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Free-roaming dog population management: The operative and post-operative health and welfare implications associated with dogs undergoing vasectomy versus orchiectomy in catch neuter return intervention programmes.

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Submitted in part fulfilment of the requirements of the degree of Bachelor of Science in Zoology

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Abstract
Domestic dogs are present globally, and approximately 75% of the population is defined as "free-roaming". Free-roaming dog population management intervention is required to reduce the risks they present to humans, wild-animals and their own populations. Intervention methods include catch neuter return (CNR), culling and sheltering. Presently, in CNR programs, welfare assessments are often absent, and alternative neutering methods are not considered as a way to improve health and welfare. This study aims to determine the factors affecting the health and welfare and speed of recovery of male dogs undergoing the primary neutering technique; orchiectomy, and vasectomy, which is rarely used in practice. Operative and post-operative health and welfare data were collected from 152 male and female dogs undergoing gonadectomy and vasectomy procedures. A key finding of this study was that the duration of neutering surgery significantly increased throughout the day (Wald=5.678, df=1, p=0.017). Additionally, the duration of vasectomy surgery decreased over the 10-day intervention period (Wald=4.014, df=1, p=0.045). This result is due to the first-time vasectomy surgeons improving their skills in surgical technique over the intervention period. The underlying true cause of the effect of surgery start time is thought to be fatigue in the surgeons carrying out high-volume neutering surgery. The factors shown to influence the change in post-operative health and welfare should be interpreted with caution due to ramifications with the qualitative post-operative assessment protocol. This study is the first of its kind which has highlighted some critical areas for potential future research to improve the effectiveness of CNR intervention programs.

Key words: Vasectomy, Orchiectomy, Free-roaming Dogs, Dog Population Management, Welfare, Catch Neuter Return.

List of Abbreviations:
ABC: Animal Birth Control
CNR: Catch Neuter Return
CNVR: Catch Neuter Vaccinate Return
GLMM: Generalized Linear Model
GSFCPS: The Glasgow Short Form Composite Pain Scale
OHE: Ovariohysterectomy
TNR: Trap Neuter Return

1. Introduction

1.1. The Free-Roaming Dog Population

The global dog (*Canis familiaris*) population is estimated to be approximately 700 million individuals, with approximately 75% of those classified as "free-roaming" (Hughes and Macdonald, 2013). Dog population dynamics and ownership levels vary between countries. Broadly there are two ownership states – owned or unowned (Hiby *et al*., 2017); however there is crossover in their definitions. The majority of owned dogs are restricted and fully reliant on their owner for food, water and shelter. In some countries, however owned dogs may be unrestricted and free to roam, with one or more owner (e.g. "community dogs") (Matter and Daniels, 2000). Unowned dogs do not have an owner, but may still rely on humans for food, water and shelter directly or indirectly (e.g. food indirectly from human waste) (Smith *et al*., 2019). These dogs may either be restricted to a shelter environment or entirely free to roam (Hiby *et al*., 2017). The free-roaming dog population, therefore, comprises both unrestricted owned and unrestricted unowned dogs (Smith *et al*., 2019).

Free-roaming dogs pose many issues to human, wild animal and their own populations through disease transmission, road traffic accidents, dog bite injuries, noise and fouling, especially in areas where they are densely populated (Bacon *et al*., 2017). Dog bites and rabies present significant problems to human health; it is estimated that 60,000 people die from rabies annually (Hampson *et al*., 2015), with over 99% these deaths instigated by dog bites leading to viral transmission of rabies to humans (WHO, 2013). The health and welfare of free-roaming dog populations is generally low (Smith *et al*., 2019). Compromised health and welfare is particularly prevalent in unowned free-roaming dogs due to the low quality and quantity of food and water and lack of veterinary care (Matter and Daniels, 2000). In India around 49% of free-roaming dogs have an emaciated body condition as a result of poor health and welfare (Totton *et al*., 2011), putting dogs at higher risk of disease (Rautenbach *et al*., 1991). These issues are magnified as the ever-growing population is free to mate and reproduce (Smith *et al*., 2019). Dog population management is therefore crucial in controlling the population growth of free-roaming dogs, maintaining a good state of health and welfare in the population and minimising the risks presented by dogs to public health and other animals (Hiby *et al*., 2017). There are many different intervention methods designed to manage the free-roaming dog population, including surgical or chemical sterilisation, culling and sheltering. Which method is selected will depend on the population dynamics, dog ownership practices and disease risks in the area (Hiby *et al*., 2017).

Current research into intervention programmes is highlighting areas where animal welfare is compromised and the efforts that can be made to maximise the welfare of the dogs involved. These areas include, but are not limited to: ineffective surveying and planning resulting in ineffective programmes; and inappropriate handling and interpretation of behaviour due to lack of experience, training and resources (Bacon, 2019). In terms of surgery, issues can arise due to poor preparation resulting in injury, and poor anaesthetic planning or monitoring, resulting in consciousness during surgery (Urfer and Kaeberlein, 2019). Injury and distress during capture, transport, and kennelling are mainly due to human interaction and dogs being subject to unusual restricted environments (Bacon *et al*., 2017). Within the spectrum of surgical CNR interventions, many areas exist where welfare problems could arise.

This review will examine the literature available on the physical and physiological challenges that dogs may face during surgical sterilisation in catch neuter return (CNR) intervention programmes, with a focus on the associated potential welfare implications and how they may be alleviated. I will then go onto analyse the welfare issues in a CNR intervention programme in Bila Tserkva, Ukraine, discussing how they may be mitigated, and the potential barriers to improving dog welfare in CNR programmes.

1.1.1. Terminology

In the interest of clarity and uniformity, for the scope of this dissertation, I will use “neutering” as a sex-neutral term for any procedure intended to cause temporary or permanent loss of fertility. “Gonadectomy” is a sex-neutral term for the surgical removal of the gonads in both sexes, “orchiectomy” is a sex-specific term for gonadectomy in males, “ovariohysterectomy” (OHE) is a sex-specific term for gonadectomy in females. “Vasectomy” is a sex-specific term for male surgical sterilisation where the testes remain intact. The acronym CNR will be used in this review as an umbrella term to describe the following population management intervention methods: catch-neuter-return (CNR); trap-neuter-return (TNR); catch-neuter-vaccinate-return (CNVR); animal-birth-control (ABC); and all surgical sterilisation methods with underlying variation in catching methods and neutering techniques. CNR intervention programmes include capturing, housing/caging, neutering, in some cases vaccinating, then appropriate post-operative care during recovery before returning the individual to its origin (Jackman and Rowan, 2007).

1.2. Dog Population Management Intervention Methods

In some parts of the world efforts to manage free-roaming dog populations to reduce risks and maintain welfare standards will always be required. Ethical and animal welfare concerns must be taken into consideration when selecting the intervention method (Bacon *et al*., 2017). The specific design of the method should take into account local dog population dynamics, local risks the dogs pose to humans, wildlife and the environment as well as ownership practices (Hiby, 2019). CNR and catch and removal are stray animal population management methods recommended by the European Convention for the Protection of Pet Animals of 1988 (Council of Europe, 1987). CNR intervention programmes use a variation of catching and neutering techniques to create healthier and more controlled free-roaming animal populations (ICAM, 2015). CNR is widely regarded as being a welfare-friendly method of high-volume surgical sterilisation. In the long-term improvements to the health and welfare of the population following CNR will occur, however, the short-term risks to health and welfare during the intervention are high (Bacon *et al.*, 2019). The measure of effectiveness and success of a CNR programme will depend upon the level of health and welfare of the dogs throughout the programme, and the long-term impact on the free-roaming dog, human, and wild animal populations (Jackman and Rowan, 2007).

Catch and removal methods involve various catching approaches (e.g. hand catching or sack and loop) to take animals from their unrestricted environment for re-homing, sheltering, culling or euthanasia (Mannhart *et al.*, 2007). Catch and removal methods are beyond the scope of this review; the focus will be on CNR population management methods.

1.3. Neutering Techniques

Broadly, while neutering can be either surgical or non-surgical (Maenhoudt *et al.*, 2014), the most widely utilised form of neutering is surgical. Several surgical neutering techniques exist, the more commonly utilised procedure, which involves the removal of gonads: gonadectomy; and the procedures that do not: vasectomy, hysterectomy and salpingectomy (Urfer and Kaeberlein, 2019). Gonadectomy is the primary method of male (orchiectomy) and female (ovariectomy or OHE) sterilisation used in CNR programmes (Jackman and Rowan, 2007). Following gonadectomy gamete production ceases and the production of sex hormones, such as testosterone in males, is significantly reduced (Howe, 2006).

Orchiectomy is the surgical process of removal of the testes in male dogs through an open or closed-end technique (Urfer and Kaeberlein, 2019). Recently Miller *et al.,* (2018) evaluated a new sutureless scrotal (SLSC) approach for use in juvenile male dogs, where a single incision is made. They concluded that the SLSC approach was safe and a faster procedure when compared to orchiectomy, with the potential to improve morbidity and mortality rates and reduce costs in high-volume neutering programmes (Miller, et al., 2018). However, the study sample size was small, with only 36 dogs. Further research into the effect of the SLSC approach procedure in free-roaming dogs of varying ages and larger sample sizes needs to be conducted before it can be successfully utilised in CNR programmes.

Ovariectomy and OHE are two surgical processes which both remove the ovaries in female dogs. OHE is the removal of the ovaries and uterus and is the preferred method in older females (Howe, 2006). Both procedures require laparotomy to access the surgical site (Urfer and Kaeberlein, 2019), followed by either the more commonly used midline incision, or the flank incision approach. The flank incision is usually only utilised in dogs with significant mammary development, narrow-bodies or in shelter surgical settings (McGrath *et al.*, 2004).

Both methods use ligation and isolation of the ovarian pedicle and removal of the ovaries. This is followed by ligature of the uterine body and cervix, and removal of the uterus in the case of OHE (Urfer and Kaeberlein, 2019).

Vasectomy is an uncommon alternative form of sterilisation surgery in male dogs where the vas deferens is cut to stop the movement of sperm and prevent successful reproduction (Pérez-Marín *et al*., 2006). The gonads are not removed in vasectomy; therefore, sperm and sex hormones, such as testosterone are still produced, maintaining libido and other male-specific behaviours (Urfer and Kaeberlein, 2019). The methods include open and endoscopic vasectomy (Silva *et al*., 1993).

There have been suggestions of additional benefits to carrying out vasectomy over orchiectomy, including higher levels of welfare (Daniels, 1983) and lower costs due to reduced surgery time (Molento, 2004). A study in cats (*Felis catus*) revealed that vasectomy might lead to an effective and efficient reduction in population turnover (McCarthy *et al*., 2013). However, little is known about the effects of vasectomising free-roaming dog populations in CNR interventions. This highlights the need for further study to determine the effectiveness of using vasectomy versus orchiectomy in managing free-roaming dog populations and the associated short- and long-term health and welfare implications.

1.4. Welfare risks throughout the CNR process

Potential welfare challenges are present at each stage of any CNR programme, and the extent of these challenges depends on the focus of the programme and the primary concern for animal welfare (Warden, 2012). Nonspecific welfare issues include discomfort, distress, disorientation, thirst, pain or surgical complications (Siracusa *et al*., 2008). Outlined below are the specific short-term welfare challenges faced at each stage of CNR.

Capturing of dogs requires planning and consideration to minimise negative welfare implications in the short- and long-term (Bacon *et al*., 2017). Prior to the intervention, population behaviour should be monitored to assess whether hand catching can occur or if equipment is required (Lee, 2015). Capturing by hand is the most humane method, but full staff training on dog approach, handling and capture technique is essential (Bögel *et al*., 1990) If the equipment is required, the sack and loop or net methods are considered the most humane for fearful dogs (Bacon *et al*., 2017). Dogs have expressed increased fear and escape behaviours in programmes utilising capture equipment (Lee, 2015). This raises welfare concerns, including unnecessary suffering and irresponsible actions towards animals occurring in public, against the focus of the intervention (Bacon *et al*., 2017).

Transportation in CNR programmes can present serious welfare challenges including hyperthermia, injury from aggression, thirst and starvation (Bacon *et al*., 2017). The risk of hyperthermia is particularly prominent as many intervention programmes are carried out in countries with high environmental temperature and humidity (Warden 2012). Careful planning of journey times and specialised vehicles are required to reduce journey time, maintain an ambient temperature and suitably house dogs (Hanneman *et al*., 1977).

