to congregate preferentially along one fence in each field was observed. In field #5, cattle spent 27, 52 and 61% of their time within 5, 10 and 25 m from the top fence, whilst for field #9 these percentages were 20, 33 and 37%, respectively. In both fields, this was at the highest elevation and in proximity to water troughs. Likewise, cattle spent 49 and 52% of their daylight time in the highest quarter of the field for fields #9 and #5, respectively.

**Conclusion:** Cattle preferentially spend time in small areas of the field and close to specific fences (greatest height) and in the highest sectors of the field.

**Acknowledgments:** Work funded by BBSRC, through Soil to Nutrition (BBS/E/C/00010320), and by Lawes & Gilbert studentship.

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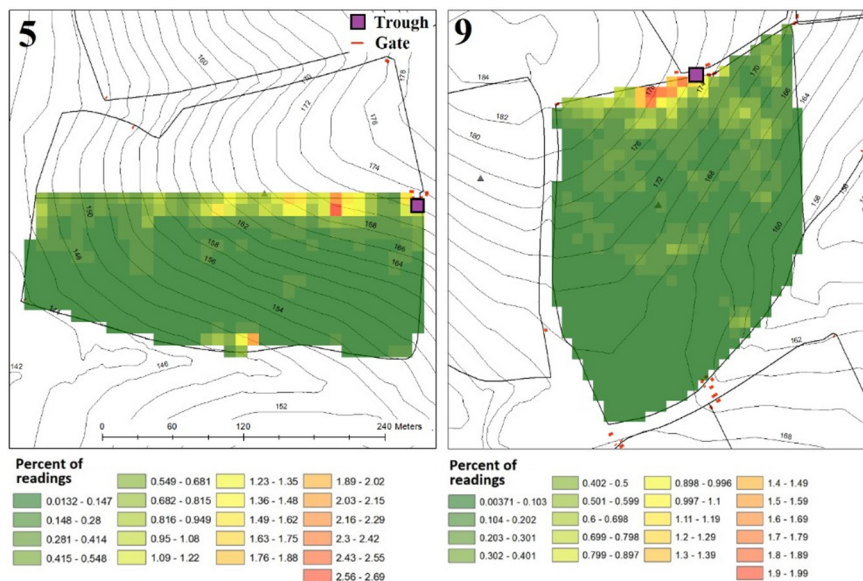


Fig. 1. Percentage of cattle GPS readings recorded in each 100x100 cell in two fields.

## 21. On-farm and abattoir predictors of intra-muscular fat in commercial UK slaughter cattle

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**Application:** On-farm ultrasound measurements do not accurately predict intramuscular fat (IMF) in commercial slaughter cattle. Marbling scores awarded in the abattoir could rank carcass IMF with moderate accuracy.

**Introduction:** Within high-end beef supply chains, it would be valuable for processors to identify animals that will yield cuts with higher IMF, associated with improved eating quality, enabling a more value-based marketing system. This study tested previously-developed IMF-predictors (Breedplan on-farm ultrasound scanning algorithms (US-IMF; <http://breedplan.une.edu.au>) and USDA marbling scores of cut carcasses (USDA-MS; <https://www.usda.gov>) for their ability to determine IMF levels (measured by FOSS Meat Analyser; Anderson, 2007) across a range of typical UK slaughter cattle.

**Materials and methods:** Finishing cattle (n = 159, range of breeds/crossbreeds) were recorded on two commercial farms supplying a commercial abattoir. Finished indoors, with diets varying across farms/season, cattle were slaughtered in batches from September 2019 to April 2020 (age range 506-908d, carcass weight 275-397 kg). On-farm US-IMF measurements were taken pre-slaughter by a Breedplan-approved technician (unavailable for 28 cattle in final 2 batches). Three ultrasound images were taken parallel to the spine, from last rib to 2nd lumbar vertebrae, and IMF% determined using a predefined algorithm. Subcutaneous fat depth (UFD) was measured at 12/13th rib. Post-slaughter, carcasses were graded for fat class (MLCF - converted to numerical 15-

point scale, Kempster et al., 1986) and scored by abattoir staff for marbling (1-6; USDA-MS) on the left side at the cut between 5th/6th rib. A slice (~100 g) was removed from the left Longissimus muscle at 5th/6th rib and frozen, then later thawed and assessed for IMF% using a FOSS<sup>TM</sup> FoodScan Meat Analyser (FOSS-IMF). Correlations and regressions were performed in Genstat (VSN International, 2015) to relate US-IMF and USDA-MS to laboratory-measured IMF (FOSS-IMF).

**Results:** Correlations with FOSS-IMF were 0.33 for US-IMF and 0.55 for USDA-MS (regressions, Fig. 1). These were higher than correlations of FOSS-IMF with total carcass fat predictors (0.30 for UFD and 0.44 for MLCF), whilst predictors of IMF and carcass fat were only moderately correlated (US-IMF vs UFD  $r = 0.37$ ; USDA-MS vs MLCF  $r = 0.44$ ). Samples from carcasses scored USDA\_MS = 1 had significantly ( $P < 0.05$ ) lower FOSS-IMF than those scored USDA\_MS = 2, which in turn had lower FOSS-IMF than those scored  $\geq 3$ . There was a non-significant trend for increased FOSS-IMF from USDA\_MS of 3 to 5 ( $P > 0.05$ ). US-IMF had a correlation of 0.39 with USDA-MS. [1,2,3].

**Conclusion:** Across commercial UK slaughter cattle, from different sources, diets and breeds, on-farm US-IMF predictions are unlikely to accurately predict IMF%. Scope exists for USDA marbling scores to identify carcasses with different IMF levels, to help sort beef cuts for potential eating quality.

**Acknowledgments:** Funded by Scottish Government's Knowledge Transfer and Innovation Fund. Thanks to PR Duff and commercial farmers for data collection.

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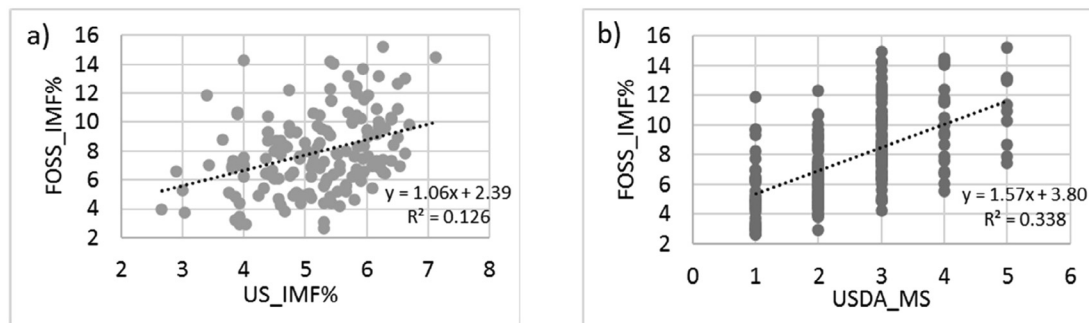


Fig. 1. Regressions of FOSS-IMF on a) US-IMF and b) USDA-MS.

## 22. Replacement of ad-libitum concentrates with fermented whole crop wheat (WCW) and red clover (RC) silages in intensive finishing diets for British blue cross Holstein steers

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**Application:** Performance and profitability is similar for finishing steers fed ad-libitum concentrates or diets containing 750 g/kg DM fermented WCW. At current prices inclusion of RC silage is not cost effective.

**Introduction:** Beef cattle are often intensively finished on *ad-libitum* concentrates based on cereals and vegetable protein supplements, with feed being the main variable cost [1]. However, the UK is only 30 % self-sufficient in vegetable protein sources, with most being imported from overseas. Fermented WCW and RC silages are high energy and protein forages that can be widely grown in the UK. The objective was to investigate the effect of replacing *ad-libitum* concentrates with fermented WCW and RC silages in intensive finishing systems for British blue cross Holstein steers.

**Materials and methods:** Whole crop wheat (*Var. Graham Redigro Pro 2018*) was harvested in July and RC silage (*Var. Atlantis*) in August 2020 at 500 and 328 g/kg dry matter (DM) respectively, using a forage harvester. The WCW was treated with Wholecrop Gold and the RC was treated with Axcool (Biotol Ltd) at 2.0 litres/tonne and ensiled in Ag-bags. Sixty British Blue cross Holstein steers were allocated to four treatments consisting of *ad-libitum* concentrates (C), or one of three diets containing 750 g/kg DM forage as either 100:0 (WCW:RC), 670:330 (RC33) or 300:670 (RC67). Four concentrates were formulated to supply 140 g/kg DM CP in the overall diet. Steers were housed in groups of five in straw bedded pens, with three pens per treatment. All diets were fed *ad-libitum* as total mixed rations. Dry matter intake (DMI) was monitored by recording the amount offered and refused twice weekly. Live weight (LW) was recorded initially, at 30-day intervals until slaughter at 600–650 kg LW, fat class 3. Diet costs and margins were calculated and data analysed by ANOVA as a randomised block design using Genstat 18 (Table 1).

Table 1

Effect of dietary fermented WCW and RC silage proportion on animal performance and diets costs.

	C	WCW	RC33	RC67	SED	P
Initial LW (kg)	458	459	462	460	4.27	0.834
Final LW (kg)	652	652	648	650	5.29	0.862
Days to slaughter	134	139	139	153	9.5	0.290
Daily LW gain	1.45	1.39	1.35	1.23	0.066	0.075
Dry matter intake (kg/d)	9.38	12.89 <sup>a</sup>	14.68 <sup>b</sup>	14.09 <sup>ab</sup>	0.642	<0.001
Feed conversion ratio (kg/kg)	6.45	9.31	10.93 <sup>a</sup>	11.45 <sup>a</sup>	0.503	<0.001
Killing out % age	54.4	52.2 <sup>a</sup>	53.3 <sup>a</sup>	52.5 <sup>a</sup>	0.004	0.008
Diet costs £/tonne DM	233	151	152	156	—	—
Margin (£/kg LW gain)	1.25 <sup>a</sup>	1.00 <sup>ab</sup>	0.96 <sup>bc</sup>	0.71 <sup>b</sup>	0.117	0.021

**Results:** Both the WCW and RC silages were well fermented and contained 84 vs 182 g/kg DM CP and 290 vs 19 g/kg DM starch, respectively. The DMI and feed conversion ratio (FCR) of steers offered diet C were lower than that of those offered diet WCW, which were lower than that of those offered diets RC33 and RC67. In addition, steers offered diet C had a higher killing out % age. No other performance parameters were significantly different. Numerically, days to slaughter increased and daily LW gain decreased on the forage-based treatments and with increasing RC proportion.

**Conclusions:** Replacement of concentrates with 750 g/kg DM fermented WCW or RC silage had limited effects on animal performance, but increased DMI and FCR, particularly with RC inclusion. It also reduced diets costs, such that margins were similar for diets C and WCW. At current prices inclusion of RC silage was not cost effective.

**Acknowledgments:** The authors acknowledge funding from AHDB Beef and Lamb.

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### 23. Citizen opinions regarding beef production and beef production systems

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**Application:** Beef production stakeholders should consider that citizens expressed higher approval for regenerative grazing beef production systems. To improve the current citizen's negative attitudes towards beef production, stakeholders need to do more to educate citizen's of the positive aspects of beef production systems: welfare, environmental and health.

**Introduction:** Beef production systems differ in land and animal management practices and housing, which overall entails different productivity, animal welfare and their environmental impacts [1]. Their production efficiency also differs and, differences in efficiency of resources used also influence productivity and environmental impact (Godfray & Garnett, 2014). The aim of the study was to explore citizens' opinions regarding beef production systems.

**Materials and methods:** Chilean participants (n = 1084) received brief information regarding aspects (space per animal, land use, productivity, soil erosion and pollution and biodiversity) of different beef production systems (confinement, extensive grazing, or regenerative grazing). They were asked their level of approval using Likert scale (1 to 5): (Q1a) Do you approve this beef production system? (Q1b) Why? (Q2) Would you

approve that your beef comes from this beef production system? (Q3) Do you approve that the system described should be the future beef production system? Then they were asked their level of agreement (0 totally disagree to 4 totally agree or "don't know"): (Q4) Need to reduce beef consumption; (Q5) Greenhouse gases are emitted in beef production; (Q6) Beef production is negative for the environment; (Q7) Beef is bad for human health. They were asked (Q8) Where do you get information regarding beef production systems? (Q9) Do you trust this source? Q1a, Q2 and Q3 were analysed using Kruskal Wallis and also descriptive data are presented.

**Results:** Regenerative grazing system had the highest approval levels and confinement the lowest (P < 0.05, Fig. 1a). The main reasons for Q1a were related to animal welfare (16%), lack of knowledge (14%) and the environment (11%). > 60% of the participants agreed on Q4 to Q6, but 42% disagreed on Q7 (Fig. 1b). The information sources most mentioned were documentaries (18%), social media (17%) and people you know (15%), but only 52%, 45% and 48% considered them reliable. Books (5%), scientific reports (11%) and universities (8%) were considered the most reliable (68%, 63% and 63%, respectively).

**Conclusion:** Characteristics of the regenerative grazing system were the most approved by citizens, whose main concerns were related to animal welfare. Participants had negative attitudes towards beef production, but disagreed that beef is bad for human health.

**Acknowledgement:** The authors acknowledge funding from CONICYT, Chile, (REDI 170086 and FONDECYT 1160697).

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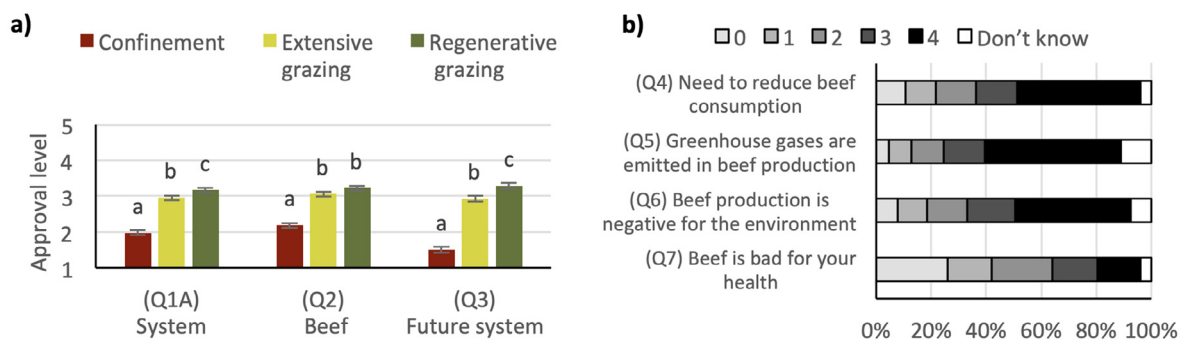


Fig. 1. Citizens a) approval level of different beef production systems, beef produced and as a future systems and b) agreement of different beef production aspects.



## 24. Evaluation of international models for predicting total dry matter intake in growing cattle

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**Application:** A comparison of the predictions given by empirical equations considering both animal and diet-related factors may provide key insights on drivers modulating total dry matter intake (TDMI) responses in growing cattle

**Introduction:** Dry matter intake is the most important variable influencing ruminant's production and therefore accurate predictions of this is a pre-requisite of effective ration formulation. However, measuring intake, especially on-farm conditions is not always an easy task and alternatives have been proposed. The aim of this study was to evaluate six international TDMI models of varying complexity using group-based observations of growing bulls collected from long-term studies conducted at the Agri-Food and Biosciences Institute (AFBI).

**Materials and methods:** Group-basis (n = 85 pens) data (dry matter intakes, live weights, feed composition and silage fermentation quality) were collected from five long-term studies involving 343 Holstein bulls fed seven grass silage-based diets aiming in all cases to study their production responses during growing and finishing phases (7.4 to 15.9 months of age). Perennial ryegrass silage (n = 19) was the predominant forage source in all experiments (mean forage-to-concentrate DM ratio of 31:69). Six international TDMI models of varying complexity were evaluated: 1) Finnish models [1], where three empirical equations were developed based on an additive approach considering metabolic live weight, neutral detergent fibre concentration in the diet, and silage quality factors; 2) The Australian model (CSIRO, 2007), which is a potential intake model built from tabulated values to obtain standard reference weights (SRW) for selected breeds; 3) The French model (INRA,

2018), where intake capacity is calculated from allometric coefficients; and 4) The American model (NASEM, 2016), which estimates TDMI required (DMIR) based on net energy (NE) requirements for maintenance and growth. Model evaluation was conducted using mixed model regression (SAS version 9.4; PROC MIXED procedure). Observed values were adjusted for random study effect. Mean biases were evaluated by the deviation of regression intercepts from zero while the deviation of the slopes of the regression equations from unity was used to determine the presence of linear biases. Model accuracy was assessed by the calculation of the root mean square prediction error (RMSPE).

**Results:** Among the six evaluated intake models, the simplest empirical equation proposed by Huuskonen et al. (2013), displayed the best accuracy (lower RMSPE = 1.75 kg/d; Table 1) for predicting TDMI responses. The American model (NASEM, 2016) predicted with reasonable accuracy TDMI at the specified level of concentrate supplementation.

**Conclusion:** In this evaluation, the simpler non-linear model by Huuskonen et al. (2013;  $TDMI (kg/d) = 0.191 \times LW^{0.627}$ ), showed the best accuracy for predicting TDMI intake in growing Holstein bulls, thus confirming that LW (live weight) is a major driving factor of this trait.

**Acknowledgements:** This work was funded by the Agriculture and Horticulture Development Board (AHDB) and The UK's Livestock Innovation Centre (CIEL) as part of the FiBNUT project.

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Table 1

Evaluation of source of bias and prediction error of different intake prediction models with mixed model regression using group-basis data (n = 85 pens).

Model Ref.	Mean bias	P-value	Slope bias	P-value	RMSPE <sup>1</sup>	CCC <sup>2</sup>	Error distribution		
							Mean	Slope	Random
Huuskonen-1 (2013)	1.44	<0.001	1.00	<0.001	1.753	0.533	0.754	0.000	0.246
Huuskonen-2 (2013)	3.02	<0.001	0.88	<0.001	2.535	0.285	0.849	0.006	0.144
Huuskonen-3 (2013)	2.94	<0.001	1.04	<0.001	3.403	0.156	0.943	0.000	0.056
CSIRO (2007)	9.67	<0.001	-0.08	0.090	4.397	0.015	0.768	0.147	0.086
INRA (2018)	1.17	<0.001	1.13	<0.001	2.342	0.424	0.872	0.018	0.109
NASEM (2016)	2.26	<0.001	0.82	<0.001	1.841	0.899	0.625	0.020	0.355

## 25. Profitability of commercial beef farms of superior herd terminal and maternal genetic merit

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**Application:** Commercial Irish beef farms of higher herd-average terminal and maternal genetic merit are, on average, more profitable.

**Introduction:** While several studies have validated the usefulness of breeding goals in the selection of more productive beef cattle at the individual animal level, no such study has been attempted at the herd-level. Using herd-level data enables the costs and income sources not directly attributable to individual animals (e.g. fixed costs) to be included in the analyses. Moreover, previous animal-level validation studies applied a single economic value, common across all farms, to each trait within the selection indexes; however, this is not a true representation of reality. The objective of the current study was to quantify the association between herd-average terminal and maternal genetic merit with profitability, using farm financial data from a large number of commercial Irish beef herds.

**Materials and methods:** Farm physical and financial performance data were available from 2,308 herd-years representing 1,311 unique beef herds across the years 2016 to 2019, inclusive. Herd-average genetic merit for the Irish beef terminal and maternal indexes from the February 2020 national genetic evaluations were generated for the cattle on each farm. Gross profit was the value of livestock sales less purchases, plus the value of any net inventory change in livestock numbers, minus variable costs; variable costs included veterinary expenses, fertilizer,

concentrate feed, and other miscellaneous costs. Net profit was gross profit minus fixed costs; fixed costs included depreciation, repairs and maintenance, hired labour, utility expenses, and rent. Associations between profit metrics and herd-average genetic merit were determined using a series of linear mixed models. Model fixed effects included year, production system, herd size, stocking rate, concentrate input, and the two-way interactions between production system and herd size, stocking rate, and concentrate input; herd nested within the county of Ireland was included as a repeated effect. Genetic merit variables included in the model were either the terminal index of progeny, or both dam maternal index and sire (of progeny) terminal index included simultaneously. **Results:** Higher herd-average terminal genetic merit was associated with greater herd profitability per hectare, and per livestock unit (Table 1). Each €1 increase in terminal index was associated with a €1.41 (SE = €0.23) increase in gross profit per livestock unit, however, this association was less than the expected coefficient of 2 ( $P < 0.05$ ). Herd-average dam maternal and sire terminal genetic merit were positively associated with gross profit per hectare, and per livestock unit (Table 1).

**Conclusion:** Results from the present study at the herd level concur with previous validation studies at the animal level, thereby providing more confidence to stakeholders as to the expected improvement in profitability associated with superior herd beef genetic merit.

**Acknowledgements:** Funding from the Department of Agriculture, Food and the Marine, Ireland Research Stimulus Fund Ref: 17/S/235 (GreenBreed) is gratefully acknowledged

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Table 1

Regression coefficients (b; standard errors in parentheses) and the significance of the regression coefficients from zero for herd financial performance on terminal index value, or on both dam maternal index and sire terminal index value.

Financial variable	Terminal index	Dam maternal index	Sire terminal index		b (SE)	P value
	b (SE)	P value	b (SE)	P value		
Gross profit per hectare, €/ha	2.33 (0.39)	< 0.001	1.40 (0.48)	0.004	1.30 (0.29)	< 0.001
Gross profit per livestock unit, €/LU	1.41 (0.23)	< 0.001	0.76 (0.29)	0.008	0.76 (0.18)	< 0.001
Net profit per hectare, €/ha	2.23 (0.48)	< 0.001	0.75 (0.61)	0.216	1.33 (0.37)	< 0.001
Net profit per livestock unit, €/LU	1.37 (0.30)	< 0.001	0.50 (0.39)	0.194	0.70 (0.23)	0.003

## 26. Genome wide association study for component traits of compensatory growth in Irish beef cattle

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**Application:** The exploitation and manipulation of compensatory growth (CG) is common practice in beef production systems worldwide, particularly in pastoral systems such as in Ireland where there is an opportunity to reduce overall feed costs by rebalancing feed demand. Single nucleotide polymorphisms (SNPs) or genetic variants associated with enhanced ability to display CG in beef cattle could be integrated into breeding programme to breed for the genomics selection of improved CG in beef cattle.

**Introduction:** Compensatory growth is a physiological process whereby an organism accelerates its growth after a period of restricted development, usually due to reduced feed intake (Hornick et al., 2000). The objective of this study was to detect genetic variants affecting CG in Irish cattle. Genome wide association studies (GWAS) have proved to be a powerful tool to achieving this by enabling the detection genetic variants affecting economically important traits (Goddard and Hayes, 2009).

**Material and methods:** The average daily gain (ADG) of Holstein Frisian bulls during a restricted growth period and a re-alimentation CG growth period was recorded over 2 years on 6 farms across Ireland. All cattle were genotyped on a 50k SNP chip panel. 43898 variants and 955 cattle remained following filtering and quality control. Principal component analysis was conducted to examine population admixture. Phenotypic values were adjusted for animal age at restricted feeding, age at CG phase, farm, breed composition and year of testing. GWAS analyses were performed using mlma (mixed linear model association) and mlma-loco (leaving one chromosome out) options in the GCTA package. The genomic relationship matrix was constructed with the markers of the 50k variant panel. This resulted in 2 sets of GWAS results for each of the three traits of interest; ADG in the feed restriction phase (ADG res), ADG in the CG phase (ADG CG) and ADG res – ADG CG.

**Results:** A total of 200, 218 and 221 SNPs were identified ( $P < 0.005$ ), in the mlma analysis for ADG res, ADG CG and ADG res - ADG CG respectively. In the mlma-loco analysis 242, 240 and 255 SNPs were identified. For the three traits respectively. Genes located within linkage

disequilibrium of the lead SNP were considered as candidate genes associated with the trait. Gene ontology annotation of these genes in the ADG CG analysis included processes such as nutrient transport, ATP binding and nucleotide binding.

**Conclusion:** Following validation, statistically significant SNPs may be included into the national genomic selection programme as DNA based biomarkers for the selection of cattle with an improved CG potential and thus reduce on farm feed cost whilst simultaneously selecting for more energy efficient and environmentally sustainable animals (Fitzsimons et al., 2013).

**Material and methods:** The average daily gain (ADG) of Holstein Frisian bulls during a restricted growth period and a re-alimentation CG growth period was recorded over 2 years on 6 farms across Ireland. All cattle were genotyped on a 50k SNP chip panel. 43898 variants and 955 cattle remained following filtering and quality control. Principal component analysis was conducted to examine population admixture. Phenotypic values were adjusted for animal age at restricted feeding, age at CG phase, farm, breed composition and year of testing. GWAS analyses were performed using mlma (mixed linear model association) and mlma-loco (leaving one chromosome out) options in the GCTA package. The genomic relationship matrix was constructed with the markers of the 50k variant panel. This resulted in 2 sets of GWAS results for each of the three traits of interest; ADG in the feed restriction phase (ADG res), ADG in the CG phase (ADG CG) and ADG res – ADG CG.

**Results:** A total of 200, 218 and 221 SNPs were identified ( $P < 0.005$ ), in the mlma analysis for ADG res, ADG CG and ADG res - ADG CG respectively. In the mlma-loco analysis 242, 240 and 255 SNPs were identified. For the three traits respectively. Genes located within linkage disequilibrium of the lead SNP were considered as candidate genes associated with the trait. Gene ontology annotation of these genes in the ADG CG analysis included processes such as nutrient transport, ATP binding and nucleotide binding.

**Conclusion:** Following validation, statistically significant SNPs may be included into the national genomic selection programme as DNA based biomarkers for the selection of cattle with an improved CG potential and thus reduce on farm feed cost whilst simultaneously selecting for more energy efficient and environmentally sustainable animals (Fitzsimons et al., 2013).

**Acknowledgements:** Authors acknowledge funding from Science Foundation Ireland (13/CDA/2182).

## 27. Genome wide association study for compensatory growth in Irish beef cattle

Yvonne Mullins<sup>a,b</sup>, Alan Kelly<sup>b</sup>, David Kenny<sup>a</sup>, Sinead Waters<sup>a</sup>

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<sup>b</sup>University College Dublin, Belfield, Dublin 4, Ireland

**Application:** The exploitation and manipulation of compensatory growth (CG) is common practice in beef production systems worldwide, considering the economic importance of CG and that Ireland is a pasture based country; CG at pasture is an extremely important trait to include in a breeding index. Identifying and determining the relationships between genetic markers and an animals CG ability would allow for an increased accuracy in predicting the genetic value of future cattle which are genotyped, without the need for costly and delayed phenotyping measurements (Hayes and Goddard, 2001, Hayes et al., 2009).

**Introduction:** Compensatory growth (CG) is one mechanism by which beef production can become more efficient. Compensatory growth is a naturally occurring physiological process whereby an organism accelerates its growth and efficiency upon re-alimentation after a period of restricted feed intake [1]. However the genetic architecture of CG remains unknown and no genome wide association study (GWAS) has been carried out for the CG traits and there is currently no heritability estimation available. This studies objective was to detect genetic variants affecting CG in Irish cattle.

**Material and methods:** The residual CG response of Holstein Frisian bulls was recorded over 2 years across 6 Irish farms. All bulls were genotyped on a 50k SNP chip. Quality control measures were carries out and 43898 variants and 955 cattle remained following filtering. Principal component analysis was conducted to inspect the population admixture.

Phenotypic values were adjusted for season (spring or autumn born calves), farm, breed composition and year of testing. GWAS analyses was performed using the mlma (mixed linear model association) option in the GCTA package. Genomic heritability was estimated using the REML package. The genomic relationship matrix was constructed based on the markers from the 50k variant panel.

**Results:** A total of 197 significant SNPs were identified ( $P < 0.005$ ) in the mlma analysis for the residual compensatory growth response trait. Genes located within linkage disequilibrium of the lead SNP were considered as candidate genes associated with the trait. Functional annotation of these genes included processes such as appetite regulation, nutrient transport, and ATP binding.

**Conclusion:** This study shows there is an opportunity to breed cattle with an improved CG potential giving rise to improved feed efficiency, increase profitability and reduced environmental footprints. Following validation the significant SNPs identified in this study may be included in genomic selection programmes as DNA based biomarkers for the selection of cattle with an improved CG potential.

**Acknowledgements:** Authors acknowledge funding from Science Foundation Ireland (13/CDA/2182).

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## 28. The effect of phenotypically ranking beef cattle for residual methane output on daily methane emissions, intensity and animal productivity

Paul E. Smith<sup>a,b</sup>, Sinéad M. Waters<sup>a</sup>, David A. Kenny<sup>a</sup>, Stuart F. Kirwan<sup>a</sup>, Stephen Conroy<sup>c</sup>, Alan K. Kelly<sup>b</sup>

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**Application:** Beef cattle ranked as having low residual methane output had lower emissions intensity and similar overall productive performance as their high emissions ranking contemporaries. The concept of residual methane output is proposed as an appropriate trait to more equitably identify animals on the basis of low emissions beef production. **Introduction:** The sustainability of ruminant livestock production can be enhanced with the inclusion of methane (CH<sub>4</sub>) output in a breeding index. Residual methane output (RCH<sub>4</sub>) is a promising CH<sub>4</sub> phenotype to aid more balanced selection, as it is strongly correlated with daily CH<sub>4</sub> (g/day) but independent of feed intake and body weight [1]. The objective of this study was to assess the impact of ranking cattle in terms of RCH<sub>4</sub> on CH<sub>4</sub> output and production traits in a population of similarly managed cross bred beef cattle.

**Materials and methods:** Intake, feed efficiency (FE) and CH<sub>4</sub> output was measured on 282 cross bred beef cattle (heifers n = 154; steers n = 128) housed at the Irish Cattle Breeding Federation (ICBF) performance test centre (Kildare, Ireland). Cattle were offered *ad libitum* access to a TMR diet (75% concentrate and 25% hay). Daily dry matter intake (DMI), average daily gain (ADG), feed conversion ratio (FCR) and residual feed intake (RFI) were calculated for all animals over a mean test period duration of 91 days (71–128 days). Enteric CH<sub>4</sub> emissions, using the GreenFeed System (C-Lock Inc) were estimated over a 21 day period during the performance test. RCH<sub>4</sub> was calculated by regressing CH<sub>4</sub> (g/day) on DMI and body weight on test day 30 with contemporary group (CG) included as a fixed effect. Cattle were ranked in terms of RCH<sub>4</sub> as high (RCH<sub>4</sub> > 0.5 SD above the mean), medium (RCH<sub>4</sub> ±

Table 1

Methane phenotypes of cattle ranked as high, medium and low in residual methane output.

	High RCH <sub>4</sub>	Medium RCH <sub>4</sub>	Low RCH <sub>4</sub>
CH <sub>4</sub> (g/day)	264.97 <sup>a</sup>	224.03 <sup>b</sup>	184.39 <sup>c</sup>
CO <sub>2</sub> (g/day)	8745.38 <sup>a</sup>	8292.25 <sup>b</sup>	8070.83 <sup>b</sup>
Residual CH <sub>4</sub> (g/day)	37.95 <sup>a</sup>	-0.11 <sup>b</sup>	-40.34 <sup>c</sup>
CH <sub>4</sub> (g/ kg DMI )	25.19 <sup>a</sup>	21.60 <sup>b</sup>	17.70 <sup>c</sup>
CH <sub>4</sub> (g/ kg ADG)	191.26 <sup>a</sup>	167.09 <sup>b</sup>	144.06 <sup>c</sup>
CH <sub>4</sub> (g/ kg Carcass Weight)	0.81 <sup>a</sup>	0.67 <sup>b</sup>	0.57 <sup>c</sup>

<sup>a, b, c</sup> Means within a row with different superscript differ (P < 0.05)

0.5 SD above and below the mean), and low (RCH<sub>4</sub> > 0.5 SD below the mean). The effect of RCH<sub>4</sub> ranking group (correcting for breed type, gender and CG) on various production traits was calculated in SAS (version 9.4) using Mixed Model ANOVA.

**Results:** No difference was observed for DMI, ADG, feed efficiency or carcass weight (CW) between the high and low RCH<sub>4</sub> groups. Low RCH<sub>4</sub> animals produced 30.4% less CH<sub>4</sub> (g/day) and 29.6% less CH<sub>4</sub> (g/kg CW) relative to high RCH<sub>4</sub> animals (Table 1). Amongst the CH<sub>4</sub> phenotypes investigated, RCH<sub>4</sub> was the strongest predictor of daily CH<sub>4</sub> output (r = 0.86; P < 0.0001).

**Conclusion:** Animal productivity was not impacted by RCH<sub>4</sub> ranking however a reduction in all measures of CH<sub>4</sub> output was observed in the low vs. high RCH<sub>4</sub> animals. Selecting animals for a low RCH<sub>4</sub> phenotype may therefore act to reduce both CH<sub>4</sub> (g/day) and CH<sub>4</sub> (g/kg CW) whilst maintaining animal performance.

**Acknowledgements:** Funding is gratefully acknowledge from the FACCE ERA-GAS project 'RumenPredict' (16/RD/ERAGAS/1RUMENPREDICT-ROI2017) and the EU Horizon 2020 funded MASTER project (818368).

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## Invited Speaker Summary

### 29. Precision sheep management in the uplands

Claire Morgan-Davies

Scotland's Rural College (SRUC), Scotland

**Introduction:** My presentation will focus on the recent initiatives around the implementation of Precision Livestock Farming tools and innovative technologies in upland sheep systems management in the UK and beyond. Based on examples from current and past research projects and collaborations, among them the H2020 project

TechCare (Innovative technologies along the value chain to improve small ruminant welfare management), H2020 Thematic Network Sm@RT (Small Ruminant Technologies), and InnovateUK SmartSheep, the presentation will address the various management challenges faced by upland sheep farming industries, and propose examples of potential solutions based on innovative and/or digital technologies. The presented ideas and initiatives will allow the upland sheep farming sector to rethink how technologies could be embraced in such systems, and what challenges or barriers lay ahead.

### 30. Trends among young and educated dairy sheep farmers in Italy regarding technology application in farm management

Michael Odintsov Vaintrub, Matteo Chincarini, Lydia Lanzoni, Isa Fusaro, Melania Giammarco, Giorgio Vignola

University of Teramo, Teramo, Abruzzo, Italy

**Applications:** This study highlights that more needs to be done to educate young farmers on the benefits of using technology in their farming systems

**Introduction:** Young farmers, new entry farmers (first generation), and educated individuals are frequently considered a key population of early adopters for Precision Livestock Farming (PLF) technologies. In the case of dairy sheep farming, they are viewed as important clients falling into the category frequently described as “Early adopters”. This category is considered a key group in gaining a market foothold for developed and under development PLF technologies.

**Materials and Methods:** In the current study a 25 items questionnaire of yes/no questions and linear scale scoring (1–5) was distributed among Italian extensive dairy sheep farmers (143 sent, 78 received), targeting young farmers via local network and professional associations. Items included both questions for systemic planning, use of software, and attitude towards technology. Descriptive statistics analysis was conducted using Microsoft Excel software.

**Results:** Reported flock sizes were small (20–100 ewes/farm) with no dedicated lamb production. The owned land surface was limited as well with only 15–40 ha per farm. The average farmer’s age was 32 (74% age 20–30) and many had high education levels (65% had a B.A./BSc

degree). The majority of farmers were “new entries” who bought (38%) or permanently leased (37%) their land as a first-generation. Among established farmers, a subdivision of land was more common (21%) than obtaining the management of an entire farm (3.8%). All the farmers applied to the “Young farmer” incentive scheme promoted by the Italian Common Agricultural Policy (CAP), with 64% receiving it within the due time.

Many farmers reported traits linked to systemic planning such as yearly production evaluation (88%) and yearly adaptation to market change (91%). However, only 24.5% reported having written management protocols, while 0% used computers for activity planning. On the other hand, computers were frequently used for marketing (84%) and contact with customers (91%). None of the farmers used dedicated software or PLF technologies, although 47% were exposed to both products. Additionally, half of the farmers (50%) reported mistrust in technological systems they cannot maintain themselves. Only a few (17%) were exposed to direct marketing attempts from commercial companies while 32% were aware of subsidized farm modernization CAP schemes for infrastructure and technology.

**Conclusions:** Observed farms were of small size, even in comparison to the average in the Mediterranean region (140–350 ewes/farm). This trend might be linked with the minimal requirements for CAP “Young farmer” incentives they applied for. While many of the young farmers are familiar with technology, their preferred use of it is for marketing and not for farm management. Long term planning rarely includes technological integration, and the availability of reliable information remains limited.

**Acknowledgments:** This study received no funding.

### 31. Field trials of the use of melatonin implants during pregnancy as a tool to improve lamb performances

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<sup>a</sup> CEVA Animal Health, Glenorie, NSW, Australia

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<sup>c</sup> Instituto Universitario de Investigación en Ciencias Ambientales (IUCA), Zaragoza, Spain

**Application:** Implanting pregnant ewes during the second half of the pregnancy with melatonin implants improves survival and growth rates of the lambs until weaning. This is an alternative to the traditional use of implants for purely reproductive purposes.

**Introduction:** Melatonin is widely used to improve reproductive success of sheep, particularly for out-of-season mating, so that pregnancy rate is increased around 30% [1]. Recently, several attempts to test the ability of exogenous melatonin to enhance lamb performances through the implantation of the ewes during pregnancy have been published. Thus, [2] observed an improvement of colostrum quality when implanting the ewes at the fourth month of pregnancy, and [3] reported a reduced neonatal mortality, an improvement of weaning rates, and twin-lamb survival by increasing tolerance of prolonged parturition in extensively managed sheep flocks, implanting with melatonin between day 70 and 120 of pregnancy. The aim of this work was to test the effect of exogenous melatonin during pregnancy on lamb survival and growth rates, in two field trials in extensive Merino flocks in Australia.

**Materials and Methods:** Two field trials with a similar protocol were carried out in 2019 and 2020 in two different flocks of Merino sheep

in Australia (Table 1). Ewes were diagnosed as pregnant by trans-abdominal scanning 84 days after ram introduction. At that moment, ewes were implanted (M group) or not (C group) with a subcutaneous 18-mg melatonin implant (Regulin, CEVA Animal Health, Glenorie, Australia). Lambs were weighted at birth and at weaning, when 3 months of age. Growth rate was calculated as the difference between both weights divided by the age at weaning. Survival rates from birth to weaning were also calculated. Lamb weights and growth rates were compared by analysis of variance, and survival rate by the X<sup>2</sup> test. In a second analysis, data from both years were pooled.

**Results:** Survival rate was significantly higher ( $P < 0.05$ ) in M lambs in 2020 (Table 1), so that ewes implanted with melatonin produced 7% more lambs than non-implanted ewes. In 2019, M lambs had a higher growth rate than C lambs ( $P = 0.07$ ), and as a consequence, lambs from implanted dams presented 1.3 extra kg at weaning ( $P = 0.01$ ). Considering both years together, lambs born from implanted ewes had higher survival ( $P < 0.05$ ) and growth rates ( $P = 0.07$ ), compared with lambs born from non-implanted animals.

**Conclusion:** Results of this study indicate that when melatonin implants are used during the second half of pregnancy in ewes, survival and growth rates from birth to weaning of Merino lambs can be improved.

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Table 1

Number of ewes and lambs included in the field trials and live weights (LW) and growth and survival rates observed (mean  $\pm$  S.E.). \*  $P < 0.10$ ; \*\*  $P < 0.05$ .

	2019		2020		2019–2020	
	M	C	M	C	M	C
No. ewes	250	175	114	117	364	292
No. lambs	352	254	190	202	542	456
Survival rate	86%	82%	75%**	68%	82%**	76%
Birth LW	5.15 $\pm$ 0.06	5.13 $\pm$ 0.05	4.33 $\pm$ 1.17	4.32 $\pm$ 0.19	4.85 $\pm$ 0.10	4.77 $\pm$ 0.08
Weaning LW	31.2 $\pm$ 0.30**	29.9 $\pm$ 0.40	26.4 $\pm$ 0.22	27.3 $\pm$ 0.34	29.5 $\pm$ 0.28	28.9 $\pm$ 0.4
Growth rate	293 $\pm$ 3*	284 $\pm$ 4	233 $\pm$ 10	238 $\pm$ 8	274 $\pm$ 6*	266 $\pm$ 6



### 32. What sources are used by stakeholders in Ireland and Europe to obtain information to improve flock profitability?

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<sup>e</sup>HAO RIAS, Pella, Greece

<sup>f</sup>SRUC, Criarlarich, Scotland, UK

<sup>g</sup>Ondokuz Mayıs University, Turkey, Turkey

<sup>h</sup>Idele, Manosque, France

**Application:** To achieve effective communication to all stakeholder groups involved in sheep production many sources of media/communication are required.

**Introduction:** Poor adoption of technologies maybe due to stakeholders being un-aware of their existence. Successful transfer of findings/technology from research is critical to improving efficiency within any farm enterprise. Keady et al. [1] concluded that to achieve successful communication of technical/practical information the choice of media depends on the target stakeholder group. The objective of this survey was to identify the main sources that stakeholders (e.g., farmers, veterinarians) use to obtain information on flock nutrition and health.

**Materials and methods:** Eurosheep, an EU/international Thematic Network (grant agreement No. 863056) on sheep production designed to stimulate exchange of knowledge, best practice and innovation between research and stakeholders, with the objective of increasing flock profitability. From May to July 2020, a survey was circulated online to stakeholders involved in sheep production in all Eurosheep countries (Ireland, France, Greece, Hungary, Italy, Spain, Turkey and UK); language used was country specific. One of the questions asked was to 'rank in order of importance the main information sources that you use to get information on flock nutrition and health'. A score was assigned to each of the 13 choices: the first choice (most important) was scored 13 to the thirteenth choice (least important) scored 1. The final rank of the main sources of information was based on the sum of the scores.

**Results:** A total of 1163 surveys were completed. The respondents were classified into three groups, namely Europe (Eur: France, Greece, Hungary, Italy, Spain, Turkey and UK), Ireland farmers (IE-F, n =

Table 1

Sources used to obtain information on flock profitability.

	IE-F	IE-O	Eur
Peer to peer	1	4	5
Farming press	2	2	2
Technical advisors/consultants	3	1	3
Farming websites	4	11	6
Discussion groups	5	6	11
Veterinarians	6	8	1
Farm open days	7	4	12
Scientific papers	8	7	4
Sheep/agricultural shows	9	12	10
Social media (eg facebook, twitter, youtube)	10	10	9
Seminars/conferences/workshops	11	9	8
Professional learning	12	3	7
Technical sales personnel	13	13	13

125) and Ireland other (IE-O, n = 38). The main sources of information on flock health and nutrition are presented in Table 1. All groups ranked peer-to-peer, farming press and technical advisors/consultants as key sources of information. Whilst veterinarians were identified as the most important source of information in Eur, IE-F and IE-O respondents ranked them 6th and 8th, respectively. IE-O ranked farm open days 4th whilst IE-F ranked them 7th which may be a reflection of the proportion of each stakeholder group attending such events. Whilst IE-F ranked discussion groups as the main source of information on ewe productivity [1], discussion groups were ranked 5th in this survey. Similar to the findings of Keady et al. [1] the 3 categories of respondents ranked technical sales personnel and social media as the least important sources of information. Respondents ranked seminars/conferences/workshops as unimportant sources of information - being ranked 11th, 9th and 8th by IE-F, IE-O and Eur groups, respectively.

**Conclusion:** Whilst interactive communication and peer-to-peer are important media, the best sources of information differed between the categories of respondents. Technical sales personnel were ranked as the poorest source of information by all of respondents.

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### 33. What are the main challenges to improve flock profitability in Ireland and Europe?

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**Application:** There are similar challenges to improving sheep profitability across Europe. EuroSheep will identify appropriate solutions to improve sheep profitability across Europe based on the results of this survey.

**Introduction:** Eurosheep is an EU/international Thematic Network (grant agreement No. 863056) designed to stimulate knowledge, best practice and innovation exchange between stakeholders with the objective of increasing flock profitability. The objective of this paper is to present results of a survey undertaken in 8 countries to identify what are the main challenges/needs to improve sheep health and nutrition, thus flock profitability.

**Material and methods:** From May to July 2020, a survey was circulated online to stakeholders, in their own language, involved in sheep production in Eurosheep countries (Ireland, France, Greece, Hungary, Italy, Spain, Turkey and UK). The respondents were asked to rank, in order of importance, a maximum of five main challenges/needs regarding nutrition of adult sheep (choice of 12 options), lambs (10 options) and replacements (9 options); health of adult sheep/replacements (12 options) and lambs (10 options); and flock health and nutrition management issues (12 options). A score was assigned as follows: the first choice (most important) was scored 5 to the fifth choice (least important) scored 1. The final rank of challenges/needs was based on the sum of the scores.

**Results:** A total of 1163 surveys were completed. The respondents were classified into three groups, namely, Europe (Eur; France, Greece, Hungary, Italy, Spain, Turkey and UK), Ireland farmers (IE-F, n = 125) and Ireland other (IE-O, n = 38). The main challenges/needs identified to improve ewe and lamb nutrition, health, and flock management are presented in Tables 1 to 5. There was general agreement among the 3 categories of respondents on the main challenges/needs in nutrition, health, and management to improve flock profitability. Knowledge of nutrient requirement, forage feed value, grazing management, mineral nutrition and lamb performance targets were the main challenges identified for nutrition. Multi species swards were not identified as an important issue. Internal parasites, lameness, clostridial diseases, respiratory disorders and joint ill were the main health challenges. Flock health plan, anthelmintic resistance and sheep shed management were the main management challenges identified.

**Conclusions:** Similar challenges to improving sheep profitability exist across Europe but order of importance differs by region. As some challenges are region specific, due to differences in environment and production system, solutions may exist elsewhere.

Table 1

Main challenges/needs to improve ewe nutrition.

	IE-F	IE-O	Eur
Grazing management	1	1	2
Knowledge - nutrient requirements	2	2	1
Mineral and vitamins	3		4
Forage feed value	4	4	3
Diet crude protein concentration	5		
Conserved forage production		3	
Forage: concentrate ratio of diet		5	5

Table 2

Main challenges/needs to improve lamb nutrition.

	IE-F	IE-O	Eur
Grazing management	1	2	
Lamb performance targets	2	1	2
Knowledge - nutrient requirements	3	3	1
Growth targets - fattening lambs	4	4	3
Post weaning management	5	5	4
Concentrate feed efficiency			5

Table 3

Main challenges/needs to ewe health.

	IE-F	IE-O	Eur
Lameness (e.g. footrot)	1	1	2
Internal parasites (e.g. worms)	2	2	1
Clostridial disease (e.g. pulpy kidney)	3	3	5
External parasites (scab, lice)	4	4	
Mastitis	5		4
Poor body condition		5	
Respiratory disorders			3

Table 4

Main challenges/needs to improve lamb health.

	IE-F	IE-O	Eur
Lameness (e.g. footrot)	1	1	
Internal parasites	2	2	5
Accute death eg clostridial disease	2		3
Joint ill	3	5	
External parasites eg lice, blowfly	4	3	
Neonatal disease	5		1
CoccidiosisRespiratory disease	5	4	24

Table 5

Main challenges/needs to improve management.

	IE-F	IE-O	Eur
Flock health plan	1	1	2
Anthelmintic resistance	2	2	3
Outdoor welfare (shelter, shade)	3		4
Sheep-shed management	4	3	1
Lamb management	5		
Biosecurity		4	
Ration formulation		5	
Feed distribution (eg mixed ration)			5

### 34. Using urinary creatinine to predict daily urine output in sheep fed fresh forages

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**Application:** The new equation can be used to accurately predict daily urine output of sheep grazing fresh forages, including forages with very low dry matter (DM) that result in great urine output.

**Introduction:** Muscle creatinine (CRA) is catabolised at a constant daily rate and is exclusively excreted via urine. CRA is therefore commonly used to predict urine output (L/d) by the ratio between daily CRA excretion per unit of liveweight (LW) and urinary CRA concentration [1]. However, CRA excretion/kg LW does not appear to be constant and urine output in sheep is greatly underestimated on forage crops with low DM content (personal observation). The objective of this study was to develop an equation to predict urine output in sheep fed fresh pasture and alternative forages.

**Materials and methods:** Data ( $n = 155$ ; i.e., LW, LW change, feed DM, total urinary output (kg/d) and CRA concentration) was collected from seven sheep total excreta collection trials involving 29 dietary treatments (e.g. ryegrass-based pasture, forage brassicas and fodder beet) [2,3]. To predict daily urine output (kg/d), simple linear-, all subsets- and multiple- linear regressions were carried out (GenStat 19th edition). The best model was chosen based on percentage variance accounted for, square root of mean square prediction error (RMSPE) and based on

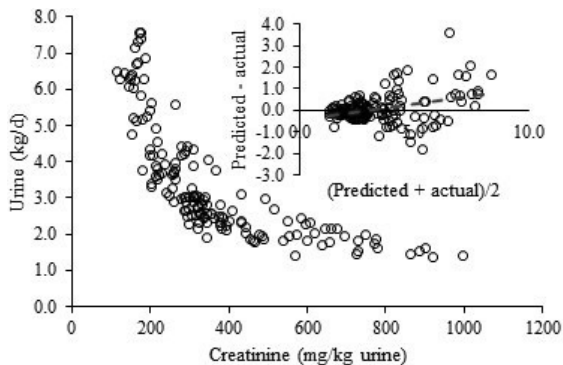


Fig. 1. Scatter plot of daily urine output (kg/d) vs urine creatinine concentration (mg/kg) and residual plot (small graph).

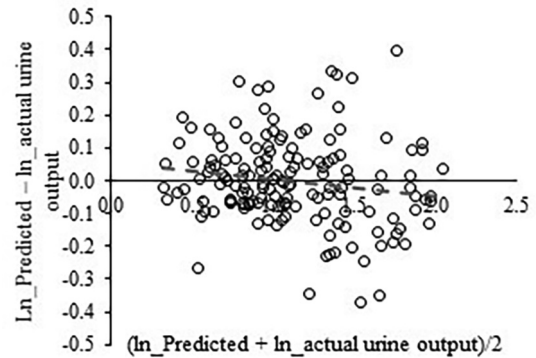


Fig. 2. Residual plot of actual vs. predicted urine output based on the equation:  $\ln_{\text{urine output}} = 5.474(0.133) - 0.8718 (0.232)\ln\text{CRA concentration} + 0.01663(0.00139)LW$ ; RMSPE = 0.53 g/d, 0.4% mean bias, 0.0% slope bias, 99.6% random error.

residual plots with least mean bias, slope bias (calculated as % of MSPE) and with homoscedastic variance. Scatter and residual plots revealed a non-linear relationship between urine output and urine CRA concentration (mg/kg) resulting in fan shaped residuals (Fig. 1). Therefore, urine output and CRA concentration had to be transformed using natural logarithms (ln).

**Results:** Based on all subsets regression it was found that  $\ln\text{CRA}$  concentration alone explained 81% of variation in  $\ln_{\text{urine output}}$  (RMSPE = 0.72 g/d), and this increased to 90% when LW was included in the prediction equation (RMSPE = 0.53 g/d), which also reduced slope bias from 5 to 0% of MSPE (Fig. 2).

**Conclusion:** Prediction of daily urine output was improved by ln transformation of measured urine output and including CRA concentration and LW in a multiple regression compared to the existing equation based on CRA excretion per unit of LW.

**Acknowledgments:** The animal trials were financially supported by PGgRc.co.nz, NZAGRC.co.nz and MPI.govt.nz/SLMACC and the laboratory analysis was carried out at Lincoln University.

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### 35. The half-life of cobalt in lamb plasma following oral administration of a cobalt supplement

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**Application:** The effect of orally-administered Co on plasma-Co concentration has essentially disappeared by approximately day 21 post treatment, regardless of dose

**Introduction:** Cobalt is an essential trace element for ruminants; its concentration in herbage on sheep farms often fails to supply the daily requirements of growing lambs. Consequently, lambs require supplementary Co, especially post weaning; this is commonly supplied by a Co drench. Since plasma vitamin B<sub>12</sub> concentration is significantly related to Co concentration (Hession and Kendall 2017; and unpublished data), the half-life of plasma Co is relevant; data from a recent study were used to provide evidence on this aspect of Co supplementation.

**Materials and methods:** Data on plasma Co concentration, from the study by Hession et al. (2021), were for lambs drenched with either 31.5 mg (n = 9) or 63 mg (n = 8) Co (as CoSO<sub>4</sub>·7H<sub>2</sub>O<sub>2</sub>) at intervals of 21 and 42 days, respectively, over an 84-day period, plus untreated controls (n = 12). Lambs (mean BW 39.5, s.d. 5.46, kg) had been weaned ~ 2 months prior to the study and were rotationally grazed as a flock on permanent pasture. Blood was collected every 7 days, from day 7 post initial drench, and analysed for Co concentration (details in Hession et al. 2021). Cobalt concentration was log<sub>e</sub> transformed prior to statistical analysis and regressed on sample day to estimate half-life; statistical analyses used mixed-model procedures (Proc MIXED; SAS, version 9.4; ©2002-2012) with fixed effects for treatment and treatment sequence, and random terms for regression for each animal. [2,3].

**Results:** Time trends in the Co concentration for control animals were significant (P < 0.05); estimates of half-life were based on the difference in regression coefficients between treated and control groups. The regression on time was quadratic (P < 0.001) in the case of the 63 mg Co dose (Table 1). To directly compare the two treatments, the data

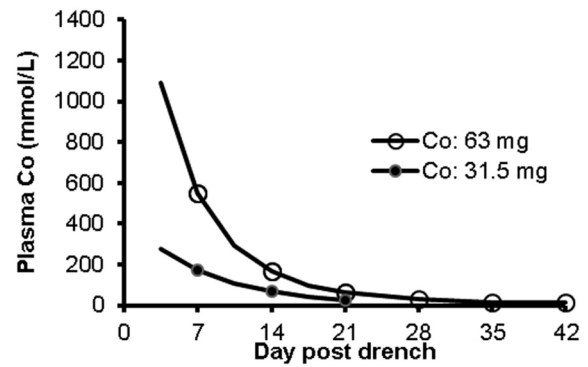


Fig. 1. Effect of cobalt drench on plasma cobalt relative to that for untreated controls.

were restricted to the first 21 days post drenching. The decline was linear and greater in the 63 mg group (P < 0.05). Estimates of half-life, with 95% confidence intervals, are presented in Table 1 and indicate that increasing the Co dose reduced half-life. This is consistent with Williams et al. (2017) whose estimates were from data over the first 13 days post drench. The plasma profiles showing the difference between treated and control lambs are in Fig. 1. It is evident that by days 21 to 28 post drench the effect of the Co drench on plasma Co has essentially disappeared.

**Conclusions:** The half-life of Co in plasma is reduced when the initial Co dose is increased. At the recommended dose (31.5 mg), or twice that amount, the effects on plasma Co concentration have essentially disappeared by day 21.

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Table 1

Regression of plasma cobalt (log<sub>e</sub> scale) on day post drench and estimates of cobalt half-life in plasma.

Co dose	Sample time points(days post-drench treatment)	Regression coefficients ± SE	Estimated half-life (days)	
			Mean	95% Confidence interval
31.5 mg	7,14,21	Linear: -0.105 ± 0.0081	6.6	6.0 - 7.4
63 mg	7,14,21	Linear: -0.124 ± 0.0095	5.0	5.0 - 6.4
63 mg	7,14,21,28,35,42	Linear: -0.159 ± 0.0160	4.9	3.7 - 6.2
		Quadratic: 0.00176 ± 0.00032		

### 36. Substitution of soybean meal with canola meal, cottonseed cake and Vicia faba L. Minor seeds and its effects on milk production and quality traits in dairy sheep

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**Application:** Soybean meal (SOY) constitutes a widely used protein supplement in dairy sheep diets. Replacing SOY with locally produced feed-stuff to enhance the circular agriculture model is a significant endeavor.

**Introduction:** Reducing the dependency on SOY in diets for food-producing animals has emerged due to economic and environmental issues. However, the effects of SOY replacement on the productivity of dairy sheep are not sufficiently documented. The objective of this study was to assess the effects of replacing SOY with a mixture of canola meal (CAN), cottonseed meal (COT) and Vicia faba L. minor (VFM) seeds, on milk yield (MY) and quality traits (MQT) in intensively reared dairy sheep.

**Materials and methods:** Sixty-four Frizarta (Farm A) and 48 Chios (Farm B) intensively reared purebred dairy ewes were enrolled in the study. In each farm, postweaning ewes were allocated into two equal groups, the control (CR) and the experimental ration (ER) group, based on their lactation number and MY. In the ER, 3/4 of SOY was replaced by a mixture of CAN, COT and VFM, to produce a ration of similar nutritional value

(16.0% crude protein, NEL 8.16 MJ). Each group was permanently housed in separate pen and isoenergetic/isoproteic quantities of concentrates and roughages were provided twice a day. Following a 30-day adaptation period to the diets, MY, MQT [daily fat-, protein-, lactose-, and total solids- yield, log of somatic cell counts (logSCC), pH and electrical conductivity (EC)] and body condition score (BCS) were recorded for each individual ewe monthly, for 4 months. Descriptive statistics were calculated and the following mixed linear regression model was used in SPSS v23 to estimate the effect of the two rations on MY and MQT (only main effects were considered):

$$Y_{ijklm} = \mu + F_i + G_j + P_k + S_l + a_1 \times BCS + E_m + \delta_{ml} + e_{ijklm}$$

where,  $Y_{ijklm}$  = dependent variables of MY and MQT,  $\mu$  = intercept,  $F_i$  = fixed effect of the farm,  $G_j$  = fixed effect of the ration ( $j = 2$  levels),  $P_k$  = fixed effect of the parity number,  $S_l$  = fixed effect of the sampling occasion ( $l = 4$  levels),  $a_1$  = fixed effect of the regression coefficient of BCS,  $E_m$  = random variation of the  $m^{\text{th}}$  ewe,  $\delta_{ml}$  = repeated variation of the  $m^{\text{th}}$  ewe in the  $l^{\text{th}}$  sampling occasion, and  $e_{ijklm}$  = residual error.

**Results:** No significant differences were observed between the two rations in most parameters except for daily fat yield (DFY), which was increased by 9 g ( $P < 0.05$ , 95% CI, 0.7 to 16.4 g) and the EC which was decreased by 85  $\mu\text{S}/\text{cm}$  ( $P < 0.001$ , 95% CI, 40.1 to 130.4  $\mu\text{S}/\text{cm}$ ) in the ER group (Table 1).

**Conclusion:** Partial replacement of SOY by a mixture of CAN, COT and VFM did not adversely affect DMY and MQT. Contrary, a favourable effect on daily fat yield was observed. A cost-efficiency analysis is imperative for the commercialization of the proposed ration.

**Acknowledgments:** Funded by the Agricultural Cooperative 'Agrinio Union' within the Regional Operational Program 'Western Greece 2014-2020-RIS3', Project code: MIS-5040306.

Table 1

Milk yield and milk quality traits for the two ration groups, and the effects of the explanatory

variables used in the mixed linear model.

Response variables	CR group Mean $\pm$ SD	ER group Mean $\pm$ SD	P-value			
			Ration	Sampling	Farm	BCS <sup>1</sup>
Daily milk yield (l)	1.1 $\pm$ 0.51	1.1 $\pm$ 0.50	ns	***	ns	ns
Daily fat yield (g)	68.1 $\pm$ 27.97	77.3 $\pm$ 29.63	*	***	ns	ns
Daily protein yield (g)	61.4 $\pm$ 28.18	65.6 $\pm$ 28.95	ns	***	ns	ns
Daily lactose yield (g)	50.0 $\pm$ 25.51	51.5 $\pm$ 24.97	ns	***	ns	ns
Daily total solids yield (g)	187.9 $\pm$ 82.89	204.8 $\pm$ 84.53	ns	***	ns	ns
Log of SCC	5.7 $\pm$ 0.77	5.7 $\pm$ 0.61	ns	***	***	ns
pH <sup>†</sup>	6.6 $\pm$ 0.15	6.6 $\pm$ 0.15	ns	***	ns	**
Electrical conductivity ( $\mu\text{S}/\text{cm}$ ) <sup>†</sup>	1537.6 $\pm$ 178.99	1449.4 $\pm$ 211.07	***	***	***	***
BCS	2.8 $\pm$ 0.33	2.8 $\pm$ 0.32	NA	NA	NA	NA

CR: control ration; ER: experimental ration; SD: standard deviation; ns: not significant; \* $P < 0.05$ ; \*\* $P < 0.01$ ; \*\*\* $P < 0.001$ ; <sup>†</sup>measured at 20 °C; BCS: body condition score; NA: data not available; <sup>1</sup>specified value of covariate in the model: BCS = 2.8

### 37. Effect of dietary inclusion of a mannan-oligosaccharide (MOS) on ewe performance

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**Application:** Dietary inclusion of MOS may increase colostrum and milk component yield in lactating ewes.

**Introduction:** Since the EU ban on antibiotics as growth promoting agents in animal feeds there has been increased interest in the use of prebiotics as an alternative. Prebiotics are defined as “non-digestible feed ingredients that beneficially affect the host by selectively stimulating the growth of one, or a limited number of microbes in the gut” [2]. They include non-digestible oligosaccharides and their mode of action is thought to include alteration of the intestinal microflora to reduce colonisation by pathogens and enhancing intestinal integrity. Actigen® is a commercially available MOS derived from the *Saccharomyces cerevisiae* that has been shown to enhance gut health, immune status, and performance in various species. However, limited work has been conducted in ruminants. The objective was to investigate the effect of dietary inclusion of Actigen® on ewe performance.

**Materials and methods:** Forty-two twin bearing Suffolk × Mule ewes were housed individually from week -6 *pre-partum* to week +4 *post-partum* and allocated by parity, live weight (LW) and body condition score (BCS) to one of two treatments. All ewes were offered chopped straw *ad-libitum*, together with concentrates containing either no Actigen® (C) or 1.0 g/kg Actigen® (A). Concentrates were fed to provide a rising plane of nutrition during late pregnancy and 3.0 litres of milk during early lactation [1]. Straw intake was recorded by weighing the amount offered daily and weighing back refusals twice weekly. Ewe LW and BCS were recorded weekly and colostrum and milk yield were estimated at 16 hours and 21 days. Lambs were separated from the ewes, which were then injected with 1.0 ml oxytocin and machine milked until the udder was empty. The procedure was repeated 4 hours later, and milk secretion rate calculated. Samples of colostrum and milk were collected and stored at -20 °C prior to further analysis. The data were analysed by ANOVA as a randomised block design using Genstat 18 (Figs 1 and 2; Table 1).

**Results:** Inclusion of Actigen® (1.0 g/kg) had no effect on LW and BCS change *pre-partum*, but increased LW loss *post-partum*. Inclusion of Actigen® also increased colostrum and milk components yields by increasing colostrum yield (3.55 vs 3.11; SED 0.172 kg/day) and composition, and milk yield (3.16 vs 2.78; SED 0.337 kg/day). However, it had no effect on milk composition.

**Conclusions:** Inclusion of 1.0 g/kg Actigen® increased ewe colostrum and milk component yield. This may partly be explained by increased LW loss. The mode of action of MOS in ruminants requires further investigation.

The authors gratefully acknowledge the support of Alltech Limited for funding this study.

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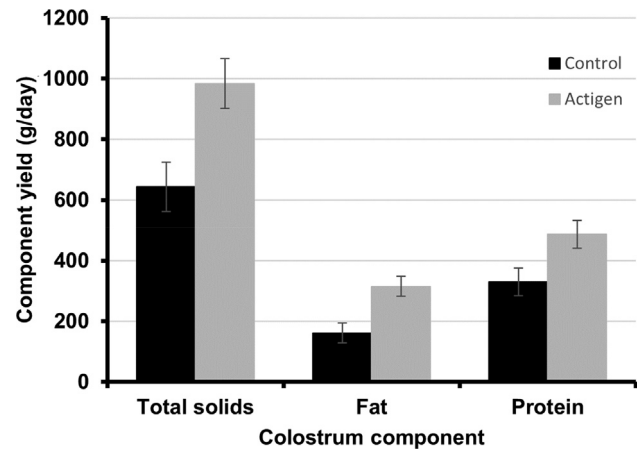


Fig. 1. Effect of Actigen on colostrum component yields.

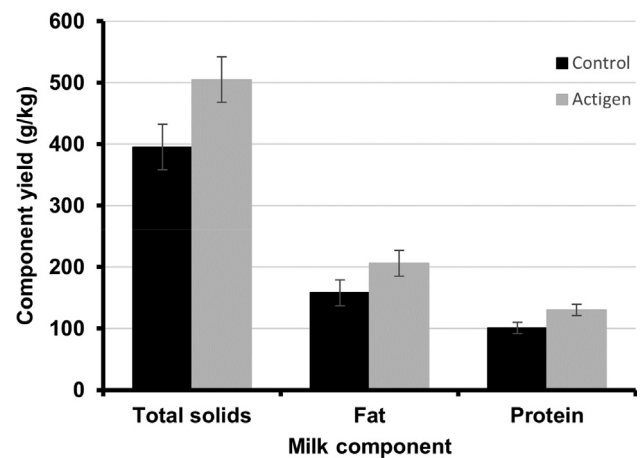


Fig. 2. Effect of Actigen on milk component yields.

Table 1  
Effect of Actigen® on ewe LW and BCS change.

		C	A	SED	P
Pre-partum	LW (kg)	10.2	9.3	0.87	0.296
change	BCS	-0.17	-0.27	0.064	0.107
Post-partum	LW (kg)	-3.2	-7.2	1.10	0.003
change	BCS	-0.21	-0.27	0.075	0.514

### 38. The impact of nutrition on intramuscular omega-3 fatty acid composition of lamb meat: A systematic review and meta-analysis

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**Application:** The omega-3 (n-3) polyunsaturated fatty acid (PUFA) profile of lamb meat can be comparably enhanced through pasture-based feeding and the use of rumen protected and marine dietary supplements. Lamb products, enriched in n-3 PUFA, present the opportunity to improve human health with little change to habitual dietary choices.

**Introduction:** The n-3 PUFA composition of meat is directly influenced by the diet of the animal [2]. There has been much interest in pasture feeding and n-3 rich supplementation including oilseeds, marine (fish oil/meal and microalgae) and rumen protected fats [1]. This study uses meta-analysis to identify trends and strengths of relationships from published research evaluating the effect of lamb diets, including nutritional supplements, on n-3 PUFA composition in muscle.

**Materials and methods:** A search strategy including key words such as lamb, diet and omega-3 was developed. Web of Science, ScienceDirect and OVID Medline were searched identifying 1768 studies, of which 22 were eligible for inclusion. Alpha-linolenic acid (18:3n-3), eicosapentaenoic acid (20:5n-3), docosapentaenoic acid and docosahexaenoic acid n-3 data were extracted from each individual treatment (n = 66) within the 22 included studies. Data were analysed using Rev-Man 5.3 software which used a random effects model to determine differences between mean and standard deviation between study treatments. An  $I^2$  statistic to level of heterogeneity was also assessed.

**Results:** The most prominent results from the study were the effect of pasture and marine and n-3 rumen protected fats, using 8 and 9 studies, respectively (see Fig. 1). Pasture-based feeding has a positive effect on 18:3n-3 (ES = 1.99; CI = 1.29, 2.70;  $I^2$  87%) and a uniquely strong effect on 20:5n-3 (See Fig. 1; ES = 2.46, CI = 1.56, 3.35;  $I^2$  87%). In addition, a positive, yet comparable effect was determined when feeding marine and rumen protected supplements on 20:5n-3 (Fig. 1; ES = 2.37, CI = 0.94, 3.80;  $I^2$  91%).

**Conclusions:** This study recognised the positive impact of pasture feeding on 18:3n-3 and 20:5n-3. Similarly, the effective use of marine and rumen protected supplements to elevate levels of 20:5n-3 in muscle were identified. This is unique as pasture does not naturally possess high levels of 20:5n-3, especially compared to marine supplements. This was a significant finding which was facilitated through the power of the meta-analytical approach and has also identified the benefit of larger sample numbers in animal studies to enhance statistical power.

**Acknowledgments:** This PhD studentship was funded by The Department for the Economy (DfE).

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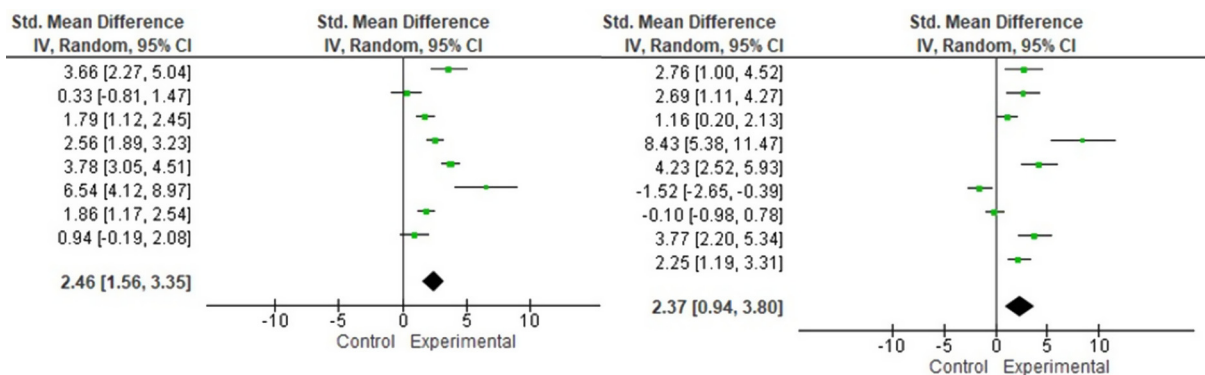


Fig. 1. Effect of pasture diets (left) and marine and rumen protected supplements (right) on 20:5 n-3 concentration in meat.

### 39. Dose-response effects of microalgae supplementation on feed intake and weight gain in finishing lambs

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**Application:** Microalgae supplementation at high levels can cause negative effects on feed intake of finishing lambs

**Introduction:** In livestock ruminant systems, feeding costs account for 60–70% of total expenditure, thus, it is of utmost importance to maximise feed utilisation and nutrient availability for the animal to assure future food security. In lambs, microalgae supplementation has shown effects on palatability and animal performance. It is still unclear how the inclusion of increasing levels of microalgae may affect BW gain and dry matter intake (DMI). The study aims to investigate the effects of supplementing a microalgae meal on growth and feed intake of finishing lambs.

**Materials and methods:** Fifty-six male lambs, Texel/Scottish black face, were blocked into 14 groups of 4 lambs according to age and body weight (BW), and then randomly assigned within each group to one of the four treatment diets. Concentrate included: 0% (Ctrl), 1.2% (Alg-L), 2.4% (Alg-M) and 3.6% (Alg-H) microalgae (DM basis). Treatments were gradually introduced by replacing silage grass (DM basis) over a period of 7 days. All sheep were fed TMR diets of 50:50 grass silage and concentrate for 35 days pre-slaughter. Individual DMI and BW were recorded daily and weekly, respectively. Average daily gain (ADG), relative growth, gross energy intake (GEI) and feed conversion ratio (FCR) were individually calculated. Univariate analyses were performed using a linear mixed model (LME) methodology via the REML estimation method in the NLME package in R (R version 3.6.2). All variables were analysed using treatment as the sole fixed effect and lamb as random effect. Resulting LME models were evaluated using an analysis of

Table 1

Characterisation of feed intake and growth performance in finishing lambs supplemented with different levels of microalgae (n = 56).

	Ctrl.	Alg-L.	Alg-M.	Alg-H.	SEM <sup>1</sup>	P-value.
Initial BW, kg.	31.4.	31.5.	32.6.	31.0.	0.92.	0.646.
Final BW, kg.	39.1.	40.6.	39.7.	38.6.	0.92.	0.453.
Relative growth <sup>2</sup> , %	22.2 <sup>ab</sup> .	25.1 <sup>a</sup> .	20.0 <sup>b</sup> .	21.9 <sup>ab</sup> .	1.32.	0.068.
Average daily gain, g/d.	220.0 <sup>ab</sup> .	257.8 <sup>a</sup> .	204.3 <sup>b</sup> .	217.1 <sup>ab</sup> .	12.19.	0.020.
DM intake, kg/d.	1.17 <sup>a</sup> .	1.18 <sup>a</sup> .	1.19 <sup>a</sup> .	1.05 <sup>b</sup> .	0.03.	0.006.
GE intake, MJ/d.	22.1 <sup>a</sup> .	22.4 <sup>a</sup> .	22.8 <sup>a</sup> .	20.0 <sup>b</sup> .	0.59.	0.009.
DMI/BWG (FCR), kg/kg.	5.6.	4.7.	6.3.	5.0.	0.45.	0.081.

<sup>a-c</sup> Least square means within a row with different superscripts differ significantly ( $P < 0.05$ ). <sup>1</sup>SEM = standard error of the mean. <sup>2</sup>Relative growth (%) =  $(W2 - W1)/(W1 + W2)/2 * 100$ ; W1 = Initial BW, W2 = Final BW.

variance (ANOVA). Predicted means from the model, together with estimates of the standard error of the mean and pairwise comparisons (Tukey test) were obtained using the PREDICTMEANS package of R. Statistical significance was declared at a P-value  $\leq 0.05$ .

**Results:** The ADG was greater ( $P = 0.020$ ) and the relative growth tended ( $P = 0.068$ ) to be greater in Alg-L than in Alg-M. The DMI and GEI were lower ( $P \leq 0.009$ ) in Alg-H than the other diets (Table 1). The FCR tended ( $P = 0.081$ ) to be lower in Alg-L and Alg-H compared to Ctrl and Alg-M. No differences ( $P \geq 0.453$ ) between Ctrl and Alg-fed lambs were observed for initial BW, final BW.

