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**THE USE OF HORSES FOR UNDERGRADUATE
PRACTICAL TEACHING: ANIMAL WELFARE AND
TEACHING IMPLICATIONS**

A thesis presented in partial fulfilment of the requirements for the degree of

Doctor of Philosophy

in

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ABSTRACT

Teaching horses are used at Massey University, New Zealand during practical classes for equine and veterinary science students to develop, improve and refine their skills. The purpose of this thesis was to investigate the management and use for teaching of these horses and to assess the potential impact of the teaching-related activities on the horses' behaviour and welfare.

The knowledge and competency of students in the veterinary programme at entry level and later in their programme were studied using a questionnaire to provide information on the level of competency of students at entry to the qualification and later after exposure to horses during teaching. The results confirmed previous findings about these students' background (i.e. mostly urban upbringing, mostly female). Confidence around horses and experience with horses were limited for most students entering the veterinary programme. First-year students had greater difficulty in interpreting a horse's behaviour, less understanding of equine learning mechanisms and poorer self-assessed equine handling skills compared to 4th-year students. The students' correct interpretation of equine behaviour was associated with a history of pet ownership, the presence of horses on the students' family property while growing up and the year of study (i.e. students' advancement in the programme).

The use of the horses kept at Massey University for teaching was studied retrospectively over a calendar year. There were seven different types of equine practical teaching classes but each of the three teaching herds was used only for a specific subset of practical class types. A relatively low frequency of teaching-related activities was reported, although there was some variation in the type and number of student interactions and frequency of use of individual herds and horses.

The behavioural activities, i.e. time budgets and herd dynamics, of the teaching horses at pasture were explored at the beginning, during and at the end of a semester of practical teaching. The horses' time budgets were similar to that of free-ranging populations with a majority of feeding and resting behaviours. Social interactions were mostly submissive, and of mild intensity

when agonistic. In addition, hierarchies were relatively linear and stable across time, and a high behavioural synchronisation was reported between pairs of nearest neighbours.

The teaching horses' perception of humans was investigated at the beginning, during and at the end of a semester of practical teaching, through a human-approach test. Horses' positive responses to human approach and contact were associated with a slow pace, straight arms and gaze directed at the horse's shoulder. Horses were significantly less likely to accept human contact if they had been used for teaching more often in the weeks prior to the test.

The behavioural and physiological responses of the horses were evaluated during three types of practical teaching classes (i.e. animal handling, medical rectal- and mare reproductive rectal examinations). Heart rates during practical teaching classes were consistent and in the range of a resting horse. Horses spent most of the time eating hay but ate less during an interaction with students in the mare reproductive rectal examination class compared to being in stocks with no interaction. No change in behaviour was reported in medical rectal examination classes.

The results reported in this thesis provide significant insight about the use for teaching of horses in equine and veterinary science degree programmes. The findings suggest that the teaching horses experienced limited physical, physiological and behavioural stress due to their use for practical teaching classes. Therefore, there may be an opportunity to increase the horses' use for teaching to enhance equine and veterinary students' learning outcomes. Additional work, however, is required to identify other equine welfare indicators that could be applied during equine practical teaching classes to further evaluate the impact of the student-horse interaction. In order to optimise the horses' use for teaching, more research is also warranted to identify the most efficient practice to teach students safe and appropriate equine handling skills and to improve their confidence around horses.

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Oral presentations (* speaker)

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CHAPTER 1. GENERAL INTRODUCTION

PROBLEM STATEMENT

Often explained by the size of horses and their flight-wired response to novelty and danger, many equine-related injuries in people occur during handling (Hawson et al., 2010). The use of inadequate handling techniques may also increase the risk of injury for the horse and elicit stress and fear responses (Klupiec et al., 2014; McGreevy, 2007), which in turn may increase the risk of welfare compromise to the horse. Therefore, skills in horse handling are important to ensure person and horse safety during interactions.

Developing such skills are thus important learning outcomes for a variety of animal-related qualifications, such as equine and/or veterinary science degree programmes. Given the complexity of animal behaviour (Chapman et al., 2007; Gronqvist et al., 2016; MacLeay, 2007), alternatives to the use of live animals, such as simulators or mannequins, cannot be solely used in animal and veterinary science curricula. Therefore, live horses are used to teach equine handling skills in many institutions to provide hands-on experience to students (Austin et al., 2007; Cawdell-Smith et al., 2007; Chapman et al., 2007; Cockram et al., 2007; Hanlon et al., 2007; MacLeay, 2007; McGreevy et al., 2007; Stafford & Erceg, 2007; White & Chapman, 2007). When they enter the programme, veterinary science students often lack experience with horses and may also have limited understanding of horse behaviour. Given that horses' reactions to interactions with humans are influenced by the behaviour and skills of the handler (Hausberger et al., 2008), practical teaching classes may potentially compromise person and horse safety and horse welfare.

Animal-related undergraduate degree programmes offered at Massey University include a 3-year Bachelor of Animal Science (BAnSci) and an accredited 5-year Bachelor of Veterinary Science (BVSc). Massey University's veterinary faculty is the only veterinary school in New Zealand, and undergraduate veterinary students are trained in a broad range of species. Contrary to some veterinary schools which can be more specialised (e.g. training focused on large-animal

species at Charles Sturt University, Australia) (Austin et al., 2007) or where students select their track area for the clinical years, it is only during their final year – dedicated to clinical placements – that Massey University’s students have an opportunity to specialise their training.

Like other universities offering equine and veterinary programmes, Massey University has horses that are specifically kept for teaching (Stafford & Erceg, 2007). This population of teaching horses includes 25 horses (14 mares, 10 geldings and one stallion) retired from sport or racing that are grouped into three herds (except the stallion). These horses are managed and kept at pasture all year round due to the temperate climate in New Zealand (Rogers et al., 2017). However, little has been published about the use, behaviour and welfare of horses used for educational purposes and the potential impact interactions with students during practical classes may have on the horses.

RESEARCH AIM

Therefore, this research aims to study the management and use of the horses kept at Massey University for educational purposes, and to assess the potential impact of these teaching-related activities on the horses’ behaviour and welfare.

THESIS STRUCTURE

The thesis starts with a background to the research question and a review of the current literature discussing the existing knowledge relevant to the research questions and identifying the gaps in the existing literature. The aims and objectives of the thesis are then presented.

Five observational and experimental chapters then follow. These research chapters form a series of published papers which have been formatted into the style of the thesis for consistency. Therefore, some repetition might occur in the introduction and methods sections of some chapters. First, the knowledge and competency of students in the veterinary programme at entry level and later in their programme were studied to provide information on the level of competency of students at entry to the qualification and later after exposure to horses during teaching (**Chapter**

2). **Chapter 3** describes the frequency and use of horses to set a background for the subsequent chapters which go on to investigate how the horse activity and herd dynamics (**Chapter 4**), behavioural responses (**Chapter 5**) and stress indicators (**Chapter 6**) varied with use for teaching.

The thesis ends with a general discussion of the implications of the findings for the use and management of teaching horses and for future research in this area.

REVIEW OF THE LITERATURE

1. INTRODUCTION

This chapter reviews the current literature relevant to horse behaviour, the equine and veterinary science degree programmes and teaching of equine handling skills, and animal welfare. The first section focuses on the interaction between humans and domestic horses, and on the importance of a good knowledge of equine behaviour to safely handle horses. In the second section of this chapter, the necessity to teach these skills to veterinary students is highlighted and the animal handling programme of the equine and veterinary science curricula is examined. This chapter ends with the consideration and measurement of animal welfare and how it can be applied for the use of live horses in the equine and veterinary science teaching environments.

2. HORSES, BEHAVIOUR AND COMMUNICATION

Behaviour and social organisation

Given that horses do not exist as truly wild animals anymore, research on horse behaviour has been carried out on feral (i.e. previously domesticated animals that have reverted to a wild existence) and free-ranging horses to provide insights into the natural behaviours of domestic horses. Such studies are subsequently often used to compare the behaviour of domestic and feral horses.

Time budget of the feral/free-ranging horse

Free of any human management, feral horses divide their time between activities revolving around feeding, resting, locomotory, maintenance and social behaviours. The proportion of time spent performing each behavioural activity is called a 'time budget' (Duncan, 1985) and has extensively been studied in feral and free-ranging horses (Berman, 1991; Boyd et al., 1988; Boyd & Keiper, 2005; Duncan, 1980, 1985; Keiper & Keenan, 1980; Tyler, 1972). Horses are herbivores and preferential grazers who evolved to feed on a high fibre, low starch diet (Harris,

2005). As a result, feral horses are engaged in feeding activities most of their waking time. Although numbers vary, authors agree that feral horses allocate at least 50% of their time (i.e. 12 hours per day) to grazing behaviours (Boyd et al., 1988; Crowell-Davis et al., 1985; Duncan, 1980; Salter & Hudson, 1979).

Making up 30% of the daily time budget, resting behaviours, either in standing position or in recumbency, are the second most observed behaviours in feral horses (Duncan, 1980). The necessity for feral horses to find water and feed resources brings locomotion behaviours as the third largest amount of their daily time budget. The average distance travelled by feral individuals is 15.9 ± 1.9 kilometres per day and up to 55 km/day depending on resource availability (Hampson et al., 2010). The rest of the daily time budget of horses is distributed between grooming, elimination, alert and social behaviours.

Time budgets are not a fixed measure and variations can occur in free-ranging horses due to environmental parameters and individual characteristics. The time of day (Boyd et al., 1988; Ransom et al., 2014; Rubenstein, 1981; Tyler, 1972), climatic conditions (Boyd et al., 1988; Crowell-Davis et al., 1985; Tyler, 1972), season (Berger et al., 1999; Lamoot & Hoffmann, 2004; Rubenstein, 1981; Tyler, 1972), availability of forage type (Rubenstein, 1981), age (Boy & Duncan, 1979; Salter & Hudson, 1979; Tyler, 1972) and reproductive status (Boyd, 1988) are all factors reported within the literature that can influence the time horses spend feeding or displaying other behaviours. Several studies investigating free-ranging horses' time budgets revealed that most foraging behaviours were performed during the daylight period (Berger et al., 1999; Duncan, 1985). Feeding behaviours often occur close to dusk and dawn, and are not evenly distributed during the day, with the least amount of time spent feeding at noon in summer (Boyd et al., 1988; Rubenstein, 1981; Tyler, 1972). Increased feeding activity at night may be explained by higher temperatures during the day (Berger et al., 1999). Authors frequently suggest that the lower abundance of forage available during autumn and winter is an explanation for the higher proportion of time spent grazing reported in autumn and winter (Berger et al., 1999; Lamoot & Hoffmann, 2004; Tyler, 1972). Elimination, grooming, social and alert behaviours occupy shorter

periods of time and generally do not vary with environmental parameters as much as resting, locomotion and feeding activities.