Caging is a practical method of housing free-roaming dogs during CNR intervention programmes as long as the environment remains clean, safe, comfortable, dry, draught-free and quiet (Looney *et al*., 2008). Restraining dogs allows CNR staff to monitor pre- and post-operative health and welfare easily. If signs of ill health become apparent, efforts can be made to treat them (Bacon *et al*., 2017). If left untreated, anaesthetic risks might arise during surgery and infection may spread to other dogs or even humans, generating zoonotic disease risk (Bacon *et al*., 2017). Welfare concerns may be raised however, as the confinement of free-roaming dogs results in behavioural and social restriction (Beerda *et al*., 2000).

Research efforts have focused on the welfare impacts of housing conditions in shelter environments. The experiences are similar in CNR programmes, such as new surroundings and routines, unfamiliar sights, sounds, smells, people and other dogs (Hewson *et al*., 2007). A study by Kiddie *et al*., (2015) utilised a validated quality of life assessment tool to recognise management and environmental factors that may affect the quality of life of domesticated kennelled dogs in rehoming centres (Kiddie and Collins, 2015). The results were in line with similar studies suggesting that dogs adjusted to the new kennel environment over time (Rooney *et al*., 2007; Stephen and Ledger, 2006) and environmental enrichment improved welfare (Wells *et al*., 2007; Schipper *et al*., 2008). Enrichment includes the addition of raised platforms in kennels to improve the dog's view and increase the complexity of the usable space (Hubrecht, 1993). Similar tools could be applied to the housing environment in CNR interventions to improve welfare standards during the relatively short stay duration (typically 24 hours to 7 days). Research should focus on applying these resource-based indicators in CNR intervention programmes and assessing their effectiveness in maximising welfare.

The neutering process is a stressful and physically demanding experience, particularly for free-roaming dogs that may never have been confined in this environment (Bacon *et al*., 2017). Appropriate handling, including physical and chemical restraint, preparation, and provision of food and water before surgery are measures that can be taken to minimise any potential welfare issues (Looney *et al*., 2008). The surgical risk may be higher in free-roaming dogs compared to domestic dogs, as the health history and health status when entering the intervention will be unknown (Bacon *et al*., 2017). To minimise these risks, full health examinations are carried out by CNR surgeons prior to surgery (Looney *et al*., 2008).

Analgesic drugs should be selected to provide both intra- and post-operative analgesia, to minimise pain, as well as post-operative monitoring (Bernarski *et al*., 2011). In a study by Lascelles *et al*., (1998) analgesics administered pre-operatively were recorded to be more effective compared to administration post-operatively in reducing pain following OHE. If pain persists following the operation, there will be an increased likelihood of post-operative infection and increased interference with the incision. This risk can be reduced by following surgery protocol, adequate training and accurate monitoring of behaviour, specifically pain behaviour in the post-operative period (Bernarski *et al*., 2011).

Short-term complications of orchiectomy are rare, and if they occur are mostly limited to the usual nonspecific surgical and anaesthetic complications such as infection, bleeding, swelling, and hematoma (Howe, 2006). The risk of these complications can increase with longer surgical duration due to a prolonged exposure to anaesthetic agents (Brodbelt, 2009), which is costly. Therefore reducing surgical duration is an important consideration in high-volume CNR intervention programmes (Reece *et al*., 2012).

Specific short-term complications of vasectomy appear to be rare in dogs but can include sperm granuloma, spermatocele (Pérez-Marín *et al*., 2006) and testicular degeneration (Whyte *et al*., 1997). OHE in the bitch is a routine procedure with similar risks of nonspecific complications, but it has been reported to have the highest complication rate of all elective surgeries (Pollari and Bonnett, 1996). The complications include uterine and ovarian stump granulomas with or without sinus tract formations, 'stump' pyometritis, recurrent oestrus, urinary incontinence and secondary vaginal haemorrhage (Pearson and Gibbs, 1973). While the midline OHE approach is more common, the flank approach has comparable benefits, including earlier release time, improved wound healing due to increased vascularity of sutured tissue and absence of wound tension from the weight of abdominal contents (Reece *et al*., 2012). In the case of lactating bitches coming into CNR interventions, a flank approach should be used to minimise trauma to the wound from suckling activity (Bacon *et al*., 2017). Davidson *et al*., (2004) reported lower pain associated with laparoscopic OHE procedures, due to the minimally invasive procedure. Laparoscopic OHE is possible, but little research effort has gone into this approach due to the substantial costs associated with it and the inability for most CNR intervention programmes to be able to absorb these costs (Bacon *et al*., 2017).

During the post-operative period complications can arise, including infection, hypothermia and post-operative pain (Turk *et al*., 2015; Goldberg *et al*., 2014) which may inhibit recovery (Fossum, 2013). It is, therefore, essential that dogs are observed for signs of post-operative complications (Looney *et al*., 2008). To minimise stress and the risk of complications, the animals should be provided with a clean and comfortable recovery area, analgesia 12-24 hours post-surgery, and regular monitoring by an observer trained in pain assessment (Goldberg *et al*., 2014). The observer must be familiar with the broad range of 'normal' behavioural patterns of free-roaming dogs as the absence of normal behaviour is a marked sign of pain (Anil *et al*., 2002). Sufficient provision of analgesia will alleviate the pain and thus reduce the negative impact on welfare (Bacon *et al*., 2017). Appropriate allocation of food and water should be offered, determined by adequate neurologic status (Looney *et al*., 2008).

Dogs should undergo an individual final assessment to ensure they have fully recovered and adequately analgised before being discharged and returned to the place of capture (Looney *et al*., 2008). Monitoring after returning is advised, using a record of identification created at the start of the intervention (Looney *et al*., 2008). Many methods to identify neutered individuals exist, including ear tags, collars, ear tip or notch, tattoo or microchip (Anchel, 1990).

1.5. Assessing welfare in free-roaming dogs

An animal's welfare is described as its ability to cope at a particular moment in time (Broom, 1986). The assessment of animal welfare is potentially measurable through scientific means (Broom, 2011); however a method has not yet been developed. Therefore assessment currently relies upon the application of relevant resource- and animal-based indicators (Bacon *et al*., 2019). The indicators selected must be valid, reliable and feasible to highlight compromised welfare. Resource-based indicators focus on housing and surrounding conditions (Hiby *et al*., 2017). They are more repeatable, and objective compared to animal-based measures, but they only indicate a risk of welfare problems without providing a measure of welfare (Barnard *et al*., 2014). Animal-based indicators include physical and physiological health and behavioural measures and are widely regarded as robust and representative indicators of an animals welfare state (Dawkins, 2003). Analysing behavioural measures of welfare, such as pain behaviour, relies on accurate evaluation, which is currently hindered by a lack of knowledge, understanding and training (Bacon *et al*., 2019).

In all CNR intervention programmes there are risks of negative welfare experiences during; capture, transport, surgical procedures, the post-operative period and release (Hennessy *et al*., 2001; Scheifele *et al*., 2012; Barnard *et al*., 2014). The potential suffering, although short-term, may have long-term impacts on the animal's welfare state (Hawkins *et al*., 2011). Thus, it is crucial in all CNR interventions that measures are taken where possible to mitigate experiences that contribute to negative welfare.

Recognition of negative welfare states, such as pain or stress is necessary before appropriate remedial actions can be applied (Winckler *et al*., 2003). Stress can be used to measure welfare levels through behavioural indicators such as increased restlessness, low body posture, yawning, and oral behaviours (Beerda *et al*., 1998). Using these measures requires a reliable establishment of baseline responses through non-invasively sampling cortisol levels (e.g. saliva, urinary, faecal), which are considered a sign of stress in most mammals (Mormède *et al*., 2007).

Pain is defined as "an unpleasant sensory and emotional experience associated with actual or potential tissue damage" (Merskey and Bogduk, 1994). The behaviours associated with pain are especially useful in assessing the welfare state of dogs in CNR programmes; however sensitive methods of assessment are required due to similar behaviours being associated with stress (Bacon *et al*., 2017). Composite scales have been used as a universal tool for detecting varying levels of pain behaviour in dogs, including The Glasgow Short Form Composite Pain Scale (GSFCPS)(Appendix 1) (Reid *et al*., 2007); Colorado State University Canine Acute Pain Assessment; and the Facial Action Coding System (Ekman and Rosenberg, 1997). Scales are necessary as individuals respond to and experience different levels of pain, even when exposed to identical environments and stimuli (Hansen, 2003). The GSFCPS is a questionnaire encompassing six scaled behavioural categories to physically assess acute pain in dogs (Holton *et al*., 2001). It was developed using psychometric principles for the measurement of pain, quality of life and intelligence (Reid *et al*., 2007). Greenaway and Reece (2013) trialled the GSFCPS in free-roaming dogs, however, it was suggested that high levels of anxiety associated with human interactions during capturing and pre-operative handling heavily influenced the results. It has also been shown that neuroticism in dogs can negatively influence the results of GSFCPS through inhibition of pain expression (Lush and Ijichi, 2018). Neurotic behaviour is not uncommon in the free-roaming dog population (Bacon *et al*., 2019); thus the indicators utilised in the GSFCPS may lack feasibility in accurate pain recognition in free-roaming dogs. Current research is highlighting the importance of developing a universal or even country-specific pain behaviour scale for free-roaming dogs. Reliable facial grimace scales have been developed and validated in several species, but not yet in free-roaming dogs (Bacon *et al*., 2019). Bacon *et al*., (2019) specify that a visual scale centred around body posture and facial tension would provide a non-invasive, practical and reliable way of detecting pain in free-roaming dogs. The homologous phenotype of these dogs makes the use of facial tension very practical (Descovich *et al*., 2017). Development of such a scale requires a greater understanding of the 'normal' behaviours of free-roaming dogs (Bacon *et al*., 2019).

Animal care professionals have the responsibility to recognise behavioural signs of poor welfare and act upon them in order to safeguard the animals under their care. Bacon *et al*., (2019) highlighted the disparity of canine animal carers in objectively identifying pain behaviours; UK veterinary professionals and shelter staff had comparably better skills than experienced CNR staff. This may be explained by global variation in training and veterinary care (Bacon *et al*., 2019). In countries where CNR interventions are carried out, resource restrictions limit the ability of CNR staff to be adequately trained in recognising pain behaviours (Urfer and Kaeberlein, 2019). Without detection of these behaviours, they will continue to be expressed and therefore perceived by staff as 'normal' behaviours (Anil *et al*., 2002).

Mitigation of pain behaviours may be further restricted by drug limitations, leading to lower standards of veterinary care (Bacon *et al*., 2019). Furthermore, cognitive dissonance may influence pain behaviour recognition as attitudes or beliefs towards a situation may conflict with the actions an individual is required to carry out (Festinger, 1962). Pain is the primary indicator of poor welfare in dogs. Without recognising it, no remedial measures can be taken, putting CNR intervention programmes at risk of not maximising welfare standards (Beerda *et al*., 1998). This highlights the need for CNR interventions to focus resources on training staff who handle and interact with dogs in recognising behaviours of compromised welfare (Bacon *et al*., 2017). Enabling successful recognition and measurement of the negative emotional state of dogs facilitates the provision of resources to remedy this state (Tod *et al*., 2005).

1.6. Conclusion

The short- and long-term health and welfare benefits of neutering free-roaming dog populations are clear, to not only their own population but humans and wild animals as well (Urfer and Kaeberlein, 2019). Once free-roaming populations are reduced, the majority of the direct and indirect problems emanating from them will be reduced, emphasising the importance of focusing resources into effective CNR interventions.

Planning a CNR programme requires an understanding of the local dog population in order to develop specific targets associated with the population (Hiby, 2019) and select the correct method of intervention. Many different methods exist under the umbrella term of CNR, with each intervention programme having underlying variation in capture and neutering techniques (Hiby *et al*., 2017). With this variation comes a range of associated welfare implications present at each stage of intervention (Bacon, 2019). Currently, the vast majority of literature available on population management focuses on gonadectomy. There have been suggestions that vasectomising dogs may be more beneficial in terms of higher welfare and lower costs when compared to gonadectomy (Daniels, 1983). However, this has not yet been validated and highlights the need for research efforts to be focussed on comparing the effectiveness and the short- and long-term health and welfare implications associated with different neutering techniques in CNR programmes. In order to investigate these welfare implications, the development of a welfare assessment tool specific to free-roaming dogs is required in the form of a reliable facial grimace scale (Bacon *et al*., 2019). This will be a practical measure due to the homologous phenotype of free-roaming dogs; however it will require a greater understanding of the range of normal behaviours expressed by free-roaming dogs to be generated (Descovich *et al*., 2017). Once the assessment tool is developed, its application will require resources to be focused on training observers in accurate recognition of compromised welfare states (Urfer and Kaeberlein, 2019). This is currently limited in the majority of countries carrying out CNR interventions due to the cost and resource restrictions (Bacon *et al*., 2019).