**Conclusions:** The present study showed that supplementation of high levels of microalgae in the diet reduced DMI without effects on ADG and FCR of finishing lambs. While, the low and medium levels of microalgae in the diet did not affect DMI when compared to controls, but low supplementation levels resulted in improved animal growth when compared to medium levels of microalgae inclusion. Further studies are required to assess such effects during the whole finishing growth period of lambs.

**Acknowledgements:** The authors acknowledge funding from European Union (H2020-SFS-2018-1 project MASTER-818368) and Devenish Nutrition.



#### 40. Post-weaning growth performance of piglets differing in weaning age (lactation length) and post-weaning dietary zinc oxide content

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AFBI, Hillsborough, UK

**Application:** Extending lactation can increase piglet weaning weight. Piglet growth performance was unaffected by the removal of Zinc Oxide (ZnO) from post-weaning diets.

**Introduction:** From 2022 the medicinal use of ZnO will be banned in the EU. Therefore producers must seek alternative methods to maintain piglet growth performance and health without increasing the use of antibiotics. Evidence suggests later weaning may reduce diarrhoea and increase piglet growth [1]. This work evaluates the impact of extending lactation length and post-weaning diets without ZnO on piglet performance.

**Materials and Methods:** Sows and their respective piglets ( $n = 347$ ) were assigned to one of two experimental groups balancing for sow parity, body weight (BW), and piglet birth weight: Lact28, litters weaned at day 28 of lactation and Lact35, litters weaned at day 35 of lactation. At weaning, piglets were allocated into pens (10 piglets/pen) according to weaning BW and provided with starter diets with ZnO (16.5 MJ DE/kg, 22.5% CP) or without ZnO and reduced CP (Control: 15 MJ DE/kg, 18.5% CP). Resulting in a  $2 \times 2$  factorial arrangement: Lact28-Control, Lact28-ZnO, Lact35-Control, and Lact35-ZnO. At 42 days of age piglets were offered Starter diet 2 (ZnO: 15.8 MJ DE/kg, 22% CP; Control: 14.88 MJ DE/kg, 18.7% CP) until 50 days of age, and a Grower diet until 10 weeks of age. Diets were formulated to a similar Energy-Lysine ratio.

Piglet BW and pen feed intake were recorded at 28, 35, 40d (Stage 1) and 10 weeks post-farrowing. Average daily gain (ADG), average daily feed intake (ADFI) and feed conversion ratio (FCR) were calculated. Performance data were analysed by REML for fitting linear models using GenStat version 19. Weaning age and ZnO content were included in the model as fixed effects as well as a two way interaction between Weaning age  $\times$  ZnO content. Average BW at 28d and replicate were included as random effects and pen was considered the statistical unit.

**Results:** No significant interaction effects were reported, only main effects are presented (Table 1). Lact28 piglets had higher ADFI and ADG after weaning. Lact35 piglets were heavier at 35d and Stage 1 and had improved FCR between 35d-Stage 1. Piglets in both treatments were of similar BW by 10 weeks. Piglets offered Control diets displayed increased ADFI and higher FCR between 35d-Stage 1. However, overall growth performance of piglets up to week 10 was not effected by ZnO content.

**Conclusions:** Extending lactation increased piglet BW at weaning and early post-weaning, but due to reduced ADFI and ADG of Lact35 piglets, piglets were similar in BW by week 10. While removing dietary ZnO did not affect piglet growth performance, we must consider impacts on gut health and the incidence of diarrhoea.

**Acknowledgements:** Research funded by DAERA.

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Table 1  
Effects of weaning age and ZnO diet content on piglet growth performance from 28 days to 10 weeks post-farrowing.

	Lact28	Lact35	SED	Significance	ZnO	Control	SED	Significance
Average weight 28d, kg	9.13	9.15	0.324	n.s	9.22	9.05	0.324	n.s
Average weight 35d, kg	9.90	11.64	0.096	<0.001	10.81	10.72	0.096	n.s
Average weight stage 1, kg	11.87	13.34	0.137	<0.001	12.62	12.49	0.136	n.s
Average weight week 10, kg	28.58	28.71	0.448	n.s	28.94	28.34	0.450	n.s
ADFI <sup>1</sup> 35d-Stage 1, kg	0.53	0.21	0.029	<0.001	0.35	0.40	0.029	<0.05
ADFI 35d-10wk, kg	0.77	0.67	0.025	<0.001	0.72	0.73	0.255	n.s
ADG <sup>2</sup> 35d-Stage 1, kg	0.32	0.26	0.022	<0.05	0.31	0.30	0.022	n.s
ADG 35d-10wk, kg	0.56	0.51	0.014	<0.001	0.54	0.52	0.014	n.s
FCR <sup>3</sup> 35d-Stage 1, kg/kg	1.68	0.82	0.137	<0.001	1.15	1.42	0.137	<0.05
FCR 35d-10wk, kg/kg	1.40	1.33	0.044	n.s	1.35	1.38	0.044	n.s

<sup>1</sup> Average daily feed intake

<sup>2</sup> Average daily gain

<sup>3</sup> Feed conversion ratio

#### 41. Dietary inclusion of galacto-oligosaccharides improves weaner pig growth performance

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**Application:** Dietary inclusion of galacto-oligosaccharides at levels 0.5 – 1% w/w GOS dry solids/feed may be efficacious at improving weaner pig growth performance.

**Introduction:** Galacto-oligosaccharides (GOS) are registered feed materials for animal diets, with prebiotic properties [1]. Improving pig gut health using prebiotics is a potential strategy in the management of abrupt weaning for overcoming challenges such as pathogenic diarrhoea. The aim was to evaluate the efficacy of GOS supplementation for improved pig growth performance during the pre- and post- weaning periods.

**Materials and methods:** Approval was granted by Harper Adams University Research Ethics Committee. A randomised block (batch) design to test the effect of dietary GOS (Nutrabiotic®, Dairy Crest Ltd, UK) at four levels: T1 (negative control, NC) – 0.00% product inclusion (0.00 % w/w GOS dry solids/feed); T2 – 0.96% product inclusion (0.5% w/w GOS dry solids/feed); T3 – 1.92% product inclusion (1.0% w/w GOS dry solids/feed); T4 – 3.84% product inclusion (2.0% w/w GOS dry solids/feed), was used. Pre-weaning, 12 litters/treatment (over two farrowing batches) of 12 ( $\pm$  1) piglets were offered creep feed containing one of the four treatments for 14 days prior to weaning. At weaning (~28 d/age), with the exception of batch 1, 35 piglets per treatment were balanced for weight, litter and sex. These were housed in groups of five, mixed sex animals, providing 7 pen replicates/treatment. In batch 1, there was 1 pig per pen extra. After the first week, 1 pig/pen in batch

1 (closest male to average pen weight) was humanely killed for tissue samples. Treatments were fed for 36 days post-weaning and were formulated by Primary Diets Ltd (UK) and manufactured by Target Feeds (UK). Diets were iso-nutritive. Body weights (BW) were recorded at farrowing, day 14 and at weaning. Post-weaning BW and feed disappearance were recorded at day 7, 14, 22 and 36. Data were subject to one-way ANOVA, in GenStat (17th edition). If significant ( $P < 0.05$ ), Tukey test separated means. A contrast comparison compared T1 (NC) and T2 (test substance).

**Results:** There was no significant difference in average pig weaning weight ( $p = 0.72$ ) or DLWG ( $p = 0.24$ ) pre-weaning. Seven days post-weaning, pigs on T4 were lighter in BW ( $P < 0.05$ ) than those fed the lower doses of GOS but were not significantly different ( $P > 0.05$ ) to NC (Table 1). At 36 days post-weaning, pigs fed T1 were 3.3% heavier compared to the NC ( $P = 0.048$ ).

**Conclusions:** Supplementing piglets with GOS at 0.5 – 1% for up to 5 weeks post-weaning may improve post-weaning growth performance. Increasing GOS to 2% may reduce growth performance to the level of the NC, suggesting an upper limit of inclusion.

**Acknowledgments:** HAU pig unit technicians (Richard Hooper, Fred Baker, Sarah Icelly and Joshua Osmond). Mr Simon Hunt and Dr Neville Fish of Dairy Crest Ltd provided technical assistance. Funding received from Dairy Crest Ltd.

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Table 1

The effect of Nutrabiotic® GOS supplementation (w/w GOS dry solids/feed) on weaner pig performance.

Diet	T1 (0%)	T2 (0.5%)	T3 (1%)	T4 (2%)	LSD (51 d.f.)	p-value	CV%
Mean weaning weight (kg)	8.57	8.57	8.60	8.58	0.081	0.85	1.2
Mean BW (kg) d7 PW	9.82 <sup>ab</sup>	9.98 <sup>b</sup>	9.88 <sup>b</sup>	9.55 <sup>a</sup>	0.235	0.004	3.2
Mean BW (kg) d14 PW	12.92	13.08	12.80	12.59	0.488	0.25	5.0
Mean BW (kg) d22 PW	17.59	17.95	17.81	17.21	0.607	0.09	4.5
Mean BW (kg) d36 PW	28.96	29.94	29.50	28.70	0.977	0.06	4.4

LSD = Least significant difference; different superscript letters within rows are different ( $p < 0.05$ ); BW = body weight; PW = post-weaning.

## 42. Finishing performance of pigs fed dry feed vs. wet/dry feed

Ramon Muns

Agri-Food and Biosciences Institute, Hillsborough, Northern Ireland, UK

**Application:** Finishing pigs fed a Wet/dry system show a productive and efficiency performance similar to dry fed pigs.

**Introduction.** Wet/Dry system consists on delivering water and compound feed at a desired ratio but keeping them separate up to the point of delivery. When compared to dry feeding, nursery pigs fed in a wet/dry system showed increased body weight, average daily gain (ADG) and average daily feed intake (ADFI), and similar feed conversion ratio (FCR) [1]. Different systems often result in different type of feeders, influencing feeder or space allowance, in turn influencing number of fights [2]. Using a wet/dry feeding system that delivers water and feed separately in the trough, the present study aimed to compare productive performance, body lesions (BL), and pen dirtiness score (pDS) of finishing pigs fed either dry feed or wet/dry feed.

**Materials and methods:** Under the Animals (scientific procedures) act 1986, 260 Duroc (LD × LW) pigs were distributed to one of two treatments at 12 weeks of age, balancing for body weight and gender: Dry) pigs were offered dry feed; Wet 3:1) pigs were offered a wet/dry diet at a 3:1 ratio. Pigs were allocated in 18 pens of 14–16 animals. All pigs were offered the same finisher diet during the trial (14 MJ DE/kg, 15.5% CP) fed *ad libitum*. Due to facilities characteristics, pigs on the Wet 3:1 treatment had one feeder (400 × 1,250 cm), while pigs on the Dry treatment had two feeders (300 × 380 cm) per pen. Pigs were weighed at 12, 17 and 22 weeks of age. ADFI, ADG, and FCR was recorded. At 12 and 22 weeks, BL score (0–2) was assessed at anterior, mid and posterior locations of the body; and pDS (0–4) was assessed according to the proportion of the pen being soiled. Data were analysed by ANOVA with pen as experimental unit employing GenStat 16th edition. (Table 1).

**Results:** Pigs fed Wet 3:1 were lighter at 22 weeks of age. No differences between treatments were observed for ADG, ADFI or FCR. At 22 weeks, Wet 3:1 had more BL and higher pDS.

Table 1

Effect of treatment on pigs' body weight, average daily gain (ADG), average daily feed intake (ADFI), feed conversion ratio (FCR), lesion score, and pen dirtiness score.

	Dry/control	Wet 3:1	SEM	p-value
<i>n</i> ° of pigs/pens	<b>n = 129/9</b>	<b>n = 130/9</b>		
12 weeks weight, kg	38.5	38.5	0.61	n.s.
17 weeks weight, kg	66.8	66.1	0.59	n.s.
22 weeks weight, kg	106.3	103.7	0.87	0.036
ADG 12 to 17 weeks, kg	0.77	0.73	0.050	n.s.
ADG 12 to 22 weeks, kg	0.98	0.96	0.069	n.s.
ADFI 12 to 17 weeks, kg	1.88	1.79	0.045	n.s.
ADFI 12 to 22 week, kg	2.41	2.36	0.060	n.s.
FCR 12 to 17 weeks	2.54	2.46	0.132	n.s.
FCR 12 to 22 weeks	2.46	2.46	0.055	n.s.
Lesion Score 12 weeks	0.29	0.27	0.058	n.s.
Lesion Score 22 weeks	0.88	1.35	0.120	0.006
Pen Dirtiness Score 12 weeks	2.52	2.81	0.167	n.s.
Pen Dirtiness Score 22 weeks	0.31	1.46	0.283	0.012

<sup>a,b</sup>Values within row with different superscript are statistically different from each other

**Conclusions:** Although statistically significant, weight at 22 weeks of age between groups only differed by less than 3 kg. Despite differences in feeder space, pigs fed a wet/dry diet at a 3:1 ratio showed similar performance indicators and efficiency than pigs fed dry. Pigs increased BL with age/weight. Difference in BL is most probably due to difference in feeder space. However, feeding pigs a Wet/dry diet increases pen dirtiness at the end of the fattening period.

**Acknowledgments:** Research funded by DAERA and Pig Regen Ltd.

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### 43. Effect of soyameal replacement by Black Soldier Fly (*Hermetia illucens*) meal in grower-finisher pig diets on productive performance and body composition

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**Application:** The inclusion of Black Soldier Fly meal in pig diets as a replacement of soya may reduce the pressure on deforestation in South America and increase the feed security in the UK

**Introduction:** The use of soymeal as the major protein source in animal feeds raises concerns due to the increased pressure on the food-feed-fuel competition and deforestation [1]. Insects have the potential to contribute to a circular economy, although further nutritional information is required from animal production perspective. Black Soldier Fly (BSF) (*Hermetia illucens*) represents the most promising insect species produced for large-scale replacement of soyameal in pig nutrition. This study aimed to assess differences in the productive performance and body composition between soyameal and BSF meal as the primary protein source in grower-finisher pig diets.

**Materials and methods:** A total of three experimental diets were formulated to have the same net energy and essential amino acids with increasing levels of BSF larva meal in substitution of 0, 33% and 66% (C, BSF5% and BSF10%, respectively) of soyameal. The diets were formulated to meet the nutrient requirements according to NRC (2012). Feed and fresh water were provided *ad libitum*. A total of 36 (Dan Duroc × (Large white × Landrace)) pigs (50% male/50% female) were individually weighed (initial body weight: 45.3 ± 2.90 kg) and

randomly allotted in individual pens to the three previously described dietary treatments (n = 12). The pigs were weighed every week until the end of the trial (LW). The trial lasted eight weeks and was conducted under the Animals (Scientific Procedures) Act 1986 and procedures were approved by the Animal Welfare and Ethical Review Body. The average daily gain (ADG), average daily feed intake (DFI) and feed conversion ratio (FCR) were calculated for the whole experimental period. After slaughter, carcass composition was assessed using DXA and images were analysed following Kipper et al. (2018). Data were analysed by a one-way ANOVA with the diet as the main treatment employing SPSS 25th edition (Table 1).

**Results:** No significant differences were observed between the dietary treatments in any of the productive parameters or in the body composition proportions after the 8-week period. However, the direct comparison between C and the BSF10% diet revealed a trend (P < 0.10), suggesting that ADG was improved through BSF inclusion. [1,2].

**Conclusions:** The replacement of up to 66% of the soyameal by partially defatted BSF larva meal revealed no detrimental effect on grower-finisher pigs' productive performance and body composition. BSF larvae meal can be considered as a potential sustainable source of protein in pig nutrition. Further research should focus on the potential benefits of BSF meal on gastrointestinal microbiota.

**Acknowledgments:** This research was funded by Agri-Food Quest Competence Centre programme (INI-11-12-17-004-AFQCC) funded by Invest NI and Finnebrogue, Moypark, Cranswick.

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Table 1

Effects of dietary BSF larva meal inclusion on growth performance and body composition parameters of the pigs.

	Diet	C	BSF5	BSF10	SEM	P	Contrasts			
							1	2	3	4
LW (g)	Initial	46.2	45.3	44.8	0.531	0.595	0.511	0.319	0.730	0.340
	Final	105.8	106.9	108.8	0.957	0.854	0.636	0.205	0.421	0.314
ADG (g)		1.22	1.26	1.31	0.019	0.145	0.362	<b>0.051</b>	0.280	0.098
DFI (g)		2.98	3.01	3.05	0.074	0.853	0.779	0.576	0.780	0.627
FCR (g/g)		2.45	2.39	2.34	0.033	0.350	0.458	0.150	0.476	0.208
Lean %		73.8	73.3	73.3	0.368	0.788	0.560	0.545	0.983	0.493
Fat %		24.0	24.5	24.5	0.459	0.815	0.585	0.578	0.992	0.525
Bone %		2.03	2.03	2.03	0.006	0.844	0.793	0.753	0.564	0.976

SEM: standard error of the mean; P: p-value. Contrasts: 1 = C vs BSF5; 2 = C vs BSF10; 3 = BSF5 vs BSF10; 4 = C vs BSF diets

#### 44. Effect of changing water to diet ratio over time on nursery pigs' performance fed wet/dry diets

Ramon Muns

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**Application:** Gradually decreasing water to feed ratio after weaning in a Wet/Dry system increased body weight, average daily feed intake (ADFI), average daily gain (ADG), and feed conversion ratio (FCR) of pigs from 4 to 10 weeks of age compared to dry feeding.

**Introduction:** While liquid systems mix water and feed on a tank before delivering it to the pig, by keeping water and feed separate up to the point of delivery Wet/Dry system might prevent formation of biofilms and degradation of aminoacids. When compared to dry feeding, pigs fed a Wet/Dry ratio of 2L of water for 1 kg of diet (2:1) from 4 to 6 weeks and a 3:1 ratio from 6 to 10 weeks of age, increased ADG and ADFI, and had similar FCR [1]. The present work aimed to study whether starting with a high dilution at weaning and gradually decreasing it would help piglets cope with weaning process and improve growth performance.

**Materials and methods:** Trial was performed under Animals (scientific procedures) act 1986. At weaning, 1,011 piglets (Duroc × (LD × LW)) were distributed to one of the following treatments, balancing for body weight and gender: Dry) pigs were offered dry feed; Wet 3:1) pigs were offered a Wet/Dry diet at a 3:1 ratio; Wet 3:1/1:1) pigs were offered a Wet/Dry diet at a 3:1 ratio for 1 week, after which they were

offered a 1:1 ratio for 5 weeks; Wet 3:1/2:1/1:1) pigs were offered Wet/Dry at 3:1 then 2:1 then 1:1 ratios for 2 weeks each, respectively. Pigs were weaned at 28 days of age to a weaner accommodation and allocated in pens of 29 animals. Pigs were offered 3 kg/pig of Starter diet 1 (16.5 MJ DE/kg, 22.5% CP), followed by 6 kg/pig of Starter diet 2 (16.3 MJ DE/kg, 21.5% CP) and then a Grower diet (14 MJ DE/kg, 18.6% CP) until the end of the trial (10 weeks of age). All pigs received the same diets only differing in the proportion of water. Pigs were weighed at weaning, 6 and 10 weeks of age. ADFI, ADG, and FCR were recorded. Data were analysed by ANOVA with pen as experimental unit employing GenStat 16th edition (Table 1).

**Results:** Dry Pigs had reduced body weight, ADG, ADFI and FCR throughout the trial compared to Wet/Dry treatments. Wet/Dry treatments did not differ among them.

**Conclusions:** Feeding nursery pigs a Wet/Dry diet gradually reducing the water content from 3:1 to a 1:1 ratio improved body weight, growth and intake compared to dry fed pigs. However, Dry pigs showed better feed efficiency. Future work on the use of co-products in Wet/Dry would be of interest.

**Acknowledgments:** Research funded by DAERA and PigRegen Ltd.

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Table 1  
Effect of treatment on pigs' body weight, average daily gain (ADG), average daily feed intake (ADFI) and feed conversion ratio (FCR) during the nursery period.

<i>n</i> <sup>o</sup> of pigs/pens	Dry/control <i>n</i> = 283/9	Wet 3:1 <i>n</i> = 282/9	Wet 3:1/1:1 <i>n</i> = 222/8	Wet 3:1/2:1/1:1 <i>n</i> = 224/8	SEM	p-value
Wean weight	9.23	9.21	9.21	9.22	0.13	n.s.
6 weeks weight	12.7 <sup>a</sup>	13.7 <sup>b</sup>	13.7 <sup>b</sup>	13.7 <sup>b</sup>	0.15	<0.001
10 weeks weight	27.6 <sup>a</sup>	30.5 <sup>b</sup>	30.6 <sup>b</sup>	30.4 <sup>b</sup>	0.15	<0.001
ADG wean to 6 weeks, g	270 <sup>a</sup>	354 <sup>b</sup>	350 <sup>b</sup>	343 <sup>b</sup>	7.9	<0.001
ADG 6 to 10 weeks, g	555 <sup>a</sup>	637 <sup>b</sup>	610 <sup>b</sup>	609 <sup>b</sup>	21.0	0.012
ADG wean to 10 weeks, g	461 <sup>a</sup>	543 <sup>b</sup>	524 <sup>b</sup>	519 <sup>b</sup>	13.0	0.002
ADFI wean to 6 weeks, g	277 <sup>a</sup>	379 <sup>b</sup>	373 <sup>b</sup>	374 <sup>b</sup>	15.3	<0.001
ADFI 6 to 10 weeks, g	906 <sup>a</sup>	1117 <sup>b</sup>	1040 <sup>b</sup>	1053 <sup>b</sup>	35.5	0.029
ADFI wean to 10 weeks, g	698 <sup>a</sup>	871 <sup>b</sup>	817 <sup>b</sup>	827 <sup>b</sup>	21.6	0.006
FCR wean to 6 weeks	1.02 <sup>a</sup>	1.10 <sup>b</sup>	1.07 <sup>b</sup>	1.11 <sup>b</sup>	0.052	0.016
FCR 6 to 10 weeks	1.64 <sup>a</sup>	1.79 <sup>b</sup>	1.75 <sup>b</sup>	1.77 <sup>b</sup>	0.051	0.003
FCR wean to 10 weeks	1.52 <sup>a</sup>	1.63 <sup>b</sup>	1.67 <sup>b</sup>	1.62 <sup>b</sup>	0.088	0.018

<sup>a,b</sup>Values within row with different superscript are statistically different from each other

#### 45. Impact of red beetroot supplemented diets on the gut bacterial composition of weaned pigs

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**Application:** Red beetroot supplemented diet at 20g/kg proffer a richer and more diverse gut microbiota thus healthier compared to diet supplemented with a high level of zinc oxide

**Introduction:** Weaning induces pathogenic colonization of the gut and oxidative damages to the intestinal structure and function which results in gut inflammation, microbiota disruption and diarrhoea. However, dietary modulation of the gut microbiota remains a viable strategy to promote the growth of beneficial microorganisms and thus a healthy gut and pig. Currently, weaned pig diet is supplemented with zinc oxide but with the upcoming ban on its use, exploring other alternatives is needful. Red beetroot is a rich source of plant bioactives with reports of grow promotion, high antioxidant, antimicrobial and anti-inflammatory abilities. But, evidence on modulation of gut microbiota is lacking. On this premise, we assessed the effects of red beetroot supplemented diets on changes in the gut microbiota of weaned pigs.

**Materials and methods:** Digesta samples were collected from the jejunum, ileum and caecum of 4 × 8 weaned pigs (aged 28 days) that were fed either a control diet (CON) or diet with zinc oxide (3000 mg/kg, ZNO) or with two different levels of red beetroot (20 g/kg, RB2; 40 g/kg, RB4) for 14 days. Following DNA extraction from the digesta samples, genomic sequencing of the V4 region of the 16 s rRNA gene was conducted and sequence reads were processed using Mothur. Data was analysed in R (with Phyloseq, Vegan and DeSeq2 statistical packages).

**Results:** Inclusion of red beetroot to weaned pig diet increased the richness and diversity of the pig gut microbiota when measured with the Chao 1 index ( $P < 0.05$ ). Highest diversity was shown in pigs fed RB2 indicating 47 and 21 genera more than those fed the control and ZNO diet respectively. Also, diets RB2 ( $R^2 = 0.080$ ,  $P = 0.006$ ) and ZNO ( $R^2 = 0.063$ ,  $P = 0.042$ ) significantly influenced the bacterial composition of the caecum ( $R^2 = 0.177$ ,  $P = 0.013$ ). However, pigs fed diet supplemented with 4% red beetroot showed a significant increase in the Phylum Proteobacteria ( $P < 0.05$ ), with a high

Table1

Differential analysis of operational taxonomic units (OTUs) with increased genus abundance in the caecum of weaned pigs relative to pigs fed control diet. (\*P values adjusted by Benjamin-Hochberg correction).

OTU number	Genus	Fold change (Log <sub>2</sub> )	*P value
<b>Increased with pharmacological level of zinc oxide (ZNO diet)</b>			
OTU00123	<i>Candidatus soleaferrea</i>	11.10	4.3 <sup>E-02</sup>
OTU00279	<i>Clostridiaceae unclassified</i>	9.09	1.8 <sup>E-03</sup>
OTU00056	<i>Erysipelotrichaceae unclassified</i>	8.66	9.94 <sup>E-07</sup>
OTU00234	<i>Blautia</i>	7.44	3.1 <sup>E-02</sup>
OTU00230	<i>Terrisporobacter</i>	7.20	1.1 <sup>E-02</sup>
<b>Increased with addition of 20 g/kg red beetroot (RB2 diet)</b>			
OTU00040	<i>Anaerovibrio</i>	11.29	1.03 <sup>E-03</sup>
OTU00032	<i>Selenomonadaceae unclassified</i>	10.82	1.3 <sup>E-04</sup>
OTU 00,089	<i>Allisonella</i>	7.56	2.9 <sup>E-02</sup>
OTU 00,132	<i>Intestinibacter</i>	5.82	3.36 <sup>E-11</sup>
<b>Increased with addition of 40 g/kg red beetroot (RB4 diet)</b>			
OTU00040	<i>Anaerovibrio</i>	10.33	1.23 <sup>E-08</sup>
OTU00089	<i>Allisonella</i>	9.90	1.95 <sup>E-06</sup>
OTU00184	<i>Campylobacter</i>	7.83	1.6 <sup>E-04</sup>
OTU00102	<i>Enterobacteriaceae unclassified</i>	6.77	9.48 <sup>E-05</sup>
OTU00127	<i>Pasturellaceae unclassified</i>	6.56	1.6 <sup>E-02</sup>
OTU00132	<i>Intestinibacter</i>	5.67	3.2 <sup>E-02</sup>

abundance of the families Enterobacteriaceae, Pasturellaceae and Campylobacteraceae (Table 1). Whilst they are natural commensals of the gut, some genera from these families are pathogenic. There were no differences in the bacterial composition of the jejunum and ileum ( $R^2 = 0.041$ ,  $P = 0.051$ ).

**Conclusion:** RB2 diet increased the richness and diversity of the gut microbiota, however inclusion levels above 20 g/kg as observed in RB4 diet should be avoided due to emergence of pathogenic genera.

**Acknowledgments:** We appreciate the funds provided by the Commonwealth Scholarship Commission.

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#### 46. Maternal heat stress during gestation results in differentially expressed genes associated with transporter activity and metabolism in pig placenta

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**Application:** Findings of this study provide novel insights into the molecular regulation of pig placenta in response to maternal heat stress during gestation. Ultimately, this may improve our understanding in reducing summer infertility in sows.

**Introduction:** The increasing prevalence of extreme heatwaves negatively affects farm animal production. Accumulating evidence shows that maternal heat stress during gestation compromises sow fertility and alters offspring phenotype. Placental insufficiency is a common consequence of maternal heat stress, possibly due to reduced uterine blood flow during animal heat adaptation. In a previous study, we have shown that maternal heat exposure during early-mid gestation (d40 to 60) reduced placental efficiency (fetal/placental weights), altered placental morphology and fetal phenotype, especially the skeletal muscle [1]. However, little is known about molecular regulation underlying differences in placental efficiency induced by maternal thermal stress. In this study, we hypothesised that exposing pregnant pigs to controlled heat stress during early-mid gestation would alter global expression of genes associated with placental efficiency and function.

**Materials and methods:** Ten placental tissues were collected on d60 of gestation from gilts (Large White × Landrace) exposed to thermoneutral control (constant 20 °C; n = 5) or heat stress (cyclic 28 to 33 °C; n = 5) conditions between d40 and d60 of gestation. Placental efficiency (fetal/placental weights) was calculated and compared using Student's t-test. Placental total RNA was extracted followed by RNA sequencing (RNA-seq). Differential expression analysis was performed using DESeq2 to identify gene expression differences in placenta between the thermoneutral and heat stress groups. Genes with a false discovery rate (FDR) ≤ 0.1 were considered differentially expressed. Function enrichment analysis of the differentially expressed genes was performed in PANTHER database.

**Results:** Maternal heat stress decreased placental efficiency (P = 0.023). Differential expression analysis identified 169 differentially expressed genes (FDR ≤ 0.1) in placenta between the thermoneutral control and heat stress groups (Fig. 1). Compared to the thermoneutral group, 134 genes were downregulated whereas 35 genes were upregulated by maternal heat stress. Gene ontology enrichment analysis revealed that the downregulated genes were associated with ion transmembrane

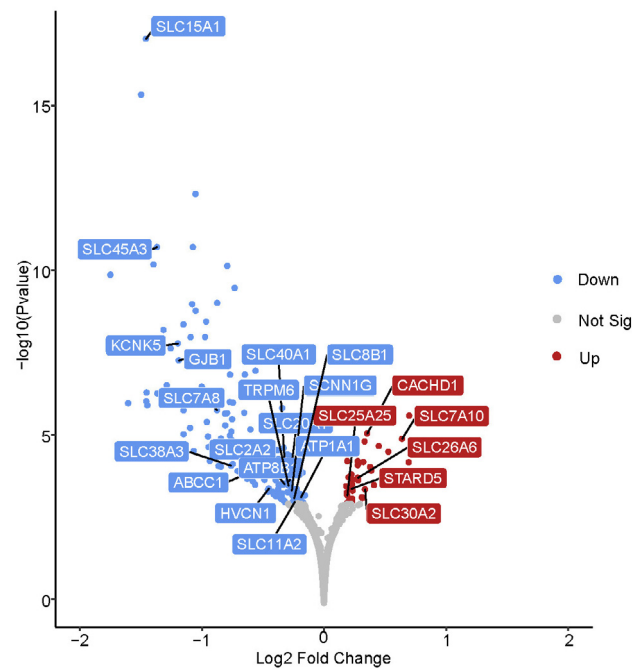


Fig. 1. Volcano plot of placental differentially expressed genes (heat stress compared to thermoneutral control). Genes associated with placental transport activity are labelled.

transporter activity (FDR = 0.042). Pathway enrichment analysis identified that SLC-mediated transmembrane transport (FDR = 0.008) and glycosaminoglycan biosynthesis (FDR = 0.027), which modulates placental stroma synthesis, were enriched by the downregulated genes. Neither gene ontology nor pathway terms were significantly enriched by the upregulated genes.

**Conclusion:** Current RNA-seq data show that maternal heat stress during early-mid gestation altered placental transcriptome profiles, especially for those genes associated with placental transport and metabolism. These findings suggest the underlying molecular mechanisms of placental insufficiency due to maternal thermal stress.

**Acknowledgments:** The authors acknowledge Australian Pork Limited and University of Melbourne for funding this work.

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#### 47. Effect of environmental enrichment and group size on the water use and waste in grower-finisher pigs

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**Application:** Pig production contributes 19% to the global waterfootprint of farm animal production. Global freshwater supplies are limited, thus, water-use in pig production chain should be optimized.

**Introduction:** Drinking water accounts for 80–87% of on-farm water use, grower-finishers account for 64% of total on-farm herd water use (Matlock, 2014). This water is partly consumed by pigs and partly wasted as pigs often manipulate drinkers without drinking. Drinking water usage and wastage is affected by different factors. Evidence suggests pigs in larger groups drink more, we hypothesise that provision of a high level of enrichment will reduce non-drinking manipulation of drinkers. This study investigated how group sizes and levels of enrichment affect water usage (ingested + wasted), water wastage and behaviour in grower-finisher pigs.

**Fig. 1.**

**Materials and Methods:** Pigs (n = 672) were assigned from 11 weeks of age ( $77 \pm 2$  days) until slaughter. The study had  $2 \times 3$  factorial design; effect of group size (SMALL (12 pigs), MEDIUM (24 pigs), and LARGE (48 pigs)) was assessed across two levels of enrichment (HIGH, LOW). Pigs on LOW enrichment received one wooden post and one hanging rubber toy/12 pigs. Pigs on HIGH enrichment received same, with the addition of one rack/12 pigs containing fresh grass. The experimental room contained 4 pens for each group size. Water meters were installed in all pens, covering both wet-dry feeder and drinker next to it. Water use was monitored automatically. Once per week, the overflow (waste) from each drinker was collected and measured. The number and duration of drinking bouts at each drinker, and total drinker occupancy, was extracted from 1hr video recording (day 25). To calculate water use/pig, 24hours water use/pen was divided by number of pigs present on each day. Data were analysed using linear mixed models (PROC MIXED) in SASv9.4.

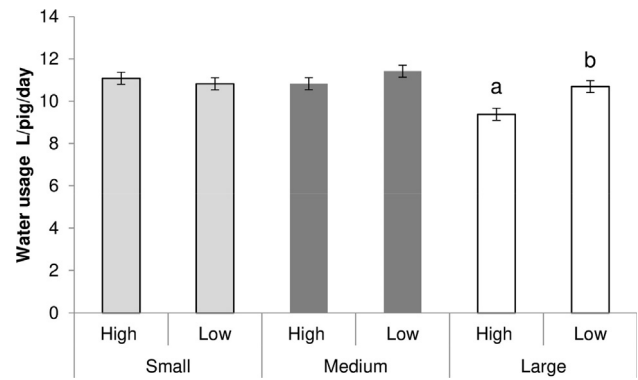


Fig. 1. Effect of group size and enrichment on water usage (ingested + wasted)/pig/day (LSmeans  $\pm$  SE). Group size (pigs/pen): Large (48), Medium (24), Small (12), Enrichment: Low (Wood + Hangingtoy), High (Wood + Hangingtoy + Grass). <sup>abc</sup>Different letters denote significant differences picked up by Tukey test ( $p < 0.05$ ).

**Results:** There was no effect of group size on water use or wastage. Pigs with HIGH enrichment ( $10.4 \pm 0.4$  L/pig/day) used less water than LOW enrichment ( $11.0 \pm 0.4$  L/pig/day), because of more wastewater ( $P < 0.05$ ). Group size didn't affect drinking bout number, but LOW enrichment had a greater number of drinking bouts (24.4 bouts/drinker/hr) than HIGH enrichment (15.5 bouts/drinker/hr,  $P = 0.037$ ). However, drinking bout duration tended to get shorter as pen size increased ( $P = 0.084$ ) while enrichment didn't affect bout duration. The total occupancy/hr was not affected by group size, but LOW enrichment pigs spent more time occupying the drinker than HIGH enrichment pigs ( $P < 0.05$ ).

**Conclusion:** Use of fresh grass (80% moisture) reduced water usage and wastage. Thus, this type of HIGH enrichment allowance may have benefits for both the environment and animal welfare.

**Acknowledgments:** Project funded by Teagasc Walsh Fellowship (2017147).

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#### 48. Temporal trends in stallion semen quality: The development of the horse as a bio-monitor species

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**Application:** Elucidating trends in stallion semen quality could advance reproductive approaches to mitigate economic impacts of perturbed fertility. Through application to livestock species, knowledge of reproductive health will be furthered, developing the equine model as a bio-monitor for terrestrial and agroecosystem environmental health.

**Introduction:** The equine sector is a lucrative global industry, with breeding forming a foundational component. Despite the impact of poor semen quality on a breeding stallions' economic value, the industry promotes sire selection based on performance and conformation, with little consideration given to fertility [1]. Current research represents fertility statuses in the late 1900's, with limited understanding of semen quality trends in the last two decades. In Breton Draught and Anglo-Arab Thoroughbred stallions, semen volume declined 2% annually between 1981–1996 and 1985–1995 respectively [2]. Trends are paralleled in comparable herbivorous species; dairy bulls display a decline in daily ejaculate volume, sperm concentration, and sperm output between 1970–1985 [3].

Presented here are results from a novel inquiry evaluating current temporal trends in stallion semen quality. Study aims: (1) determine temporal trends in stallion semen quality in a population of breeding stallions, (2) develop the equine model as a bio-monitor of terrestrial and agroecosystem environmental health.

**Materials and methods:** Retrospective data on 11,387 ejaculates; from 1,036 stallions of mixed ages and breeds were obtained from a U.K. breeding facility (2001–2020). Semen analysis methods included

microscopy (X100 lens) and were consistent throughout the study period. Preliminary analyses to determine temporal trends in raw motility (%), volume (ml) and density ( $\times 10^6/\text{ml}$ ) were obtained using a simple linear regression model (IBM SPSS Statistics 26). Each parameter represented an isolated variable in the model. Trends were assessed through the regression coefficient ( $B$ ).

**Results:** A significant decline of  $\approx 10\%$  ( $B = -0.663$  ( $p < 0.001$ )) was reported for raw motility. Over the same study period, semen volume remained consistent ( $B = -0.003$  ( $p < 0.001$ )) and, sperm density increased marginally ( $B = -0.003$  ( $p < 0.05$ )).

**Conclusions:** This novel approach presents invaluable insights into (1) the current status of stallion semen quality and (2) furthers the development of the equine model for broader application to livestock production. Future statistical analysis will determine the influence of potential effect modifiers to aid understanding of aetiological factors underlying temporal trends.

**Acknowledgments:** Special thanks to Tullis Matson and Pamela Humphreys, Stallion AI Ltd<sup>1</sup> for collaborative support.

<sup>1</sup> Stallion AI Services Ltd, Whitchurch, Shropshire, SY13 4BP

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#### 49. Ovarian activity and secretion of somatotrophic hormones in periparturient horses

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**Application:** The results of this study provide insights into causes for a delay of first postpartum ovulation in horses

**Introduction:** In horses, resumption of ovarian function after parturition usually occurs early and the majority of mares (90%) ovulate within two weeks after parturition. This allows a species with a mean gestation length of 340 days to produce offspring at one year intervals. Previous studies suggested that the early postpartum increase in the peripheral concentration of prolactin and somatotrophic hormones is a major stimulus for resumption of ovarian activity and ovulation [1].

**Materials and Methods:** Ovarian follicular development and concentrations of somatotrophic hormones were assessed in mares from 15 days before until 15 days after uneventful parturition. A total of seven healthy pregnant Haflinger mares (mean age  $9.2 \pm 3.8$  years) was included into the study for two consecutive years. All mares were suckled by their healthy foals from parturition onwards. Transrectal ultrasonography of ovarian activity was performed and blood samples for determination of prolactin, growth hormone and insulin like growth factors (IGF) 1 and 2 by ELISA were collected at 2 day-intervals. The relative abundance of mRNA for IGF1 and IGF2 was analysed by qPCR in placental tissue. Statistical analysis was performed by analysis of variance comparing data from mares that ovulated ( $n = 5$ ) and did not ovulate ( $n = 9$ ) within 15 days after ovulation. Values are means  $\pm$  SD.

**Results:** Parturition occurred later in the year in mares ovulating than in mares not ovulating (day of the year  $84 \pm 29$  vs.  $50 \pm 20$ ,  $p < 0.05$ ). No differences between groups with regard to gestation length, placental weight and size as well as foal maturity were detected. Concentrations of prolactin and IGF1 increased from five days before parturition and peaked at day 1 after parturition when plasma prolactin concentration was higher ( $13.0 \pm 6.3$  vs.  $7.9 \pm 4.4$  ng/ml,  $p < 0.05$ ) and IGF1 concentration was lower ( $350 \pm 65$  vs.  $409 \pm 71$  ng/ml,  $p < 0.05$ ) in mares that ovulated than in mares that did not ovulate, respectively. Relative abundance of IGF1 in amniotic tissue was lower ( $p < 0.05$ ) in mares that ovulated than that did not ovulate. On day 15 before parturition, follicles up to a diameter of 2 cm were already present on both ovaries in mares of both groups. Growth of follicles to preovulatory size, however, was only detectable in the postpartum period and was more pronounced in mares that ovulated than that did not ovulate (interaction day  $\times$  group  $p < 0.05$ ).

**Conclusions:** The early occurrence of postpartum ovulation in mares is stimulated by increasing day length. Although ovarian follicular growth is already detectable before foaling, only after parturition and associated changes in hormone secretion follicular growth progresses to preovulatory size. In agreement to the situation in pigs, increased plasma concentrations of IGF1 may contribute to reduced ovarian activity in lactating mares.

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## 50. Effects of blue LED light directed at one eye on equine pregnancy

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**Introduction:** The horse is a seasonally breeding species. Exposing mares to artificial light advances onset of seasonal reproductive activity. Short wavelength blue light is particularly effective at suppressing melatonin secretion. Blue LED light directed at one eye can not only advance the ovulatory season in mares but also shorten gestation length. It was the aim of this study to assess the effects of blue LED light on equine pregnancy outcome, fetal development and foal viability.

**Materials and Methods:** Warmblood mares (n = 20) with singleton pregnancies were randomly assigned to two groups and studied over two years in a cross-over design. In year 1, group 1 received an artificially extended photoperiod using 50 lux of blue light (468 nm) directed at a single eye from 08:00 until 23:00 daily via individual head-worn light masks, while group 2 were exposed only to natural photoperiodic changes. In year 2, the treatment changed. Treatment commenced mid December. Gestation length, colostrum quality, foal size and maturity were determined and compared by Wilcoxon test. Values are means  $\pm$  SEM.

**Results:** Gestation length was shorter (light 333.2  $\pm$  1.0, control 337.9  $\pm$  1.8 days; p < 0.05) and IgG content in colostrum lower (p < 0.05) in

mares equipped with blue light masks compared to control pregnancies. In both groups of mares, colostrum quality was, however, adequate to ensure sufficient IgG transfer to their newborn foals. Neither placental size nor surface differed between treatment and control pregnancies.

Foals born to mares equipped with blue LED light were smaller in height at withers (102.5  $\pm$  1.1 vs. 105.2  $\pm$  1.0 cm; p < 0.01) and elbow to carpus distance (30.6  $\pm$  0.3 vs. 31.8  $\pm$  0.3 cm; p < 0.05) than control foals while there was no difference in body weight and chest circumference. The neutrophil/lymphocyte ratio at birth was higher in foals born to mares equipped with light masks than in foals from control pregnancies (3.2  $\pm$  0.2 vs. 2.7  $\pm$  0.2; p < 0.05), suggesting stimulatory effects of blue light on fetal maturation in horses.

Hair length of foals was reduced when mares had been exposed to light treatment during gestation compared to foals from control pregnancies (13.1  $\pm$  0.8 vs. 20.9  $\pm$  0.1 mm; p < 0.001). This demonstrates that external light effects do reach the fetus. Hair regrowth in a previously clipped area in mares was less pronounced when mare underwent blue light treatment (5.2  $\pm$  0.2 vs. 6.9  $\pm$  0.5 mm; p < 0.05) while hair length outside the clipped area did not differ.

Prolactin concentration in plasma of mares increased steadily over 10 days before parturition, peaked on the day of foaling and decreased rapidly thereafter (p < 0.001) but at no time differed between light treatment and control pregnancies, thus no effects of blue LED light on prolactin release could be demonstrated. The concentrations of glucose (p = 0.001) and insulin (p < 0.001) in plasma of mares increased transiently at foaling, but showed no significant difference between blue light and control pregnancies.

**Conclusions:** Blue LED light directed at one eye advanced the onset of foaling and resulted in the birth of slightly smaller but mature and healthy foals and was thus without negative effects.

### 51. Effects of blue light diodes directed at one eye of pregnant mares on first postpartum ovulation and pregnancy rate

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**Application:** A long-day light program using blue LED light directed at one eye of pregnant mares advances the time of first postpartum ovulation

**Introduction:** Photoperiod is the major environmental factor influencing reproductive seasonality in mares. Increasing day length decreases melatonin secretion and initiates the onset of cyclic ovarian activity in non-pregnant mares. The aim of this study was to assess the effects of a long day light program applied via blue LED light directed at one eye in late gestational mares on the first postpartum ovulation.

**Materials and Methods:** Nineteen late pregnant Warmblood mares were included in a two-year crossover design study. Mares either received a long-day light program via individual head-worn masks or were left untreated and exposed to natural photoperiodic changes as controls. Blue LED light (50 lux; 468 nm) was provided to the right eye from 08:00 to 23:00. In both years, treatment started mid of December,

corresponding to days  $237 \pm 5$  of pregnancy, and ended 20 days after foaling. During the last two weeks of gestation and until 20 days after foaling blood was collected for analysis of insulin-like growth factor 1 (IGF1). An ultrasound examination of the ovaries and uterus was performed on day 3 postpartum and daily from day 6 until first ovulation. During first postpartum oestrus, 12 light-treated and 11 control mares were inseminated and checked for pregnancy 14 days after ovulation. Statistical analyses were undertaken using SPSS. Differences between groups were assessed by Wilcoxon tests. For repeated samples, comparisons were made by ANOVA with a general linear model for repeated measures with time as within subject factor and group as between subject factor. Alpha values  $< 0.05$  were considered significant.

**Results:** Time from parturition to first postpartum ovulation was shorter in mares under additional blue light (mean  $\pm$  SEM, light:  $12.0 \pm 0.5$ , control:  $17.2 \pm 2.3$  days,  $p < 0.01$ ). Fifteen out of 19 mares ovulated within 20 days postpartum. Diameter of the largest follicle increased from days 3 to 10 after foaling ( $p < 0.001$ ). Diameter of the largest follicle did not differ between groups from day 3 to day 9 but was larger in mares receiving additional light on day 10 ( $p < 0.05$ ). Follicle diameter did not differ between groups during the last 4 days before ovulation. The concentration of IGF1 increased during the last days of gestation, peaked at parturition and decreased thereafter ( $p < 0.001$ ) but did not differ in response to blue LED light and control treatments. Pregnancy rate during first postpartum oestrus did not differ and was 83% and 73% when mares were exposed to blue LED light or served as controls, respectively.

## Invited Speaker Summary

### 52. The relation between early luteal function and conceptus development in the pregnant mare

Christine Aurich

University for Veterinary Medicine, Vienna, Austria

**Introduction:** In the pregnant mare, nourishment of the conceptus until approximately day 40 of pregnancy depends on endometrial gland secretions (histotroph) alone. Recently, the importance of an adequate postovulatory progesterone concentration for histotroph production and conceptus development has been investigated in more detail: There is a direct relationship between early post luteal progesterone concentration and the onset of down-regulation of endometrial progesterone receptors with higher progesterone concentrations resulting in earlier receptor downregulation. An experimentally reduced postovulatory progesterone concentration contributed to changes in the amino acid composition of the histotroph as well as delayed conceptus development that was detectable from day 14 after ovulation onwards. On day 34, conceptuses from progesterone-deprived mares were smaller, lighter and development of the auricular hillock, footpad, respiratory tract and heart was

delayed compared to conceptuses from untreated controls. A larger conceptus size on day 14 indicated a higher chance of normal development on day 34. Luteal tissue resumption in mares with experimentally impaired postovulatory luteal function allowed for a slow increase in progesterone towards a physiologic concentration in plasma. This response was, however, not sufficient to counteract a delay in conceptus development. An impaired conceptus development in progesterone-deprived pregnancies is in agreement with the recently published finding that the crown rump-length of equine conceptuses that underwent early pregnancy loss was smaller than from ongoing pregnancies (Kahler et al. Equine Vet J. 2020;00:1–12). Low postovulatory progesterone concentration are thus involved in the pathogenesis of early pregnancy loss in the horse. Treatment of oestrous mares with ovulation-inducing agents or the substitution of progestins during the early postovulatory phase stimulated endometrial function. A systematic assessment of progesterone concentration in plasma of mares early in pregnancy, i.e. on day 5 after ovulation may help to identify mares that might benefit from such a treatment.

### 53. A preliminary investigation of limb coronal movement range in horses and relationship to pelvic symmetry

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**Application:** Evaluation of manual chiropractic adjustment to correct pelvic asymmetry may have implications for assessing and improving horse gait and performance

**Introduction:** Pelvic asymmetry is often used in objective lameness assessment. Gait analysis often focusses on movement in the sagittal plane, but the coronal plane movement (adduction/abduction) may provide further information. This study investigates coronal range of limb movement in sound horses and any relationship with pelvic axial asymmetry and its correction by manual chiropractic adjustment. Previous studies support chiropractic method in improving pelvic axial symmetry.

**Materials and Methods** 13 sound, healthy riding horses from same yard and routine were acclimatised to wearing boots daily for 1 week. On trial day, triplicate tuber coxae(TC) heights (nearest 0.5cm) were measured from the dorsal aspect to level ground stood square, using a plumb line. Difference between means indicated direction and value of asymmetry relative to symmetry of zero. Pegasus triaxial accelerometer/data logger assessed sagittal and coronal limb movement of metatarsal and metacarpal hind and fore limbs at trot, on level ground over 10 strides, before and after receiving pelvic rotation and tilt manual chiropractic adjustments by a fully qualified McTimoney animal practitioner and

relative to palpation rotation and tilt assessment. From GaitSmart analysis, minimum and maximum angles of metatarsus and metacarpus produced coronal range of motion (CRM) degrees for each limb using average stride data. A smaller CRM indicates a straighter limb movement. Data sets were tested for normality. Statistical analyses included paired T-Test, Pearsons Moment Correlation.

**Results:** There was a significant TC height discrepancy reduction after treatment ( $p=0.002$ ). Mean TC height discrepancy was  $1.69 \text{ cm} \pm 0.99$  before and  $0.42 \text{ cm} \pm 0.61$  after treatment.

Before treatment, there were no significant differences between CRM for either left hindlimb ( $12.77 \pm 2.53$ ) and right hindlimb ( $13.83 \pm 3.8$ ), or left forelimb ( $16.17 \pm 6.41$ ) and right forelimb ( $18.36 \pm 8.25$ ) ( $p=0.26$  in both cases).

After treatment, CRM for left hindlimb( $10.82 \pm 2.05$ ) was significantly less than right hind limb ( $12.98 \pm 2.84$ ) and CRM for right forelimb ( $15.40 \pm 7.37$ ) was significantly less than left forelimb ( $22.22 \pm 9.73$ ) ( $p=0.03$  and  $p=0.005$  respectively).

Before treatment there was a significant correlation ( $p=0.03$ ) between pelvic TC height asymmetry and left hind CRM. Following chiropractic adjustment there were no associations between TC height asymmetry and CRM of forelimb or hindlimb ( $p>0.05$ ).

There was no significant difference between saggital ROM of left or right hock, fore and hind cannon before or after treatment ( $p>0.05$ ).

**Conclusions** Correction of pelvic axial asymmetry by chiropractic method has an immediate effect on coronal movement range symmetry of hind and fore limbs that was not associated with saggital ROM. Further study would be required with larger cohorts and effects over time to establish a relationship between pelvic symmetry and coronal movement range.

**Acknowledgments:** There was no external funding for this project

#### 54. Ridden water submersion training – A useful adjunct to current condition training protocols in elite performance horses?

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**Application:** Given that tendon is a slowly healing and poorly regenerative tissue, prompt detection of subclinical damage and timely adaptations to training protocols may have a greater outcome for performance horses than advances in treatment. The use of temperature manipulation during exercise for injury prevention purposes may be of particular clinical benefit to performance horses

**Introduction:** Tendon injuries are the most common musculoskeletal injury reported in equestrian jumping disciplines [2]. In an attempt to reduce tendon injuries, Ridden Water Submersion Training (RWST) involves submerging the horse up to sternum height in water and trotting for set intervals. It is used by trainers to increase cardiovascular fitness, whilst potentially minimising the tendon temperature increase typically reported during traditional condition training sessions on land. The objective of this preliminary study was to assess the effects of RWST on the cardiovascular and musculoskeletal systems of the horse and determine whether RWST limits distal limb temperature increase reported in traditional condition training sessions.

**Materials and methods:** Ethical approval was obtained from the University of Portsmouth Ethics Committee prior to data collection. 15 international event horses, acclimatised to RWST training, were recruited. Polar V800 Equine Heart rate monitors measured heart rate, speed, pace and altitude. Skin temperature of front limbs was measured using FLIR E800 infrared thermal imaging camera, in accordance with published guidelines [1,3].

Statistical analysis was carried out using IBM SPSS (v26). Significance level was set as  $P < 0.05$ . Mean HR/HR<sub>max</sub> values were calculated for

warm-up, main work and cool-down. Three-way mixed ANOVA's with post hoc analyses (Bonferroni adjustment) were conducted to determine statistically significant differences between phases of training, and which between-subject factors significantly affected results. Pearson's Correlation was conducted to determine strength and direction of a linear relationship between WGBT/H2O Temperature and distal limb temperature within each condition.

**Results:** Intervals of RWST produced a total mean HR<sub>max</sub> value of  $65.18 \pm 3.76\%$ ; within parameters for increasing aerobic stamina. Mean HR increased significantly ( $p < 0.01$ ) while mean distal limb temperature decreased significantly ( $p < 0.01$ ) between conditions warm-up, RWST and post work, contrasting with positive correlations previously reported during gallop training on land.

**Conclusion:** In this study, RWST challenged the cardiovascular system adequately to increase aerobic fitness of the horse, whilst limiting the increase of distal limb temperature commonly associated with equivalent forms of high-intensity exercise on land. Therefore, this form of training may be a useful adjunct to current condition training protocols and further investigation is warranted.

**Acknowledgments:** The authors would like to thank Doncker's Stables and staff for participating in the study.

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## 55. The effect of daytime blue-enriched polychromatic light and nighttime red light on peripheral circadian clock gene rhythmicity in horses

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**Application:** By understanding how different lighting impacts circadian (24-h) rhythms in equine tissues, advances can be made in designing stable lighting regimes that support the circadian system and ultimately benefit equine health

**Introduction:** The strength of the photoperiod signal influences the robustness of molecular, cellular and behavioural rhythms in animals [1]. Pineal melatonin production is an important mediator of photoentrainment. Equine studies demonstrate a dose dependent suppression of melatonin by blue light [4] and the facilitation of its normal nightly rise by dim red light [2]. This study assessed internal circadian synchrony by examining clock gene expression patterns in hair follicle cells of horses housed under standard incandescent lighting or a regime consisting of timed blue-enriched polychromatic light and nighttime red light (Fig 1).

**Materials and methods:** Ten healthy Thoroughbred racehorses (aged 3–4 years) were used in a randomised controlled trial at a single training establishment, under the same regularly timed daily feeding and exercise regime. On week 0, baseline samples were collected by plucking 10–20 mane hairs (follicles attached) at 4-h intervals for 24 h. Samples were stored in RNAlater at  $-20^{\circ}\text{C}$ . Horses were blocked for

gender and assigned to treatment ( $n = 5$ ) or control ( $n = 5$ ) lighting groups. The treatment group were provided timed polychromatic white LED light and supplemental blue LED light (450 nm) by day, red light ( $<5$  lux, 625 nm) at night and gradual light transitions at dawn and dusk. The control group had light provided by incandescent bulbs that were used as needed by staff (morning and evening stables and feeding times). On week 20, hair samples were collected at 4-h intervals for 24 h. RNA was isolated and samples interrogated by quantitative PCR for the clock genes; *ARNTL*, *CRY1*, *PER1*, *PER2*, *NR1D2* and *DBP*. 24-h temporal variation of transcript means was evaluated using a Cosinor programme [3] based on the least squares cosine fit method.

**Results:** Cosinor analyses revealed 24-h rhythmicity for *NR1D2*, *PER2* and approached significance for *CRY1* ( $P = 0.013$ ,  $P = 0.013$  and  $P = 0.051$ , respectively) in the treatment group on week 20 only. No rhythmicity was detected on week 0 or on week 20 in control horses.

**Conclusions:** Exposure to a customised LED lighting regime stimulates 24-h clock gene rhythmicity in equine hair follicle cells. These rhythms are absent in racehorses maintained under standard lighting regimes.

**Acknowledgments:** This research was funded by an Irish Research Council Enterprise Partnership Scheme Postgraduate Scholarship award and by Equilume Ltd.

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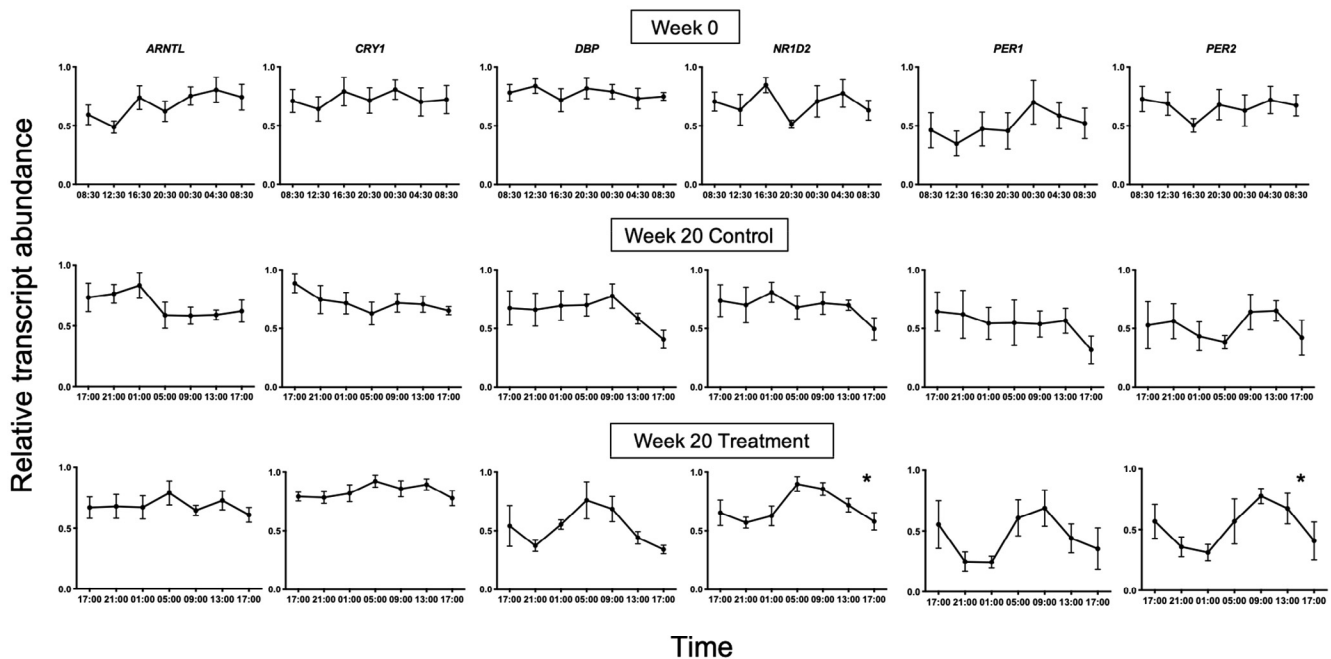


Fig. 1. 24-h profiles of mRNA expression. Plotted are normalised mRNA levels of the candidate genes relative to reference genes *GAPDH* and *PPIA* in equine hair follicles on Week 0 (baseline), Week 20 (Control) and Week 20 (Treatment). Data are presented as means  $\pm$  SEM.



## 56. Genome-wide approaches to dissecting the genetic architecture of equine exertional rhabdomyolysis (ER)

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**Application:** Exertional rhabdomyolysis is a common disease of horses (prevalence 7–10% in racehorses [5]), and presents both a significant welfare concern and huge financial cost to equine sport. Prophylaxis and disease management are challenging, and aetiopathogenesis remains unclear

**Introduction:** Equine exertional rhabdomyolysis (ER) is a syndrome characterised by exercise-induced episodic myofibre necrosis. ER is clinically heterogeneous, with variation in clinical signs and disease severity. Despite ER-susceptibility being heritable ( $h^2 = 0.44-0.49$ ; Norton et al., 2016), causative genes have not been identified. Genome-wide association studies (GWAS) in US [1] and Japanese [4] Thoroughbred horses revealed distinct regions of interest. Studies in other breeds have not been conducted. The aim of this study was to identify disease susceptibility-associated genetic markers in alternative athletic horse breeds, and determine whether ER-susceptibility is a polygenic trait.

**Materials & methods:** 96 Warmblood (WB) horses and Connemara ponies (CP) (cases and controls) were genotyped using a 670 k SNP array, and 38 further WBs and CP were whole-genome sequenced (20X paired-end using Illumina HiSeqX). Case-control GWAS and

regional heritability mapping (RHM) with 10, 20, 40 and 60 SNP windows were conducted across- and within-breeds on the combined set of 127 horses including age, sex and registered studbook as covariates, using a Bonferroni-corrected significance threshold and suggestive threshold per Lander and Kruglyak [2,3].

**Results:** GWAS revealed no disease-associated markers across breeds, but two suggestive-significant markers in CP (ECA8 and ECA16, Fig. 1-B) and one in WB (ECA7, Fig. 1-C). In the across-breed RHM, one genome-wide significant region was identified (ECA11 Fig. 2-A) and 5 suggestive-significant regions (ECA8 and ECA11). The RHM for WB identified 9 suggestive-significant regions (ECA3, ECA11 and ECA12, Fig. 2-B).

**Conclusions:** Several genomic regions associated with ER were successfully identified by GWAS and RHM, suggesting that ER is likely a polygenic disease. Genes linked to muscle function and energy metabolism were located close to these markers making them good candidates for future functional studies aiming to identify causal variants.

**Acknowledgements:** The authors thank the RVC Mellon Fund and the Thoroughbred Breeders' Association for funding this work.

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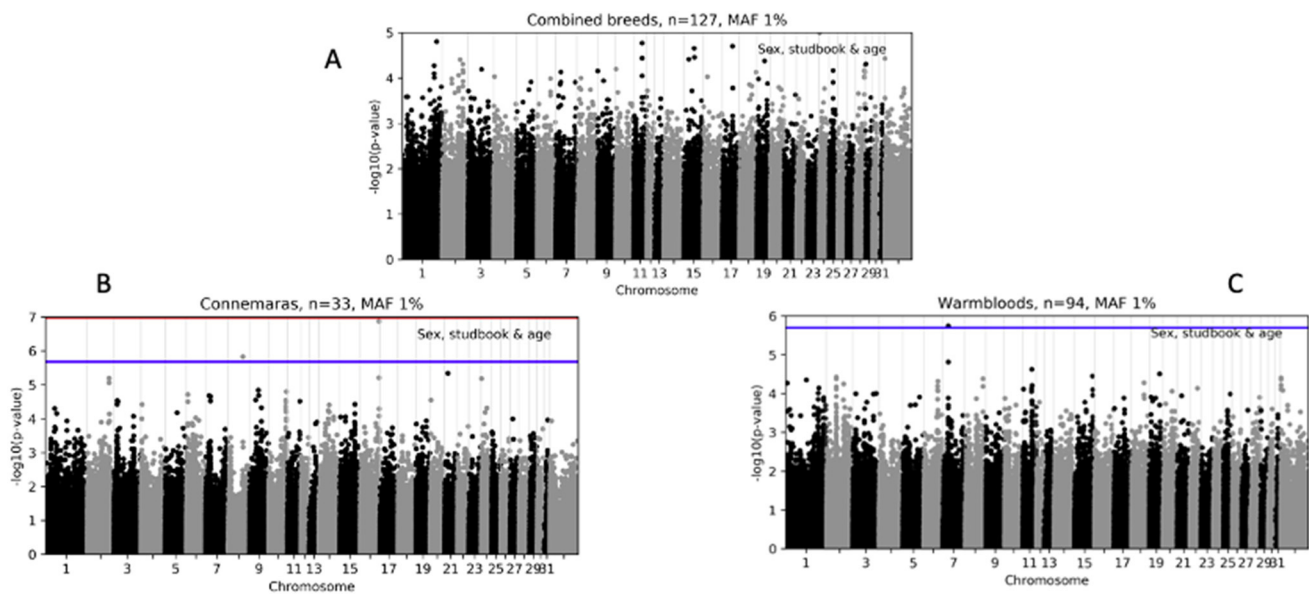


Fig. 1. Manhattan plots of marker location by  $-\log_{10}(P\text{-value})$  of GWAS of A) WB classic cluster versus controls B) CP classic cluster versus controls C) WB non-classic cluster versus controls D) CP non-classic cluster versus controls. The red line indicates the significance threshold (Bonferroni-corrected), and blue line the suggestive threshold (per Lander and Kruglyak (1995)).

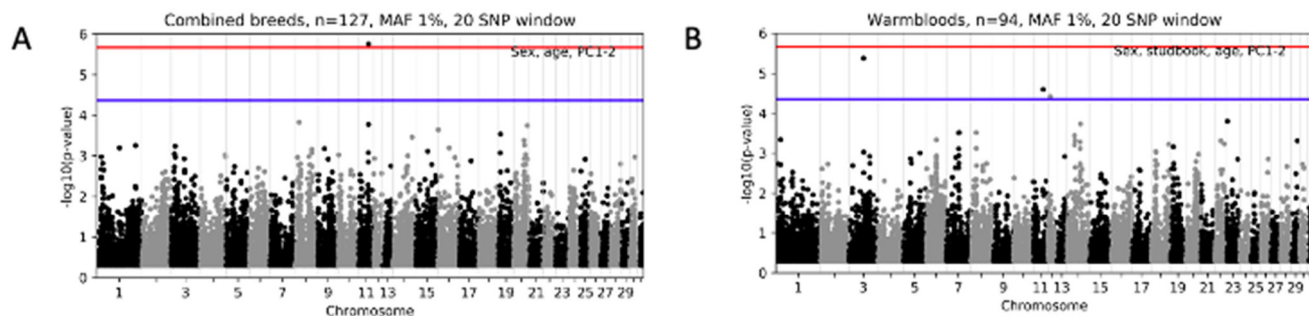


Fig. 2. RHM results of marker location by  $-\log_{10}(P\text{-value})$  A) across breeds and B) in WB. The red line indicates the significance threshold, and blue line the suggestive threshold.

## Invited Speaker Summaries

### 57. Equine parasite control: How to strike that balance?

Martin Nielsen

University of Kentucky, M.H. Gluck Equine Research Center, USA

**Introduction:** This presentation will summarize current recommendations for equine parasite control and describe the scientific evidence behind these. Proper parasite control has become a balancing act between treating too much and too little. On one hand, anthelmintic resistance is ever-increasing in important equine parasites, and more treatments will only accelerate this development. On the other hand, treating too little might lead to reoccurrence of more pathogenic parasitic infections, such as the bloodworm, *Strongylus vulgaris*. Current approaches for parasite control make pronounced demands to the portfolio of diagnostic techniques and their performance. Substantial improvements have been made in developing new diagnostics and validating them in recent years, but several needs have not yet been met. The presentation will describe classic parasite disease complexes, summarize current levels of resistance found in equine parasites, and highlight recent advances in parasite diagnostics. Finally, current unmet needs will be outlined and avenues of future research will be discussed.

### 58. How can livestock systems improve biodiversity?

Mariecia Fraser

Aberystwyth University, United Kingdom

**Introduction:** Focusing on grassland-based production, this presentation will consider opportunities to improve the diversity and abundance of different taxa impacted by management decisions made within livestock systems. It will explore the role of factors such as sward mix, fertilizer inputs, and defoliation pattern when preparing and managing sown pasture. Ways in which differences in the grazing behaviour of different species and breed of grazer can be exploited to deliver beneficial changes in sward composition and related habitat value will also be explored, particularly in relation to native vegetation communities. Opportunities to simultaneously deliver improved biodiversity and increase production system efficiency will be highlighted wherever possible.

### 59. The debatable future of our livestock

Imke de Boer

Wageningen University & Research, the Netherlands

**Introduction:** Livestock have been an integral part of our food systems for thousands of years, and contributed to the formation of societies and cultures. They have contributed, and still do, to food security by providing high-quality macro- and micronutrients, manure and draught power, and by generating income. An increasing body of literature, however, argues for moderating or even eliminating the production and consumption of animal-source food (ASF). Nowadays, ASF is at the heart of fierce debates on healthy and sustainable food systems, as animals impose a significant burden on the environment, consume food edible for humans, cause public health risks, and their keeping and killing raise moral concerns. In order to enhance a transparent societal debate and make informed decisions about the future role of livestock in food systems, we need to better understand the multiple, and often complex roles they play in food systems. Here I address this need by reviewing the multiple benefits and costs of livestock for human health, planetary health, and the health and wellbeing of (farm) animals.

### 60. Funding initiatives for stimulating international collaboration in agricultural research

Margaret Gill

University of Aberdeen, United Kingdom

**Introduction:** There is an increasing awareness of the importance of local context when developing solutions to the Societal Challenges of the 21st century. There is a wide diversity of geographical, economic and environmental ecosystems across the world which impact on Food Systems and livestock production in particular. Hence, since a key research principle when testing hypotheses is to explore extremes, international collaboration can make a major contribution in facilitating comparisons between contexts and hence challenge the concept of “one size fits all” as a solution to sustainable livestock production. The benefits of international collaboration have long been appreciated by researchers but in the 21st century funders have also increased collaboration across national boundaries. The European Commission DG Research has led the way in driving research collaboration through specifying country requirements for partners in the various framework programmes, but BBSRC, the Scottish Government and Defra have a history of holding “Tetrapartite” meetings of agricultural research funders with their French and Canadian equivalents. This talk will outline the history of these mechanisms and others and analyse some of the risks and opportunities they present. For example, elections may prove disruptive in terms of research priorities, but globally agreed goals such as the Sustainable Development Goals (agreed to by >190 nations in 2015) provide a more stable framework. Agricultural research for development is increasingly requiring completed “Theories of Change” and the talk will explore how such constructs can help in framing research with consortia from different countries.

### 61. The importance of international collaboration in research on heat stress of livestock

Frank Dunshea

The University of Melbourne, The University of Leeds, United Kingdom

**Introduction:** Heat stress of livestock is a global issue that is going to become even more important as a result of climate change and resultant more frequent and severe heat events. Development of adaptation and mitigation strategies are reliant upon cross and trans-disciplinary, -institutional and jurisdictional approaches. No single organisation has all the expertise required to develop these strategies across disciplines including, but not limited to, animal and plant physiology, nutrition, immunology, inflammation, engineering, animal health, animal and plant genetics, nano-technology, food science, social science, climate change, economics. International collaboration is one means to put teams together to tackle heat stress and other climate-related problems. For example, a Web of Science search based on Dunshea F\* and heat results in publications across 13 countries including Australia, USA, UK, India, Ghana, France, Thailand, Germany, Ireland, Italy, China, New Zealand and Italy. This research has involved students from additional countries such as Chile, Brazil, Malaysia, Taiwan, Iran, Pakistan, The Netherlands, Sri Lanka, The Philippines, Bolivia, Nigeria, Botswana, Colombia and Mexico. Similarly, prominent heat stress researchers Robert Collier and Lance Baumgard have published papers with collaborators across 21 and 13 countries, respectively. Working across countries and regions allows study of different plant and animal species, cultivars or breeds that may be better adapted to heat stress than local breeds. Also, research across hemispheres can provide research opportunities under prevailing summer conditions year around thereby facilitating more rapid assessment of adaptation and mitigation strategies. In conclusion, international collaboration is essential to tackle most if not all of the global issues including heat stress.

## 62. The Animal Task Force and its role in international collaboration in animal science

Frank O'Mara

Teagasc, Ireland

**Introduction:** The Animal Task Force (ATF) is a European Public-Private Partnership of research, farmer and industry organisations. It plays an important role in international science collaboration by bringing together these organisations from 19 countries to develop and share a common vision and to propose research and innovation priorities to contribute to the environmental and societal challenges. Its goals are to stress the importance of sustainable livestock production for Europe's future; to propose science-based solutions to the major challenge of global food security in the context of a limitation and mitigation of global warming, restoration of biodiversity and efficient resource use; to support knowledge development and innovation by enhancing cooperation and knowledge exchange; and to set the agenda for research and innovation in the animal domain. The ATF vision is for a sustainable, competitive & diverse livestock production sector in EU through innovation & knowledge transfer. It is based around the concept of circular and sustainable agri-food systems in which livestock are an essential component with a key role to facilitate crop diversification and the provision of food and organic fertilizers. The diversity of livestock systems in Europe is recognised and can be considered an asset that allows resilience and a diversity of markets to be supplied. A strategic research and innovation agenda (SRIA) has been developed following consultation with members and with collaboration from a number of European Technology Platforms, European initiatives (EIP Agri, FACCE JPI, SCAR) and international organisations (e.g. FAO, World Organisation for Animal Health, EAAP, Global Research Alliance on Greenhouse Gases). This SRIA provides input to the EU research and innovation agenda, Horizon Europe. This is facilitated by dialogue with key stakeholders, and the ATF also organises events such as workshops, seminars, a session at the annual EAAP meeting, and maintains an informative website.

## 63. Modelling for pandemic preparedness: A need for a one health approach

Lisa Boden

Global Academy of Agriculture and Food Security at the University of Edinburgh, United Kingdom

**Introduction:** The current COVID-19 pandemic has illustrated the role of mathematical models as dominant sources of policy evidence and standard decision-support tools during various stages of disease management at strategic, tactical and operational policy levels. Other types of normative (value-driven) exploratory approaches may better identify and control the influence of longer-term drivers of disease spread, yet may be less highly prioritised at critical stages of decision-making. It has been 20 years since the UK FMD outbreak in 2001, in which the animal health sector learned important lessons about the use of models in outbreak disease emergencies. EPIC, Scottish Government's Centre of Expertise on Animal Disease Outbreaks was created as a result of a wider recognition that the UK needed to improve disease outbreak preparedness. EPIC exemplifies a model of research provision for policy-making that utilizes academic partners, working closely with Government policy-makers, to contribute to evidence-based decision-making for outbreak science.