Social dynamics and interactions in groups of horses

Horses are social animals who keep close contact with their conspecifics and live in large herds, which consist of several family groups, up to 35 individuals (Boyd & Keiper, 2005; Keiper, 1986). These groups are also called harem bands and usually comprise one mature breeding male (stallion), several mature breeding females (mares) and their young offspring (McDonnell, 2003). In feral horses, juvenile males that have not yet held a harem and males without broodmares form groups called bachelor bands (McDonnell & Haviland, 1995).

Because of limited resources, such as grazing areas, water or shelter, social orders are formed within bands through bilateral dominance relationships, which are established between pairs of horses (McDonnell, 2003). Although harem bands are more stable over time than bachelor bands, hierarchies have been described in both groups. As stated in a recent report of the International Society for Equitation Science (ISES, 2017), it is unlikely that horses have awareness of the concept of rank order or hierarchy. However, the study of each bilateral (dyad) relationship within a herd provides an understanding of the social dominance between individuals and at the herd-level (Langbein & Puppe, 2004). Drews (1993) attempted to define the concept of dominance and summarised it as follows:

Dominance is an attribute of the pattern of repeated, agonistic interactions between two individuals, characterised by a consistent outcome in favour of the same dyad member and a default yielding response of its opponent rather than escalation. The status of the consistent winner is dominant and that of the loser subordinate.

This definition suggests that the concept of dominance between individuals should first be evaluated at the dyad-level (Langbein & Puppe, 2004). Within a pair of horses, this level refers to the asymmetry of the displayed agonistic behaviours and is

the start point to any further description of a group's social relationships. Langbein and Puppe (2004) proposed to describe social dominance at two other levels for a global understanding of the structure of the group. The second level of analysis is at the group level and can provide information on the strength and stability of bilateral relationships; while the third level places each individual in relation to the other individuals with which it has interacted (e.g. often attributes a rank position). To describe social dynamics in groups of horses, authors generally focus on the group (strength and stability of rank position (Ingólfssdóttir & Sigurjónsdóttir, 2008; van Dierendonck et al., 2004), Directional Consistency Index, DCI (Vervaecke et al., 2006)) and/or individual levels (dominance index (Krueger et al., 2014; Schneider & Krueger, 2012), rank order (Hauschildt & Gerken, 2015; Heitor et al., 2006)) and do not always specifically describe the types of dyads (i.e. unknown, one-way, two-way, tied) observed within the group. When reported, this information is crucial to better understand social dynamics within groups of horses. Indeed, some measures of the group or individual levels (e.g. DCI) have no or limited sensitivity to unknown or tied relationships.

Varying from subtle to more violent, agonistic interactions in domestic and free-ranging horses refer to behaviours such as displacement, threat (bite or kick), chase, bite, kick, push or rear (Christensen et al., 2011; Heitor et al., 2006; Ransom & Cade, 2009). Because of the potential high cost for the animals, the number and intensity of agonistic interactions decrease when the hierarchy is formed in favour of spontaneous avoidance and displacements (Heitor et al., 2006). Agonistic interactions are reported to be of higher intensity between two animals of similar ranks than between individuals where a clear dominance relationship has been established (Keiper & Receveur, 1992). Unstable bands, on the contrary, have an increased risk for bite and kick injuries (Knubben, Gygax, Auer, et al., 2008).

Although agonistic interactions occur when inter individual distances decrease, i.e. an animal's personal space is invaded (Tyler, 1972), horses are frequently seen less than two meters away from a conspecific, regardless of group composition and enclosure size. This spatial

proximity in individuals' personal space is usually observed between animals of similar rank and age (Clutton-Brock et al., 1976) and generally do not exceed two social preferred partners per individual (Monard & Duncan, 1996).

Linear or triangular dominance relationships can be observed in horses, the latter more frequently in large bands of feral horses. In a triangular social organisation, an individual A can be dominant over individual B, itself dominant over individual C, which is dominant over A. Dominance relationships have been shown to remain stable for up to 18 months (Houpt & Wolski, 1980) and three years (Keiper & Sambraus, 1986). However, dominance relationships are a dynamic process and tend to be influenced by environmental (Berman, 1991; Franke Stevens, 1990; Ransom & Cade, 2009) and individual characteristics (Tyler, 1972). Whereas most studies agree on a positive correlation between position in the herd and age (i.e. older individuals are of higher position in the herd) (De Vries et al., 1994; Heitor et al., 2006; Houpt et al., 1978; Ingólfssdóttir & Sigurjónsdóttir, 2008; Keiper, 1988; Keiper & Sambraus, 1986), the influence of size and weight is less clear (De Vries et al., 1994; Houpt & Keiper, 1982; Houpt et al., 1978; Ingólfssdóttir & Sigurjónsdóttir, 2008; Tyler, 1972). In a captive herd of Icelandic horses, De Vries et al. (1994) found no influence of height, whereas weight was correlated with rank in the study of a similar population by Ingólfssdóttir and Sigurjónsdóttir (2008). Authors also report, in both feral and captive horses, a positive correlation between time in the group (i.e. longer residency) and an individual's relative position in the herd (De Vries et al., 1994; Keiper & Sambraus, 1986). Additionally, temperament may be a significant indicator of rank (Houpt et al., 1978), and either stallions or mares can be the highest-ranked animals depending on the context (Houpt et al., 1978; Keiper & Sambraus, 1986; Tyler, 1972).

Behaviour of the domestic horse

Despite over 7,000 years of domestication, horses managed by humans seem to display all the behaviours that are essential to feral horses' survival (Fraser, 2010). Duncan (1985) suggested that variations in time spent engaged in various activities may reflect the animals' ability to cope

with changes in their environment. When disturbed, variations in time budgets of domestic horses are often due to housing and management practices.

The effects of management on horse behaviour

In the literature, despite no clear definition and consensus in the use of the terms, a paddock is used to describe an outdoor enclosed area from less than 1ha up to 10ha (Benhajali et al., 2008; Flauger & Krueger, 2013; Heleski et al., 2002; Jørgensen et al., 2009; Majecka & Klawe, 2018), whereas large enclosures refer to bigger areas up to 1300ha (Duncan, 1980; Keiper & Receveur, 1992; Tyler, 1972). Horses kept at a high density (200 individuals/ha) in a paddock showed altered time budgets (i.e. animals engaged in more locomotion) compared to feral horses (Benhajali et al., 2008). These results highlighted the importance of the enclosure size and space available per animal when comparing time budgets of domestic and feral or free-ranging horses.

When kept in groups, most of the aggressive interactions between horses occurring in stable groups consisted of threats (Jørgensen et al., 2009). For horses managed at pasture, a larger size of enclosure and available area per horse has been linked to less aggressive interactions between conspecifics (Flauger & Krueger, 2013; Jørgensen et al., 2009; Majecka & Klawe, 2018). Higher numbers of agonistic interactions per hour (8-10/hour) have been reported for free-ranging herds kept in semi-natural conditions (i.e. large enclosures of 35-350ha, requiring no or low supplementary feeding) (Keiper & Receveur, 1992) compared to feral or free-ranging bands managed in bigger areas (i.e. 5000-10,000ha), where this number averages 1.5/hour (Clutton-Brock et al., 1976; Houpt & Keiper, 1982). Through the observation of 11 different groups of domestic horses kept in enclosure of various sizes (0.04-1ha), Flauger and Krueger (2013) reported a negative association between the size available per horse and the level of aggression. The authors observed lower numbers of agonistic interactions (2-7/hour) in enclosures with more than 331m² per horse, intermediate numbers of agonistic interactions (8-22/hour) in enclosures with 106-331m² per horse, and high numbers of agonistic interactions (23-85/hour) in enclosures

with less than 106m² per horse. An enclosure size of more than 331m² per horse was even suggested to reduce aggression close to zero (Flauger & Krueger, 2013).

As a highly sociable species, horses benefit from a management in groups which allows contact with conspecifics and performance of a range of social behaviours (Yarnell et al., 2015). Horses kept alone in a pen, with no conspecifics in the adjacent pen, showed increased locomotion behaviours, at the expense of feeding time, suggesting an impact of social isolation (Houpt & Houpt, 1988). Nevertheless, single stalls or boxes are the predominant housing system in most European countries (Hartmann et al., 2015; Hockenhull & Creighton, 2015; Knubben, Gygax, & Stauffacher, 2008; Larsson & Müller, 2018; McGreevy et al., 1995). This management system drastically restricts movements, reduces the foraging opportunity and creates social isolation. Individually-stabled ponies who were provided with *ad libitum* hay and opportunity to interact and see conspecifics showed time budgets similar to that of feral horses (Sweeting et al., 1985). On the contrary, individual stabling affected the behaviour of young horses previously kept in groups and stabled for the first time (Visser et al., 2008). Three weeks after being stabled, 67% of the individually-stabled horses developed a stereotypy, compared to none for the pair-housed horses. Stereotypies have been defined as repetitive, invariant behaviour patterns and appear to serve no obvious function (Dantzer, 1986; Mason & Latham, 2004). They are not reported in feral horses. The occurrences of nibbling, neighing and snorting behaviours also increased in individually-stabled horses compared to pair-housed horses (Visser et al., 2008).