If vasectomising a reasonable percentage of a dog population proves to be an effective method of controlling the birth rate, a novel welfare advantage could be achieved for the free-roaming dog population (Urfer and Kaeberlein, 2019). This will occur through a more drastic reduction in their numbers per unit of resource utilised, a reduction in euthanasia, and education towards the care of free-roaming dogs (Molento, 2004).

1.7.Study Aims and Objectives

The overarching research questions I aim to answer are:

1. What factors influence the duration of vasectomy and orchiectomy procedures?
2. What factors influence the post-operative health and welfare of dogs following vasectomy versus orchiectomy surgery?
3. What factors influence the post-operative speed of recovery following vasectomy versus orchiectomy surgery?

These will be answered through investigation of the hypotheses in Table 1.

Table 1 Hypotheses

I	Vasectomised dogs will have a longer surgery duration than orchiectomised dogs.
II	There is a difference in the duration of surgery between the two surgeons.
III	Surgeries completed earlier in the intervention will have a shorter duration.
IV	Surgeries completed earlier in the day will have a shorter duration.
V	Orchiectomised dogs will have a greater improvement in post-operative health and welfare compared to vasectomised dogs.
VI	There will be a difference in post-operative health and welfare of dogs operated on by surgeon 1 and surgeon 2.
VII	Dogs undergoing surgeries of longer duration will see a greater deterioration in post-operative health and welfare.
VIII	Dogs undergoing surgeries earlier in the intervention will have a greater improvement in post-operative health and welfare.
IX	Dogs undergoing surgeries earlier in the day will have a greater improvement in post-operative health and welfare.
X	Greater time between assessment 1 and assessment 2 will lead to a greater improvement in post-operative health and welfare.
XI	Dogs staying for a longer duration will have a greater deterioration in post-operative health and welfare.
XII	Vasectomised dogs will take a longer time to wake after surgery than orchiectomised dogs.
XIII	There is a difference in the recovery time for dogs operated on by the two different surgeons.
XIV	Surgeries completed earlier in the intervention will lead to a shorter recovery time.
XV	Surgeries completed earlier in the day will lead to a longer recovery time.
XVI	Shorter duration of surgery leads to a shorter recovery time.

2. Materials and Methods

This study was observational. The data analysed in this dissertation was collected by a team of volunteers for the charity, VIER PFOTEN International, in Bila Tserkva, Ukraine. I carried out data cleaning and statistical analyses on the data based on my hypotheses. I assisted PhD student Lauren Smith with similar method of data collection in L'viv, Ukraine, using photographic mark-recapture to estimate the free-roaming dog population.

2.1. Pre-data collection

2.1.1. Training

Lisa Collins and Lauren Smith provided the Stray Animal Care volunteer team for VIER PFOTEN International with training on the procedure of data collection and storage in Bila Tserkva. This included details of the study sites, survey routes, data collection techniques and processing procedures for collected data. An application called Animal-ID (Kopach, 2017) was used to record the observations of dogs.

Two trained VIER PFOTEN International veterinary surgeons carried out vasectomy and gonadectomy neutering procedures in this study. They have carried out gonadectomy in CNR intervention programmes in Ukraine since 2013. Neither surgeon had prior experience in the surgical techniques of vasectomy, therefore DVM Johanna Painer provided training at the VIER PFOTEN International clinic in Zhytomyr, Ukraine prior to the intervention.

2.1.2. Study location

The study took place in Bila Tserkva, Ukraine. The city is 67km^2 and has a human population of 199,163 (Review, 2020). A low level of CNR has been carried out by a local enterprise previously, however, VIER PFOTEN has not carried out CNR in this location.

To compare the post-operative health and welfare of male dogs undergoing vasectomy versus orchiectomy, the city was split into two separate surgical intervention areas. This aimed to match the dog demographics and CNR history across the two intervention areas. To determine these areas the city was divided into three sections; the west, the centre, and the east. To avoid dogs mixing from the two different surgical intervention areas, no intervention was carried out in the centre section of the city. Male dogs caught in the west of the city were orchiectomised and male dogs caught in the east of the city were vasectomised. Females caught from both sections of the city underwent OHE. All dogs were returned to the locations they were caught.

The city was further divided into six sectors (A-F), three in the west of the city (A-C) and three in the east of the city (D-F). The free-roaming dog population was estimated in each sector using the photographic mark-recapture method. The methodology is detailed in Appendix 2.

2.2. Catch Neuter Return Process

The CNR intervention was carried out by VIER PFOTEN International, Ukraine between 22nd July – 2nd August 2019. Using the total populations of each sector (A-F), the number of males and females caught in each sector was calculated according to the percentage of the populations planned to be neutered. In sectors A and F, 20% of the sectors male and female population were caught to be neutered. In sectors C and D, 10% of the sectors male and female population were caught to be neutered (Table 2). No dogs were caught for neutering in sectors B and E. The number of males and females vasectomised and gonadectomised in each sector is detailed in (Table 2).

Table 2 Visual representation of the percentage of the dog population neutered in each sector across the city.

Sector	A	B	C	CENTRE	D	E	F
	WEST				EAST		
Male Neutering Procedure	Orchiectomy		Orchiectomy	Buffer Zone	Vasectomy		Vasectomy
% of male population neutered	20%		10%		10%		20%
Number of males neutered	27		24		12		22
% of female population neutered	20%		10%		10%		20%
Number of females neutered	21		17		11		18

Catching and housing

The dogs were caught by hand in most cases, however some were darted and anaesthetised for both dog and handler safety. They were then transported to the clinic in shared cages (no more than four dogs to a cage) in a van. Sample size varied based on population estimates calculated from the baseline population surveys. Each animal entering the intervention was assigned a unique numeric identity number and were caged with one or two other dogs. Water was provided in each cage *ad libitum* and an appropriate fasting time was allowed before individual dogs entered surgery.

Neutering Techniques

Prior to surgery preparation standards were followed by the two VIER PFOTEN International surgeons; sedation (1ml/kg 2% Xylazine), anaesthesia (1ml/kg Zoletil), pre-anaesthetic pain management (15-20 minutes before incision) and application of local anaesthetic (7-10 minutes before incision). Surgical preparation of the site occurred which included shaving, iodine soap scrub, application of chlorhexidine and covering with surgical drapes. Surgical start and end time for each operation was measured from the first incision to the cutting of the last suture. Timings were measured with a digital watch and recorded to the nearest minute. The surgeon, date, dog identity number, sex and type of surgical technique was recorded for each dog on the post-operative protocol (Appendix 3). The specific surgical techniques are detailed in Appendix 4. Following surgery all dogs received amoxicillin antibiotics, vitamins and anti-inflammatory drugs to aid recovery. Gonadectomised male and female dogs were fitted with a blue ear tag, vasectomised male dogs were fitted with a green ear tag for future identification.

Post-operative Care and Return

Post-surgery the animals were transferred to a warehouse where they recovered in shared cages (no more than three dogs to a cage) with free access to water. Dogs requiring longer post-operative care (over 24 hours) were provided with food. To monitor health and welfare two post-operative assessments were conducted against a set assessment protocol (Table 3). Individual dogs were given a score from 0 to 4 for categories A-F and a score of 0 or 1 for categories G and H. Assessment one was carried out approximately two hours after the neutering procedure and assessment two was carried out between 12 and 20 hours after assessment one. One individual carried out all post-operative assessment to avoid issues associated with inter-observer reliability and accuracy. All operative and post-operative data was recorded on the sheet in Appendix 3.

Once recovered, dogs received a rabies vaccination and a microchip and were returned by van to their capture site. The time the dogs were returned was recorded.

Table 3 Final Post-operative Assessment protocol.

<table>
<tr><th colspan="2"></th><th>0</th><th>1</th><th>2</th><th>3</th><th>4</th></tr>
<tr><td>A</td><td rowspan="2">Dog in crate</td><td>Quiet</td><td>Crying or whimpering</td><td>Groaning</td><td>Screaming</td><td></td></tr>
<tr><td>B</td><td>Ignoring wound or painful area</td><td>Looking at wound or painful area</td><td>Licking wound or painful area</td><td>Rubbing wound or painful area</td><td>Chewing wound or painful area</td></tr>
<tr><td>C</td><td rowspan="2">Overall state of the dog</td><td>Happy and content/ bouncy</td><td>Quiet</td><td>Indifferent/non-responsive to surroundings</td><td>Nervous/ anxious/ fearful</td><td>Depressed/ non-responsive to stimulation</td></tr>
<tr><td>D</td><td>Comfortable</td><td>Unsettled</td><td>Restless</td><td>Hunched or tense</td><td>Rigid</td></tr>
<tr><td>E</td><td>Wound score</td><td>Perfectly healing wound</td><td>Mild redness on the skin surrounding the wound</td><td>Swelling or discharge or exposed subcutis</td><td>Partial opening</td><td>Complete opening</td></tr>
<tr><td>F</td><td>Post-op complications</td><td>Not complications</td><td>Surgical site specific, relating to wound</td><td>Minor complications requiring observation</td><td>Intermediate complication, requiring intervention & extension of post-op</td><td>Major complications requiring surgical intervention or death</td></tr>
<tr><td colspan="2"></td><td colspan="2">0</td><td colspan="2">1</td><td>/</td></tr>
<tr><td>G</td><td>Water</td><td colspan="2">Did drink</td><td colspan="2">Did not drink</td><td>Not given water or Spilled water</td></tr>
<tr><td>H</td><td>Other</td><td colspan="2">Did not vomit</td><td colspan="2">Did vomit</td><td>N/A</td></tr>
</table>

2.3. Data analysis

The data collected in this study were entered into Excel by the volunteer team. In the post-operative assessment, I condensed sections G (water) and H (other) as drinking water and vomiting were not considered responses that could be measured in a scale form, therefore they were changed to a 0 or 1 categorical option (Table 3). I calculated the procedure duration, time to wake following surgery, time between post-operative assessment 1 and assessment 2, time between the end of surgery and assessment 2, total post-operative score at assessment 1 and assessment 2 and the change in post-operative score, in Excel. The change in post-operative score indicates the change in health and welfare between assessments. A change in score of zero indicates no change in health and welfare between assessments. A negative change in the score indicates an improvement in health and welfare. A positive change in the score indicates a deterioration in the health and welfare.

Exploratory analyses were performed on the data to select the most appropriate statistical tests for the research questions. Initially, assumptions for ANCOVA were tested; there was linearity and homogeneity of regression slopes. However, the assumption of heteroscedasticity was not met; the data were transformed by $\log_{10}$ and re-tested, however failed to achieve heteroscedasticity, therefore three Generalized linear models (GLMM) were used. The normality of the data sets for each objective was examined using the Kolmogorov Smirnov test. Normality was confirmed through the p value ($p>0.05$) and visualisation of histograms and Normal Q-Q plots. The response variable and predictor factor and covariate variables were entered into the three models as outlined in Appendix 5. All models excluded missing data (Appendix 6). Bonferroni Post-hoc tests were carried out where necessary. Each variable's significance was tested in the model using the Wald test. Statistical significance was set at $p<0.05$. Statistical analyses were conducted in IBM SPSS Statistics version 20.

After the initial statistical analysis of the GLMM outputs of all objectives, box plots and graphs were created to examine if any further pair-wise interactions may be present between variables that had not been previously hypothesised. The three original GLMM's were re-run to include these interactions (Appendix 7). This further analysis was hypothesis-free and enabled me to explore beyond the scope of my original hypotheses.

Once all statistical modelling was completed to answer the original hypotheses, I ran three models including variables for gonadectomised females, hypothesis free (Appendix 8). This provided a baseline comparison to determine whether any significant effects determined by the original models were directly due to the variables included, or factors not controlled for.