This presentation will focus, through examples, on the importance of multidisciplinary and multi-sectoral One Health approaches and the need to reflect on lessons learned in animal health if we are to improve long-term contingency planning for the "next" pandemic.

## 64. Change for the better by integration across the disciplines and good stakeholder engagement

Lorna Dawson

James Hutton Institute, SEFARI Gateway, FFCC, United Kingdom

**Introduction:** COVID-19 has been the biggest threat to global human health since the Spanish Flu in 1918. We also face the existential threats of biodiversity loss and climate change, both the focus of international COP Conferences later this year. This One Health session at BSAS2021 is providing us with an opportunity to showcase some of the work being undertaken by SEFARI (the Scottish Environment, Food and Agriculture Institutes) and the Food Farming and Countryside Commission (FFCC) to address these global challenges. For example, SEFARI scientists have stepped up to help the NHS by offering frontline COVID testing, representing the only veterinary node in the UK to do so. SEFARI scientists have also been instrumental in developing a "Genetic Scorecard" as a template to help understand and conserve genetic diversity in some of Scotland's most precious wild species, with international applicability and SEFARI scientists are leading the way in measuring, monitoring and mitigating greenhouse gas emissions from agriculture, including emissions from our iconic ruminant livestock. The FFCC have shown through their Farming for Change: mapping a route to 2030 report, based on the results of IDDRI Ten Years for Agroecology modelling, that, with the right enabling conditions and dietary change, the UK can grow enough healthy food for a future population while reducing synthetic fertilisers and pesticides, increasing the amount of land available for green and ecological infrastructure, releasing some current agricultural area for more flexible use, reducing greenhouse gas (GHG) emissions from agriculture, all without compromising food security or associated environmental impacts. Health and wellbeing of animals and humans are central to this, to allow us to meet net zero carbon targets, while promoting nature, biodiversity, livestock, farming enterprises and food security, but to achieve this we must act in concert, in collaboration, co constructing with key stakeholders.

## 65. The role of veterinary professionals in a changing world

Laura Higham

Vet Sustain, United Kingdom

**Introduction:** The world is facing a multitude of complex and pressing sustainability challenges including climate change, biodiversity depletion and public health pandemics that present existential threats to human civilisation and the natural world. The international scientific community warns that transformative changes are urgently required to limit the catastrophic consequences of climate change and ecological collapse. Zoonotic diseases, such as the virus responsible for the COVID-19 pandemic, account for the majority of new infectious diseases impacting human health, and animal welfare problems persist in domestic animals as a result of entrenched practices in our pursuit of companionship and animal-source foods. These crises demand a robust and urgent response, together with a fundamental and sustained shift in our behaviours at a personal and societal level.

Veterinary professionals occupy an extraordinary niche at the animal-human-environment interface, and deliver a wide range of sustainability services to society. Examples of these services include upholding animal welfare, reducing waste in food chains associated with livestock disease, supporting wildlife health and conservation, and reducing the risk to public health of antimicrobial resistance, through responsible medicine use.

As veterinary professionals and members of the vet-led team, we also hold an opportunity to use our skills as multi-disciplinary scientists and our position of trust to leverage change towards a more sustainable

future, for our own professions, for the sectors we influence and for wider society. A 2019 survey by the Royal College of Veterinary Surgeons placed vets amongst the most trusted professionals by the UK public, and according to the British Veterinary Association, 89% of vets wish to play a more active role in the sustainability agenda. In this presentation, I will outline how veterinary professionals can leverage their skill set, position of trust and intent to act collectively through their work and as citizens to help tackle some of the most critical sustainability concerns we face.

## 66. Farming for 1.5 – An independent inquiry on farming and climate change in Scotland (“What are Scotland doing!?”)

Nigel Miller

Independent Farming for 1.5C and UK Ruminant Health & Welfare Group, Veterinary Surgeon and Livestock Farmer, United Kingdom

**Introduction:** In November, 2020, Farming for 1.5 °C, the independent inquiry on farming and climate change in Scotland, unveiled its report “A Transformation Pathway” on the best way forward for Scottish agriculture to meet the challenge of limiting global warming to 1.5 °C. The report takes a holistic approach - a positive pathway for farms and crofts to reach net zero but also to deliver gains for biodiversity and rural communities.

Everyone and every sector must deliver on Scotland’s climate change ambition to be net zero by 2045, at the latest, and to support nature’s recovery. Farming, uniquely, has the opportunity to not just improve its own performance by reducing emissions from agricultural activity, but to impact positively on wider societal emissions through good soil and land management, by locking up carbon in trees and soil, and by supporting ecosystems. Without the engagement of the agricultural community, with its ability to absorb emissions and not just cut them, it will be impossible for Scotland to deliver against its targets. This engagement must have political and financial recognition.

The Farming for 1.5 °C Inquiry, jointly sponsored by NFU Scotland and Nourish Scotland, is innovative in its make-up of farmers, scientists, activists and environmental NGOs. For the last 18 months they have listened to experts from across the agricultural and climate change arena in order to recommend how Scottish agriculture can become the champions of the fight against climate change. It is not an easy task, nor are implementing the solutions, but it is crucial for the success and resilience of the industry, nature and Scotland as a whole.

## 67. Classification and productivity of grazing ecosystems in China

Fujiang Hou

Key Laboratory of Grassland Livestock Industry Innovation, Ministry of Agriculture, College of Pastoral Agriculture Science and Technology, Lanzhou University, China

**Introduction:** In China, grassland occupies 41.7% of national land area with an average productivity of 18.04 APU/ha (APU, animal product unit, equivalent to 1 kg body weight gain of a medium-fat grazing beef cattle) and annual national productivity of 6.35 billion APU. According to comprehensive sequential classification system of grassland, the main types of grazing ecosystem are classified 9 groups in China. (1) Alpine grazing ecosystem, mainly grazed by reindeer, bison, Yak and Tibetan sheep/goat in the high latitude or high altitude areas. The average productivity is about 15.21 APU/ha at an appropriate stocking rate (ASR) in the Qinghai-Tibetan Plateau. (2) Desert grazing ecosystem in the inland arid area. Dominant livestock include goat and camel with an average productivity of 3.36 APU/ha at an ASR. (3) Semi-desert grazing ecosystem, distributing between desert and typical steppe and mainly grazed by Mongolian sheep, Mongolian goat, camel and beef cattle with an appropriate productivity of 8.66 APU/ha. (4) Subtropical forest and

shrub grazing ecosystem, mainly grazed by goat, beef cattle, sheep, and buffalo. This system covers about 39.2% of shrubland and 29.2% of woodland at regional scale and with a productivity of 28.68 APU/ha at an ASR. (5) Typical steppe grazing ecosystem, mainly grazed by sheep, beef cattle and dairy cow with 15.15 APU/ha at an ASR. (6) Meadow steppe and meadow grazing ecosystem, accounting for 11.3% of total grassland area with a productivity of 20.71 APU/ha at an ASR. (7) Temperate forest and shrub grazing ecosystem, grazed by beef cattle, sheep and dairy cows with 28.88 APU/ha at an ASR. (8) Tropical forest and shrub grazing ecosystem, mainly grazed by goat and buffalo with an appropriate productivity of 23.67 APU/ha. (9) Sown grassland ecosystem, being developed rapidly with rotational grazing and contributed higher productivity than rangeland.

## 68. One world, one approach: Sustaining animal agricultural productivity for a changing world

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**Introduction:** The world is facing common challenges straddling its animal agriculture and human interphase. The human population is increasing, projected to be 9 billion by 2050. With the increase, are changes in patterns of consumption by way of increasing demand of livestock products, largely due to urbanization, increasing incomes, nutritional and environmental concerns and the growing threat of disease transmission to and from agricultural animals. Overconsumption in the developed world and undernutrition in the developing world coupled with the need to reduce impacts of climate change currently shape what research agendas to pursue and what policies to formulate. Livestock being the largest land use sector has a significant role to play in providing solution to these emerging challenges. Demand for livestock products will increase due to the factors stated earlier, coupled with increase in consumption of per capita of meat compared to cereals. Proffering solutions to sustainable feeding of the world depends on how we manage the animal agriculture sector. Local production under current yield trends in many parts of the world, like Sub-Saharan Africa (SSA) and parts of Asia, will have to increase. Animal numbers will have to increase to meet local demands for meat and milk. The challenge of green house gas emission will have to be mitigated by reduced emissions across commodities. Water use will increase, so concerted efforts must be made to ensure conservation of water sources. While food production can be attained under current productivity and demand trends, progress in sustainable environmental and social changes may be slowed.

## 69. Animal welfare in Latin America: How far have we come

Tamara Tadich

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**Introduction:** Animal Welfare has gained increased interest in Latin America, especially during the last 20 years. This is reflected through an increase in scientific activity, but also in changes applied by the livestock sector. The main drivers for the implementation of animal welfare practices in the region have been its close relationship with food security, food safety, product quality and sustainability of the livestock sector. This has implied an increase in animal welfare policy development in each country in order to align local regulations with the OIE standards. Human capacity building has also been an important aim and training of focal points in each country has been conducted together with local training of different stakeholders. Scientific research has also increased significantly, with countries such as Brazil, México, Chile, Argentina and Uruguay being the main contributors. The most studied aspects have been animal welfare during the production process (farm,

transport, slaughter) with an important focus in cattle, poultry and pigs. It is important to highlight that many Latin American countries already count with production systems that comply with different animal welfare assurance schemes increasing their opportunities to export and comply with societal ethical expectations. In terms of veterinary education, since 2013 there is an agreement between Veterinary Medical Schools to include at least one mandatory animal welfare course, this according to the latest data has been accomplished by over 90% of the Faculties surveyed. Currently the Latin American Association for Animal Production (ALPA) gathers twelve local associations. During 2020 ALPA has increased its efforts to promote collaboration among countries, a group on animal welfare, ethology and precision livestock farming has been organized. The group is formed by 75 researchers from different countries and meets every other month to share information and discuss issues that affect the region in order to analyze possible future actions for continued improvement in this subject.

## 70. One world - one animal science: A European (EAAP) and global (WAAP) perspective

Isabel Casasús, Philippe Chemineau

President of the European Federation of Animal Science & President of the World Association for Animal Production, Italy

**Introduction:** The European Federation of Animal Science (EAAP) is an association of members from 35 countries which aims to promote scientific research, cooperation and dissemination of results concerning livestock farming amongst scientific communities, the agri-food industry and civil society. Europe is a hotspot for diversity, also in terms of livestock species and breeds, agri-environmental landscapes and farming systems, which are currently facing multiple challenges posed by global change. Science, innovation and dialogue are doubtlessly the answer to help this wide range of systems cope and thrive sustainably, while taking into account the industry and consumers, environmental and societal demands. EAAP encourages this with a multiple approach, where several Scientific Commissions and Working Groups consider different species and disciplines, regional aspects and emerging topics. A particular emphasis is placed on early career scientists at all levels within EAAP, to facilitate their interaction, training and engagement with the global animal science community. Our activities include annual meetings, occasional workshops and training courses with the industry, and dissemination through scientific journals, all of them in close collaboration with other scientific networks with which we organize joint meetings, exchange speakers and promote science at different levels. This collaboration with sister societies falls under the umbrella of the World Association for Animal Production (WAAP), which congregates 17 societies related to animal science from all over the world. The WAAP promotes cooperation for the advancement of science, technology and

education in animal agriculture at a global level. This interaction is brought about through a periodic World Conference on Animal Production and the dissemination of common activities in newsletters and scientific publications. Both for EAAP and WAAP, the exchange of experience and collaboration amongst scientific societies is not only deemed necessary for their mutual benefit, but also the only way forward in an increasingly globalized world.

## 71. The FEFAC Sustainability Charter 2030

Asbjørn Børsting

European Feed Manufacturers' Federation (FEFAC)

**Introduction:** FEFAC launched in September 2020 a new Sustainability Charter 2030 where we based on many analyzes and discussions covers the main aspects of sustainability in the feed sector. The charter contains 5 chapters each covering an area of our sustainability challenges and opportunities – We call each of them an ambition.

Each ambition matches specific key elements in the EU Green Deal Proposals – and some of the 17 UN Sustainability Development Goals. The 5 ambitions in the FEFAC Sustainability Charter are:

1. To contribute to climate neutral livestock and aquaculture production through feed. According to FAO feed production represents 45 % of the carbon footprint of livestock products globally.
2. Foster sustainable food systems through increased resource and nutrient efficiency. Feed plays an important role here.
3. Promote responsible sourcing practices. FEFAC has in February 2021 launched the next generation of our Soy Sourcing Guidelines (SSG 2021), where there is a strong focus on no- deforestation goals.
4. To contribute to improving farm animal health and welfare through feed.
5. Enhance the socioeconomic environment and the resilience of the livestock sector.

We have decided some of the key industry indicators where we will measure the development in the feed sector concerning the sustainability ambitions. The aim is to do that on both national and aggregated FEFAC- level. The indicators we are starting with are 1. Feed conversion rate, 2. The share of non-edible feed materials – it is a very important goal for the feed sector to make valuable use of co- and byproducts from the food- and bioenergy production, 3. Share of responsible and deforestation free soy, 4. GHG reduction potential and 5 is the share of European protein on total feed protein use.

Looking at these indicators there can – and will be some “everything else equal” considerations and in some cases dilemmas where a little lower score on one indicator could bring positive effects on others. But with explanations this toolbox can really make sense to measure the sustainability development.

## 72. Prediction of CO<sub>2</sub> emissions from dairy cattle using calorimeter chamber data

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<sup>a</sup>Livestock Production Science Branch, Agri-Food Biosciences Institute, Hillsborough, Down, UK

<sup>b</sup>School of Computing, University of Ulster, Jordanstown, Antrim, UK

**Application:** Accurate prediction of CO<sub>2</sub> emissions from dairy cattle in naturally ventilated dairy house is essential for robust quantification of air flow rates through the house and then CH<sub>4</sub>, N<sub>2</sub>O and NH<sub>3</sub> emissions from dairy herds.

**Introduction:** Quantification of air flow rates in naturally ventilation dairy house for estimating gas emissions from dairy cows poses a big challenge. The recent VERA protocol [1] recommends the adoption of the CO<sub>2</sub> balance technique to predict air flow rates for such livestock houses. This method requires information on CO<sub>2</sub> emissions from housed cattle and CO<sub>2</sub> concentrations inside and outside the house. The former is derived from heat production of cows which is calculated from energy metabolism models of energy feeding standards. However, these energy models are developed for the average cows in national dairy herds, with little consideration of individual animal variations. The objective of the present study was to develop prediction equations for CO<sub>2</sub> output from lactating dairy cows using data (e.g., DM intake (DMI), live weight (LW) and milk yield (MY)) derived from calorimeter chamber studies.

**Materials and Methods:** Data (n = 401) used were collated from 17 calorimeter chamber studies with lactating dairy cows undertaken at the Agri-Food and Biosciences Institute between 1993 and 2002, and comprise 365 Holstein-Friesian and 36 Norwegian dairy cattle. Cows were offered *ad libitum* mixed diets of concentrate and forage (292 cows fed grass silage, 52 grass silage/maize silage and 57 grass). Energy intake and outputs including CO<sub>2</sub> emissions were measured using indirect open-circuit respiration calorimeter chambers. The REML procedure was used to develop prediction equations for CO<sub>2</sub> emissions, with the effect of experiment being removed.

**Results:** Cows had a mean LW of 548 kg (s.d. 62.2), ranged from 385 to 733 kg, and the corresponding data for DMI are 16.1 (3.23) from 7.7 to 24.9 kg/d, dietary forage proportion 0.605 (0.2222) from 0.180 to 1.000 kg/DM, MY 22.0 (7.96) from 3.8 to 49.1 kg/d, and CO<sub>2</sub> emissions 5870 (981.1) from 3475 to 8855 (L/d), respectively. The CO<sub>2</sub> emissions are positively and significantly related to DMI, LW and MY (P < 0.001), with a high R<sup>2</sup> value (0.85) with DMI while relatively low R<sup>2</sup> value (0.62 or 0.60) with LW or MY (Table 1 and Fig. 1). These relationships

Table 1

Equations for prediction of CO<sub>2</sub> output.

Equations	R <sup>2</sup>
CO <sub>2</sub> = 237.8 DMI + 2040	0.85
CO <sub>2</sub> = 5.43 LW + 2768	0.62
CO <sub>2</sub> = 54.4 MY + 4712	0.60
CO <sub>2</sub> = 5.53 LW + 55.8 MY + 1574	0.68
CO <sub>2</sub> = 5.47 LW + 47.3 MY - 4.52 ADF + 2892	0.70

Unit = L/d for CO<sub>2</sub>, kg/d for DMI and MY, kg for LW, and g/kg DM for ADF.

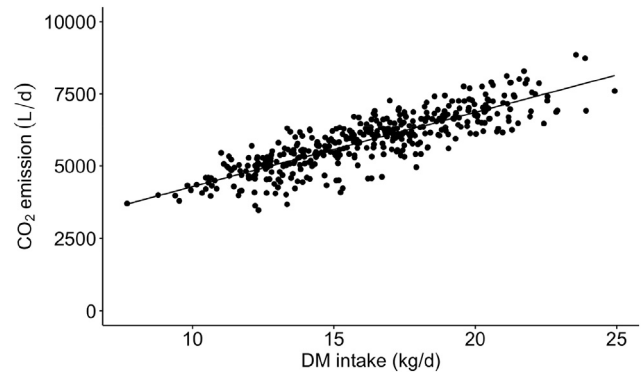


Fig. 1. The relationship between CO<sub>2</sub> output and DM intake in dairy cows.

indicate that increasing 1 kg of DMI, LW or MY could increase CO<sub>2</sub> emissions by 237.8, 5.43 or 54.4 L/d. A combination of LW and MY as predictors increased the R<sup>2</sup> to 0.68, and a further inclusion of dietary ADF concentration raised the R<sup>2</sup> to 0.70.

**Conclusion:** The present study provides novel information for accurate quantification of CO<sub>2</sub> emissions from feed intake for lactating dairy cows. Where feed intake data are unavailable, live weight, milk yield and dietary ADF concentration data can also be used to predict CO<sub>2</sub> emissions for lactating dairy herds.

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### 73. Detection of methane eructation peaks in dairy cows using signal processing

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**Application:** Mobile gas analysers provide a potential method for measuring individual animal enteric methane emissions by detecting eructation peaks.

**Introduction:** Cattle are a notable source of enteric methane emissions. The objective of this study was to investigate using signal processing to detect eructations during milking and compare measurements from two gas analysers that differ in volume of air sampled and speed of processing sampled air.

**Materials and methods:** A total of 5,512 methane spot measurements were obtained from 65 cows over a 3 week period. Methane in air (parts per million) was measured every second from the feed bin of the milking station. The following two gas analysers sampled air at the same time: Guardian SP (T<sub>90</sub> response time < 30 seconds; Edinburgh Instruments Ltd., Livingston, UK) at 0.75 L/min and IRMAX (T90 response time < 4 seconds; Crowcon Detection Instruments Ltd., Abingdon, UK) at 0.75 L/min in week 1, 375 L/min in week 2 and 750 L/min in week 3 (representing a 5 fold increase in air speed each week from 0.5 in week 1 to 5 m/s in week 3). Peak analysis software (Matlab) (version R2018a, The MathWorks, Inc., Natick, USA) was used to extract the maximum gas concentration amplitude (ppm) during each milking. A linear mixed model in Genstat (version 19.1), with fixed effects of analyser × week of

study, lactation number, days since calving and the random effect of cow ID was used for the analysis. Repeatability of methane measurements was assessed by  $\sigma^2_{\text{animal}} / (\sigma^2_{\text{animal}} + \sigma^2_{\text{residual}})$ , where  $\sigma^2$  is the variance. Between-cow and residual coefficients of variation (CV) were calculated from variance components as root mean square error divided by the mean. Significance is attributed at  $P < 0.05$ .

**Results:** The high association ( $R^2 = 0.94$ ) between methane measurements from both analysers. At the same air sampling volume there was no difference in methane concentration between analysers (Table 1). Increasing volume of air sampled by the IRMAX resulted in a higher concentration measured by the IRMAX and a lower concentration measured by the SP ( $P < 0.001$ ). Repeatability of measurements was 0.65 for the SP and 0.53 for the IRMAX. Between-cow CV were 0.22 for the SP and 0.16 for the IRMAX, and residual CV were 0.16 for the SP and 0.15 for the IRMAX.

**Conclusion:** The Guardian SP and IRMAX infrared analysers provided similar measures of amplitude of methane eructation peaks. The Guardian SP provided a more repeatable methane measure and detected more variation among cows compared to the IRMAX [1].

**Acknowledgments:** This PhD studentship and research was funded by the government of Sultanate of Oman.

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Table 1

Effect of analyser and week of study on methane concentrations parts per million (PPM). Means with different superscript letters in the same row differ ( $P < 0.05$ ).

Variable	Mean			F statistic	SED	P
Analyser (PPM)	SP	IRMAX		67.1	9.4	< 0.001
	546 <sup>a</sup>	622 <sup>b</sup>				
Week	1	2	3	1.6	12.2	0.198
	575	594	584			
Analyser x Week (PPM)	SP wk1	SP wk2	SP wk3	IRMAX wk1	IRMAX wk2	IRMAX wk3
	576 <sup>a</sup>	545 <sup>b</sup>	517 <sup>b</sup>	573 <sup>a</sup>	643 <sup>c</sup>	651 <sup>c</sup>
				18.9	16.7	< 0.001

#### 74. Estimating greenhouse and ammonia gas emissions in a natural-ventilated dairy cow house

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<sup>b</sup> AFBI, Belfast, UK

**Application:** The photoacoustic and CO<sub>2</sub> balance method can be used to estimate with high accuracy CO<sub>2</sub>, CH<sub>4</sub>, N<sub>2</sub>O and NH<sub>3</sub> emissions from the natural-ventilated livestock house.

**Introduction:** In the UK and Ireland, ruminant livestock production poses the largest environmental challenge within the agri-food sector. The understanding of NH<sub>3</sub> and greenhouse gas emissions in cattle housing systems is thus essential for the development of management practices to reduce their emissions. This study aims to estimate and analyse NH<sub>3</sub>, CO<sub>2</sub>, CH<sub>4</sub> and N<sub>2</sub>O gases emitted in a natural-ventilated dairy barn using a multichannel photoacoustic and CO<sub>2</sub> balance method.

**Materials and methods:** In summer 2020, gases emissions were monitored during 3 weeks in a natural-ventilated dairy house in AFBI Hillsborough. The barn (70x21 m) has a loose housing system with cubicles and solid-slurry-floor system (3 h scraping intervals). During the measuring period, 36 cows were housed, with live weight 646 ± 48 kg, milk yield 26.4 ± 1.70 kg/d, and DM intake 21.8 ± 0.02 kg/d. Concentrations of NH<sub>3</sub>, CO<sub>2</sub>, CH<sub>4</sub> and N<sub>2</sub>O inside and outside the house were measured using a photoacoustic monitor (INNOVA 1512, LumaSense Technologies) equipped with a multipoint sampler (INNOVA 1409, LumaSense Technologies), allowing 24-sampling locations (18 indoor and 6 outdoor) with a measurement interval of 72 seconds/point, according to VERA's protocol. Indoor environmental conditions were: temperature 15.8 ± 2.32 °C, relative humidity 83 ± 7.9% and wind speed 0.19 ± 0.177 m/s. Carbon dioxide emissions from cows were predicted from measured DM intake [1]. Nitrogen (N) excretion in faeces and urine were estimated from measured N intake. Air flow rates were calculated using the CO<sub>2</sub> balance (VERA, 2018) and used to determine NH<sub>3</sub>, CO<sub>2</sub>, CH<sub>4</sub> and N<sub>2</sub>O emissions.

**Results:** Gases analysed are presented in Table 1. Methane and CO<sub>2</sub> estimation data were within the range of those measured in respiration chambers published elsewhere. Ammonia-N/urine-N estimations showed a small daily variation, but within the range of those used in the UK inventory models for updating annual NH<sub>3</sub> inventories [2]. Estimated N<sub>2</sub>O emissions showed a small daily variation in comparison with published data. Positive weak relationships were observed between temperature and NH<sub>3</sub>-N/urine-N or N<sub>2</sub>O-N/manure-N (Fig. 1).

**Conclusions:** The results showed a high accuracy in CH<sub>4</sub> and CO<sub>2</sub> estimations using the photoacoustic gas monitor and CO<sub>2</sub> balance method, while small variations were observed for NH<sub>3</sub> and N<sub>2</sub>O estimations with respect to reports elsewhere. Increasing temperature increased NH<sub>3</sub>-N/urine-N and N<sub>2</sub>O-N/manure-N. The utilised methodology proved to accurately estimate gas emissions from natural-ventilated livestock houses, however, long-term measurements are required to develop a stronger model.

**Acknowledgements:** The authors acknowledge funding from DAERA NI, AgriSearch NI and Moy Park (17-4-03).

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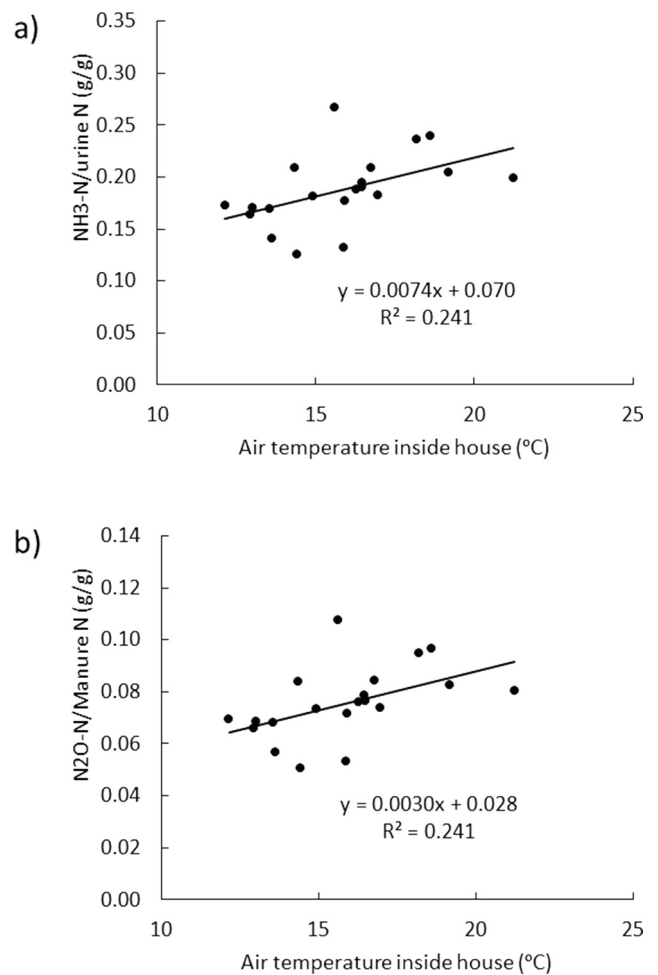


Fig. 1. The relationship between air temperature inside the house (°C) and a) NH<sub>3</sub>-N/urine-N (g/g), and b) N<sub>2</sub>O-N/manure-N (g/g).

Table 1  
 Analysis of gasses emitted in natural-ventilated dairy house<sup>1</sup>.

	Mean	S.D.	Minimum	Maximum
CO <sub>2</sub> (g/cow/d)	14210	528.3	13123	15269
CO <sub>2</sub> /DMI (g/kg)	653	12.7	634	689
CO <sub>2</sub> /ECMY (g/kg)	500	27.7	453	562
CH <sub>4</sub> (g/cow/d)	448	21.8	409	494
CH <sub>4</sub> /DMI (g/kg)	20.6	0.82	19.4	22.8
CH <sub>4</sub> /ECMY (g/kg)	15.8	0.99	14.2	17.6
N <sub>2</sub> O (g/cow/d)	7.1	0.41	6.4	7.7
N <sub>2</sub> O-N/manure N (g/g)	0.08	0.014	0.05	0.11
N <sub>2</sub> O/ECMY (g/kg)	0.25	0.019	0.22	0.28
NH <sub>3</sub> (g/cow/d)	41.6	7.70	28.2	58.9
NH <sub>3</sub> -N/urine N (g/g)	0.19	0.035	0.13	0.27
NH <sub>3</sub> /ECMY (g/kg)	1.47	0.296	0.99	2.15

<sup>1</sup> DMI - DM intake; <sup>2</sup> ECMY - energy corrected milk yield.



## Invited Speaker Summary

### 75. Sustainability in practice – you can't manage what you can't measure!

Stephen Ross

Alltech E-CO<sub>2</sub>

**Introduction:** The European Green Deal has established ambitious targets for the agricultural industry, addressing issues such as nutrient loss and food waste. Furthermore it sets goals to address climate change while promoting environmental, social and economic sustainability. The challenge for agriculture is to achieve these goals whilst still maintaining the upward trajectory of increased production efficiency we've witnessed over recent decades. Setting specific targets drives innovation and improvement, but farmers and the industry as a whole first need to understand the baseline they are starting from, and how practical on-farm management can provide mitigation opportunities in the bigger picture.

Alltech E-CO<sub>2</sub> specialises in conducting on-farm assessments in order to examine the carbon footprint and efficiency of livestock production systems. Working across the entire industry incorporating feed mills, farmers, processors and retailers, we've seen that sustainability is not an option in business today, it's a prerequisite - but if you can't measure it, you can't manage it! Life Cycle Assessment methodology enables us to account for the environmental impacts of processes and products, from raw materials up to leaving the farm gate.

Emissions are a quantifiable indicator of waste, therefore carbon assessment should not be seen as a burden to industry, but an opportunity to reappraise our production efficiency. Furthermore, instead of focussing on just one pollutant, or improving one process 100%, we should ask ourselves can we also improve 100 things by 1%? Identifying a mitigation strategy from this life cycle perspective will not only benefit the farmer, but will resonate over the supply chain, leading to cumulative gains. Our message should be a common one: a more efficient production system is a more sustainable system, and a more profitable farm.

## 76. Methane emission of beef cattle raised in different types of pastures in southwest of england

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**Application:** Explore potential of pasture species to reduce enteric methane emissions from beef cattle as strategy at farm level and for policy.

**Introduction:** Enteric methane (CH<sub>4</sub>) emission (EME) from ruminants has a substantial impact on the UK's greenhouse gas emissions inventory, representing 10% of the total CO<sub>2</sub>-eq emissions[1]. Previous studies have demonstrated that some plant species can be beneficial, due to changes in the efficiency of feed conversion by ruminants resulting in mitigation of CH<sub>4</sub> emissions [3]. Therefore, based on these arguments this study aimed to test the hypothesis that cattle raised on different types of pastures, i.e., fertilized permanent pasture (PP) vs. unfertilised mixed sward with high sugar perennial ryegrass (PRG) and white clover (G-WC), produce different EME.

**Materials and methods:** The two campaigns were carried out in September-October 2019 at the Rothamsted Research North Wyke Farm Platform (UK). Treatments corresponded to two types of pastures grazed by cattle: PP, dominated by PRG (*Lolium perenne* L.) and *Agrostis stolonifera* L. receiving 200 kg N/ha/year (April to August); G-WC, comprised PRG (cv. AberMagic), and white clover (*Trifolium repens* L.) (target soil cover 70:30, respectively). Fifty-eight beef cattle (Stabilizer breed, heifers and steers) with an initial liveweight (LW) 475 ± 52 kg grazed the two pastures. On the first week in September and October, EME was measured in 4 heifers and 4 steers on each treatment (sample size defined with power calculations) for 6 days using the

sulphur hexafluoride (SF<sub>6</sub>) tracer technique [2]. Animals were weighed every 30 days. Statistical analysis (performed with SAS 9.4) included treatment effects (pasture type), period (months) and interaction treatments X period as fixed effects, with sex included as a random effect in the model. All averages are presented as least squares methodology and were separated by the PDIFF option of SAS 9.4 The treatment effect was considered significant at P < 0.05.

**Results:** No significant differences were identified (P > 0.05) between the PP and G-WC treatments regarding the EME, 171.8 vs. 196.9 g/day, nor concerning to the average daily gain (ADG), 230.3 vs. 229.5 g CH<sub>4</sub>/kg ADG, nor the LW, 0.32 vs. 0.36 g CH<sub>4</sub>/kg LW, respectively.

**Conclusion:** The EME did not differ in beef cattle raised on a fertilized permanent pasture or an unfertilised mixed sward with high sugar PRG and white clover relative to animal average daily gain and liveweight.

**Acknowledgments:** Biotechnology and Biological Sciences Research Council (BBSRC) ISP Soil to Nutrition (BBS/E/C/000I0320) at Rothamsted Research.

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## 77. Feed nitrogen utilisation efficiency of mid-lactation Holstein cows fed varying dietary crude protein levels

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**Application** Using appropriate dietary crude protein (CP) levels for dairy cows is an important first step in maximising feed nitrogen (N) utilisation efficiency, and hence reduced N losses to the environment.

**Introduction:** Dairy farming contributes significantly to N losses to the environment in Europe. About 16–36% of N consumed by cows on European dairy farms is retained in milk, with the rest mainly excreted in faeces and urine. As the feed N intake relative to requirements increases, N conversion into milk (feed N utilisation efficiency) reduces, and N excretion (especially urine-N) increases. At a given level of feed intake, feed N utilisation efficiency (consequently, the extent of urinary and faecal-N excretion) is influenced by animal and dietary factors (e.g. dietary ME and N concentrations). This study was designed to evaluate feed N utilisation efficiency of Northern Irish Holstein cows in mid-lactation fed diets with varying CP levels.

**Materials and Methods:** This study was conducted at the Agri-Food and Biosciences Institute, Hillsborough, UK using 24 lactating Holstein cows (17 multiparous and 7 primiparous). The cows were 146–200 days in milk (DIM) averaging 2.4 parity, 645 kg live weight, and 32 kg milk/day. The cows were grouped by parity, DIM, milk yield, and live weight into 6 blocks of 4 cows, and randomly assigned to 4 total mixed rations (grass silage:concentrate ratio, 48:52 DM basis) containing 141, 151, 177 and 210 g CP/kg DM. Two concentrate meals (containing 134

and 252 g CP/kg DM) were used. The cows were *ad libitum*-fed to ensure 10%orts, for 25-day experimental periods. The first 20 days were for adaptation to treatment diets, followed by 5 days for collecting N balance data. Data were analysed using the PROC MIXED procedure of SAS (2003) for repeated measures. The model included block, diet, and diet × day interaction as fixed effects. Pre-treatment milk yield was included as a covariate for the milk N, and N balance data. Linear and quadratic effects of CP level were determined.

**Results:** N intake, and milk N increased in a linear and quadratic manner ( $P < 0.05$ ), while urine N increased linearly, and faecal-N increased quadratically as CP level increased (Table 1). The increase in CP level from 177 to 210 g/kg DM resulted in decreased milk-N, and increased urine-N. N balance was negative (except for the 201 diet) and similar ( $P > 0.05$ ) across CP levels. As a percentage of N intake, urine-N increased linearly ( $P < 0.05$ ), while faecal-N, and milk-N (feed N utilisation efficiency) decreased linearly as CP level increased.

**Conclusion:** Increasing dietary CP levels from 141 to 177 g/kg DM did not significantly affect N output rates in urine and milk. However, further increase in CP level from 177 to 210 g/kg DM resulted in decreased milk-N and increased urine-N excretion, with a linear decrease in feed N utilisation efficiency [1].

**Acknowledgments:** Department of Agriculture, Environment and Rural Affairs.

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Table 1  
Effect of CP level on feed N utilisation efficiency.

	Dietary CP Content, g/kg DM				SEM	P-Value		
	141	151	177	201		Diet	Linear	Quadratic
N intake, g/day	436 <sup>b</sup>	457 <sup>b</sup>	637 <sup>a</sup>	633 <sup>a</sup>	21.1	<0.0001	<0.0001	0.015
N output, g/day								
Urine	146 <sup>c</sup>	156 <sup>c</sup>	243 <sup>b</sup>	287 <sup>a</sup>	13.8	<0.0001	<0.0001	0.652
Faeces	185 <sup>b</sup>	175 <sup>b</sup>	234 <sup>a</sup>	193 <sup>b</sup>	11.8	0.016	0.110	0.023
Milk <sup>1</sup>	127 <sup>b</sup>	138 <sup>ab</sup>	161 <sup>a</sup>	149 <sup>ab</sup>	7.68	0.033	0.022	0.048
N balance, g/day	–24	–15	–5	9	22.9	0.674	0.232	0.992
N output, % N intake								
Urine	33.7 <sup>b</sup>	35.0 <sup>b</sup>	38.7 <sup>ab</sup>	45.6 <sup>a</sup>	2.58	0.008	0.001	0.501
Faeces	42.2 <sup>a</sup>	38.7 <sup>ab</sup>	36.7 <sup>b</sup>	30.2 <sup>c</sup>	1.55	<0.0001	<0.0001	0.551
Milk	29.6 <sup>a</sup>	29.9 <sup>a</sup>	25.5 <sup>ab</sup>	23.3 <sup>b</sup>	1.62	0.021	0.003	0.953

<sup>abc</sup>Least square means in the same row with different superscript letters differ ( $P < 0.05$ ). SEM = standard error of least square means.

<sup>1</sup> Determined as milk protein yield/6.38.

## 78. Predicting enteric methane production from the rumen microbial composition of beef steers fed either a high concentrate or cut grass diet

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**Application:** This research shows the potential of the rumen microbiome as a proxy for enteric methane (CH<sub>4</sub>) yield from cattle fed two contrasting diet types.

**Introduction:** The development, testing and implementation of strategies to reduce or mitigate enteric CH<sub>4</sub> emissions would greatly benefit from the development of reliable, large scale methods to measure emissions from individual animals. Genetic sequencing of the rumen microbiome has received increasing attention as a potential proxy that could fulfil this requirement. The objectives of this study were to assess the differences in the rumen microbial composition between cattle on contrasting diets and to assess the diet specific potential of relative abundances of 16S rRNA gene sequenced microbiota as a proxy for enteric CH<sub>4</sub> production.

**Materials and Methods:** Thirty-six beef steers (18x Limousin cross and 18x Aberdeen Angus cross) were allocated to either a high concentrate total mixed ration (TMR, 136:864 forage:concentrate) or a fresh cut grass diet (ryegrass and clover mown once per day). Cattle were housed one pen per treatment, bedded on sawdust and fed ad libitum. Animals were allocated to six indirect open-circuit respiration chambers over a 6-week period. Concentrations of CH<sub>4</sub> in chamber exhaust air was measured by infra-red absorption spectroscopy and daily feed intake was recorded. Rumen fluid samples were collected after exiting the respiration chambers using a naso-gastric tube. DNA was extracted from the rumen samples and subjected to 16S rRNA gene sequencing. Microbial

genera with different abundance amongst the two diets were identified by a projection to latent structure regression discriminant analysis (PLS-DA). The sign and magnitude of the differences between diets were tested using a Wilcoxon Rank Sum test and corrected p-values by Bonferroni procedures. A linear PLS analysis was used to identify those microbial genera abundances that best explained CH<sub>4</sub> emissions within each diet group.

**Results:** There was a significant difference ( $p < 0.001$ ) in CH<sub>4</sub> emissions between diets, which was independent of breed. A total of 31 microbial genera were identified in the DA-PLS model as discriminating between diets. The differential abundances between selected genera showed a clear separation between diets. The PLS-DA model was based on a single component and displayed a 94.5% discriminating ability and 93.6% after cross-validation. The Wilcoxon Rank Sum test elucidated significant differential abundances between diets in 16 microbial genera ( $p < 0.1$ ), of which 15 were also identified in the PLS-DA, corroborating these results. The PLS analysis for the TMR diet identified a total of 29 microbial genera in a model based on a single component explaining 89.0% of the variation in CH<sub>4</sub> yield and 70.6% after cross-validation. Following the same approach for the Grass diet, 26 microbial genera explained 69.2% of CH<sub>4</sub> yield variation and 42.4% after cross-validation.

**Conclusion:** A strong divergence in rumen microbial genera was observed between two extreme diets. Prediction of CH<sub>4</sub> yield using rumen microbial genera abundances was more accurate for the high concentrate than the grass diet.

**Acknowledgments:** This work was funded by the Scottish Government's Rural Affairs, Food and the Environment Strategic Research Programme.

### 79. Comparison of enteric methane emissions between Holstein heifers and steers using respiration chamber data

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**Application:** Accurate prediction of CH<sub>4</sub> emission from growing cattle can provide useful information for development of robust CH<sub>4</sub> emission inventories and appropriate mitigation strategies for cattle production systems.

**Introduction:** Cattle young stock are significant contributors to total greenhouse gas emissions in cattle production systems, however, there is little information available on CH<sub>4</sub> emission rates in different gender groups. The objective of the present study was to evaluate if there was any difference in enteric CH<sub>4</sub> emissions between Holstein heifers and Holstein steers.

**Materials and Methods:** Data (n = 152) used were collated from 8 respiration chamber studies undertaken at the Agri-Food and Biosciences Institute and the University of Reading between 1997 and 2012. The dataset comprises data from 88 Holstein heifers (live weight (LW) = 143 to 599 kg) and 64 Holstein steers (LW = 145 to 649 kg). The animals were offered either forage alone (n = 24) or a mixture of forage and concentrates (n = 128) at production feeding levels. The forage offered included grass silage (n = 120), maize silage (n = 16) and Lolium perenne-based forage mixtures (n = 16). Energy intake and CH<sub>4</sub> emission were measured using indirect open-circuit respiration calorimeter chambers. The REML procedure was used to evaluate if there was any significant difference between Holstein heifers and steers in animal performance and CH<sub>4</sub> emission rates, with the effect of experiment being removed.

**Results:** There were no significant differences between heifers and steers in LW, feed DM intake (forage, concentrate or total), DM intake/LW, dietary forage proportion (0.774 vs. 0.779 kg/kg DM), DE/GE or ME/GE (0.627 vs. 0.634 MJ/MJ) (Table 1). Daily CH<sub>4</sub> output of heifers tended to be less (P = 0.061) than that of steers, which might be partially due to the uneven distribution of diet/forage type between the heifer and steer groups. However, CH<sub>4</sub> output as a proportion of LW, DM

Table 1  
Comparison of enteric CH<sub>4</sub> emissions between Holstein heifers and steers\*.

	Heifer	Steer	SED	P-value
Live weight (kg)	401	406	8.9	0.564
DM intake/live weight (g/kg)	18.0	17.9	0.40	0.942
CH <sub>4</sub> (g/d)	176	186	5.4	0.061
CH <sub>4</sub> /live weight (g/kg)	0.45	0.46	0.011	0.595
CH <sub>4</sub> /DM intake (g/kg)	24.3	25.1	0.51	0.110
CH <sub>4</sub> /OM intake (g/kg)	26.7	27.6	0.56	0.109
CH <sub>4</sub> -E/GE intake (MJ/MJ)	0.074	0.074	0.0014	0.885
CH <sub>4</sub> -E/DE intake (MJ/MJ)	0.106	0.105	0.0157	0.663

\* CH<sub>4</sub> or CH<sub>4</sub>-E = methane or methane energy output.

Table 2  
The linear regression analysis (with a common intercept) of CH<sub>4</sub> or CH<sub>4</sub>-E against feed or energy intake between Holstein heifers and steers.

	Equation*	R <sup>2</sup>	P-value
Heifer	CH <sub>4</sub> = 20.1 <sub>(1.61)</sub> DMI	+ 29.6 <sub>(21.77)</sub>	0.855
Steer	21.6 <sub>(1.59)</sub> DMI		0.005
Heifer	CH <sub>4</sub> -E = 0.064 <sub>(0.0045)</sub> GEI	+ 1.01 <sub>(1.298)</sub>	0.870
Steer	0.065 <sub>(0.0043)</sub> GEI		0.642
Heifer	CH <sub>4</sub> -E = 0.078 <sub>(0.0062)</sub> DEI	+ 2.25 <sub>(1.508)</sub>	0.848
Steer	0.079 <sub>(0.0059)</sub> DEI		0.641

\* Values in subscript parentheses are SE. CH<sub>4</sub> or CH<sub>4</sub>-E = methane (g/d) or methane energy (MJ/d) output; DMI = DM intake (kg/d); GEI or DEI = GE or DE intake (MJ/d).

intake or OM intake, or CH<sub>4</sub> energy (CH<sub>4</sub>-E) output as a proportion of GE intake or DE intake did not differ significantly between heifers and steers. The linear regression analysis of CH<sub>4</sub> or CH<sub>4</sub>-E against intake of DM, or GE or DE found a similar result (Table 2). With a common intercept, there was no significant difference in the two slopes between heifers and steers, in the relationship between CH<sub>4</sub>-E and GE intake or between CH<sub>4</sub>-E and DE intake. Although steers had a significantly greater slope than heifers in the relationship of CH<sub>4</sub> emission with DM intake, the difference between the two slopes was relatively small.

**Conclusion:** The present result indicates there is little difference in enteric CH<sub>4</sub> emission rates between Holstein heifers and steers when evaluated at similar conditions (LW, feed intake and dietary quality).

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## 80. Effect of breed and ruminal fraction on rumen bacterial and archaeal populations in sheep

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**Application:** Breed and ruminal fraction have effects on microbial abundance and community diversity with implications for breeding programs, rumen sampling and feeding strategies.

**Introduction:** Cattle and small ruminant studies have indicated links between the rumen microbiome, animal performance and methane emissions. Understanding factors that impact on rumen microbial populations is critical for improving animal production and alleviating environmental impacts. While breed has been reported to influence the structure and activity of the microbial populations within the bovine rumen [1], this has not been widely investigated in sheep. Furthermore, distinct microbial profiles have been observed between ruminal fractions [2], potentially introducing bias when sampling the rumen. To investigate the effect of breed and ruminal fractions on the rumen microbiota in sheep, we compared rumen bacterial and archaeal populations across 4 sheep breeds in solid, liquid and epithelial ruminal fractions, and also across fractions in breeds.

**Materials and methods:** In total, 36 lambs across 4 breeds (Cheviot (n = 10), Connemara (n = 6), Lanark (n = 10) and Perth (n = 10)) were used for this study. Lambs were offered a nut based concentrate diet ad-libitum with access to silage throughout the trial period. Rumen samples (n = 90) were collected at slaughter consisting of epithelial (n = 28) liquid (n = 30) and solid (n = 32) samples across all the breeds

ruminal samples and were subjected to 16S rRNA sequencing to investigate rumen bacterial and archaeal communities. Reads were processed using the DADA2 pipeline. Alpha diversity measures (Shannon, inverse Simpson, Faiths PD and observed ASVs) were explored using a two-way ANOVA. Weighted and unweighted UniFrac distances were used to calculate dissimilarity in community composition. Differences between groups was tested using PERMANOVA. Differential abundances was conducted using both the likelihood ratio test (LRT) and the Wald's test from the DESeq2 package (Table 1).

**Results:** Bacterial alpha diversity was affected by breed in the solid and epithelial ruminal fractions (ANOVA, P < 0.05), and affected by fraction in the Lanark and Perth breeds (ANOVA, P < 0.05). The LRT revealed effects of breed on bacterial abundances in both solid and epithelial but not liquid ruminal fractions (LRT, P.adj < 0.05). Similarly, ruminal fraction also exhibited an effect on bacterial abundances in Cheviot, Lanark and Perth breeds (LRT, P.adj < 0.05). Pairwise analysis revealed most abundance changes occurred between Perth and Cheviot in solid and liquid ruminal fractions, and between Perth and Lanark in epithelial ruminal fractions, while most changes between fractions was observed between solid and epithelial.

**Conclusion:** Our study indicates that breed can be an important factor influencing microbial populations within the rumen of sheep. Furthermore, the differences observed in the distribution of bacteria taxa between ruminal fractions, particularly between solid and epithelial fractions, highlights a rumen fraction bias which may have implications for rumen sampling strategies.

**Acknowledgments:** The Authors would like to acknowledge the Teagasc WF Scheme (Ref:2017128)

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Table 1  
Microbial taxa identified as differentially abundant across breeds (LRT).

	Tax. Rank	Cheviot	Connemara	Lanark	Perth	LRT, P.adj	Fraction
Sharpea azabuensis	ASV	28 ± 24	387 ± 769	164 ± 332	409 ± 327	0.01	Solid
F_Lachnospiraceae	ASV	284 ± 337	395 ± 345	144 ± 146	748 ± 1006	0.02	Solid
Sharpea	Genus	31 ± 29	346 ± 680	140 ± 259	391 ± 291	0.046	Solid
Firmicutes	Phylum	10729 ± 3045	6630 ± 4269	10506 ± 7893	16486 ± 9445	0.01	Epithelial
Family XIII	Family	479 ± 223	716 ± 272	1671 ± 1058	517 ± 184	0.04	Epithelial
F_Family XIII	ASV	20 ± 25	28 ± 11	156 ± 132	18 ± 24	0.01	Epithelial

### 81. Compare two methods to analyse total nitrogen from freeze-dried dairy cattle manure

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**Application:** Total nitrogen concentration (TN%) measured from mass spectrometer (two detectors) can be used to replace LECO nitrogen (N) analysis for freeze-dried dairy manure samples.

**Introduction:** The TN% from manure is an important parameter concerning cattle production sustainability (nitrogen utilization and pollution). Previous work studied nitrogen isotopes and TN% from the same manure sample used mass spectrometer (MS) and LECO N analysis, respectively [1,2]. This is partly due to MS uses a very small sample (i.e., ug) for analysis and it may not produce a reliable TN% results when the sample is low in homogeneity. Therefore, the objective of this study was to investigate the use of freeze-dried and ground dairy manure samples to perform TN% analysis on micro-scale MS/TCD vs macro-scale LECO N.

**Materials and methods:** Total of 128 manure samples used for analysis were collected at Dookie campus dairy farm, The University of Melbourne, Australia. They were subjected to an *in vitro* incubation trial to investigate ammonia emission rate over 15 days period. Manure was sampled daily and freeze-dried at  $-20^{\circ}\text{C}$ , and ball milled by Qiagen

Tissue Lyser II, 25 mL hardened stainless steel capsules. The manure samples were sub divided into two samples. The first sample (3 mg) analysis was performed on a Thermo Flash 2000HT elemental analyzer paired to a Thermo Delta V Advantage MS. The second one (300 mg) was executed on a LECO Trumac CN at a furnace temperature of  $1250^{\circ}\text{C}$ . All data were analyzed by a linear regression analysis and Lin's concordance correlation coefficient (CCC) analysis in Genstat.

**Results:** There was a wide range of TN% detected in this study. The TN% performed on elemental + MS was highly correlated with the TN% from LECO N analysis (Fig. 1,  $n = 256$ ,  $R^2 = 0.983$ ,  $P = 0.000$ ,  $\text{CCC} = 0.967$ ).

**Conclusion:** Under the current condition, there was a strong and significant correlation of TN% analyzed by the elemental + MS and LECO N methods, for the dried and ground dairy manure samples. Elemental + MS method with less sample size requirement, can be used to replace LECO N for TN% analysis of dried and ground dairy cattle manure samples.

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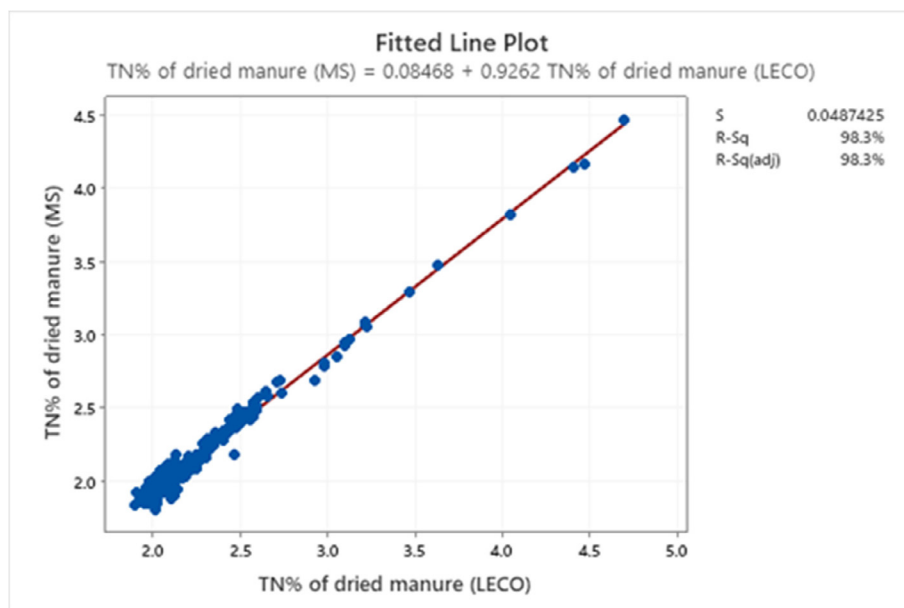


Fig. 1. The regression analysis of TN% from elemental analyzer paired to mass spectrometer (Y axis) and LECO Trumac CN (X axis) methods.

## 82. Effect of feeding Mesquite (*Prosopis* spp.) pods on productive performance and methane emissions in small ruminants: a systematic review and meta-analysis

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**Application:** Mesquite pods are an alternative feed to provide energy and protein to small ruminants in arid regions.

**Introduction:** Small ruminant producers in arid regions use mesquite pods to provide their herds with a low-cost source of energy and protein and improve production parameters. This legume is a good source of protein, fiber, and sugars, which makes it a good ingredient for nutrition [2]. Furthermore, mesquite pods are rich in secondary bioactive substances such as alkaloids, flavonoids, terpenes and phenolic compounds [1]. Results of studies that assess effects of mesquite pods inclusion on ruminant rations has been inconsistent. The aim of this study was to carry out an analytical review to quantify the effect of the inclusion of mesquite pods on productive performance and methane (CH<sub>4</sub>) emissions of small ruminants.

**Materials and methods:** A compressive and structured search of articles was performed using the following electronic databases: Google Scholar, Primo (UAEH), PubMed, Science Direct and ISI Web of Knowledge. The final database includes 37 studies with the following response variables: dry matter intake (DMI), average daily gain (ADG), feed conversion rate (FCR), digestibility (g/100 g DM) and CH<sub>4</sub>. To explore heterogeneity, a meta-regression analysis was performed, including the following *a priori* selected explanatory variables: dosage, species (goat and sheep), breed, body weight and processing of mesquite pods (ground, chopped, extract and meal). All analyses were performed in the R environment for statistical computing using the 'meta' package [3].

**Results:** Inclusion of mesquite pods in small ruminant rations had a positive effect on ADG (+9.94 g/d), digestibility (1.45 g/100 g DM), FCR (-21.2 g/g) and DMI (+68.57 g/d) (Fig. 1). The heterogeneity was substantial to all variables (>80.4), however, this was reduced when the explanatory variables were added to mixed-effect model. An increase of CH<sub>4</sub> emission was observed to mesquite group (+0.57 g/d), likely associated with better DMI and digestibility of supplemented animals. There was a positive relationship between level of incorporation of mesquite pods and DMI, ADG and CH<sub>4</sub> response. The largest effect size to DMI in response to mesquite supplementation was observed to meal processing following by ground and extract (Fig. 2).

**Conclusion:** Supplementation with mesquite pods has a positive effect on productive performance in small ruminants, but its response varies according to the level of incorporation and processing of mesquite pods.

**Acknowledgments:** Authors are grateful to the Mexican Ministry of Education (SEP) for the funding of this research Project under grant UAEH-PTC-823.

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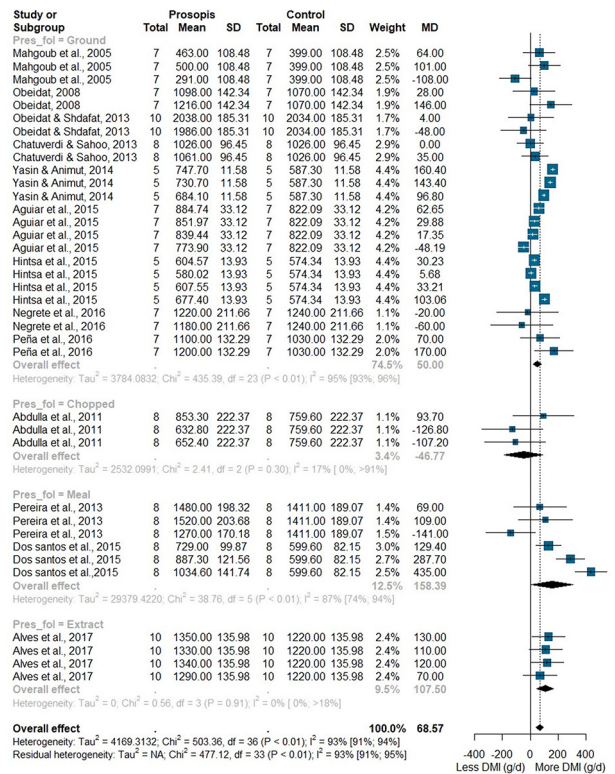


Fig. 1. Forest plot of DMI for studies focused in *Prosopis* spp. supplementation of small ruminants.

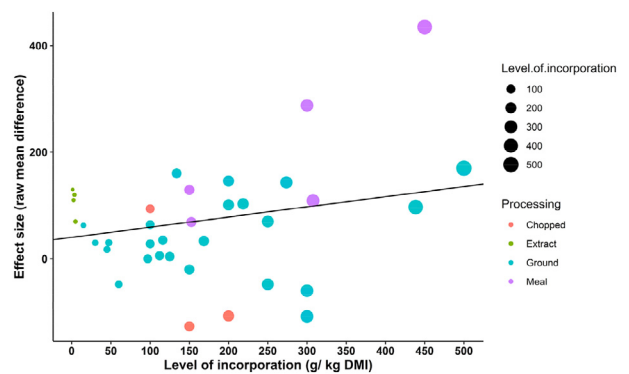


Fig. 2. Bubble plot of raw mean difference to DMI by the level of supplementation and processing of mesquite pods. Each point represents a study.

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### 83. Effect of Nannochloropsis supplementation on ruminal bacterial diversity and on biohydrogenation indicators in lambs

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**Application:** Microalgae are a promising food resource in ruminant production. Because it is a novel food resource, the impact in the ruminal microbiome must be explored.

**Introduction:** Nannochloropsis is a 20:5n-3 rich microalga. Dietary 20:5n-3 and 22:6n-3 affect 18:2n-6 and 18:3n-3 biohydrogenation in the rumen [3], reducing their extension or completeness. It is still unclear which microorganisms determine *trans*-10 shift occurrence in the rumen, but some have been correlated with *trans*-10 18:1 [2]. Our goal was to investigate the effect of dietary Nannochloropsis supplementations on the family abundances of lamb ruminal bacterial community, the ruminal fatty acid profile, and the relationship between bacterial family abundances and biohydrogenation indicators.

**Materials and methods:** Twenty-eight lambs were fed with 4 isoenergetic diets consisting of a control diet without microalgae addition and 3 diets with Nannochloropsis incorporated as oil, spray dried and freeze-dried biomass. After a 3-week trial, the animals were slaughtered, and rumen contents were collected and analysed by 16S sequencing. DNA extraction was performed using the FastDNA™ SPIN Kit for Soil and sequencing on Illumina MiSeq platform. Data were submitted to MGnify pipeline (2.0). Diversity analysis was performed using R vegan package. Differences in bacterial families abundances between treatments were analysed using Proc Glimmix from SAS based in a negative

binomial distribution. Fatty acid methyl esters were analysed by gas chromatography with flame ionization detection and statistics using Proc Mixed by SAS. A Pearson correlation was performed between families and biohydrogenation indicators (i.e. *trans*-10 shift expressed as the *trans*-10/*trans*-11 18:1 ratio) and C18 biohydrogenation completeness as defined by [1] within diets Table 1.

**Results:** In total, 107 bacterial families were identified. Simpson index was similar between diets ( $P = 0.328$ ) with an average of 0.76. *Trans*-10 shift and C18 biohydrogenation completeness were lower in Nannochloropsis treatments ( $P = 0.013$  and  $P = 0.010$ , respectively). The highest correlation with *trans*-10 shift was found for *Eubacteriaceae* ( $P < 0.001$ ) and with C18 biohydrogenation completeness for *Succinivibrionaceae* ( $P = 0.008$ ).

**Conclusion:** Simpson index indicates the presence of a high diversity but no apparent diet influence in the ruminal bacteria family abundances. *Trans*-10 shift and C18 biohydrogenation completeness were lower in Nannochloropsis treatments which indicates a lower content of *trans*-10 18:1 and a lower reduction to 18:0 in the presence of high levels of eicosapentaenoic acid. The role of *Eubacteriaceae* on the *trans*-10 shift should be further explored.

**Acknowledgments:** The authors acknowledge funding Project CIISA UIDB/00276/2020 and PTDC/CAL-ZOO/29654 from FCT - Fundação para a Ciência e Tecnologia IP.

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Table 1

Five most abundant bacterial families (%) in the different treatments/results/Values correspond to the mean  $\pm$  standard error of the mean.

Family	Treatment			
	Control	Oil	Spray-dried	Freeze-dried
<i>Prevotellaceae</i>	22.77 $\pm$ 5.314	28.52 $\pm$ 5.891	29.21 $\pm$ 4.484	29.80 $\pm$ 3.463
<i>Lachnospiraceae</i>	28.76 $\pm$ 3.509	26.49 $\pm$ 3.757	22.42 3.319	31.35 $\pm$ 4.369
<i>Ruminococcaceae</i>	13.51 $\pm$ 2.373	13.67 $\pm$ 2.381	14.69 $\pm$ 3.005	11.20 $\pm$ 1.784
<i>Erysipelotrichaceae</i>	9.53 $\pm$ 3.590	5.05 $\pm$ 2.199	3.38 $\pm$ 0.923	4.27 $\pm$ 1.651
<i>Succinivibrionaceae</i>	3.22 $\pm$ 1.388	3.98 $\pm$ 2.375	3.50 $\pm$ 1.662	6.64 $\pm$ 3.077

#### 84. Effect of different levels of microalgae supplementation on methane emissions, ruminal fermentation, dietary digestibility and nitrogen retention in finishing lambs

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**Application:** Microalgae supplementation can decrease feed intake, and increase digestibility and ruminal VFA concentrations, but has no effect on methane emissions or N retention of finishing lambs.

**Introduction:** In the rumen, dietary fermentation provides energy and protein to the host through volatile fatty acids (VFA) and microbial protein production. However, methane production contributes to greenhouse gas accumulation and represents a loss of dietary energy. In sheep, microalgae supplementation has shown to decrease methane production and increase VFA concentrations in the rumen, with changes in pH and VFA profiles. However, excessive microalgae supplementation might decrease DM intake (DMI) and digestibility. This study assessed the effect of microalgae supplementation at different levels on ruminal fermentation, methane emissions, feed digestibility and nitrogen retention in finishing lambs.

**Materials and methods:** Twenty-four male lambs, Texel/Scottish black face, were blocked into 6 groups of 4 lambs by age and body weight (BW), and randomly assigned into one of four diet treatments. All treatments were fed TMR diets of 50:50 of grass silage and concentrate, with concentrate meals including 0% (Ctrl), 1.2% (Alg-L), 2.4% (Alg-M) and 3.6% (Alg-H) microalgae (DM basis). The DMI and BW were recorded daily and weekly, respectively. At day 24 of treatment, lambs were placed in digestibility crates for 7 days, with urine and faeces collected the final 6 days. Crates were moved inside respiration chambers days 4–7. Methane emissions were measured during the final 48 h. Rumen samples were taken at the end of day 7. Response variables were analysed using linear mixed models (LME) via the REML estimation method, with Treatment as fixed effect and Run and Chamber as random effects. Pairwise differences between treatments were examined using Fisher's LSD test if the overall treatment effect was significant ( $P = 0.050$ ). GenStat (version19) was used to carry out all analyses.

**Results:** The DMI tended to decrease ( $P = 0.078$ ) in Alg-H compared to the other diets (Table 1). Digestibility of DM, OM, NDF, ADF and Energy

Table 1

Effects of microalgae supplementation on feed intake, feed digestibility, methane emissions and rumen fermentation in finishing lambs ( $n = 24$ ).

	Ctrl	Alg-L	Alg-M	Alg-H	SEM <sup>1</sup>	P-value
Body weight, kg	33.81	36.14	34.95	34.02	1.379	0.253
DM intake, kg/d	1.10	1.15	1.11	1.02	0.049	0.078
DM digestibility, %	69.5 <sup>a</sup>	74.1 <sup>b</sup>	70.2 <sup>a</sup>	73.8 <sup>b</sup>	0.988	0.017
OM digestibility, %	71.3 <sup>a</sup>	75.9 <sup>b</sup>	72.1 <sup>a</sup>	75.8 <sup>b</sup>	0.905	0.017
NDF digestibility, %	70.6 <sup>ab</sup>	73.5 <sup>c</sup>	68.9 <sup>a</sup>	73.0 <sup>bc</sup>	1.271	0.007
ADF digestibility, %	73.9 <sup>ab</sup>	75.9 <sup>b</sup>	72.3 <sup>a</sup>	76.2 <sup>b</sup>	1.171	0.044
Energy digestibility, %	66.0 <sup>a</sup>	72.8 <sup>b</sup>	68.7 <sup>ab</sup>	72.8 <sup>b</sup>	1.008	0.030
Nitrogen digestibility, %	86.9	87.4	85.8	86.9	0.660	0.655
Retained Nitrogen, %	20.59	22.31	20.44	18.96	0.871	0.325
Methane emission, g/d	17.16	17.17	16.62	16.07	1.612	0.143
Methane/DMI, g/kg	15.52	14.93	15.01	16.19	1.513	0.465
Rumen pH	7.33 <sup>ab</sup>	7.47 <sup>b</sup>	7.10 <sup>a</sup>	7.13 <sup>a</sup>	0.063	0.024
Total VFA concentration, g/L	2.47 <sup>a</sup>	1.98 <sup>a</sup>	2.58 <sup>ab</sup>	3.25 <sup>b</sup>	0.257	0.012
Acetate, %	64.42	63.41	58.29	59.43	0.919	0.078
Propionate, %	19.42	20.43	22.50	23.16	0.630	0.157
Butyrate, %	13.02	14.02	15.17	13.56	0.721	0.533

<sup>1</sup>SEM = standard error of the mean. <sup>a-c</sup> Least square means within a row with different superscripts differ significantly ( $P < 0.050$ ).

were greater ( $P \leq 0.030$ ) in Alg-L and Alg-H than in Ctrl and Alg-M, while N digestibility and retention were similar ( $P \geq 0.325$ ) between treatments. Ruminal pH was lower ( $P = 0.024$ ) in Alg-M and Alg-H than in Alg-L. Total VFA concentration was greater ( $P = 0.012$ ) in Alg-H than in Ctrl and Alg-L. Acetate proportions tended to decrease ( $P = 0.078$ ) with increasing microalgae supplementation. Methane emissions and methane/DMI were similar ( $P \geq 0.143$ ) between treatments.

**Conclusions:** The present results suggest that microalgae supplementation reduced rumen pH and increased VFA concentrations, with a trend to shift acetate to propionate production, but had no effect on methane emissions. However, feeding different levels of microalgae showed unclear patterns on nutrient and energy digestibility. Therefore, further studies are required to assess the effect of microalgae supplementation in more integrated diets of finishing lambs.

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## 85. Effect of feeding sheep graded levels of forage rape on methane emissions and rumen fermentation

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**Application:** Feeding forage rape (*Brassica napus*) greatly reduces methane (CH<sub>4</sub>) yield when fed as a sole diet. Fermentation pathways away from methanogenesis appeared to be upregulated.

**Introduction:** Feeding forage rape has repeatedly been shown to reduce CH<sub>4</sub> emissions when fed as a sole diet compared with sheep fed perennial ryegrass [2]. In one trial, increasing forage rape in the diet decreased CH<sub>4</sub> yield (g CH<sub>4</sub>/kg dry matter intake) linearly [3]. The objective of this study was to confirm the linear reduction in CH<sub>4</sub> yield, and evaluate ruminal fermentation, when graded levels of ryegrass are substituted with forage rape.

**Materials and methods:** Wether lambs (n = 14/treatment; 42 ± 0.4 kg live weight) were fed ryegrass (*Lolium perenne*) substituted with 0, 25, 50, 75 and 100% forage rape (FR0%, FR25%, FR50%, FR75%, FR100%, respectively) on a dry matter basis. CH<sub>4</sub> and hydrogen (H<sub>2</sub>) emissions, and dry matter intake (DMI) were measured for 48 h in respiration chambers after 15 d of adaptation to the diets. Rumen fluid was sampled via esophageal-stomach tubing, pre-feeding, for volatile fatty acids analysis by gas chromatography. Fourteen sheep/treatment were required to detect 3 g/kg CH<sub>4</sub> yield difference at 0.8 power for significant treatment effect (p = 0.05). Treatment, linear and quadratic effects

were analysed by a mixed model using 'predictmeans' package in R Core Team [1] Table 1.

**Results:** The CH<sub>4</sub> yield (g/kg DMI) decreased quadratically (t = -6.37) while H<sub>2</sub> yield increased quadratically (t = 3.20) with increasing forage rape inclusion in the diet. The CH<sub>4</sub> yield was lower and H<sub>2</sub> yield higher in sheep FR100% compared to any of the other treatments. Rumen pH decreased quadratically (t = 3.00), while rumen fermentation shifted linearly (t = -9.98) toward more propionate and less acetate with increasing forage rape inclusion in the diet.

**Conclusion:** CH<sub>4</sub> reduction was associated with increased H<sub>2</sub> emissions and greater propionate concentration relative to acetate, which, when taken together, suggest that significant upregulation of fermentation pathways away from CH<sub>4</sub> formation in sheep fed FR100%.

**Acknowledgments:** This research was funded by the New Zealand Agricultural Greenhouse Gas Research Centre (NZAGRC) and Maria Della Rosa was financially supported by a NZAGRC LEARN award.

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Table 1

Mean dry matter intake (DMI), gas emissions and fermentation characteristics from sheep fed ryegrass-based pasture substituted with 0, 25, 50, 75 and 100% of forage rape (FR0%, FR25%, FR50%, FR75%, FR100%, respectively).

	DMI	CH <sub>4</sub>	H <sub>2</sub>	pH	Ac	Bu	Pr	A:P
	[kg/d]	[g/kg DMI]		% of total VFA <sup>2</sup>				
FR0%	0.98 <sup>c</sup>	19.18 <sup>a</sup>	0.05 <sup>c</sup>	7.05 <sup>a</sup>	67.73 <sup>a</sup>	10.07 <sup>c</sup>	16.76 <sup>d</sup>	4.05 <sup>a</sup>
FR25%	1.01 <sup>bc</sup>	18.65 <sup>a</sup>	0.08 <sup>bc</sup>	6.95 <sup>a</sup>	65.13 <sup>b</sup>	11.16 <sup>b</sup>	18.89 <sup>c</sup>	3.49 <sup>b</sup>
FR50%	1.08 <sup>ab</sup>	18.09 <sup>ab</sup>	0.12 <sup>b</sup>	6.96 <sup>a</sup>	62.41 <sup>c</sup>	11.93 <sup>b</sup>	21.65 <sup>b</sup>	2.92 <sup>c</sup>
FR75%	1.10 <sup>a</sup>	17.02 <sup>b</sup>	0.12 <sup>b</sup>	6.65 <sup>b</sup>	60.87 <sup>c</sup>	13.83 <sup>a</sup>	21.54 <sup>b</sup>	2.88 <sup>c</sup>
FR100%	1.06 <sup>abc</sup>	12.67 <sup>c</sup>	0.37 <sup>a</sup>	6.18 <sup>c</sup>	53.64 <sup>d</sup>	15.92 <sup>a</sup>	26.68 <sup>a</sup>	2.14 <sup>d</sup>
SED	0.03	0.66	0.11	0.13	1.09	1.84	1.15	0.18
p-value	< 0.01	< 0.01	< 0.01	< 0.01	< 0.01	< 0.01	< 0.01	< 0.01
FR inclusion	—contrast t-values <sup>3</sup> —							
linear	3.00	-9.80	10.16	-6.84	-10.65	5.75	6.93	-9.98
quadratic	-1.93	-4.66	3.20	-3.00	-2.44	1.17	0.88	0.32

SED: average standard error of differences,<sup>1</sup>log transformed variable for statistical analysis, reported values were back transformed.

<sup>2</sup> Total VFA includes: acetate (Ac), propionate (Pr), butyrate (Bu), valerate, caproate, isobutyrate and isovalerate.

<sup>3</sup> linear and quadratic effects were considered significant at t-value > 3.

<sup>a,b,c</sup> treatment means within a column with different letters are significantly different (p < 0.05).

## 86. Using nitrogen isotopic fractionation to estimate *in vitro* ammonia emissions from dairy cattle manure

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**Application:** Nitrogen isotopic fractionation ( $\delta^{15}\text{N}$ ) can be used directly to estimate *in vitro* ammonia ( $\text{NH}_3$ ) emissions from dairy cattle manure without the effects of feed ration.

**Introduction:** Manure ammonia ( $\text{NH}_3$ ) emissions management and mitigation is an important aspect in dairying. The  $\delta^{15}\text{N}_{\text{Air}} \text{‰}$  from manure is a significant parameter concerning dairy cattle manure nitrogen (N) losses. Hristov et al. [2] showed there was a positive relationship between the manure  $\delta^{15}\text{N}$  and accumulative  $\text{NH}_3$  emissions of cattle manure in a 14-day incubation trial ( $p < 0.001$ ,  $r^2 = 0.76$ ). However, Lee et al. [1] only demonstrated the similar relationship from the first 6 days of their incubation trial, and after that, limited relationship was found. This may be due to the differences in their manure properties and incubation condition (e.g., manure nitrogen content, temperature). Therefore, the objective of this study was to establish the relationship between the accumulative  $\text{NH}_3$  emission and the  $\delta^{15}\text{N}$  of dairy cattle manure, when changing manure property.