Management decisions also seem to affect horse behaviour towards humans. Søndergaard and Halekoh (2003) investigated the approachability of young horses either stabled individually or in groups directly after weaning, and found single-housed animals easier to approach by humans. On the contrary, adult horses housed individually and with no possible contact with conspecifics were more difficult to handle than grouped-housed, pair-housed and single-housed horses with semi-contact with conspecifics allowed (Yarnell et al., 2015). Vigilance behaviours, which can be an indicator of acute (Morgan & Tromborg, 2007) or chronic stress (Carlstead et al., 1993), increased in single-housed horses vs pair-housed horses (Visser et al., 2008). However,

in single-housed horses, the presence of a neighbour through grids decreased the frequency of alert postures (Lesimple et al., 2019), suggesting lower level of stress in presence of a conspecific.

To date, most of the behavioural and social research on domesticated horses kept in groups at pasture has focused on social interactions between individuals (Burla et al., 2016; Flauger & Krueger, 2013; Jørgensen et al., 2009; Majecka & Klawe, 2018; Pierard et al., 2019) or specific behaviours, such as feeding and drinking (Crowell-Davis et al., 1985) or locomotion (Kurvers et al., 2006). The time spent engaged in each type of activity, however, has received very little attention except for night-time behaviours (Houpt et al., 1986), horses kept at high density in a bare paddock (Benhajali et al., 2008), horses grouped with other species (Arnold, 1984), or to evaluate the effect of pasture management (Maisonpierre et al., 2019). More work is therefore required to describe the time budgets of domesticated horses managed in stable groups and kept at pasture year-round.

Horse communication and learning

As a prey species, horses use subtle cues to communicate with their conspecifics. These cues may take the form of changes in their body posture, such as the position of their ears, tail, mouth, head and feet (Goodwin, 1999). Their subtlety is such that, although negative states seem easier to recognise than positive ones (Bell et al., 2019), even horse caregivers do not always correctly identify the behavioural stress indicators of horses (Bell et al., 2019; Horseman et al., 2016). Moreover, the behavioural signals used by horses to communicate are based on an escalation ladder from very subtle to more explicit (Goodwin, 2007), which an untrained or inexperienced handler might not be able to recognise. Lastly, horses are flight animals with a response to danger based on avoidance and escape behaviours. This may explain why the species is often considered as unpredictable by people unable to identify early behavioural changes in horses. A good knowledge of equine behaviour and attention to the environment are widely encouraged to increase the predictability of horses and mitigate the risks of human injury (Carmichael et al., 2014).

The number of people who interact on a daily basis with horses professionally and recreationally is large (van Dierendonck & Goodwin, 2005) and each interaction between a human and a horse constitutes a potential learning experience for the horse, as horses learn through a trial and error system (Pearson, 2015a). Learning can be defined as the “process whereby experience produces a relatively permanent change in the response to a stimulus” (McGreevy & Boakes, 2011). Learning theory, which documents the way animals learn, includes two types of learning: non-associative (i.e. habituation and sensitisation) and associative (i.e. classical and operant conditioning) learning (Starling et al., 2016). Non-associative learning refers to the repeated use of a single stimulus to change the strength of the behavioural response. Through the process of habituation animals become progressively accustomed to a stimulus; there is a reduction in their response to the stimulus over time (McLean & Christensen, 2017). Associative learning involves establishing an association between two previously unrelated stimuli. In classical conditioning, first described by Pavlov (1927), innate behaviours are triggered by using a previously neutral stimulus. In operant conditioning, stimuli are used as reinforcers or suppressors to increase ('reinforcement') or decrease ('punishment') the frequency, duration or intensity of a specific behavioural response. This is done by applying (i.e. 'positive') or removing (i.e. 'negative') a desirable/pleasant or noxious/aversive stimulus (McBride et al., 2017). Based on the interaction between a stimulus, a response and a consequence (Cooper, 1998), this learning process allows the animals to associate two events and respond voluntarily.

With horses, negative reinforcement has historically been and is still commonly used by caregivers, trainers and riders (McLean, 2005; McLean & Christensen, 2017). There seem to be two major reasons for the common use of negative reinforcement in horse training. The first reason is the inherent nature of horses for which any novel stimulus can be perceived as aversive. Removing the stimulus when the desired behaviour is shown, therefore immediately eliminates the associated discomfort (McLean & Christensen, 2017) and reinforces the behaviour. The second reason comes from the difficulty of applying positive reinforcers (such as food) when riding horses, compared to when handling horses. However, when they can be applied, positive

reinforcement methods have been proven to be efficient and beneficial in the training of horses. Therefore, both positive and negative reinforcement methods can improve husbandry and handling (Innes & McBride, 2008), and facilitate veterinary procedures (Pearson, 2015a).

Low stress handling

Low stress handling techniques have been promoted for handling small animals (e.g. dogs, cats, rabbits, rodents) in the veterinary practice, as they aim to reduce fear and pain responses for the animals (Lloyd, 2017; Rodan et al., 2011). The application of these techniques in the handling of horses undergoing veterinary procedures have been investigated more recently with increasing evidence of their benefits (Foster & CAAB, 2017; Pearson, 2015a, 2015b; Watson & McDonnell, 2018).

One official guideline on the physical restraint of animals is the AVMA's (American Veterinary Medical Association) policy which recommends using "the least restraint required to allow the specific procedure(s) to be performed properly, and [to] protect both the animal and personnel from harm" (AVMA). In anticipation of potentially dangerous situations, chemical restraint can be used as the preferred method when handling horses. However, it was suggested that chemical restraint methods, although decreasing the horses' stress behaviours might not eliminate the risk of injury due to a reduction of people's vigilance (MacLeay, 2007).

Good handling skills have been defined by Coleman and Hemsworth (2014) as a general knowledge of the animal's needs, a practical experience in the care of the animal and an ability to quickly identify changes in behaviour, health or performance as well as addressing those appropriately. The application of these skills should aim to increase human and animal safety by limiting the stress reactions of both the animal and the handler (Chastain, 2017). In the context of a veterinary procedure (Watson & McDonnell, 2018), the handler, the patient and the veterinarian may be at risk of injury. Most (95%) surveyed equine-specialised veterinarians in the United Kingdom indicated working at least several times per month with horses that they perceived difficult (Pearson et al., 2020). Aversions to stimuli specific to the veterinary context (e.g.

injections, clippers) were one of the most common unwanted behaviours reported by the veterinarians. In response, chemical and physical restraint were the most frequent methods used. As the authors of this study suggested (Pearson et al., 2020), the high prevalence of injuries in this population of veterinarians indicate that physically or chemically restraining horses for examination and treatment may not be the most appropriate approach. Using both effective and safe techniques may help minimise the pain responses and reactivity levels of the animals (Doherty et al., 2017; Hanlon et al., 2007; McGreevy, 2007). The use of learning theory techniques has shown its value in the veterinary environment (Pearson, 2015a, 2015b) but their application requires knowledge and skill to provide the desirable outcomes. To apply low stress handling techniques, handlers and veterinarians are therefore likely to benefit from an understanding of the learning mechanisms in horses.

In the literature, for equestrians and veterinarians, there is often a discrepancy between the understanding of learning theory concepts and the understanding of the terminology of these concepts. Habituation and classical conditioning may be the easiest concepts to understand, as most (97% and 91%, respectively) equine-specialised veterinarians who perceived that they understood these concepts correctly identified the scenarios provided (Pearson et al., 2020). However, different results were reported for positive and negative reinforcement and punishment. Brown and Connor (2017) reported a relatively good understanding of positive reinforcement amongst professional (over 90% of correct and partially correct) and amateur equestrians (over 90% of correct and partially correct) in the UK. However, positive reinforcement was described as negative reinforcement by 50.6% of professional equestrians in Australia (Warren-Smith & McGreevy, 2015). Similarly, negative reinforcement was described as positive punishment by 22.5% of UK-based amateur equestrians (Brown & Connor, 2017) and as punishment by 51.5% of professional equestrians (Warren-Smith & McGreevy, 2015). Despite a perceived good understanding of positive and negative reinforcement, respectively only 19% (of the 84% who thought they understood) and 33% (of the 80% who thought they understood) of equine-specialised veterinarians identified the correct scenarios (Pearson et al., 2020). These

discrepancies between the understanding of concepts and the understanding of the terminology were also reported for positive and negative punishment in these studies. While positive punishment was correctly described by just over a third of professional equestrians and by a fraction of amateur equestrians (just over 10%) (Brown & Connor, 2017), Warren-Smith and McGreevy (2015) indicated that 82% of professional equestrians described negative punishment as negative reinforcement. In addition, Pearson et al. (2020) reported that less than half of equine-specialised veterinarians perceived that they understood what punishment was, whether positive or negative, and that respectively only 43% (of the 38% who thought they understood) and 67% (of the 47% who thought they understood) were able to correctly identify the scenarios.

Despite the body of research highlighting a misunderstanding or potential lack of knowledge of the terminology of positive and negative reinforcement and punishment, these studies present one major limitation. Indeed, they are questionnaire-based and potentially do not properly investigate the techniques used by professionals. Therefore, the results of these studies may not fully reflect the application of learning theory concepts and techniques by equestrians and veterinarians. However, when equine veterinarians were asked about the restraint methods they find useful, Pearson et al. (2020) highlighted that an emphasis was made on physical restraint in the free comments section, along with no mention of learning theory techniques. This may suggest that learning theory techniques are less frequently used by equine veterinarians compared to physical restraint, but further research objectively assessing the type of techniques used by these professionals on horses is required to confirm this hypothesis and to determine the reason behind this lower frequency of use.

Application of signals

The key principles involved in the operant conditioning type of learning are the timing, the predictability of the response and the reward of the correct response. Although they have a good long-term memory (Wolff & Hausberger, 1996), horses have a poor short-term memory (Pearson, 2015a)). For optimal results, the application or removal of the stimulus must be as close as

possible to the desired behavioural response. Due to horses' ability to discriminate between stimuli, the stimulus chosen for a particular response must be specific to that response and trainers must aim for maximum consistency (McCall, 1990). Poor timing, inconsistency of and/or inadequate signals may lead to a dissociation between the stimulus and the desired response. This can lead to confusion for the horse (McLean & Christensen, 2017; McLean & McGreevy, 2010; Overmier & Wielkiewicz, 1983) and result in the development of conflict behaviours, also referred to as evasions and resistances, such as mouth opening, head shaking or excessive tail swishing in the ridden horse (Górecka-Bruzda et al., 2015; McGreevy et al., 2005). Increased arousal levels are linked to the horse's flight response and therefore dangerous responses from the horse may occur (Bartolomé & Cockram, 2016; McLean & Christensen, 2017; Starling et al., 2013).