3. Results

Descriptive statistics

A total of 152 dogs were neutered in this study by two surgeons, 51 were orchiectomised, 34 were vasectomised and 67 OHE (Table 4). Of the 152 dogs, 19 (12.5 per cent) had identifiable problems pre- or immediately post-surgery; five of those received Atipamezole, a drug indicated for the reversal of the sedative and analgesic effects of the sedative (Volger, 2006). One had to have a hind leg injury sutured, this individual remained for a third assessment and was released at a later time (Table 4). Appendix 6 outlines the percentage of missing data for each variable.

Table 4. The total number of vasectomy, orchiectomy and OHE procedures carried out by surgeon 1 and surgeon 2 during the 10-day intervention period and percentage of complications dogs experienced before, during or after surgery.

Sector	Surgeon 1	Surgeon 2	Total	% of Complications
Vasectomy	18	16	**34**	**14.7**
Orchiectomy	25	24	**49**	**13.7**
OHE	27	40	**67**	**10.5**
Total	**70**	**80**	**150**	**12.5**

Vasectomy, orchiectomy and OHE surgery occurred over 10 days from 22nd July to 1st August 2019 (Figure 1a and b, Appendix 9). During which there was one day where no surgery occurred (28th July). The total number of procedures per day, number of procedures performed per surgeon per day and number of orchiectomies, vasectomies, and OHE varied from day to day (Figure 1a and b, Appendix 9). The surgery start time, assessment 1 and 2 times and release time varied for each dog in the intervention (Figure 2a and b).

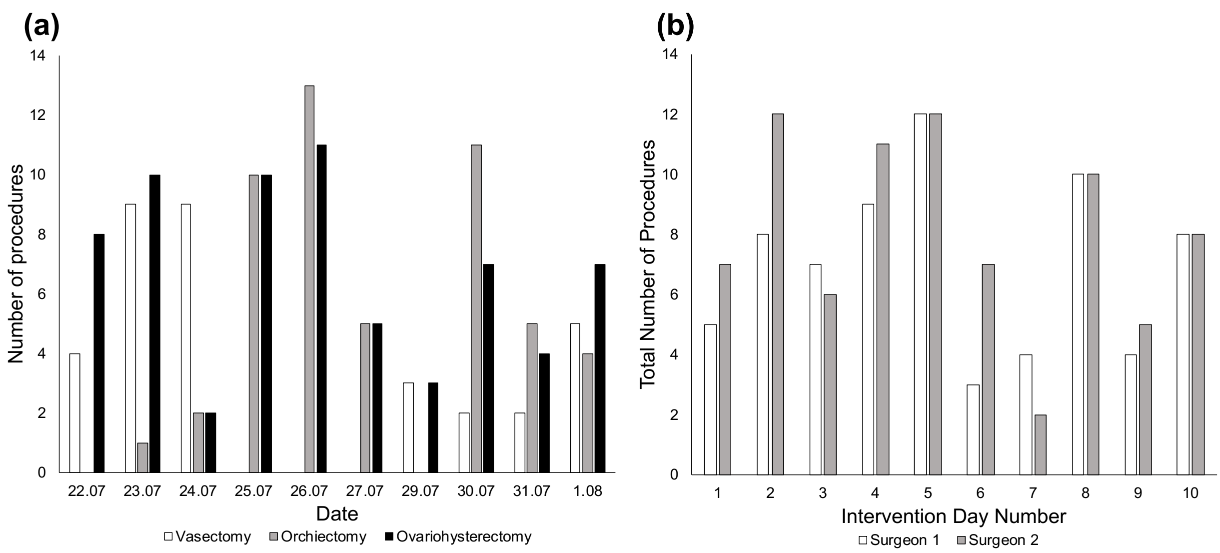

Figure 1 (a) The total number of male and female neutering procedures occurring over the 10-day intervention period. **(b)** The total number of surgeries performed by surgeon 1 and surgeon 2 per day throughout the 10-day intervention period.

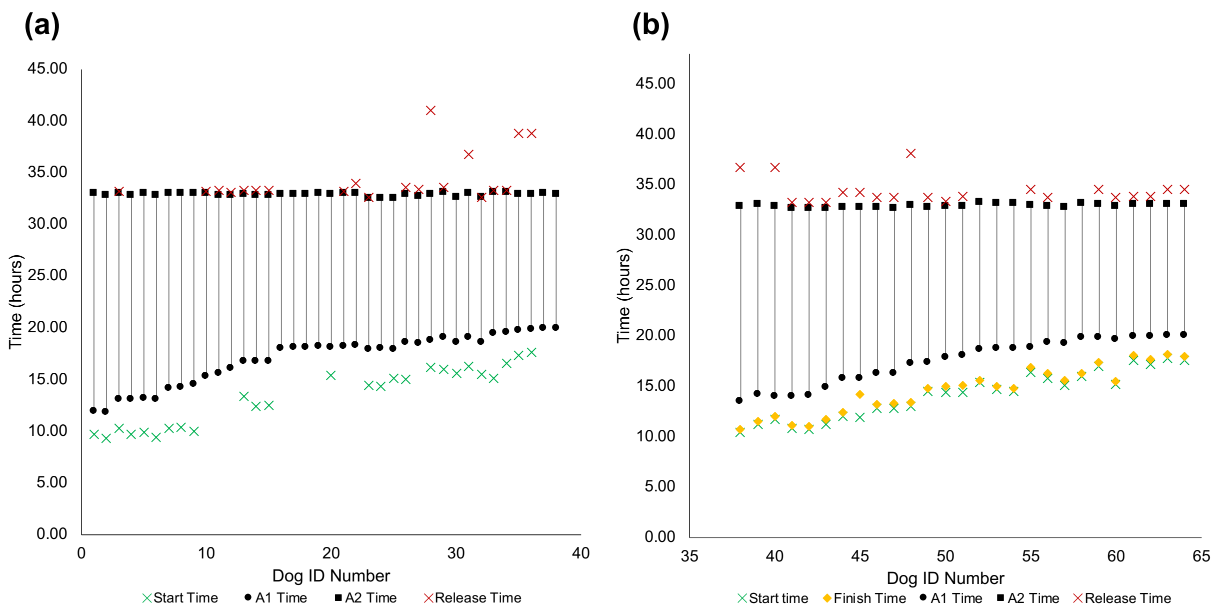

Figure 2 Comparison of surgical and assessment timing for individual dogs undergoing orchiectomy and vasectomy. Missing points are due to missing data. **(a)** Time scale for orchiectomised dogs. Surgery finished so soon after start time that it could not be included in the graph. **(b)** Time scale for vasectomised dogs.

3.1. Duration of surgery

Table 5 Descriptive statistics for the duration of surgery (N = 117).

TYPE OF SURGERY	N	MEAN (hh:mm:ss)	SD (hh:mm:ss)	MINIMUM (hh:mm:ss)	MAXIMUM (hh:mm:ss)
VASECTOMY	27	00:23:13	00:06:45	00:16:00	00:40:00
ORCHIECTOMY	35	00:04:38	00:02:31	00:02:00	00:15:00
OHE	55	00:14:38	00:04:30	00:08:00	00:25:00

The mean duration of vasectomy surgery (23:13 mm:ss, SD ±06:45 mm:ss) is significantly different from the duration of orchiectomy surgery (04:38 mm:ss, SD ±02:31 mm:ss, p=0.001) (Table 5).

Table 6 The GLMM output for the response variable; surgery duration. Significance was set at p<0.05.

Variables in the model	B	Regression slope	S.E.	Wald	*df*	Sig.
Procedure	17.716	+	1.3395	174.921	1	0.001
Surgeon	5.112	+	2.2878	4.994	1	0.025
Day number	0.339	+	0.4890	0.481	8	0.488
Start time	0.621	+	0.0089	4.964	1	0.307

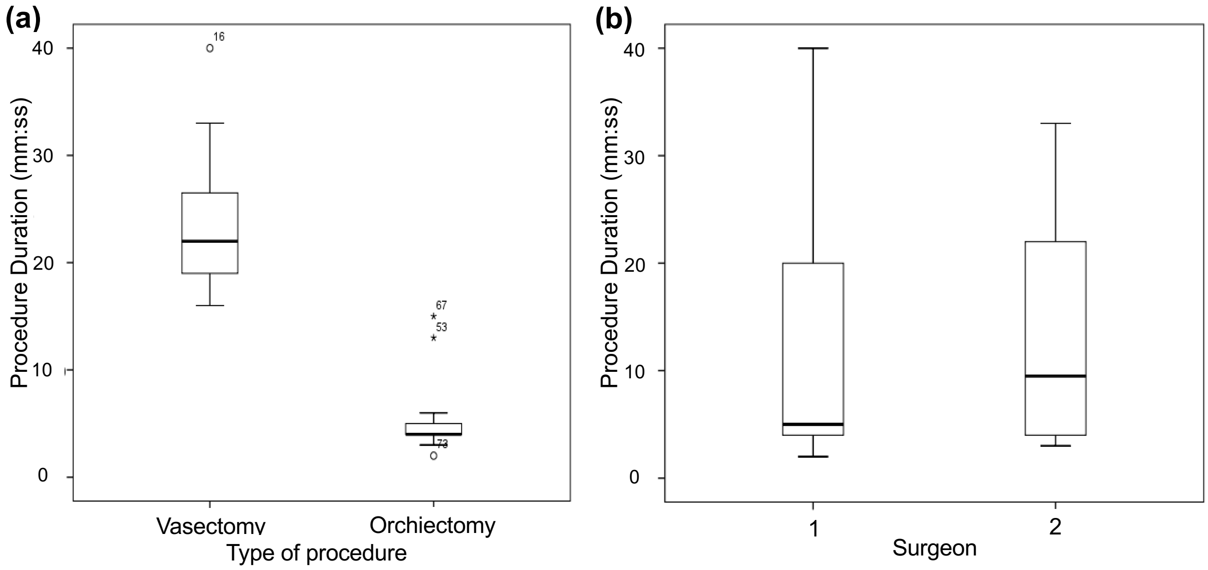

Figure 3 The duration of surgery for each procedure and each surgeon. The lower and upper quartiles represent the variability in the procedure duration. **(a)** Duration of vasectomy and orchiectomy surgery. **(b)** Duration of neutering surgery carried out by surgeon 1 and surgeon 2.

The results from the GLMM looking at the duration of surgery showed a significant effect of the type of procedure. The duration of orchiectomy surgery was significantly shorter and less variable compared to the duration of vasectomy surgery (Wald=174.921, df=1, p=0.001) (Figure 3a). Post hoc tests using the Bonferroni correction revealed that orchiectomy had a shorter surgical duration (05:01 mm:ss SD ±00:17 mm:ss) compared to vasectomy (22:53 mm:ss SD ±01:18 mm:ss) (Wald=172.8, df=1, p=0.001). It was also revealed that the duration of surgery carried out by surgeon 1 (12:59 mm:ss SD ±00:41 mm:ss) was significantly shorter compared to surgeon 2 (14:39 mm:ss SD ±00:45 mm:ss) (Wald=8.938, df=1, p=0.003) (Figure 3b).

There was no effect of the day surgery was carried out on, on the duration of surgery (Wald=0.481, df=8, p=0.488) or the surgery start time (Wald=4.964, df=1, p=0.307).

3.2. Change in post-operative score

All dogs were monitored for a maximum of 28:54 hh:mm and a minimum of 23:25 hh:mm following surgery.

Table 7 Descriptive statistics for the change in post-operative score following surgery (N = 152).

TYPE OF SURGERY	N	MEDIAN	SD	MINIMUM	MAXIMUM
VASECTOMY	34	0	1.35	-4	4
ORCHIECTOMY	51	0	1.34	-2	3
OHE	67	0	1.41	-5	3

Table 7 highlights that there was no difference in the median post-operative scores between the different procedures.

Table 8 The GLMM output for the change in post-operative score between assessment 1 and 2. Significance was set at $p<0.05$.

Variables in the model	B	Regression slope	S.E.	Wald	*df*	Sig.
Procedure	-1.095	-	1.1762	0.866	1	0.352
Surgeon	0.287	+	0.4264	0.454	1	0.500
Surgery Duration	0.061	+	0.0594	1.069	1	0.301
Day number	-0.007	-	0.4497	0.00	1	0.987
Start time	0.00	N/A	0.0004	0.135	1	0.713
Time between A1 and A2	0.006	+	0.0047	1.467	1	0.226
Stay Duration	-0.010	-	0.0253	0.150	1	0.699

The results from the GLMM looking at the change in post-operative score showed no significant difference for the variables in the model; procedure, surgeon, surgery duration, intervention day, surgery start time, time between post-operative assessment and stay duration (Table 8). Therefore, further exploratory statistical analyses were carried out and are detailed below.