**Materials and methods:** Urine and faeces samples were collected via vulva stimulation and passing hand through the anus from total 20 random-selected dairy cattle fed with pasture, wheat hay and concentrate feed. An acid trap based *in vitro* dairy cattle incubation system was set

up similar to Lee et al. [1]. Fresh urine and faeces samples were collected from Dookie Campus Robotic Dairy Farm, The University of Melbourne. The experiment was conducted for 15 days and designed based on altering ratio of urinary N to faecal N (UN: FN) (i.e., 1 UN: 1 FN vs. 2 UN: 1 FN). Each treatment group had 4 replications and each replication contained 600 g of manure. Daily manure and acid trapped samples were collected to quantify  $\delta^{15}\text{N}$  and  $\text{NH}_3$  emissions, respectively. All data were analyzed by linear regression analysis and Lin's concordance correlation coefficient (CCC) analysis in Genstat and Minitab, respectively.

**Results:** The accumulative  $\text{NH}_3$  emission was correlated with the manure  $\delta^{15}\text{N}$  (Fig. 1,  $n = 120$ ,  $R^2 = 0.630$ ,  $P = 0.000$ ,  $\text{CCC} = 0.011$ ). The equation was  $Y = -1.183 + 0.1900X$ , while  $Y =$  The accumulative  $\text{NH}_3$  emission (ranged from 0.077 g to 1.594 g) and  $X = \delta^{15}\text{N}$  (ranged from 6.4‰ to 13.9‰).

**Conclusion:** Manure  $\delta^{15}\text{N}$  can estimate *in vitro* ammonia ( $\text{NH}_3$ ) emissions from dairy cattle manure over the 15-day incubation period.

**Acknowledgments:** The authors acknowledge the funding from The University of Melbourne and Australia-China Joint Research Centre.

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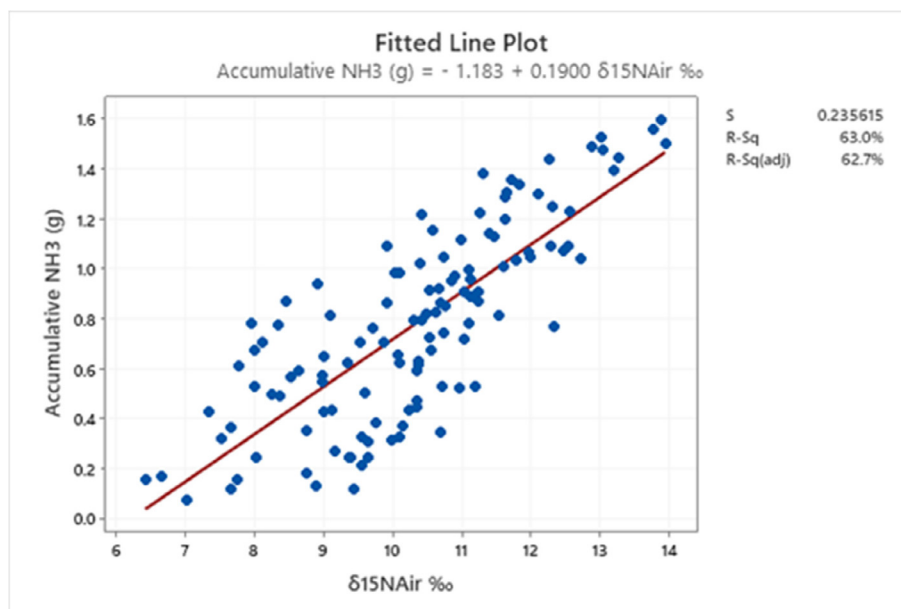


Fig. 1. The regression analysis of the accumulative  $\text{NH}_3$  emission paired with the manure  $\delta^{15}\text{N}$ .

### 87. An investigation of anti-methanogenic effects and digestibility of seaweeds using the rumen simulation technique (RUSITEC)

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**Application:** Enteric methane (CH<sub>4</sub>) production from ruminants represents a major environmental concern. Assessing anti-methanogenic compounds *in vitro* for their effects on methane production, productivity and overall digestibility is essential to validating these additives *in vivo*. In this study, *Asparagopsis taxiformis* reduced CH<sub>4</sub> parameters whilst the other seaweeds increased CH<sub>4</sub> parameters and cannot be recommended for methane mitigation.

**Introduction:** Agriculture accounts for > 34% of Irish greenhouse gas (GHG) emissions, > 60% of which come from ruminant derived enteric methane. Phlorotannins contained in brown algae have shown to inhibit CH<sub>4</sub> production [3], while red algae contain bioactives - bromoform and dibromochloromethane, which have also shown to reduce methanogenesis *in vitro* [1]. The objective of this study was to evaluate the effects of inclusion of red and brown seaweeds, using *Asparagopsis taxiformis* as a positive control.

**Materials and methods:** The experiment was conducted using three RUSITEC systems (8 vessels/system) as previously described by Kirwan et al. [2]. Treatments included: Control (CO - no seaweed inclusion), *Asparagopsis taxiformis* (ASP), *Bifurcaria bifurcata* (BIB), *Chondrus crispus* (CHC), and *Pelvetia canaliculata* (PEC), all supplemented at 10 g kg<sup>-1</sup> dry matter (DM). The *in vitro* basal diet consisted 50:50 grass silage:concentrates on a DM basis. Rumen inoculum was sourced from four rumen-cannulated Holstein Friesian lactating cows on a total mixed ration diet. The study lasted for 21 days, including a 14 day adaptation

period with gas volume, CH<sub>4</sub> and pH recorded daily. Data were analysed using the Mixed procedure in SAS 9.4. Statistically significant differences were considered when P < 0.05, while P < 0.1 was considered as a tendency towards significance Table 1.

**Results:** There was no difference observed between treatments for organic matter digestibility (OMD) and total gas volume (P > 0.05). *A. taxiformis* inclusion decreased total CH<sub>4</sub> gas percentage values (P < 0.05) compared to all treatments, however; it had no effect on reducing CH<sub>4</sub> when expressed as mmol/day or mmol/d DOM<sup>-1</sup> (P > 0.05) relative to the control. Including BIB, PEC, and CHC increased CH<sub>4</sub> mmol/day, mmol/g DOM, and CH<sub>4</sub> volume (P < 0.05) relative to the control diet.

**Conclusion:** Inclusion of seaweed species had no effect on nutrient digestibility. In accordance with Kinley et al. [1], daily CH<sub>4</sub> % was reduced by the addition of *Asparagopsis taxiformis*, in this study the observed reduction was 40%. Apart from *A. taxiformis*, supplementing 50:50 grass silage:concentrate diet with all other seaweeds predominantly increased CH<sub>4</sub> output.

**Acknowledgments:** This project was funded by DAFM via ERA\_NET project SEASOLUTIONS and 'METH-ABATE' (RSF contract no. 2019R479).

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Table 1  
Effect of seaweed supplementation on OMD, gas and CH<sub>4</sub> production.

	CO	ASP	BIB	CHC	PEC	SEM	p-value
OMD	70.37	71.54	72.82	71.24	72.29	0.717	0.2131
Total gas (L/d)	1.68	1.83	2.07	1.78	1.66	0.107	0.0674
Methane parameters							
CH <sub>4</sub> mmol/d	5.85 <sup>b</sup>	4.02 <sup>b</sup>	10.23 <sup>a</sup>	8.31 <sup>ac</sup>	6.43 <sup>bc</sup>	0.578	<0.001
CH <sub>4</sub> mmol/g DOM	0.4 <sup>ac</sup>	0.27 <sup>a</sup>	0.7 <sup>b</sup>	0.58 <sup>bd</sup>	0.43 <sup>cd</sup>	0.038	<0.001
CH <sub>4</sub> %	8.03 <sup>b</sup>	4.79	12.03 <sup>a</sup>	11.33 <sup>a</sup>	7.6 <sup>b</sup>	0.399	<0.001
CH <sub>4</sub> (L/d)	0.14 <sup>b</sup>	0.1 <sup>b</sup>	0.25 <sup>a</sup>	0.2 <sup>ac</sup>	0.16 <sup>bc</sup>	0.014	<0.001

<sup>ab</sup> values within a row with different letters differ P ≤ 0.05.

### 88. Effects of including different seaweeds species in a 50:50 forage: Concentrate diet, on methane production and organic matter digestibility *in vitro* using artificial Rumen Simulation Technique (RUSITEC)

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**Application:** Some seaweeds have the potential as a mitigating strategy to reduce methane emissions in ruminant systems.

**Introduction:** The environmental impact of methane (CH<sub>4</sub>) from ruminant production systems is a growing concern. In Ireland, agriculture accounts for ~ 34% greenhouse gas (GHG) emissions with enteric CH<sub>4</sub> responsible for 56 % of total emissions. Recent studies suggest that macroalgae may contain bioactive compounds that have the potential as a mitigating strategy in reducing enteric CH<sub>4</sub> production in ruminant production systems [2,3]. Red and brown seaweeds differ in their compositions of bioactive compounds with Phlorotannins (PT) a polyphenolic compound found exclusively in brown seaweeds, while halogenated compounds are found within both species [1]. Therefore, the objective of this study was to evaluate the effects of supplementing a 50:50 forage: concentrate diet with different seaweeds species, on CH<sub>4</sub> production and organic matter digestibility *in vitro* using artificial Rumen Simulation Technique (RUSITEC).

**Materials and Methods:** The experiment consisted of two incubation periods using three RUSITEC systems (8 vessels/system). Each vessel was considered an experimental unit, in a completely randomized design. Dietary treatments were Control no seaweed (CO), *Alaria esculenta* (ALE), *Ascophyllum nodosum* (ASC), *Asparagopsis taxiformis* (ASP),

*Fucus serratus* (FUS), *Fucus vesiculosus* (FUV), *Himantalia elongate* (HIM), *Palmaria palmata* (PAL), *Ulva lactuca* (ULI), with seaweeds supplemented at 10 g kg<sup>-1</sup> DM. The *in vitro* basal diet consisted of 50:50 grass silage: concentrate on a dry matter (DM) basis. Rumen inoculum was sourced from four rumen-cannulated dairy cows on a TMR diet. Data was analysed using Proc Mixed procedure in SAS. Statistically significant differences were considered when P < 0.05, while P < 0.1 was considered as a tendency towards significance Table 1.

**Results:** Seaweed inclusion had no negative effect on organic matter digested (OMD), while ASC tended to increase OMD (P < 0.1). *A. taxiformis* inclusion decreased daily CH<sub>4</sub>% compared to all other treatments except ALE (P < 0.001), while including FUS increased daily CH<sub>4</sub>% compared to the control (P < 0.05). Methane volume and production (mmol/d) decreased with ASP inclusion compared to FUS and ULI treatments (P < 0.01), whereas FUS increased CH<sub>4</sub> volume compared to HIM (P < 0.05). When CH<sub>4</sub> production was expressed as OMD, ASP produced less CH<sub>4</sub> mmol/g OMD (P < 0.01).

**Conclusion:** Under *in vitro* conditions, the inclusion different seaweeds had no negative effect on OMD. Compared to the control ASP was the only seaweed species to reduce daily percentage of methane by 36% while no difference between methane L/d and mmol/g OMD.

**Acknowledgments:** This project was funded by ERA\_NET project SEASOLUTIONS and DAFM project METHABATE.

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Table 1

The effect seaweed inclusion on methane production and organic matter digestibility *in vitro* using artificial Rumen Simulation Technique (RUSITEC) system.

	CO	ALE	ASC	ASP	FUS	FUV	HIM	PAL	ULI	SEM	P-value
OMD	70.37	73.62	75.75	70.37	73.12	72.37	71.17	71.93	70.91	1.241	0.078
Total gas (L/d)	1.90	2.14	1.70	1.82	2.33	1.70	1.66	1.98	2.11	0.197	0.161
Methane (mM/d)	5.85 <sup>ab</sup>	10.78 <sup>a</sup>	7.03 <sup>ab</sup>	4.04 <sup>bc</sup>	12.02 <sup>ad</sup>	8.50 <sup>ab</sup>	7.93 <sup>ab</sup>	9.52 <sup>ab</sup>	10.91 <sup>bd</sup>	1.470	0.005
Methane %	8.03 <sup>a</sup>	12.34 <sup>ab</sup>	10.08 <sup>a</sup>	5.12 <sup>b</sup>	12.94 <sup>a</sup>	12.11 <sup>a</sup>	11.41 <sup>a</sup>	11.84 <sup>a</sup>	12.60 <sup>a</sup>	1.017	<0.001
Methane (L/d)	0.14 <sup>ab</sup>	0.26 <sup>ab</sup>	0.17 <sup>ab</sup>	0.09 <sup>b</sup>	0.29 <sup>c</sup>	0.21 <sup>ab</sup>	0.19 <sup>bd</sup>	0.23 <sup>ab</sup>	0.27 <sup>dc</sup>	0.036	0.005
Methane (mmol/g OMD)	0.40 <sup>ac</sup>	0.73 <sup>bc</sup>	0.47 <sup>ac</sup>	0.27 <sup>a</sup>	0.81 <sup>bc</sup>	0.58 <sup>ac</sup>	0.55 <sup>ac</sup>	0.66 <sup>ac</sup>	0.76 <sup>bc</sup>	0.095	0.002

<sup>abc</sup> Rows with different superscripts differ (P < 0.05).

### 89. Assessment of NIRS and wet chemistry analytical methods for nutritive value analysis of microwave treated feeds

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**Application:** Microwave (MW) technology is a non-ionic electromagnetic heating technique that has been applied to different feed resources to improve the nutritive value [2].

**Introduction:** Traditionally, wet chemistry is used to analyze feed nutritive value. However, researchers and many analytical laboratories have changed their preference from slow, hazardous, and expensive wet chemistry analysis methods to near-infrared spectroscopy (NIRS) to evaluate feed nutritive value [1]. The objective of this study was to evaluate the consistency and reliability of the NIRS method for nutritive value analysis of MW treated feed (roughage and concentrates) in comparison to the traditional wet chemistry method Fig 1.

**Materials and Methods:** Five roughage hays (made from lucerne, canola, pasture, oat, and wheat) and two concentrate grains (faba bean and wheat) were used in this study. All the feed types had three samples of 50 gm each for MW treatment. The feed samples were treated within an MW oven operating at 1.1 kW and 2.45 GHz frequency for 0, 20, 40, 60, and 80 seconds and 0.15, 30, 45, and 60 seconds treatment time for roughage and grain, respectively. The three samples of each feed type were combined into one composite sample for nutritive value analysis.

Samples (n = 35) were analysed with wet chemistry and NIRS to compare the nutritive value parameter of determining dry matter digestibility (DMD), digestibility of organic matter in the dry matter (DOMD), crude protein (CP), acid detergent fibre (ADF), and neutral detergent fibre (NDF). We compare the results by doing a simple linear regression analysis in GenStat 16.0.

**Results:** Overall, there was a strong relationship between the wet chemistry and NIRS for all the measured parameters ( $R^2 = 0.89-0.93$ ,  $P < 0.001$ ). The weakest correlation was in the digestibility of organic matter in the dry matter parameter (0.89). All the other parameters showed a correlation of more than 0.90 ( $P < 0.001$ ).

**Conclusion:** Regarding accuracy, NIRS showed good consistency for all the measurement parameters when correlating to wet chemistry. However, further study is required with a large number of data for the extensive use of NIRS as an alternative to the slow wet chemistry process to measure the nutritive value of feeds treated with microwave technology.

**Acknowledgments:** The author would like to acknowledge the NSW Department of Primary Industry Feed testing lab and Dookie Chemistry Lab for providing assistance with the analytical work.

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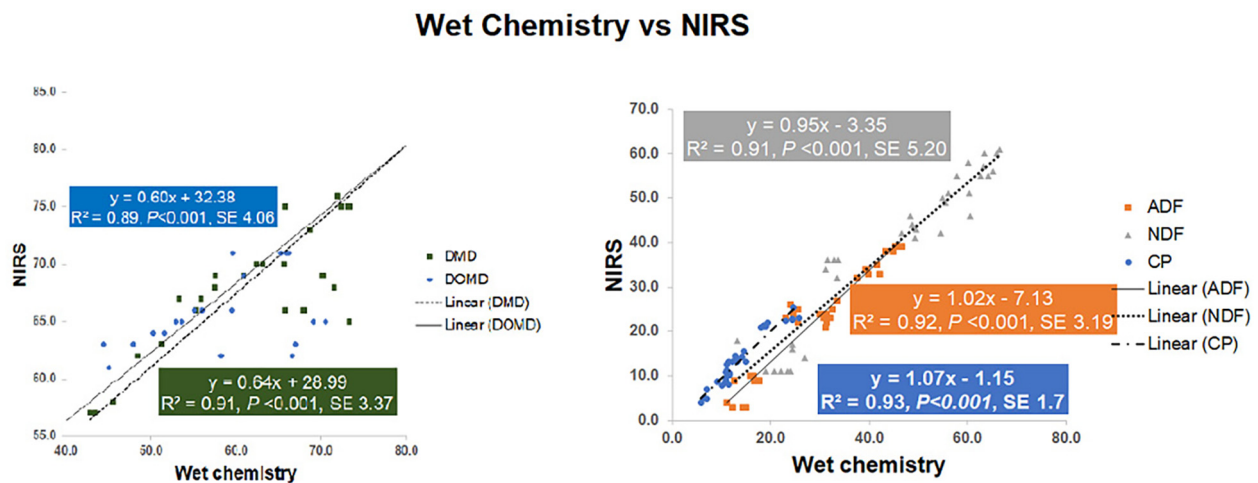


Fig. 1. Comparison between Microwave (MW) treated feed nutritive value parameters analyzed by NIRS and wet chemistry method.

## 90. A survey to understand methods of rearing goat kids away from their dams

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**Application:** This survey suggests that there are likely differing production system needs being encountered by smaller-scale bottle fed versus larger-scale ad libitum milk feeding systems for goat kids. These findings of variable management practices highlight the need for deeper research into the welfare of kids reared in these systems in order for best practices to be identified and communicated, and to ensure welfare is optimised.

**Introduction:** The global goat population has been increasing over the last decade, yet little is known about methods being used to feed milk to kids reared away from their dams and wean them from these systems. Previous surveys investigating management practices have focused solely on dairy goats, whereas this survey included kids reared away from dams for any purpose. The aim of this survey was to quantify and characterise kid rearing and weaning methods currently being used, to enable future research to be suitably targeted towards establishing relevant best practice for both productivity and welfare.

**Materials and methods:** A recall-based survey investigating current on-farm kid-rearing practices was distributed online (in English, French and Dutch) and on paper (UK only). Relationships between farm characteristics and husbandry practices were analysed using Chi-square tests in IBM SPSS version 15.

**Results:** A total of 242 responses from 16 countries were collected. Countries with <10 responses were categorised as 'other'. Small farms

Table 2  
Milk feeding and weaning systems (all countries).

Milk feeding system, n (%)	Bottle		Ad libitum		Other	
	141 (58)		74 (31)		27 (11)	
Weaning method by system, n (%)	Abrupt	Gradual	Abrupt	Gradual	Abrupt	Gradual
	19 (13)	121 (87)	48 (65)	26 (35)	0 (0)	27 (100)

represented a high proportion of respondents (Table 1). A significant relationship between number of kids reared and milk feeding system was identified,  $p < 0.001$ , with farms rearing >100 kids more likely to use *ad libitum* systems. Target weaning ages and weights varied by country (Table 1).

Kids were more likely to be abruptly weaned from *ad libitum* milk than from bottle feeding ( $p < 0.001$ ; Table 2). Strategies for gradual weaning included reducing milk quantity on 150 (93%) and diluting milk on 6 (4%). Enrichment that met the survey's definition was supplied by 155 (64%), with items that allowed climbing/hiding being the most common, provided by 161 (92.5%).

**Conclusion:** Whilst this survey found that bottle feeding was the most common milk feeding method, followed by *ad libitum* systems, it also identified that farms rearing >100 kids were significantly more likely to *ad libitum* milk feed compared to those rearing <100 kids. Abrupt weaning, known to cause stress in other species, was more likely from *ad libitum* milk feeding and could represent a welfare concern.

**Acknowledgments:** The authors acknowledge funding from the University of Reading and the West Country Dairy Awards.

Table 1  
Responses by country, number rearing < 100 kids and >100 kids per year and use of weaning targets.

	All	USA	UK	Australia	Canada	NZ	Other
Total responses, n (%)	242	72 (30)	71 (29)	33 (14)	23 (10)	20 (8)	23 (10)
< 100 kids, n (%)	180 (74)	67 (93)	44 (62)	28 (84)	12 (53)	17 (85)	12 (52)
> 100 kids, n (%)	62 (26)	5 (7)	27 (38)	5 (16)	11 (47)	3 (15)	11 (48)
Use target age, n (%)	165 (68)	55 (76)	37 (53)	37 (53)	19 (83)	13 (65)	12 (52)
Median age, days	84	84	56	84	56	98	60
Use target weight, n (%)	88 (36)	16 (22)	25 (36)	8 (24)	14 (61)	9 (45)	16 (70)
Median weight, kg	16	18	15	16	15.5	20	17
Total responses, n (%)	242	72 (30)	71 (29)	33 (14)	23 (10)	20 (8)	23 (10)



### 91. Effects of the addition of bael (*Aegle marmelos*) on the viability of probiotic bacteria and the physicochemical characteristics of fermented goat milk over the refrigerated storage

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**Application:** The inclusion of up to 20% bael (*Aegle marmelos*) in fermented goat milk may increase the survivability of added probiotics over 21 days of storage.

**Introduction:** Goat milk products have become increasingly popular as a substitute for cow milk. Due to its acceptable pH, good buffering capacity and high nutritional profile, goat milk matrix facilitates long-term survival of probiotics. However, maintaining viability during storage is challenging due to post-acidification. Bael is a sub-tropical fruit belongs to Rutaceae family which is widely utilised in traditional medicinal systems such as *Siddha*, *Unani* and *Ayurveda*. The objective of this study was to determine the microbial and physicochemical properties of bael-incorporated fermented goat milk products containing the probiotic *Lactobacillus rhamnosus* GG (LGG) over 21 days of refrigerated storage Fig. 1.

**Materials and methods:** Four formulations of probiotic fermented goat milk (FGM) namely, FGM containing 0% (CON), 5% (5FGM), 10% (10FGM), and 20% (20FGM) bael were evaluated over 21 days of refrigerated (4 °C) storage. The viability of LGG, colour change and the pH were determined at 1, 7, 14 and 21 days of storage. All the analyses were

conducted in triplicates, and the whole experiment was repeated twice. Data were submitted to statistical analysis as randomized complete blocks arranged in a 4 × 4 factorial scheme (4 treatments and 4 lengths of storage times). Multiple Repeated Measures ANOVAs and separate One-way ANOVAs were performed to determine the simple main effects for time and treatment on physicochemical and microbial viability data, respectively. Means were separated using Tukey Test and the significant differences were determined at  $\alpha = 0.05$ .

**Results:** Goat milk contained 13.43 ± 0.32% total solids, 4.03 ± 0.06% fat, 3.42 ± 0.00% protein, 4.21 ± 0.26% nitrogen-free extract, and 1.77 ± 0.02% ash. Both the storage time and treatment had a significant effect on the pH, colour and viability ( $P < 0.05$ ). The decline in pH over the storage was lower in bael added samples compared to the control. Moreover, increasing levels of bael had resulted in a proportionate decrease in pH. There was no difference in brightness values ( $L^*$ ) among the treatments ( $P > 0.05$ ). However, the addition of bael significantly increased the redness ( $a^*$ ) and yellowness ( $b^*$ ) ( $P < 0.05$ ). The viability of LGG in all four treatments was decreased significantly over the storage period ( $P < 0.05$ ) although the viable counts remained  $> 10^6$  CFU/mL. After 21 days of storage, the viable counts in the bael-incorporated formulations (5FGM, 10FGM and 20FGM) were significantly higher ( $P < 0.05$ ) than that of the formulation without bael (CON). The highest viable counts ( $> 10^7$  CFU/mL) were observed in the sample having 20% bael (20FGM).

**Conclusion:** Results concluded that the addition of bael significantly increased the viability of LGG during refrigerated storage in addition to having positive effects on colour and post-acidification.

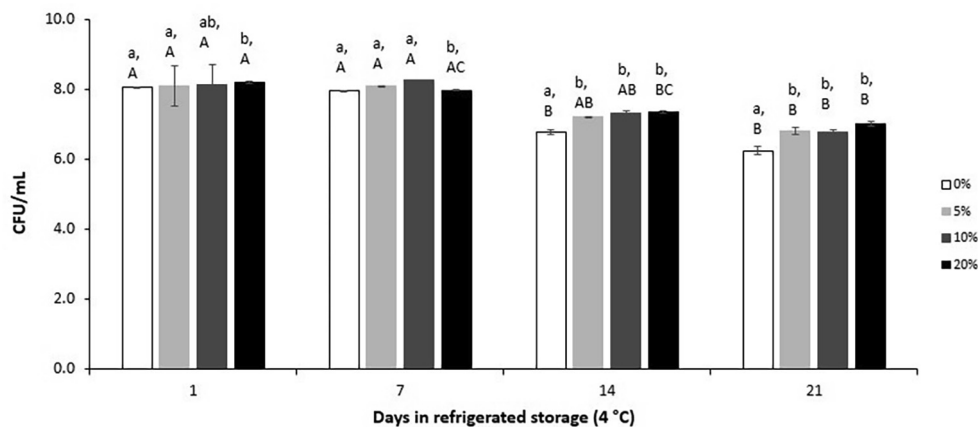


Fig. 1. Viable counts of LGG of fermented goat milk formulations during 21 d of refrigerated storage (4 °C). Results are mean ± SD. <sup>ab</sup>Means without common superscripts in a particular time point are different ( $P < 0.05$ ). <sup>ABC</sup>Means without common superscripts between time points of a particular treatment are different ( $P < 0.05$ ).

## 92. Sustainable intervention for the improvement of goat health and socio-economic outcomes in rural communities in central Botswana

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**Application:** Across rural Botswana, the majority of households rear goats, positioning these animals as strategic contributors to food and financial security in these communities. Improvements to goat health and performance outcomes therefore are likely to yield socio-economic benefits to these communities.

**Introduction:** Goat performance outcomes in rural Botswana are poor; kid mortality is 33%, birth rate 39%, and off-take 7% [1–3]. This study aims to develop sustainable practices to rectify this issue by understanding the socio-economic structure of goat ownership and identifying nutritional and disease management interventions to improve animal performance and ultimately socio-economic outcomes.

**Materials and methods:** Socio-economic analysis and nutritional analysis approaches were combined to address goat herd performance challenges. A socio-economic survey was conducted across 800 households focussed on goat ownership demographics; income food security, goat herd dynamics and goat management practice. Forage nutritional analysis was conducted on samples collected from nine grazing areas, with an emphasis on crude protein and metabolisable energy.

**Results:** Survey results highlighted the importance of goats within these communities. Goat ownership was 56% and more common than any other animal husbandry. Goat holding was found to be negatively associated with the household's concern regarding food security. The main reason for keeping goats was for financial value (80% of respondents)

with an off-take rate of 8.6%. Furthermore, 49% of respondents found that their income was only just sufficient to meet their expenses and 33% indicated it as insufficient. Clear demographic trends were also apparent, with care for goats more likely to fall to older individuals than other groups; 32% of owners were over 65 years of age.

Nutritional analysis identified the importance of browse consumption as a source of protein and energy. The potential for dietary supplementation with other plants, such as *Viscum*, was also apparent due to high concentration of protein (23%, s.d. 3.8) and favourable energy content (16% s.d. 0.07). The nutritional composition of plants varied across both time and space, with lower crude protein concentrations in the dry season. Crude protein concentrations of forages across sites on Hardveld and Sandveld soil types were found to be significantly different ( $p = 0.007$ ).

**Conclusions:** Results highlight both the need for intervention due to poor performance outcomes a lack off food and financial security. However, results also highlight the significant challenges in accomplishing this, notably a lack of financial and technical means and high variability in nutritional resources. Overlaying understandings of different components of the system will enable an improved understanding of sustainable methods for improving goat health and socio-economic outcomes for rural communities.

**Acknowledgments:** Funded by BBSRC (BB/S014748/1)

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### 93. Survival of the fittest? Fitness assessment of *Teladorsagia circumcincta* of known anthelmintic resistance status

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**Application:** There were significant fitness differences between isolates of *Teladorsagia circumcincta*. Further research will explore if these can be exploited to encourage reversion to anthelmintic susceptibility.

**Introduction:** In recent years, the frequency of multi-drug resistant *T. circumcincta* has increased. Detection of resistance to benzimidazole drugs was first documented in the 1960s, followed by levamisoles in the 1970s, and ivermectins in the 1980s [1]. Thus, the search for alternative methods of control and new active drug groups became increasingly important. Repeated use of drug-active groups, incorrect dosing, and moving animals to clean pasture immediately following drenching are some factors contributing to the proliferation of multi-drug resistant nematode populations. Strategies to combat the alarming levels of resistance seen worldwide are essential. Several studies worldwide have investigated reversing the increase in drug resistance, with documented evidence for reversion toward susceptibility in some cases. It has been hypothesised that the accumulation of mutations responsible for resistance may result in an ecological fitness cost. The study aimed to test the hypothesis that resistance mutations result in an ecological fitness cost.

**Materials and Methods:** The fitness of four *T. circumcincta* isolates of known anthelmintic resistance status was assessed using *in vitro* assays:

Isolates R1 and R2 were multi-drug resistant, whilst isolates S1 and S2 were pan-susceptible. The resistance status of the four isolates was confirmed using the larval migration inhibition assay. The *in vitro* assays were designed to assess ex-sheathment rate in artificial rumen fluid, egg hatch rate, larval length (a proxy for fecundity), egg to L3 development after cold-stress, and migration after heat-stress.

**Results:** Isolates R1 and S1 had higher ex-sheathment than S2 after 24 hours incubation in artificial rumen fluid ( $P < 0.01$ ). R2 had a higher egg hatch rate than S2 ( $P = 0.03$ ). S2 was longer than all other isolates ( $P < 0.005$ ), while R2 was longer than R1 ( $P = 0.005$ ). After storage at 4 °C all isolates exhibited decreased egg development. Timepoints after the initial measurement of 0 days are significantly different from the initial measurement for all isolates ( $P < 0.005$ ). All isolates displayed reduced migration ability after exposure to heat-stress (figure 1). Timepoint had a statistically significant effect on all isolates ( $P < 0.005$ ).

**Conclusion:** Despite significant differences between isolates being observed for various fitness traits of *T. circumcincta*, reduced fitness was not definitively associated with multi-drug resistance. Variation in fitness parameters between isolates was large, making it difficult to determine whether resistant genotypes come with lower fitness. Future studies will focus on separating genotypes within isolates to overcome the variation in inter-isolate fitness.

**Acknowledgments:** The authors would like to thank AgResearch, Teagasc, and UCD for their support with this research.

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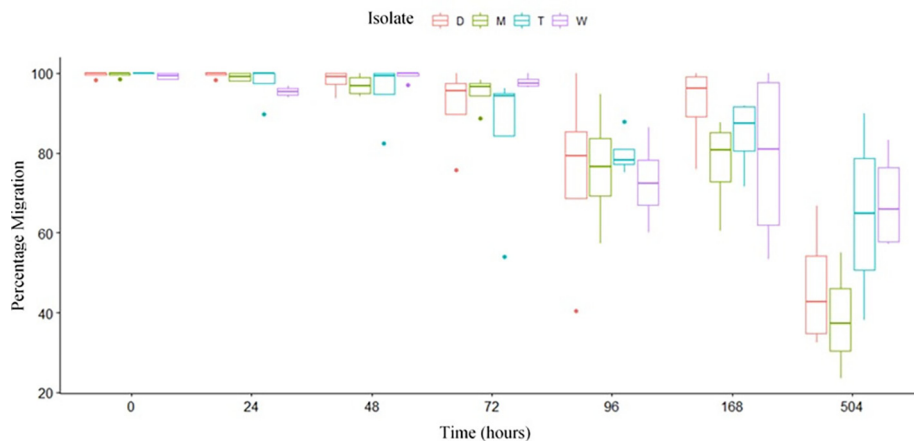


Fig. 1. Percentage migration of stage-three larvae after exposure to 30 °C for a specific length of time (hours), for four isolates of known anthelmintic resistance status.

#### 94. New insight into the role of ovine hoof shape and damage on the susceptibility to *Dichelobacter nodosus* infection

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**Application:** Understanding the role hoof shape and damage can play in the susceptibility of hooves to infectious lameness could help elucidate why sheep in similar environments are not equally susceptible to lameness.

**Introduction:** Footrot accounts for approximately 70% of lameness in English flocks [3]. The primary cause, *Dichelobacter nodosus*, is thought to invade and sequester in damaged or misshapen hooves [1,2]. Our aim was to investigate the associations between hoof shape and/or damage and presence of *D. nodosus*.

**Materials and methods:** Four commercial sheep flocks (A-D) were recruited. No flocks were foot trimmed. Fifteen ewes were initially convenience selected from each flock and sampled over four visits between September 2019 and September 2020. In the absence of ewes (i.e. culled) at a visit, previously unsampled ewes were included to achieve a minimum of 15 ewes/flock/visit. At each visit, all hooves of ewes were scored for sole and heel shape and/or damage, hoof wall shape and/or damage and wall overgrowth. For each variable, a hoof was defined as 'good' when the maximum score was 0 and 'poor' when score was  $\geq 1$ . One sterile cotton swab was also taken from the interdigital skin of each hoof. Genomic DNA was extracted and a real-time PCR assay for the detection of *D. nodosus* was performed. Associations between hoof shape and/or damage variables (fixed effects) and presence of *D. nodosus*

were investigated using a binomial mixed effects model, constructed in R using the "glmer" function from the "lme4" package. Random terms included 'Unique ewe ID' (e.g. 856A) and 'flock ID and visit' (e.g. A1). **Results:** In total, 972 hooves and swab DNA samples were analysed from 243 sheep-level observations of 85 individual ewes. On average, ewes were sampled 2.9 times (95% CI: 2.6–3.1). Approximately 25% of hooves were positive for *D. nodosus*. Hooves with sole and heel damage had an increased risk of being positive for *D. nodosus*, whereas hooves with wall overgrowth had a reduced risk of being positive (Table 1). Hooves with wall damage were no more likely to have *D. nodosus* present.

**Conclusion:** Hooves with misshapen or damaged sole and heel areas are more likely to be infected with *D. nodosus*, which could increase susceptibility to clinical disease. These hooves could act as 'silent spreaders' of *D. nodosus* to healthy feet of other sheep. Interestingly, wall overgrowth had a protective effect on *D. nodosus* infection. Further investigation is required to elucidate the mechanisms behind these findings.

**Acknowledgments:** With thanks to Harper Adams University, MSD Animal Health and Perry Foundation for their funding.

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Table 1  
Binomial mixed effects model of associations with presence of *D. nodosus* from 972 hooves.

Variable	Number of feet	Odds ratio	Lower 95% CI	Upper 95% CI	P
Sole and heel shape and/or damage					
Good	767	Ref			
Poor	205	1.84	1.10	3.08	0.020
Wall shape and/or damage					
Good	241	Ref			
Poor	731	0.94	0.59	1.51	0.800
Wall overgrowth					
Good	268	Ref			
Poor	704	0.55	0.32	0.94	0.028

CI: confidence interval for odds ratio; Ref: baseline category for comparison.

## 95. Evaluation of infrared thermography as a diagnostic tool for the detection of foot lesions in dairy sheep

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**Application:** Early and accurate diagnosis of foot lesions in dairy sheep is important to tackle lameness and ensure welfare. Infrared thermography (IRT) is a non-invasive nature technology, currently utilized by practitioners for the diagnosis of systematic diseases.

**Introduction:** Ovine interdigital dermatitis (OID), footrot and white line disease (WLD) are the most observed foot-related lameness causes in intensive dairy sheep systems. The associated lesions are revealed during routine and/or exploratory foot-trimming. Hence, considering the non-invasive of infrared thermography, our objective was to assess its diagnostic accuracy for the detection of foot lesions in dairy sheep.

**Materials and methods:** One hundred multiparous randomly selected dairy ewes from each of 6 enrolled farms were used in the study (n = 600 ewes). Data were recorded during routine foot-trimming and included lactation number, body condition score (BCS), OID, footrot and WLD lesions. Thermographic images were captured by an infrared thermographic camera (FLIR 8) and processed with Flir Tools software to estimate ambient (AT) and the maximum interdigital skin temperature (MIST), at the foot level (n = 2400 feet). The difference between AT and the MIST (DAMT) for each foot was calculated. SPSS v23 was used for data analyses, that included i) descriptive statistics, ii) two sets of binary regression analyses with lactation number, BCS (covariate) and

MIST (covariate) used as predictors of the outcome variables (occurrence of OID, footrot, and WLD); in the second set MIST was replaced by DAMT, and iii) receiver operating characteristic (ROC) analyses to compare the diagnostic performance of sound foot (0) and foot with lesions (1) and to calculate the optimal efficiency threshold, sensitivity (Se) and specificity (Sp) values when the predicted (by the models) probabilities were considered.

**Results:** Overall, prevalence of OID, footrot, and WLD at the foot level was 8.1% (195/2400), 2.2% (52/2400), and 13.6% (326/2400), respectively. Mean MIST and DAMT were  $33.8 \pm 0.08$  and  $14.5 \pm 0.08$  °C, respectively. Both MIST and DAMT were significant predictors of the occurrence of OID, footrot, and WLD lesions ( $P < 0.001$ ). In the case of MIST, one-degree Celsius increase of IST was associated with 1.39, 1.21, and 1.05 times increased probability of OID, footrot, and WLD lesions occurrence, respectively. The respective values for DAMT were 1.25, 1.16, and 1.08. The most effective models for setting threshold values for diagnosis of foot lesions were the ones that included MIST as covariate. The area under the ROC curve was 0.754, 0.698, and 0.567 for OID, footrot, and WLD lesions, respectively ( $P < 0.001$ ). Optimal efficiency threshold values for the prediction of OID, footrot, and WLD based on the first set prediction equation estimates were 0.0926 (Se: 71.3% and Sp: 68.9%), 0.0237 (Se: 71.2% and Sp: 61.8%), and 0.1318 (Se: 68.4% and Sp: 41.4%), respectively.

**Conclusion:** IRT is a promising method for the early detection of OID and footrot in dairy sheep. However, assessing additional sites on the foot may further improve diagnostic performance and value.

**Acknowledgments:** This research is co-financed by Greece and the European Union (European Social Fund- ESF) through the Operational Programme «Human Resources Development, Education and Lifelong Learning 2014-2020», project code MIS5048473.

## 96. Ethiopian medicinal plants have *in vivo* anti-parasitic activity

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**Application:** Results reported here support the view that Ethiopian small-holder farmers can expect anthelmintic benefits from using *Adenia* sp. and *Cissus ruspolii* plants

**Introduction:** Ethno-medicinal knowledge claims many plants control livestock parasitism [1]. We previously demonstrated anti-parasitic activity *in vitro* of two such Ethiopian plants, i.e. *Adenia* sp. and *Cissus ruspolii* [2]. Here, we assessed their *in vivo* anti-parasitic properties in a mouse model for sub-clinical parasitism in livestock [3].

**Materials and methods:** We used 32 eight-wk-old female C57BL/6 mice, with average body weight (BW) of  $19.1 \pm 0.14$  g, each inoculated with 200 *Heligmosomoides bakeri* infective larvae (d0). Four experimental drench treatments (n = 8) used were de-ionized water (negative control), *Adenia* sp. extract, *C. ruspolii* extract, or ivermectin (positive control). Plant extracts dosing, planned on d22 and d29 at 500 mg/kg BW, unexpectedly resulted in severe side-effects in one and three of the first four mice dosed with *Adenia* sp. and *C. ruspolii*, respectively. Further dosages were reduced by 50% at each step for one mouse at the time, subject to absence of moderate side effects. Resulting dosing averaged 250 and 219 mg/kg BW over two applications for *Adenia* sp. and *C. ruspolii*, respectively. Individual mouse data was collected on d22 and d36 on faecal egg count (FEC, eggs per gram, epg), BW, haematology and worm burdens (d36 only). Parasitological data were log-

transformed prior to statistics. Data was analysed via analysis of variance, with d22 data as covariate. This experiment, approved by SRUC's Animal Welfare and Ethical Review Body, was conducted under Home Office Authority (PPL 60/4395).

**Results:** *Adenia* sp. and *C. ruspolii* treatments reduced log-transformed FECs and worm burdens (Table 1), with associated backtransformed mean FEC reduced by 54% and 56%, and worm burdens by 62% and 25%, respectively, largely in female worms. *Adenia* sp. treatment increased red blood cell concentrations, whilst *C. ruspolii* treatment increased segmented neutrophil and reduced lymphocyte percentages (Table 1). Final BW did not differ between treatments, averaging  $21.8 \pm 0.37$  g.

**Conclusions:** Both *Adenia* sp. and *C. ruspolii* extracts showed anthelmintic properties, supporting their ethno-medicinal use. Further studies are required to determine minimal effective dose and longer-term benefits on resilience to parasitism.

**Acknowledgments:** We thank BBSRC/DFID/SG and SRUC's International Engagement Strategy for funding and UoE March building staff for technical assistance.

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Table 1  
Parasitology and haematology after water, *Adenia* sp. extract, *C. ruspolii* extract or ivermectin drench.

Parameter	Drench treatments <sup>a</sup>				s.e.d.
	Water (n = 8)	<i>Adenia</i> sp. (n = 7)	<i>C. ruspolii</i> (n = 4)	Ivermectin n = 7)	
Faecal egg count (log epg)	3.95	3.59 <sup>**</sup>	3.56 <sup>**</sup>	0.87 <sup>***</sup>	0.11
Total worm burden (log n)	1.83	1.42 <sup>***</sup>	1.69 <sup>#</sup>	0.11 <sup>***</sup>	0.08
Female worms (log n)	1.72	1.29 <sup>***</sup>	1.54 <sup>*</sup>	0.09 <sup>***</sup>	0.07
Male worms (log n)	1.22	0.86 <sup>***</sup>	1.19	0.04 <sup>***</sup>	0.07
Red Blood Cells ( $\times 10^9$ /ml)	7.3	8.6 <sup>#</sup>	7.9	6.9	0.66
Lymphocytes (% of WBC <sup>b</sup> )	85.6	87.9	75.9 <sup>*</sup>	91.6	4.07
Neutrophils (% of WBC)	10.9	7.2	18.2 <sup>*</sup>	6.5	3.48
Monocytes (% of WBC)	2.7	4.2	4.9	1.8	1.44
Eosinophils (% of WBC)	0.7	0.8	1.0	0.1	0.61

<sup>a</sup> Significance relative to water: <sup>#</sup>P < 0.10; <sup>\*</sup>P < 0.05; <sup>\*\*</sup>P < 0.01; <sup>\*\*\*</sup>P < 0.001; <sup>b</sup>White blood cells.

## Invited Speaker Summary

### 97. Protein level reduction and alternative feed ingredients to improve the sustainability of poultry production

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**Introduction:** The recent increase in raw material prices has motivated nutritionist to search for strategies to lower feed cost while maintaining profitability of animal production. One of the strategies

is to reduce dependence on imported soybean meal and use more of local alternative ingredients supplemented with available synthetic amino acids. This strategy has also other positive implications on gut health, litter quality, incidence and severity of footpad dermatitis, animal welfare, carbon footprint, ammonia emission and overall, the sustainability of poultry production. This paper we will highlight some recent research on crude protein reduction, current and future feed ingredient alternatives, and the practical application within the industry.

### 98. The effect of mezquite pod flour in a wheat-based diet on broiler chicken growth performance

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**Application** Inclusion of Mezquite pod flour in broiler chicken diets significantly reduced weight gain, this could inform a recommendation against the use of non-processed Mezquite in broiler chicken diets.

**Introduction:** Mezquite is an under-utilised crop and a possible alternative to soybean. Mezquite is part of the genus *Prosopis*, a group of leguminous trees. Their pods, containing seeds, are valued for their nutrition and are often ground into flour with 7–17% protein [1]. There are few studies looking at feeding pods to monogastric species, but Girma et al. [2] found that a 30% inclusion reduced growth and feed intake in broiler chickens, while a 10 or 20% inclusion had no significant effect. This study aimed to assess the impact of a 15% inclusion of Mezquite pod flour on broiler chicken growth and performance.

**Materials and Methods** This trial was approved by the Animal Welfare & Ethical Review Body at the University of Nottingham. Eighty, one day old Ross 308 Broiler chickens were group housed for 6 days before being split into 20 pens of 4. Birds were fed either *ad-libitum* wheat-soya based diet (CON n = 40), or 15% inclusion of non-processed ground Mezquite pod (*Prosopis juliflora*) (MEZQ n = 40); collected from the Baringo area, Kenya. Both diets were balanced for equivalent energy and protein content, predominantly replacing wheat. Feed intake and weights were recorded twice weekly, before culling on days 35 and 36. The right

breast muscle and liver were dissected and weighed. Data was analysed by one-way (treatment) or two-way (time × treatment) ANOVA (Genstat 19th edition), with significance taken as  $p < 0.05$ .

**Results:** A 15% inclusion of Mezquite pod flour in the diet negatively affected weight gain and FCR. A two-way ANOVA showed a significant interaction between time and feed for weight gain (data not shown), due to significantly higher growth of CON broilers. Mezquite inclusion therefore significantly decreased the final bodyweight ( $P < 0.001$ , one-way ANOVA). Total feed intake was similar, resulting in a significantly higher FCR for the MEZQ group ( $p < 0.001$ , one-way ANOVA). CON breast muscle weights were significantly higher ( $p < 0.001$ , one-way ANOVA). Liver weights were taken as an indication of effects on metabolic active tissues, but these were similar (Table 1).

**Conclusion:** These results suggest that Mezquite pod inclusion affects feed utilisation. This could be due to antinutritional factors, such as tannins or trypsin inhibitors, inhibiting digestion. Further study is needed to identify if antinutritional factors are responsible, and whether post-harvest processing or the use of exogenous enzymes reduces the negative impact.

**Acknowledgments:** The work was funded by GCRF – Research England (RIS1775401), the University of Nottingham and BBSRC.

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Table 1

The effect of inclusion of Mezquite pod flour on broiler chicken growth, expressed as an average per bird at 36 days.

	Initial BWt (g)	Final BWt (kg)	Total Feed intake (kg)	FCR	Breast muscle wt (% of BWt )	Liver wt (% of BWt )
Control	129.1	1.99	2.93	1.42	0.061	0.025
Mezquite	131.5	1.57	2.90	2.03	0.054	0.024
SED <sup>1</sup>	2.3	0.07	0.13	0.08	0.002	0.001
P-value	0.298	< 0.001	0.847	< 0.001	< 0.001	0.240

<sup>1</sup>SED = standard error of the differences of the means. BWt = Body weight, wt = weight.



### 99. Relationship between crude protein content and larval growth in *Tenebrio molitor*

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**Application:** The stage of development at which *Tenebrio molitor* (yellow mealworm) are harvested affects the crude protein content, which is important for inclusion in animal feeds

**Introduction:** There is increasing pressure to find alternatives to traditional feed ingredients for livestock [1]. *Tenebrio molitor* larvae (TM) could be used as a partial replacement of protein-rich feed ingredients (e.g. soya) due to similar composition. However, as a growing larval species there is a large growth phase before metamorphosis. The study aim was to determine changes in TM protein content over larval growth stages to identify the optimal harvesting age for highest protein content.

**Materials and Methods:** TM (Monkfield Nutrition Ltd) maintained on wheat bran (WB) until pupation. TM were kept in a dark incubator (25 ± 2 °C, 60% humidity) and water supplied through cotton wool. 225 pupae were collected, transferred to three plastic dishes (n = 75/dish) and left to metamorphose. After 2 weeks beetles were removed, leaving only eggs in the WB. Hatched TM were present after 2 weeks, which was taken as day 0. TM were left to grow on *ad libitum* WB. Starting at week 6, a 2 g live sample of TM were taken for analysis and killed by cold exposure (-20 °C). This was repeated weekly until

week 13, when pupation started to occur. Samples were freeze dried and analysed for crude protein (EA1112 Elemental Analyser). Data were statistically analysed by one-way ANOVA (Genstat 20th Edition), and when significant (P < 0.05) a post-hoc Bonferroni test was carried out. **Results:** Dry matter of TM significantly increased with increasing age (Table 1). The TM crude protein content and total crude protein when expressed per unit weight were also significantly affected by age, with the former decreasing between weeks 6 and 11 and the latter increasing between weeks 6 and 12.

**Conclusion:** Although dry matter crude protein content decreased with increasing larval age, total protein content per unit weight of TM increased due to increasing dry matter, potentially due to growth and metabolic changes before metamorphosis. This suggests harvesting TM close to pupation is appropriate for use as a feed ingredient. Further work is needed to determine the changes in crude protein content and the relationship to total fat content.

**Acknowledgments:** The work was gratefully supported by funding from AB Agri, AB Vista and BBSRC.

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Table 1  
Changes in dry matter and crude protein content of live TM during larval growth.

Week	6	7	8	9	10	11	12	13	SED	P
Dry Matter (gDM/gTM)	0.24 <sup>a</sup>	0.30 <sup>ab</sup>	0.35 <sup>abc</sup>	0.40 <sup>abcd</sup>	0.43 <sup>bcd</sup>	0.46 <sup>cd</sup>	0.51 <sup>d</sup>	0.46 <sup>bcd</sup>	0.043	< 0.001
Crude Protein (g/gDM)	0.60 <sup>a</sup>	0.58 <sup>ab</sup>	0.57 <sup>ab</sup>	0.56 <sup>ab</sup>	0.53 <sup>ab</sup>	0.51 <sup>b</sup>	0.57 <sup>ab</sup>	0.52 <sup>ab</sup>	0.024	0.013
Total crude protein (g/gTM)	0.15 <sup>a</sup>	0.17 <sup>ab</sup>	0.20 <sup>ab</sup>	0.22 <sup>ab</sup>	0.23 <sup>ab</sup>	0.24 <sup>ab</sup>	0.30 <sup>b</sup>	0.24 <sup>ab</sup>	0.031	0.006

### 100. Influence of malted millet (*Pennisetum glaucum*) on the growth performance, carcass characteristics and economy of feed conversion of broiler chickens

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**Application:** Malting can be used to improve the nutritional properties of millet; and the weight gain, feed conversion ratio and sensory properties of broiler meat.

**Introduction:** Maize, which forms the bulk of energy source in poultry feeds, is in short supply as a result of industrial and human needs, leading to high cost of animal products [1]. This can be replaced by millet, which cheaper production cost. Processing of cereal grains through malting has been found to improve its nutritional value for livestock [2]. The aim of this present study was to evaluate the effect of feeding varying levels of malted and unmalted millet on the growth performance, carcass characteristics, sensory properties and the economy of feed conversion of broiler chickens.

**Materials and Methods:** The malted millet was prepared by washing cleaned millet grains with water, and allowing it to sprout after 36 hours. Six isoproteinous and isocaloric diets were formulated: Diet 1 consisted of a maize based diet and served as the control; Diets 2, 3, 4, 5 and 6 consisted of malted and unmalted millet incorporated into the diets at 0:100 %, 25:75 %, 50:50 %, 75:25 % and 100:0 % respectively. A total of 216 day old CHI strain of broiler chicken were randomly allotted to the six diets, with each diet consisting of three replicates of 12 birds per replicate. The diets were fed to the birds *ad libitum* for eight weeks and data collected on growth performance. After the feeding trial, one bird per replicate was randomly selected, slaughtered through cervical dislocation and analyzed for their carcass

characteristics. The meat from each treatment was then evaluated for its sensory properties such as appearance, flavour, tenderness and juiciness using a nine-point Hedonic scale. Data collected were analyzed using a one way analysis of variance, based on a completely randomized design model.

**Results:** Malting improved the protein content of the millet grains from 10.42 to 11.90 %; reduced the crude fibre content from 2.40 to 1.30 %; and reduced the nitrogen-free extracts from 82.39 to 78.00 %. Birds fed diet containing 50 % malted and 50 % unmalted millet recorded significantly ( $P < 0.05$ ) higher values in body weight gain (1647.22 g), with better feed conversion ratio (1.46) than the control diet (2.28). Dressed weight, weight of kidney, proventriculus, gizzard and heart were significantly ( $P < 0.05$ ) affected by the dietary treatments; and the meat of birds fed the 50 % malted millet diet was significantly ( $P < 0.05$ ) more acceptable than those of the other diets. The feed cost per kg weight gain were significantly ( $P < 0.05$ ) lower for the malted millet diets than for the control diet.

**Conclusion:** Birds fed 50 % malted and 50 % unmalted millet diet had optimal weight gain, better feed conversion ratio and improved sensory properties; due to the improved nutritional quality of the grains.

**Acknowledgments:** The authors acknowledge the Federal University of Technology Minna for using their Research Farm.

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### 101. Effect of dietary probiotics on the growth performance of Cobb 500 broiler chickens

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**Application:** Probiotics are a possible alternative to antibiotics as growth promoters, without residual effect on chicken meat.

**Introduction:** There is growing interest in searching for alternatives to the use of antibiotics in poultry production as growth promoters. The use of antibiotics for growth promotion has been banned in the EU, UK and USA, but its use as a growth promoter has not been officially banned in many countries of the world including Nigeria. Probiotics are live microbial feed additives with beneficial properties like improvement in animal health and growth performance [1]. The effectiveness of probiotics used may be dependent on the species of microorganism, the form it was used either wet or dry, the dosage and the environment in which the birds are raised [2]. This study aims to evaluate the use of probiotics for growth promotion in broilers.

**Materials and methods:** The experiment was conducted at the Research Farm of the Federal University of Technology Minna, Niger State, Nigeria. One hundred Cobb 500 broiler chicks were used for the study. The chicks were weighed and randomly allotted to five treatments in a completely randomized design. This study was approved and followed the standard ethics of the animal production department of the university. The probiotics used contained  $10^9$  CFU/g of *Lactobacillus*. Treatment 1 served as the control without probiotics (0%) while treatments 2, 3, 4 and 5 were diets supplemented with 1.25 g (0.125%),

2.5 g (0.25%), 3.75 g (0.375%) and 5 g (0.5%) of probiotics per 1 kg of feed, respectively. The probiotics dosage for T2 was  $(1.25 \times 10^8$  CFU/kg); T3 ( $2.5 \times 10^8$  CFU/kg); T4 ( $3.75 \times 10^8$  CFU/kg) and T5 ( $5.0 \times 10^8$  CFU/kg). The birds were managed under a deep litter system and were fed iso-caloric and iso-nitrogenous diets with feed and water provided *ad libitum*. Broilers were weighed weekly, feed intake was measured daily, while feed conversion ratio was calculated from the data obtained. All data were subjected to the Analysis of Variance (ANOVA). **Results:** The study conducted showed that the daily feed intake, daily weight gain, feed conversion and final live weight were not significantly ( $P > 0.05$ ) affected by dietary treatments, notwithstanding, the probiotics did not negatively affect the broilers, since the effect did not differ from the control (Table 1).

**Conclusion:** Researchers using probiotics in poultry have reported variable results. The results obtained in this study not been significant may be indicative of the type of microbe used for the probiotics or the fact that the probiotics inclusion levels were low to produce any significant effect, possibly higher levels of probiotics may yield a significant effect on broiler growth performance. Further research may focus on comparing the use of different types of probiotics on broiler growth promotion.

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Table 1  
Effect of probiotics on growth performance of broilers.

Parameters	Treatments					SEM	P value
	T1	T2	T3	T4	T5		
Initial weight (g)	109.00	108.50	108.25	108.00	105.98	0.45	0.07
Final weight (g)	2202.20	2173.40	2191.60	2492.80	2180.80	54.90	0.11
Daily weight gain (g)	49.84	49.16	49.61	56.78	49.40	1.30	0.10
Daily feed intake (g)	98.46	98.65	98.89	97.37	97.54	0.32	0.22
Feed conversion ratio	1.98	2.01	2.00	1.74	1.98	0.04	0.06

## 102. Challenge of change – Effect of *Vernonia amygdalina* as alternative to antibiotics on lymphoid cells in broiler chicken

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**Application:** Health promoting efficacy of phytochemicals to reduce therapeutic antibiotics use.

**Introduction:** Plant extracts, also known as phytochemical feed additives consist of herbs, spices, essential oils and oleoresins elicit varied effect on poultry performance. These include, stimulating blood circulation, improved intestinal microbial composition [5], enhance immune response and intestinal quality [1]. Aqueous *Vernonia amygdalina* extract previously reduced enteropathogenic bacteria in ileum of broiler chickens [3]. This study aimed at determining the effect of aqueous *V. amygdalina* for health promotion on lymphoid cells in spleen of 28 days-old broiler chickens to reduce therapeutic antibiotics use in poultry production.

**Materials and methods:** One-hundred and fifty day – old Cobb 2000 broiler chicks were randomly distributed into three treatment groups having five replicates and ten birds per replicate in a completely randomized design. Treatment 1 served as control group (T1); birds administered antibiotics (Gentaryl-D: a combination of antibiotic and vitamins for growth stimulation/vitamin supplement during periods of disease, convalescence and stress) were assigned to treatment 2 (T2). Birds in treatment 3 (T3) were administered 1 ml of aqueous *V. amygdalina* on days 8, 15 and 22. Feed and water were supplied *ad-libitum*, antibiotics was administered on day 15 for five days (1 gm /1.5 litres of drinking water). One bird per replicate per treatment was sacrificed on day 28

to collect the spleen. This was fixed in 10% neutral buffered formalin for routine histological processing [4] prior to histomorphology analysis of lymphocyte counts in the follicles and peri-arteriolar lymphoid sheet cells [2]. Animal experimentation/data collection were according to ethics approved by Niger Delta University research ethics committee. Data collected were subjected to one-way analysis of variance using IBM SPSS version 25 and significant means separated with Duncans multiple range test.

**Results:** Lymphocyte counts in T3 (140.54) was lower ( $p < 0.05$ ) than T1 (187.83). A value of 182.59 was recorded for T2 which was not different ( $p > 0.05$ ) from values recorded for T1 and T3 (Fig. 1).

**Conclusion:** Although there is no fixed range for lymphoid cells in the body. Increased lymphoid cells number represents an unhealthy state of an animal indicating the animal may be trying to produce more antibodies to fight an infection. Lower lymphoid cells number may be indicative of healthier tissues in physiology which was observed in spleen of broiler chickens administered 1 ml aqueous *V. amygdalina* extract.

**Acknowledgment:** Staff of the teaching and research farm, Niger Delta University are gratefully acknowledged.

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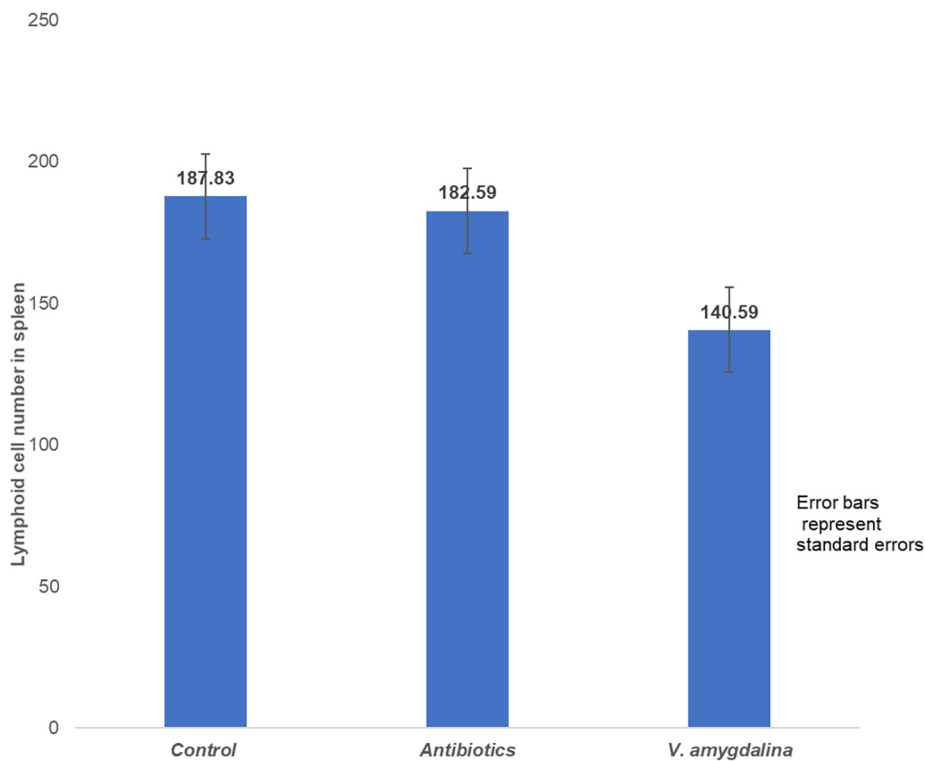


Fig 1: Lymphoid cell number in 28 days-old broiler chickens administered antibiotics and aqueous *Vernonia amygdalina* extract

### 103. Reproductive physiology of heritage turkey toms administered aqueous *Moringa olerifera* leaf and seed extracts

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**Application:** The use of *Moringa* leaf and seed extracts on heritage male turkeys to improve their fertility

**Introduction:** Sexual dimorphism among male and female turkeys poses a serious threat that makes commercial turkey farms to rely solely on artificial insemination to achieve better fertility. Turkeys are more vulnerable to per oxidation due to activities of antioxidants enzymes found to protect the sperm cells from peroxidation by mopping up the oxygen species found within the system [1]. To alleviate these problems, using *Moringa olerifera*, a plant loaded with anti oxidants, vitamins and minerals will go a long way to improve the reproductive potentials of heritage turkey toms. This plant extracts have been used by Cajuday and Pocsidio [3] to improve the fertility of male mice.

**Materials and methods:** A total of 36 male turkey of about 8–9 weeks old were used for the 154 days study. There were nine turkeys per treatment groups, grouped as; T<sub>1</sub> (control) T<sub>2</sub> (1% leaf w/v) T<sub>3</sub> (0.5% leaf and 0.5% seed w/v) and T<sub>4</sub> (1% w/v). Groups was replicated three times with three turkeys per replicate. Semen quality was checked at twenty six (26) weeks of age all the male turkey from each treatment were trained for semen collection using the abdominal massage techniques of Bakst and Long [2]. Collected samples were analyzed for semen colour, sperm motility, total sperm per ejaculation, sperm proportion and sperm concentration. Finally the testes were examined histologically.

Data collected were subjected to analysis of variance using SAS (2016) (Table 1).

**Results:** There were significant difference ( $P < 0.05$ ) on the semen consistency, pH, viable spermatozoa among the treatment groups. The left epidymal weight and corpus also differed significantly ( $P < 0.05$ ) among the extra gonadal parameters assessed. The histological sections of the testis from different treatments reveal the presence of intact seminiferous tubules and interstitial spaces which are highly convoluted lined by germinal epithelium.

**Conclusion:** Single use of the leaves extract promoted the quality of semen better in terms of concentration, progressive motility, viability, corpus weight as well as length of the testes. The seminiferous tubules density was slightly more in the control group. The quality of the semen was better in T<sub>4</sub>. Therefore it is safe to conclude that, using *Moringa* leaves alone has great potential in improving the semen as well as general reproductive performance of local male turkeys.

**Acknowledgement:** Tetitary Education Trust Fund (Federal Polytechnic Oko Unit).

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Table 1  
Mean values of semen traits of heritage turkey toms administered aqueous *Moringa* leaf and seed extracts.

Parameter	T <sub>1</sub>	T <sub>2</sub>	T <sub>3</sub>	T <sub>4</sub>
Semen Volume (ml)	0.29 ± 0.06	0.26 ± 0.07	0.23 ± 0.06	0.20 ± 0.03
Semen Consistency	3.00 ± 0.00 <sup>a</sup>	2.33 ± 0.34 <sup>b</sup>	2.00 ± 0.00 <sup>b</sup>	3.00 ± 0.00 <sup>a</sup>
Semen pH	7.46 ± 0.038 <sup>b</sup>	7.57 ± 0.02 <sup>ab</sup>	7.64 ± 0.03 <sup>a</sup>	7.55 ± 0.08 <sup>ab</sup>
Progressive Motile Spermatozoa (%)	74.40 ± 0.87 <sup>c</sup>	90.83 ± 0.48 <sup>a</sup>	82.93 ± 0.81 <sup>b</sup>	2.87 ± 0.04 <sup>a</sup>
Viable Spermatozoa (%)	87.68 ± 1.24 <sup>c</sup>	96.23 ± 1.53 <sup>a</sup>	91.40 ± 0.8 <sup>b</sup>	96.93 ± 1.22 <sup>a</sup>
SpermConc. (× 10 <sup>9</sup> )	5.24 ± 0.19	5.41 ± 0.16	5.07 ± 0.04	5.93 ± 0.45
Total No of cell per Ejaculation (× 10 <sup>9</sup> )	1.52 ± 0.36	1.44 ± 0.43	1.187 ± 0.31	1.23 ± 0.25
Total Viable Sperm (× 10 <sup>12</sup> )	11.23 ± 2.70	13.01 ± 3.94	9.77 ± 2.49	11.34 ± 2.34
Normal Sperm Proportion (%)	99.52 ± 0.30	99.62 ± 0.17	99.85 ± 0.08	99.56 ± 0.14

Means with different superscripts, a, b or c on the same row are significantly different ( $P < 0.05$ ).

**104. Effect of two probiotics (*Lactococcus lactis* and *Lactobacillus plantarum*) during the starter period on the prevalence of antimicrobial resistance of *Escherichia coli* in the broiler caecum**

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**Application:** The high prevalence of ampicillin and tetracycline resistance by cecal coliforms needs to be controlled. This may not be achieved through the administration of lactic acid bacteria (LAB).

**Introduction:** *Escherichia coli* (*E. coli*) has been used as a sentinel species for the determination of antimicrobial resistance (AMR) [2]. The prevalence of AMR *E. coli* threatens both human and animal health. Probiotics such as *Lactobacillus plantarum* and *Lactococcus lactis* have demonstrated the beneficial effects on the prevention of pathogen growth [1]. Two species of LAB, *Lactobacillus plantarum*, LP, and *Lactococcus lactis*, LL, have been isolated from chicken caeca in previous study and have demonstrated an inhibitory effect on the growth of *E. coli in vitro* [3]. The objective of this study was therefore to determine the effect of administering either LL or LP on the prevalence of AMR of *E. coli*.

**Materials and methods:** A total of 240 chicks were used in a 28 d feeding trial, with birds maintained according to the breeder's recommendations. All chicks were blocked by liveweight, randomly placed in one of 24 pens (10 chicks per pen, eight replicate pens per treatment) and fed a starter diet until the birds were 14 days of age. Water was provided either untreated (Control) or with a preparation of LL or LP ( $10^7$  cfu/ml water offered) via a nipple drinker for three days each week for 14 d. On day 14, birds were weighed and changed to a grower/finisher diet.

One bird per pen was sacrificed at random when birds were 7, 9, 11, 14 and 28 days of age. Samples of caecal digesta were taken and a subsample (1 g) was then serially diluted with 0.01 M Phosphate-buffered saline (PBS) and 100  $\mu$ l of the suspension spread on MacConkey agar. Colonies were transferred 24 h later using the replicate plating method to four separate MacConkey plates, containing (ampicillin, nalidixic acid, tetracycline or chloramphenicol, all 20  $\mu$ g/ml) to assess AMR. The effect of bird age and probiotics on the proportion of AMR coliforms was determined by ANOVA (without repeated measures, general linear model Minitab 19).

**Results:** Little resistance to nalidixic acid or chloramphenicol was observed, but there was a high prevalence of resistance to both ampicillin and tetracycline (Table 1). This increased as birds aged from 7 to 11 d (ampicillin,  $P < 0.001$ ) or from 9 d to 11 d (tetracycline,  $P < 0.01$ ). Neither LL nor LP affected the prevalence of AMR.

**Conclusions:** Even in absence of antibiotic exposure, birds are susceptible to colonisation with coliforms resistant to both ampicillin and tetracycline. The administration of probiotics did not control this colonisation.

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Table 1  
Effect of bird age on the prevalence of AMR *E. coli* (CFU/g).

Antibiotic	Bird age (d)					SEM	P-value	
	7	9	11	14	28		Age	Treatment
Ampicillin	60.43 <sup>bc</sup>	44.47 <sup>c</sup>	82.43 <sup>a</sup>	73.34 <sup>ab</sup>	71.81 <sup>ab</sup>	4.89	< 0.001	0.29
Tetracycline	88.99 <sup>b</sup>	98.8 <sup>a</sup>	100 <sup>a</sup>	95.57 <sup>ab</sup>	99.4 <sup>a</sup>	1.85	0.001	0.13
Nalidixic acid	11.96	7.17	13.05	24.2	8.98	5.13	0.17	0.10
Chloramphenicol	0.00	0.00	0.59	0.00	0.00	0.31	0.41	0.37

### 105. Estimation of body weight via gizzard weight in growing broilers

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**Application:** Body weight of Ross 308 broilers could be predicted from gizzard weight with acceptable prediction error.

**Introduction:** Body composition data have been used in system approaches and modelling, which have been gaining popularity as tool in animal nutrition studies. The accuracy of such models in predicting performance or organs, and nutritional requirements relies upon detailed body composition data. Previously, association of gizzard dimensions with the body weights in meat poultry has been reported by Mobini [1]. However, very little information is available on the assessment of relationship or association of gizzard weight with body weight. A heavier gizzard may be related to better body weight. The aim of this study is to assess the association of gizzard weight with body weight and determine the ability of gizzard weight to predict live body weight in Ross 308 broilers at 21 days post-hatch (Table 1).

**Material and methods:** The study is extracted from a project ethically approved by the Ethical Review Committee, Nottingham Trent University, where 50 day-old as-hatched Ross 308 broiler chickens were reared with free access to standard broiler diet meeting the nutritional requirements for Ross 308 broilers, and water for 21 days. On day 21, all birds were weighed, euthanized by cervical dislocation and gizzard weight was recorded. Gizzard weight was regressed on live body weight to produce the linear and non-linear prediction models using statistical software R (3.6.1) with lm package in it. Regression model was preferred over simple correlation values to calculate the prediction equation.  $R^2$  and 95% percentile prediction error (PE) was used to assess

Table 1

Comparison of two models for estimating body weight via gizzard weight in growing broilers.

Model type	Simple linear	Box-cox
Equation	Body weight (g) = 321 + 26.4*gizzard weight (g)	Log <sub>10</sub> (body weight (g)) = 2.62 + 0.015*gizzard weight(g)
R <sup>2</sup>	0.617	0.602
RSE	99.3	0.057
PE 95 % percentile	22.5	3.28

RSE = Residual standard error, PE = prediction error, Simple Linear Model p-values (Model < 0.001, Intercept < 0.001, Slope < 0.001), Box-Cox Model p-values (Model < 0.001, Intercept < 0.001, Slope < 0.001).

prediction accuracy for the model. Box-cox transformation was used to identify the best transformation which increases the linearity of the data.

**Results:** Mean (and standard deviation) of body weight and gizzard weight was 773 g (159 g) and 17.1 g (4.73 g), respectively. Body weight and gizzard were highly correlated ( $r = 0.7854$ ,  $p < 0.001$ ). Gizzard weight predicted body weight of 95% of the birds with prediction error of 22.5%. When gizzard weight was log<sub>10</sub> transformed, the PE of the model substantially decreased to 3.28%.

**Conclusion:** The gizzard weight has tendency to predict the body weight. Transformation of gizzard weight predicts body weight of Ross 308 growing broilers at 21 days of age with predictability error of 3.28%. Further studies are required for the validation of model.

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### 106. Using a multi-state modelling (MSM) approach to predict dairy calf health status using on-farm precision technology data

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**Application:** Mean motion index, drinking speed, daily live weight and birth weight were found to be important variables for predicting calf health with evidence to suggest prediction is potentially easier after the third week of life.

**Introduction:** Calf mortality is a major issue within the UK livestock sector [3], with dairy heifer mortality rates observed up to 12% in the first month of life [1]. Farm-based automatic monitoring systems collect behavioural and physiological data in real-time offering the opportunity for earlier detection of health issues. This study explored the application of MSM for predicting calf ill-health from data obtained from on-farm precision technologies matched with base metadata and health records.

**Materials and methods:** Feeding behaviour and activity data was collected via automatic feeders and IceRobotics® IceQube® sensors from sixty group housed Holstein-Friesian calves over the first ten weeks of life. Birth weight, gender and daily live weight was recorded together with sickness, defined as day animal treated (based on visual indicators for respiratory and enteric disease). To capture the time characteristics observed in the activity data, “summary measures” were derived [2]. To explore data leading to an event (healthy/sick), time-lagged data was engineered. The predictive capability of MSM was explored using RStudio (Version 1.3.1056). The Likelihood Ratio Test was applied to determine covariate inclusion in the model. The probability of a calf moving from healthy to sick was derived using the MSM package. Hazard Ratios with 95% confidence intervals (CIs) were produced per covariate for each state transition. Performance was evaluated using the confusion matrix, sensitivity & specificity and the area under the receiver operating characteristic curve (AUC).

Table 1  
Hazard ratios with 95% CIs.