3. MANAGING VETERINARY STUDENTS' LEARNING ABOUT EQUINE BEHAVIOUR AND HANDLING SKILLS

Equine-related injuries

Horses are one of the most dangerous domestic species due to their size and flight-wired response to danger. They were reported as the main cause of animal-related fatalities after excluding zoonotic disease and motor vehicle collisions involving animals (Lathrop, 2007), and as the second cause (95/350 fatalities) in a study of work-related deaths due to animal-related events (Langley & Hunter, 2001). When involved in non-fatal accidents of riders or handlers, horses can be responsible for severe injuries and significant hospitalisation stays (Abu-Zidan & Rao, 2003). The definition of significant injury differs among authors, therefore, comparisons between the injury rates reported in different studies should be considered cautiously. Some define injuries as resulting in a major impact on work because a hospital admission was required, work-time was lost or the usual working pace could not be maintained for a few days (Fritschi et al., 2006; Nienhaus et al., 2005). Others define an injury resulting in sought or self-administered medical treatment (Landercasper et al., 1988). Multiple human body parts are frequently affected during an accident involving horses, similarly to other large-animal species accidents (Davidson

et al., 2015). The injury rate to humans from horses can range from 16 to 233 per 100 000 people (Jagodzinski & DeMuri, 2005; Smartt & Chalmers, 2009; Williams & Ashby, 1992), and activities other than riding are responsible for up to 50% of equine-related injuries (Hawson et al., 2010). Records of patients who suffered from an equine-related injury highlight being kicked (23%) or crushed by or trampled on (8%) as the most non-riding horse-associated injuries (Davidson et al., 2015). Both handlers and riders can suffer from head trauma due to a horse-related accidents, whereas handlers often sustain more injuries to the abdomen (17% vs 7%) and or the face (22% vs 5%) and riders have more injuries to the lower extremities (28% vs 12%) and chest (16% vs 9%) (Carmichael et al., 2014).

Handling horses in the veterinary environment

Carrying out an effective examination of an animal is a significant risk for veterinarians, who are responsible by law for their own safety and, that of their personnel, as well as patients and clients (Chapman et al., 2007; MacLeay, 2007; McGreevy & Dixon, 2005). In part, due to the amount of time they spend around animals, veterinarians from equine, large-, mixed- and small animal practices perceive the risk of accidents as fairly or very high (Reijula et al., 2003). Studies investigating injuries sustained by veterinarians support this observation. Compared to medical professionals, veterinarians and their associated personnel are 9.2 (95% CI 8.12-10.42) times more likely to experience a severe occupational accident (Nienhaus et al., 2005) and it is expected that the average veterinarian will sustain 7-8 serious work-related injuries over a 30-year career (British Equine Veterinary Association, 2014; Jeyaretnam et al., 2010). Injuries sustained by veterinarians working with large animals tend to be more common, as indicated in studies which report an increased likelihood (1.2-10+) of having had a recent injury for large- and mixed-animal (small and large) veterinarians compared to small animal veterinarians (Epp & Waldner, 2012; Fritschi et al., 2006; Nienhaus et al., 2005).

From all types of practitioners, equine veterinarians are the most likely to view animal-related accidents as a significant risk (Reijula et al., 2003). In a survey of Dutch equine

veterinarians, 67% of the self-reported diseases or injuries were related to work, which is significantly higher than most employees in Europe (35%) (Loomans et al., 2008). With a loss of working time that can range from 0 to 180 days (mean 7.4 ± 0.1 days) (Lucas et al., 2009), equine-related injuries can severely impact a veterinarian's activity. The majority of horse-related accidents suffered by equine veterinarians occurred during interventions of low risk requiring little attention (Pasquet & Denoix, 2005). The major causes of injury reported by Australian equine veterinarians were being kicked or struck by a horse (79%), being crushed, pushed or stepped on (12%), and bitten (2%) (Lucas et al., 2009).

Equine and veterinary science students also face the risks of equine-related injury. Findings similar to those reported for equine veterinarians were described at an Australian University in a survey of 260 veterinary and animal science students (Riley et al., 2015), with 8.5% of the respondents reporting equine-related incidents resulting in injuries. At the time of the incident, students were either standing near a horse (26%), performing a handling or husbandry procedure (27%), or performing a non-invasive physical examination procedure (17%). Major causes of injury included being trampled on (30%), kicked by hind limb (30%), crushed, knocked over or struck by the horse's head (13%), and bitten (13%) (Riley et al., 2015), which is similar to causes reported for Australian equine veterinarians (Lucas et al., 2009).

The factor of experience

Factors mitigating animal-related injuries are still debated in the literature. Whereas cumulative exposure to animals could be associated with an increased risk of serious injury (Kriss & Kriss, 1997), experience does not appear to moderate the severity of injury (Ball et al., 2007). Hawson et al. (2010) suggested that "experience in itself would not be preventative if new knowledge has not been incorporated into practice". However, experience is believed by some to be a protective factor for the risk of injury when interacting with animals (Gabel & Gerberich, 2002). Fritschi et al. (2006) conducted a cross-sectional survey of Australian veterinarians and revealed that recent veterinary graduates were 1.8 times more likely to have had a recent injury

than less recent graduates. The lack of experience was also reported as the second most important factor (39%, 9 respondents) associated with equine-related injuries of surveyed veterinary and animal science students in Australia (Riley et al., 2015). In this survey, seven students (30%) also identified the behaviour of the horse (distressed or fearful) at the time of the incident as another risk factor. Fear or stress responses from the horse were associated with 20% of handler injuries in another study by Williams and Ashby (1992) investigating injuries involving horses.

Several authors have suggested that increased knowledge of horse behaviour and increased training in equine learning theory may reduce the prevalence of horse-related injuries (Lathrop, 2007; Pearson et al., 2020). Gronqvist et al. (2017) found an association between inexperience and inability to correctly identify equine negative behavioural cues in first-year veterinary and equine science students. In order to avoid, or at least mitigate, human injuries, it seems essential to teach students what stimuli could lead to an unexpected response of the animals they work with and how to address this response (MacLeay, 2007). However, applying the appropriate signal at the adequate time and recognising the desired behavioural response may not be possible for novices. A sufficient knowledge of equine behaviour may therefore be required for a correct use of learning theory. More research is therefore required to assess the amount of knowledge students that work with horses during their undergraduate curriculum have of equine behaviour.

Teaching handling skills to the equine and veterinary science students

Students' background

Many studies report the importance of teaching handling and equine behaviour in the veterinary curriculum (Calder et al., 2017; Klupiec et al., 2014; Loomans et al., 2008; Miller et al., 2004). A change of behaviour is often the first sign of a pathology (Horwitz, 2017) and, therefore, the knowledge of equine behaviour and the ability to identify normal behaviours are especially crucial for veterinarians in the health assessment of an animal. In a recent study, graduating veterinary students felt increasingly more prepared for their 1st day of practice if they had received sufficient training in the identification of abnormal behaviours (Calder et al., 2017).

In addition, there is a need for equine veterinarians to recognise and manage behavioural issues as they frequently interact with horses that they consider difficult, i.e. that present unwanted behaviours, during their examination (Pearson et al., 2020). Many horses develop behavioural issues due to inadequate handling in previous interactions (Pearson, 2015a, 2015b). Not standing still, shyness (needle, head and clipper) and kicking or striking with foot were reported by Pearson et al. (2020) as the most common behavioural issues encountered by equine veterinarians on at least a monthly basis. Many of these common equine behavioural issues can be avoided or managed through equine behaviour education and low stress handling techniques (Pearson, 2015a, 2015b).

Veterinarians in private equine practice in North America reported using several handling techniques (e.g. attach halter and lead, elevate limb, restrain) on a weekly basis and expected a high level of proficiency for these techniques from new graduates (Hubbell et al., 2008). There is a consensus across veterinary schools that animal handling is a necessary skill for students to master (Austin et al., 2007; Cawdell-Smith et al., 2007; Chapman et al., 2007; Cockram et al., 2007; Hanlon et al., 2007; MacLeay, 2007; McGreevy et al., 2007). By demonstrating good knowledge in animal behaviour and handling animals adequately, veterinarians will inspire confidence in their clients and, in addition, will be capable of teaching clients the right methods of handling (Klupiec et al., 2014; McGreevy, 2007).

A demographic shift in the students enrolling in veterinary and animal science degrees has occurred in the past two decades (Cawdell-Smith et al., 2007). In several countries, students from urban areas enrolling in these degrees outnumber those who have grown-up in rural environments (Heath, 2008; Ostovic et al., 2017; White & Chapman, 2007). In New Zealand, only 28% of veterinary science and veterinary nursing students come from either small towns, villages or rural areas (Wake et al., 2006). It is hypothesised that students from an urban background may have a reduced exposure to a range of domestic animal species compared to students from more rural backgrounds. Wake and Stafford (2006) reported that undergraduate students from rural areas were more likely to have lived with, or owned, a dog prior to starting their degree. The change in

demographic profile of veterinary students suggests that some students may never have been close to horses or production animals before starting their degree (Chapman et al., 2007; Old & Spencer, 2011). This was confirmed by Riley et al. (2015) who surveyed undergraduate veterinary and animal science students and reported 60% of horse exposure in this population of students prior to starting their degree, with a heterogeneous exposure duration of 340 ± 811 days (Riley et al., 2015). Students enrolling in veterinary and animal science degrees therefore come from a variety of environments and social backgrounds, have various levels of experience with animals (Klupiec et al., 2014) and knowledge of equine behaviour (Gronqvist et al., 2017), and show low confidence in handling horses and other large animals (White & Chapman, 2007).