3.3. Recovery Time

Table 9 Descriptive statistics for the recovery time following surgery (N = 133).

TYPE OF SURGERY	N	**MEAN** (hh:mm:ss)	**SD** (hh:mm:ss)	**MINIMUM** (hh:mm:ss)	**MAXIMUM** (hh:mm:ss)
ORCHIECTOMY	51	01:06:28	00:34:09	00:06:00	00:02:17
OHE	55	00:52:20	00:23:00	00:12:00	00:01:45
VASECTOMY	27	01:09:26	00:48:07	00:06:00	00:02:17

There was little difference in the recovery time following orchiectomy (01:06:28 hh:mm:ss SD ±00:34:09 hh:mm:ss) compared to vasectomy (01:09:26 hh:mm:ss SD ±00:48:07 hh:mm:ss) (Table 9).

Table 10 The GLMM output for the response variable; recovery time following surgery. Significance was set at $p<0.05$.

Variables in the model	B	Regression slope	S.E.	Wald	*df*	Sig.
Procedure	39.254	+	28.5993	1.884	1	0.170
Surgeon	3.355	+	10.8227	0.096	1	0.757
Surgery Duration	-1.931	-	1.4811	1.700	1	0.192
Day number	3.560	+	10.4282	0.117	1	0.733
Start time	0.001	+	0.0019	0.116	1	0.733

There was no significant difference in the recovery time following surgery for any of the independent variables; procedure, surgeon, surgery duration, intervention day and surgery start time (Table 10).

3.4. Additional exploratory analysis

Procedure duration

Table 11 The exploratory GLMM output for the response variable; surgery duration. Significance was set at p<0.05.

Variables in the model	B	Regression slope	S.E.	Wald	*df*	Sig.
Procedure	21.351	+	2.3721	81.018	1	0.00
Surgeon	4.478	+	2.2367	4.007	1	0.045
Day number	0.017	+	0.5299	0.362	1	0.547
Start time	0.00	N/A	0.0009	3.548	1	0.060
Surgeon*Start time	0.00	N/A	0.00051	5.678	1	0.017
Procedure*Day number	-0.796	-	0.3973	4.014	1	0.045
Procedure*Surgeon	0.00	N/A	0.0	3.020	1	0.047
Procedure*Start time	0.00	N/A	0.00084	0.448	1	0.503

There was a significant difference in the duration of surgery carried out by each surgeon when the surgery start time was considered (Wald=5.678, df=1, p=0.017)(Table 11).

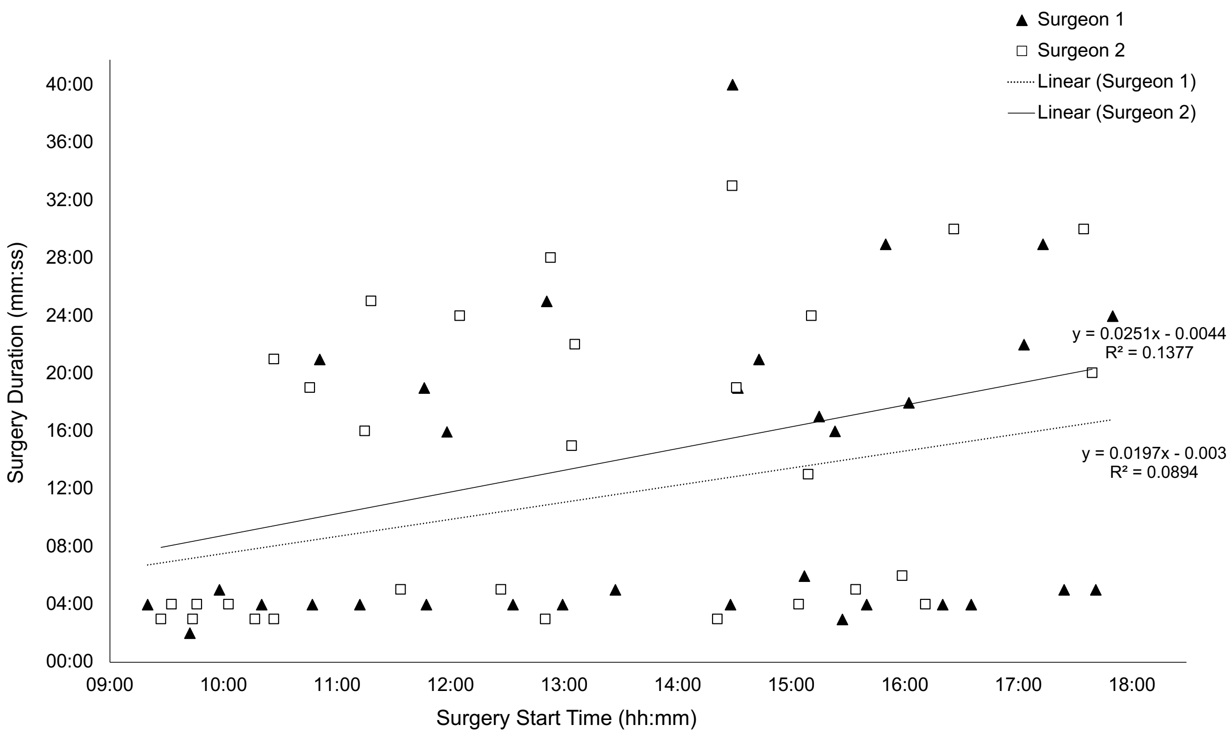

Figure 4 The change in surgical duration carried out by surgeon 1 and surgeon 2 throughout the day.

Figure 4 depicts that for both surgeons 1 and 2 the duration of surgery was lower when it was started earlier in the day and duration increased as surgery was carried out later in the day. Surgeon 1 had a shorter duration of surgical procedures in comparison to surgeon 2 (Figure 4).

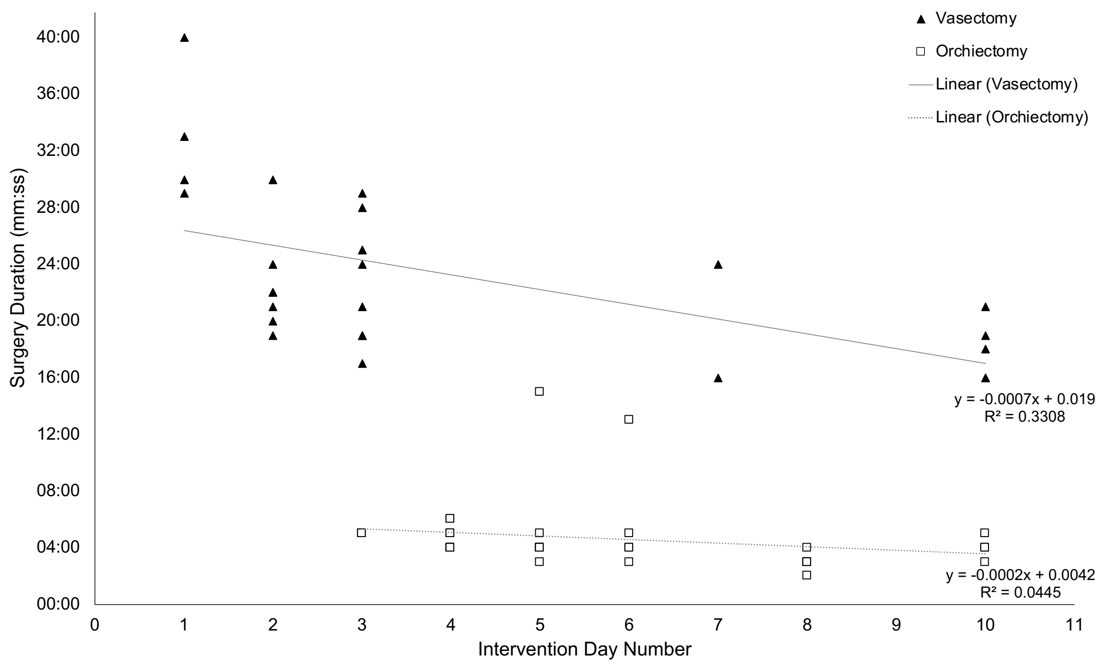

Figure 5 The change in surgical duration of vasectomy and orchiectomy across the 10-day intervention period.

There was a significant difference in the duration of surgery carried out on different days when the type of procedure was considered (Wald=4.014, df=1, p=0.045) (Table 11). Vasectomy surgery carried out later in the intervention had a shorter duration compared to vasectomy surgery carried out earlier in the intervention. This was confirmed through a significant negative regression slope (B=-0.796, Wald=4.014, df=1, p=0.045). Figure 5 shows that there was a small reduction in the duration of orchiectomy surgery when carried out later in the intervention.

Change in post-operative score

Table 12 The exploratory GLMM output for the response variable; change in post-operative score. Significance was set at p<0.05.

Variables in the model	B	Regression slope	S.E.	Wald	*df*	Sig.
Procedure	-0.819	-	1.1364	0.520	1	0.471
Surgeon	-6.149	-	3.1064	3.918	1	0.048
Procedure Duration	0.041	+	0.0582	0.504	1	0.478
Day Number	-0.345	-	3.3978	0.005	1	0.941
Time A1 to A2	0.044	+	0.0222	3.962	1	0.047
Duration of stay	0.017	+	0.0308	0.323	1	0.570
Start time	0.00	N/A	0.0004	0.087	1	0.768
Day number*Start time	-0.00053	-	0.0003	0.027	1	0.869
Surgeon*Start time	0.00	N/A	0.0005	4.662	1	0.031
Surgeon*Day number	0.189	+	0.1637	1.332	1	0.248
Day number*Time A1 to A2	0.001	+	0.002	0.081	1	0.776
Time A1 to A2*Stay duration	-0.003	-	0.0001	3.471	1	0.062

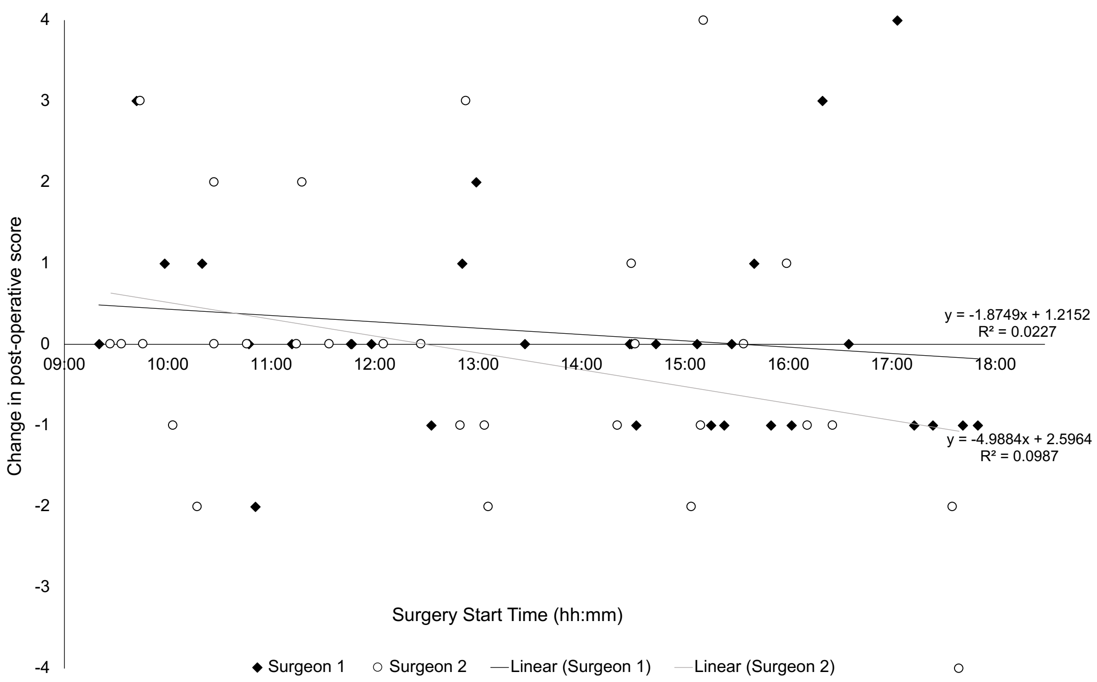

Figure 6 The change in post-operative score over time following surgery carried out by both surgeon 1 and surgeon 2.