	Healthy to Sick	Sick to Healthy
Daily live weight	0.8949 [0.8602,0.9309]	1.0162 [0.9776, 1.0563]
Mean motion index	0.9620 [0.9459, 0.9784]	1.0048 [0.9911, 1.0186]

**Results:** The MSM classifier comprised daily live weight, mean motion index, birth weight and drinking speed, in order of importance. The prevalence plot suggests it is potentially easier to predict calf health after third week of life (Fig. 1), coinciding with biological and behavioural factors. The Hazard Ratios (Table 1) suggest an increase of 1Kg in daily weight is associated with 11% less risk of moving from healthy to sick ( $P < 0.05$ ); an increase of 1 unit of mean motion index is associated with 4% less risk of moving from healthy to sick ( $P < 0.05$ ). Validating the classifier on unseen data gave an AUC score of 0.86.

**Conclusion:** Using a MSM approach, real-time data from precision technologies can be used to aid prediction of calf ill-health offering earlier interventions and individualised treatment.

**Acknowledgements:** The authors gratefully acknowledge the Livestock Production Sciences Branch, AFBI. This study involved the analysis of data produced from a PhD project funded by AgriSearch.

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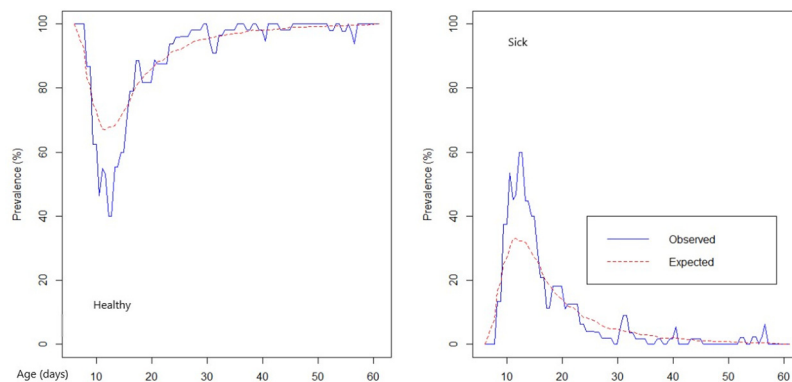


Fig. 1. Prevalence plot - observed versus predicted by age (in days)



### 107. Predicting male dairy calf live-weight for use in calf management decision support

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**Application:** Planning is paramount during the calving period to ensure the availability of ample housing, feeding facilities and labour. Results suggest it is possible to predict calf live-weight and, therefore, the age at which a male dairy calf will reach a target live-weight thus facilitating the planning process.

**Introduction:** Male dairy calves are often deemed to be a low value product. Consequently, producers habitually want to sell these calves as quick as possible, however, purchasers of male dairy calves often stipulate a minimum calf live-weight. Retaining calves to achieve such target weights often results in increased pressure on available farm resources. The objective of the present study was to develop a relatively simplistic statistical model to inform producers when an animal will likely reach a minimum target weight thus aiding in farm planning and management.

**Materials and Methods:** After edits, live-weight data were available on 602 male dairy calves between the ages of 10 to 42 days originating from 34 herds. A linear regression model was developed regressing calf live-weight onto age at weighing (days), dam parity number, gestation length (days), and herd of origin as well as three estimated breeding values for birth size, birth weight and carcass weight. Animals were ran-

domly assigned into 10 smaller datasets and a 10-fold cross-validation was undertaken. The model was applied to each individual fold as well as across all data.

**Results:** The mean (standard deviation) live-weight and age of all male calves in the dataset was 56.61 kg (SD = 8.66 kg) and 26 days old (SD = 5.77 days). The correlation between the actual and predicted live-weight varied from 0.64 to 0.82 per fold and was 0.76 across all data. The root mean square error of the predicted live-weight was 5.65 kg across all data, ranging from 4.40 kg to 6.66 kg per fold. The regression coefficients were relatively consistent across all folds (Fig. 1). Across all data, live-weight increased by, on average, 0.61 kg and 0.22 kg per day of age and gestation length, respectively. Live weight increased, on average, by 0.75, 0.18 and 0.10 kg per unit increase in estimated breeding value for birth weight, birth size and carcass weight, respectively. Simple algebra can be used to transform the prediction equation into one that can predict when a calf will reach a given target weight. Coupled with calving date predictions from service and pregnancy diagnosis data, it would also be possible to estimate when and how long the calf will be resident on the farm.

**Conclusion:** The robustness of the statistical model presented suggests there is potential to transform the prediction equation into one capable of estimating the number of days required for a calf to reach a stipulated target weight. This information can then be used to inform producers of the anticipated workload and infrastructure requirements.

**Acknowledgement:** Funding from VistaMilk is gratefully acknowledged.

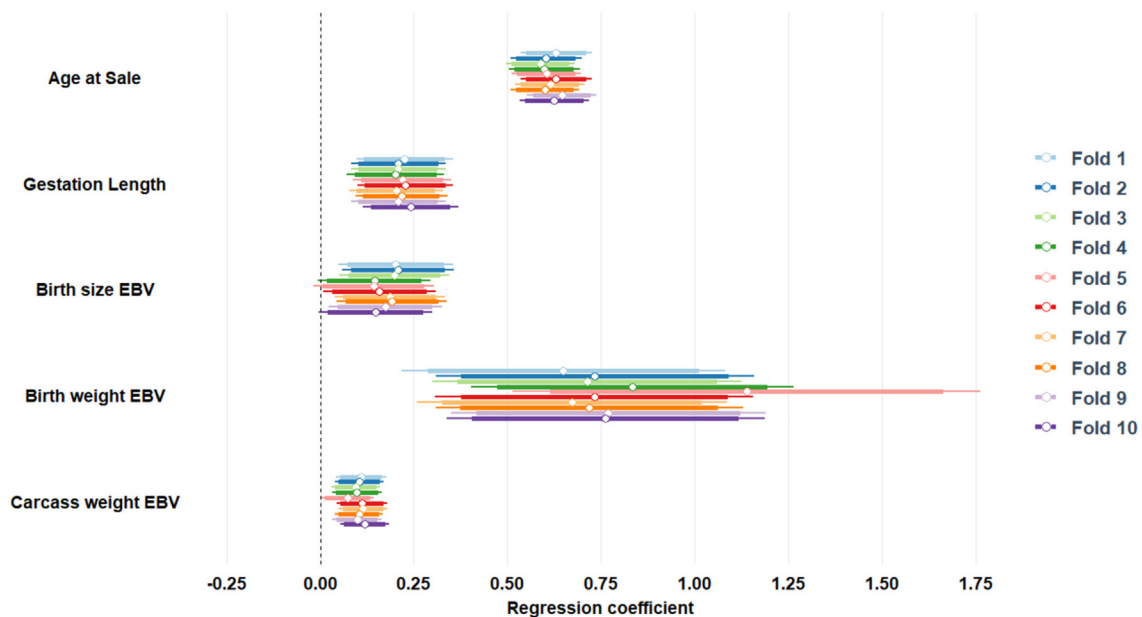


Fig. 1. Model regression coefficients (symbols) for the covariates fitted in each of the ten folds; thick and thin lines represent 90% and 95% confidence intervals.

### 108. Effect of environmental enrichment on feed intake, feed efficiency and cross-sucking of dairy bred calves offered high amounts of milk replacer twice daily

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**Application:** Cross sucking (CS) was almost eliminated in calves reared in pairs by bedding calves with straw directly following milk feeding, while feed efficiency (FE) was not affected.

**Introduction:** Artificially reared calves commonly suck each other following milk consumption [3], which is indicative of poor welfare [1,2]. This study aimed to compare individually and pair housing calves, along with the effect of environmental enrichment (EE) on CS in pair reared calves on feed intake, growth rate and FE of dairy bred calves.

**Materials and methods:** Calves ( $n = 48$ ) were randomly selected and allocated according to birth date and live weight (LW) ( $49.3 \pm 0.45$  kg) to housing in pairs or individually ( $2.5 \text{ m}^2$ ) at 7 to 10 d of age. At 5 weeks of age calves were offered one of three EE's; Cereal bedding straw (Straw) and Manila rope (Rope) for one hour directly following twice daily milk feeding (max 11.5 l/d) until weaning at 12 weeks of age. Dry matter intake (DMI), LW and FE were assessed bi-weekly, while behaviour was assessed twice daily for 10 mins pre and 30 mins post milk feeding [3]. DMI, LW and FE were normally distributed and assessed using GLM AVONAs, while behaviour was not normally distributed and assessed using Kruskal Wallis commands (Minitab, 17.0). Animal was applied as a random effect, while housing and EE were

applied as fixed effects in the models. Mean and medians are presented with relevant P values (Table 1).

**Results:** Calves housing calves in pairs performed more CS (Table 1), unless offered straw bedding following milk feeding. Offering straw directly following milk consumption, reduced CS and increased play, nutritive behaviour, while manilla rope did not. Housing, calves individually or in pairs, EE and oral engagement with straw had no effect on starter intake or FE.

**Conclusion:** Calves housed in pairs performed greater amounts of CS compared with individually housed calves which undertake more non-nutritive oral behaviour. This CS was almost eliminated by offering straw bedding as an EE directly following milk feeding, but not by offering manilla rope. Individual and pair housing of calves and offering bedding straw and rope as an EE did no effect DMI, LW and FE of dairy bred calves.

**Acknowledgement:** Suputo (formerly Dairy Crest) for partially funding of this research.

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Table 1

Mean dry matter intake (DMI) and feed efficiency (DMI / weight gain) and median behaviour of calves housed either individually or in a pair and offered environmental enrichment (EE); None, manilla rope (Rope) or bedding straw (Straw) directly following milk replacer meals.

	Individually	Pair housed			SEM	P value	
	None	None	Rope	Straw		Housing	EE
Calves, No.	12	12	12	12	-	-	-
Starter intake, g/d	0.69	0.70	0.73	0.76	0.041	0.101	0.704
Milk intake, L/d	9.5	9.5	9.5	9.5	0.07	0.989	0.989
Growth rate, g/d	0.91	0.90	0.91	0.92	0.031	0.801	0.795
Cross sucking, s	0.0 <sup>b</sup>	21.0 <sup>a</sup>	20.5 <sup>a</sup>	0.0 <sup>b</sup>	-	<0.001	<0.001
Non-nutritive behaviour, s	72.0 <sup>a</sup>	49.2 <sup>b</sup>	47.4 <sup>b</sup>	11.0 <sup>c</sup>	-	<0.001	<0.001
Nutritive behaviour, s	0.0 <sup>b</sup>	0.0 <sup>b</sup>	0.0 <sup>b</sup>	0.02 <sup>a</sup>	-	0.358	<0.001
DMI/LW gain, kg/kg	1.54	1.58	1.57	1.55	0.31	0.711	0.650

<sup>a, b, c</sup> - Means in the same row that do not share a letter are significantly different at  $P < 0.05$ .

### 109. A preliminary evaluation of British farmers' perception of dairy cow-calf rearing

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**Application:** Assessing farmers' perception towards cow-calf rearing is important to understand their incentive to adopt alternative rearing strategies, or the obstacles to doing so.

**Introduction:** A common component of the British conventional dairy system is that cow and calf are separated in order to collect saleable milk from the dam, however, the issue of early separation remains contentious [1]. Cow-calf rearing systems, whereby calves are reared entirely or in part by the dam or foster cow, are receiving increasing attention amongst consumers and producers alike but current uptake of these systems in the UK is minimal. The aim of the present study was to gain insight into why some dairy farmers adopt cow-calf rearing strategies and why others do not, what they perceived were the main benefits or challenges of these, and whether they thought cow-calf rearing was a feasible option in modern dairy production.

**Materials and methods:** Semi-structured interviews (average 86 minutes long) were carried out with 11 British dairy farmers. Farms recruited varied in calf rearing system (conventional-artificial /cow-calf rearing), geographic location and herd size (20–800 + cows), milking system (robotic/parlour milking). All interviews were recorded with consent and transcribed using NVivo software (QSR International Pty Ltd. (2018) NVivo Version 12). Data collection and analysis overlapped in an iterative approach and a pilot study was conducted to identify flaws in the interview schedule prior to data collection. Data were coded thematically and key trends identified. All transcriptions were checked against the recordings for accuracy and a sample of transcriptions were sent to colleagues to confirm assessment.

**Results:** Most farmers stated that there were benefits for cow-calf rearing, including reduced labour costs and improved calf growth, health, and welfare, but were conflicted on the issue of separation stress and on the loss of saleable milk which was the primary deterrent to adopting cow-calf rearing systems. Conventional farmers expressed concerns around perceived lack of control during the milk feeding period, particularly surrounding colostrum management, and the impact on farm economics when adopting cow-calf rearing systems. But farmers rearing calves with cows stated that the benefits outweighed potential challenges, in particular improved calf health and growth, which afforded economic benefits in the long term. Conventional farmers stated that foster cow rearing was the most commercially attractive option, since it did not result in the loss of saleable milk and would be their preferred approach, providing uptake was incentivised.

**Conclusion:** The study suggests that there is a disparity in attitudes towards cow-calf rearing amongst dairy farmers and that there is no consensus regarding the potential benefits or challenges associated with the system. This suggests more research is needed to provide clarity regarding the effects of cow-calf rearing.

**Acknowledgments:** The author gratefully acknowledges the farmers involved in this study, and funding from Devenish Nutrition and the Silcock Fellowship for Livestock Research.

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### 110. Effect of oligosaccharides on calf health, antimicrobial use, live-weight gain, feed intake and feed efficiency of dairy bred calves

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**Application:** Growth rate of dairy bred bull calves was greater when oligosaccharides were added to milk replacer.

**Introduction:** Morbidity in young calves is largely attributed to enteric disorders and respiratory disease [1] and antimicrobials are an integral part of livestock production. However, human mortality due to antimicrobial resistant (AMR) bacteria is projected to be 10 million by 2050, eclipsing those due to cancer by 1.8 million annually [2] and the development of natural alternatives to antimicrobials are essential for calves. The aim of this study was to compare the performance of dairy bred bull calves offered milk replacer (MR) without and with the addition of oligosaccharides.

**Materials and methods:** A total of 60 dairy bull calves were allocated, at 10 ( $\pm 3$ ) d of age, to one two treatments according to birth date and live weight (LW) (n = 30 calves/treatment) and offered MR without ( $50.4 \pm 1.09$  kg) and with the addition of oligosaccharides ( $49.3 \pm 1.09$  kg). Calves were offered *ad libitum* access to stored colostrum till 10 ( $\pm 3$ ) d of age, followed by which calves were housed in pairs and individually offered MR at 5 up to 11 L/d (150 g MR/L) between 10 and along with *ad libitum* access to water and calf starter (16 % CP and 12.4 MJ ME /kg DM). Calves were gradually weaned at 12 weeks of age. Data was normally distributed and analysed using general linear mixed models procedure (GLM ANOVA), applying as a fixed effect and animal as a random effect in the model. Differences were assessed by Tukey's test with a confidence interval of 95% and differences reported at  $P < 0.05$ .

Table 1

Mean electrolyte and antimicrobial use, dry matter intake (DMI), live-weight gain (Gain) and feed efficiency of dairy bred dairy bull calves of milk replacer without (MR) and with oligosaccharides (MR + OGs).

	MR	MR + OG	P value
Electrolyte use, d	1.46 (0.354)	1.55 (0.341)	0.560
Antimicrobial use, d	20.0	20.0	0.425
Milk replacer intake, kg/d	1.21 (0.009)	1.22 (0.009)	0.696
Starter intake, kg DM/d	2.60 (0.101)	2.72 (0.108)	0.451
Weight gain, kg/d	1.15 (0.023)	1.22 (0.024)	0.035
DMI/ gain, kg DM/kg	2.07 (0.075)	2.04 (0.080)	0.794
Gain/DMI, kg/kg DM	0.50 (0.020)	0.50 (0.021)	0.978

**Results:** Calf growth rate was greater when oligosaccharides were added to milk replacer, but had no effect on electrolyte and antimicrobial use, or feed intake and feed efficiency (Table 1).

**Conclusion:** Calf growth rate was greater when oligosaccharides were added to milk replacer. However, the inclusion of oligosaccharides in milk replacer had no effect on electrolyte and antimicrobial use or feed intake and feed efficiency.

**Acknowledgement:** Suputo (formerly Dairy Crest) and Denkavit for providing products for the completion of this research.

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### 111. Accounting for individual cow variation in milk composition and dry matter intakes when allocating concentrates on a feed-to-yield basis

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**Application.** Increasing precision of concentrates allocation within a feed-to-yield system did not improve energy corrected milk yield or efficiency of energy use for milk production.

**Introduction.** While a feed-to-yield (FTY) approach aims to improve 'precision' by allocating concentrates according to individual cow milk yields, it may be possible to improve efficiency by taking account of additional parameters such as energy balance [1]. The current study hypothesised that production and efficiency could be improved by taking account of individual cow milk composition and dry matter intake (DMI) within a FTY system.

**Materials and methods.** Mid-lactation housed Holstein dairy cows (24 primiparous, 55 multiparous) balanced for lactation, previous two week milk yield and composition, and intake were used in a 12 week continuous design study. All cows were offered a 'basal' mixed ration of grass silage and concentrates, the latter included at a rate of 5.3 (4.3) kg/d for cows (heifers) through individual feed-boxes mounted on weigh scales. Additional concentrates were then offered on a FTY basis through an out-of-parlour feeder, using three different approaches: CON, the basal ration was assumed to meet each cow's maintenance energy requirements plus a fixed amount of milk based on group intake of basal ration: average 20.8 (14.4) kg milk/day for cows (heifers), with additional concentrates offered to individual cows at 0.43 kg/kg milk not supported by the basal ration; Precision 1, used the same approach as CON, with

additional concentrates offered on the basis of individual cow milk energy output (per Tyrrell and Reid [2]); Precision 2, milk supported by the basal ration was calculated on an individual basis, and additional concentrates offered on the basis of individual milk energy output. Concentrate allocations were calculated weekly based on the previous week's milk yield and intakes and the previous two week's composition. Mean weekly data were analysed using REML, with cow as experimental unit and week as repeated measure.

**Results.** Total DMI was not affected, but CON cows had lower concentrate DMI ( $P = 0.036$ ) and lower milk protein content ( $P = 0.003$ ). No other parameters were affected by treatment. All parameters changed over time. There were significant Week  $\times$  Treatment interactions for concentrate and total DMI ( $P < 0.001$ ) and energy balance ( $P = 0.021$ ) (Table 1).

**Conclusions.** While concentrate DMI and milk protein content were higher within the Precision treatments, neither ECM or ECM/ME intake were improved by 'precision feeding' of concentrates compared to conventional FTY. This experiment provides no evidence that accounting for milk composition or individual DMI when allocating concentrates improves feed use efficiency.

**Acknowledgments:** Co-funded by Department of Agriculture, Environment and Rural Affairs and AgriSearch.

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Table 1  
Effect of concentrate allocation strategies on intake, milk production and energy use efficiency of dairy cows.

	CON	Treatments		SED	P values		
		Precision 1	Precision 2		Trt.	Week	Week $\times$ Trt.
Concentrate DMI (kg/d)	8.8 <sup>a</sup>	10.2 <sup>b</sup>	9.9 <sup>b</sup>	0.43	0.036	<0.001	<0.001
Total DMI (kg/d)	21.2	21.8	21.5	0.24	0.113	<0.001	<0.001
Milk yield (kg/d)	32.9	34.5	34.3	0.68	0.181	<0.001	0.002
Fat (g/kg)	45.1	44.9	43.1	0.81	0.055	<0.001	0.767
Protein (g/kg)	32.7 <sup>a</sup>	33.5 <sup>b</sup>	33.1 <sup>b</sup>	0.24	0.003	0.059	0.910
Energy corrected milk (kg/d)	34.6	37.0	36.3	1.93	0.563	<0.001	0.578
Energy balance (MJ/d)	8.7	10.9	11.1	2.51	0.592	<0.001	0.021
Body weight (kg)	626	644	645	19.8	0.416	<0.001	0.181
Energy corrected milk <sup>1</sup> /ME intake (kg/MJ)*	0.14	0.14	0.13	0.002	0.984	<0.001	0.187

ME, metabolizable energy

$$^1 \text{Energy corrected milk} = (((0.0376 \times \text{milk fat}\%) + (0.0209 \times \text{milk protein}\%) + 0.9048) \times \text{milk yield}) / 3.1$$

## 112. Effect of concentrate intake and cow genotype on milk composition within a feed-to-yield system

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**Application:** Cows offered a high level of concentrates on a feed-to-yield basis had reduced milk fat content, which will reduce the value per kg of milk produced by these cows.

**Introduction:** While feed-to-yield (FTY) systems seek to improve the precision of concentrate supplementation, this approach inevitably results in the highest yielding cows receiving high levels of concentrates. There is some evidence [1] that higher yielding cows managed within FTY systems have poorer milk composition which may impact the profitability of these cows. This on-farm study was designed to identify if higher yielding cows offered concentrates within a FTY system had lower milk fat and protein content, and if cow genotype (predicted transmitting ability (PTA) fat and protein %) contributed to this.

**Materials and methods:** This study was conducted on 27 dairy farms during winter 2018/19, and involved 3547 early lactation cows. All cows were housed and offered concentrates on a FTY basis. Each farm was enrolled in a milk recording scheme which provided monthly test-day milk yield and milk composition data. Information on concentrate intakes for each cow during the week prior to milk recording was obtained, either from automated concentrate feeding systems, or calculated using information on feeding practices on farm at that time. Individual cow genetic information was also collected from a subset of

pedigree registered herds (n = 18). Within each of lactations 2 and 3 the data set was split into six equal groups according to the concentrate intake of each cow. Data were analysed using a linear mixed model, with farm and animal within farm fitted as random effects, and month of lactation, concentrate intake and their interaction fitted as fixed effects.

**Results:** As concentrates were offered in response to milk yield (FTY), milk yield increased with increasing concentrate level (Table 1). Milk fat content decreased with increasing concentrate level in lactation 2 (P < 0.001), and tended to decrease in lactation 3 (P = 0.066). This decrease in milk fat content can only be explained in part by the fall in PTA for milk fat in cows offered higher levels of concentrate, indicating that diet was also likely to be a contributing factor. Milk protein content was not affected by concentrate level in either lactation, although PTA for milk protein % decreased with increasing concentrate level in lactation 2 cows.

**Conclusions:** The reduction in milk fat concentration at higher concentrate feed levels can be explained by both genetic and nutritional effects. However, milk protein content was unaffected by concentrate feed level.

**Acknowledgment:** This work was co-funded by DAERA and AgriSearch through the Research Challenge Fund.

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Table 1

Effect of concentrate intake group on milk composition of lactation 2 and 3 cows managed on a FTY basis, and PTA values for cows within those groups (mean data for months 2–5 post-calving).

	Concentrate intake group (kg DM/cow/day)						SED	P Value
	< 7.7	7.7–9.1	9.1–10.3	10.3–11.6	11.6–13.4	> 13.4		
Lactation 2								
Milk yield (kg/day) <sup>1</sup>	26.9	30.5	33.2	36.0	38.9	44.4		
Fat (%)	4.17 <sup>c</sup>	4.08 <sup>bc</sup>	4.06 <sup>ab</sup>	4.07 <sup>b</sup>	3.98 <sup>ab</sup>	3.96 <sup>a</sup>	0.050	0.003
Protein (%)	3.32	3.31	3.29	3.30	3.29	3.27	0.014	0.110
PTA fat (%)	0.08 <sup>c</sup>	0.08 <sup>c</sup>	0.07 <sup>c</sup>	0.06 <sup>bc</sup>	0.05 <sup>b</sup>	0.02 <sup>a</sup>	0.011	< 0.001
PTA protein (%)	0.04 <sup>b</sup>	0.04 <sup>b</sup>	0.04 <sup>b</sup>	0.03 <sup>b</sup>	0.03 <sup>b</sup>	0.01 <sup>a</sup>	0.005	< 0.001
Lactation 3								
Milk yield (kg/day) <sup>1</sup>	< 8.7	8.7–10.3	10.3–11.4	11.4–12.6	12.6–14.6	> 23.8		
Milk yield (kg/day) <sup>1</sup>	28.5	33.1	36.1	38.8	42.1	47.8		
Fat (%)	4.14	4.07	4.09	4.04	4.05	3.96	0.06	0.066
Protein (%)	3.25	3.26	3.26	3.26	3.24	3.24	0.017	0.823
PTA fat (%)	0.07 <sup>b</sup>	0.06 <sup>b</sup>	0.06 <sup>b</sup>	0.06 <sup>b</sup>	0.03 <sup>a</sup>	0.01 <sup>a</sup>	0.012	< 0.001
PTA protein (%)	0.04	0.04	0.04	0.03	0.02	0.03	0.006	0.128

DMI; dry matter intake; PTA; predicted transmitting ability

<sup>1</sup> Mean of concentrate group

### 113. A survey of Northern Ireland farms practising selective dry-cow therapy

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**Application:** The results of this survey provides insight into how and why selective dry-cow therapy (SDCT) is practiced on Northern Ireland (NI) farms

**Introduction:** While the use of intramammary antimicrobials at drying-off is standard practise on most farms. There is evidence that not all cows require an antibiotic at drying-off [1]. Consequently a small number of farmers have adopted SDCT, whereby antimicrobials are only used on cows considered 'high risk'. This survey was conducted to provide insight into SDCT practises being used on farms in NI, and to gain insight into farmer experiences and motivations.

**Materials and methods:** A survey of NI dairy farmers (n = 32) who were practising SDCT was conducted, with farmers recruited through vets and CAFRE dairy advisors. Farmers completed the questionnaire either in 'hard copy' and returned by post (n = 7), or online (n = 25) created using Snap® survey software (Snap version 11). Data were exported to Excel and summary statistics produced.

**Results:** Farm data indicated SDCT has been adopted on smaller and larger herds, with both moderate and high yielding cows (Table 1). The majority of farms participated in a milk recording scheme (88%), with

54% recording on a monthly basis. On average, 64% of cows in herds were dried-off without antibiotics.

The SCC thresholds used by farmers to select cows varied, being 100,000, 150,000 and 200,000 cells/ml on 23, 33 and 27% of farms respectively, with threshold levels in place for 3 months prior to drying-off. Eighty % of farmers agreed that a cow should have no cases of mastitis during the 3 months prior to drying-off. Over 60% of farmers used one or more additional indicators to identify cows for SDCT (Fig. 1).

Procedures applied when drying cows off without antibiotics varied, 69% treated drying-off as a 'separate task' to milking; 63% washed and dried teats; 38% cleaned and dried back teats first; 66% dip/sprayed teats prior to drying-off; 91% cleaned teat ends with surgical spirit/disinfectant before applying teat sealants; 94% used teat sealant. The main reason cited for adopting SDCT was the understanding that not all cows require an antibiotic at drying-off (31%). Over 70% of farmers agreed that the adoption of SDCT had not had a negative impact on herd mastitis incidence or SCC.

**Conclusions:** The survey highlights the potential opportunities within the industry to standardise SDCT drying-off protocols to maintain hygiene and help reduce the risk of drying-off without antibiotic use.

**Acknowledgments:** Farmers that participated in the survey and the contribution by local vets and CAFRE dairy advisors. This work was carried out in partnership with Agrisearch, Animal Health and Welfare Northern Ireland, LMC and Farm Vet systems, and co-funded by DAERA.

Table 1

Mean herd size, annual milk sold/cow and somatic cells counts of participating farms.

	Mean	SD	Minimum	Maximum
Herd size (cows)	152	82	30	390
Milk sold/cow/year (kg)	7,952	1,481	5,200	10,500
Somatic cell count ('000/ml)	132	38	64	200

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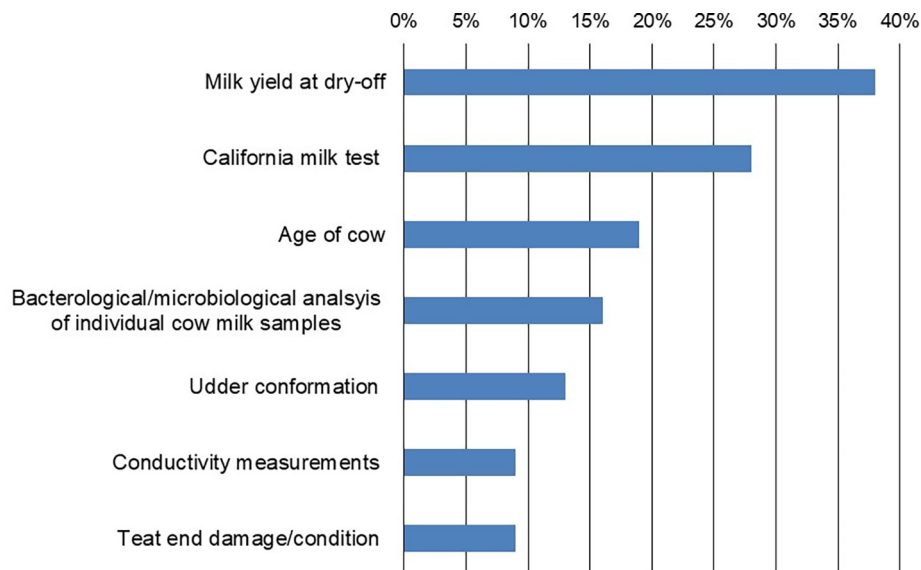


Fig. 1. Additional indicators used by farms to identify cows for SDCT (% of farms)

#### 114. Effect of grazing and feeding management on milk mineral concentrations

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**Application.** Pasture intake and non-Holstein genetics were negatively correlated with milk I, Mn, and Cu, and positively correlated with milk Ca and P; but other feeds and mineral supplementation also influenced milk mineral concentrations.

**Introduction.** Milk is a good source of minerals (Ca, Mg, P and K) and trace elements (I, Se, Zn), which are essential for optimum health [2]. Pasture intake may increase milk Ca and P concentrations but decrease Cu and Se concentrations, although pasture composition, plant maturity, and animal/environmental factors will also have an effect [1]. This study aimed to (i) investigate the milk mineral concentrations in herds with different pasture intakes, and (ii) assess the relative impact of individual feeds.

**Material and methods.** Bulk-tank milk samples (n = 359) were collected monthly (over 12 months) from 30 dairy farms, and animal diet and breed were gathered via questionnaire. Three groups of 10 farms represented contrasting grazing management between April-September: high pasture intake (HP; 28–65% DMI), standard pasture intake (SP; 5–18% DMI), outdoors with low/limited pasture intake (LP; 0–3% DMI). Milk mineral concentrations were determined using ICP-MS. Analysis of variance by linear mixed effects models used pasture intake (HP, SP, LP), months (12 months), and their interaction as fixed factors. Farm ID was used as a random factor. A multivariate redundancy analysis (RDA; CANOCO 5) assessed the relative impact of breed and feeds on milk mineral concentrations.

**Results.** When compared with HP and LP farms, milk from SP farms contained significantly less Ca (–50 mg/kg and –45 mg/kg, respectively) and P (–25 mg/kg and –29 mg/kg, respectively) (Table 1). The RDA indicated that pasture intake and non-Holstein genetics were negatively correlated with milk I, Mn, and Cu, and positively correlated with milk Ca and P. Grass/grass-clover silage and dry-straights were positively correlated with I, Mn, Cu, Mo, Zn and negatively correlated with Na. Intakes of maize silage, blends, moist by-products, oils and minerals were negatively correlated with Mn, Cu, Mo, Zn, Ca and P. Milk from low/no grazing periods contained, on average, more I (+21.1%), Mn (19.5%), Cu (+16.9%), Zn (+6.1%) and Mo (+5.5%), than milk produced from the grazing season, although between-month differences were not always significant.

**Conclusions.** Cow diet, which affects mineral intakes, and breed, which may affect mineral utilisation, had significant effects on milk mineral concentrations. The overall effect of grazing management was not significant, but seasonal variation and correlations between pasture intake and mineral concentrations illustrate a potential effect of grazing on milk mineral concentrations.

**Acknowledgements:** Authors acknowledge financial support from EIT Food, the innovation community on Food of the European Institute of Innovation and Technology (EIT), a body of the EU, under the Horizon 2020, the EU Framework Programme for Research and Innovation (activities/projects: NMR MIRACLE 19118/19118-20/20095, INSPIRE 20092); and EIT Food's Regional Innovation Scheme Talents. Special thanks to dairy producers who assisted with data/sample collection.

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Table 1  
Effect of grazing management on milk mineral concentrations.

Major minerals	HP	SP	LP	SE	P-value	Trace elements	HP	SP	LP	SE	P-value
Ca (mg/kg milk)	1015.7 <sup>a</sup>	966.2 <sup>b</sup>	1010.8 <sup>a</sup>	9.77	0.001	Cu (ug/kg milk)	62.1	61.9	58.4	3.52	0.815
K (mg/kg milk)	1364.8	1352.5	1380.1	12.74	0.246	I (ug/kg milk)	301.8	314.4	384.2	21.21	0.626
Mg (mg/kg milk)	95.5	95.4	97.4	1.03	0.511	Mn (ug/kg milk)	54.2	49.4	43.4	8.53	0.460
Na (mg/kg milk)	341.6	340.8	348.7	5.01	0.652	Mo (ug/kg milk)	62.2	59.5	61.2	2.55	0.713
P (mg/kg milk)	799.7 <sup>a</sup>	774.8 <sup>b</sup>	803.8 <sup>a</sup>	8.37	0.045	Zn (mg/kg milk)	5.07	5.07	5.1	0.21	0.989

HP, high pasture intake (28–65% DMI); SP, standard pasture intake (5–18% DMI); LP, outdoors with low/limited pasture intake (0–3% DMI). Significant differences were declared at P < 0.005. Means with different lower case letter are significantly different according to Fischer's least significant difference test (P < 0.05).



### 115. Increasing the frequency of ejaculate collection in young dairy bulls increases semen production and field fertility

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**Application:** Increasing ejaculate collection frequency may be a useful strategy to satisfy market demands for semen from young genomically selected sires.

**Introduction:** There is a trend in the cattle industry to market semen from younger genomically selected bulls, which have limited ability for sperm production and often fail to meet market demands [1]. The objective of this study was to assess the effect of ejaculate collection frequency on semen output, sperm quality and field fertility in young bulls under commercial conditions.

**Materials and methods:** Holstein-Friesian bulls aged  $366 \pm 8$  days (mean  $\pm$  SEM) were assigned one of two ejaculate collection frequencies; namely high frequency (HF,  $n = 14$  bulls), where ejaculates were collected twice a day, five days in each two-week period and low frequency (LF,  $n = 12$  bulls) where ejaculates were collected once a day, two days per week. The trial period continued until each bull reached 20 ejaculates and 1000 marketable semen straws. Subjective motility was assessed on all ejaculates pre-freeze and post-thaw (both immediately and post-stress where sperm were held at  $37^\circ\text{C}$  for 2 h). A subset of ejaculates was assessed post-thaw for motility and kinematics by computer assisted sperm analysis (CASA) and for viability, membrane fluidity, acrosome integrity, reactive oxygen species and DNA fragmentation by flow cytometry. A total of 13,846 inseminations (9,541 for HF and 4,305 for LF) were carried out on dairy cows and heifers.

**Results:** HF reached 1000 straws 41 days earlier than LF ( $P < 0.01$ ), with the same number of ejaculates. Ejaculate volume and sperm concentration were not affected by treatment but the first ejaculate of the day (HF only) had a greater volume ( $P < 0.001$ ) and sperm concentration ( $P < 0.05$ ) than the second ejaculate. HF had higher pre-freeze total ( $P < 0.01$ ) and gross ( $P < 0.05$ ) motility than LF. HF had higher post-stress total and gross motility than LF ( $P < 0.05$ ). Ejaculate rejection rates did not differ significantly between treatments. There was no effect of treatment, week or ejaculate number of the day (HF only) on post-thaw motility and kinematic parameters assessed by CASA or sperm viability, membrane fluidity, acrosome integrity and DNA fragmentation assessed by flow cytometry. However, HF had lower superoxide production than LF ( $P < 0.05$ ). Pregnancy rate was  $64.5 \pm 1.0\%$  for HF and  $59.9 \pm 1.1\%$  (mean  $\pm$  SEM) for LF bulls ( $P = 0.05$ ).

**Conclusions:** Collecting ejaculates more frequently from young bulls, significantly reduced the number of days to obtain 1000 straws, increased semen quality in terms of lower superoxide production and increased field fertility. This may be a useful strategy to obtain a greater number of semen doses from young bulls in a shorter period of time.

**Acknowledgements:** The authors acknowledge funding from the Irish Research Council (Project: EBPPG/2019/173) and Science Foundation Ireland (Project: 16/IA/4474).

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### 116. Production performance of dairy cows offered rumen-protected (calcium salt) fat supplements differing in proportion of palmitic and oleic fatty acids

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**Application:** Supplementation with calcium salt fat supplements increases milk yield and milk fat production without negative effects on body condition.

**Introduction:** Calcium salts are common rumen-protected fat supplements offered to dairy cows to increase energy density of diets. Responses to fat supplements are influenced by their fatty acid profile and recent data have highlighted contrasting production responses with differing fatty acid blends [1]. The objective of this study was to evaluate the production effects of two calcium salt supplements differing in palmitic to oleic ratio when offered to dairy cows.

**Materials and methods:** Eighteen early-mid lactation (mean 61 days in milk; range 33 to 94) Holstein-Friesian dairy cows, mean initial milk yield 43.3 kg/d (range 33.2 to 57.3), were split into three dietary treatment groups in a Latin Square design study with 28-d periods. Two 3 × 3 Latin squares were used. Three groups of six cows were established based on similarity of milk production, live weight, days in milk and parity, and cows in each group were allocated at random to one of six experimental groups. Three of these groups were then assigned to one Latin square template while the other three groups were assigned to the second Latin square template. The diet treatments were (1) Control, no fat supplement; (2) Control ration plus Megalac (48% C16:0, 36%

C18:1); or (3) Control ration plus Mega-Max (58% C16:0, 28% C18:1) (Megalac and Mega-Max are calcium salt brands manufactured by Volac International Ltd). Target intake of fat supplements was 500 g/cow/d. The Control total mixed ration contained grass silage, maize silage, whole crop wheat, crimped wheat, field beans and dairy concentrate. Dry matter intake was measured for the first five and the last five days of each 28-d feeding period. Milk yield and composition were recorded daily and data from the last five days in each feeding period used for statistical analysis. Live weight and condition score of each cow were measured weekly. Data were analysed by ANOVA using GenStat 14 (VSN International).

**Results:** Fat supplementation increased ( $P < 0.001$ ) milk yield; cows offered the Megalac formulation produced an additional 3.1 kg/d and those offered Mega-Max an additional 1.6 kg/d, with no significant change to body weight or condition (Table 1). Dry matter intake was similar ( $P > 0.05$ ) between treatments both in the initial changeover and end of period recordings. Fat supplementation increased ( $P < 0.01$ ) milk fat yield and maintained milk protein yield ( $P > 0.05$ ), though milk protein concentration was reduced ( $P < 0.001$ ).

**Conclusion:** Supplementation with calcium salts of differing palmitic to oleic ratio effectively increased milk yield and fat production. No negative effects on dry matter intake were recorded following an immediate changeover from a non-fat supplemented diet to calcium salt-supplemented diets.

#### Reference

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Table 1  
Production performance of cows offered Control or fat-supplemented diets.

	Diet			SED	P
	Control	Megalac	Mega-Max		
Dry matter intake (kg/d) : initial 5d of each period	22.9	22.3	21.9	0.66	NS
Dry matter intake (kg/d) : final 5d of each period	22.7	23.0	22.5	0.53	NS
Milk yield (kg)	38.8 <sup>a</sup>	41.9 <sup>b</sup>	40.4 <sup>c</sup>	0.71	< 0.001
Milk fat (g/kg)	40.1	39.1	40.5	0.82	NS
Milk fat (kg/d)	1.520 <sup>a</sup>	1.633 <sup>b</sup>	1.612 <sup>b</sup>	0.0345	< 0.01
Milk protein (g/kg)	32.0 <sup>a</sup>	30.5 <sup>b</sup>	30.8 <sup>b</sup>	0.27	< 0.001
Milk protein (kg/d)	1.228	1.275	1.231	0.0286	NS
Live weight (kg)	656	658	650	3.9	NS
Condition score <sup>#</sup>	1.98	2.00	1.99	0.029	NS

# 5-point scale Differing subscripts within a row denote significant difference

### 117. The effect of a zero-grazed perennial ryegrass, perennial ryegrass and white clover, or multispecies sward on the dry matter intake and milk production of dairy cows

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**Application** Multispecies swards can increase the milk production of dairy cows while requiring less nitrogen fertiliser to grow.

**Introduction:** Multispecies swards can reduce nitrogen losses from dairy systems through a reduction urinary nitrogen excretion from grazing dairy cows [3], and a lower nitrogen fertiliser requirement [1]. However, their effect on milk production is unclear. Some studies have found no difference, while in studies that reported increased milk production, increased dry matter intake (DMI) was not concurrently reported [2]. Our objective was to investigate differences in DMI and milk production of dairy cows offered varying sward types in mid-late lactation.

**Materials and methods** A perennial ryegrass (PRG), PRG and white clover (PRGWC) and multispecies sward containing PRG, timothy, WC, red clover, chicory and plantain (MULTI), were compared using 24 cows in a 3-period crossover experiment. Animals were blocked on days in milk and balanced for parity, BCS and pre-experimental milk production. Cows were acclimatised to dietary treatments for 14 days, before entering a 21-day sampling period. Cows were housed indoors throughout the experiment, with access to sward treatments via computerized feeding boxes which recorded daily forage fresh weight intake. A concentrate nut formulated for grazing dairy cows (14% CP; 3 kg as fed/day), was offered in addition to forage. Swards were cut using a zero-grazer and feed boxes filled throughout the day. Sward DM was determined daily and forage DMI subsequently calculated. Milk yield was recorded daily, and composite milk samples taken weekly during sampling periods. Data was analysed in SAS using PROC MIXED and checked for normality using UNIVARIATE. Day was included as a repeated measure and cow within sequence as a random measure. Model selection via linear regression was set at  $P < 0.25$ .

**Results:** Forage DMI was higher for MULTI and PRGWC compared to PRG ( $P < 0.05$ ). Cows offered MULTI had significantly higher milk yield

Table 1

Effect of sward type on forage DMI and milk production.

Item	Treatment			SEM	P-value
	PRG	PRGWC	MULTI		Sward
Forage DMI (kg/d)	15.41 <sup>a</sup>	16.35 <sup>b</sup>	15.95 <sup>b</sup>	0.427	<0.001
Milk Production (kg/d)					
Milk yield	20.33 <sup>a</sup>	20.03 <sup>a</sup>	21.63 <sup>b</sup>	0.728	<0.001
Energy corrected milk	24.50 <sup>a</sup>	23.57 <sup>a</sup>	25.90 <sup>b</sup>	0.576	<0.001
Fat	0.94	0.89 <sup>a</sup>	0.96 <sup>b</sup>	0.029	0.047
Protein	0.74 <sup>a</sup>	0.73 <sup>a</sup>	0.78 <sup>b</sup>	0.019	0.002
Milk solids	1.67 <sup>a</sup>	1.61 <sup>a</sup>	1.75 <sup>b</sup>	0.044	0.002
Lactose	0.98 <sup>a</sup>	0.97 <sup>a</sup>	1.06 <sup>b</sup>	0.022	<0.001
Milk Quality (%)					
Fat	4.64 <sup>a</sup>	4.54	4.33 <sup>b</sup>	0.119	0.025
Protein	3.75 <sup>a</sup>	3.74 <sup>a</sup>	3.59 <sup>b</sup>	0.053	<0.001
Lactose	4.57	4.56	4.57	0.032	NS

<sup>a,b</sup> denotes significant differences between treatments ( $P < 0.05$ ).

( $P < 0.001$ ), ECM ( $P < 0.001$ ) and milk solids ( $P < 0.05$ ). Both PRG and PRGWC had significantly higher milk protein ( $P < 0.001$ ), and PRG also had a significantly higher milk fat concentration ( $P = 0.022$ ), compared to MULTI. Sward type had no effect on milk lactose concentration (Table 1).

**Conclusion:** Sward type significantly affected the milk production and DMI of dairy cows offered zero-grazed forage. These results show that multispecies swards can increase milk production when zero-grazed and offered to cows housed indoors. These swards may provide a means of maintaining milk production while also reducing nitrogen fertiliser use in grazing dairy systems.

**Acknowledgement:** This work was funded by the DAFM and commercial industry.

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### 118. Dietary starch concentration influences milk protein via the phenylalanine and tyrosine metabolism in early lactation Holstein Friesian dairy cows

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**Application:** Diets containing high levels of starch alter phenylalanine and tyrosine metabolism and increase milk protein concentration. This information can be used to more accurately formulate diets to alter milk protein content in dairy cows.

**Introduction:** Starch is frequently included in dairy cow rations to fulfil energy and protein requirements. Studies have reported changes in phenylalanine metabolism as a result of varying dietary starch concentrations in lactating Holstein-Friesian dairy cows [1] but subsequent effects on milk protein have yet to be determined. This study aimed to examine the metabolic changes in plasma associated with varying dietary starch concentration in early lactation Holstein-Friesian dairy cows.

**Materials and methods:** Sixteen Holstein-Friesian dairy cows that were 38.9 ( $\pm$  3.4) days in milk and yielding 42.6 ( $\pm$  1.6) kg/d were blocked according to milk yield in the week prior to being allocated to 1 of 2 dietary treatments. Cows remained on study for a period of 98 days, and the 2 diets were formulated to contain dietary starch concentrations of either 231 (HS) or 103 g/kg of DM (LS). Plasma samples collected

during weeks 0, 1, 2, 4, 6, 8 and 14 were assessed using flow infusion electrospray high resolution mass spectrometry on a Q Exactive hybrid quadrupole-Orbitrap mass spectrometer for high throughput, sensitive, non-targeted metabolite fingerprinting. Variables of importance for the projection (VIP) scores ( $>$  1) in Partial Least Squares – Discriminant Analyses (PLS-DA) were used to indicate mass-ion ( $m/z$ ) values which discriminated between the classes. Performance parameters, including milk composition, were measured by t.test repeated measures using Genstat (v18).

**Results:** PLS-DA and hierarchical cluster analysis of the data discriminated between treatments. In total, 43 of these discriminatory metabolites were identified using the mummichog algorithm within MetaboAnalyst 4.0. Metabolite set enrichment analysis (MSEA) using over representation analysis demonstrated the significant effects ( $P < 0.05$ ) of dietary starch on phenylalanine and tyrosine metabolism (Figs. 1 and 2). The mean milk protein concentration of cattle on treatment HS was 3.07 g/kg ( $P < 0.05$ ) higher than the LS treatment, with mean concentrations of 32.51 and 29.44 g/kg, respectively. No differences in other performance parameters ( $P > 0.05$ ) were observed.

**Conclusion:** Feeding high starch diets decreased concentrations of multiple metabolites relating to phenylalanine and tyrosine metabolism, and was positively correlated with milk protein concentration. This suggests that phenylalanine metabolism is altered due to low amino acid supply within the low starch diets.

**Acknowledgment:** The authors gratefully acknowledge funding from KESS 2 and the AHDB.

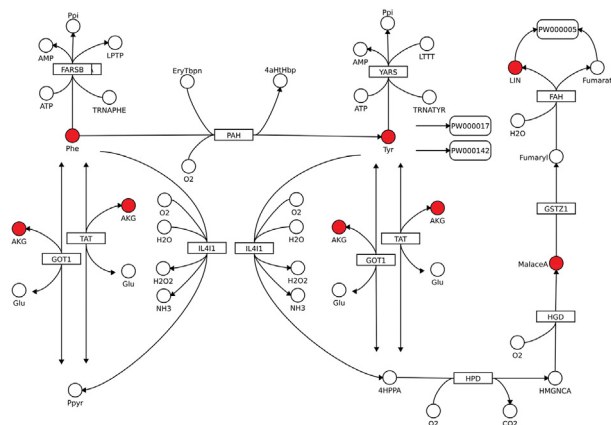


Fig. 1. MSEA demonstrates the effect of starch concentrations on the phenylalanine and tyrosine metabolism. Significantly affected metabolites (VIP score  $>$  1) are highlighted in red.

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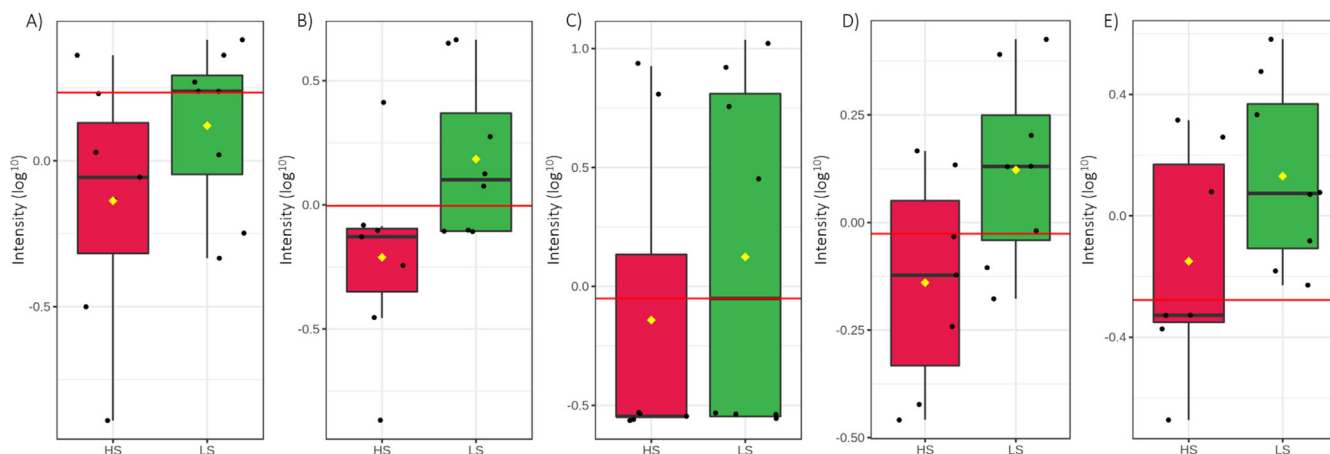


Fig. 2. Boxplots showing the effect of HS (red) and LS (green) starch diets on A) acetoacetic acid, B) maleylacetoacetic acid, C) oxoglutaric acid D) phenylalanine and E) tyrosine.

### 119. The impact of farm area fragmentation and stocking rate on the grazing platform on the length of the grazing season, milk and herbage production in pasture-based dairy systems

Friederike Fenger<sup>a,b</sup>, Imelda Casey<sup>b</sup>, James Humphreys<sup>a</sup>

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**Application:** Fragmented pasture-based systems can be managed without a loss in productivity up to a grazing platform stocking rate of 4 cows/ha when the non-grazing platform land is used for silage production to fill feed deficits during the grazing season.

**Introduction:** The majority of Irish farms are fragmented and the grazing platform; the land accessible with grazing dairy cows, represents approximately 60% of the total farm area. On a fragmented farm with a moderate overall farm stocking rate, there is typically a much higher stocking rate on the grazing platform (GPSR). The objective of this study was to determine how GPSR affects pasture production, milk production, the length of the grazing season and components of the diet when silage produced on outside parcels is used to fill feed deficits in the diet of lactating dairy cows during the grazing season.

**Materials and methods:** The experiment was conducted over three years between 2017 and 2019 at Solohead Research Farm (52°51'N, 08°21'W). There were four grazing systems, each with an overall stocking rate of 2.5 cows/ha but differing in the proportion of area available as grazing platform (100%, 82%, 70% and 61%) resulting in four different GPSRs of (GP25) 2.5 (GP30) 3.0 (GP35) 3.5 and (GP40) 4.0 cows/ha, respectively. The grazing platform was available for grazing and grass-silage

production. The remaining area (outside parcels) was used solely for grass-silage production. The experimental area was divided into six blocks. One paddock from each block was randomly assigned to each system. All systems were compact spring-calving. Each spring cows (24 per system) were blocked based on lactation number and calving date before one cow from each block was randomly assigned to each herd. Nitrogen fertiliser application (280 kg N/ha, all paddocks), concentrate supplementation and target post-grazing height (4 cm) was consistent across systems. Differences between systems were analysed in PROC MIXED (SAS 9.4) with system and year as fixed effect and block as random effect. Paddocks were experimental units for herbage measurements, individual cows for milk production variables.

**Results:** Total herbage production was not different between systems (15.3 ± 0.52 t DM/ha,  $P = 0.98$ ). With higher GPSR less silage was harvested from the grazing platform but more from the overall system area (incl. outside parcels) ( $P < 0.01$ ). The difference in days at pasture and silage intake between systems (Table 1) was mainly caused by feed deficits between August and November. Milk production was not affected by grazing system (Table 1).

**Conclusions:** Similar milk production per cow was achieved within the range of GPSR tested albeit a higher proportion of silage in the diet and fewer days at pasture with increasing GPSR. The majority of supplemental silage in the higher stocked systems was fed in the second half of lactation where dietary requirements of dairy cows are lower than in early lactation.

**Acknowledgement:** This work was supported by the Walsh Scholarship Scheme.

Table 1  
Effect of grazing system on three-year mean days at pasture, silage intake and milk production.

Item	Grazing system (GS)				SEM	Probability		
	GP25	GP30	GP35	GP40		GS	Year	GS × Year
Days at pasture (days/cow)	243 <sup>a</sup>	232 <sup>b</sup>	216 <sup>c</sup>	205 <sup>d</sup>	1.52	<0.001	<0.001	<0.001
Silage intake (kg/cow)	1497 <sup>d</sup>	1674 <sup>c</sup>	1894 <sup>b</sup>	2066 <sup>a</sup>	20.4	<0.001	<0.001	<0.01
Milk yield (kg/cow)	5916	5945	6000	5803	159.2	0.40	0.43	0.03
Yield of milk fat and protein (kg/cow)	487	490	492	479	13.6	0.54	0.19	0.12

SEM = standard error of grazing system means<sup>a-d</sup> Mean values in the same row with different superscripts differ between grazing systems ( $P < 0.05$ ).

## 120. Short-term feed restriction and re-feeding alters rumen metabolism and performance of high yielding dairy cows fed different concentrate patterns and either with or without a live yeast

Catherine Johnson<sup>a</sup>, Tim Snelling<sup>a</sup>, James Huntington<sup>a</sup>, Jules Taylor-Pickard<sup>b</sup>, Helen Warren<sup>b</sup>, Liam Sinclair<sup>a</sup>

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**Application** Short term periods of feed restriction reduce performance in dairy cows, therefore management strategies should be put in place to avoid occurrence on farm.

**Introduction:** Short-term feed restriction (FR) occurs on over 1/3 of dairy farms in the UK [1], due to insufficient allocation or restricted access to feed [2]. Upon re-feeding cows can overeat, increasing concentrate intake, enhancing the risk subacute ruminal acidosis (SARA); 58% of UK dairy herds have shown evidence of significant diet selection [1], potentially further increasing negative effects of FR. Active dry yeasts can scavenge oxygen and reduce lactic acid concentration in the rumen. The objective was to determine the effect of short-term FR and re-feeding on rumen metabolism and performance in dairy cows fed different concentrate patterns either with or without a live yeast.

**Materials and methods:** Four multiparous Holstein-Friesian dairy cows fitted with rumen cannulas were used in a 2x2 factorial Latin square designed study, with four periods each lasting 35 days. All cows were fed a partial mixed ration (PMR) containing grass and maize silage with forage: concentrate ratio of 60:40. An additional 4 kg concentrate/cow/d were fed as an even pattern (E; 1 kg every 4 h between 06:00 and 17:00) or uneven pattern of allocation (U; 4 kg as one meal at 0600), diets were unsupplemented (-) or supplemented (+) with Yea-Sacc® (Alltech Inc., USA) at the rate of 1 g/cow/day. Baseline measurements

for DMI, milk yield, rumen pH, and VFA were taken on day 30. On day 31 cows received 75% of previously recorded PMR intake. On day 32 PMR was reintroduced at 125% of previously recorded intake (recovery day 1), performance and rumen pH were recorded on day 33 (recovery day 2). All data were analysed using GenStat 18.1 (VSN International Ltd, Oxford, UK), as a 2x2 factorial Latin Square design with main effects of time (days), pattern of concentrate allocation, yeast supplementation and their interaction.

**Results:** Pattern of concentrate allocation and yeast supplementation had no effect ( $P > 0.05$ ) on performance or rumen metabolism, and only the effects of time are presented. Relative to the baseline, DMI was 5 kg lower ( $P < 0.001$ ) during FR, and 5.1 kg higher on rec d1, returning to baseline on rec d2 (Table 1). Milk yield decreased ( $P < 0.001$ ) by 3.1 kg/d on rec d1 and was 1.7 kg/d below baseline on rec d2. Mean rumen pH was lower ( $P < 0.001$ ) during rec d2 by pH 0.088 compared to baseline, FR had no effect ( $P > 0.05$ ) on total VFA concentration.

**Conclusion:** Following a period of feed restriction, re-feeding increased dry matter intake, however, milk yield and rumen pH decreased, VFA concentration was unchanged. Pattern of concentrate allocation or yeast had no effect on any of the parameters.

**Acknowledgement:** The authors gratefully acknowledge Alltech for funding this study.

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Table 1  
Mean performance and rumen metabolism in dairy cows during a short term feed restriction period.

	Baseline	FR	Rec d1	Rec d2	SED	P-value
DMI, kg/d	22.1 <sup>a</sup>	17.1 <sup>b</sup>	27.2 <sup>c</sup>	22.2 <sup>a</sup>	0.44	<0.001
Milk yield, kg/d	39.2 <sup>a</sup>	38.5 <sup>ac</sup>	36.1 <sup>b</sup>	37.7 <sup>c</sup>	0.49	<0.001
Mean rumen pH	5.82 <sup>a</sup>	–	5.84 <sup>a</sup>	5.73 <sup>b</sup>	0.023	<0.001
Total VFA concentration, $\mu\text{M}$	122.8 <sup>a</sup>	–	122.5 <sup>b</sup>	–	3.78	0.921

DMI = Dry matter intake, VFA = Volatile fatty acids, FR = Feed restriction, Rec d1 = recovery day 1, Rec d2 = Recovery day 2<sup>a, b, c</sup> Means in rows with different superscript letters differ significantly

## Invited Speaker Summary

### **121. Future dairy – The role of agri tech in optimising cow health and welfare**

Duncan Forbes

Agri-EPI Centre, UK

The presentation will provide a snapshot of current and emerging technology that is being developed to optimise dairy cow health and welfare.

The Agri-EPI Centre has invested in state-of-the-art facilities at its South West Dairy Development Centre where it has established a 180 cow dairy unit with high levels of automation and robotics. It provides the platform to research, develop, test and demonstrate the benefits and potential of emerging agri tech to improve cow welfare and productivity.

## 122. Preliminary study of effect of sample site on hair cortisol concentration in Holstein-Friesian Dairy Cows

Hannah Wicks<sup>a</sup>, Rachel Abrahall<sup>b</sup>, Abduselam Mohammed<sup>c</sup>, Paul Ward<sup>b</sup>

<sup>a</sup>Trichotech, Boscastle, Cornwall, UK

<sup>b</sup>Duchy College, Rural Business School, Callington, Cornwall, UK

<sup>c</sup>Cansford Laboratories, Cardiff, Wales, UK

**Application:** The creation of a method to measure cortisol levels in cattle to provide an objective measurement of stress for use in future work assessing cortisol and stress levels in cattle. [Fig 1.](#)

**Introduction:** With the interest in ensuring milk is produced as ‘naturally’ as possible and the advent of ‘free-range’ milk, an objective method of measuring stress in cattle maybe useful. Ultimately it may be possible to prove cows are not stressed, and/or pick up on chronic stress that may affect everything from fertility to health to milk production. Several questions need to be assessed before this point is reached. (Other studies are looking at the use and usefulness of hair cortisol in humans.) Sharma *et al* (2019) and Nedic *et al* (2017) have shown variations in hair cortisol concentrations with, stress levels, cattle breed and milk production levels. Hair cortisol gives an overall picture of the situation, which is not affected by acute factors (stress) on the day of sampling. Hair samples are relatively quick and easy to obtain and are envisaged to be used in conjunction with other sample types. Before assessing whether cortisol can be used as an early warning marker for fertility and health issues (e.g. Do cows with fertility or health problems have raised cortisol concentrations?), further studies will assess how cortisol levels change with lactation number, and actual and potential milk production.

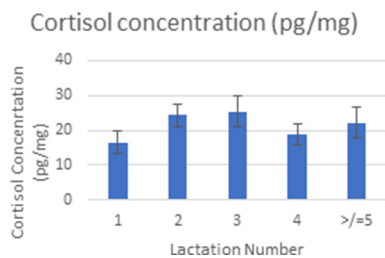


Fig. 1. Hair Cortisol Concentrations (pg/mg) by Lactation number

Table 1  
Hair cortisol concentrations (pg/mg) for sample sites.

Variable	Cortisol (pg/mg)	sem
Sample Site	P < 0.001	
Neck (n = 32)	32.69a	2.699
Side (n = 30)	15.50b	2.106
Rump (n = 30)	16.00b	2.640

The objective was to assess whether sample site affects concentration of cortisol in hair samples, before larger sample numbers were collected from additional cows.

**Method:** 32 Holstein-Friesian dairy cattle, randomly selected from the Duchy College dairy herd (at Stoke Climsland), were sampled in two groups January (n = 12) and July 2020 (n = 20). Hair samples were taken from 3 sites on each cow, the neck, the side and the rump; using a standardised set of battery-operated clippers. Hair from the clip area (2x2 cm) was placed into individual envelopes. Average hair length was 1.23cm (SD 0.516). Cortisol concentrations were assessed by Cansford Laboratories using the LC-MS/MS technique (Binz *et al* 2016). An analysis of variance was used to assess the importance of sample site, with factors of lactation number and sample group included.

**Results:** Overall mean cortisol concentration was 21.64 pg/mg (+/- 1.663, n = 92), values ranged from 3 to 77pg/mg. Cortisol concentration (Table 1) was significantly (P < 0.001) higher in neck samples (32.69 pg/mg +/- 2.699) compared to side (15.50pg/mg +/- 2.106) or rump (16.00pg/mg +/- 2.640). Additionally, results showed first lactation heifers had lower concentrations of cortisol (16.58 pg/mg +/- 3.22) when compared to second (24.33pg/mg +/- 3.29) and third (25.45pg/mg +/- 4.46) lactation cows. There were no significant interactions between sample site, lactation number or sample group.

**Conclusion:** Sampling site is important when using hair to measure cortisol concentrations in dairy cattle.

### Uncited references

[1,2,3].

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### 123. Impact of adopting selective dry-cow therapy on cow performance and udder health

Anna Lavery<sup>a</sup>, Aimee Craig<sup>a</sup>, Alan Gordon<sup>b</sup>, Conrad Ferris<sup>a</sup>

<sup>a</sup> Agri-Food and Biosciences Institute, Hillsborough, UK

<sup>b</sup> Agri-Food and Biosciences Institute, Belfast, UK

**Application:** Selective dry-cow therapy can offer dairy farms the opportunity to reduce antibiotic use, without negative effects on productivity or cow health.

**Introduction:** While the use of intramammary antibiotics is standard practice on most farms, improvements in milking hygiene and cow genetics have resulted in a decrease in herd somatic cells counts (SCC). This may provide an opportunity for many farms to adopt selective dry-cow therapy (SDCT), which involves limiting antibiotic treatment at drying-off to cows considered 'high risk'. This study examines the effects of adopting SDCT at drying-off on cow productivity and udder health during the first 5 months of the subsequent lactation.

**Materials and Methods:** This study was conducted within the dairy herd at AFBI Hillsborough during two successive years, involving 223 cows in year 1 and 172 cows in year 2. Cows were allocated to treatments at drying-off based upon their 'risk' category. Cows were considered 'high risk' (n = 110) if they had a SCC > 200,000 cells/ml, or at least one case of mastitis during the three months prior to drying-off. These 'high risk' cows were treated with Conventional dry-cow therapy (DCT, intramammary antibiotics and teat sealant). Cows were identified as 'low risk', if they had a SCC < 200,000 cell/ml and no cases of mastitis during the

three months prior to dry-off. 'Low risk' cows were allocated to one of two treatments at drying-off; Conventional DCT (n = 130), or SDCT (teat sealant only, n = 155). Cows were monitored for the first 5 months post-calving with daily milk yield, monthly milk composition (fat, protein and SCC) and mastitis incidence recorded. Milk yield and milk composition were analysed using linear mixed model methodology using REML estimation, while mastitis incidence was analysed using generalised linear mixed model methodology using binomial distribution and logit function. In all analyses, cow and year were included as random effects, treatment as a fixed effect in the model. Data were analysed using Genstat (20th Edition).

**Results:** Mean lactation number was greater for the 'high risk' group compared to the 'low risk' groups (4.0 vs. 3.0, respectively). Neither milk yield nor milk fat or protein content were affected by drying-off approach adopted (Table 1). 'High risk' cows had a higher SCC than 'low risk' cows during the first five milk recordings post-calving (P < 0.001). Drying-off treatment had a significant effect on mastitis incidence (P < 0.001). On average, 39% of 'high risk' cows would develop mastitis, while 'low risk' cows dried-off using either SDCT or conventional DCT had a similar incidence of mastitis (12% and 17%, respectively).

**Conclusion:** Drying-off 'low risk' cows without antibiotics did not negatively impact cow performance or udder health.

**Acknowledgments:** This work was co-funded by DAERA RCF, and carried out in partnership with Agrisearch, Animal Health and Welfare Northern Ireland, LMC and Farm Vet systems.

Table 1  
Mean daily milk yield, monthly milk composition and mastitis incidence over the first five milk recordings for 'high risk' cows which received Conventional DCT, and for 'low risk' cows subject to either SDCT or Conventional DCT.

	High risk (Conventional DCT)	Low risk (SDCT)	Low risk (Conventional DCT)	SED	P-value
Milk yield (kg/day)	38.9	37.9	39.0	0.62	0.123
Fat (%)	3.88	3.93	3.91	0.054	0.593
Protein (%)	3.33	3.35	3.35	0.021	0.477
SCC ('000/ml)	285	84	81	-	-
SCC <sub>log10</sub> ('000/ml)	1.77 <sup>b</sup>	1.65 <sup>a</sup>	1.57 <sup>a</sup>	0.043	<0.001
Cows with mastitis (%)	39	12	17	-	<0.001

## 124. Quantifying temperament using behavioural testing in adult dairy cattle

Nick Britten, Gareth Pearce

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**Application:** Consistent behavioural variation between individuals has been widely used to predict factors such as overall mortality, disease susceptibility and social interactions in a variety of species. This study evaluated the validity of using existing frameworks for characterising individual behavioural variation (temperament) in adult dairy cattle using behavioural data.

**Introduction:** Temperament has been characterised across many animal species as either continuous 'traits' or categorical 'styles'. [2] proposed five traits-based attributes: shyness/boldness, exploration/avoidance, sociability, aggression and activity whereas [1] categorised individuals as either 'active' or 'passive' temperaments in the coping-style framework. Standardised behavioural tests have been used in cattle to evaluate temperament but it is not clear which of these frameworks is most effective in interpreting and combining test outcomes in order to categorise specific temperament types in cattle.

**Materials and Methods:** Standardised temperament tests (runway test, crush test and flight speed, novel arena test, startle test, novel object test and human approach test) were carried out with each of 67 lactating Holstein cows over 10 weeks. 595 aggressive interactions (displacement, butt, push and threat) were recorded at the feed face over the 2 hour period following feed delivery on 31 days. Scaled principal component analysis was used to extract traits from the behavioural observations using an eigenvalue of  $4/\sqrt{3}$  to retain components and a k-means cluster analysis was used to aggregate cows into categories of temperament across all test results. Both 2 and 3 group analysis was conducted to investigate the possibility of the 'intermediate' coping style. All data were analysed using RStudio 1.3.1093.

**Results:** Six principal components representing the traits-based attributes of activity, sociability, avoidance, exploration, shyness-boldness and vocalisation explained 51.89% of total variation. As shown in Figs. 1 and 2, there were significant differences between groups of cattle in activity ( $P < 0.001$ ) and sociability ( $P < 0.001$ ) but not in any other

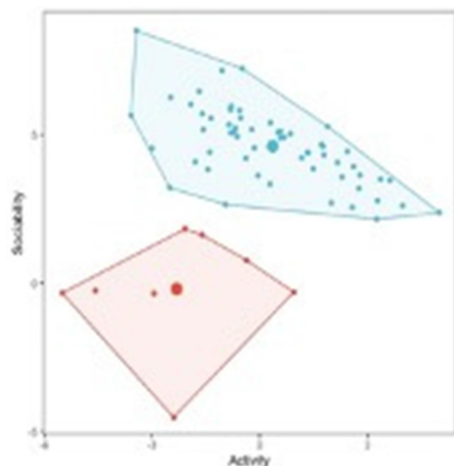


Fig. 1. Two group cluster analysis (right of next paragraph)

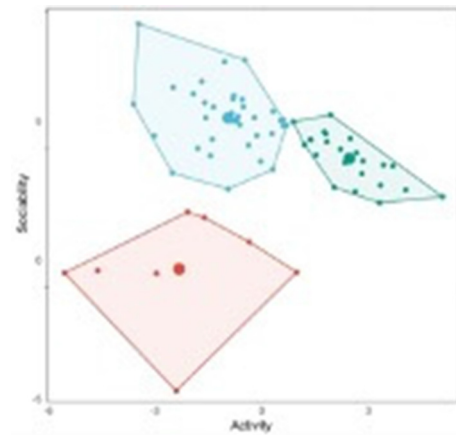


Fig. 2. Three group cluster analysis (immediately below Figure 2 to right of text)

behavioural trait in both the 2 and 3 group cluster analysis. The two clusters could be identified as 1. a high activity - high sociability 'active' temperament type and 2. a low activity - low sociable 'passive' temperament type. The 3 group analysis showed two similar groups plus a third high sociability - low activity 'intermediate' temperament type.

**Conclusion:** Standardised behavioural testing can be used to both quantify and categorise dairy cow temperament and combining the outcomes of these tests using statistical methods such as scaled principal component and k-means cluster analyses supports the use of both the 'traits'-based and the 'styles'-based frameworks for classifying animal temperament.

**Acknowledgments:** The authors gratefully acknowledge funding from BBSRC.

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## 125. Temperament, health and fertility outcomes in dairy cattle

Nick Britten, Gareth Pearce

University of Cambridge, Cambridge, UK

**Application:** Individual differences in dairy cow temperament are associated with differences in fertility, lameness and somatic cell counts. This offers the potential for improving dairy cow health through selection and management changes based on individual temperament measurement.

**Introduction:** Differences in individual temperament in cattle can be quantified using standardised behavioural tests and statistical dimension reduction techniques [2]. These behavioural differences are associated with variations in physiological stress responses that impact on fertility and health outcomes [1]. This study examined the relationships between temperament and measures of reproductive performance, lameness and udder health in commercial dairy cattle.

**Materials and methods:** Standardised temperament tests (runway test, crush test and flight speed, novel arena test, startle test, novel object test and human approach test) were carried out with 130 lactating Holstein cows over 10 weeks. Principal component analysis was used to combine test results. Monthly milk recording somatic cell counts (SCC), mobility scores (MS), days post-partum (DPP) and number of services (NS) to conception were recorded for each animal. A threshold eigenvector of  $4/\sqrt{3}$  was used to determine which principal components to retain in a multivariate model, simplified using stepwise selection based on the Akaike Information Criterion. This process was used to quantify the relationship between  $SCC > 200\ 000$  cells/ml,  $MS \geq 2$ , DPP and NS and retained principal components. Age was included in all models and days in milk in models of SCC and MS. All analysis was conducted using RStudio version 1.3.1093.

**Results:** Five principal components identified as activity, sociability, neophobia, exploration and shyness-boldness met the threshold eigenvector for retention in the models, explaining 44.13% of the sample variance. Highly neophobic cows showed significantly less lameness during the study period ( $P = 0.018$ ) (Fig. 1) but took significantly longer ( $P = 0.025$ ) (Fig. 2) and required significantly more services to conceive ( $P = 0.01$ ). Bolder cows took significantly fewer services to conceive ( $P = 0.007$ ) and tended to conceive earlier ( $P = 0.068$ ). Highly exploratory cows had significantly more SCCs above 200,000 cells/ml ( $P = 0.009$ ) (Fig. 3) as did highly social cows ( $P = 0.047$ ).

**Conclusions:** Temperament differences in commercial lactating dairy cattle are associated with significant variations in important fertility and health parameters. This suggest that selection and management of dairy cattle based on variations in individual temperament offers significant potential for improvement in economically important characteristics.

**Acknowledgments:** The authors gratefully acknowledge funding from BBSRC.

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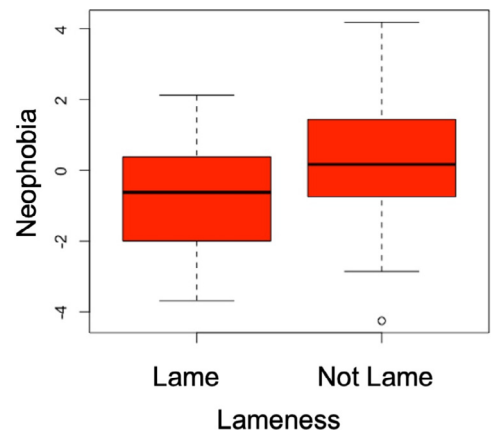


Fig. 1. Boxplot of Lameness by Neophobia.