The equine handling programme

Undergraduate veterinary and animal science degrees commonly last between 3 and 6 years and basic animal handling skills are often taught within the first two years of a veterinary programme (Austin et al., 2007; Cawdell-Smith et al., 2007; Chapman et al., 2007; Cockram et al., 2007; McGreevy et al., 2007; Stafford & Erceg, 2007). After being provided with the basic safety principles for working around horses and the basic aspects of animal behaviour and welfare, students in many Australasian programmes learn how to approach and catch, lead, groom, pick up both front and hind feet, and put a rug (Austin et al., 2007; Cawdell-Smith et al., 2007; Chapman et al., 2007; Cockram et al., 2007; McGreevy et al., 2007; Stafford & Erceg, 2007). Animal handling is a core subject of veterinary and animal science degrees taught as a specific topic and as a component of other subjects. By participating in various activities that represent an animal handling learning opportunity, such as physical examination, blood collection or hoof trimming, students can refine their technique. Teaching these basic skills in practical classes given early in the programme aims to develop veterinary and animal science students' ability to handle and restrain animals. It also ensures that students have adequate technique before leaving campus for extramural placements (Austin et al., 2007; Cockram et al., 2007).

Equine practical teaching classes

A variety of different types of practical teaching classes are held during the equine and/or veterinary science degree programmes: animal handling (Austin et al., 2007; Cawdell-Smith et al., 2007; Chapman et al., 2007; Cockram et al., 2007; Hanlon et al., 2007; MacLeay, 2007; McGreevy et al., 2007; Stafford & Erceg, 2007; White & Chapman, 2007), lameness investigation (MacLeay, 2007), rectal examination (Berghold et al., 2007; van Vollenhoven et al., 2017), dental examination and treatment (Austin et al., 2007). Many universities keep dedicated group- or single-housed ‘teaching horses’ specifically for these classes (Austin et al., 2007; Chapman et al., 2007; Hanlon et al., 2007; Stafford & Erceg, 2007). Because of the diversity in both equine experience and confidence between veterinary students (Austin et al., 2007; Cockram et al., 2007; Stafford & Erceg, 2007; White & Chapman, 2007), the horses are cautiously selected based on their temperament and sedate characteristics. Therefore, anxious or distressed animals are not used (McGreevy et al., 2007; Stafford & Erceg, 2007). Furthermore, teaching horses are assigned to types of practical teaching classes based on their tolerance of the teaching procedures and apparent ability to cope with their use.

To date, reports of the use of horses in practical teaching in equine and veterinary science degree programmes have been limited to one type of practical class (animal handling) and have involved teaching across a range of species (Austin et al., 2007; Cawdell-Smith et al., 2007; Chapman et al., 2007; Cockram et al., 2007; Hanlon et al., 2007; MacLeay, 2007; McGreevy et al., 2007; Stafford & Erceg, 2007; White & Chapman, 2007). Reports focus on the topics covered and provide few details on the horses used. When reported, information regarding the horses and their use in equine handling practical classes may include the total number of horses available (Austin et al., 2007; Chapman et al., 2007; MacLeay, 2007; McGreevy et al., 2007), the student-horse ratio (Austin et al., 2007; Cawdell-Smith et al., 2007; Chapman et al., 2007; Cockram et al., 2007; Stafford & Erceg, 2007), the number of practical classes per student (Austin et al., 2007; Cockram et al., 2007), the frequency of practical classes (Cockram et al., 2007) and the duration of practical classes (Austin et al., 2007; Cawdell-Smith et al., 2007; Chapman et al., 2007;

Cockram et al., 2007; Hanlon et al., 2007; Stafford & Erceg, 2007). However, information on the nature and frequency of the teaching horses' use, or details for other types of practical class have not been reported. Furthermore, while parameters to describe the workload of horses have been reported for other working horse populations, such as horses used for recreational riding or sport horses (Bolwell et al., 2015; Lonnell et al., 2014; Lonnell et al., 2012; Munsters et al., 2013; Rogers et al., 2007; Sloet van Oldruitenborgh-Oosterbaan et al., 2006; Verhaar et al., 2014; Walters et al., 2008), parameters for describing the workload of teaching horses, such as the type, frequency and regularity of their interactions with students, have not been defined or measured.

During practical classes, horses are being handled by novices, who might have poor handling skills and show inconsistencies in their approach and use of signals. Individual horses are likely to vary in their perception of these novice interactions and their response to them. This will add further to the variation in individual experience amongst horses that occurs because of disparities in regularity, frequency and type of interaction with equine and veterinary students. More research is required to investigate what effect, if any, repeated and varied interactions with students may have on the horses used for practical teaching.

4. ANIMAL WELFARE, A MULTI-DIMENSIONAL CONCEPT

Animal welfare and measures of assessment

The use of live animals

As stated in the US Code of Federal Regulations, the “handling of all animals shall be done as expeditiously and carefully as possible in a manner that does not cause trauma, [...] behavioural stress, physical harm, or unnecessary discomfort” (U.S. Code of Federal Regulations, 2019). The use of animals in research and teaching is regulated by the 3Rs. Formulated by Russell et al. (1959), the 3Rs stand for Replacement (“any scientific method employing non-sentient material”), Reduction (“right choice of *strategies* in the planning and performance of whole lines of research”) and Refinement (“reduction to an absolute minimum the amount of distress imposed

on those animals that are still used”). This tenet advocates for “the humanest possible treatment of experimental animals”, which the authors viewed as “a prerequisite for a successful animal experiment”.

Based on the 3Rs tenet, Animal Ethics Committees, composed of professional veterinarians, scientists and non-animal users, review and approve any procedure requiring animals. The decision-making process of these committees strives to regulate the animals’ safety and well-being (McGreevy et al., 2007) by controlling the number of animals and type of procedures, and by maximising the cost-benefit ratio. Through appropriate justification, this utilitarian view allows the use of and subsequent potential compromises of welfare of some animals for the gain in practical skills for the wider benefit of other animals, and therefore requires informed data to determine the costs (Mellor & Reid, 1994).

Welfare, concept and definition

Broom (2011) proposed that the term ‘animal welfare’ describes “a potentially measurable quality of a living animal at a particular time and hence is a scientific concept”. However, three views of animal welfare have been identified during recent decades, and with them, their related assessment methods: biological functioning (health, growth and productivity), affective state (pain, suffering, other emotions and feelings) and natural living (expression of normal behaviours) (Fraser, 2003). The complexity of the abstract concept that is animal welfare may explain the significant body of research available on its attempt to provide a reliable and repeatable assessment. Hence, a unique measure or method cannot cover all the dimensions of animal welfare and the literature agrees that a holistic approach is mandatory in the evaluation of an animal’s welfare state.

Assessment of welfare

Formulated in 1994 by Mellor and Reid (1994) and progressively refined thereafter (Green & Mellor, 2011), the Five Domains Model was first developed in order to assess animal welfare

compromise in experimental animals. This welfare assessment model covers four overlapping physical domains (i.e. nutrition, environmental challenge, physical health, behaviour) as well as a “mental state” domain (i.e. anxiety, fear, pain and distress). Therefore, an individual’s welfare can be assessed in the short or long term using a multi-dimensional approach. Protocols developed to assess the welfare of animals on-farm, such as the European Animal Welfare Indicators Project (AWIN, 2015) or the European Union Welfare Quality® project (Welfare Quality® Protocol, 2009), generally use a combination of animal-based (“indicators taken directly from the animal”), resource-based (“measures taken regarding the environment in which the animals are kept”) and management-based measures (“measures which refer to what the manager does on the animal group and what management processes are used”) (AWIN, 2015; EFSA Panel on Animal Health and Welfare, 2012).

Environmental-based parameters are inherent to one accepted definition of animal welfare formulated by Broom (1986), which defines welfare as “the state of an individual and its attempts to cope with its environment”. Coping mechanisms can require considerable effort from the animals, and a failure or a difficulty to manage environmental conditions may have an impact on their survival and fitness (Broom, 1986). Measured in the animal’s environment, these parameters focus on the availability of essential resources. They can be resource- (e.g. food and water provision, size of enclosure) or management-based (e.g. health plan, time in paddock, group size). As indicated by Dalla Costa et al. (2014), in some situations, such as the assessment of the absence of prolonged thirst, resource-based parameters “are the most valid, reliable, and feasible indicators for on-farm assessment of this criterion”. However, environmental-based parameters are indirect measures of welfare, as the presence of a resource does not guarantee its use by the animals (Beggs et al., 2019). The combination of several parameters is therefore likely to provide more informed interpretation (Hawkins et al., 2011).

Animal-based parameters, however, provide direct information about the effects on the animal (AWIN, 2015). The European Food Safety Authority even states that these parameters are “the most appropriate indicators of animal welfare and a carefully selected combination of animal-

based measures can be used to assess the welfare of a target population in a valid and robust way” (EFSA Panel on Animal Health and Welfare, 2012). For this reason, and because they are more direct measures of an animal’s welfare, animal-based parameters, which provide information on physical health, physiology and behaviour of the animals, are used more frequently.

Physical health

Important animal welfare indicators are the presence of disease or injury as they are often associated with negative experiences, such as pain discomfort or distress (Rousing et al., 2001). In horses, welfare indicators of an animal’s physical health may be the use of body condition scores (BCS), presence of lameness or lesions, hair coat and hoof condition, consistency of manure, abnormal breathing or coughing (AWIN, 2015). These indicators require no, or minimal, handling of the horses and are therefore considered as readily applied robust and feasible animal-based measures used in animal welfare assessment at farm level (AWIN, 2015).

Physiology

The evaluation of stress responses as a reflection of the animals’ mental state is frequently used to provide information on animal welfare. In the veterinary context, Fraser et al. (1975) defined stress as a state in which an animal “is required to make abnormal or extreme adjustments in its physiology or behaviour in order to cope with adverse aspects of its environment and management”. To provide information on the level of stress of the animals, a combination of multiple animal-based parameters, such as body or eye temperature (Hall et al., 2014; Yarnell et al., 2015), salivary endocrine levels (Erber et al., 2013; Hall et al., 2014; Ille et al., 2016; Schmidt, Hodl, et al., 2010; Yarnell et al., 2015), heart rate (Erber et al., 2013; Ille et al., 2016; Leiner & Fendt, 2011; Rietmann et al., 2004; Schmidt, Hodl, et al., 2010; Strand et al., 2002) or behaviour (Erber et al., 2013; Leiner & Fendt, 2011; Strand et al., 2002; Yarnell et al., 2015), is often used by authors.