There was a significant difference in the change in post-operative score following surgery carried out by the two surgeons when the surgery start time was considered (Wald=4.662, df=1, p=0.031) (Table 12). Figure 6 shows that health and welfare improved (reduction in the post-operative score between assessment 1 and 2) to a greater extent following surgeries started later in the day by both surgeons. There was a greater improvement in health and welfare following surgery carried out by surgeon 2.

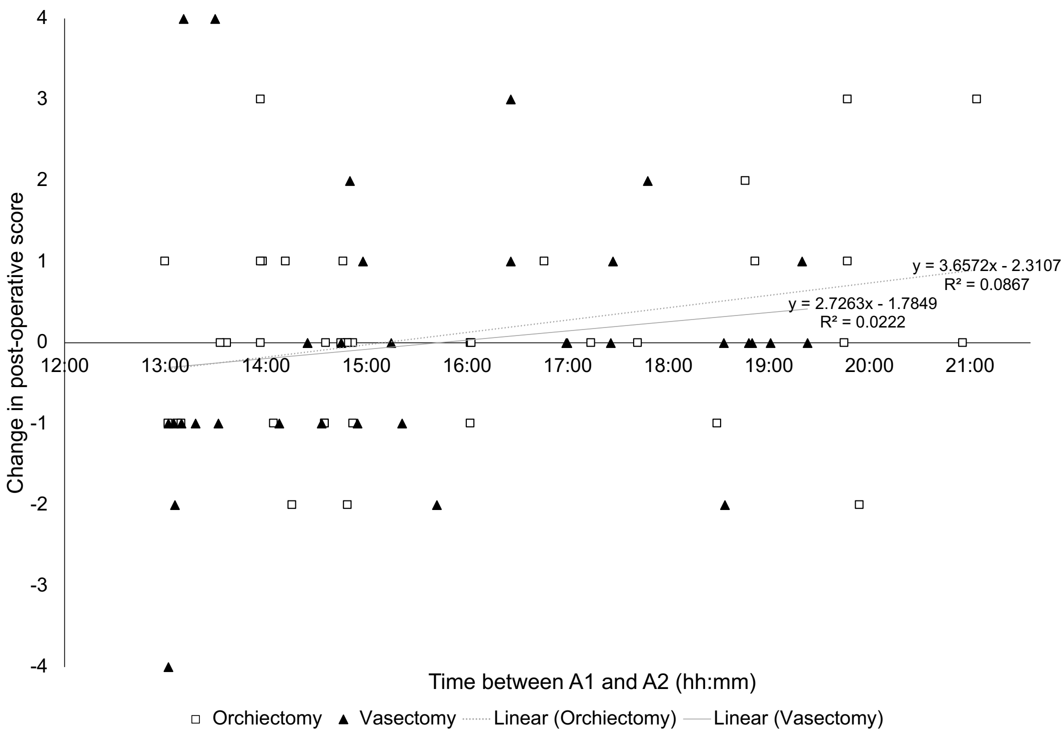

Figure 7 The change in post-operative score as the time between assessment 1 and 2 changes for Vasectomy and Orchiectomy.

Additionally there was a significant difference in the change in post-operative health and welfare with different durations between the two assessments (Wald=3.962, df=1, p=0.047). The weak positive regression slope (B=0.044, df=1, p=0.047) showed that with more time between the two assessments there was a greater deterioration in the post-operative health and welfare (Figure 7).

Recovery Time

Table 13 The exploratory GLMM output for recovery time following surgery. Significance was set at p<0.05.

Variables in the model	B	Regression slope	S.E.	Wald	*df*	Sig.
Procedure	103.853	+	210.664	0.075	1	0.785
Surgeon	-61.474	-	171.341	0.242	1	0.622
Procedure Duration	-9.513	-	5.1798	5.263	1	0.022
Day Number	-32.264	-	44.974	1.002	1	0.317
Start time	-0.003	-	0.0047	0.416	1	0.519
Day number* Procedure duration	1.701	+	0.800	4.515	1	0.034
Surgeon*Start time	0.002	+	0.0028	0.432	1	0.511
Day number* Procedure	-22.451	-	12.396	3.280	1	0.070
Day number*start time	-0.0097	-	0.0002	0.204	1	0.652

There was a significant increase in the recovery time following surgery of a greater duration, carried out later in the intervention (Wald=4.515, df=1, p=0.034)(Table 13).

Table 14 outlines the results of each of my original hypotheses based on the statistical analyses.

Table 14 The results for each of the original hypotheses based on the statistical analyses.

	Hypothesis	**Result**
I	Vasectomised dogs will have a longer surgery duration than orchiectomised dogs.	Supported
II	There is a difference in the duration of surgery between the two surgeons.	Supported
III	Surgeries completed earlier in the intervention will have a shorter duration.	Not supported
IV	Surgeries completed earlier in the day will have a shorter duration.	Supported
V	Orchiectomised dogs will have a greater improvement in post-operative health and welfare compared to vasectomised dogs.	Not supported
VI	There will be a difference in post-operative health and welfare of dogs operated on by surgeon 1 and surgeon 2.	Supported
VII	Dogs undergoing surgeries of longer duration will see a greater deterioration in post-operative health and welfare.	Not supported
VIII	Dogs undergoing surgeries earlier in the intervention will have a greater improvement in post-operative health and welfare.	Not supported
IX	Dogs undergoing surgeries earlier in the day will have a greater improvement in post-operative health and welfare.	Not supported

X	Greater time between assessment 1 and assessment 2 will lead to a greater improvement in post-operative health and welfare.	Not supported
XI	Dogs staying for a longer duration will have a greater deterioration in post-operative health and welfare.	Not supported
XII	Vasectomised dogs will take a longer time to wake after surgery than orchiectomised dogs.	Not supported
XIII	There is a difference in the recovery time for dogs operated on by the two surgeons.	Not supported
XIV	Surgeries completed earlier in the intervention will lead to a shorter recovery time.	Supported
XV	Surgeries completed earlier in the day will lead to a longer recovery time.	Not supported
XVI	Shorter duration of surgery leads to a shorter recovery time.	Supported

Female Analyses

Table 15 The exploratory GLMM output for the response variable; OHE duration. Significance was set at $p<0.05$.

Variables in the model	B	Regression slope	S.E.	Wald	*df*	Sig.
Surgeon	567.412	+	332.898	2.905	1	0.088
Day number	43.107	+	44.909	0.688	1	0.407
Start time	0.013	+	0.0048	5.071	1	0.024
Day number*Start time	-0.001	-	0.0009	1.846	1	0.174
Surgeon*Day number	-11.918	-	22.176	0.289	1	0.591
Surgeon*Start time	-0.004	-	0.0059	0.402	1	0.526

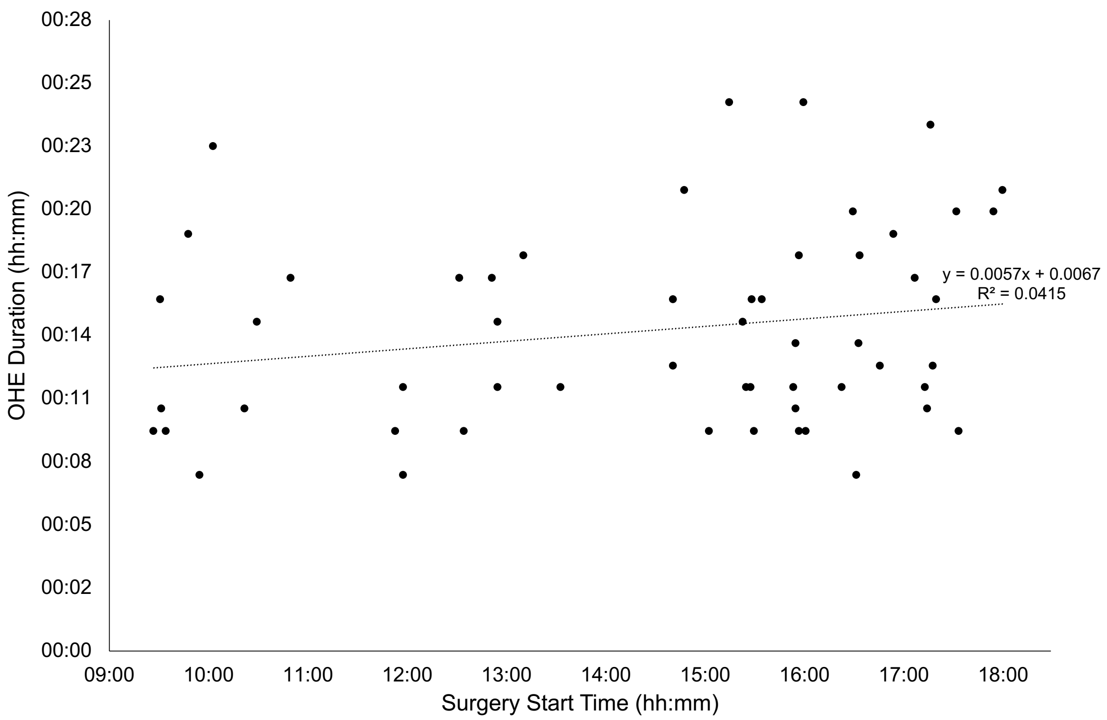

Figure 8 The change in OHE surgery duration throughout the day.

The results from the GLMM looking at the duration of female OHE surgery showed a significant effect of surgery start time (Wald=5.071, df=1, p=0.024)(Table 15). Surgeries carried out later in the day led to an increase in surgery duration (Figure 8).

Table 16 The exploratory GLMM output for the recovery following surgery. Significance was set at p<0.05.

Variables in the model	B	Regression slope	S.E.	Wald	*df*	Sig.
Surgeon	16.311	+	8.3964	3.774	1	0.052
Surgery Duration	-0.013	-	0.0263	0.249	1	0.618
Day number	12.876	+	5.666	5.165	1	0.023
Start time	0.001	+	0.0006	1.118	1	0.290
Day number*Start time	0.00	N/A	0.0001	4.246	1	0.039
Day number*Surgery duration	-0.001	-	0.0049	0.079	1	0.779

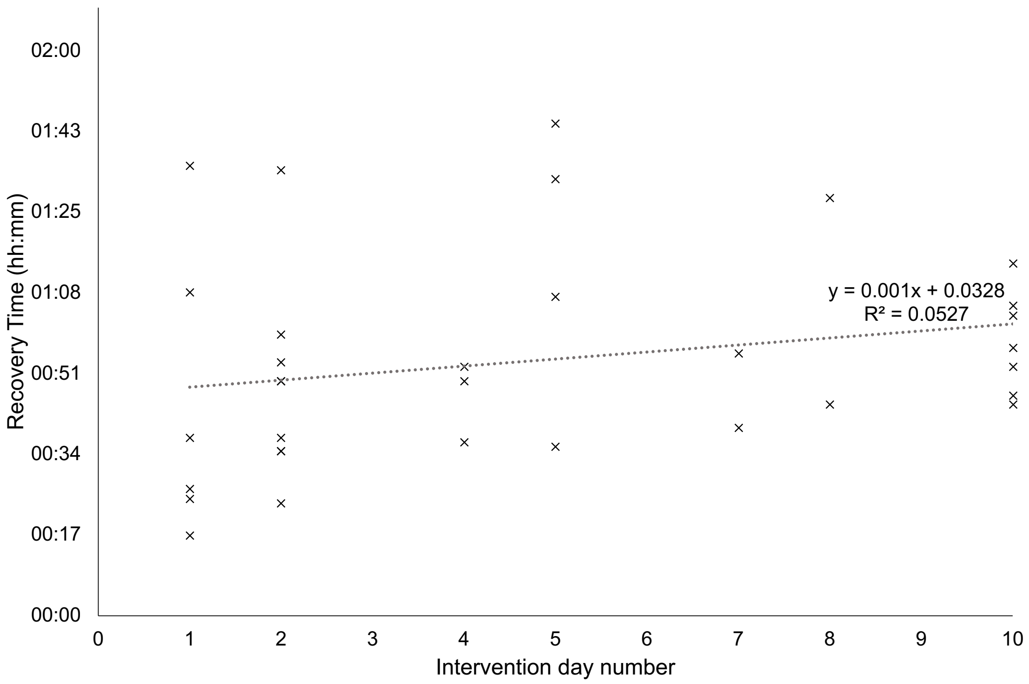

Figure 9 Change in the recovery time following surgery as the intervention goes on (surgery start time and day number).