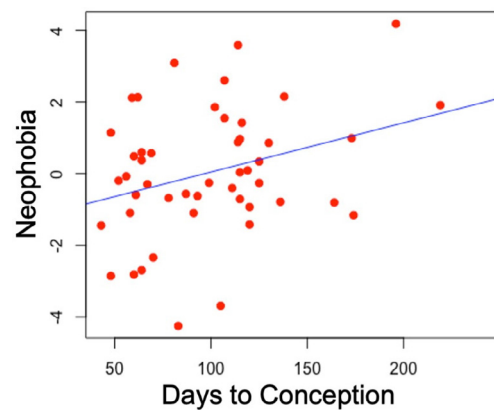


Fig. 2. Plot of Days to conception by Neophobia

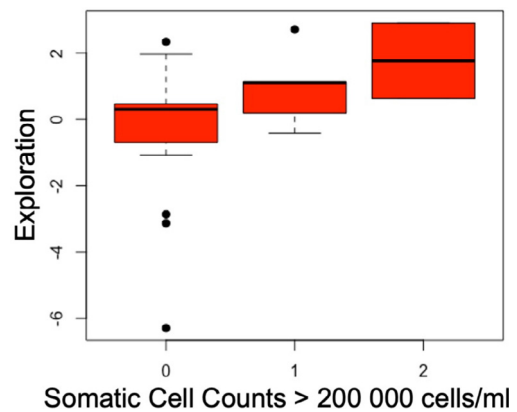


Fig. 3. Plot of Somatic Cell Counts by Exploration (Ideally to all go side by side)

## 126. Corroboration of rumination duration between the Lely collar and the Rumiwatch sensor

Zelin Li<sup>a</sup>, Long Cheng<sup>a</sup>, Brendan Cullen<sup>b</sup>

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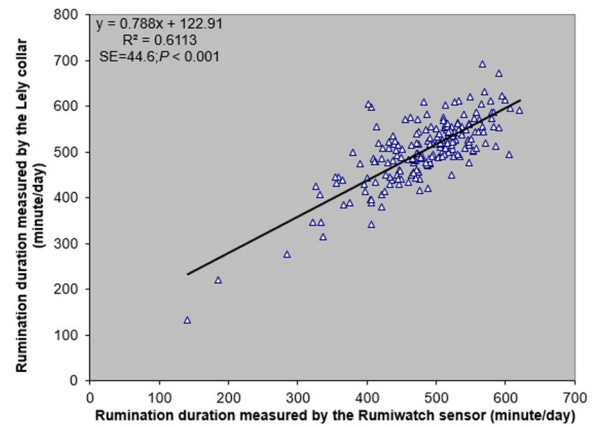
**Application:** The Lely collars can substitute the high-precision sensor (the RumiWatch sensor) in commercial farms to monitor the rumen health and functions of dairy cows.

**Introduction:** With the development of sensor technologies, many sensors can be used to measure animal behaviours. Behaviour data can be used to quantify the grazing intake of dairy cows, monitor the health status, and assist farmers in achieving better management in the farms [1,2]. The rumination duration is one of the important parameters, reflecting the rumen function and feed intake. Although some sensors reached high accuracy (CCC = 1.00) in measuring rumination behaviours with visual observation [2], special wearable devices may also affect normal dairy cows' normal behaviours. As a standard wearable device in the automatic milking system, Lely collar (Lely Industries NV, Maastrand, The Netherlands) can measure the rumination duration with minimised interference. Therefore, the objective of this study is corroborating the Lely collar and the RumiWatch sensor for measuring rumination duration.

**Materials and methods:** Twenty Holstein cows were mounted with the RumiWatch sensors and Lely collars for 14 days to provide a dataset. The concordance correlation coefficient (CCC) between Lely collar and the RumiWatch sensor was analysed by Genstat 18<sup>th</sup> (VSN International; Hemel Hempstead, United Kingdom).

**Results:** Lely collar has achieved a high concordance (CCC = 0.75;  $R^2 = 0.61$ ;  $C_b = 0.96$ ; SE = 44.6;  $P < 0.001$ ) with RumiWatch sensor (RWS; Itin + Hoch GmbH, Liestal, Switzerland) in measuring rumination duration (Fig. 1).

**Conclusion:** The Lely collar applied within the Lely automatic milking system can be used as a daily wearing device to measure rumination duration with reasonable accuracy and minimal interference. The recording rumination data can show the rumen health state and



**Figure 1.** The correlation between the Rumiwatch sensor and Lely collar for the rumination duration. The prediction equation of the rumination duration from the Lely collar is rumination duration<sub>Lely collar</sub> (minute/day) = 0.788 × rumination duration<sub>Rumiwatch sensor</sub> (minute/day) + 122.91 (SE = 44.6;  $P < 0.001$ ).

potentially predict the grazing intake. This study indicated that the Lely collar could be commercially used in the farms to monitor rumen health and functions by checking daily updated data.

**Acknowledgments:** We would also like to extend our gratitude to Mr. Nathan Anderson and Miss. Jess Reeks from Dookie dairy farm, The University of Melbourne for their help in conducting this study.

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## 127. Dairy cow motivation for access to open lying space

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**Application:** Cows are motivated to access an open lying space, which could inform the design of future cow housing.

**Introduction:** With dairy cows being housed longer indoors [1], it is important to ensure that cow lying comfort and welfare are not compromised. A previous study showed that when choosing where to lie down, an open lying space is more important to cows than their preferred lying surface [2]. This study aims to quantify that preference, measuring cow motivation for an open lying space using two different surface types, straw and mattress, with walking distance as a measure of motivation Table 1.

**Materials and Methods:** Thirty Holstein-Friesian dairy cows were allocated to one of six experimental periods ( $n = 5 \times 6$ ) and housed in a robotic milking unit, given ad libitum feed and water and access to six mattress cubicles with sawdust bedding. Cows were given access to a one-way indoor raceway, at a short distance (~30 m), leading to an open lying area (9 m  $\times$  5 m) of either deep-bedded straw or mattress bedded with sawdust. Following a training period, cows had a 3 day choice period to access the open lying surface via the raceway or lie in the cubicles for 'free'. The raceway distance was change and this was repeated at a medium distance (~67 m) followed by a long distance (~112 m). The raceway was changed back to the short distance and the above protocol repeated for the second open lying surface. Order of surface type was alternated for each experimental period.

Linear mixed effects modelling was used to assess the significance of raceway distance, open lying surface type and individual cow in explaining variations in lying time on the open lying area and in the cubicles.

**Results:** Cow lying time reduced on the open lying surface as walking distance to access it increased ( $P < 0.001$ ), with cows increasing the

Table 1

Average number of hours per day spent lying down on the cubicles and open lying surface during each treatment (open mattress access and straw yard access).

Distance	Lying Time for Open Mattress Access (hr/d $\pm$ SEM)		Lying Time for Straw Yard Access(hr/d $\pm$ SEM)	
	Cubicle	Mattress	Cubicle	Straw Yard
Short	3.2 $\pm$ 0.8	10.3 $\pm$ 0.9	1.4 $\pm$ 0.5	12.4 $\pm$ 0.6
Medium	4.0 $\pm$ 0.8	9.0 $\pm$ 1.0	2.1 $\pm$ 0.6	11.3 $\pm$ 0.9
Long	6.0 $\pm$ 0.9	6.7 $\pm$ 1.0	3.6 $\pm$ 0.8	8.2 $\pm$ 1.0

time they spent lying on the cubicles as this distance increased ( $P < 0.001$ ). Individual cow had no effect on lying time for the open lying area ( $P = 0.8081$ ) or cubicles ( $P = 0.979$ ).

**Conclusion:** Walking distance has an effect on the lying times for the open lying surfaces and the cubicles. As walking distance increases, lying times on the open lying surfaces decrease but increases on the cubicles, indicating a lowering of motivation to access an open lying space with increased walking distance. However, cows are still motivated at the long distance to lie down on the open lying surfaces for an average ~ 7.5hrs per day, highlighting the importance of an open lying space for cows.

**Acknowledgments:** The authors gratefully acknowledge funding from AHDB Dairy.

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## 128. Comparison of behaviour of in-calf Holstein Friesian heifers out-wintered on fodder beet or grass with those housed over winter

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**Application:** Out-wintering dairy replacements offers a low cost alternative for rearing in-calf heifers. However, public perception and heifer welfare are concerns. This study uses activity monitors to assess behaviour of out-wintered in-calf heifers.

**Introduction:** Out-wintering dairy replacement heifers is perceived to be an option that has low variable costs and health benefits for the heifers [2]. Growth rates and subsequent lactation performance are similar to winter-housed heifers [1]. However, inclement weather has implications for heifer welfare and public perception of this practice [3]. Lying behaviours have been shown to indicate cattle welfare [4]. The aim of the present study was to monitor the activity of out-wintered, in-calf Holstein-Friesian heifers.

**Materials and Methods:** The study was conducted at Harper Adams University, Newport, UK from November 2013 to January 2014. In-calf Holstein-Friesian heifers (n = 24; liveweight (mean  $\pm$  sed) 455  $\pm$  7.1 kg; BCS 2.38  $\pm$  0.037) were randomly allocated to three treatments: housed, fed baled grass silage and concentrate (H), out-wintered on grass with baled grass silage (G) or out-wintered on fodder beet with baled grass silage (FB). Groups were balanced for stage of pregnancy and liveweight. Activity monitors (IceTag™, Ice Robotics Ltd, Roslin, UK) were attached to the back right leg of heifers to determine the amount of time they spent standing, lying or active per day during 20 day observation periods. Each heifer was monitored in two alternate periods. The number and frequency of lying bouts were also calculated. Data (means for each 20 day period) were analysed in R (version 4.0.3) with variables fitted by REML using the lme4 package. The model included fixed effect of treatment (H, G, and FB), with heifer and period as random effects. Tukeys tests were performed post hoc.

**Results:** Outwintered heifers (FB and G) spent approximately 3 hours/day less lying than housed heifers, and spent over 2 more hours active

Table 1

Time (mean) spent lying, standing still or active and lying bout number and duration in heifers either outwintered on fodder beet (FB) or grass (G) or housed (H).

Parameter	Treatment			sed	significance
	FB	G	H		
Lying (h / day)	10.3 <sup>b</sup>	10.0 <sup>b</sup>	13.5 <sup>a</sup>	0.41	P < 0.001
Standing (h/day)	5.8 <sup>a</sup>	4.4 <sup>b</sup>	4.5 <sup>b</sup>	0.31	P < 0.001
Active (h/day)	8.0 <sup>a</sup>	8.7 <sup>a</sup>	6.1 <sup>b</sup>	0.36	P < 0.001
Number of lying bouts	7.4 <sup>b</sup>	8.0 <sup>b</sup>	12.7 <sup>a</sup>	0.75	P < 0.001
Duration of lying bouts (mins)	92.0 <sup>a</sup>	90.6 <sup>a</sup>	70.6 <sup>b</sup>	6.13	P = 0.003
ADG (kg)	1.37 <sup>ab</sup>	1.09 <sup>b</sup>	1.39 <sup>a</sup>	0.114	P = 0.023
BCS change	0.13 <sup>ab</sup>	0.06 <sup>b</sup>	0.31 <sup>a</sup>	0.090	P = 0.031

Means within rows with different superscripts are significantly different (p < 0.05); ADG = average dairy gain, BCS = body condition score (1 to 5 scale).

(Table 1). G and H heifers spent less time standing still than FB heifers. H heifers had more lying bouts of shorter duration than outwintered heifers. ADG and BCS change were greatest in H heifers

**Conclusions:** The present study demonstrates that out-wintering in-calf heifers alters their behaviour compared to housed heifers. However, behaviours were within the expected range for cattle. Further work will demonstrate the effects of weather on the behaviours of outwintered heifers.

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### 129. Altered rumen microbiota profiles in cattle infected with and vaccinated against the parasitic nematode *Ostertagia ostertagi*

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**Application:** The comprehensive understanding of host-parasite-microbiome interactions is paramount for the development of infection-mitigation strategies against *Ostertagia ostertagi*, one of the major causes for economic losses in cattle production systems

**Introduction:** *O.ostertagi* is a cattle-parasitic nematode that causes impaired gastrointestinal function, (e.g. inappetence) at subclinical levels of infection. Immunity to *O.ostertagi* takes long to develop, with cattle remaining susceptible even after long grazing periods. In developed production systems this parasite is controlled using anthelmintics, and vaccines are being developed. Parasite- and vaccine-induced alterations to the gastrointestinal microbiota, implications on the infection and on the effective host immune response development are not yet clear (Li *et al.*, 2011). The objective of this study was to evaluate the impact of *O.ostertagi* parasitism at subclinical levels and of a native *O.ostertagi* vaccine on the microbiota of dairy cattle.

**Materials and methods:** An experiment was performed to investigate the effects of an experimental vaccine based on parasite excretory-secretory material against *O.ostertagi*. Animals (4–5 months old) were allocated to treatment groups (balanced for body weight) receiving either vaccine or adjuvant-only, and later challenged with trickle infection; an additional group acted as control (unvaccinated and unchallenged). From this experiment, a subset of animals was chosen for whole metagenome sequencing of post-mortem rumen digesta samples, consisting of four treatment groups of four calves each: UNF – unvaccinated, unchal-

lenged; CHE and CLE – adjuvant-only, challenged, selected based on their high and low average faecal egg count (cFEC), respectively, upon infection; and VAC – vaccinated, challenged. Centred-log-ratio-transformed abundances of 899 microbial genera of CHE, CLE and VAC were compared against UNF using Partial Least Squares-Discriminant Analyses. Significantly discriminative microbes were identified by variable importance in projection scores ( $VIP > 1$ ).

**Results:** PLS-DA identified 314, 294 and 330 microbes important for the discrimination of CLE, CHE, and VAC, respectively, from UNF (Fig. 1). A set of 86 microbes discriminated UNF from all other groups, e.g. *Butyrivibrio* (butyrate-producer) was always at least 16% more abundant, whereas *Fimbriimonas* (potential pathogen) was always at least 39% less abundant in UNF than in any other groups. Additionally, 82, 109 and 121 microbes exclusively discriminated UNF from CHE, CLE, or VAC, respectively. For example, CHE showed 29% more *Leptospira* (potential pathogen) and 26% less *Fibrobacter* than UNF; CLE had 41% more *Lysobacter* (reported association to feed inefficiency) and 11% less *Megasphaera* than UNF, and VAC had 41% more *Bordetella* (potential pathogen) and 27% fewer *Prevotella* than UNF animals.

**Conclusion:** Our results highlight that the infection and vaccination affected the rumen microbiota profiles; this impact differed according to the pathogen burden of the animals. This suggests that microbiota profiles are potentially suitable to evaluate infection level and vaccine success, underlining the need for consideration of the rumen microbiota in the development of efficient strategies against *O.ostertagi* infection.

**Acknowledgments:** The project was funded by the Scottish Government and BBSRC.

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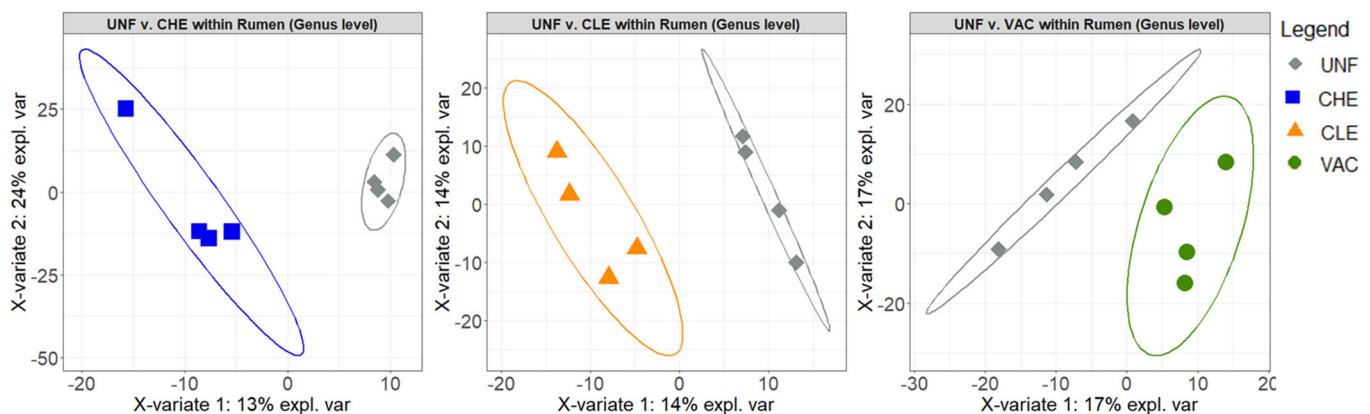


Fig. 1. Partial Least Squares-Discriminant Analyses plots discriminating high (CHE), low (CLE) egg count and vaccinated (VAC) from uninfected (UNF). Classification error was 0% for all models.

### 130. Prediction of subclinical ketosis based on automated milking systems (AMS) in dairy cattle

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**Application:** Within automated milking systems (AMS), milk yields and milking intervals may be used to predict ketosis.

**Introduction:** For herd management, utilizing different available information sources is valuable with regard to metabolic disorders. AMS have the advantage of providing daily information on dairy performance traits. This abundance of data in conjunction with other information may be used for health alerts. The aim of the present work was to derive a model for ketosis prediction. For this purpose, daily milk yields and milking intervals and the results of keto-tests were used from AMS farms.

**Materials and Methods:** In 2014 and within the project “Efficient Cow”, ketosis was diagnosed using ELANCO's milk test, which measures  $\beta$ -hydroxybutyrate (BHB). The test was performed twice during early lac-

Table 1

Considered effects, estimates for  $\text{logit}(p_i)$  with  $p_i$  as the probability for diseased and p-values (pre-analysis dataset).

Effect	Estimate	P-value
Intercept	1.9901	0.110
Lactation no. 1	-0.5818	0.035 <sup>1</sup>
Lactation no. 2	0.0545	
Lactation no. 3	0.1394	
Lactation no. 4	-0.4314	
Days in milk	-0.1775	0.097
Milking interval (min)	0.0014	0.269
Milk Index 1 <sup>1</sup>	-1.9386	< 0.001
Milk Index 2 <sup>1</sup>	13.6505	< 0.001

<sup>1</sup> P-value for the effect lactation number, <sup>2</sup> Milk Index 1 and 2 represent the Cow and Mate Index (Paudyal et al., 2018), respectively.

Table 2

Sensitivity (SE) and specificity (SP) as well as area under the curve (AUC) for training and test datasets and two thresholds for classification “diseased”.

Threshold	Training data sets		Test data sets	
	0.5	0.3	0.5	0.3
SE (%) (Min – Max) <sup>1</sup>	54.1 (31.8–63.8)	84.2 (69.7–91.5)	51.5 (12.5–92.9)	75.2 (12.5–100.0)
SP (%) (Min – Max) <sup>1</sup>	77.1 (71.3–89.8)	44.6 (29.8–59.1)	73.2 (59.1–91.9)	32.1 (7.1–67.6)
AUC (Min – Max) <sup>1</sup>	0.73 (0.72–0.75)	0.64 (0.52–0.78)		

<sup>1</sup> Min = Minimum, Max = Maximum.

tation. For this analysis, data from 11 farms with AMS and 372 cows with results from the first keto-test were used. A BHB content of  $\geq 100 \mu\text{mol/l}$  resulted in the classification “diseased”. For the prediction of the ketosis risk, the SAS procedure logistic was used. Effects considered in the model were based on a pre-analysis ( $p < 0.30$ ). The final model included the fixed effect lactation number (1 to 5 + ) and the covariates days in milk, milking interval at keto-test day and two milk yield indices. The indices followed those of [1] for rumination time. For prediction and calculation of sensitivities and specificities, the dataset was repeatedly split into training and test dataset (9 and 2 farms). Further, two thresholds regarding the predicted disease risk (0.5 and 0.3) and the assignment “diseased” were applied.

**Results:** In total, 36.9% of the cows were classified as diseased (within farm ranging from 0.0 to 76.2%). Estimates and p-values for the pre-analysis are illustrated in Table 1.

In Table 2, results are shown for the training and test datasets. With the ketosis risk threshold of 0.5, the average sensitivity was only slightly above 50% in the test dataset ranging from 12.5 to 92.5% depending on combination of farms. Lowering the threshold to 0.3 markedly increased the sensitivity ( $> 75\%$ ) but resulted in a notable drop in specificity (73.2 vs. 32.1%) and would thus result in a high number of false positives.

**Conclusion:** Information from AMS systems have the potential of being utilized as predictors for ketosis. However, finding the balance between correctly detecting diseased animals and a reasonable false positive rate is challenging. However, the results could help target potential risk animals.

**Acknowledgments:** Funding by BMLRT, the Provinces of Austria, ZAR and its organisations (Project Efficient Cow) and BMVIT, BMDW and the Provinces of Lower Austria and Vienna (COMET Project D4Dairy, handled by FFG) is acknowledged.

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### 131. Effect of mobility at parturition on subsequent mobility, milk yield, feed intake and feed efficiency of lactating dairy cattle

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**Application:** Half of cows had imperfect or impaired mobility at parturition, which persisted throughout almost the whole of early lactation.

**Introduction:** Lameness causes substantial pain for dairy cow welfare [2], which has implications for consumer perception and economic resilience of the dairy industry [1]. However, previous studies have not assessed mobility score at parturition (MSAP) and its effect on subsequent animal performance. This study aimed to assess MSAP, the time cows had suboptimal mobility and its effect on milk yield, dry matter intake (DMI) and feed efficiency. Table 1.

**Materials and methods:** This study was completed by observing 51 Holstein-Friesian dairy cattle (LW 611 ± 6.09 kg) that were selected at random (11,000 L/cow/year) from the University of Nottingham dairy herd according to calving date and allocated according to MSAP to one of three groups; MS: < 0.5, (n = 17), MS: 0.5 to 1 (n = 17) and MS: > 1 (n = 17). Mobility (0 to 4), DMI, body condition score (BCS), milk yield and composition were measured on an individual

animal basis up to 100 d pp. Data was normally distributed and analysed using AVOVA GLM, with differences between means being assessed using Tukey's test, with a confidence interval of 95%.

**Results:** Cows with an impaired MSAP had a greater milk (6.8 kg/d), EC milk and component yields and DMI than cows with good and imperfect MSAP. Cows that had impaired MSAP were older and had completed more parturitions /lactations than cows with a good MSAP. There was no effect of MSAP on the efficiency with which feed was converted into milk yield, protein, fat and solids yield.

**Conclusions:** Cows with impaired MSAP were older that had completed more lactations and had greater milk yield and DMI, but lower BCS gain than cows that had good and imperfect MSAP. However, feed efficiency did not differ between cows that had good, imperfect and impaired MSAP. However, half of the cows had imperfect or impaired MSAP and this continued for 94.5 % (± 3.31) of the first 100 d of lactation.

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Table 1

Mean milk, milk composition and component yield, body condition score (BCS: 0 to 5), feed intake and feed efficiency (± se) between 0 and 100 d postpartum of dairy cattle that had a good (<0.5) imperfect (0.5 and 1) and impaired (2 & 3) mobility at parturition.

	Mobility score (0 to 4) at parturition		P	
	< 0.5	0.5 to 1	> 1	
ECM yield, kg/d <sup>1</sup>	40.5 (0.81) <sup>b</sup>	42.2 (0.74) <sup>b</sup>	47.1 (0.67) <sup>a</sup>	<0.001
Milk fat, kg/d	1.47 (0.022) <sup>b</sup>	1.50 (0.021) <sup>b</sup>	1.70 (0.022) <sup>a</sup>	<0.001
Milk protein, kg/d	1.19 (0.011) <sup>b</sup>	1.26 (0.011) <sup>b</sup>	1.39 (0.012) <sup>a</sup>	<0.001
Milk solids, kg/d <sup>2</sup>	2.66 (0.056) <sup>b</sup>	2.76 (0.052) <sup>b</sup>	3.09 (0.047) <sup>a</sup>	<0.001
Parity, No.	1.75 (0.250) <sup>b</sup>	2.21 (0.261) <sup>a,b</sup>	3.09 (0.359) <sup>a</sup>	0.005
DMI, kg /hd/d	20.5 (0.34) <sup>b</sup>	21.3 (0.31) <sup>b</sup>	22.8 (0.28) <sup>a</sup>	<0.001
ECM <sup>1</sup> /DMI, kg/kg	1.98 (0.051)	1.98 (0.041)	2.07 (0.044)	0.584
Protein <sup>3</sup> /DMI, kg/kg	0.058 (0.0015)	0.059 (0.0012)	0.060 (0.0013)	0.550
Solids <sup>3</sup> /DMI, kg/kg	0.13 (0.003)	0.13 (0.003)	0.14 (0.003)	0.510
BCS, 0 100 d pp	0.49 (0.133) <sup>a</sup>	0.34 (0.130) <sup>a,b</sup>	0.05 (0.120) <sup>b</sup>	0.049

<sup>1</sup> - ECM: Energy corrected milk yield, kg = 0.327 × milk yield kg + 12.95 × fat kg + 7.21 × protein kg. <sup>2</sup> - Milk solids = kg milk fat + kg milk protein, kg/d <sup>3</sup> - Milk protein and milk solids.

<sup>a, b, c</sup> - Means that do not share a letter are significantly different at P < 0.05.

### 132. Genetic differences exist in the rate of maturity among grazing dairy cows

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**Application:** Genetic differences were identified in the rate at which dairy cows reach mature milk production and the rate at which lactation yield reduces thereafter. These differences may be exploited to select for cows that retain high lactation yields for more parities.

**Introduction:** Improvements in genetic merit for reproductive performance, achieved since the inclusion of reproductive performance in dairy cow genetic evaluations, should manifest in longer productive lives. Risk factors of culling, especially in older cows, are therefore transitioning from infertility/subfertility to low yield. The objective of the present study was to understand the dynamics of lactation yields in dairy cows across parities and to quantify the potential to alter the trajectory through breeding.

**Materials and methods:** Estimated 305-day milk yields were available post-editing for 3,470,520 lactations from 1,162,473 dairy cows calving in 8,566 Irish herds. The parity in which individual cows reached maximum production was determined using a phenotypic random regression model. Contemporary group, heterosis, recombination loss, age, and a cubic polynomial regression on parity were all fitted as fixed effects. A cubic random regression on parity was fitted to represent the cow effect. The random regression model fitted to estimate the (co)variance components was as per the phenotypic analyses, with the addition of a quadratic random regression on both the additive genetic effect of sire and the cow permanent environmental effect in place of the random regression on cow effect. Estimated breeding values (EBV) for 305-day milk production per parity were calculated for each sire with  $\geq 30$  daughters reaching  $\geq 5$ th parity.

**Results:** Of the cows included in the analysis, 13.5%, 79.7%, and 6.1% reached mature 305-day milk production in either fourth, fifth, or sixth

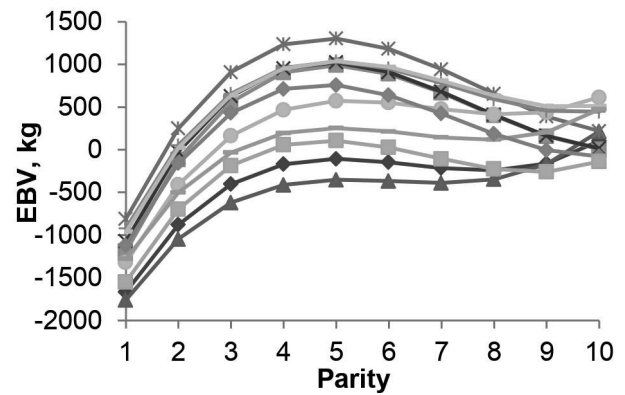


Fig. 1. Estimated breeding values for sires with the most extreme EBVs for parity 305-day milk production.

parity, respectively. The heritability of 305-day milk yield ranged from 0.19 (8th parity) to 0.35 (1st parity). Genetic correlations between 305-day milk yield in different parities varied from  $-0.035$  to  $0.997$ , the strength of the genetic correlation being inversely related to the interval between the compared parities. Differences in EBV for 305-day milk production per parity existed among sires (Fig. 1). Differences in the shape of EBV profile of sires across parities became more pronounced after fifth parity.

**Conclusion:** Differences exist between dairy cows in the parity where maximum milk production is reached. Additionally, genetic differences exist between sires for daughter rate of maturity and the rate of decline in milk production after the parity of maximum production.

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### 133. Prediction of genetic merit for live-weight and body condition score in dairy cows using routinely available linear type and carcass data

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**Application:** Without ample phenotypic data, accurate estimated breeding values (EBVs) are not achievable. Results suggest that readily available linear type trait data are a useful source of information to predict live-weight and body condition score (BCS) with minimal additional predictive value from also considering carcass data.

**Introduction:** Accurate genetic evaluations are predicated on phenotypic data. Multi-trait genetic evaluations can, however, exploit information from correlated traits when predicting EBVs. Interest in efficiency of production is intensifying and two contributors to this are live-weight and BCS. The objective here was to explore the usefulness of routinely recorded data, namely linear type traits and carcass data in the pursuit of accurate genetic evaluations for both live-weight and BCS in dairy cows.

**Materials and methods:** The data post-editing consisted of on-farm records of live-weight and BCS (assessed using both visual and tactile cues) from 33,242 dairy cows. These data were complemented with information on six body-related linear type traits (stature, angularity, chest width, body depth, body condition score, rump width) scored on primiparae and three cull cow carcass measures (carcass weight, conformation and fat cover) on multiparae. (Co)variance components were estimated using animal linear mixed models. A multi-trait genetic evaluation was undertaken where the goal trait was either live-weight or BCS and the predictor traits were type traits only, carcass traits only,

or a combination of both; validation was undertaken in an independent population.

**Results:** The genetic correlation between stature, angularity, body depth, chest width, rump width, and body condition score with live-weight was 0.68, -0.28, 0.43, 0.64, 0.61, and 0.44, respectively. The genetic correlation between angularity and BCS measured on farm (based on both visual and tactile appraisal) was -0.79; BCS is also visually assessed as part of the linear assessment and the genetic (phenotypic) correlation between this visual assessment with BCS assessed using both tactile and visual cues was 0.90 (0.27). The genetic (phenotypic) correlation between carcass weight and live-weight was 0.81 (0.21) while the genetic (phenotypic) correlation between carcass fat cover and BCS was 0.44 (0.12). The regression of phenotypic live-weight on its EBV from the multi-trait evaluations was 1.00 when the EBV was generated using just linear type traits data and less than one (0.83) when using carcass data; the partial correlation between phenotypic live-weight and its EBV ranged from 0.38 to 0.43. While the prediction of phenotypic BCS from its respective EBV was relatively good when using just linear type trait data (regression coefficient of 0.83; partial correlation of 0.22), the predictive ability of BCS EBV derived from carcass data was poor.

**Conclusion:** Both linear type traits and carcass traits are useful sources of information to predict live-weight and BCS. Live-weight can be equally well predicted using linear type traits data or carcass data; in contrast, it is not recommended to predict BCS from carcass data alone.

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### 134. Relationships between energy balance parameters, genetic indexes and fertility traits in multiparous dairy cows

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**Application:** A better understanding of key energy balance (EB) parameters could help improve our understanding of dairy cow fertility.

**Introduction:** Intense genetic selection for milk production has contributed to a decline in fertility performance in modern dairy cows. Energy is the major nutrient driving animal performance, and an inadequate energy supply during early lactation can have a detrimental impact on reproductive activity. The objective of this study was to calculate phenotypic relationships between EB parameters modelled at an individual-cow level, and genetic indexes and reproduction performance in multiparous Holstein cows offered grass silage-based diets.

**Materials and methods:** Data used in this analysis was obtained from a sub-set of studies (n = 23) conducted at the Agri-Food and Biosciences Institute (AFBI) in Northern Ireland over a 20-year period (1996–2016). Data from 143 multiparous cows (156 individual lactations: second to sixth) were used in the analysis. Dry matter intake (DMI), animal performance and feed composition data available from four until at least 90 days in milk (DIM) allowed the calculation of EB for each cow on a daily basis using equations within 'Feed into Milk'. An EB curve for each cow was then estimated using the polynomial regression model of Ali and Schaeffer (1987). From each individual EB curve, four parameters were

calculated: 1. Nadir EB (MJ of ME/day), 2. Time to nadir EB (days), 3. Interval for return to positive EB (days), and 4. Total energy deficit (TED; MJ of ME). Furthermore, for individual lactations to be included in the analysis, genetic indexes, and fertility data, including a positive fertility outcome, was also required. Milk progesterone data (to determine start of luteal activity; SLA) were available for 130 lactations. Partial correlations among these variables were determined using MANOVA in PROC GLM of SAS.

**Results:** On average, multiparous cows reached nadir EB (-61 MJ of ME/d) at 9.4 days in milk (DIM) and returned to positive EB at 52 DIM (Table 1). The large variation in genetic indexes reflects the period over which data was collected (1996 to 2016). Start of luteal activity (SLA) occurred at 33 DIM with first observed heat (FOH) 13 days later. Partial correlations among variables are presented in Table 2. While trends were largely as expected, only a small number were significant. A smaller nadir EB was associated with increased PTA fertility (0.328) and a reduction in days FOH (-0.220), and days from start breeding season until pregnancy (DPSB; -0.185). There was a positive correlation between days to nadir EB and PLI (£), while increasing total energy deficit (TED) was associated with a decrease in PTA for fertility.

**Conclusion:** Cows with smaller nadir EB had a higher PTA for fertility, earlier FOH and fewer days to pregnancy, while a decreasing PTA for fertility was associated with a greater TED. These results demonstrate clear correlations between EB and fertility traits.

**Acknowledgments:** This work was funded by DAERA as part of the DAFM NutriGen project.

Table 1  
Means and standard deviations of modelled energy balance (EB) parameters, genetic indexes, and reproductive traits.

EB parameters <sup>1</sup>			Genetic index <sup>2</sup>			Reproductive traits <sup>3</sup>		
Item	Mean	SD	Item	Mean	SD	Item	Mean	SD
Nadir EB, MJ of ME/d	-61	36.9	PTA Milk (kg)	-5.72	228.9	SLA	33	18.5
Time to Nadir EB, d	9.4	4.95	PTA Fat (kg)	3.65	9.10	FOH	46	27.2
Return to positive EB, d	52	37.0	PTA Prot (kg)	5.26	7.30	N. Serv.	2.3	1.32
TED, MJ of ME	1498	1447.3	PTA Fertility	-0.40	4.62	DSBP	65	54.2
			PLI (£)	80.4	154.5			

<sup>1</sup> TED = total energy deficit; <sup>2</sup> Genetic indexes were sourced for the cows from Animal Horticulture Development Board (AHDB), UK, during December 2018; PTA<sup>2018</sup> = Predicted transmitting ability of milk production (kg) and fertility; PLI<sup>2018</sup> = Profitable life index (£); <sup>3</sup> SLA = Start of luteal activity (d); FOH = First observed heat (d); N.Serv. = number of services needed for the cow become pregnant (maximum 6); DSBP = Days from start breeding season until pregnancy.

Table 2  
Partial correlations among EB parameters, genetic indexes and reproductive traits<sup>1</sup>.

EB Parameters <sup>2</sup>	Genetic index <sup>2</sup>					Reproductive traits <sup>2</sup>			
	PTA Milk (kg)	PTA Fat (kg)	PTA Prot (kg)	PTA Fert.	PLI (£)	SLA	FOH	N. Serv.	DPSB
Nadir EB	-0.040	-0.077	-0.116	0.328***	0.140	-0.032	-0.220*	-0.103	-0.185*
Time Nadir EB	0.164	0.141	0.140	0.095	0.211*	-0.088	-0.172	-0.027	0.003
Return posit. EB	0.125	0.079	0.090	-0.091	-0.020	-0.127	-0.039	0.113	0.063
TED	0.092	0.046	0.080	-0.254**	-0.103	-0.085	0.123	0.146	0.164

<sup>1</sup> Correlations controlled for the effects of lactation number and individual study within the dataset; \*P < 0.05, \*\*P < 0.01, \*\*\*P < 0.001; <sup>2</sup> A full description of abbreviations is provided in Table 1 and its corresponding footnote.

## Invited Speaker Summaries

### 135. Is it enough for the UK dairy reach net zero alone?

Nic Parsons

Agriculture and Horticulture Development Board (AHDB), United Kingdom

Where are we now as a UK dairy sector in meeting our Net Zero commitments? Setting out a baseline as an industry is crucial to the future success of measuring impact also to be able to portray to consumers and influencers of the improvements towards Net Zero gained over the coming years. Can the dairy industry move as one sector, or might this have to be at the rate of the slowest? How do we motivate the dissenters in the industry to adopt and support the direction of travel of the majority stakeholders in our industry?

'Managing through measuring' greenhouse gases alone will not provide the dairy sector with enough of a justification to farm while ignoring the sustainability and wider environmental improvements that consumers and detractor's demand. Challenges to the customer's right to consume will require a wholesale adoption of wider environmental and sustainable behaviours that position dairy farming in a strong position for the future.

Dairy processors and supply-chains, push and pull of the primary producer is crucial to meet the expectations of Government and Devolved Administrations. Meeting targets, measuring progress and reflecting a new collective attitude to the new low carbon economy is critical.

What role does govt /Devolved govt play in the direction of travel? Does the industry have to be driven by policy and legislation or can we take a world lead in advances in carbon reduction technologies, adoption of best practices in productivity, efficiency and off-setting measures?

### 136. The functionality of milk and dairy foods at key life stages

Ian Givens

University of Reading, Food Chain Nutrition and Director, Institute for Food, Nutrition and Health

There is increasing evidence that dietary exposure in early life can influence chronic disease risk in later life. Milk/dairy foods are important suppliers of a range of key nutrients with some being particularly important at certain life stages. Milk protein can stimulate insulin-like growth factor-1 essential for longitudinal bone growth and bone mass acquisition in young children, thus reducing the risk of stunting. Low milk consumption during adolescence contributes to sub-optimal intake of calcium, magnesium, iodine, and other important nutrients. Given the generally low vitamin D status of European populations, this may have already impacted bone development with any resulting reduced bone strength not becoming apparent until they are much older. Sub-optimal iodine status of females of child bearing age has been reported together with observational studies showing an association with poorer cognitive development by the offspring. There is now good evidence that consumption of milk/dairy foods does not lead to an increased risk of cardiovascular diseases and type 2 diabetes and indeed some negative associations are seen, notably between yoghurt consumption and type 2 diabetes. For the elderly, more attention is needed on dietary approaches to reduce loss of muscle mass, its metabolic functionality and bone protecting effect. Whey protein has been shown to be particularly effective for reducing muscle loss and this needs development to provide simple dietary regimes for the elderly to follow. There is an ongoing, often too simplistic debate about the relative value of animal vs. plant food sources for protein in particular. It is important that judgements on the replacement of dairy products with those from plants also include the evidence on relative functionality which is not expressed in simple nutrient content (e.g. hypotensive and muscle synthesis

stimulation effects). Only by considering such functionality will a true comparison be achieved.

### 137. Precision Dairy monitoring technology opportunities and challenges

Jeffrey Bewley

Analytics and Innovation Scientist, Holstein Association USA

Precision Dairy Farming is the use of technologies to measure physiological, behavioral, and production indicators on individual animals to improve management strategies and farm performance. Technologies included within Precision Dairy Farming range in complexity from daily milk yield recording to measurement of specific attributes (e.g. fat content or progesterone) within milk at each milking. Many Precision Dairy Farming technologies, including daily milk yield recording, milk component monitoring (for example, fat, protein, and SCC), pedometers, automatic temperature recording devices, milk conductivity indicators, automatic estrus detection monitors, and daily body weight measurements, are already being utilized by dairy producers. Other Precision Dairy Farming technologies are being developed to measure rumen pH and contractions, feeding behavior, body condition score, animal positioning and activity, locomotion, lying behavior, odor, progesterone, heart rate, greenhouse gas emissions, and respiration rate. With Precision Dairy Farming, the trend toward group management may be reversed with focus returning to individual cows through the use of technologies. The main objectives of Precision Dairy Farming are maximizing individual animal potential, early detection of disease, and minimizing the use of medication through preventive health measures. Precision Dairy Farming is inherently an interdisciplinary field incorporating concepts of informatics, biostatistics, ethology, economics, animal breeding, animal husbandry, animal nutrition, and process engineering.

### 138. Nutrition and feeding management of hyperprolific sow

Uislei Orlando

PIC North America, Global Nutrition Programs

This presentation aims to summarize the current knowledge on nutrition and feeding highly prolific sows to maximize lifetime productivity and herd profitability. Different production phases such as gilt development, body condition management, gestation, lactation, and wean-to-estrus interval are discussed and recommendations are based on peer-reviewed large-scale commercial research. Four pillars of a successful gilt development program include achieving age at puberty in less than 195 d and breeding targets of 135–160 kg of body weight, 200–220 d of age, and second or third estrus. Another key aspect of a high-performance sow farm is to properly manage sow body condition, with the caliper being an easy-to-use tool to accurately measure body condition while taking the subjectivity out of visual scoring. Over-conditioned females at farrowing are costly from a feed perspective and because they have poorer lactation feed intake, compromised subsequent reproductive performance, and overall lower retention and productivity up to parity 3. Thus, body condition should be used as guidelines for gestation feeding, with attention needed in the first 30 days to not feed under maintenance or over two times maintenance. In late gestation, if unable to obtain a caliper reading, it is recommended to maintain the feeding level from the previous period. During lactation, females should be allowed ad libitum feeding to maximize feed intake, which is critical to optimize litter and subsequent performance. Recent trials have shown that providing 2.7 kg or 36.4 MJ of metabolizable energy per day for weaned sows with a caliper reading of ideal is enough to maximize subsequent reproductive performance. A web application was developed to be used by nutritionists and production managers to compare their current feeding

practices to PIC recommendations for highly prolific sows. This will aid in the decision-making process regarding nutrition and feeding programs considering productivity and profitability outcomes.

### **139. Nutritional and other management of the progeny of hyper-prolific sows**

John Pluske

The University of Melbourne/Australasian Pork Research Institute Ltd

Breeding from hyperprolific sows that produce very large litters is now commonplace in commercial pork production. Nevertheless, the presence of a positive relationship between a larger litter size and piglet mortality means that in order to curtail the health and welfare challenges associated with larger litters to in turn maximise the economic potential of having more piglets, various genetic, nutritional and management interventions are needed. Different breeding objectives will play a key role in addressing the issue of raising more viable and surviving piglets from hyperprolific sows, but there are numerous interventions that can be applied to both the sow and the litter, both before and after parturition, to assist in improving piglet survival and health. Dealing with piglets only, then treating complications during farrowing is obviously important for the number of piglets born alive and for sow health. Once piglets are born, then the imperative is to maximise colostrum intake such that each piglet in the entire litter receives sufficient colostrum (at least 200 g) within 12–16 h from parturition. Management strategies such as split suckling, cross fostering/nurse sows, and colostrum dosing can assist with this. Of critical importance and in parallel with these practices, optimising the piglets' thermal environment is essential. It is important to prevent hypothermia immediately at and after birth for hyperprolific litters, or mortality will increase. Once piglets are established, then artificial energy and nutrient supply systems, e.g., feeding milk replacer in the farrowing pen, can assist supernumerary piglets in obtaining energy and nutrients for growth and development. As piglets approach weaning, then offering (or continuing to offer) an energy and nutrient source can assist with the post-weaning transition. Unfortunately, there is at present no one-size-fits-all solution to managing larger litters, and a combination of the various strategies is required to more fully achieve the potential economic benefits of increased numbers of piglets born.

### **140. Welfare and housing of hyperprolific sows and progeny**

Vivi Aarestrup Moustén

SEGES Danish Pig Research Centre, and appointed Honorary, Animal Production, Pigs, University of Copenhagen

How can we contribute to higher welfare for hyper-prolific sows and their progeny? It will to some extent depend on the definition of welfare. However, most farrowing and lactating sows are housed indoors in crates with limited possibilities to nest-build and without the option to turn around or to walk away from their lying area to defaecate.

For the pig industries across the world, it's a lot easier to continue using crates compared to the challenge of changing to more loose housing, and especially if loose housing of lactating sows is to become the new normal. However, the stone age did not end because we ran out of stone...but because other opportunities arose.

Loose housing of lactating sows is an opportunity, but not an easy one to overcome. Pens are larger than crates, so either the production capacity decrease or farmers need to build new buildings. When farmers build a farrowing house, it lasts for 20–30 years, so they want to ensure they have the 'right' pen design before building. So far building for loose housing is more expensive, there is a risk of higher piglet mortality and the opportunity for a premium for the product is limited.

A few countries have had loose housing of lactating sows for a number of years, but it's countries that produce for the home-market, which means the farmers obtain a higher price for their product compared to farmers in countries producing for the global market.

However, it's no longer a question whether the lactating sows will be loose housed, the question is when and how. For the last decade or more, there has been research in loose housing of lactating sows and the impact on sows and piglet survival. Systems are developing which can increase the welfare of hyper-prolific sows and their progeny.

### **141. What do future beef farms need to look like?**

Jude Capper

Livestock Sustainability Consultant

Sustainable beef production must balance three factors: environmental responsibility, economic viability and social acceptability, although the environmental aspect currently garners the greatest attention.

Recent reports have highlighted the importance of improving productivity to allow cattle to finish earlier, thereby reducing environmental impacts. However, little information exists regarding the relative impacts of other key performance indicators, e.g. birthweight, age at first calving, mature cow bodyweight, longevity or calving interval. Inter-sector communication must also improve such that finishing performance and carcass data is fed back down the chain from processors to store and suckler producers. Cattle health must continue to be a priority – the productivity impacts of disease are well-documented, yet few producers are able to quantify the relative economic and environmental cost:benefit ratio of animal health practices (e.g. targeted antimicrobial treatment vs. vaccination) and therefore make evidence-based decisions. It will be crucial for beef producers to approach or exceed carbon net zero targets, therefore they will need to assess their baseline greenhouse gas (GHG) emissions, the opportunities for improvement and the relative importance of different management choices. Although GHG assessment tools are available, no industry standard currently exists, which is a significant threat as the lack of standardization may create discord at the farm or sector level. In future, it will also be essential to broaden the environmental focus and include other metrics (e.g. biodiversity, soil erosion and health, nutrient loss, air and water pollution and odour) in assessment tools.

The future is not challenge-free, but with clear engagement, forward-planning and due diligence with regards to sustainability issues the challenges may be successfully overcome. This requires a culture of continuous improvement, with sustainable increases in cattle productivity; improved health and welfare; adoption of new and existing technologies; and better stakeholder communication both within and outside the beef sector.

### **142. Using machine vision to improve dairy cow welfare standards and reduce green house gas emissions**

Terry Canning

CattleEye

CattleEye, an 'AI first' company, have harnessed the capabilities of advanced Artificial Intelligence in video analytics to deliver the world's first hardware independent autonomous livestock monitoring platform. The platform will transform how billions of livestock are managed with a radically new approach to monitoring which will meet a rapidly growing market demand driven by trends in protein consumption, antimicrobial usage reduction, animal welfare and a need for increased livestock efficiencies.

Founded in 2019 the company has rapidly established a world class multi-disciplinary team, combining deep sectoral knowledge within

Agtech, repeat entrepreneurial experience and world class deep learning image analytics experts

CattleEye provides a completely new way of monitoring and gaining insights on dairy cows simply by walking them under a security camera connected to the Internet. Our completely hands-free solution will monitor a cow's welfare and performance without the need for collars or pedometers that need regular maintenance and commissioning.

Launching in June 2021 the CattleEye mobility scoring module will monitor lameness levels on cows and highlight any individual cows that may require treatment. Using this tool to select cows for foot trimming and treatment will reduce lameness levels on your herd. Research from the UK Animal Health & Welfare Technical Directorate has demonstrated that a 10% decrease in lameness levels on an average dairy herd would create a saving of £91.25 per cow per year and a carbon reduction of 0.57t per cow per year. Research from the University of Liverpool has concluded that CattleEye is "at least as accurate as a human expert in identifying lame cows". Giving dairy farmers a brand new way of improving lameness on herds.

#### **143. The haptic cow: A novel use of innovative technology and research to solve an old problem**

Sarah Baillie

University of Bristol

Simulation is increasingly used in training for many clinical and practical skills and haptic technology provides an innovative solution for some of the more challenging techniques. The presentation will introduce an example of haptics in veterinary education, describe the rationale for using the technology, explain how the simulator was designed and summarise the research studies, including an evaluation of the benefits for student learning. Further examples and opportunities to utilize haptic technology will be discussed.

#### **144. Energy requirement models for beef cattle**

Xianjiang Chen

Agri-Food and Biosciences Institute

Beef energy rationing models of AFRC (1993) were developed using calorimeter chamber data of cattle obtained in 1970 s. However, there is scientific evidence indicating that these models underestimate maintenance energy requirements for modern beef herds, possibly due to the improvement in cattle genetic merit and change of cattle feeds during the last 50 years. The objective of this task are to develop updated maintenance energy requirements for modern beef cattle, and review AFRC (1993) models for quantification of ME requirement for live weight gain. Energy utilization data (n = 297) used were collated from 14 calorimeter chamber studies with steers (n = 176), heifers (n = 88) and dry suckler cows (n = 34), undertaken in AFBI and the University of Reading from 1995 to 2015. The linear regression technique was used for statistical analysis with the effects of research centre, experiment, trial year and other dietary and animal factors being removed. The results indicated that that AFRC (1993) models under-predict maintenance energy requirement for modern beef herds. Dry suckler cows had a lower efficiency of ME utilization than steers or heifers, while live

weight group had no effect. Forage type (grass silage vs. non grass silage) had no effect on energetic efficiency, and forage only diets had a marginally lower energetic efficiency than forage-concentrate diets.

#### **145. Using genetic evaluations to improve descriptions of the composition of growth**

Richard Dewhurst

Scotland's Rural College (SRUC)

Estimates of energy requirements for growing and finishing cattle depend first on predictions of daily liveweight gain and then on the composition (lipid and protein content) of that gain. In this presentation, we will first investigate how well existing models predict growth rates of modern genotypes, focusing on the Reference Animal categories used in the most recent INRA growth models. We will then explore potential to use estimated breeding values (EBVs) for 'carcass traits' to provide better descriptions of animal types than the current (ARC) 'small', 'medium' or 'large' breed ('early', 'medium' and 'late' maturing) descriptors. Models of the composition of liveweight gain are based on serial slaughter studies, which have not been conducted in recent times because of cost and ethical concerns. There is a concern that beef nutrition models are hampered by incorrect representation of the composition of weight gain of modern genotypes because modern/continental genotypes were not represented in data generated before 1980. We have much more information about the composition of animals at slaughter, which is easily gathered at a large scale. Body composition at slaughter is the sum of body composition at all previous stages of growth and finishing and so can provide useful information for scaling of models of growth composition. National genetic evaluations use information from large numbers of slaughter animals to produce EBVs for days to slaughter, average daily carcass gain, carcass weight, carcass conformation and fat class. We will use information from animals finished at SRUC to evaluate use of this information as input to models of the composition of growth.

#### **146. Use of on-farm data alongside nutritional models**

Jenna Bowen

Beef & Sheep Research Centre, Scotland's Rural College (SRUC)

Recent advances in technology has changed how on-farm data is collected, the speed at which it is collected and transformed into real time feedback. Agri-tech or precision livestock farming (PLF) has allowed for automatic and continuous recording of various parameters including animal production, environmental monitoring and animal health and welfare on individual animals, with combinations of data different data streams improving decision making capabilities. The use of on-farm generated data shows great potential to allow for individually tailored nutritional modelling, either at the individual animal level or at the farm/holding level. This presentation will summarise current technologies available to monitor intake, body composition, growth, carcass, and ruminal pH and discuss how on-farm generated data can be used to further refine models.

### 147. Estimation of milk yield based in udder measures of Pelibuey sheep using artificial neural networks (ANN)

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**Application:** The implementation of ANN to Pelibuey sheep data using udder measures as input allows an adequate estimation of milk yield opening the possibility of using image-data in the future.

**Introduction:** Pelibuey sheep is a hair breed widely distributed in Central America countries, principally reared in tropical regions [1]. This breed is mainly focused to produce meat; however, recently has been an increasing interest in using sheep's milk to produce high-quality dairy products. Udder measures have been used to estimate milk yield of sheep through classical methods of estimation [2]. ANN is capable to deal with complex non-linear relationships between input and output variables, with no need for rigid *a priori* models. In the current study ANN were applied to udder measures from Pelibuey ewes to estimate their milk yield and compared with predictions from the classical linear regression.

**Materials and methods:** A total of 357 milk yield records with its corresponding gland udder measures was used in the present study. Ewes were milking manually once a day and udder measures were recorded twice a week. Following udder measures were taken before and after milking: udder circumference (UC), udder width (UW), udder height

(UH), teat length (TL), diameter of the teat (TD) and udder volume (UV). A supervised learning was used to train and teach the network using a two-layer ANN with 10 neurons each one (Fig. 1). The globally convergent algorithm based on the resilient backpropagation was used to calculate ANN in *neuralnet* package [3] in R software. The fit of ANN was compared with the best multiple regression model (MRM) fitted previously by Arcos-Alvarez et al. [2] using same input variables in both models. Goodness of fit was evaluated using the mean square prediction error (MSPE), correlation between actual and predicted values ( $r$ ) and Akaike's Information Criterion (AIC).

**Results:** ANN showed better estimations of milk yield in comparison with MRM. Lower values of MSPE (0.008 vs. 0.013) and AIC (-1629.01 vs. -507.22) were observed to ANN. The  $r$  value was higher in ANN (0.87) than in MRM (0.79).

**Conclusion:** The study reveals that ANN is a powerful tool to estimate milk yield when udder measures are used as input variables and showed better goodness of fit in comparison with classical regression methods.

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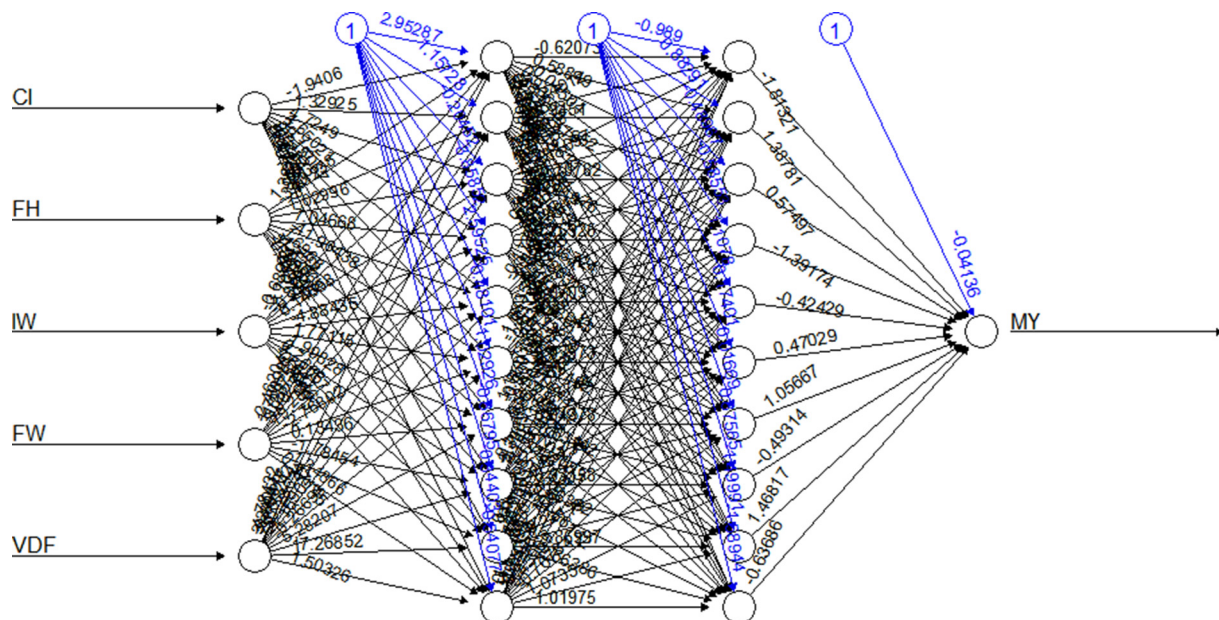


Fig. 1. Architecture of used ANN with two-layers and 10 neurons to estimate milk yield (MY) and initial udder circumference (CI), final udder height (FH), initial udder width (IW), final udder width (FW), and difference initial and final udder volume (VDF) as input variables.



#### 148. Polymorphisms of the melatonin receptor 1A (*MTNR1A*) gene do not modify social strategies of Rasa Aragonesa rams in spring

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**Application:** Carrying certain genotypes of the receptor MT1 of the melatonin hormone produces an improvement of sexual performance of rams in spring, although we have demonstrated that this is not related with a higher social rank or behavior of these rams.

**Introduction:** Melatonin secretion controls reproductive seasonality in sheep through specific receptors, MT1 being involved in the regulation of reproductive functions [5]. It is encoded by the *MTNR1A* gene with two different polymorphisms (Rsal and MnlI), that affect the seasonal reproductive function of the female sheep [2]. Since we have previously demonstrated that polymorphisms of the *MTNR1A* gene sequence influence the reproductive performance of young and adult rams [1], the aim of this work was to study the effect of carrying specific genotypes of the *MTNR1A* gene on their social strategies.

**Materials and Methods:** Thirty-one Rasa Aragonesa rams housed indoors, were genotyped for their Rsal and MnlI polymorphism of the *MTNR1A* gene: CC (n=19), CT (n=5), and TT (n=7), and GG (n=20), GA (n=6), or AA (n=5), respectively. In mid-June, during the anestrus season, maintenance and social behaviour of the rams [3] was recorded through direct observations, with a combination of instantaneous scan sampling (recording behaviour of an individual in a group at predetermined time intervals) and continuous behaviour sampling methods. Maintenance behaviours (standing still, lying, drinking, feeding, walking or stereotyping) were observed at 10-minute intervals, and the continuous method was used to record agonistic interactions among rams. Index of Success (IS) and Displacement (ID) were calculated as IS = number of rams he displaces / (number of rams he displaces + number of rams which displace him), and ID = number of times a ram displaces another ram / (number of times it displaces another ram + number of times he is displaced). Rams were divided into three ranking categories according to their IS: low (0.0-0.33), medium (0.34-0.66), and high rank (0.67-1.0) [4]. Anova and X<sup>2</sup> were applied.

**Results:** No statistical differences among the three genotypes of each polymorphism were observed, so that they performed and received a similar number of aggressions with and without contact, and their active and passive behaviours were also similar. For the IS and ID, genotypes presented no differences in their values (Fig 1), and the proportion of rams in each success category was similar among groups (Table 1).

**Conclusion:** Results of this study indicate that the previously observed best sexual performance of rams carrying certain genotypes of the *MTNR1A* gene is not related with differences of social strategies of these particular rams.

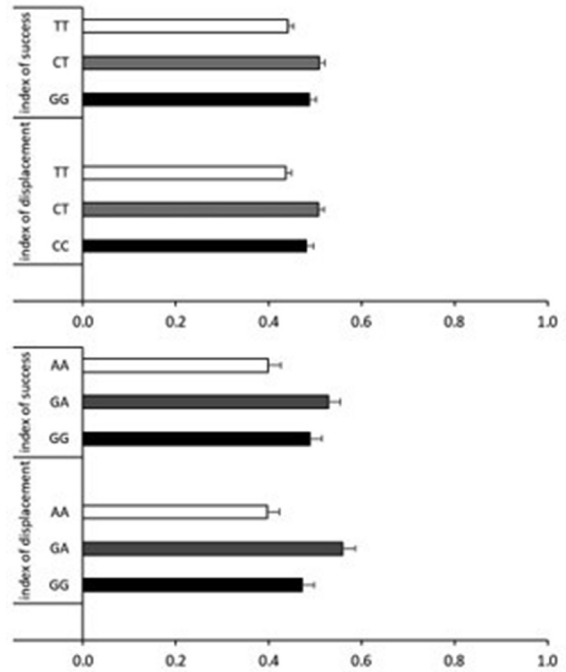


Fig. 1. Index of Success and Displacement achieved by each genotype

Table 1  
Social rank (by Index of success) of each genotype

	Low	Medium	High
CC	32%	47%	21%
CT	40%	40%	20%
TT	0%	57%	43%
GG	30%	40%	30%
GA	33%	67%	0%
AA	0%	60%	40%

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### 149. Early diagnosis of bovine respiratory disease (BRD) using clinical respiratory signs and thoracic ultrasonography in artificially reared calves

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**Application:** The detection of sub-clinical BRD in calves with lung lesions without showing clinical respiratory signs, can only be confirmed using thoracic ultrasonography.

**Introduction:** Despite widespread use of clinical respiratory signs for diagnosing BRD, lung lesions are detected, using thoracic ultrasonography (TUS) or following post-mortem, in calves showing no clinical signs. This highlights the limitation of clinical respiratory signs as a method of detecting sub-clinical BRD.

**Materials and methods:** A total of 53 male dairy-bred calves, comprising of 28 Herford × Aberdeen Angus (24 (SD; 7.1) days old; 53.5 (SD; 8.8) kg) and 25 Holstein-Friesian (21 (SD; 4.6) days old; 51.7 (SD; 5.4) kg), were purchased from 13 different commercial farms during February and March 2020 and transported by road to the research centre (transport duration ranged from 30 min to 240 min). Following arrival (d 0), calves were randomly accommodated indoors in straw-bedded individual pens, in a naturally ventilated area (n = 32) or in a fan ventilated area (n = 21), until weaning at 75 (SD; 8.8) days of age (d 53). Recording of clinical respiratory signs (rectal temperature, cough, ear position, nasal and eye discharge) and of TUS evaluations were performed by the same trained research veterinarian on d 0, 7, 14 and 30 after arrival.

**Results:** Clinical BRD (based on Wisconsin clinical respiratory score and/or rectal temperature > 39.6 °C) was detected in 43% and lung lesions were detected in 64% of calves from purchase (23 (SD; 6.2) days of age) until weaning, 53 days post-arrival. Calves with clinical BRD were treated. The classification of calves with clinical BRD (cBRD) and TUS score (TUSS) 1 to 4 from day (d) 0 to d 14 (TUS14) and d 30 (TUS30) post-arrival is represented in Table 1. Sixty-one per cent calves affected with clinical BRD had lung lesions 10.5 days (median) before

Table 1

Distribution of calves according to clinical bovine respiratory disease (cBRD) and thoracic ultrasonography score.

TUSS <sup>1</sup>	TUS14 <sup>2</sup>			TUS30 <sup>3</sup>		
	cBRD- <sup>4</sup>	cBRD+ <sup>5</sup>	TOTAL	cBRD- <sup>4</sup>	cBRD+ <sup>5</sup>	TOTAL
TUSS1	22	6	28	15	4	19
TUSS2	3	8	11	9	8	17
TUSS3	2	5	7	3	6	9
TUSS4	3	4	7	3	5	8
Sum of TUSS 1 to 4	30	23	53	30	23	53

<sup>1</sup>Thoracic ultrasonography score (from 1 to 4) (TUSS1 (normal lung or comet tails); TUSS2 (Lung lesion < 2 cm<sup>2</sup>; TUSS3 Lung lesions ≥ 2 cm<sup>2</sup>; TUSS4 Consolidated lung lobe and/or emphysema). <sup>2</sup>Classification of calves considering the ultrasound evaluations performed from arrival to day 14. <sup>3</sup>Classification of calves considering the ultrasound evaluations performed from arrival to day 30. <sup>4</sup>cBRD- calves that had no BRD treatment from arrival until weaning. <sup>5</sup>cBRD + calves that had BRD treatment from arrival until weaning.

detection of clinical signs. Moderate correlations ( $r_{sp}$  0.40 to 0.70;  $P < 0.05$ ) were found between cough and lung lesions ≥ 2 cm<sup>2</sup> on days 0 and 7 post-arrival, and between rectal temperature > 39.6 °C and lung lesions ≥ 2 cm<sup>2</sup> on days 7 and 14 ( $r_{sp}$  0.40;  $P < 0.05$ ) post-arrival. Mean average daily live weight gain (ADG) of calves from purchase to weaning was 0.75 (SD; 0.10) kg; calves with or without clinical BRD did not differ in ADG ( $P > 0.05$ ), whereas ADG of those with severe lung lesions (lung lobe completely consolidated or pulmonary emphysema) was 0.09 kg/d less ( $P < 0.05$ ) than calves without lung lesions.

**Conclusion:** Thoracic ultrasonography detected lung consolidation in calves that did not show signs of respiratory disease. The presence of severe lung lesions was associated with reduced pre-weaning growth. These findings emphasise the importance of using TUS in addition to clinical respiratory scoring of calves for an early and accurate detection of clinical and sub-clinical BRD.

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### 150. Relationships between early lactation energy balance and performance indicators, blood metabolites and fertility traits in multiparous dairy cows

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**Application:** Early lactation energy balance is a key driver of return to cyclicity in dairy cows, but not of conception rate.

**Introduction:** While the decline in dairy cow fertility in recent decades has often been linked to a greater negative energy balance (EB), many 'fertility studies' have involved relatively small numbers of cows. The current study used a large individual cow dataset collected over a 20-year period to examine the relationships between early lactation EB (days 4 – 21 in milk, generally corresponding to nadir EB), cow performance, blood metabolites, and a number of fertility traits.

**Materials and methods:** A meta-analysis of data from 1020 Holstein-Friesian dairy cow lactations, collated from 27 experiments conducted at AFBI Hillsborough over a 20 year period, was undertaken. Cows were in lactations 2 – 6, and had dry matter intake (DMI), performance and feed composition data available from calving until a mean of 129 days in milk (DIM), thus allowing daily EB (MJ of ME/d) to be calculated for this period. Fertility data was available for all cows, while milk progesterone (to determine start of luteal activity; SLA) and periodic blood metabolite data were available for 722 and 860 lactations, respectively.

The dataset was divided into quartiles (Q1 – Q4) according to the average EB during 4 – 21 DIM (Q1, –191 to –79; Q2, –79 to –48; Q3, –48 to –22 and Q4, –22 to 93 MJ/d), and differences between EB quartiles for production and fertility traits were compared using GenStat, Version 20.1 (VSN International Limited, 2019).

**Results:** Moving from Q1 to Q4 (increasing EB), mean DMI and milk protein content increased, while energy corrected milk yield, milk fat content, milk fat : protein ratio, plasma non-esterified fatty acid and  $\beta$ -hydroxybutyrate concentrations decreased (Table 1). Similar significant trends ( $P \leq 0.01$ ) were observed over days 4 – 150 DIM (data not presented), with the exception of DMI ( $P = 0.082$ ). For the subset of cows for which milk progesterone data was available, interval from calving to SLA decreased from Q1 to Q4, while peak progesterone concentration at SLA increased. Days to first observed heat (FOH) decreased from Q1 to Q4, while the percentage of cows with FOH before 42 DIM increased from Q1 to Q4. For each 10 MJ/day decrease in mean EB during 4–21 DIM, FOH was delayed by 0.8 days. However, neither days to first artificial insemination (AI) nor the percentage of cows that conceived to first AI, were affected by early lactation EB, perhaps reflecting first AI taking place at approximately 70 DIM in all EB quartiles.

**Conclusion:** While interval from calving to SLA and to FOH were reduced with improving EB in early lactation, conception rate to first AI was unaffected by early lactation EB.

**Acknowledgments:** This work was funded by DAERA and DAFM as part of the DAFM NutriGen project.

Table 1

Performance indicators (days 4 to 21 post calving) and fertility traits of cows within energy balance (EB) quartile (the latter based on mean daily EB during 4 to 21 days in milk; DIM).

Item <sup>1</sup>	Quartiles (mean daily EB during 4 to 21 DIM)				SED	P-value <sup>2</sup>
	Q1 (-191 to -79 MJ/d)	Q2 (-79 to -48 MJ/d)	Q3 (-48 to -22 MJ/d)	Q4 (-22 to 93 MJ/d)		
<b>Performance indicators (4 – 21 DIM)</b>						
Total dry matter intake (kg/d)	15.5 <sup>a</sup>	16.8 <sup>b</sup>	16.8 <sup>b</sup>	18.0 <sup>c</sup>	0.24	< 0.001
Energy corrected milk (kg/d)	41.9 <sup>d</sup>	38.3 <sup>c</sup>	34.7 <sup>b</sup>	30.9 <sup>a</sup>	0.54	< 0.001
Milk fat (g/kg)	49.6 <sup>c</sup>	46.0 <sup>b</sup>	45.3 <sup>b</sup>	42.6 <sup>a</sup>	0.58	< 0.001
Milk protein (g/kg)	34.8 <sup>a</sup>	35.3 <sup>b</sup>	35.7 <sup>bc</sup>	36.0 <sup>c</sup>	0.26	< 0.001
Fat: protein ratio	1.43 <sup>c</sup>	1.30 <sup>b</sup>	1.27 <sup>b</sup>	1.19 <sup>a</sup>	0.017	< 0.001
Plasma NEFA (mmol/mL)	0.70 <sup>d</sup>	0.58 <sup>c</sup>	0.52 <sup>b</sup>	0.42 <sup>a</sup>	0.028	< 0.001
Plasma BHB (mmol/l)	0.82 <sup>c</sup>	0.74 <sup>b</sup>	0.69 <sup>b</sup>	0.61 <sup>a</sup>	0.038	< 0.001
<b>Fertility traits</b>						
Interval from calving to SLA	35.7 <sup>b</sup>	29.4 <sup>a</sup>	31.0 <sup>a</sup>	29.8 <sup>a</sup>	1.29	0.003
Peak progesterone conc. at SLA (ng/mL)	24.4 <sup>a</sup>	25.9 <sup>ab</sup>	27.4 <sup>b</sup>	28.1 <sup>b</sup>	2.14	0.026
Cows with SLA pre day 42 (%)	70 (59–80)	83 (75–90)	78 (68–86)	79 (69–86)	-	0.103
Days to FOH	49.0 <sup>b</sup>	44.9 <sup>ab</sup>	42.9 <sup>a</sup>	41.6 <sup>a</sup>	2.07	0.012
Cows with FOH pre day 42 (%)	41 (29–53)	49 (38–61)	55 (43–66)	58 (46–69)	-	0.038
Days to first AI	72.7	69.9	68.9	68.6	3.04	0.557
Conception to first AI (%)	28 (22–35)	29 (24–36)	26 (21–33)	33 (26–40)	-	0.672
<b>Genetic traits</b>						
Profitable lifetime index (EPLI)	-64.9	-55.8	-43.4	-39.2	11.23	0.140
PTA milk (kg)	40.6 <sup>a</sup>	45.2 <sup>ab</sup>	55.8 <sup>b</sup>	58.7 <sup>b</sup>	6.83	0.048
PTA fertility	-0.5	-0.7	-0.5	-0.5	0.17	0.270

<sup>1</sup> NEFA = non-esterified fatty acids; BHB =  $\beta$ -hydroxybutyrate; FOH = first observed heat; SLA = start of luteal activity; PTA = Predicted transmitting ability; <sup>2</sup>Values within a row with different superscript differ at  $P < 0.05$ ; Binomial data, upper and lower confidence limit in parenthesis.\*This poster was presented as part of the Developing talent session at the BSAS conference.

### 151. Effects of regrouping of Scottish Holstein-Friesian cows on milk production, physical activity, rumination time and cortisol concentration in a robotic milking system

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**Application:** Farmers should be aware that switching primiparous cows between groups may cause a short-term milk yield drop, and a significant decline in milk fat in all cows

**Introduction:** In the commercial dairy industry, cows are commonly regrouped. Regrouping of unfamiliar animals poses animals to a new social environment which may negatively influence their welfare. Consequently, animal welfare impairment can induce negative effects and may lead to a reduction in milk production (Hasegawa et al., 1997). We investigated whether regrouping altered dairy cows' physical activity, rumination time and impacted on their stress levels, milk yield and quality in a robotic milking system.

**Material & Methods:** Fifty-two lactating cows (n = 17 primiparous; 35 multiparous) were moved in groups of 3–5 individuals into established pens of approximately 100 cows. Average daily milk yield (ADMY), fat and protein, physical activity and rumination time data were extracted from the Lely T4C management program for 4-time blocks (day-prior (d-1), day-of regrouping (d0), day-after (d + 1) and 6-days after (d + 6) regrouping) and milk samples were collected for cortisol concentration analysis. Linear Mixed Effect Modeling ('NLME' package) in R

was used to determine the effect of the regrouping on milk yield, fat, protein, water, dry matter, gross energy (GE), cortisol, activity and rumination time as the dependent variables; with regrouping days, parity and their interaction as the fixed effects.

**Results:** Neither regrouping nor parity statistically influenced dry matter, water content, GE or cortisol concentration ( $P > 0.05$ ). Primiparous cows produced ( $3.80 \pm 2.42$  kg (12.2%),  $P = 0.006$ ) less ADMY on d + 1 compared to baseline level, whereas multiparous cows did not change ADMY (Fig. 1C). Fat % decreased significantly for both groups following regrouping and remained low up to d + 6 (Fig. 1D), but a decrease in protein was only detected on d + 6 compared to d-1 in both groups. Activity in both primiparous and multiparous cows increased significantly on d0 compared to d-1 (Fig. 1A). Multiparous cows significantly decreased their time spent ruminating on d0 compared to d-1, but the values returned to baseline by d + 6 (Fig. 1B).

**Conclusion:** This study indicated that whilst regrouping increased activity and decreased rumination time, it only negatively affected the milk production of primiparous cows in the short term and a long-term drop of over 0.2% in fat content in both parities. There was no impact on ADMY in multiparous cows and it did not influence the stress responses in both parities. Multiparous cows may therefore benefit from previous experience.