In horses, both heart rate (HR) and heart rate variability (HRV) measurements can be affected by factors such as physical activity (Visser et al., 2002), restraint (Vitale et al., 2013), age (Munsters et al., 2012), temperament and reactivity (Visser et al., 2001). Heart rates ranging from 25-60 beats per minute (bpm) have been reported in resting horses (Clayton, 1991), up to a maximum of 212-240 bpm for horses at full work (Evans et al., as cited in König von Borstel et al., 2017). HR and HRV measurements may also reflect other stressors, pain or arousal levels (Dawkins, 2003; Hockenhull & Whay, 2014). These are limitations to the sole use of heart rate and heart rate variability, in addition to the potential impact of the measurement itself.

In horses, associations between the changes in heart rate, heart rate variability indices and behavioural indicators have been reported during novelty (i.e. horses confronted to novel objects or situations) and handling tests. Rietmann et al. (2004) investigated horses' behavioural and physiological responses during a continuous 3-min ground backward walk and found associations between HRV parameters and HR and most of their selected behavioural indicators. Correlations between heart rate and behavioural responses were also reported in a study looking at the effectiveness of three training methods (Christensen et al., 2006). Therefore, in studies investigating horses' stress responses, physiological measurements are often completed with an assessment of behavioural indicators to provide a more robust evaluation.

Behaviour

By providing a sound knowledge of the species-typical repertoire of normal behaviours, the observation of animal behaviour is one of the most informative animal-based indicators of welfare (Mench & Mason, 1997). Normal behaviours of domestic animals, i.e. behaviours normally displayed to attain functional goals (Rousing et al., 2001), are defined by studying wild or free-ranging animal species. Therefore, the welfare of domestic animals is commonly evaluated by comparing with the behaviour and social interactions of wild or feral animals (Veasey et al., 1996). Any variation (increased or decreased frequency or duration) from behaviour observed under natural conditions may therefore provide indications on the welfare of the animal. To

evaluate the stress responses of horses, behaviour can be assessed in situations such as transportation, social isolation, riding, or confinement (Hovey et al., 2021; Kaiser et al., 2006; Kay & Hall, 2009; Mal et al., 1991; Padalino et al., 2012; Reid et al., 2017; Waran & Cuddeford, 1995). Believed to be indicative of stress in horses, the presence and frequency of behaviours such as vocalisation, movements of the head, ears and tail (raised or swishing), pawing, locomotion, defecation and feeding/drinking are commonly reported (Kaiser et al., 2006; Kay & Hall, 2009; Reid et al., 2017; Rietmann et al., 2004). Using a behavioural assessment coupled with physiological measurements, Young et al. (2012) developed a scale of behavioural indicators of stress for the purpose of welfare assessment in stabled domestic horses. This validated scale ranged from 1 (“no stress”) to 10 (“high stress”) and was used to evaluate horses’ stress responses through the observation of specific behaviours.

In horses, the possibility to express a range of behaviours is evaluated through the possibility for social contact between horses (AWIN, 2015), whereas the presence of abnormal behaviours, such as stereotypies, may be an indicator of poor welfare (Broom & Kennedy, 1993). However, it is worth noting that a wild (or feral) environment *per se* does not always provide conditions for optimum welfare, and variations between wild and captive individuals in the types of behaviours performed might reflect adaptation rather than poor welfare (Cooper & Albentosa, 2005). Hence, measurements of the emotional states of horses, covered by the “mental state” domain of the Five Domains Model, are increasingly used, and include fear tests (to determine the fearfulness of the horses), Qualitative Behaviour Assessment (general description of an animal’s behaviour and posture) and Human-Animal Relationship tests (to assess the quality of the relationship) (AWIN, 2015).

Using live animals for educational purposes

The human-horse relationship

A “relationship”, as defined by Hinde (1979), refers to a succession of inter-individual interactions. These interactions can be either positive, neutral or negative (Hinde, 1979) and of

various types (e.g. visual, olfactory). Horse temperament, previous experiences with humans and human's attitude, behaviour and skills at the time of the interaction are factors identified to impact on a horse's reaction to a human interaction (Hausberger et al., 2008).

The quality of the human-horse relationship can be assessed by measuring the behavioural and/or physiological responses of the horse to the presence of humans. These behavioural responses can refer to the horse's acceptance of human contact, the horse's approach towards the person or the horse's avoidance of the person. The parameters measured include for example a score of approachability or allowance of physical contact (Burn et al., 2010; Pierard et al., 2017; Søndergaard & Halekoh, 2003) and a latency to approach (Maros et al., 2010; Seaman et al., 2002; Søndergaard & Halekoh, 2003), and may be evaluated through behavioural tests, also referred to as 'human-horse relationship tests' (Dalla Costa et al., 2015; Hausberger et al., 2012). Reviewed in detail in Hausberger et al. (2008), multiple tests are available to assess horses' reactions to humans, the most frequently used being the 'motionless person test', 'the approach test' or the 'stroking test'.

Several human factors influencing the approachability of horses have been identified, such as the lateralisation and speed of approach. Lateralised reactivity to stimuli was first suggested in a novelty test where more emotional horses spent more time fixating the novel object with their left eye (Larose et al., 2006). The lateralised responses of horses to an approaching human was confirmed in a fear test where horses moved further away when approached from the left side with an open umbrella (Austin & Rogers, 2007). Young and untrained horses also showed more negative reactions (i.e. avoidance, escape, threats) when approached at normal pace from the left side (Sankey et al., 2011). Using a population of semi-feral ponies, speed of approach was investigated by Birke et al. (2011), along with other parameters of approach. The authors reported that a fast approach was associated with increased flight distance and faster flight pace. As indicated by Hausberger et al. (2008), the number of studies that clearly indicate these parameters is low and other parameters of approach, such as gaze direction and posture, are still debated within the literature. While Birke et al. (2011) reported increased flight distance when the person

approaching was looking away from the tested semi-feral ponies, Verrill and McDonnell (2008) found no effect of the gaze direction (either directed on the ponies' face and eyes, or on their body) during approach at a relaxed pace of semi-feral and group-housed ponies. In both studies, there was only one handler approaching the ponies. Seaman et al. (2002) evaluated the impact of maintenance of visual contact during a motionless person test. Although the authors reported no difference in approach times between visual contact or not, it should be noted that the posture and head position of the person was different in each condition (i.e. no visual contact, shoulders rounded and head down vs visual contact, shoulders back and in erect, head up and rigid posture) and may have confounded the results. This thinking is supported by the results of a recent study investigating the effect of a submissive (e.g. slouching, hunched shoulders) vs dominant (e.g. squared shoulders, standing tall) human body posture on the approach preference of horses (Smith et al., 2017). Horses were more likely to approach humans displaying submissive postures, but no difference in latency to approach was reported.

Hama et al. (1996) found lower heart rates in horses when touched by confident people and of positive attitudes towards animals compared to people of negative attitudes towards animals, suggesting that horses are sensitive to people's emotions and attitudes. When walked by hand, horses also seem to react to some of their handler's parameters of approach and attitudes. When they investigated the behavioural reactions of horses to their handler's confidence, Chamove et al. (2002) reported that the mare was more likely to show relaxed and comfortable behaviour indicators when the veterinary students had a positive attitude towards horses, were confident and presented subsequent relaxed parameters of approach. These results are important in the context of veterinary teaching, but more research is required to investigate teaching horses' responses to veterinary students in a routine practical handling class. Chamove et al. (2002) indeed included veterinary students as participants and one horse from the university's teaching herd, but the procedure did not reflect the conduct of a typical practical class.

The student-horse interaction

During a practical class, teaching horses may interact with inexperienced students learning different skills. This inexperience of students is likely to increase the inconsistency and unpredictability of their interaction with the horses. To mitigate the welfare cost to the horses in the veterinary teaching environment, Gronqvist et al. (2016) suggested to achieve an optimal balance between the averseness of the veterinary procedure performed by students, the frequency and the predictability of the student-horse interaction. These metrics are currently lacking, and there are limited data in the literature investigating the responses of teaching horses to procedures performed by veterinary students. This knowledge is crucial to determine which veterinary procedures may be perceived by teaching horses as aversive.

Horses' responses to veterinary procedures

Teaching horses' physiological stress responses in the context of a veterinary student-horse interaction were recently investigated by van Vollenhoven et al. (2017) during a transrectal palpation of the reproductive tract. Until then, most studies investigating horses' responses to transrectal examinations had been conducted with short procedures performed by experienced veterinarians or researchers. Ille et al. (2016) conducted a study on 21 mares transrectally palpated during 185 seconds by experienced veterinarians. The authors reported a slight increase in heart rate, no difference in HRV parameters (i.e. SDRR: standard deviation of the beat-to-beat interval, and RMSSD: square root of the mean squared differences of successive beat-to-beat intervals) and a maximum two-fold increase from baseline in salivary cortisol 15 minutes after the procedure. The combined interpretation of these parameters led the authors to conclude the presence of a stress response from the horses, but they acknowledged that the transrectal palpation was associated with a lower stress response than has been reported for transportation (Schmidt, Biau, et al., 2010; Schmidt, Hodl, et al., 2010), weaning (Erber et al., 2012) or social isolation (Erber et al., 2013). Another study investigated the stress responses of pregnant and/or lactating mares to a transrectal ultrasound examination (Schonbom et al., 2015). In pregnant non-lactating

mares, an increase in the HRV parameter LF/HF ratio (i.e. ratio of low to high frequencies) was reported during the examination, as well as an increase in salivary cortisol 15 minutes after the examination and a two-fold maximum from baseline after 30 minutes. In this group of mares, heart rate and one HRV parameter (i.e. RMSSD) did not differ from baseline values during the examination. In pregnant lactating and non-pregnant lactating mares, similar salivary cortisol levels, heart rates or HRV values (i.e. RMSSD and LF/HF ratio) were reported before and during the examination. The authors concluded that the transrectal ultrasound examination was a “modest temporary stressor” only in pregnant non-lactating mares, and that lactation may induce a decreased response to the procedure. These studies however, derived their heart rate and HRV conclusions only from the difference between baseline (i.e. pasture and/or stocks) values and values during the procedure, without taking into consideration the magnitude of variations observed. In both studies, HR values were not greater than 60 beats per minute (bpm), which remains in the range of 25-60 bpm described for a resting horse (Clayton, 1991). The lack of definition of the terms used to describe the horses’ stress responses to the procedures (“minor”, “moderate”, “major”), and the lack of agreement on what threshold refers to a stress response may lead to potential conflicts in the interpretation of the results of similar studies. Without a concrete agreement and by interchangeably using these terms, it may be difficult to clearly communicate the results.