The results from the GLMM looking at the recovery time following OHE showed that there was a significant difference in the recovery time following surgery when the start time and intervention day number were considered (Wald=4.246, df=1, p=0.039) (Table 16) (Figure 9).

4. Discussion

This study aimed to answer the following questions: What factors influence the duration of vasectomy and orchiectomy procedures? What factors influence the post-operative health and welfare of dogs following vasectomy versus orchiectomy surgery? What factors influence the post-operative speed of recovery following vasectomy versus orchiectomy surgery? The results suggest that the duration of male neutering surgery was affected by the type of procedure, the surgeon carrying out the procedure, the start time of surgery and the intervention day number. The duration of surgery was seen to have an effect on the recovery time following surgery, as well as the intervention day number surgery was carried out on. The change in post-operative score was independently affected by the surgeon carrying out the procedure, the time between post-operative assessments and the surgery start time.

The first question in this study sought to determine the factors affecting the duration of surgery. As predicted in hypothesis (iv)(Table 1), a correlation was present between the surgery start time and the duration of surgery, with procedures that started later in the day more likely to have a longer surgery duration. This association was also illustrated in the results of the hypothesis-free analyses of OHE in females (Figure 8). The effect on surgery duration was seen to a greater extent when male neutering surgery was carried out by surgeon 2 (Figure 4), supporting hypothesis (ii)(Table 1), that there is a difference in the duration of surgery between the two surgeons (Figure 3b). One possible explanation for these findings may be that the surgeons were fatiguing over the day. Fatigue has the ability to impair key neurocognitive functions (Sugden *et al*., 2012), thus affecting surgical performance leading to an increase in surgery duration. This is the first study, to show that surgeon fatigue may have an effect on the post-operative health and welfare of a non-human animal and reflects findings in human studies. The results of the human studies show that the total number of hours a surgeon is operating per day is associated with an increased risk of complications (Thomas *et al*., 2012; Kelz *et al*., 2009; Sugden *et al*., 2012). The factors associated with the risk of complications include the duration of surgery (Thomas *et al*., 2012). Conversely, several studies contradict this relationship (Sessler *et al*., 2011; Boscá *et al*., 2019; Lu *et al*., 2017). The effect of fatigue on surgical performance affects individual surgeons differently depending on levels of self-regulation and professional judgement (Lu *et al*., 2017).

The results support the hypothesis which stated that the duration of vasectomy surgery will be significantly longer than the duration of orchiectomy surgery (i)(Table 1), due to the surgeon's inexperience in carrying out vasectomy procedures. An interaction was found between the type of neutering procedure, and the intervention day surgery was carried out on. Dogs undergoing vasectomy surgery earlier in the intervention were more likely to have a longer duration of surgery, whereas dogs vasectomised later in the intervention were more likely to have a shorter surgery duration (Figure 5). There was very little change in the duration of orchiectomy surgery over the 10-day intervention period (Figure 5). The association did not support hypothesis (iii)(Table 1); which stated that surgery completed earlier in the intervention will have a shorter duration compared to surgeries completed later in the intervention. This result reflects other research, such as a study of first-time ovariectomy surgical performance in dogs by Runge *et al*., (2014) which modelled that with each surgery, surgical time would be expected to decrease by 27%. They concluded that an experienced laparoscopic surgeon might anticipate achieving improved skills in their surgical techniques after performing approximately eight procedures (Runge *et al*., 2014). This was illustrated in the study in the form of a reduction in the duration of vasectomy surgery for both surgeons 1 and 2 over the 10-day intervention period, by 48% and 44% respectively (Figure 5). Additionally, the results of another study into the duration of ovariohysterectomy in dogs and cats and orchiectomy in dogs by Freeman *et al*., (2017) clearly indicates the value of repetition to improve surgical skills (as measured by surgery time) overtime (Freeman *et al*., 2017). The association between first-time vasectomy surgical performance on surgery duration in dogs has not previously been reported. However, I can confidently conclude that the reduction in vasectomy surgery duration over time was due to improving skills in surgical technique in first-time vasectomy surgeons.

The duration of surgery is a crucial consideration for high-volume CNR intervention programmes, as shorter surgical durations reduce the risk of anaesthesia-related complications (Brodbelt, 2009) and reduces costs (Reece *et al*., 2012). Molento, 2004 suggested that vasectomy surgery may have lower associated costs due to reduced surgery duration, in comparison to orchiectomy. However, to date, no research has been conducted into the duration of vasectomy versus orchiectomy surgery in CNR interventions. This study highlights the need for further research of a similar nature; ensuring the surgeons have equal and substantial prior experience of orchiectomy and vasectomy surgical procedures. This will enable firm conclusions to be drawn on how surgery duration is affected by the type of procedure; surgeon fatigue; surgery start time; and the intervention day.

This is also the first study to my knowledge to investigate the factors affecting the change in post-operative health and welfare following vasectomy versus orchiectomy, the second question of the study. The most surprising finding was that a longer duration between post-operative assessments led to a greater deterioration in the health and welfare of dogs (Figure 7). This is contrary to what was hypothesised, which was that there will be a greater improvement in post-operative health and welfare with a longer duration between assessments (Table 1x). As most dogs recovered, it was expected that the post-operative health and welfare should improve over time (Bacon *et al*., 2019), therefore a longer time between assessments would likely lead to a greater improvement in health and welfare, rather than a reduction in health and welfare as shown by the association.

Previous research in human medicine has reported that surgery start time did not measurably affect the short-term outcomes of the patients (Lu *et al*., 2017). However, the results of this study suggest that dogs operated on later in the day had a greater improvement in post-operative health and welfare (ix)(Table 1**.** While any inferences over this difference are made with caution, and in need of further investigation, surgeries which were started later in the day had longer durations and subsequently had greater improvements in post-operative health and welfare. It would be interesting to confirm this association with a more robust data set in further research.

The results of this study do, however, support the hypothesis that the post-operative health and welfare differs when surgery is carried out by surgeon 1 compared to surgeon 2 (Table 1vi). It was speculated that the greater improvement in the health and welfare of dogs operated on by surgeon 2 (Figure 7) may be due to longer surgery duration. However, a study by Wagner *et al*., (2008) reported that the duration of gonadectomy surgery did not have any effect on the expression of post-operative pain-related behaviours (Wagner *et al*., 2008).

Contrary to the hypotheses that state that the type of procedure (Table 1v), duration of surgery (Table 1vii), intervention day number (Table 1viii) and length of stay in the intervention (Table 1xi) would affect post-operative health and welfare, the model found no significant difference. It was not possible to draw any firm conclusions on the factors affecting the post-operative health and welfare from the present study due to many confounding issues with the assessment scoring as I will discuss, and the lack of previous research.

As specified in the literature review, globally, there is much variation in the perception of pain in animals (Bacon *et al*., 2019). However, behaviours associated with pain are the main behaviours used to detect compromised welfare in animals. Pain is an experience rather than an objectively quantifiable physiological response, making its assessment quite difficult (Hansen, 2003). The use of scales to assess health and welfare in free-roaming dogs is challenging (Bacon *et al*., 2019). This is reflected in the results of this study as very little change in health and welfare was recognised between assessments; the median for the change in post-operative score following vasectomy, orchiectomy and OHE was zero (Table 7), and very little statistical association was found (Table 8). However, neutering is a painful and invasive surgical procedure being performed on free-roaming dogs with little to no previous human contact. It is expected that the health and welfare of the dogs may be compromised in the short term (Bacon *et al*., 2019). Therefore caution should be taken when interpreting these results and it should not be presumed that there were no short-term effects. The changes in post-operative health and welfare may not have been detected in this study due to; the design of the post-operative assessment (Table 3), the limited training in detecting pain behaviour, and the variation in timing of the two post-operative assessments, detailed below. Once these factors are recognised, they could be easily altered, to significantly improve the effectiveness of post-operative health and welfare assessment in CNR interventions.

The post-operative assessment utilised in this study (Table 3) was based on a qualitative scale, the GSFCPS (Appendix 1). The GSFCPS (Srithunyarat *et al*., 2016) has been recently reported to lack feasibility in accurate pain recognition, specifically in free-roaming dogs (Bacon *et al*., 2019; Greenaway and Reece, 2013). A study by Conzemius *et al*., (1997) compared the use of qualitative and quantitative pain measures pre- and post-operatively. They concluded that the qualitative measures of pain do not predictably reflect the severity of post-operative pain in dogs compared to quantitative measures (including heart rate and blood pressure). Thereby suggesting that clinicians should not rely on only these variables when assessing the severity of post-operative pain in dogs (Conzemius *et al*., 1997). Instead, clinicians should use a mixture of non-invasive quantitative measures and refined qualitative measures. Bacon *et al*., (2019) suggest refining post-operative assessments to focus more definitively on pain-specific aspects of behaviour, including orbital tightening and wound interference, rather than general muscle tension.

In order to mitigate negative welfare states, training of assessors in recognising these complex and specific behaviours is crucial (Bacon *et al*., 2019). The importance of focusing resources to train assessors was detailed in the literature review. In this study, only one assessor was used to eliminate intra-observer variability; however, no training was provided due to time and cost constraints. Further research is required to provide evidence of the importance of assessors to receive behaviour recognition training, particularly in LEDCs where most CNR intervention programmes are carried out.

It is difficult to arrive at any firm conclusions associated with the change in post-operative health and welfare due to the variation in time between assessments (Figure 2). This was a limitation in the study design, which could not be rectified as returning the dogs to their origin had to occur the following morning before a specific time. This meant that dogs caught later in the day had significantly less time between post-operative assessments, compared to dogs caught and neutered earlier in the day, potentially affecting the results of this study. The first post-operative assessment occurred soon after surgery finished, but at no specific time point. It should also be noted that the lack of observations in assessment 1 (shown by the missing data in Appendix 6) may be explained by pain behaviours being masked by continued sedative effects from surgery (Radisavljević and Vučinić, 2012; Leach *et al*., 2009). In future studies, it would be preferable if the experimental set up allowed a set time for the two post-operative health and welfare assessments.

Overall, post-operative assessments are a quick, easy and non-invasive way to assess the health and welfare of dogs in CNR programmes, but this study has highlighted some significant areas of improvement in detecting compromised health and welfare in future research. The protocol utilised in this study was not able to detect evidence of change in the health and welfare in dogs between the two assessments due to; the design of the protocol, the lack of observer training and the different time of the post-operative assessments. Bacon *et al*., (2019) suggest a free-roaming dog-specific health and welfare assessment protocol is required. Additionally, resources available in CNR programmes should be focussed on training assessors to detect behavioural indicators of poor health and welfare accurately. Finally, a time scale should be developed to determine when post-operative assessments should be carried out, to account for the time sedative and anaesthetic effects of surgery take to wear off. This will allow for more accurate determination of the post-operative health and welfare of free-roaming dogs following neutering surgery for CNR intervention programmes.

The final question in this study sought to determine the factors affecting the speed of recovery as measured by the time taken to wake following surgery. The analyses highlighted that recovery time was longer following surgeries of longer duration carried out later in the intervention period. These results support the hypotheses which state that a shorter duration of surgery leads to quicker recovery time (Table 1xvi) and that surgeries completed earlier in the intervention period will have a shorter recovery time (Table 1xiv). The latter association was also illustrated in the results of the hypothesis-free analyses of OHE in females (Figure 9). This highlights for the first time how recovery time is influenced by the interaction between intervention day surgery was carried out on and the duration of surgery following neutering in CNR intervention programmes. In a study by Pottie *et al*., (2007) of the 69 males and females gonadectomised, most patients experienced a mild degree of hypothermia after a relatively short period of general anaesthesia, which subsequently led to an increase in the recovery time (including the time taken to wake following surgery). However, without data on total anaesthetic time and body temperature before, during and after surgery, no conclusions can be drawn for this association.