**Acknowledgements:** The authors acknowledge funding from CSC UK.

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#### Tables

\*This poster was presented as part of the Developing talent session at the BSAS conference

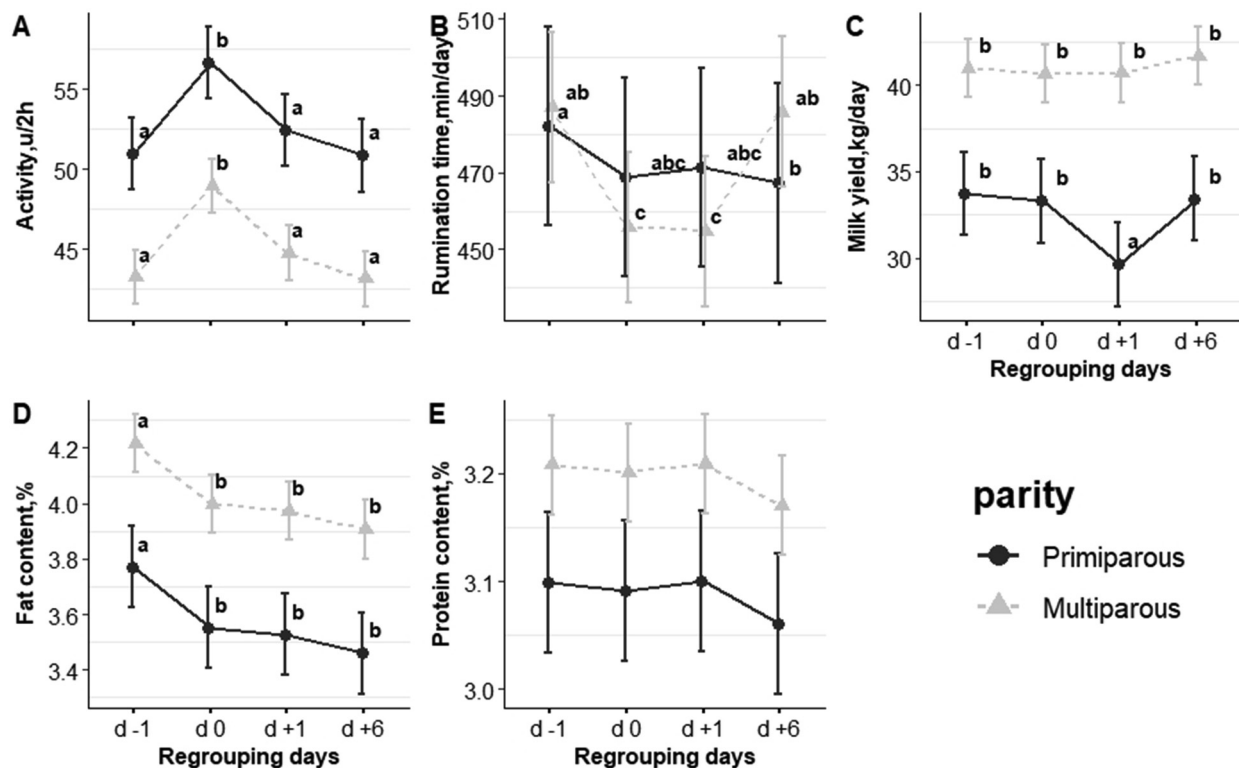


Fig. 1. Least square means (± SEM) of the physical activity (A), rumination time (B), average daily milk yield (C), fat (D) and protein (E) (n = 52) of the cows during the experimental study periods. The plot illustrates the effect of regrouping (represented by the days relative to the regrouping in the x-axis) and parity. <sup>a,b,c</sup> superscripts represent a significant difference ( $P < 0.05$ ) between the regrouping days and within parity.

### 152. Comparison of growth, yield and nutrient composition of Napier hybrids cultivars Sampoorna (*Pennisetum pedicellatum* × *P. americanum*) and CO5 (*Pennisetum glaucum* × *P. Purpureum schumacheri*) harvested at five intervals

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**Application:** Current findings demonstrated that hybrid Napier cultivar CO5 can be used as a potential forage for ruminant feeding in Sri Lanka.

**Introduction:** Nutritionally poor quality tropical forages are the predominant varieties available for ruminant feeding in Sri Lanka and as a result, average milk production per cow/day is less than 4 kg. Two hybrid Napier cultivars; Sampoorna (SP) and CO5 introduced by Tamilnadu University of Agriculture and Indian Grassland and Forage Institute respectively have been described as superior to previously introduced hybrid Napier varieties of CO3 and CO4. The objectives of the current study were to evaluate growth, yield and nutrient composition of SP and CO5 grown under rain fed conditions and harvested at five intervals.

**Materials and Methods:** A 2 × 5 factorial design (2 varieties, each harvested at 4, 6, 8, 10 and 12 weeks) was laid out in 3 blocks (3 replicates). The plot size was 5 × 5 m. Soil analyzed prior to the establishment, recommended basal fertilizer added (Premarathne and Premalal, 2006) and two healthy nodes per harvest were planted at 1 × 1 m spacing. At each harvest, fodder fresh matter yield at 1 m<sup>2</sup> measured and representative sub samples (± 1 kg) obtained for laboratory analysis. Metabolizable

energy (ME) contents were evaluated using *In-vitro* digestibility method (Menke and Steingass 1988). Data analyzed using MINITAB (16th Version) as a generalized linear model with a 2 factor ANOVA to compare the mean differences between cultivars harvested at 5 intervals.

**Results:** Dry matter yield (DMY) was not different ( $p = 0.981$ ) between SP and CO5 at each harvest, but increased ( $p < 0.004$ ) from 8th to 10th week (Table 01). DM content of two cultivars increased ( $p < 0.001$ ) from 8th to 10th week and cultivar SP had more DM ( $p = 0.00$ ) than CO5 at 10th and 12th weeks of harvest. Crude protein (CP) content gradually declined with forage maturity, but was high ( $p < 0.002$ ) in CO5 compared to SP at 8th and 12th weeks of harvest. Compared to the 4th week, ash content of two cultivars were low ( $p < 0.001$ ) at 12<sup>th</sup> week of harvest. Crude fiber (CF) content increased for both cultivars from 8 to 12th weeks of harvests ( $p < 0.002$ ), and at 12th weeks, CO5 had less ( $p < 0.003$ ) CF than SP. CO5 had higher ( $p < 0.002$ ) ME content compared to the SP at both 10th and 12th weeks of harvest.

**Conclusions:** Hybrid Napier CO5 harvested at 12th week recorded high CP content, ME and low CF compared to the cultivar SP. Therefore, CO5 harvested at 3 months of age is recommended to be introduced in to Sri Lanka as a potential fodder variety for ruminant feeding.

**Acknowledgments:** The authors acknowledge funding from the National Science Foundation of Sri Lanka (Grant No: RG/2017/AG/04).

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Table 01

Dry matter yield, proximate composition and ME content of SP and CO5 harvested at 5 intervals.

Parameter	Variety (V)	Harvesting Interval : HI (Weeks)					S.E.M	Significance		
		4	6	8	10	12				
DMY	SP	0.25 <sup>b</sup>	0.17 <sup>b</sup>	0.88 <sup>b</sup>	4.78 <sup>a</sup>	5.25 <sup>a</sup>	V	0.23	V	0.981
(MT/ha)	CO5	0.19 <sup>b</sup>	0.17 <sup>b</sup>	0.31 <sup>b</sup>	4.63 <sup>a</sup>	5.13 <sup>a</sup>	HI	0.37	HI	0.004
DM (g/100 g)	SP	12.12 <sup>c</sup>	12.72 <sup>bc</sup>	15.82 <sup>bc</sup>	17.13 <sup>a</sup>	23.01 <sup>a</sup>	V	0.51	V	0.001
	CO5	11.70 <sup>c</sup>	12.51 <sup>c</sup>	13.58 <sup>c</sup>	15.92 <sup>b</sup>	18.72 <sup>b</sup>	HI	0.81	HI	0.000
CP (g/100 g)	SP	17.32 <sup>ab</sup>	18.34 <sup>a</sup>	11.43 <sup>c</sup>	9.44 <sup>cd</sup>	7.56 <sup>d</sup>	V	0.19	V	0.002
	CO5	19.29 <sup>a</sup>	17.54 <sup>a</sup>	15.46 <sup>b</sup>	10.65 <sup>c</sup>	10.15 <sup>c</sup>	HI	0.30	HI	0.001
Ash (g/100 g)	SP	17.82 <sup>a</sup>	15.45 <sup>bc</sup>	11.65 <sup>d</sup>	11.31 <sup>d</sup>	11.18 <sup>d</sup>	V	0.39	V	0.002
	CO5	19.00 <sup>a</sup>	17.38 <sup>ab</sup>	14.01 <sup>d</sup>	12.03 <sup>abc</sup>	11.53 <sup>d</sup>	HI	0.61	HI	0.001
CF (g/100 g)	SP	23.34 <sup>a</sup>	24.10 <sup>a</sup>	27.86 <sup>b</sup>	32.46 <sup>c</sup>	40.87 <sup>d</sup>	V	0.62	V	0.002
	CO5	23.62 <sup>a</sup>	23.98 <sup>a</sup>	28.94 <sup>b</sup>	34.11 <sup>c</sup>	38.29 <sup>c</sup>	HI	0.53	HI	0.003
ME	SP	8.77 <sup>bcd</sup>	8.81 <sup>bcd</sup>	8.84 <sup>bcd</sup>	8.04 <sup>de</sup>	8.74 <sup>bcd</sup>	V	0.20	V	0.004
(MJ/Kg/DM)	CO5	8.64 <sup>cde</sup>	7.74 <sup>e</sup>	9.62 <sup>ab</sup>	10.06 <sup>a</sup>	9.96 <sup>a</sup>	HI	0.09	HI	0.002

Means followed by different superscripts within a row are significantly different ( $p < 0.05$ ).

### 153. Are male calves lazier than female calves?

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**Application:** Sensors are increasingly being used for monitoring behavioural changes as a disease indicator in calves. However, housing method, age, and sex influence calf behaviour and should be considered when estimating behavioural changes in response to disease events.

**Introduction:** Research has shown that behavioural data have potential to indicate disease in individual calves [1]. This study aimed to characterise the effects of housing system, sex, and age period on the activity of dairy calves aged 5 to 65 days using leg-mounted sensors.

**Materials and methods:** Across seven data sets from AFBI Hillsborough, UK, 85 calves were individually housed [2], 123 calves were housed in groups of initially 5 animals, gradually increasing up to 15 animals, and 28 were pair housed until 54 days, then group housed. Calves were weaned between 50 and 76 days of age depending on trial. Calf health was assessed daily, and only days where no disease was recorded were used for this study. A total of 9023 healthy days from 236 dairy calves (117 female and 119 male) were available for analysis. Activity was measured as Motion Index (MI, no unit) using IceQube + accelerometers (IceRobotics Ltd, South Queensferry, Edinburgh, UK). Data were split into three periods: d5-24 (P1), d25-44 (P2), and d45-65 (P3). Effects were estimated in a mixed model (R-package lme4) with sex, housing method, period, and their 3-way interaction as fixed effects and calf as a random effect. Contrasts are reported with standard error (SE).

**Results:** Females exhibited numerically higher MI than males in every period and housing system (Fig. 1). However, the 3-way interaction was significant ( $X = 19.7$ ,  $df = 4$ ,  $P < 0.001$ ), thus females showed significantly higher MI than males only when group housed during P1 ( $890 \pm 137$ ,  $P < 0.001$ ) and P2 ( $525 \pm 136$ ,  $P < 0.05$ ). As expected, and likely due to increased space allowance and ability for social contact, group housed calves showed higher MI than pair housed calves ( $1087 \pm 118$ ,  $P < 0.001$ ), which showed higher MI than individually housed ones ( $503 \pm 136$ ,  $P < 0.001$ ). MI decreased with age, except for individually housed calves and for group housed males, whose mean MI peaked during P2.

**Conclusion:** Young, group housed male calves were significantly less active than female calves. Innate behavioural differences between sexes and housing system should be accounted for when designing experiments and developing sensor-based health monitoring systems.

**Acknowledgments:** We gratefully acknowledge the Centre for Innovation and Excellence in Livestock, UK, for supporting this study as part of their seed funding program 2020-2021.

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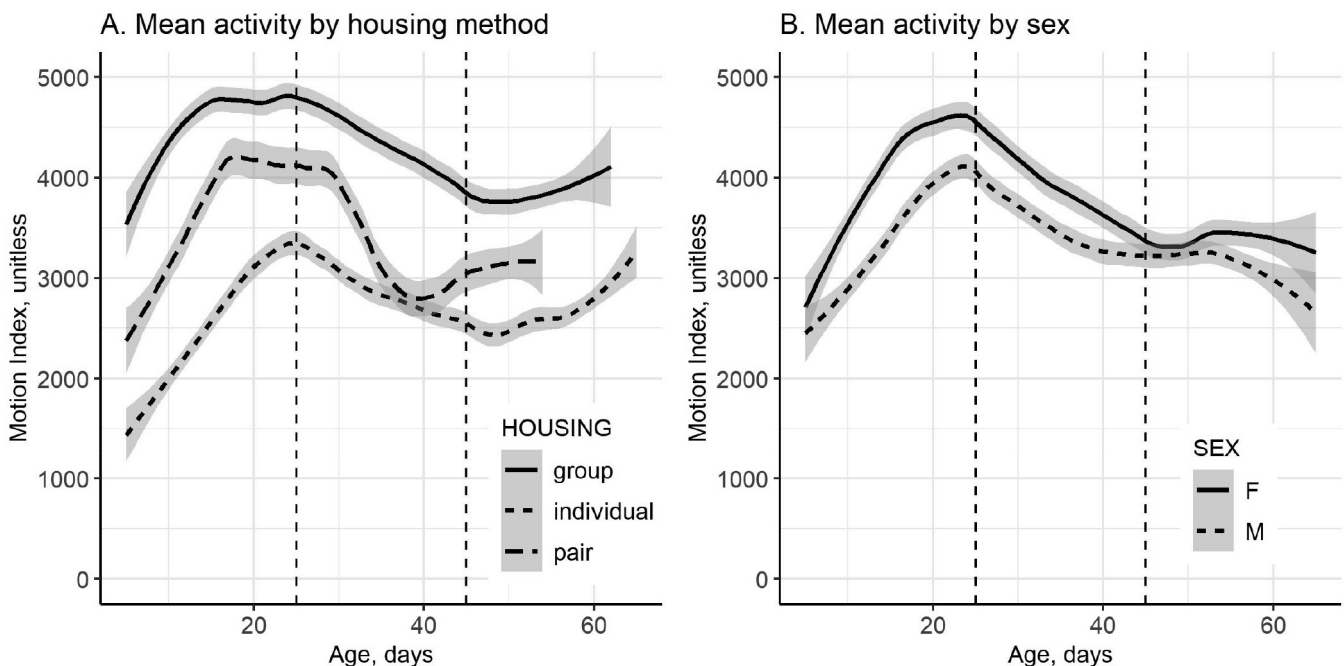


Fig. 1. Mean daily activity of healthy calves relative to age in days, pooled by A) housing method and B) sex, F: female, M: male,  $n = 9032$  days, vertical lines separate periods 1, 2 and 3. Loess smoothing (span = 0.4) applied.

### 154. Impact of supplementation dose and form on selenium partitioning and composition in urine and faeces of sheep

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**Application:** Supplementation dose is a greater determining factor than supplement form with respect to selenium retention in sheep and selenium flux in grazing pasture systems.

**Introduction:** Ruminant excreta provide important nutrients for growing pasture in grazing livestock systems. Additionally, with sustained interest in increasing mineral provision to ruminants through supplements, there is a need to better understand how different forms and doses of mineral supplements, which reflect different bioavailability, affect micronutrient excretion and subsequent flux within the environment.

Table 1

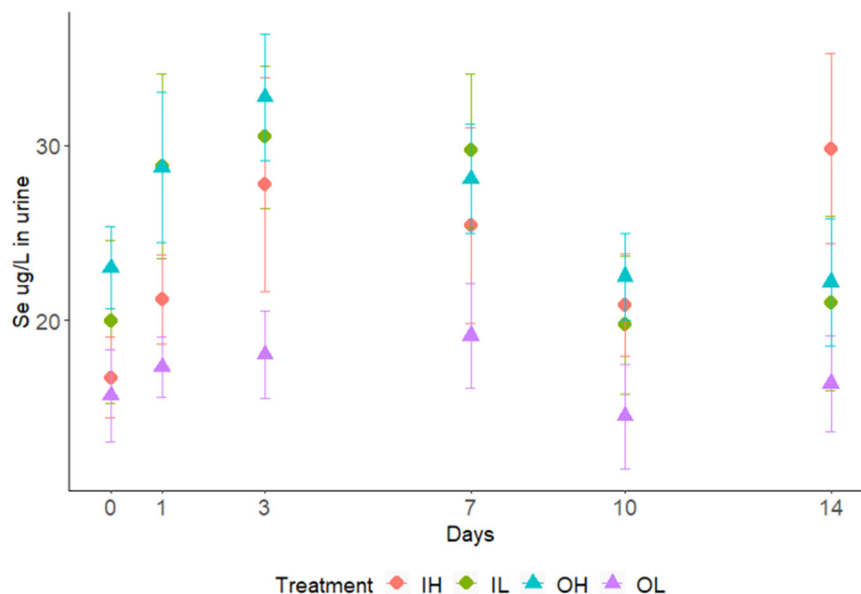
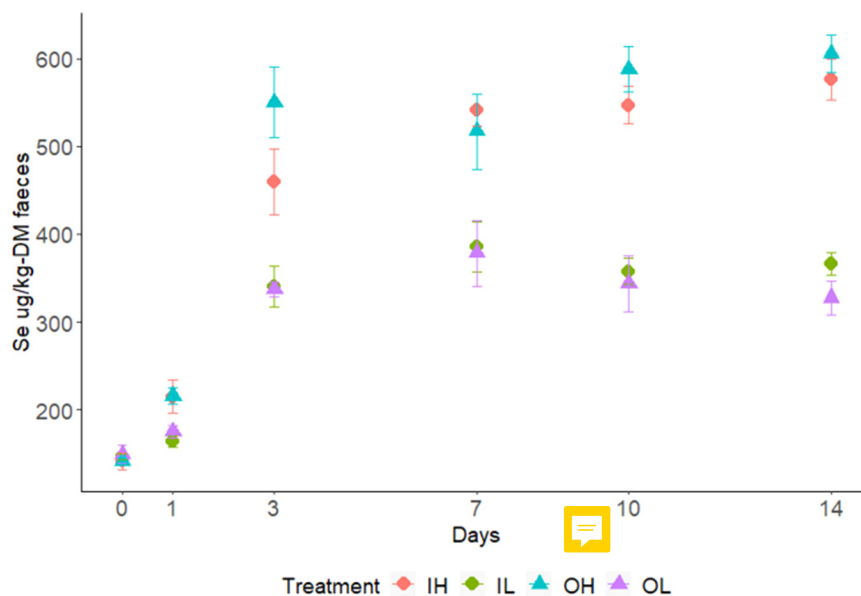
Selenium provision (mg/animal/day) from different intake sources during the experiment.

Treatment	Silage	Background concentrate	Mineral supplement	Total intake
IL	0.022 (9%)	0.094 (38%)	0.133 (53%)	0.249
IH	0.020 (4%)	0.095 (19%)	0.391 (77%)	0.506
OL	0.021 (10%)	0.078 (37%)	0.110 (53%)	0.209
OH	0.023 (5%)	0.095 (20%)	0.368 (76%)	0.486

Table 2

Selenium partitioning (proportion of total intake) in urine and faeces and retention in sheep.

Treatment	Urine	Faeces	Retained
IL	0.11 ± 0.01	0.52 ± 0.02	0.37 ± 0.02
IH	0.09 ± 0.01	0.46 ± 0.03	0.45 ± 0.03
OL	0.13 ± 0.00	0.51 ± 0.02	0.36 ± 0.02
OH	0.08 ± 0.01	0.43 ± 0.03	0.49 ± 0.02
Se forms (Pr > F)	0.9361	0.5106	0.4777
Se levels (Pr > F)	0.0010***	0.0156*	0.0004***
Forms × levels (Pr > F)	0.0638	0.8717	0.3765



The aim of this experiment, therefore, was to determine the effects of supplementary doses and forms (organic/inorganic) of key minerals on mineral partitioning across different excretory routes in sheep. The selenium results are presented here.

**Materials and methods:** Concentrates containing inorganic minerals: manganese oxide, copper sulphate pentahydrate, zinc oxide and sodium selenite or organic minerals: manganese, copper, zinc chelates of protein hydrolysate, and selenised yeast (Selplex®) were given to 24 individually-penned male Charolais × Suffolk-Mule sheep offered a ration of grass silage: concentrate (60:40; DM basis of pre-determined 0.9 *ad libitum* silage DM intake). The concentrates with premixed minerals of 100% or 80% of typical industrial levels of inclusion (IND) were offered for two weeks, with Se offered 0.6 or 0.2 mg-Se/kg-concentrate, respectively, based on the permitted upper-level of organic Se supplement. This resulted in four treatments: organic minerals at higher (OH) or lower (OL) IND and inorganic minerals at higher (IH) or lower (IL) IND. The sheep were allocated randomly into six blocks according to their body weights, and offered silage and different treatments of concentrate in individual feeding buckets. Complete urine and faecal collection was performed separately and individually before the morning feed. The total element analysis of samples was carried out using ICP-MS. An ANOVA model (two-factor factorial design in a randomized complete block) was performed in R software to test the influence of mineral forms and supplementary dose on the response variables.

**Results:** More than 50% of the Se came from the supplementation as opposed to the basal diet (Table 1). The Se partitioning in urine and faeces, as well as retention, was significantly affected by supplementation dose (Table 2). Faeces was the major excretion route (Table 2) and the concentrations of Se in faeces were significantly influenced by supplementation dose (Fig. 1 top). No significant impact of supplement forms was observed on the excretion and partitioning of Se, nor on that of other minerals observed (data not shown).

**Conclusion:** Up to the regulated upper levels of organic Se supplementation, the form of supplement showed no significant effect, whereas the different doses significantly affected Se partitioning in sheep and Se concentration in faeces. This difference in output and level within faeces and urine will influence Se profiles in soil and potential environmental flux within grazing livestock systems.

**Acknowledgments:** This work took place at Rothamsted Research, North Wyke, which is supported by the Biotechnology and Biological Sciences Research Council (BBSRC). Alltech Ltd provided funding for the lead author's PhD and the concentrate feed used in this study.



### 155. Effect of early life nutritional status on the transcriptional profile of hepatic tissue in Holstein Friesian bull calves

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**Application:** This study highlights the effect of enhanced nutritional status during early life on the transcriptional control of liver tissue in calves. Results from this study can be further utilised to augment early life nutritional management in cattle with potential latent benefits for overall lifetime performance.

**Introduction:** Improved nutritional status in calves during the early life period has been shown to induce positive effects for many economically important traits, including improved lifetime growth and carcass composition [1]. The liver has a central role in intermediary metabolism and therefore is likely to mediate the effect of improved nutrition. However, knowledge of the role of the liver towards this effect in young calves is currently lacking. Thus, the objective of this study was to evaluate genes differentially expressed in liver tissue of Holstein-Friesian bull calves following altered plane of nutrition from two to twelve weeks of age.

**Materials and methods:** Thirty Holstein-Friesian bull calves (mean age: 17.5 days; mean weight 48.8 kg) were assigned to either a high (H; n = 15) or moderate (M; n = 15) plane of nutrition. Calves on H were individually offered 10L of milk replacer, ad libitum concentrate and 500 g/day of hay, whilst those on M were offered 4L of milk replacer, 500 g of concentrate and 500 g hay per day. Calves were euthanised at twelve weeks of age, liver tissue was harvested and snap frozen for subsequent molecular analyses. Total RNA was isolated using the Qiagen

RNeasy Plus Universal kit. cDNA libraries were subsequently prepared using the Illumina TruSeq mRNA stranded kit and sequenced using the Illumina NovaSeq sequencing platform. Following QC reads were aligned to the bovine reference genome (ARS1.2) and quantified using STAR software. EdgeR software was then utilised to determine differentially expression genes (DEG; adj.P-value < 0.1; fold change > 1.5) between H and M cohorts. The gProfiler pathway analysis tool was used to assign biological function to the DEGs identified.

**Results:** During the ten week trial period, calves on H achieved a growth rate of 0.9 kg/day whilst those on M grew at 0.6 kg/day. Plane of nutrition resulted in a total of 174 DEGs with 74 genes up-regulated and 100 genes down-regulated in H compared with M animals. Pathway analysis revealed that both the retinol metabolism and steroid hormone biosynthesis pathways were enriched (adj.P-value < 0.01) as a consequence of improved nutritional status. Additionally, gene ontology terms including steroid hydroxylase activity, long-chain fatty acid metabolic processes and extracellular space were also significantly enriched (adj.P-value < 0.05).

**Conclusion:** Enhanced early life nutritional status contributed to altered metabolic and steroid hormone processes within the liver tissue, which may lead to subsequent improved animal performance. The results of this study provide an insight into the underlying molecular response of enhanced early life nutrition in liver tissue of bull calves.

**Acknowledgments:** This work was funded by Science Foundation Ireland (16/IA/4474).

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## 156. Identification of novel variants associated with feline hypertrophic cardiomyopathy (HCM) using targeted next-generation sequencing

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**Application:** Furthering understanding of feline hypertrophic cardiomyopathy (HCM) genetics will facilitate development of targeted DNA tests allowing identification of genetically susceptible individuals prior to onset of clinical signs, enabling preventive management measures and identification of novel drug targets.

**Introduction:** HCM is the most common inherited cardiac disease in humans and cats, characterised by significant clinical heterogeneity making diagnosis challenging. HCM can be fatal in cats through heart failure, systemic thromboembolism, or sudden arrhythmic death. In humans, HCM is typically caused by mutations in cardiac sarcomeric genes. In cats, 3 causative mutations have been found in 2 sarcomeric genes, *MYBPC3* and *MYH7* (Meurs et al., 2005, 2007; Schipper et al., 2019). Nevertheless, in most cats HCM aetiology is unknown. There is an urgent need for both earlier identification of cats at risk of HCM, and a better understanding of the factors driving disease severity to allow targeted therapy. The aim of the study was to identify variants associated with feline HCM-susceptibility and assess if the cat is a good study-model for the human disease.

**Materials and Methods:** Two targeted next-generation sequencing (tNGS) experiments were performed using an 18-candidate genes panel based on human HCM genes in Birman cats (n = 31) and an across-breed cat population (n = 44). Cat phenotyping was performed by echocardiography. Genome analysis toolkit for best practice were used for variant discovery. Allelic/genotypic frequencies of the identified variants were compared between cases and controls using the Chi-squared test (P < 0.05) and the PLINK algorithm. Genomic association

analyses including breed, age and sex as fixed effects were conducted to identify further variants of interest. Significance level was set at P < 0.05 and a Bonferroni correction for multiple testing was performed.

**Results:** The tNGS analysis revealed missense variants located in *CSRP3*, which encodes the muscle LIM protein involved in cardiomyocytes signalling, and *ENSFACAG00000040035* a protein coding gene with unknown function (Table 1). Several (>100) intronic variants with a statistically significant difference between cases and controls were identified in both studies.

**Table 1:** Missense mutations associated with feline HCM across two tNGS studies.

Results from the genomic association analysis revealed the presence of SNP markers with a suggestive significant association with HCM located in *ACTN2* and *MYL2* genes in the across-breed study and with *CSRP3*, *MYH7* and *ACTN2* genes in the Birman study.

**Conclusion:** Our study highlighted numerous variants associated with feline HCM in candidate genes based on the human literature. Novel variants of interest were identified for the first time in cats in the sarcomeric genes *MYL2*, *ACTN2*, *MYH7* and the non-sarcomeric gene *CSRP3*. Our results suggest that HCM in cats is a complex polygenic disease, with a similar genetic architecture to human HCM. However, these results should be validated further in a larger cat population.

**Acknowledgments:** The authors acknowledge funding from LIDo, BBSRC and PetPlan.

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Table 1  
Missense mutations associated with feline HCM across two tNGS studies.

Cohort	Gene	Nucleotide Change	Amino acid change	> Frequency
Cohort (i) Across-breed tNGS Study	<i>CSRP3</i>	c.133 T > C	p.Ile45Val	> Controls
Cohort (ii) Birman-breed tNGS Study	<i>ENSFACAG00000040035</i>	c.64G > C	p.Glu22Gln	> Cases

### 157. Examining the potential of using feeding behaviour halters and pedometers to predict dry matter intake and energy balance of dairy cows

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**Application:** Parameters derived from feeding behaviour halters and pedometer systems were unable to predict dry matter intakes or energy balance in dairy cows.

**Introduction:** As researchers attempt to improve precision feeding within milk production systems, there is interest in strategies to predict dry matter intake (DMI) and energy balance (EB) of dairy cows. A wide range of activity and behaviour monitoring devices are being developed for use on-farm. This study examined if parameters obtained from either feeding behaviour halters or pedometer systems could predict DMI and EB.

**Materials and methods:** Holstein dairy cows (110; 30 primiparous and 80 multiparous) were offered grass silage/concentrate diets from calving until 21 weeks post calving. All cows were fitted with a pedometer system (IceRobotics, Edinburgh, UK) from 7–22 weeks of lactation, which recorded the number of steps, lying time, lying bouts, standing time, standing bouts, and ‘motion index’ each day. Data was averaged over weeks 7–10, 11–14, 15–18 and 19–22 post-calving. RumiWatch halters (ITIN + HOCH, Switzerland) were used to assess a range of feeding/rumination parameters (‘other activity time’, ‘ruminate time’, ‘eat time’, ‘drink time’, ‘other chew’, ‘ruminate chew’, ‘total eat chew’, ‘drink gulp’, ‘bolus/hr’, ‘chews/min’, ‘chews/bolus’, ‘activity’, ‘head up time’, ‘head downtime’ and ‘temperature’) on 45 multiparous cows from weeks

10–21 of lactation. Mean data was collected at hourly time points, and daily data averaged over two periods (weeks 10–15 and weeks 16–21 post calving). For each of the periods outlined above, mean daily DMI and EB was determined, the latter according to equations described within Feed into Milk. For both DMI and EB a univariate analysis of each variable recorded by the pedometers and rumination halters were undertaken during each period. Furthermore, a stepwise regression analysis for both DMI and EB was performed using forward selection with backward elimination, according to a criterion based on variance ratios, in which all variables available from the pedometer and the rumination halter were offered to the model. Analysis were undertaken using Genstat (20th edition; VSN International Limited, Oxford, UK).

**Results:** Univariate analysis of pedometer data provided no relationships with a  $R^2 > 0.30$ . Similarly, within the univariate analysis of rumination data, with the exception of the relationship between total DMI and activity during Period 1 ( $R^2 = 0.48$ ), no other parameters had a  $R^2 > 0.30$ . Multivariate analysis of the pedometer data yielded no variables to explain either DMI or EB. Multivariate analysis of the rumination data yielded no variables to explain EB, but out of the 15 explanatory variables offered to the model, ‘drink gulp’ along with ‘activity’ (Period 1) and ‘total eat time’ (Period 2), could explain some of the variation in total DMI (Table 1), although  $R^2$  values were low.

**Conclusion:** Parameters derived from feeding behaviour halters or a pedometer system were unable to make any practical contribution to predicting DMI or EB in dairy cows.

**Acknowledgement:** This work was co-funded by DAERA and AgriSearch.

Table 1  
Using multivariate analysis to predict DMI (kg/d) in dairy cows using rumination halters.

Time period.	Response.	Constant.	Explanatory. Total Eat time (min/h)	R <sup>2</sup> Drink gulp.(n/h)	Activity.(n/h)	
1.(10-15 weeks post-calving)	Total DMI (kg/d)	33.5.(2.144)	-	0.1224.(0.05181)	-0.1041 (0.01855)	0.55.
2.16-21 weeks (post-calving)	Total DMI (kg/d)	29.98.(2.118)	-0.4546.(0.13010)	0.2018.(0.05370)	-	0.40.

### 158. The effect of sire muscling EBV and lamb finishing diet on the feed efficiency, carcass traits and meat quality of lambs

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**Application:** This study highlights the need for further research into the relationship between lamb performance traits and feed efficiency to support the development of new estimated breeding values (EBVs) for feed efficiency to improve production and resource efficiency. In addition, lambs on a grass diet only reached slaughter 11.4 days later than those supplemented with concentrates, thus grass finishing was cost effective in comparison to concentrate supplementation at the levels used in this study

**Introduction:** Genetic improvement in commercial sheep flocks is largely achieved through the purchase of breeding males. The adoption of EBVs into breeding programmes allows for targeted breeding strategies which can increase performance and the rate of genetic gain within flocks. EBVs for performance traits such as muscling and fat depth have become more widely available. Yet, little is known about the effect of sire muscling EBV and its interaction with finishing diet on lamb feed efficiency.

**Material and method:** This study consisted of 142 lambs in a four week, 2 (EBV) × 2 (diets) factorial design experiment. Lambs were sired by Texel and Suffolk rams of either high or low muscling EBV and allocated to one of two finishing diets; either grass only (G), or grass plus 200 g concentrates per day (GC). Lambs had a target start weight of 30–32 kg and were offered fresh grass (harvested daily using a zero grazer)

and concentrates (pellet of 16% crude protein) through an automated indoor feed system, which recorded daily individual intakes. Lambs were weighed weekly. Total dry matter intake (DMI) and live weight gain during the experimental period were used to calculate individual feed conversion ratio (FCR). Lambs that had not reached target slaughter weight of 42 kg after the four weeks, were returned to pasture and continued to be offered the experimental diet. At slaughter, carcass characteristics, including carcass weight, conformation and fat classification were recorded. A meat sample from the loin was retained for instrumental meat quality (ultimate pH, colour, cooking loss and Warner Bratzler Shear Force (WBSF)). Statistical analyses were conducted using the REML estimation method.

**Results:** There were no statistically significant interactions between EBV group and diet and thus only the main effects are presented in Table 1. Muscle EBV had no statistically significant impact on DMI, ADG or FCR. Lambs offered the GC diet reached target slaughter weight 11.4 days earlier than G lambs. Neither sire muscle EBV nor diet had any statistically significant effect on carcass traits or instrumental meat quality.

**Conclusion:** In conclusion there was no interaction between sire muscling EBV and finishing diet on lamb feed efficiency. Furthermore, the main effects of sire muscling EBV showed no impact on feed efficiency, carcass traits and meat quality of lambs.

**Acknowledgements:** This study was co-funded by DAERA, AgriSearch and Dunbia, with support from AHDB and Sheep Ireland as part of the Ram Compare UK project.

Table 1  
DMI and performance of lambs

	Muscle EBV.		SED.	P-value.	Diet.		SED.	P-value.
	High.	Low.			G.	GC.		
Total DMI (kgDM)	1.04.	1.03.	0.017.	ns.	0.94.	1.12.	0.016.	<0.001.
Final feedbox weight (kg) <sup>1</sup> .	36.63.	36.49.	0.300.	ns.	36.46.	36.67.	0.307.	ns.
Overall ADG (g/d)	126.7.	136.1.	16.66.	ns.	133.4.	129.4.	16.54.	ns.
Overall FCR.	14.13.	16.94.	7.401.	ns.	15.55.	15.52.	7.364.	ns.
Age at slaughter (days)	273.2.	271.5.	3.764.	ns.	278.0.	266.6.	3.725.	<0.01.

G = grass diet; GC = grass + 200gFW concentrates.<sup>1</sup> = final weight when lambs left the automated feed system.

### 159. Differences in affiliative and agonistic social interactions in a bachelor pack and family pack of European grey wolves (*Canis lupus lupus*)

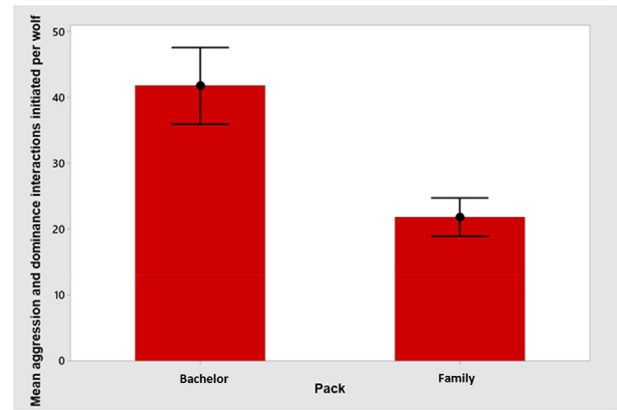
Maisie Geobey

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**Application:** Despite European grey wolves (*Canis lupus lupus*) being kept in UK zoos in both family packs and bachelor packs, there is little research on the effects that different social housing systems can have on the social behaviours that the wolves then exhibit; therefore, there could be gaps within the industry's knowledge about how these social groupings can impact the welfare of captive wolves [1].

**Materials and Methods:** A bachelor pack (n = 5) and a family pack (n = 12) were observed for 40 hours each in July and August 2019 over a period of eight consecutive days where five hours were collected on each day. An ethogram was constructed denoting affiliative, aggressive and dominance, and submissive behaviours based on criteria described by Essler et al. [3] and Ausband et al. [2]. Instantaneous focal behavioural sampling was employed to record the duration, initiator and receiver of all social interactions. Two-sample t-tests were conducted to compare group mean data.

**Results:** There was no significant difference in affiliative ( $t = 0.12$ ,  $p = 0.905$ ) or submissive ( $t = 1.33$ ,  $p = 0.254$ ) behaviours initiated by the wolves in each pack (see Fig. 1: interval plots to show the difference in the mean submissive (left) and affiliative (right) behaviours initiated by the wolves in each pack; error bars show standard error). A significant difference ( $t = 3.43$ ,  $p = 0.004$ ) was identified in the amount of aggressive and dominance behaviours initiated; the bachelor pack showed elevated levels of these behaviours (see Fig. 2: an interval plot showing the difference in the mean aggressive and dominance behaviours initiated by the wolves in each pack; error bars show standard error). Elevations in aggression can have negative consequences for the welfare of animals in captivity [4], therefore it was recommended that the bachelor pack should be monitored continuously to track the trajectory of dominance and aggression interactions occurring within the pack. The bachelor pack had only been together for six months at the time of the study after transitioning out of a family pack, which may explain their elevated aggression and dominance behaviours [5]. Therefore,

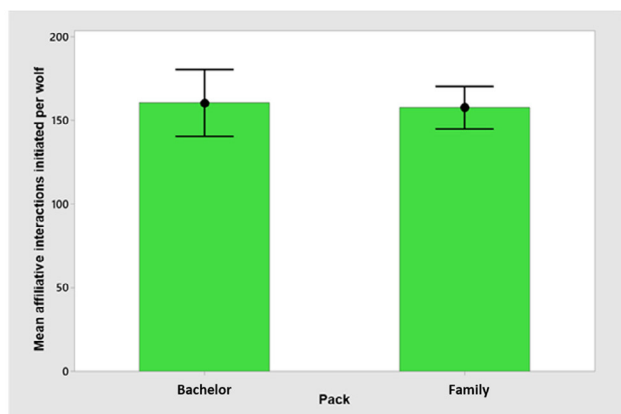
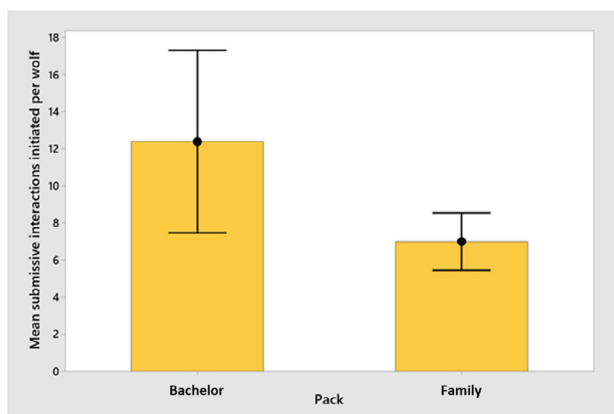


any future comparative studies should focus on contrasting already established bachelor and family wolf packs.

**Conclusion:** The study found a statistically significant increase in aggression and dominance in the bachelor pack compared to the family pack, indicating that further examination of this topic in multiple UK collections could be undertaken to better the welfare of zoo-housed wolves. The transition from family to bachelor pack may have contributed to this result, but more data should be analysed before making a more concrete conclusion.

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### 160. Lower feed quality does not impact on the nutritional composition of *Tenebrio molitor*

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**Application** Reducing feed quality and altering the environment of *Tenebrio molitor* (yellow mealworm) appears not to effect composition, indicating environmental and financial savings are possible.

**Introduction:** *Tenebrio molitor* larvae (TM) production methods and feed sources need to be sustainable for it to be an effective replacement feed ingredient. For small-scale producers, the relatively high-quality feed, chick crumb, is a common feed. However larger-scale producers commonly use lower quality wheat bran as a feed. This study compared the effects of feeding wheat bran or chick crumb in different environments on TM production and nutritional composition.

**Materials and Methods** TM (mini form) were obtained from Monkfield Nutrition Ltd and acclimatised for 3 days on wheat bran. The feeds utilised were either wheat bran (WB) or 20% chick crumb (CC) feed [1]. Two dark locations were used, an insect incubator ( $25 \pm 1$  °C, 60% relative humidity) or a room ( $18.5 \pm 3.5$  °C,  $40 \pm 3\%$  relative humidity). On day 0, TM were transferred to plastic containers ( $n = 200$ /container) and allocated to one of four treatment groups: WB-I (WB in incubator), WB-R (WB in room), CC-I (CC in incubator) and CC-R (CC in room). TM were fed *ab libitum*, with feed and water consumption measured and refreshed on days 4, 7 and 11. On day 14, final measurements were

recorded and any dead removed. After killing by cold exposure, samples were freeze dried and analysed for crude protein (EA1112 Elemental Analyser), total fat (Gerhardt Soxtherm) and total energy (Bomb Calorimeter). Data were analysed by two-way (feed  $\times$  location) ANOVA using Genstat (19th Edition). When significant ( $P < 0.05$ ) a post-hoc Bonferroni test was carried out.

**Results:** There were no significant differences in initial or final group live weights, dry matter, crude protein, total fat or total energy of the TM (Table 1). Total feed intake was significantly lower in the groups fed in the room, with the WB-R group the lowest. No differences in final live weights suggests that WB-R TM were the most feed efficient.

**Conclusion:** With no composition differences both CC and WB were equally suitable as feeds, but WB would be more cost effective and sustainable. Further work is needed to define TM production for optimal environmental and financial benefit.

**Acknowledgments:** The work was gratefully supported by funding from AB Agri, AB Vista and BBSRC.

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Table 1

Effects of feeding chick crumb (CC) or wheat bran (WB) for 14 days in an incubator (I) or room (R) on TM production and composition.

	WB-I	WB-R	CC-I	CC-R	SED	P Value
Initial Live Weight (g)	12.56	12.42	12.65	12.66	0.439	0.801
Final Live Weight (g)	18.27	19.19	19.53	20.19	0.785	0.817
Total Feed Intake (g)	39.37 <sup>a</sup>	31.29 <sup>b</sup>	38.82 <sup>a</sup>	34.76 <sup>c</sup>	0.622	<0.001
Dry Matter (gDM)	7.10	6.81	7.67	7.69	0.449	0.642
Crude Protein (g/gDM)	0.50	0.49	0.48	0.48	0.016	0.671
Total Fat (g/gDM)	0.26	0.24	0.24	0.22	0.014	0.660
Total Energy (MJ/gDM)	0.024	0.024	0.024	0.24	0.0003	0.838

\*This poster was presented as part of the Developing talent session at the BSAS conference.

### 161. Predicting milk quality traits in dairy cows from routinely available milk spectra using machine learning methods

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**Application:** Improved prediction of milk quality traits, however small, is useful for the milk processing industry to discriminate milk at the pre-processing stage enabling milk to be used for the process for which it is most suited.

**Introduction:** Mid-infrared spectroscopy (MIRS) is a tool widely used to predict the concentration of individual milk components and thanks to its characteristics (low cost, rapid and non-disruptive technique) it is used for collecting vast quantities of data at a (cow) population level. Partial least squares regression (PLSR) is the most popular method to relate MIRS data to novel milk and cow traits. In recent years, statistical machine learning (ML) methods have emerged as useful tools for prediction purposes in animal science. The novelty of the present study is the evaluation of modern statistical ML methods in predicting several cow milk quality traits from MIRS.

**Materials and methods:** Milk samples from 622 individual cows were collected across 12-month period from 7 different Irish research herds. The milk technological traits of rennet coagulation time (RCT), curd firming time (k20), curd firmness at 30 and 60 minutes (a30, a60), casein micelle size (CMS), pH, and heat stability as well as milk protein traits including alpha s1 casein, alpha s2 casein, beta casein, kappa casein, alpha lactalbumin, beta lactoglobulin A, and beta lactoglobulin B were quantified for each milk sample using gold standard methods. All samples were analysed by the same MilkoScan FT6000 to generate 1,060 transmittance values and high-noise-level regions were removed

from each spectrum. Eleven regression-based approaches were tested, such as PLSR, ridge regression (RR), least absolute shrinkage and selection operator (LASSO), elastic net, principal component regression, projection pursuit regression, spike and slab regression, random forests, boosting decision trees, neural networks (NN) and a novel post-hoc model averaging (MA) approach. The root mean square error (RMSEV) of a four-fold cross-validation was used for method comparison and the coefficient of determination ( $R^2$ ) was used to assess method accuracy.

**Results:** PLSR was the best prediction method for only pH; MA was the best prediction method for 5 of the 14 traits investigated. NN and RR were the best methods for 3 traits each, while LASSO was best for 2 traits. The well-established PLSR-based method performed competitively; nonetheless, ML methods led to a reduction in the RMSEV from between 0.18% (kappa casein) to 3.67% (heat stability). Although the ML methods investigated here slightly improved predictions over PLSR, the accuracy of the traits prediction remained moderate to low, with an  $R^2$  between 0.08 (CMS) to 0.65 (pH) for milk technological traits and between 0.19 (beta lactoglobulin A) to 0.47 (alpha s1 casein) for milk proteins.

**Conclusion:** The application of statistical ML methods improved the prediction of almost all milk traits. The MA approach had the lowest RMSEV most often and thus should be considered for such analyses.

\*This poster was presented as part of the Developing Talent session at the BSAS conference

**Acknowledgments:** The authors acknowledge funding from Science Foundation Ireland, Grant 18/SIRG/5562 and Grant 16/RC/3835 (VistaMilk).

## 162. Supplementing *Megathyrsus maximus* with seeds and whole pods of *Enterolobium cyclocarpum* reduced *in vitro* CH<sub>4</sub> gas emissions

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**Application:** The inclusion of the seeds and whole pods of *Enterolobium cyclocarpum* reduced CH<sub>4</sub> gas emissions compared to higher emissions produced by the pure grass treatment.

**Introduction:** Ruminant animal livestock contributes about 18% of the annual GHG emissions worldwide, owing to the fermentation processes that occurs in their rumen (Lascano & Cárdenas, 2010). Reducing this emission rate would fulfil the dual objective of reducing the negative impact of ruminant livestock on the climate and the energy in feeds that are lost as CH<sub>4</sub> gas. Tropical tree legume seeds and pods are feedstuffs with phenolic compounds that have CH<sub>4</sub> gas suppressing effect (Berhanu et al., 2019). However, most of these tropical tree legumes produce seeds and pods that go into waste yearly without proper utilization. This study, therefore, explored the effects that the inclusion of different proportions of seeds and pods of *Enterolobium cyclocarpum* Jacq. Griseb. has on reducing CH<sub>4</sub> gas volume.

**Materials and methods:** *Megathyrsus maximus* was harvested at 6 weeks, air dried, and milled. Seeds and whole pods of *E. cyclocarpum* were harvested, dried at 65 °C until constant weight, and milled. The separately milled seeds and pods were mixed with the milled *Megathyrsus maximus* (g/g) to create 5 composite treatment samples of the following proportions (60:40 and 80:20 grass and seeds, 60:40 and 80:20 grass and whole

pods, and 100 grass as the control sample) laid out in a completely randomized design. *In vitro* analysis was conducted on the treatment samples in triplicates using rumen buffer solution (Menke and Steingass, 1988). CH<sub>4</sub> was estimated 24 hours post-incubation by infusing 4.0 ml of NaOH into the incubated solution. This allowed for the absorption of CO<sub>2</sub> such that left-over gas was considered as CH<sub>4</sub>. The data on the total *in vitro*, CO<sub>2</sub>, and CH<sub>4</sub> gas volumes were collected and subjected to one-way analysis of variance and treatment means were separated using Tukey's HSD test.

**Result:** Treatment containing 40% seeds produced the highest ( $P < 0.05$ ) total *in vitro* and CO<sub>2</sub> gas volumes, while the other treatments produced comparable lower ( $P > 0.05$ ) total *in vitro* and CO<sub>2</sub> gas volumes with the control. At high and low proportions of the seeds and pods, the volume of CH<sub>4</sub> gas volume produced significantly ( $P < 0.05$ ) declined when compared to the control. There were no significant ( $P > 0.05$ ) differences in the CH<sub>4</sub> gas volume produced by the seed and pod treatments (Fig. 1).

**Conclusion:** Supplementing *M. maximus* with seeds and pods of *E. cyclocarpum* caused significant reductions in CH<sub>4</sub> gas emissions. Although high and low proportions of either the seeds or pods yielded similar effects on the CH<sub>4</sub> gas emission.

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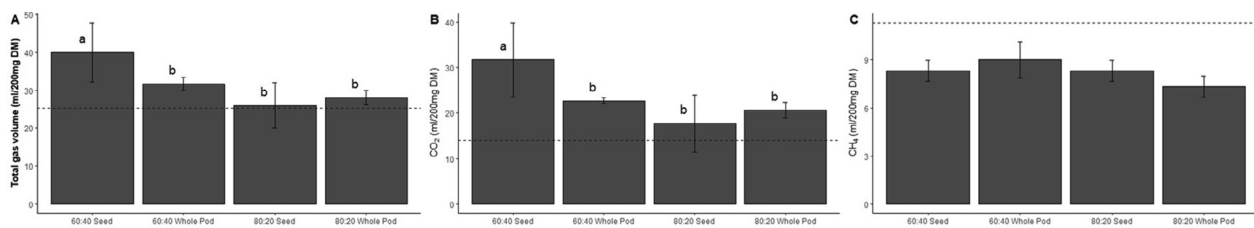


Fig. 1. Total *in vitro*, CO<sub>2</sub> and CH<sub>4</sub> gas volumes produced from composite treatment sampled made from *M. maximus* with proportions of seeds and pods of *E. cyclocarpum*. Dotted line = control treatment.



### 163. Equine exertional rhabdomyolysis: A phenotypically and genetically heterogeneous syndrome

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**Application:** The equine muscle disorder exertional rhabdomyolysis (ER) is an animal welfare concern that has substantial financial implications for equine industries. Breeders may inadvertently be selecting for the disease (Isgren et al., 2010), and identifying genetic markers could inform future breeding programmes. Recognition of specific clinical presentations across different breeds might reveal separate causes enabling more refined treatments and prophylaxis.

**Introduction:** Equine exertional rhabdomyolysis (ER) is a clinically heterogeneous myopathy. The clinical signs, which can include intermittent episodes of muscle fasciculation, myofibre necrosis, myoglobinuria, recumbency and even death, vary in character and severity between episodes and affected animals suggesting that ER might be a syndrome consisting of multiple distinct diseases (Valberg et al., 1999). Identifying phenotypic patterns might facilitate future studies into the genetic architecture, aetiopathogenesis, and treatment of these diseases.

**Materials & methods:** Signalment, clinical history and histological features from 196 ER cases from the muscle biopsy service database of the RVC Comparative Neuromuscular Diseases Laboratory were used in this study. Firstly, k-means clustering was performed on the clinical and histological features. Chi-square testing was used to identify features that varied significantly between clusters. Moreover, hierarchical clustering (Ward method) was implemented on cases and features. A support vector machine (SVM) algorithm was trained to assign 66 case horses with genotypes (Connemaras (CP): 16; Warmbloods (WB): 50;

670 k SNP array) to clusters. Case-control genome-wide association studies (GWAS) ( $n = 127$ , including 61 controls) were performed using GEMMA.

**Results:** The k-means clustering of clinical and histological features identified 4 clusters, with one not associated with any particular feature (termed the 'classic' cluster) and others associated with gait abnormalities, myalgia, paresis, ataxia and reluctance to move. A similar pattern was seen using clinical signs alone, with 5 clusters associated with repeated episodes, Thoroughbred breed, stiffness, myalgia, poor performance, gait abnormalities, ataxia and lethargy, and one 'classic' cluster. Hierarchical clustering clustered the clinical signs of poor performance, exercise intolerance, stiffness, reluctance to move, myalgia and gait abnormalities together. GWAS revealed a distinct genetic architecture for ER-susceptibility between classic and non-classic clusters (see Fig. 1).

**Conclusions:** Distinct patterns in ER disease phenotype are detectable, and specific clinical signs are consistently used to distinguish clusters. GWAS results supported that distinct phenotypic clustering reflects genetic differences between ER cases.

**Acknowledgments:** The authors thank the RVC Mellon Fund and the Thoroughbred Breeders' Association for funding this work and the horse owners and their veterinary surgeons for submission of clinical material.

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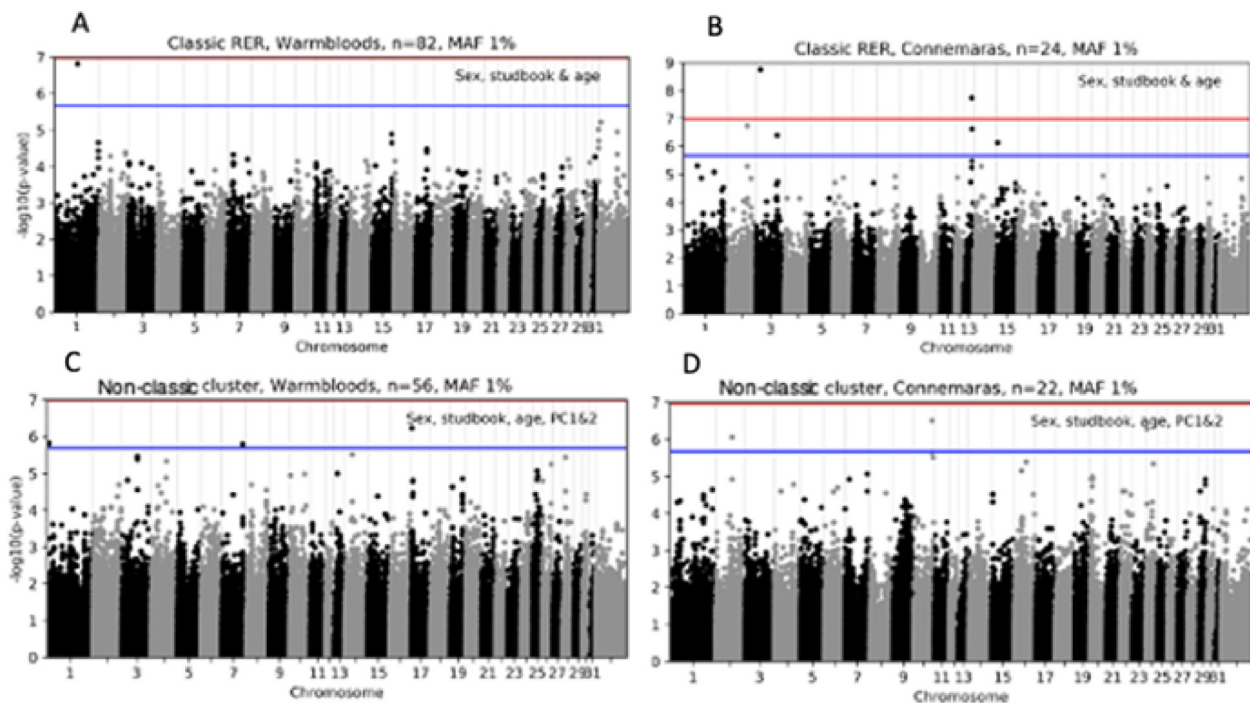


Fig. 1. Manhattan plot of marker location by  $-\log_{10}(P\text{-value})$  for GWAS A) across-breeds B) within CP and C) within WB. The red line indicates the genome wide significance threshold (Bonferroni-corrected), and blue line the suggestive threshold (one false positive per genome scan).

### 164. Effect of atypical reproductive cycles on key performance indicators of reproductive efficiency in Holstein Friesian dairy cattle

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**Application:** Reducing the incidence of atypical reproductive cycle length can increase efficiency of reproductive performance in dairy cattle.

**Introduction:** Good reproductive performance is essential to the economic efficiency of dairy production. Previous studies reported that 31 to 46% of dairy cattle had reproductive problems [1]. However, few studies have assessed reproductive cycle length of high yielding dairy cows and its effect on key performance indicators (KPI) according to calving pattern. This study aimed to assess the incidence and effect of atypical reproductive cycle length (ARCL) on subsequent KPI of reproductive efficiency in high yielding dairy cows that calve all year round (AYR).

**Materials and methods:** A total of 228 lactating Holstein Friesian dairy cows were selected from 320 cows at the Centre for Dairy Science Innovation (mean milk yield of herd 11,500 L/cow/year) at the University of Nottingham, UK. All cows had *ad libitum* access to a partial mixed ration (PMR) consisting of grass, maize and whole crop silages (50: 25: 25), soya and rapeseed meals, protected fat and mineral supplement. A pelleted concentrate was offered in a robotic milking system at 0.4 kg per litre of milk yield above 32 L/d (30 L/d for cows in Parity 1), up to a maximum of 12 kg/d. Milk samples were collected thrice weekly

between 10 and 120 d postpartum and analysed for milk progesterone (P4) concentration (ELISA, Ridgeway Science Ltd., Gloucestershire, UK). Results were used to evaluate onset of postpartum luteal activity (first P4 > 5 ng/mL) and luteal cycle length (d). ARCL was defined as delayed onset of luteal activity (>35 d), cessation of luteal activity (P4 < 5 ng/mL for > 14 d), or prolonged luteal cycles (≥20 d). Data were analysed using the ANOVA procedure of Genstat (20th Edition).

**Results:** The majority (69%) of cows had normal reproductive cycles, and 31% exhibited ARCL, which included delayed onset (6.6% of cows), cessation (4.4% of cows) and prolonged (20.2% of cows) luteal activity. Occurrence of ARCL was associated with longer intervals to first P4 rise and conception, lower conception rate at first service and longer calving interval (Table 1).

**Conclusion:** Atypical reproductive cycle length was associated with significant reductions in the KPI related to efficient reproductive performance in an AYR calving dairy herd. Incidence of ARCL was greater in older cows, but was not related to BCS at parturition. Investigation and elimination of factors that lower incidence of ARCL could improve the calving interval of the herd.

**Acknowledgments:** I.A. was in receipt of a scholarship from the government of the Kingdom of Saudi Arabia., King Faisal University Saudi Arabia.

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Table 1

Number observations and mean ( $\pm$ SE) parity, body condition score, interval to first progesterone (P4) rise, first insemination and conception, and conception rate to first insemination for cows that exhibited normal and atypical cycle lengths.

	Cycle length		P value	Difference
	Normal	Atypical		
Observations, No.	157	71	-	-
Parity, No.	2.7 (0.12)	3.2 (0.25)	< 0.050	0.52 (0.019)
Body condition score at parturition, 0 to 5	2.6 (0.04)	2.7 (0.07)	NS	0.08 (0.001)
Interval to first P4 rise postpartum, d	27.6 (0.80)	37.7 (2.61)	< 0.001	10.1 (0.45)
Interval to first insemination, d	66.2 (1.71)	77.7 (3.11)	< 0.001	11.5 (0.21)
Interval to conception, d	91.3 (5.41)	129.4 (14.02)	< 0.001	38.1 (4.51)
Conception rate at first insemination, %	38.2	23.8	< 0.050	- 14.4 ( - )
Calving interval, d	374.3 (22.3)	412.4 (40.60)	< 0.001	38.1 (1.46)

### 165. Field indicators of dry matter yield: Are sward harvest height and residue length good predictors of dry matter changes in *Megathyrus maximus*?

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**Application:** Harvesting tropical grasses at a given sward height and residue could serve as a predictive dry matter determination tool for making rapid grazing management decisions.

**Introduction:** In tropical Africa, traditional yield determination techniques are time wasteful and not feasible for making rapid grazing management decisions. On the other hand, field indicators of dry matter yield from sward traits could suffice as an easily understandable approach that can be carried out seamlessly [1]. However, field dry matter yield indicators must be developed under contrasting environmental conditions to effectively model the association between sward traits and dry matter changes. To this end, we conducted a study to determine how the changes in the dry matter yield of *M. maximus* is associated with some defoliation management decisions (harvest sward height and residue) that are known to influence dry matter accumulation.

**Materials and methods:** We investigated the yield changes in *Megathyrus maximus* during the 2020 April – November growing season at the Federal University of Agriculture, Abeokuta, Ogun State, Nigeria. The study was laid out as a randomized complete block design with three blocks. Two sward defoliation heights (80 and 100 cm) and three sward defoliation residue lengths (10, 20, and 30 cm) were considered as factors for the experiment. This totalled six treatments replicated three times per block. Plots measured 3 m × 4 m with a 0.8 m buffer

zone between plots and a 1.5 m buffer zone between blocks. All plots received a basal fertilizer dose of 200 kg N/ha two weeks before planting and 400 kg N/ha after the first 2 harvests. The plants were harvested when they attained 80 and 100 cm heights and were cut following the sward residue treatment structure (10, 20, and 30 cm). Four total harvests were made. Dry matter yield was determined at each harvest by weighing the total herbage yield from a plot. Fresh sub-samples weighing 300 g were oven-dried at 65 °C until constant weight, and the dry matter percentage was multiplied by the fresh weight value to determine the dry matter yield. Yield values were extrapolated to tonnes/hectare. Yield values from the four harvests were aggregated and subjected to two-way analysis of variance. Differences in treatment means were separated using the Tukey HSD posthoc test.

**Results:** The dry matter yield values obtained at the 80 cm and 100 cm sward heights were not significantly different ( $P > 0.05$ ); however, dry matter yield values significantly ( $P < 0.05$ ) declined as the harvest sward residue increased, with values ranging from 1.43 t/ha for harvests at 30 cm sward residue length to 2.72 t/ha for harvest at 10 cm sward residue lengths (Fig. 1).

**Conclusion:** The sward heights produced similar dry matter yield, but increasing the sward residue decreased dry matter accumulation in *M. maximus*.

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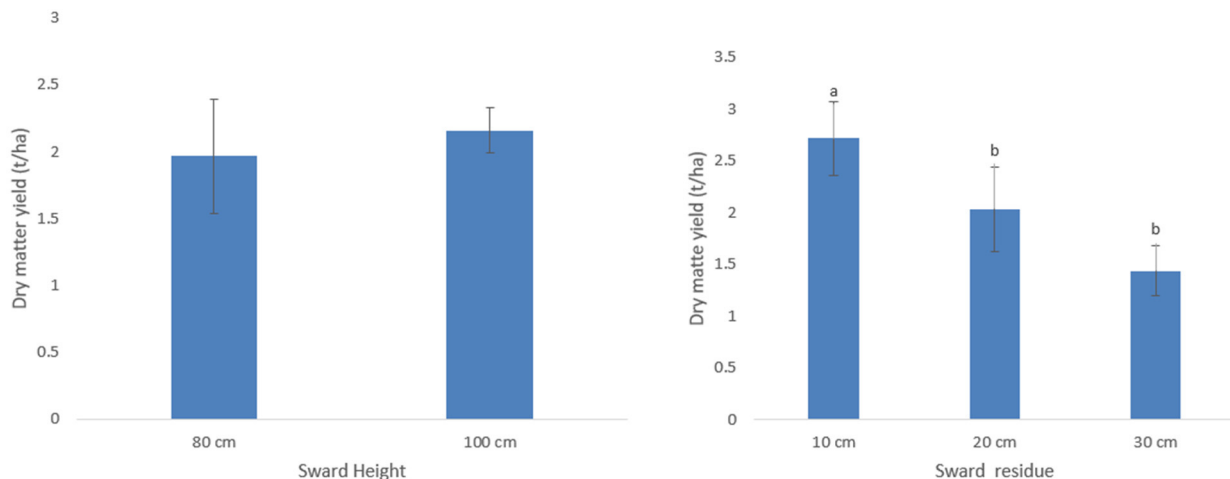


Fig. 1. Effect of harvest sward height and sward residue lengths on the dry matter yield of *M. maximus*

### 166. Relationship between some physiological growth traits and dry matter yield of 27 drought resistant maize accessions

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**Application:** Using physiological growth traits to predict dry matter changes in maize would aid in making seamless and rapid harvest decisions

**Introduction:** Dry matter yield estimation based on changes in plant growth traits is essential for making rapid harvest decisions. Plants often divert accumulated biomass to other plants parts to allow them to maintain high production capacity potential (Hokmalipour and Darbandi, 2011). This implies that a plant's physiological growth traits could serve as important predictors for measuring changes in its dry matter. Such predictive models would offset the cumbersome and time-consuming process associated with traditional dry matter determination techniques. However, such models must be developed under contrasting environmental conditions. As a result, we conducted a study that measured the association between some physiological growth traits and dry matter yield of 28 maize accessions under drought and well-watered conditions.

**Materials and Methods:** The study was conducted during the July 2019 – February 2020 growing season at the Federal University of Agriculture, Ogun State, Nigeria. 27 drought tolerant open-pollinated maize accessions seeds were sourced from the International Institute of Tropical Africa (IITA). A locally sourced variety was used as a local check. Planting occurred twice, under well-watered (July) and drought (October) conditions. In each planting season, the maize seeds were planted at two seeds per hole in three blocks with 28 plots per block (1 plot per ascension). Three physiological growth traits (plant height, leaf area, and the number of ears per plant) were measured during the growth stage on 15 plants per accessions (5 plants on each plot in each block). The plants were harvested at the ripe phonological stage (~14 weeks after planting). The fresh yield was determined at harvest and dry matter yield ( ) was determined by drying sub-samples of the fresh harvest (300g) at 65 °C until constant weight and multiplying the dry

matter percentage value by the fresh weight value. Dry matter yield values were extrapolated to tonnes/hectare. The relationship between DMY and the growth traits were measured using Pearson's product-moment correlation test.

**Results:** All but the correlation between dry matter yield and plant height under drought conditions were significant. Leaf area had the highest correlation with dry matter yield under the drought and well-watered conditions. Other measured associations with dry matter yield were moderate and low (Table 1).

Table 1: Correlation between the dry matter yield and some morphological traits of 28 maize accessions under drought and well-watered conditions

	DMY	LA	PH	NOEPP
DMY	1	<b>0.80***</b>	<b>0.50**</b>	<b>0.50**</b>
LA	0.62***	1	<b>0.67***</b>	<b>0.77***</b>
PH	0.36 <sup>NS</sup>	0.65**	1	<b>0.73***</b>
NOEPP	0.48**	0.57*	0.59***	1

Upper diagonal: well-watered condition; Lower diagonal: Drought condition

DMY: dry matter yield; LA: leaf area; PH: plant height; NOEPP: Number of ears per plant; \*\*\*:  $p < 0.001$ ; \*\*:  $p < 0.01$ ; \*:  $p < 0.05$ ; <sup>NS</sup>: Not significant ( $p > 0.05$ )

**Conclusion:** Leaf area had the highest correlation with dry matter yield under drought and well-watered conditions. The relationship between dry matter yield and the other traits were moderate to poor.

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### 167. Reducing the dietary protein content in dairy cow diets based on lucerne and maize silage does not affect performance but improves nitrogen use efficiency

Mohammed Rashed Chowdhury, Robert G. Wilkinson, Liam A. Sinclair

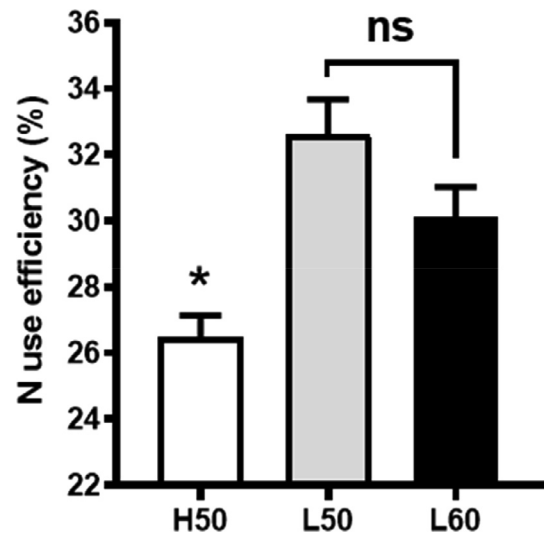
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**Application:** Low protein diets based on a 50:50 dry matter (DM) basis of lucerne to maize silage can improve nitrogen use efficiency without affecting performance unless the lucerne silage inclusion rate increases to 60% of the forage DM.

**Introduction:** Reducing the dietary crude protein (CP) concentration to around 150 g/kg DM in dairy cow diets can increase the nitrogen utilisation without affecting milk performance if the diets are formulated to supply sufficient metabolizable-protein [1]. Previous studies by Sinclair et al. [2] reported that lucerne silage could contribute up to 0.6 of the forage component (DM-basis) when fed along with maize silage without affecting performance. However, the effect of feeding a low protein diet based on lucerne/maize silage on performance and nitrogen-use-efficiency (NUE) is unclear.

**Materials and methods:** Eighteen Holstein-Friesian dairy cows yielding  $46.5 \pm 4.78$  kg milk/d with a mean live weight of  $705 \pm 59$  kg were used in a 3x3 Latin-square design with 3 periods, each of 28-d with a 21-d adaptation and a 7-d sampling period. The cows were randomly allocated to 1 of 3 dietary treatments: 1) 175 g CP/kg DM with 50:50 lucerne to maize silage (H50); 2) 150 g CP/kg DM with 50:50 lucerne to maize silage (L50); 3) 150 g CP/kg DM with 60:40 lucerne to maize silage (L60). The diets were fed as a total mixed ration and contained a forage to concentrate ratio of 52:48 (DM basis), and were formulated to be iso-energetic and contain a similar MP content. Milk and blood samples and live-weight measurement were taken during the sampling week of each period. Performance data was analysed as a Latin-square design and plasma parameters were analysed as repeated measures ANOVA in Genstat 18.1 (VSN International Ltd, Oxford, UK).

**Results:** Mean DM intake was 2 kg/d lower when cows were fed L50 or L60 compared to H50, and milk yield was 2 kg/d lower in cows fed L60 compared to H50 (Fig. 1; Table 1). There was no effect of diet on milk fat



content but milk protein, milk and plasma urea concentrations were higher in cows fed H50 compared with those fed L50 or L60. In contrast, the apparent NUE was approximately 5% units higher in cows receiving L50 or L60 compared to those fed H50 (Fig. 2).

**Table-1.** Performance of dairy cows fed lucerne/maize silage-based diets<sup>1</sup>.

Item	Diets <sup>1</sup>			SEM	Pvalue
	H50	L50	L60		
Milk yield, kg/d	40.9 <sup>a</sup>	39.8 <sup>ab</sup>	38.9 <sup>b</sup>	0.43	0.010
Milk protein, g/kg	30.7 <sup>a</sup>	30.2 <sup>ab</sup>	30.1 <sup>b</sup>	0.14	0.024
Milk fat, g/kg	38.3	38.3	39.4	0.86	0.553
Milk urea, mg/dl	24.0 <sup>a</sup>	16.6 <sup>b</sup>	17.0 <sup>b</sup>	0.53	<0.001
Plasma urea, mmol/l	4.96 <sup>a</sup>	3.51 <sup>b</sup>	3.56 <sup>b</sup>	0.124	<0.001

<sup>1</sup>H50 = 175 g CP/kg DM with 50:50 lucerne to maize silage; L50 = 150 g CP/kg DM with 50:50 lucerne to maize silage; L60 = 150 g CP/kg DM with 60:40 lucerne to maize silage on dry matter basis.

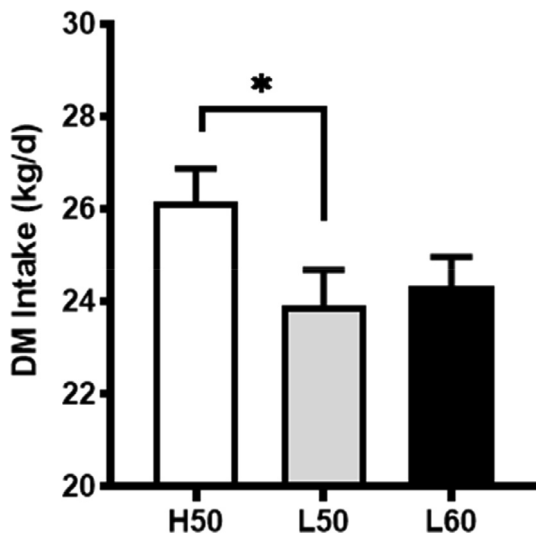
Different superscripts within a row differ significantly ( $P < 0.05$ )

**Conclusions** Reducing the dietary CP concentration from 175 to 150 g/kg DM in a 50:50 lucerne to maize silage-based ration had no effect on performance but improved NUE. However, reducing the same CP concentration in a 60:40 lucerne to maize silage-based ration reduced milk yield and milk protein concentration but did not affect feed intake.

**Acknowledgments:** The authors thankfully acknowledge funding from the AHDB.

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### 168. The application of near infrared spectroscopy to determine concentrations of organic constituents in equine faeces

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**Application:** Faecal near infrared spectroscopy (NIRS) is utilized to predict dietary intake and nutrient utilisation in animals, thus optimizing management practices and feeding regimes.

**Introduction:** Faecal NIRS has been explored in ruminant species, however few studies have analysed equine faeces. The objective of this study was to investigate the application of NIRS to determine concentrations of organic constituents, specifically dry matter, ash, and organic matter, in equine faeces. The impact of sample particle size on the accuracy of NIRS calibrations was examined through scanning each sample milled to two particle sizes, 1 mm and 0.5 mm. Particle size can cause increased scatter in spectral data, therefore, particle uniformity throughout the sample is vital [1].