The management- and procedure-related stress of mares to a gynaecological examination (rectal palpation, transrectal ultrasound examination and vaginal inspection) were investigated by Berghold et al. (2007). These procedures were regularly performed by veterinary students on the selected group of teaching mares as part of students’ training, but for the purpose of this study, only experienced researchers performed the procedures. The cortisol levels of the teaching mares remained stable during the whole study period and were consistently lower than the other groups of mares. Based on these results, the authors concluded that – contrary to the other mares who demonstrated a stress response to the procedure and to the management (e.g. transport, isolation from conspecifics) – the teaching mares were accustomed to the gynaecological examination,

which was not a “major disturbance to the [their] welfare”. However, despite assessing the responses of teaching mares, this study did not include veterinary students. The gynaecological examinations were conducted by experienced researchers, and the horses’ responses to the procedure may therefore not reflect their responses in the context of teaching.

Transrectal palpations are regularly questioned in the veterinary teaching environment due to the danger they represent for both the horses and the students if performed incorrectly (Crossan et al., 2001) and to students’ inexperience (Berghold et al., 2007). Currently, there is only one available study investigating the responses of teaching horses during a standardised 20-minute transrectal palpation performed by veterinary students (van Vollenhoven et al., 2017). The authors observed a significant increase in mean heart rate and variations in HRV parameters (i.e. RMSSD, LF norm, SDNN and mean RR) during the first five minutes of the procedure compared to pasture and/or stock baseline values. Heart rate values then decreased in the last five minutes of the procedure and were only different to that of stock baseline. Heart rate values were the highest at 65 minutes post procedure and were explained by a disruption of the routine of the horses which usually were led out of stocks directly after the procedure. The authors concluded that a stress response was elicited during the transrectal palpation without commenting on the intensity of the variations observed. However, they acknowledged the likely tolerance of the mares to the procedure. The authors also pointed out that a longer restraint than necessary elicited an equal or greater stress response (measured as mean HR and mean RR) than the procedure itself when horses were habituated to the procedure. This study of van Vollenhoven et al. (2017) was a first necessary step in the understanding of teaching horses’ responses to veterinary procedures performed by students. However, the duration and standardisation of the procedure (20 minutes) and its performance by only one student on the same mare do not reflect the routine of a typical practical teaching class where students may rotate between horses and manipulate for various durations. In the context of the veterinary teaching environment where a range of practical teaching classes involve students of various equine experience and confidence with horses,

studies still need to be conducted to evaluate both teaching horses' physiological and behavioural stress responses in a class.

5. CONCLUSION

Horses are widely used for educational purposes in universities offering equine and veterinary science degree programmes. Working with horses presents many risks for both handlers and horses. Using horses during practical teaching classes therefore increases students' risk of horse-related injury and may have welfare implications for the horses. Therefore, there is a need to improve students' skills early in their programme to reduce both the risk of injury for students and the welfare cost to the horses.

Despite this crucial use of horse for teaching, little investigation has been performed so far to describe the teaching-related activities of these horses and evaluate if this use may affect their welfare. This review of the literature also highlighted the current lack of information and metrics available to understand the management, use for teaching and responses of horses during practical teaching classes.

AIMS AND OBJECTIVES OF THE THESIS

This thesis was undertaken with the main aims of (1) describing the use for teaching and management of the horses involved in equine and veterinary science degree programmes, and of (2) assessing the potential impact of this use on the horses' behaviour. This work may optimise the welfare outcomes for the horses, the safety of students during their training, and students' educational outcomes.

Chapter 2 aims to evaluate (1) the level of knowledge students have of equine behaviour and (2) their equine handling competency when entering the veterinary programme. Furthermore, this study aims to evaluate what, if any, type of animal exposure confers a significant advantage to veterinary students' knowledge of equine behaviour and self-assessed equine handling competency.

Chapter 3 aims to describe and quantify the type and frequency of use of the teaching horses over an entire educational year.

Chapter 4 aims to provide information on the time budget and herd dynamics of the teaching horses managed in groups at pasture.

Chapter 5 aims to (1) assess the behavioural responses of the teaching horses to three different persons by studying their acceptance level of human approach and contact over a semester of practical teaching; and (2) identify any changes in these responses in relation to their use for teaching.

Chapter 6 aims to investigate the stress responses of the teaching horses during three types of routine veterinary practical teaching classes (i.e. animal handling, medical rectal examination and mare reproductive examination) by measuring physiological (i.e. heart rate) and behavioural parameters.

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PRELUDE TO CHAPTER 2

As part of their undergraduate equine and veterinary science degree programmes, students are provided with equine practical teaching classes. During these classes, students interact with live horses, whose responses may be influenced by students' attitudes, confidence, experience and competency.

Using a questionnaire, Chapter 2 evaluates 1st- and 4th-year veterinary students' knowledge of equine behaviour and their self-assessed competency of equine handling skills.

This chapter is prepared in the style format of the scientific journal *Animals*.

CHAPTER 2. BENEFITS OF ANIMAL EXPOSURE ON VETERINARY STUDENTS' UNDERSTANDING OF EQUINE BEHAVIOUR AND SELF-ASSESSED EQUINE HANDLING SKILLS

SIMPLE SUMMARY

First-year veterinary students often lack recognition of horse behavioural signals and exposure to animals. Based on self-assessments, we studied their level of knowledge of equine behaviour and their equine handling competency before starting the programme. A previous exposure to horses and/or companion animals on property seemed to confer an advantage in the interpretation of equine behaviour and self-reported equine handling competency.

ABSTRACT

Horses are one of the most dangerous animals veterinarians have to work with. For many veterinary students, first exposure to horses occurs during practical classes. To evaluate the level of knowledge students have of equine behaviour and their equine handling competency when entering the programme, 214 veterinary students were recruited to participate in a questionnaire. Participants were asked to choose one out of 12 terms that best represented the affective state of a horse in a picture, and to self-assess their equine handling skills. Half of first-year students correctly interpreted the horse's behaviour. The majority had (1) a low understanding of equine learning mechanisms and (2) poor self-rated equine handling skills. Fourth-year students were 3 times more likely to accurately interpret the horse's behaviour (95% CI = 1.33 – 7.17, $p = 0.01$) and rated their handling skills higher than first-year students ($p = 0.006$). A history of pet ownership ($p = 0.027$) and the presence of horses on property ($p = 0.001$) were significantly associated with a correct understanding of equine behaviour. These results suggest that previous

animal experience confers a considerable advantage to interpret equine behaviour and highlight the critical importance of practical training in the veterinary programme.

INTRODUCTION

Veterinarians are 9.2 (95% CI 8.12-10.42) times more likely to suffer from a severe occupational accident compared to their colleagues in the medical profession (Nienhaus et al., 2005). Over a 30-year career, the average veterinarian is expected to sustain 7-8 work-related injuries (Jeyaretnam et al., 2010), this number being higher than any other professional civilian occupation, according to the British Equine Veterinary Association (2014). Additionally, 26% of Australian veterinarians affirmed that they had sustained at least one injury in the previous year (Fritschi et al., 2006). The risk of injury is increased when working with large animals, as having a recent injury is more likely for large- and mixed- (small and large) animal veterinarians compared to small animal veterinarians (Epp & Waldner, 2012; Fritschi et al., 2006). For equine veterinarians, 67% of self-reported disease or injuries are related to work, which is higher than those reported (35%) in the European workforce (Loomans et al., 2008).

In studies that have investigated veterinarians' animal-related injuries, experience is believed to be a protective factor (Gabel & Gerberich, 2002). Recent veterinary graduates (with presumably less experience) were 1.8 times more likely to have had an injury in the past 12 months compared to graduates from previous years (Fritschi et al., 2006). Inexperience (39%) is also reported by veterinary students as a factor associated with their equine-related injury (Riley et al., 2015).

Horses are one of the more dangerous domestic species, being the prime or second cause of animal-related fatalities amongst the general population (Langley & Hunter, 2001; Lathrop, 2007), with 50% of equine injuries being unrelated to riding activities (Hawson et al., 2010). In most of reported cases, equine-related injuries were associated with the horse showing a stress or a fear response (Riley et al., 2015; Williams & Ashby, 1992). The horse is a species known for its unpredictability and innate flight response (Thompson et al., 2015), which may make horses

one of the most dangerous animals veterinarians have to work with. As a social prey species, they are extremely sensitive to subtle changes in the body posture of their conspecifics (Goodwin, 1999). The position and tenseness of the ears, tail, mouth, head and feet are major body parts used by horses to communicate, and handlers should have a good knowledge of these body language signals to anticipate the animal's responses (Myers, 2005). An inability to do so and to react accordingly may lead horses to become defensive and more difficult to manage (Baragli et al., 2015). Therefore, a correct understanding of equine behaviour is necessary to avoid miscommunication between the handler and the horse.

In the veterinary environment, a recent study suggested that the recognition of horse behavioural signals was limited for some first year students (Gronqvist et al., 2017), which may result in both higher injury risks and animal welfare issues. Indeed, animal behaviour knowledge is one component of good handling skills. Good handling skills have been defined by Coleman and Hemsworth (2014) as the “general knowledge of the animal's need, a practical experience in the care and an ability to quickly identify changes in behaviour, health or performance as well as addressing those appropriately”. Therefore, veterinarians who show good handling skills can minimise horses' stress and pain, and therefore lower their reactivity levels, resulting in an animal that is easier to examine and treat (Doherty et al., 2017; Hanlon et al., 2007; McGreevy, 2007).