None of the other hypothesised variables were shown to have an effect on the recovery time following surgery. These included the effect of the type of procedure being carried out (xii), the surgeon carrying out the operation (xiii) and the surgery start time (xv). This suggests further study is required to measure the effects of anaesthetic duration and body temperature before, during and after surgery on the speed of recovery in vasectomy versus gonadectomy in high volume CNR programmes.

This study has highlighted many potential areas for further research comparing the effects of orchiectomy versus vasectomy surgery in free-roaming dog population management interventions. Additionally, pre-operative assessments should be considered to more accurately determine the change in health and welfare following surgery. This should include assessment of body condition score and skin conditions, the most common ways to assess physical condition (Hiby *et al*., 2017). At the same time, a short human reaction test could be carried out (Barnard *et al*., 2014), to ensure behavioural reactions to humans are not mistakenly recorded as compromised welfare behaviours. Finally, it may be beneficial to measure the total incision length to investigate further the effect of incision length on operating times (Reece *et al*., 2012) and post-operative complications (Cimino-Brown *et al*., 1997).

Further research beyond the scope of this study includes the effect of neutering age on short- and long-term health and welfare (Urfer and Kaeberlein, 2019). Additionally, research into the costs associated with different methods of neutering will further aid CNR staff in selecting the most suitable neutering technique.

5. Conclusions

In conclusion, the results from this study showed that the duration of surgery increases over the course of the day. Surgeon fatigue has been identified as the likely explanation behind this association. A relationship consistent in human medical studies (Thomas *et al*., 2012; Kelz *et al*., 2009; Sugden *et al*., 2012), but recognised in this study for the first time in non-human animals. Vasectomy surgical technique improved over the 10-day period leading to shorter surgery duration in the latter part of the intervention, which is consistent with previous studies (Runge *et al*., 2014; Freeman *et al*., 2017). The factors shown to influence the change in post-operative health and welfare should be interpreted with caution due to the ramifications with the qualitative post-operative assessment protocol. This is the first study to investigate the change in post-operative health and welfare following vasectomy versus gonadectomy and has highlighted areas of focus for future study. Most importantly, the development of a robust free-roaming dog-specific health and welfare assessment, to be used by a trained assessor (Bacon *et al*., 2019). The intervention day number and duration of surgery influence the speed of recovery. The results from this study showed that recovery time was greater following surgeries of longer duration carried out later in the intervention period. Analyses of ovariohysterectomised females confirmed this influence of the intervention day on the speed of recovery. Overall, the results show the influence that surgical and non-surgical factors may have on the health and welfare and speed of recovery following neutering in CNR intervention programmes. The suggestion that dogs may experience higher levels of welfare following vasectomy in comparison to orchiectomy (Daniels, 1983) requires further investigation. There is a potential for negative health and welfare states to persist in the long-term; therefore, all interventions should make attempts to maximise their effectiveness and efficiency in terms of animal welfare wherever possible. The importance of increasing understanding of post-operative health and welfare of free-roaming dogs in CNR interventions is paramount in achieving effective canine population management.

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Appendices

Appendix 1 Glasgow Short Form Composite Pain Scale

		0	1	2	3	4
Ai	**Dog in crate**	Quiet	Crying or whimpering	Groaning	Screaming	
Aii		Ignoring wound or painful area	Looking at wound or painful area	Licking wound or painful area	Rubbing wound or painful area	Chewing wound or painful area
B	**Lead dog out of the kennel**	Normal walking	Lame	Slow or reluctant	Stiff	Refuses to move
C	**Gentle pressure to wound**	Do nothing	Look around	Flinch	Growl or guard area	Snap
Dv	**Overall state of the dog**	Happy and content/ bouncy	Quiet	Indifferent/non-responsive to surroundings	Nervous/ anxious/ fearful	Depressed/ non-responsive to stimulation
Dvi		Comfortable	Unsettled	Restless	Hunched or tense	Rigid

Appendix 2 Methodology for street survey areas and routes for population estimation

The city was further divided up into contiguous blocks of 2km^2. Out of these, one block was chosen as a study area in each sector (A-F) of the city (map). There were three study areas in the west of the city (A-C) and three in the east of the city (D-F). Photographic mark recapture was carried out in each study area to estimate the population of free-roaming dogs in each sector (A-F) of the city. In each of these study areas, survey routes were planned, lasting a maximum of two hours. Prior to data collection a pilot study was carried out to ensure there were free-roaming dogs present and that the survey routes were fully accessible (e.g. did not cover any private property).

Dog population Estimation

Population data collection was carried out by three teams of two VIER PFOTEN International observers, over two six-day periods; 10th-15th May and 15th-20th July 2019. Each study area (A-F) was surveyed three times on consecutive mornings by the same team. During each of the six-day periods, the first three days were spent surveying the study areas in the west of the city (A-C). The final three days of the period were spent surveying the study areas in the east of the city (D-F). All surveys were conducted between 6am and 10am and took a maximum of 2 hours to complete. Starting time varied between 6am and 8am, depending on the time of first light (i.e. later starting time in autumn). The surveys consisted of the teams following the planned survey routes, one individual would take photographs from multiple angles of each dog at a precautionary distance and the second individual would record biological information on the Animal ID (Kopach, 2017) application. At the same time demographic information was recorded in the application, including: GPS location, sex (male / female / unknown), age estimate (>5 years old / 5 years old / <5 years old), neutering status (Y / N based on ear tag presence / absence), year of neutering (based on ear tag colour, as different colour tags used in different years), collar (presence/absence), visible pregnancy, lactation status (visibly lactating / not visibly lactating), visible skin condition (presence / absence), visible injury (presence / absence) and body condition score on a 1-5 scale (emaciated, slightly underweight, normal, slightly overweight, obese).

Data processing

All data was processed in Excel. PhD student, Lauren Smith used the photographic mark recapture data to estimate the population of dogs in each study area, then extrapolated this to get an estimation of the population for each sector of the city (Appendix 4). The full methodology of photographic mark-recapture is explained in detail in Smith, *et al*., (in prep).

Appendix 3 Post-operative Health and Welfare Assessment Protocol

VASECTOMY STUDY
POST-OP OBSERVATION PROTOCOL

CNR – JULY/AUGUST 2019

Date: ___ / ___ / ___ Sex: Male ♂ ____ Female ♀____
Dog ID: ________ Gonadectomy ___ Vasectomy ___
Ear tag colour: ___________

Surgeon: _____________________________ Post-op start: ___ / ___ / ___ ___ : ___
Surgery start time: ___ : ___ Post-op end: ___ / ___ / ___ ___ : ___
Surgery end time: ___ : ___ Release: ___ / ___ / ___ ___ : ___

Type	Description	Assessment 1	Assessment 2	Assessment 3	Assessment 4	Assessment 5
Time	Exact time when assessments were done					
Dog in Crate	Quiet 0; Crying or whimpering 1; Groaning 2; Screaming 3					
	Ignoring any wound or painful area 0; Looking at wound or painful area 1; Licking wound or painful area 2; Rubbing wound or painful area 3; Chewing wound or painful area 4					
Overall state of the dog	Happy and content/bouncy 0; Quiet 1; Indifferent/non-responsive to surroundings 2; Nervous/anxious/fearful 3; Depressed/non-responsive to stimulation 4					
	Comfortable 0; Unsettled 1; Restless 2; Hunched or tense 3; Rigid 4					
Wound Score	Perfectly healing wound 0; Mild redness on the skin around the wound 1; Swelling or discharge or exposed subcutis 2; Partial opening of the wound 3; Complete opening of the wound 4; Not visible wound N/A					
Post-op Complications	No complications 0; Surgical site specific, relating to the wound 1; Minor complications, requiring observation 2; Intermediate complications, requiring intervention and extension of post-op 3; Major complications, requiring surgical intervention or resulting in high morbidity/death 4					
Water	Water was not given 0; Water spilled 1; Did not drink 2; Drank water 3					
Other	Urine 0; Faeces 1; Diarrhea 2; Vomit 3					

Notes:____
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Appendix 4 Surgical Neutering Methodology

Orchiectomy surgical technique

Orchiectomy involved the removal of both testis with attached epididymis, via the open and closed surgical techniques. In both of these techniques the dog was placed in dorsal recumbency and a single incision was made cranially from the scrotum through which the testicles were extracted. In dogs undergoing the open technique, the vaginal tunic was opened before ligating the vas deferens and testicular artery and removing the testicle. In the closed technique a ligature was placed around the tunic, testicular artery, cremaster muscle and vas deferens inside the tunic before removal of the testicle and surrounding structures distal of the ligature. Once both testicles were removed, the incision was sutured and closed (Urfer and Kaeberlein, 2019).

OHE surgical technique

OHE involved the removal of the uterus and two ovaries, the midline approach was used in every case. This procedure occurred through an incision in the midventral abdomen, while the dog was in dorsal recumbency. The incision began at, or no more than 1 cm caudal to the umbilicus and was long enough to reveal the ovaries and junction of the cervix and uterine body. Then the ovarian pedicle underwent isolation and ligation, and removal of the ovaries occurred. Finally, ligature of the uterine body and cervix occurred, and the uterus was removed. The incision was sutured and closed (Davidson, et al., 2004).

Vasectomy surgical technique

Vasectomy is a surgical sterilisation process in males where the gonads remain intact and there is surgical transection of the vas deferens, leading to azoospermia and thus, sterilisation without affecting hormonal and sperm production levels. An open-ended technique was used where a careful incision was made over the spermatic cord between the scrotum and inguinal rings. The common tunic was incised and opened to reveal the vas deferens. A 1-1.5cm segment of the vas deferens was then excised leaving an open testicular end which was left, the cranial end was gently ligated. The tunica vaginalis was then sutured. The subcutaneous layer and the skin were also sutured (Silva, et al., 1993).

Appendix 5 The response variable, factor predictor variables, covariate predictor variables and interaction entered into the GLMM for each hypothesised objective.

Objective	Response variable	Predictor variable	
		Factor	Covariate
1	Procedure duration	Procedure Surgeon	Day number Start time
2	Change in post-operative score	Procedure Surgeon	Day number Start time Time between A1 and A2 Time between finish and awake Procedure duration
3	Time between procedure finish and awake	Procedure Surgeon	Day number Start time Procedure Duration

Appendix 6 Percentage missing data for each variable

	Total	Orchiectomy	Vasectomy	OHE
Surgeon	1.3		3.9	
Start time	22.4	20.6	31.4	16.4
Finish time	21.1	11.8	31.4	17.9
Surgery duration	23	20.6	31.4	17.9
Awake time	21.7	17.7	21.6	23.9
Return time	30.3	23.5	33.3	31.3
A1 E	96.1	100	98	92.5
A1 G	55.9	64.7	62.8	46.3
A1 H	84.9	88.2	62.8	80.6
A1 E	86.2	82.4	96.1	80.6
A2 G	52.6	61.8	54.9	46.3
A2 H	88.8	61.8	54.9	46.3

Appendix 7 The response variable, factor predictor variables, covariate predictor variables and interaction entered into the GLMM for each non-hypothesised exploratory objective

Model	Response variable	Interaction
1	Procedure duration	Surgeon * Start Time Procedure * Day number Procedure * Surgeon Procedure * Start Time
2	Change in post-operative score	Surgeon * Start Time Surgeon * Day number Day number * Time between A1 and A2 Time between A1 and A2 * Stay duration
3	Time between procedure finish and awake	Day number * Procedure duration Surgeon * Start time Day number * Procedure

Appendix 8 Response variables, predictor variables and interactions entered into the GLMM for OHE.

Response variable	Predictor variables	Interaction
Procedure duration	Surgeon Day number Start time	Day number * Start time Surgeon * Day number Surgeon * Start Time
Time taken to wake	Surgeon Surgery duration Day number Start time	Day number * Start time Day number * Procedure duration

Appendix 9 Total number of all procedures, individual procedures and surgeon 1 and surgeon 2 procedures per day across the 10-day intervention period.

Day Number	Total	Orchiectomies	Vasectomies	OHE	Surgeon 1	Surgeon 2
1	12	0	4	8	5	7
2	20	1	9	10	8	12
3	13	2	9	2	7	6
4	20	10	0	10	9	11
5	24	13	0	11	12	12
6	10	5	0	5	3	7
7	6	0	3	3	4	2
8	20	11	2	7	10	10
9	11	5	2	4	4	5
10	16	4	5	7	8	8
TOTAL	**152**	**51**	**34**	**67**	**70**	**80**