**Materials and methods:** Freshly voided faecal samples ( $n = 60$ ) were dried and milled to pass a 1 mm mesh, a subsample of each was milled to pass a 0.5 mm mesh. Laboratory analysis was conducted on the samples to determine quantities of dry matter, ash, and organic matter. Samples were scanned at both particle sizes in reflectance mode using a Foss NIRS DS2500 system. Spectra were pre-processed with scatter corrections including standard normal variate, detrend, standard normal variate and detrend, or no correction along with mathematical treatments (1,4,4,1; 2,4,4,1; 2,4,4,2; 2,6,4,1 and 3,5,5,1). The equation with the lowest standard error of cross validation, and highest  $R^2$  and one minus variance ratio was considered the best predictive equation. Paired t-tests and bivariate Pearson correlation tests were conducted to analyse validation statistics. Multivariate general linear models were

developed to assess the effects of pre-processing techniques on chemometric data.

**Results:** Validation statistics for samples milled to 1 mm produced superior  $R^2$  and ratio performance deviation (RPD) values. Dry matter was successfully predicted ( $R^2 = 0.65$ , RPD = 1.56), however, ash and organic matter predictions were less satisfactory ( $R^2 = 0.54$ , RPD = 1.23). NIRS predictions for 0.5 mm samples were inferior, predictions for dry matter ( $R^2 = 0.23$ , RPD = 1.10) and ash and organic matter ( $R^2 = 0.39$ , RPD = 1.29) were unreliable. Mean concentrations determined by laboratory analysis included dry matter = 20.28%, ash = 8.61%, and organic matter = 91.39% with standard deviations of 2.68%, 2.57%, and 2.57% respectively. NIRS predictions for 1 mm samples were strongly correlated to reference values (dry matter  $r = 0.81$ , ash & organic matter  $r = 0.74$ ) compared to 0.5 mm sample predictions that were moderately correlated (dry matter  $r = 0.48$ , ash & organic matter  $r = 0.63$ ).

**Conclusions:** This study indicates that equine faecal material is suitable for NIRS. Findings suggest that there is no added benefit of a reduction in particle size from 1 mm to 0.5 mm to the accuracy of NIRS when analysing equine faeces.

**Acknowledgments:** This investigation was conducted in the Department of Biological Sciences, University of Limerick, Ireland. The authors thank Clonshire Equestrian Centre and Croom House Stud, Limerick for providing equine faecal samples.

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### 169. Effects of dietary supplements rich in docosahexaenoic acid on *in vitro* fermentation and microbial composition of lamb rumen fluid

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**Application:** The results suggest that the inclusion of microalgae or oil rich in DHA could be fed to lambs with no impact on ruminal fermentation.

**Introduction:** Microalgae is a potential alternative dietary source of polyunsaturated fatty acids (PUFA), especially omega-3, that could improve meat quality of the ruminants [1]. However, the impact of that dietary supplementation on the ruminal microbiota is still unclear. This study aims to evaluate the impact of the supplementation of a DHA rich microalgae and oil extracted from the microalgae on the *in vitro* fermentation patterns and microbial composition of lamb's rumen fluid Fig. 1.

**Materials and methods:** The rumen fluid of three 8-month old Texel-cross lambs fed a 50:50 energy-based control diet of grass silage and balanced concentrates was sieved and mixed with Van Soest buffer in a 1:3 ratio (v/v). The preparation was anaerobically incubated in triplicates with milled silage (0.8%) (control treatment), milled silage (0.8%) + microalgae (0.6%) (algae treatment) and milled silage (0.8%) + oil (0.5%) (oil treatment) into ANKOM gas pressure monitor jars for 48 h (39 °C – 25RPM). Samples jars were collected at four time points (0, 7, 24, 48 h) and analysed as follows: pH – each incubation time, ammonia concentration – Chaney and Marbach method (1961), microbial protein – Bradford method (1976), gas pressure - automatically, volatile fatty acid – gas chromatography, protozoa counts – Sedgewick rafter

counting slide and DNA 16S sequences – MGnify pipeline (ENA). Statistical tests (ANOVA and pairwise T test or Kruskal Wallis and pairwise Wilcoxon test according to the data distribution) for each parameter, potential correlations and graphics were run on R (4.0.3).

**Results:** The pH remained similar amongst the treatments ( $P = 0.99$ ) and decreased over time (6.40 to 6.05). The ammonia concentration increased over time (109 mg/L, 130 mg/L, 138 mg/L to 521 mg/L, 556 mg/L, 516 mg/L for control, algae and oil respectively) but no significant treatment effect was observed ( $P = 0.96$ ). A tendency for higher microbial protein was observed for the algae and oil compared to the control treatment ( $P = 0.06$ ). Total gas production increased over time but was not statistically different between treatments ( $P = 0.86$ ). Volatile fatty acids concentration increased over time but were similar between treatments ( $P = 0.82$ ). Shannon diversity index ranged from 1.95 at 0 h to 2.31 at 48 h regardless of the treatment. The protozoa count decreased over time and was similar amongst the treatments ( $P = 0.95$ ). The most predominant families were *Christensenellaceae*, *Prevotellaceae*, *Lachnospiraceae* and *Ruminococcaceae*, with a significantly higher presence of *Prevotellaceae* ( $P = 0.02$ ) but a significantly lower presence of *Lachnospiraceae* ( $P = 0.01$ ) in the control compared to the supplemented treatments.

**Conclusion:** Both microalgae and oil derived supplementation showed similar fermentation patterns to the control treatment. The changes in the microbial diversity at the family level between the control treatment and the supplemented treatments did not impact the measured fermentation parameters.

**Acknowledgements:** The authors acknowledge funding from Devenish Nutrition and the Department for the Economy.

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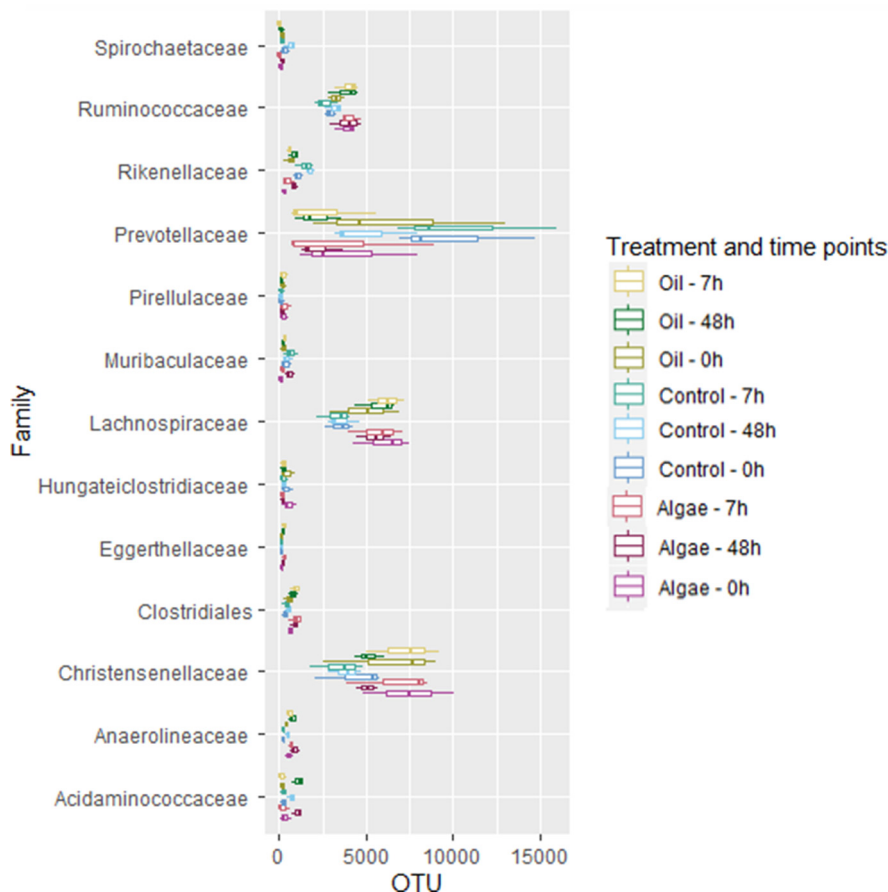


Fig. 1. 16S DNA OTU in relation to the 95% most predominant families organised by treatment and incubation time.

### 170. Investigating equestrians' perceptions of 'horse happiness': An exploratory study

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**Application:** Some equestrians' subjective interpretation of behaviours indicative of 'happiness' in horses differs from known characteristics of positive affective states in equines. This may lead to misperception of horse behaviour which could compromise equine welfare and negatively affect horse-human relationships.

**Introduction:** Happiness is a subjective feeling and associated with chronic positive affective states that can indicate good welfare in animals. Since animals, including horses, are unable to self-report 'happiness', equestrians are required to interpret their horses' emotional states, which may not necessarily reflect reality. Unawareness of or inability to correctly identify and interpret negative affective states in horses may jeopardise equine welfare. This study investigated equestrians' perceptions of 'horse happiness' in general and in given sample scenarios.

**Materials and Methods:** A cross-sectional SurveyMonkey™-survey comprising 25 questions was distributed through equestrian-related social media sites. The survey consisted of three sections: Demographics (e.g., gender); participants' perception of horse happiness (e.g., "How happy is your horse in general?"); scenarios participants considered most important to their horses' happiness (based on the five domains welfare model) (e.g., all-day turnout vs. being stalled). Participation was voluntary and minors were excluded. A total of 332 complete surveys were analysed using Statistical Package for Social Sciences. Frequency analysis was conducted within each factor category (e.g., horse ownership). Pearson chi-squared test for independence and

Fisher's exact test determined if differences occurred between the tested factor categories. Significance was set at  $P < 0.05$ .

**Results:** The majority of respondents believed that they would be able to recognise when their horses are 'happy' ( $P < 0.0001$ ). A large respondent proportion believed that their horses' overall 'happiness' states could be described as "rather happy" ( $P < 0.0001$ ). Respondents who owned horses more frequently believed they could recognise 'happiness' in their horses ( $P < 0.005$ ) compared to respondents not owning horses. Increased numbers of respondents who indicated that they are able to recognise when their horses are 'happy' also believed that their horses are happy when they **interact** with them ( $P < 0.001$ ); when they **ride** them ( $P < 0.0001$ ); when they **work** them ( $P < 0.0001$ ). A large proportion of respondents believed that "very 'forward' or energetic horses every time they are being ridden" can be best described as "rather happy" ( $P < 0.0001$ ). A large respondent number believed that "daily full-body interactions with (an)other horse(s)" and "full day (24/7) turnout with (an) other horse(s)" are more important to 'horse happiness' than "daily grooming sessions/scratches by owner/familiar human" or "daily exercise and eight hours turnout" ( $P < 0.0001$ ) respectively.

**Conclusion:** These results suggest that some equestrians may have overconfidence in interpreting equine affective states. Furthermore, some respondents may incorrectly perceive behaviours potentially indicating negative emotions (e.g., stress, pain) as indicators of 'happiness'. However, a large proportion of study participants seemed to have been aware of the positive impact of, for instance, full-body social contact to conspecifics and turnout/voluntary locomotion, may have on horses' affective states and, ultimately, welfare. Further in-depth studies involving larger sample sizes are required to explore equestrians' interpretation of horse behaviours in relation to their perceived affective states.

**Acknowledgements:** None.



### 171. A cross-sectional study of foot diseases and lesions in intensive dairy sheep farms in Greece

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**Application:** Foot diseases and lesions thereof constitute main causes of lameness in high-yielding, intensively reared dairy sheep, leading to remarkable production losses.

**Introduction:** In intensive dairy sheep farms, foot-related lameness has emerged as a significant health and welfare challenge. The objective of the present study was to assess and compare the prevalence of footrot, white line disease (WLD), hoof overgrowth and the resulting lameness, under various intensive dairy sheep farming systems in Greece.

**Materials and methods:** Thirty intensive dairy sheep farms were initially involved in the study (n = 10,630 ewes). Farmers were interviewed on-site using a structured questionnaire, and general information regarding farm characteristics were recorded. A multivariate statistical approach was used to produce the most representative typology of intensive farming systems and assign the farms into the ideal number of clusters. For this reason, principal component analysis was used with the retained principal components explaining ca. 86.5% of the observed variation. Afterwards, hierarchical cluster analysis was applied to determine the ideal number of clusters that the farms should be allocated into during k-means cluster analysis. From each cluster, three farms and 100 milking ewes per farm were randomly selected and enrolled in the study (n = 600 ewes). Occurrence of hoof wall overgrowth, foot lesions and lameness were recorded during routine foot-trimming, and the prevalence of foot lesions at the animal, the foot and the claw level were estimated. Comparisons between the clusters regarding the occurrence of foot lesions were made in SPSS v23 using the chi-square test.

Table 1

Prevalence of WLD and Footrot at the foot (n = 2400) and the claw (n = 4800) level, in the examined ewe population and the two clusters.

	Foot disease	Prevalence at the foot level (Number of feet affected)	Prevalence at the claw level (Number of claws affected)
Cluster 1	WLD <sup>1</sup>	12.5% (150)	7.1% (171)
	Footrot	2.8% (33)	1.5% (36)
Cluster 2	WLD	14.7% (176)	8.6% (206)
	Footrot	1.6% (19)	0.8% (20)
Overall	WLD	13.6% (326)	7.9% (377)
	Footrot	2.2% (52)	1.2% (56)

<sup>1</sup>WLD: White Line Disease

**Results:** Farms were classified into two clusters (15 farms each), with the major assignment factor being annual milk production per ewe (mean ± standard deviation was 398.7 ± 40.55 and 300.7 ± 59.91 kg for cluster 1 and 2, respectively). Overall, 9.0% (54/600) of the examined ewes were lame, whereas, prevalence of footrot and WLD at the animal level were 8.0% (48/600) and 37.7% (226/600), respectively. Prevalence of WLD and footrot at the foot and the claw level are presented in Table 1. Hoof overgrowth and cracks on the hoof wall were observed in 99.3% (596/600) and 15.3% (92/600) of the ewes, respectively. At the animal level, prevalence of lameness was significantly higher in cluster 1 (11.3%, 34/300) compared to cluster 2 (6.7%, 20/300) [ $\chi^2$  (1, n = 600) = 3.99, P < 0.05]. Similarly, both at the foot and at the claw level, hoof overgrowth was significantly higher in cluster 1 compared to cluster 2 [ $\chi^2$  (1, n = 2400) = 15.39, P < 0.001] and [ $\chi^2$  (1, n = 4800) = 16.01, P < 0.001, respectively].

**Conclusion:** In intensively reared dairy sheep, milk production seems to be a challenging factor for the foot health. Prospective studies to assess the long-term effects of foot diseases and lesions are crucial to better understand the epizootiology of foot-related lameness.

**Acknowledgments:** This research is co-financed by Greece and the European Union (European Social Fund- ESF) through the Operational Programme «Human Resources Development, Education and Lifelong Learning 2014-2020», project code MIS5048473.

## 172. A genome wide association study of hypertrophic cardiomyopathy in cats

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**Application:** Improving knowledge on hypertrophic cardiomyopathy (HCM) genetics will facilitate preventative management for genetically predisposed individuals. Here we present the first GWAS study performed for feline HCM.

**Introduction:** HCM is the most common inherited cardiac disease in humans and cats, characterised by significant clinical heterogeneity making diagnosis challenging. In humans, HCM is typically caused by mutations in cardiac sarcomeric genes, however 40% of cases cannot be explained by variants located in these genes. In cats, 3 causative HCM mutations have been found in 2 sarcomeric genes, *MYBPC3* and *MYH7*[1–3]. There is an urgent need for earlier identification of cats at risk of HCM, and enhanced understanding of factors driving disease severity to allow targeted therapy. Until now only candidate gene approaches have been implemented to identify causative variants in cats with limited success. Here, we implemented a genome wide association (GWAS) study to search for HCM associated variants in the cat population.

**Materials and Methods:** We performed a GWAS study for HCM-susceptibility (n = 120, 93 cases and 27 controls) across different cat breeds (Bengal (n = 18), Norwegian Forest Cat (NFC) (n = 15), Sphynx (n = 5) and Domestic shorthair (DSH) (n = 82)). Cat phenotyping was performed by echocardiography. Genotyping was performed using the

feline 60 K SNP Illumina Infinium iSelect DNA array. Quality control included minor allele frequency > 0.05, call rate > 95% and Hardy-Weinberg equilibrium ( $P > 10^{-6}$ ). The GEMMA algorithm was used to perform the GWAS using a standard univariate linear mixed model in which breed, age and sex were fitted as fixed effects and the genomic relatedness matrix among individuals was fitted as a polygenic random effect. Genome-wide significance level and suggestive levels were set at  $P < 0.05$  and one false discovery per genome scan, respectively, and a Bonferroni correction for multiple testing was applied.

**Results:** GWAS revealed the presence of one SNP located on chromosome B2 close to *CITE2* gene associated with HCM at the genome-wide level and numerous suggestive significant associations on chromosomes A1, B1, B4 and D2 (Fig. 1). *CITE2* gene has previously associated with cardiac disease in humans.

**Conclusion:** According to our results a number of SNPs across different chromosomes associated with feline HCM, suggesting that the genomic architecture of the disease is complex polygenic. This study is on-going: we are currently phenotyping and genotyping more cats to increase our sample size (n = 200) and empower the GWAS to discover more SNPs of interest and validate these preliminary results.

**Acknowledgments:** The authors acknowledge funding from LIDo, BBSRC and PetPlan.

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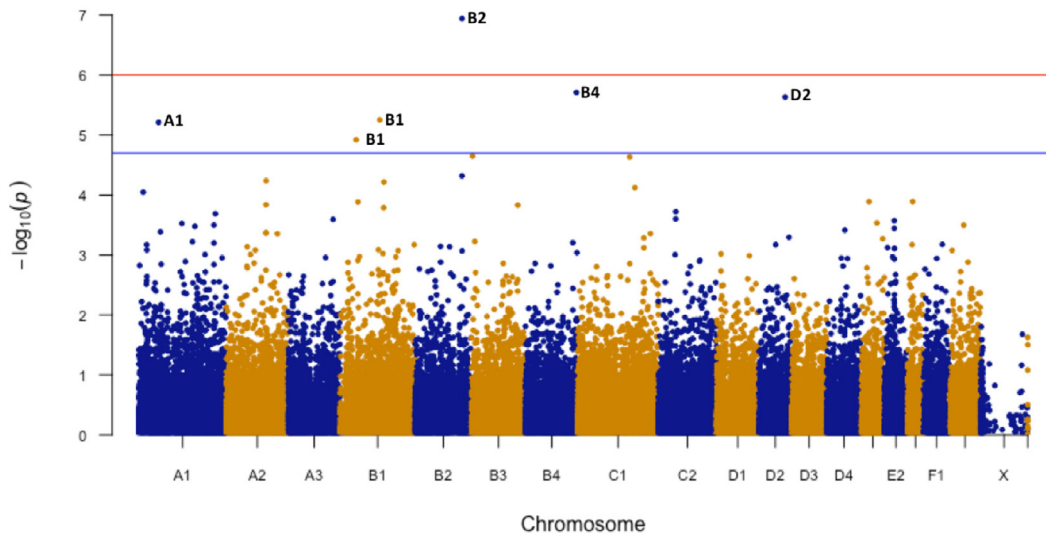


Fig. 1. Manhattan plot displaying results of the genomic association with HCM in 120 cats across breeds. Genomic location is plotted against  $-\log_{10}(p)$ . Red and blue lines, refer to genome-wide significant and suggestive significant thresholds, respectively.

### 173. Investigating the structural effects of MYBPC3 variants causative for feline and human hypertrophic cardiomyopathy (HCM)

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**Application:** The p.R820W variant found in the gene encoding cardiac myosin binding protein (MYBPC3) is known to cause hypertrophic cardiomyopathy (HCM) in both humans and cats through an unknown mechanism. Our study showed that this variant causes a loss of domain stability and may result in pathogenic structural modifications leading to the development of the disease. Improved understanding of such modifications could provide useful insights for future drug-discovery.

**Introduction:** HCM is the most common, heritable heart disease in cats and humans with prevalence of 1/7 and 1/500 respectively, characterised by excessive thickening of the left ventricular walls. The disease impacts feline welfare through development of congestive heart failure, arterial thromboembolism and arrhythmogenic death [2]. In human HCM, many disease causing variants have been identified. However in cats only 4 variants are known, two of these are found in MYBPC3 including p.R820W. The aim of this study is to determine how the R820W variant found in the C6 Fn3 domain of MYBPC3 causes increased risk of HCM in cats. Additionally, as this variant has a human homologue, this study provides further support for feline HCM as a model for human HCM. Fig. 1

**Materials and Methods:** Cardiomyocyte cell lines were produced from induced pluripotent stem cells (iPSCs) which had been edited using

CRISPR/Cas9 to contain the R820W variant in the Fn3 domain of MYBPC3. Amino acid sequencing data from these cell lines was used to produce models of the Fn3 domain under investigation. Models were produced for the wild-type, R820W-containing and a deletion variant which was produced as a by-product of the editing process that has been found in human HCM – KDel814 [1]. These models were subjected to discrete molecular dynamics simulations using the piDMD programme to determine what effects these variants had on the structural integrity and folding specificity of the C6 Fn3 domain. The output from these analyses were distinct protein folding funnels which we compared in order to gain insights on the structural effects of these variants on MYBPC3.

**Results:** Our results indicate that the R820W and KDel814 variant cause a loss of stability in the conserved and stable Fn3 domain of MYBPC3. Moreover, the KDel814 variant appears to result in alternate structural variants.

**Conclusions:** Our results indicate that these two HCM-associated variants cause a loss of stability of the C6 Fn3 domain of MYBPC3 and enable structural variants to form. These changes may explain the increased risk of HCM in cats with the R820W variant. This ongoing work, in combination with *ex vivo* work on cardiomyocyte cell lines aims to further investigate the underlying mechanism through which these variants lead to HCM.

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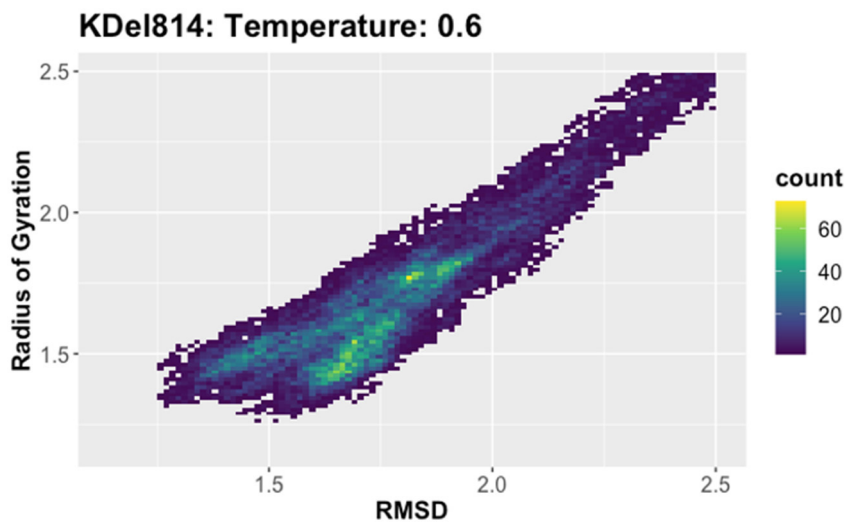


Fig. 1. Relationship between the average distance between atoms (RMSD) and protein compactness (Radius of Gyration). Three clear clusters present at the 0.6 Temperature point for the KDel814 variant indicating alternate structural variants.

### 174. The effect of the removal of pharmacological doses of zinc oxide from the post-weaning diet of outdoor bred newly weaned pigs

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**Application:** Proposed dietary alternative to Zinc Oxide (ZnO) in piglet diets may help to prevent post-weaning scours and maintain growth.

**Introduction:** ZnO is used in piglet diets to reduce enteric disorders post-weaning, however, environmental concerns have led to the imminent control of its use. Moilist *et al.* (2010) suggested the inclusion of wheatbran in piglet diets can reduce post-weaning diarrhoea, and this study assessed the effect of a feed concept for zinc removal (which included lower protein and 4% wheatbran). Additionally, there is a lack of research surrounding the consumption of straw from pigs and the consequences on nutrient digestibility. Studies on growing and weaner pigs suggest variations in consumption of 0 g to 234 g straw/day [2]. Consequently, this study assessed the effect of the dietary changes on the quantity of straw consumed and crude protein (CP) digestibility.

**Materials and Methods:** 1760 commercial, ((LR × Red Duroc) × PIC337), outdoor-bred piglets housed in straw barns were randomly allocated one of four treatments in a 2 × 2 factorial experiment from weaning at 28 days of age, balanced for litter origin and sex (55 pigs per pen, 8 replicates). Pigs were allocated to either a lower protein, +4% WB feed regime (WB) or the control feed regime (C), with or without the inclusion of 3100 mg ZnO/kg. WB feed regime: Diet 1–20% protein, 4% WB, 1.55% lysine; followed by Diet 2–20% protein, 4% WB, 1.45% lysine. Control feed regime: Diet 1–22% protein, 0% WB, 1.60% lysine; followed by Diet 2–21.5% protein, 0% WB, 1.50% lysine.

Dietary treatments were fed during Phase 1 (D0-11), with all pigs fed a standard diet during Phase 2 (D11-17), and Phase 3 (D17-31). Straw consumption and protein digestibility was estimated during phase 2 using a dual marker analysis of acid insoluble ash and TiO<sub>2</sub> which was included in the diet at 3 g/kg. Data was subject to a two-way ANOVA, blocked by shed, using GenStat software (version 19) at a 95% statistical significance level.

**Results:** Pigs fed diets including 3100 mg/kg ZnO had a significantly greater ( $P < 0.05$ ) ADLWG and ADFI D0-31. Pigs fed the WB diet did not see any performance improvement. There were no significant differences between treatments for straw consumption or CP digestibility, and pigs on average consumed 244 g/d of straw.

#### Table 1

**Conclusion:** In contrast to the work of Molist *et al.* [1], a lower protein, +4% WB diet was unable to offset performance loss from ZnO removal. However, the inclusion of ZnO had no significant effect on lifetime performance. Dietary treatment had no effect on CP digestibility or straw consumption, however, piglets appear to consume a significant amount of straw. The effect of this on nutrient digestibility requires further research.

**Acknowledgements:** Thanks to Primary Diets for supplying the feed.

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Table 1  
Effect of zinc and dietary treatment on the performance of newly-weaned piglets.

Item	Treatment				SEM	P-Value		
	WB + Zinc	C + Zinc	WB - Zinc	C - Zinc		Diet	Zinc	Diet × Zinc
ADLWG (D0-31) (g/d)	513	518	476	471	11.3	0.993	< 0.001	0.670
ADLWG (D31-101) (g/d)	988	980	949	959	16.1	0.956	0.071	0.591
ADFI (D0-31) (kg/d)	0.70	0.70	0.68	0.65	0.016	0.463	0.044	0.488
FCR (D0-31)	1.34	1.33	1.39	1.35	0.022	0.377	0.122	0.531
Straw Consumption (g/d)	232	226	266	252	37.9	0.747	0.413	0.909
CP Digestibility (%)	74.06%	74.19%	73.74%	75.30%	0.009	0.370	0.679	0.450

### 175. Intake, growth and carcass traits of steers offered grass silage supplemented with barley- or oats-based rations with or without peas and beans

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**Application:** The feeding value of oats was comparable to barley, and beans was superior to peas. Supplementation of cereals with protein feedstuffs may be unnecessary for finishing beef cattle offered good 'quality' grass silage.

**Introduction:** In Northern Europe there is increasing interest in exploiting indigenous energy and protein feedstuffs. Relatively little research has been reported on oats [2], peas and beans [1] as feed ingredients for beef cattle, particularly when used as supplements to grass silage. In terms of reducing nitrogen excretion and feed costs, it is also important to assess if protein supplements can be excluded from concentrate rations without compromising animal performance. The study objective was to determine intake and performance of beef cattle offered grass silage supplemented with barley or oats with or without peas or beans.

**Materials and methods:** Late-maturing suckler-bred steers (n = 72; 596 kg, s.d. 19.8; 21-months old, s.d. 0.9) were blocked by sire breed and weight, and within block were randomly assigned to one of six supplement treatments: 1.Rolled barley (922 g/kg), 2.Rolled barley (622 g/kg) plus flaked peas (260 g/kg), 3.Rolled barley (742 g/kg) plus flaked beans (180 g/kg), 4.Rolled oats (922 g/kg), 5.Rolled oats (642 g/kg) plus flaked peas (280 g/kg), 6.Rolled oats (732 g/kg) plus flaked beans (190 g/kg). Concentrates containing peas or beans were formulated to

be isonitrogenous (144 g crude protein (CP)/kg dry matter (DM)); the corresponding value for those not containing a protein ingredient was 116 g CP/kgDM. To decrease the high rumen degradable protein, peas and beans were flaked [3]. All concentrates contained 50 g/kg molasses and were balanced for minerals/vitamins. Steers were individually offered 4 kg concentrate DM daily (in two feeds) in addition to grass silage (725 g/kgDM digestibility, 152 g/kgDM CP) *ad libitum*, for 146 days. Animal live weight, ultrasonic measures of muscle and fat depth, and carcass weight, conformation and fat score were determined. Data were analysed using ANOVA with terms for cereal type, protein source, their interactions and block in the model.

**Results:** There were no interactions between cereal type and protein source. Intake, growth, carcass weight, conformation and fat score, and ultrasonic measures of body composition did not differ between cereal type, or cereal 'protein' and beans; however, carcass weight was lower (P = 0.05) for peas (Table 1).

**Conclusion:** The feeding value of oats was equal to barley, and beans was superior to peas. Supplementing the cereals with beans had no effect on carcass growth, whereas supplementation with peas had a negative impact.

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Table 1  
Effect of cereal type and protein inclusion on animal intake and performance.

	Cereal type		SEM	P-value	Protein source		Cereal	SEM	P-value
	Barley	Oats			Peas	Beans			
Dry matter intake (kg/day)	5.9	5.8	0.12	NS	5.8	5.8	5.9	0.14	NS
Daily live weight gain (kg)	0.64	0.64	0.027	NS	0.58	0.67	0.68	0.033	0.08
Final live weight (kg)	697	697	3.9	NS	688	701	703	4.8	0.08
Carcass weight (kg)	402	405	3.3	NS	396	405	410	4.0	0.05
Conformation score (1–15)	9.0	9.5	0.24	NS	8.7	9.7	9.3	0.29	0.07
Fat score (1–15)	7.9	7.8	0.24	NS	7.8	7.9	7.9	0.30	NS

\*This poster was presented as part of the Developing talent session at the BSAS conference.

### 176. Effects of supplementation of *Eucalyptus globulus* essential oil on methane production, dry matter intake and milk composition in Jersey cows

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**Application:** Improving dairy cow performance with supplemental eucalyptus oil can be a novel strategy in dairy cow nutrition and will contribute to the sustainability of the eucalyptus oil extraction enterprise.

#### Table 1

**Introduction:** Essential oil have a wide range of applications including antimicrobial properties, reduction in methane emission by ruminants as well as improvements to growth, milk production, and feed efficiency making them a useful replacement to in-feed antibiotics and growth promoters [1]. The aim of the study was to investigate the effects of supplementation with *Eucalyptus globulus* oil on dry matter intake, milk yield and methane production in dairy cows.

**Materials and methods:** Eight primiparous Jersey cows (140 ± 14 Days in milk) in a zero grazing system were used in a four replicated 2 X 2 Latin square design. The animals had 21 days of adaptation and 7 days for measurements. The treatments were: control and dosing with 15 g d<sup>-1</sup> *Eucalyptus globulus*, oil. Cows in both groups received 6 kg d<sup>-1</sup> of a dairy concentrate, had free access to *Eragrostis curvula* hay and clean water. Milk yield was recorded daily and milk composition determined. Methane production was measured as concentration part per million using a laser methane detector during 7 days of data collection. Measurements was performed twice a day, morning and afternoon. Data were analysed according to a 2 × 2 Latin square design using the general linear model procedures of the statistical analysis systems, 2009 for the average effects over time.

**Results:** There were no significance difference observed in dry matter intake, milk yield and milk protein concentration (p > 0.05).

Table 1

Dry matter intake, milk production and composition, and methane as affected by *Eucalyptus globulus* oil.

Parameters	Treatments	SEM	P-Value	EO
	Control			
DMI, kg/d	12.6	12.7	0.129	0.94
Forage DMI, kg/d	6.56	6.60	0.129	0.93
Forage DMI, % of BW	1.75	1.74	0.037	0.96
Milk yield, kg/d	15.4	15.3	0.350	0.84
Milk fat, %	4.57	4.86	0.206	0.04
Milk prot, %	3.96	3.74	0.079	0.04
Milk urea nitrogen, mg/dL	15.1	14.9	0.73	0.85
Energy corrected milk, kg/d <sup>2</sup>	18.5	18.8	0.70	0.80
Feed efficiency	1.47	1.50	0.050	0.73
Methane, mg/kg	93.7	75.6	6.61	0.06

SEM: Standard Error of Mean.

However, there was a significant increase and decrease decline in milk fat % and protein % (p = 0.04). Methane production (mg/kg) tended (P = 0.06) to be lower in cows supplemented cows.

**Conclusion:** Dosing mid-lactating primiparous Jersey cows with *Eucalyptus globulus* did not affect dry matter intake and milk yield but the milk fat content was increased. The reduced methane might have not spared sufficient energy that could enhance milk production. A prolonged feeding trial is recommended during early lactating to better understand the influence of supplementation of the oil.

**Acknowledgements:** The authors acknowledge funding National Research Foundation and Agricultural Research Council.

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### 177. Efficacy of closantel against a *Fasciola hepatica* isolate from sheep and its *in vitro* sensitivity to albendazole

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**Application:** To develop practical tools to detect drug resistant liver fluke parasites in sheep and cattle.

**Introduction:** *Fasciola hepatica*, liver fluke, causes acute and chronic fasciolosis in sheep and cattle, necessitating drug (flukicide) control to maximise livestock health and welfare. Triclabendazole resistance has led to a reliance on alternative flukicides, e.g. closantel and albendazole. Triclabendazole has high efficacy (>99%) from two days post infection [1], however, closantel and albendazole show lower efficacy, 91% six-nine weeks and ~80% ≥12 weeks post infection, respectively. Previously an isolate of *F. hepatica* from sheep was suspected of being closantel-resistant, however, we subsequently confirmed it as closantel-susceptible by *in vivo* trial, with >99% efficacy against ≥12-week-old fluke. We aimed to further characterise this isolate and define a) *in vivo* efficacy of closantel against fluke aged 2–27 weeks and b) *in vitro* sensitivity to albendazole by egg hatch test.

**Materials and Methods:** a) Fourteen parasite-free sheep were each infected with ~200 metacercariae; closantel (10 mg/kg) was administered at two, four- and 27-weeks post infection (n = 3 sheep per time point), a fourth group was left untreated (n = 5 sheep). Twenty-eight weeks post infection, all animals were euthanised, the mean total *F. hepatica* burden at two, four- and 27-weeks post infection calculated and compared to untreated controls by pairwise t-test. b) Five replicates of eggs (~200/well) were incubated at 25 °C in darkness, for 14 hours,

in 5, 0.5, 0.05 μM albendazole dissolved in methanol, or methanol only. Following 15 days incubation, % ovicidal activity was calculated as [(% eggs developed in control - %eggs developed after drug incubation)/% eggs developed in control] x100 and results compared by pairwise t-test. **Results:** a) The mean number of flukes was as follows: untreated controls, 67.6 ± 9.4; two weeks post infection 51.7 ± 8.1, (23.6% reduction, p = 0.102); four weeks post infection, 28.7 ± 16.7 (57.6% reduction, p = 0.003) and 27 weeks post infection, 0 (100% reduction, p < 0.001). b) For the egg hatch test, the mean egg development rate (control wells) was 84.6 ± 1.8; significant reduction in % ovicidal activity at all drug concentrations was observed; 76.8%, 43.8%, 19.7% at 5, 0.5 and 0.05 μM, respectively, (p < 0.001 at all concentrations).

**Conclusion:** Closantel has ~57% efficacy vs fluke > four weeks post infection; within the range reported previously (Swan GE. 1999). Efficacy earlier, at two weeks post infection, is >20%, although not statistically significant, this is of interest as fluke at 2 weeks post infection can cause acute fasciolosis in sheep. An ovicidal activity at a discriminating dose of 0.5 μM of 43.8% is equivocal (Ceballos et al. 2019) and indicates the isolate may be resistant to albendazole, but requires further evaluation.

**Acknowledgements:** The authors acknowledge funding from BBSRC DTP, University of Liverpool and Hybu Cig Cymru.

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### 178. Effect of aerobically spoiled silage and a mycotoxin binder on the intake, performance and rumen metabolism in dairy cows

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**Application:** Visibly spoilt silage may not have high concentrations of mycotoxins but will be lower in nutritive value, although this may not affect performance unless fed at a high inclusion level.

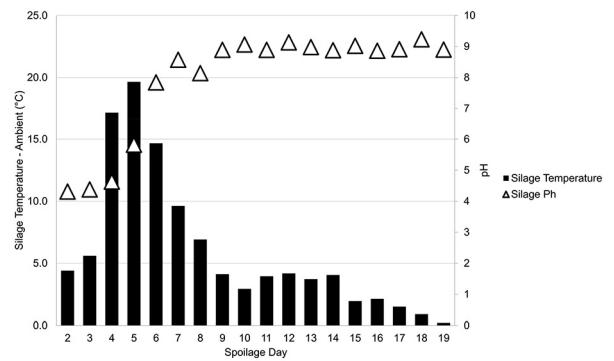
**Introduction:** Typically, when a grass silage clamp has been opened it is subject to aerobic spoilage causing a loss in nutrients and growth of microorganisms including fungal species that are capable of producing mycotoxins [1]. This has the potential to affect intake, rumen metabolism and performance of the dairy cow, and poses a risk to health of the animal. The objective of the study was to determine the effect of a spoilt silage, fed without or with a mycotoxin binder, on the performance and rumen metabolism of dairy cows.

**Materials and methods:** Six rumen-fistulated Holstein-Friesian dairy cows were used in a 3x3 Latin square design, with 3 periods each of 28 days duration. During the initial 23 days, the cows were fed a total mixed ration containing first cut grass silage and concentrates (69:31 DM basis respectively). During the final 5 days, cows received one of three treatments: C: unspoilt grass silage, S: 25% (DM basis) of the silage replaced with spoilt silage, or SM: 25% of the silage replaced with spoilt silage and a mycotoxin binder (Mycosorb A+, Alltech UK Ltd.). The spoilt silage was prepared by spreading over the floor of a temperature control room in 500 kg batches at a depth of 15–20 cm for 19 days. Milk yield and composition were measured daily during the final 7 days of each period. Rumen fluid samples were collected via ruminal cannula on days one, two and five of the final five days of each period at 0800, 1000, 1300 and 1600 hours. Data were analysed using a general linear model three-factor analysis of variance using Genstat 18.1 (VSN International Ltd, Oxford, UK).

**Results:** Aerobic spoilage led to an increase in temperature, dry matter % and pH of the silage (Fig. 1). The spoilt silage contained mean concentrations of Fusarenon X, Fumonisin B1 and Roquefortine C of 574, 879 and 39 µg/kg DM respectively.

Dry matter intake, milk yield, and milk fat and protein content were not affected by treatment ( $P > 0.05$ ) with mean values 20.3, 23.4 kg/d, 49 and 40 g/kg respectively. Rumen metabolism was also not affected by treatment ( $P > 0.05$ ), with a mean pH of 6.2, total volatile fatty acid concentration of 117 mmol/l and acetate to propionate ratio of 3.64.

**Conclusion:** At the levels used in this study aerobically spoiled silage had no effect on rumen metabolism or performance. However, levels



Figure/image 2

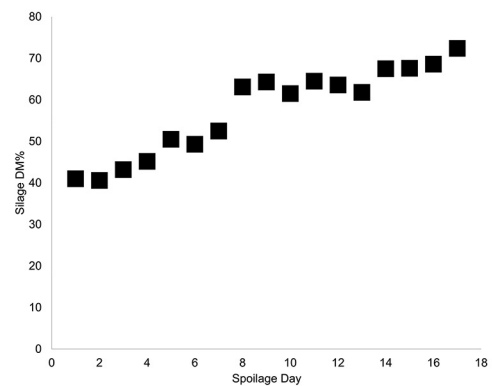


Fig. 1. Temperature and pH profile (a) and DM (b) of aerobically spoiled grass silage

of spoiled silage in a farm environment could exceed the amount used here, affecting nutrient intake and increasing the risk of mycotoxin exposure.

**Acknowledgements:** This study has been funded by Alltech UK Ltd.

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### 179. Reproductive performance of ewes following pre-mating targeted selective treatment against gastrointestinal nematodes

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**Application:** Pre-mating targeted selective treatment (TST) of ewes against gastrointestinal nematodes (GIN), based on body condition score (BCS) or liveweight can be sufficient to maximise reproductive performance whilst decreasing anthelmintic use. **Table 1.**

**Introduction:** GIN threatens animal health and productivity globally and estimated to cost the European sheep industry €157-477 million annually [2]. GIN infections are typically controlled by anthelmintics, but due to anthelmintic resistance, Sustainable Control of Parasites in Sheep discourages drenching pre-mating [1]. However, a recent survey found that this is practiced by 42% of British farmers [5]. Ewe reproductive rate greatly influences ewe profitability [4]. Studies show that TST of lambs, where only a proportion of the flock is treated against GIN, reduces resistance development, whilst maximising lamb performance [3]. This study aimed to identify if BCS and liveweight can be used as measures to identify ewes that benefit from TST against GIN pre-mating.

**Materials and methods:** 354 Welsh Mule ewes, aged 2–8, were randomly allocated into two groups (anthelmintic treatment and control) on a Welsh farm and grazed together. Flock faecal egg count (FEC) suggested low GIN burdens pre-mating (FEC = 215epg). Ewes in anthelmintic treatment group were administered 0.2 mg/kg bodyweight of ivermectin. Ewe BCS, weight, and DAG score were recorded pre-flushing, pre-mating and post-mating. Ewes were scanned 70–90 days into gestation. Ordinal regression analysis used to identify factors associated with reproductive performance and a linear regression used to identify factors associated with ewe condition post-mating. In each model, the independent variable was an interacting effect between anthelmintic treatment group and BCS/weight group.

**Results:** Mean ewe BCS and weight at mating was 3.38 and 67.4 kg, respectively. Mean flock scanned litter size was 1.81. Reproductive performance of ewes with BCS < 3, or weighing < 60 kg pre-mating benefited from anthelmintic treatment against GIN pre-mating ( $P < 0.05$ ). There was no significant effect on scanned litter size of treating ewes

Table 1

Reproductive performance of ewe treated and untreated against GIN pre-mating grouped via pre-mating BCS and liveweight.

Ewe category	Mean scanned litter size	B	S.E	Sig
Ewe BCS				
BCS $\geq 3$ + Drench	1.94	2.16	0.85	0.01
BCS < 3 + Drench	1.82	2.67	1.01	0.01
BCS $\geq 3$ + No Drench	1.81	2.11	0.85	0.01
BCS < 3 + No Drench	1.20	0.00	-	-
Ewe liveweight				
Liveweight $\geq 60$ kg + Drench	1.83	1.6	0.44	0.00
Liveweight < 60 kg + Drench	1.80	1.35	0.60	0.02
Liveweight $\geq 60$ kg + No Drench	1.85	1.66	0.45	0.00
Liveweight < 60 kg + No Drench	1.41	0.00	-	-

with an anthelmintic when BCS  $\geq 3$ , or weight  $\geq 60$  kg ( $P > 0.05$ ). Treating ewes with BCS < 3 or weight < 60 kg had no significant effect on BCS post-mating.

**Conclusions** Reproductive performance of Welsh Mule ewes with BCS < 3 or weight < 60 kg pre-mating benefited significantly from anthelmintic treatment despite low FEC, which usually implies that anthelmintic treatment has no benefit. TST based on ewe BCS or weight pre-mating may maximise flock performance whilst reducing anthelmintic use. Further research is required to identify optimum BCS and weight thresholds for TST of other breeds and production systems.

**Acknowledgements:** The study was funded by IBERS, Aberystwyth University, HCC and KESS2.

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### 180. Evaluation of the tibial bone ash and mineral concentration of Ross 308 broilers with divergent bodyweights on D7

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**Application:** This study provides insights into the differences in tibial ash and mineral concentrations of high and low bodyweight Ross 308 chicks raised on the same diet and environmental conditions.

**Introduction:** It is imperative to achieve the target body weight (BW) for broiler chicks as this has been shown to be positively associated with lifelong growth. In a previous study of Ross 308 male chicks, it was reported that 18% of chicks failed to reach the day 7 BW target of 207 g [1]. Poor growth may be related to lower feed intake or malabsorption, which may manifest as an altered mineral bone profile in underweight chicks. In a continuation of that study, the differences in the tibial bone ash and mineral concentrations of 7-day old Ross 308 broilers ranked as under-performers (UP) or super-performers (SP) were evaluated.

**Materials and methods:** Chicks were raised together in a deep litter pen and fed the same commercial starter diet; ethical approval was granted by the University of Nottingham ethical committee. The legs from SP (n = 8; average BW 270 g) and UP (n = 9; average BW 175 g) d7 chicks were collected and defleshed to extract the tibial bones and remove cartilaginous caps. The tibial bones were oven-dried at 105 °C for 24hrs to achieve a constant weight and ashed at 600 °C overnight to determine

the tibial ash, then the ash weight of individual tibial bone was expressed as a percentage of dry weight. Tibial mineral concentration was determined using the ICP-MS methods. Statistical contrasts were carried out using the student t-test of graph pad prism version 8, while significant differences were accepted at ( $p < 0.05$ ).

**Results:** This study revealed that the percentage of tibial ash was higher ( $p < 0.05$ ) in SP chicks than the UP chicks. Differences ( $p < 0.05$ ) were found in some mineral concentrations between the two groups as shown in Fig. 1. An increase ( $p < 0.05$ ) was seen in the sodium (Na) and rubidium (Rb) contents in SP compared to UP chicks. While calcium (Ca) and phosphorous (P) were not significantly different ( $p > 0.05$ ), there was a numeric increase in tibial Ca and P concentrations in the SP relative to UP chicks. Interestingly, the UP chicks had higher ( $p < 0.05$ ) cesium (Cs), cadmium (Cd), and lead (Pb) compared to the SP chicks, hence the need for further study.

**Conclusion:** SP and UP chicks showed differences in the tibial bone ash and mineral concentrations. The differences observed in the Cs, Cd, and Pb in tibial bones of the UP chicks relative to the SP chicks are not fully understood. However, it could be better explained by our ongoing ileal transcriptomic analysis of these SP and UP chicks as this would give insights into differences in the expression of genes involved in mineral transport.

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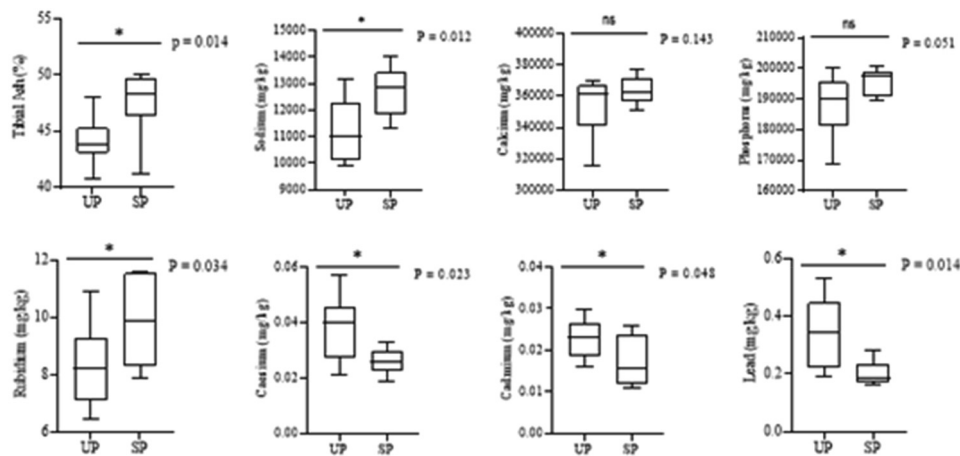


Figure 1: Tibial ash and mineral concentrations of groups ranked as under-performers (UP) or super-performers (SP). Mineral concentrations are

### 181. Dietary protein and energy requirements of Japanese quails in the tropics

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**Applications:** Dietary protein and energy are the most important nutrients in poultry diets. There is lack of information on dietary standard for tropical Japanese quails; this is a major obstacle to the growth and productivity of Japanese quails in the tropics. This study is aimed at determining the dietary energy and protein for optimal performance of Japanese quails in Nigeria.

**Introduction:** Quails is one of the poultry species of economic importance, they produce eggs and meat that are enjoyed for their unique flavour [2]. They have low maintenance cost, short generation and resistance to diseases. However, the nutritional requirements of this bird for the tropical region have not been well established. Studies have shown that the nutritional needs of poultry birds vary from species to species and from one location to another. Presently, in Nigeria, the diets fed to quails are mostly based on 20 to 24% crude protein recommended for the temperate [3]. Furthermore, there are variations in the nutrient requirements reported for this bird [1]; Abbasali et al., 2011). Thus, determining the optimal dietary protein, energy and their interaction on the productivity of Japanese quails is the justification for this work.

**Materials and methods:** A total of 576 unsexed Japanese quails aged two weeks were used for this experiment. A 4 (dietary energy levels: 2600, 2800, 3000 and 3200 Kcal metabolizable energy) × 4 (dietary crude protein levels: 20, 22, 24 and 26 %) factorial arrangement in a

completely randomized design (16 experimental units). Sixteen soya beans-based mash diets were formulated and compounded according to the design of the study. Data on body weight, feed intake were measured and were used for calculating the body weight gain, and feed conversion ratio. All data collected were subjected to a two-Way Analysis of Variance using SAS statistical package (SAS, 2019). Standard and procedure of the Federal University of Technology, Minna, Nigeria ethical guide line was strictly followed.

**Results:** The results of the main effect of different dietary protein and energy levels on the growth performance of Japanese quails are presented in Table 1. In all the performance parameters measured, there was no significant ( $p > 0.05$ ) differences observed in the treatment groups, thus no interaction.

**Conclusion:** Since there was no difference in all the parameters measured, in order to save feed cost, it is more economical to use the lowest protein and energy levels. It is, however, recommended that the same diets should be fed to laying birds to determine the requirements for optimal egg laying performance and quality.

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Table 1  
Main effect of dietary protein and energy on performance of Japanese Quails.

Treatments	Initial Weight (g)	Final Weight (g)	Weight Gain (g)	Total fed Intake (g)	FCR
Effect of crude protein (%)					
20.00	20.23	84.16	63.94	892.89	2.00
22.00	20.27	84.83	64.56	893.01	1.98
24.00	20.25	84.63	64.38	915.44	2.02
26.00	20.24	84.06	64.82	903.16	2.04
SEM	0.02	0.66	0.65	20.92	0.06
p. value	0.81	0.81	0.83	0.85	0.93
Effect of energy ME/Kcal )					
2,600	20.26	83.91	63.65	928.44	2.04
2,800	20.24	84.14	63.90	926.41	2.07
3,000	20.24	84.94	64.70	883.61	1.96
3,200	20.26	84.53	64.27	865.95	1.94
SEM	0.03	0.66	0.65	20.92	0.06
p. value	0.86	0.71	0.70	0.12	0.34

**182. Metabolomic changes in *Mycobacterium avium* subsp. *Paratuberculosis*** (MAP) inoculated Holstein-Friesian cattle suggest metabolites indicative of MAP exposure

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**Application:** Identified metabolites successfully differentiated between *Mycobacterium avium* subspecies *paratuberculosis* (MAP) inoculated and control cattle during the incubation stage of infection. If validated, these metabolites would enable farmers to identify MAP exposed cattle before shedding or clinical signs develop.

**Introduction:** Paratuberculosis, commonly known as Johne's disease, is a chronic intestinal infection of ruminants, caused by MAP. Clinical signs, including reduced milk yields, weight loss and diarrhoea, are typically absent until 2 to 6 years post-infection [1]. This study aimed to examine the effects of MAP inoculation on the metabolomic profile of serum from Holstein-Friesian cattle and correlate affected metabolites to haematological parameters.

**Materials and methods:** At approximately 6-weeks of age, 35 calves received an inoculation with MAP (clinical isolate CIT003)  $3.8 \times 10^9$  on 2 consecutive days. An additional 20 calves formed the control group. Cattle were housed indoors in two separate buildings 500 metres apart, according to their inoculation status. Serum samples were collected pre-, 2-, 3-, 6-, 10-, 12-, 16-, 20-, 24-, 28-, 31- and 33-months post MAP inoculation. Cattle were euthanised 12-, 24- or 33- months post MAP inoculation. Sera from 18 cattle euthanised 33-months post MAP inoculation were utilised. Sera were assessed using flow infusion electrospray high resolution mass spectrometry on a Q Exactive hybrid quadrupole-Orbitrap mass spectrometer for high throughput, sensitive, non-targeted metabolite fingerprinting. Following principal component analyses (PCA), time series analyses used false discovery rate adjusted two-way ANOVA tests to identify mass-ions (m/z) which significantly (p-values < 0.05) differed between experimental classes. Correlation analysis between the identified metabolites and haematology parameters,

including erythrocytes, lymphocytes and monocytes, was performed using Pearson's correlation coefficient. Metabolites were identified using the DIMEdb database of metabolites for Direct Infusion/Injection Metabolomics (<https://dimedb.ibers.aber.ac.uk/>) based on their mass-ion (m/z) values.

**Results:** PCA of the metabolomes discriminated between MAP inoculated and control cattle (Fig. 1). Repeated measures ANOVA highlighted the effect of time on metabolite levels. These results were reinforced by area under the curve (AUC) assessments which indicated that identified metabolites represented sensitive and specific changes occurring at discrete time points. Metabolite set enrichment analysis (MSEA) using over representation analysis (ORA) demonstrated that phosphatidylcholine biosynthesis was significantly affected by MAP inoculation (P < 0.05). Additionally, significant correlations were seen between identified metabolites, such as 2-oxosuccinamate, and haematology parameters, particularly monocytes (%) (P < 0.05) (one example is given in Fig. 2).

**Conclusion:** Metabolomic analysis showed clear differentiation between MAP inoculated and control cattle, in addition to significant correlations between selected metabolites and haematological parameters. Future work could include assessing the relative accumulation of these metabolites in lactating dairy cattle.

**Acknowledgments:** The authors acknowledge funding from KESS2 and ProTEM Services Limited.

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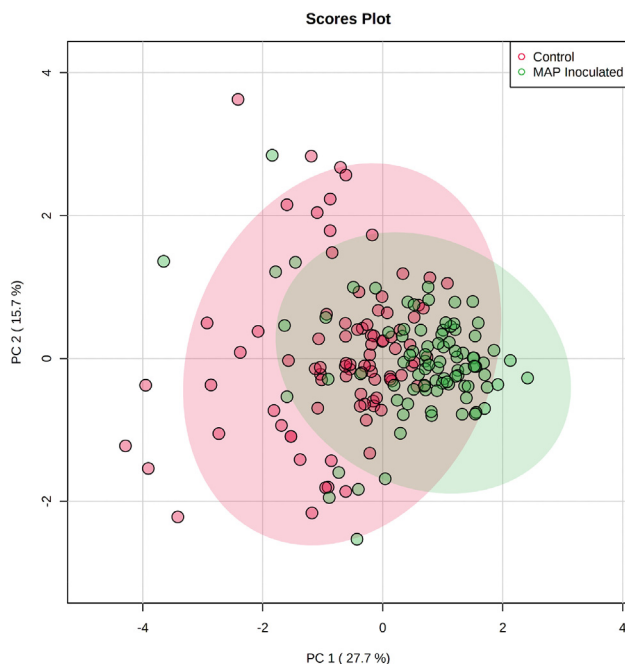


Fig. 1. PCA for MAP inoculated and control cattle in the negative ionization mode. The light red and green ellipses represent 95% confidence intervals.

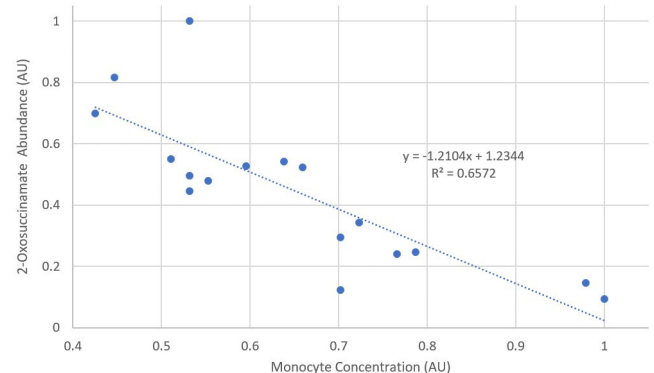


Fig. 2. Correlation between monocytes and 2-oxosuccinamate 33-months post MAP inoculation.

### 183. Enteric methane emissions from Canchim steers finished in feedlot

Paulo Meo-Filho<sup>a</sup>, Alexandre Berndt<sup>b</sup>, Taro Takahashi<sup>a</sup>, Tom Misselbrook<sup>a</sup>, Cintia Righetti Marcondes<sup>b</sup>

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**Application:** Enhanced knowledge of genetic factors contributing to bovine enteric methane emissions can inform optimal breeding/selection strategies, both at individual farm and policy scales.

**Introduction.** Genetic selection has a significant potential to reduce emissions from bovine rumen fermentation [2]. In Brazil, the Canchim breed (3/8 Zebu + 5/8 Charolais), designed to combine advantages of Zebu (adaptation to the tropical conditions) and Charolais (high growth rate and good meat quality), has two distinct lines. The 'old' line originates from 53 Charolais bulls imported from France between 1922 and 1950 and mated with local Guzera, Indubrasil and Nellore females, and has been kept closed since 1953. The 'new' line, developed in the 1990s as a cross between modern elite Charolais bulls and local Nellore females, has been kept closed since 1998. Once closed, both lines have been improved based on a same phenotypic index to achieve local breeding targets [1]. However, their relative performance with regards to enteric methane emissions is not well-understood. The present study aimed to fill this knowledge gap, with a particular focus on the trade-off between daily emissions and daily liveweight gain.

**Materials and methods:** The experiment was carried out at Embrapa Southeast Livestock station (São Carlos, São Paulo) over two seasons. Fifty-one Canchim steers (17 from the old lines and 34 from the new lines), with an initial body weight of  $354 \pm 37$  kg and an initial age of  $21 \pm 3$  months, were placed in feedlot for 105 days in 2015 and

90 days in 2016, respectively. The animals were allocated to collective pens with an electronic trough for individual dry matter intake measurements, while enteric methane emissions from individual animals were evaluated using the GreenFeed system, during the entire feedlot period for both years. Data were analysed initially evaluated using the standard t-test and then further analysed through mixed linear models using SAS's MIXED procedure.

**Results:** The new line demonstrated significantly ( $P < 0.05$ ) higher dry matter intake (10.6 vs. 9.9 kg/day) as well as daily methane emissions (202.2 vs. 169.9 g CH<sub>4</sub>/day) than the old line. However, there was no significant difference was detected in CH<sub>4</sub> emissions per kilogram of dry matter intake (18.4 vs. 18.4 g CH<sub>4</sub>/kg DMI), or in emissions per average daily gain (137.0 vs. 142.0 g CH<sub>4</sub>/kg ADG).

**Conclusion.** While the steers from the new line produce a greater volume of methane per day than the old line, this impact is offset by its greater growth performance. For system-wide comparison of climate impacts, therefore, further research is required to evaluate differences in nitrogen excretion and its potential impact regarding subsequent nitrous oxide emissions, another important greenhouse gas from cattle production.

**Acknowledgements:** Funded by Embrapa (02.12.02.008.00.02).

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#### 184. Essential amino acid concentration of soybean herbage as influenced by stage of growth

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**Application:** Forage legumes provide the base upon which livestock rations are built especially the ruminants. High-quality forage legumes have a high intake potential, along with high digestibility which transform into quality products in terms of meat and milk [Fig. 1](#).

**Introduction:** Meeting the demands for protein requirements in ruminants is fundamental to the production of meat and milk. Forages are well known as the major component of the diet for ruminant livestock [\[1\]](#). Forage legumes have been reported to play significant and profitable role in the ruminant production industry in the tropics [\[2\]](#) but the quality of these legumes are greatly influenced by the stages of growth, hence the aim to evaluate the amino acid concentration of soybean herbage in the humid south western Nigeria.

**Materials and methods:** The experiment was conducted at the Pasture Unit, Federal University of Agriculture, Abeokuta, Nigeria and the laboratory of the International Livestock Research Institute, Ibadan, Nigeria to profile amino acid of soybean (TGx-1951-3F) herbage as affected by stage of growth. The experiment was a randomized complete block design with three treatments based on the stages of growth and four replications. The seed were sown in July 2020 and the soybean samples were harvested at 10 cm above ground level three times during the growth cycle which corresponds to the pre bud, early bud and full bloom stages of growth. The harvested samples weighed and oven dried at 60

°C for 48 hours. Oven-dried samples were milled and stored for subsequent laboratory analysis. The milled oven dried samples were scanned with NIRS for the amino acid of the samples using equation for the feed sample analysis based on the mixed feed global calibration model using the software package (Win ISI II FOSS, Denmark, Model NIRS™ 5000) calibrated against convectional wet laboratory analysis. Spectral information were registered in the wavelength range 1100–2500 nm using NIR system mode 5000 scanning monochromatic infrared spectrophotometer. Data were subjected to generalized linear model. Data means were compared using Tukey HSD Test at 0.05 probability of the same statistical package and was visualized using Microsoft Excel.

**Results:** All the amino acids were significantly ( $p < 0.05$ ) influenced by the stage of growth. There was an increase in all amino acid concentrations, except for valine, lysine, phenylalanine and tryptophan, from pre-bud to early bud growth stage. All amino acid concentrations decreased between the early bud and the full bloom stage of growth. The concentration of methionine and tryptophan are lower than others

**Conclusion:** Stage of growth was found to alter the amino acid concentrations of soya forage. The low concentrates of methionine and tryptophan indicate that ruminants offered soya forage may require additional supplementation'

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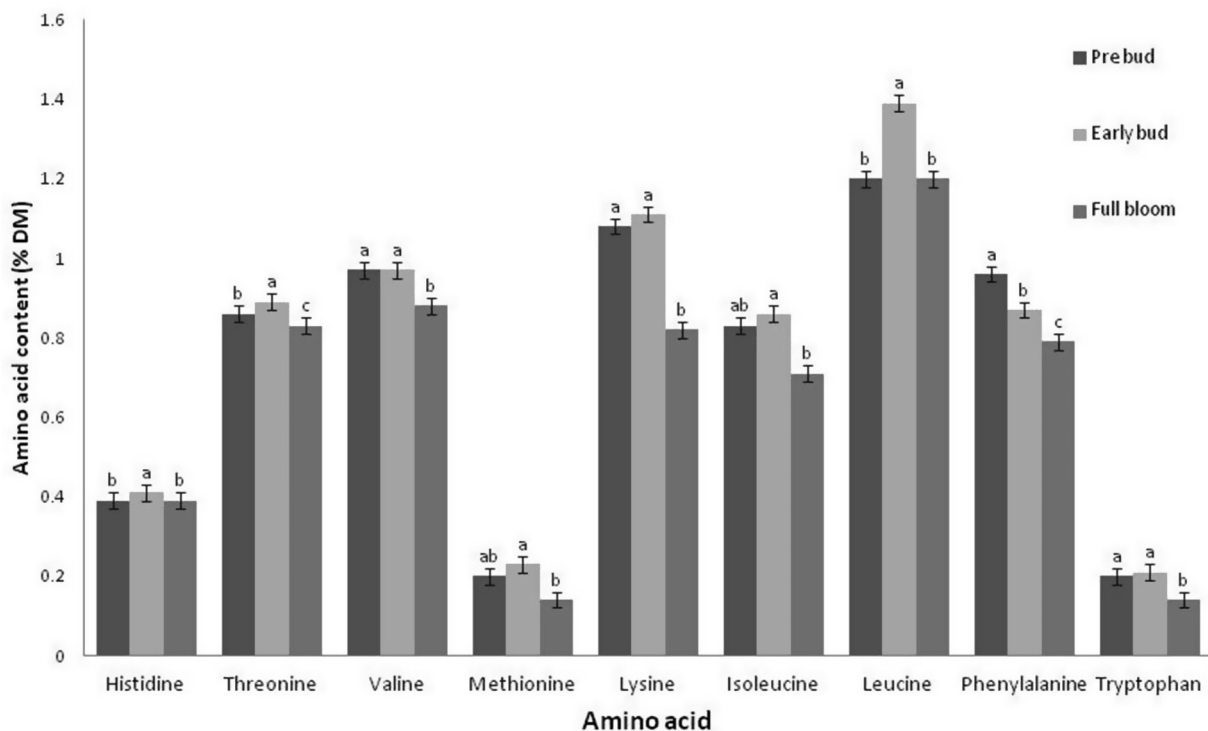


Fig. 2. Effect of stage of growth on the amino acid content of soybean herbage

### 185. Effect of concrete slats and rubber-covered slats on the intake and performance of growing 'weanling' beef cattle accommodated indoors

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**Application:** Covering concrete slats with rubber matting has no effect on performance of 'weanling' cattle.

**Introduction:** The main critical points for cattle welfare during housing have been identified by the European Commission [5], the EU Welfare Quality® project [2] and the European Food Safety Authority [1], as housing conditions, flooring type. Some suggest that conventional slatted floors without lying-areas should be replaced by alternative floor types. [3] investigated the performance and feed efficiency of finishing beef steers, which was enhanced ( $P < 0.001$ ) on rubber mat-covered slats (RM) compared to concrete slats (CS). Compared to faster-growing 'finishing' cattle there is little research evaluating the effect of floor type on the indoor winter performance of slower-growing weanling cattle, prior to turn-out in spring. The study objective was to compare the performance and feed efficiency of suckler-bred weanling beef cattle accommodated on CS and RM.

**Materials and Methods:** Two hundred early- (Aberdeen Angus and Hereford) and late-maturing (Charolais and Limousin) breed weaned suckler heifers and steers (315.6 kg, s.d. 18.8; 262 days old, s.d. 22.2), offered grass silage only ad libitum or silage supplemented with 2 kg of a barley-based concentrate, were used. Animals were blocked by breed, sex, age, weight and diet and from within block randomly assigned to one of 2 treatments for 84 days; i) CS ( $n = 20$ ) ii) RM ( $n = 20$ ) (Durapak Agri Ltd., Ballincollig, Co. Cork, Ireland). Animals were accommodated in groups of 5 at a space allowance of 2.5 m<sup>2</sup> per animal. Feed was weighed into each pen daily and refusals were recorded twice weekly. Animals were weighed at the start, and end, at 7 d intervals during the experiment; average daily live weight gain (ADG) was determined by linear regression. Pen was the experimental unit. Data were analysed using a randomised mixed model ANOVA with the MIXED

Table 1

Effect of floor type on feed dry matter (DM) intake and performance of weanling beef cattle.

	Floor Type		SEM	P-value
	Concrete slats	Rubber-covered slats		
Total DM intake (kg/day)	5.65	5.67	0.077	NS
Final live weight (kg)	362	361	2.63	NS
Live weight gain (kg/day)	0.55	0.53	0.024	NS
FCR (kg DM/ kg ADG)	10.7	11.4	0.66	NS

procedure of SAS (9.4). The model included the fixed effect for floor type and block was included as a random effect.

**Results:** No effect ( $P > 0.05$ ) of floor type on dry matter intake, ADG, feed conversion ratio (FCR) or final weight (d 84) of suckler-bred weanling cattle (Table 1). These results are in line with a previous study carried out by [4].

**Conclusion:** The addition of rubber matting to concrete slats had no effect on intake, growth or feed efficiency of weanling beef cattle.

**Acknowledgments:** Post-graduate, Cathy McGettigan, was funded under a Teagasc Walsh Scholarship.

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### 186. Analysis of mare's milk by near infrared spectroscopy

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**Application:** If shown to be effective, the use of near infrared spectroscopy (NIS) to predict milk composition in mares could help ensure optimal nutrient intake in foals and promote foal growth

**Introduction:** Mares milk is the primary source of nutrition for the newborn foal. The composition of the mare's milk changes during lactation to meet the demanding nutritional requirements of the foal. Average daily gain (ADG) in foals is highest in the first month with ADG levels recorded in excess of 1.6 kg/day. Foster mares are becoming an increasingly popular option in the equine industry, however, the effect of the different milk composition on the foal has not been well researched, due to the lengthy laboratory procedures. Near Infrared Spectroscopy (NIRS) is a technique which uses light in the near infrared region to quantify molecules based on the amount of light they absorb. Research has very successfully used NIRS for the analysis of milk in other mammals, however, to date there has been no research conducted on the application of NIRS for the determination of mare's milk.

**Materials and methods:** This study collected milk from mares at various stages of lactation. Traditional laboratory methods were used to determine total protein (kjeldahl), lactose (spectrophotometry) and pH (pH meter). Data analysis was carried out using MATLAB. Principal

component analysis (PCA) was used to identify outliers and possible groupings in the data. Pre-treatments to the data included standard normal variate (SNV), detrend, first and second derivative, and second derivative with SNV. Relationships between predicted data and reference data was analysed through partial least squares regression (PLS) using cross validation to reduce overfitting. Calibrations were developed using FOSS WinISI software. A total of 74 samples were used for the calibration for protein and 30 samples used for the calibration of lactose. Statistical measures of RPD,  $R^2$  and RMSE were used to identify the most accurate calibration model.

**Results:** The average values for protein and lactose were 2.27% and 5.68% respectively. The pH of milk ranged from 6.65 to 7.55 with an average of 7.2. The best calibration for protein was developed using second derivative Savitzky-Golay smoothing and SNV. This produced a model with RPD of 1.145,  $R^2$  of 0.97 and RMSE of 0.54. For lactose, the best model was found using second derivative Savitsky-Golay smoothing with RPD of 1.13,  $R^2$  of 0.99 and RMSE = 2.012. While, in both cases the  $R^2$  showed the data fits the model well, the RPD and RMSE values indicate the model does not predict accurately.

**Conclusion:** This study presents some preliminary data to indicate there is potential of NIR to be used as a tool to estimate protein and lactose content in mare's milk.

**Acknowledgments:** The research was conducted at the Department of Biological Sciences at University of Limerick.



### 187. An examination of forage production costs on Northern Irish dairy farms

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**Application:** Forage costs are often an ‘unknown’ for producers. The results of this exercise will enable farmers to make decisions based on the full economic cost of producing forages.

**Introduction:** While quality forages are the mainstay of ruminant production systems within Northern Ireland (NI), farmers are often unaware of the true cost of their production. Also, forage costs are often used by advisors and scientists when examining economic performance on farms or within research. This work seeks to update the detailed analysis of forage production costs undertaken almost two decades ago by [1], as management practices and costs have changed considerably since the original analysis.

**Materials and Methods:** This analysis adopted the methodology established by Keady et al. (2002), with original costs and assumptions re-examined. Costs (contractor, labour and variable) were updated using Farm Business Data (FBD) [2]. Where not available from FBD, costs were obtained from local contractors. Assumptions concerning crop management were updated using survey data or recent Agri-Food and Bioscience Institute (AFBI) research data. If no published data were available, ‘considered’ opinion was obtained from AFBI scientists and College of Agriculture Food and Rural Enterprises (CAFRE) farm managers/technologists/advisors. As per FBD, land was charged at £266/ha and adjusted for the proportion of the year the land was assumed to be utilized for. Grass and silage yields and utilisation were updated

using data from recent AFBI studies. Grass silage dry matter (DM) was increased (+20%) to reflect changes in silage harvesting practices. Applications of organic manures and inorganic fertilisers were aligned with Nutrient Action Programme Regulations (NI) 2019 and Nutrient Management Guide (RB209; Feb 2020). Silo sizes and grazing infrastructure were increased to reflect a herd size of 120 cows, with all silos now costed as having a roof.

**Results:** Grazed grass remains the lowest cost forage, while the cost of zero grazing was less than a 3 cut silage (Table 1). Grass silage costs increased with increasing harvest frequency. Silage costs were lower when harvested using a silage wagon (than a forage harvester) with full costs for 2, 3 and 4 cut grass silage being £98, £115 and £134, respectively. It was more costly to produce whole crop (WC) winter wheat silage compared to maize silage. Costs are dependent on multiple factors, such as yield and DM losses, which vary greatly between farms; furthermore, complete diet costs, including supplementary concentrates, should be taken into account when evaluating the full costs of animal diets.

**Conclusions:** Grazed grass remains the cheapest forage. While actual costs will differ between farms, these results provide a basis for making informed decisions concerning the cost of forages on-farm.

**Acknowledgments:** This work was co-funded by DAERA and AgriSearch.

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Table 1  
Calculated production costs for forages on Northern Ireland dairy farms.

	Grazed grass	Zero grazed grass	Grass silage* (2 cut)	Grass silage* (3 cut)	Grass silage* (4 cuts)	Maize silage	WC cereal silage
Establishment cost (£/ha)	61	61	102	76	61	212	178
Variable costs (£/ha)	190	181	256	367	434	521	358
Contractor costs excl. harvesting (£/ha)	134	167	95	95	108	87	167
Contractor costs for harvesting (£/ha)	-	595	380	510	600	200	163
Utilized DM yield (t/ha)	10.2	10.4	11.7	11.6	10.5	13.7	11.5
Forage cash cost (£/t DM)	38	96	71	91	114	75	75
Land charge (£/ha)	266	239	213	226	239	266	266
Forage cost inc. land (£/t DM)	64	119	89	110	137	94	99
Storage cost (£/t DM)	10 <sup>†</sup>	-	13	13	13	13	18
Forage cost incl. storage (£/t DM)	74	119	102	123	150	107	117

\*Harvested using forage harvester, <sup>†</sup>cost of grazing infrastructure.

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