Veterinary students who have grown-up in rural environments used to outnumber those coming from urban areas, but a demographic shift has occurred in the past two decades (Cawdell-Smith et al., 2007). In several countries, the majority of students who enrol in veterinary degrees nowadays come from urban environments (Heath, 2008; Ostovic et al., 2017; White & Chapman, 2007). Students' exposure to a range of domestic animal species is therefore likely to be reduced compared to students from more rural backgrounds, and some students may never have been close to horses or production animals before (Chapman et al., 2007; Old & Spencer, 2011). For this reason, many veterinary students' first exposure to large animals and horses occurs during practical classes.

Investigating first-year veterinary students' level of animal exposure prior to starting their degree would increase the current information available on their background. Although a cluster between equine experience and accurate assessment of equine behaviour has been identified in a previous study (Gronqvist et al., 2017), there are currently no reports on (1) the level of knowledge students have of equine behaviour and (2) their equine handling competency when entering the veterinary programme. The main objectives of the current study were therefore to provide information regarding these two points, and to evaluate what, if any, type of animal exposure confers a significant advantage to veterinary students' knowledge of equine behaviour and self-assessed equine handling competency.

MATERIAL AND METHODS

For this study, 124 first-year students and 90 fourth-year students enrolled in the Bachelor of Veterinary Science of Massey University for the 2018 year were recruited. Participation was voluntary and students were informed that the objective of this study was to investigate their interpretation of equine behaviour. A paper-based questionnaire was used to gather data (Appendix A).

Students were asked to provide demographic and background information (e.g. gender, type of area they spent the most time in growing up, type of large animals kept on their family property), and information regarding their general animal experience (e.g. presence of pets, previous animal-related work experience, confidence around animals) and equine experience. The type of large animals kept on students' family property was a multiple-choice question, whereas the presence of pets was a binary question and students were asked to indicate the species they were referring to.

General equine handling skills were self-rated by students using a 5-point scale (Poor, Sufficient, Good, Very good, Excellent). Using the same scale, they were also asked to assess their ability to perform basic handling tasks required in the first year of the degree (e.g. putting a head collar, leading, grooming, putting a rug, and lifting front and hind feet of a horse).

The last section of the questionnaire focused on students' understanding of equine behaviour and learning theory. Participants were shown a picture of a horse (Appendix A) and asked to circle the one term from a list of 12 that best described the horse's behaviour (Table 2.1). The list of 12 terms was adapted from the Animal Welfare Indicator's qualitative assessment descriptors (AWIN, 2015). The description of each term was provided to avoid misinterpretations. Based on the picture, students were asked to identify which one or more of the horse's body parts they used to make a decision. Following this, there were several questions aimed at determining whether students were familiar with the concepts of learning theory, including positive and negative reinforcement, and positive and negative punishment. Finally, four practical situations (Henderson, 2014) were described and students were asked if these were examples of positive/negative reinforcement or punishment. Each student was therefore given a final score of 0%, 25%, 50%, 75% or 100%, related to the number of correct answers given to these four practical situations.

Table 2.1. Pre-selected terms used in the questionnaire to evaluate students' understanding of the horse behaviour, adapted from the AWIN (2015).

| Term | Definition |
|-------------------------|--|
| Aggressive | Hostile, defensive aggression, intention to harm |
| Alarmed | Worried/tense, apprehensive, nervous, on guard against a possible threat |
| Annoyed* | Irritated, displeased, exasperated, bothered by something, upset, troubled |
| Apathetic | Having or showing little or no emotion, disinterested, indifferent, unresponsive |
| At ease | Calm, carefree, peaceful |
| Curious | Inquisitive, desire to investigate |
| Fearful | Afraid, hesitant, timid, not confident |
| Friendly | Affectionate, kind, not hostile, receptive, confident |
| Happy | Feeling, showing or expressing joy, pleased, playful, satisfied |
| Look for contact | Actively looking for interaction, interested, eager to approach |
| Relaxed | Not tense or rigid, easy-going, tranquil |
| Pushy | Assertive or forceful |

* term evaluated to be the best to describe the horse's behaviour (referred to as "correct term").

The questionnaire was pretested with 19 participants not enrolled in the Massey University veterinary programme and was also peer reviewed by 4 international experts in equine behaviour and/or veterinarians. The derived data were used to determine the correct answers to the learning theory practical situations and the horse's behaviour in picture (Annoyed will be referred later as the "correct term").

Results were entered into MS Excel for further analysis. The questionnaire was completed by (1) first-year veterinary students on their first day of the programme, during an introductory lecture; (2) fourth-year veterinary students at the start of an equine practical class. The project was evaluated by peer review and judged to be low risk by the Massey University Human Ethics Committee (Ethics Notification Number: 4000018923).

STATISTICAL ANALYSIS

An open source sample calculator was used to assess the number of respondents required to obtain a margin of error (beta) no higher than 5%. To be within a 95% confidence interval, at an alpha risk of 5%, 138 respondents were required.

Because students referred to pets as a wide range of species, only the presence of common mammal pet species (dogs, cats, rodents, ferrets, rabbits) was included in the analysis following demographic information and background description. Equids and other large animals were not considered to be pets.

Respondent general demographic information was analysed using pivot tables and pivot charts on Microsoft Excel. Statistical analyses were performed using R 3.5.3 (R studio). Treatment of missing values was performed using the 'VIM' package for graphical visualisation (Templ & Filzmoser, 2008) and the 'BaylorEdPsych' package for Little's test (Beaujean & Beaujean, 2012). Little's test null hypothesis states that missing values are likely to be missing completely at random (MCAR) (Little, 1988). In the case of a failure to reject the null hypothesis, a listwise deletion (complete case analysis) would be performed before further statistical analysis.

Multiple correspondence analysis (package ‘factoextra’) and hierarchical cluster dendrogram (package ‘ClustOfVar’) were performed to visualise the relationships between categorical variables (e.g. background, level of equine experience, presence of large animals on property). A dendrogram shows the hierarchical relationship between objects/variables and when reading such a dendrogram, the smaller the height of the link that joins two variables, the more similar these variables are (Forina et al., 2002). Using a “bootstrap” method, the ‘ClustOfVar’ package also provided the number of clusters for a maximum stability (Chavent et al., 2011).

The threshold used for statistical significance was $p < 0.05$. Associations between categorical variables were looked for using binary logistic regressions. When allowed by the data, multivariable logistic models were built with adequate variables. The package ‘car’ was used to calculate variance inflation factors (VIFs) in order to identify collinearity between categorical variables used as independent variables in the multivariable logistic models (Zuur et al., 2010). Adjusted odds-ratios (AOR) were calculated from the best-fitted model.

RESULTS

GENERAL DEMOGRAPHIC INFORMATION

Of the 214 eligible veterinary students, 115/124 (92%) first-year and 71/90 (79%) fourth-year students completed the questionnaire. The resulting calculated sampling error was 3% at the 95% confidence level. Response rates per question ranged from 80% to 100% (mean 98.5 ± 2.95). Most (75%) of the responses were not missing any information. The null hypothesis (i.e. the missing data is likely to be MCAR) was not rejected after running Little’s test ($p = 0.18$).

BACKGROUND DESCRIPTION

First-year veterinary students were mostly females from an urban environment (Table 2.2). Most respondents grew up with a pet in their household and described themselves as confident or very confident around small animals. Less than half of the students had large animals or horses on their property when growing up. Their confidence around large animals and horses was lower, with respectively only 50% and 45% of students rating it good or above. While most students had

had contact with horses and at least one horse riding experience prior to entering the programme, only 26% considered themselves experienced with horses.

None of the demographics of fourth-year veterinary students differed significantly from those of first-year students. Most were females, from an urban environment, grew up owning at least one pet, were confident with small/large animals and horses, had had a previous contact with horses prior entering the programme, had ever ridden a horse and had a low level of equine experience (Table 2.2).

Table 2.2. Number and percentage of first- and fourth-year veterinary students by gender, background, confidence around animals, and equine experience. P-values were obtained from binary logistic regressions when comparing the distribution of variables between first- and fourth-year students.

| Variables | First-year students Number (%) | Fourth-year students Number (%) | <i>p</i> -Value |
|---|-----------------------------------|------------------------------------|-----------------|
| Gender | | | |
| Male | 21 (18%) | 16 (23%) | 0.497 |
| Female | 93 (82%) | 55 (77%) | |
| Background | | | |
| Rural | 40 (35%) | 22 (31%) | 0.582 |
| Urban | 73 (65%) | 48 (69%) | |
| Grew up with pets (common and/or other*) | 99 (89%) | 65 (93%) | 0.504 |
| Large animals on property | 52 (45%) | 24 (35%) | 0.189 |
| Horses on property | 37 (33%) | 23 (32%) | 0.961 |
| Confidence (good , and above) | | | |
| With small animals | 106 (92%) | 66 (93%) | 0.584 |
| With large animals | 58 (50%) | 45 (63%) | 0.681 |
| With horses | 52 (45%) | 37 (52%) | 0.91 |
| General equine experience (yes) | | | |
| Previous contact with horses | 106 (92%) | 59 (83%) | 0.063 |
| Horse riding experience (yes/no) | 102 (89%) | 66 (93%) | 0.344 |
| Level of experience (good , and above) | 30 (26%) | 23 (32%) | 0.581 |

*Pets (common) = dogs, cats, rabbits, rodents, ferrets. Pets (other) = fish, bird, reptiles, insects.

KNOWLEDGE OF EQUINE BEHAVIOUR

Respectively, first-year and fourth-year students selected nine and six out of the 12 possible terms describing the behaviour of the horse in the picture (Table 2.3). The terms Happy, Look for contact and Pushy were never selected. Annoyed was used by just over half of the first-year students and almost 80% of the fourth-year students to describe the horse's behaviour. A positive term (e.g. At ease, Curious, Friendly, Happy, Look for contact or Relaxed) was selected by 23% of first-year students and 4% of fourth-year students (Table 2.4). To help them interpret the horse's behaviour, most first-year students chose to look at the tail, ears and hind legs (80%, 67% and 65%, respectively). Compared to first-year students, and although the percentage of students selecting each of the body parts varied, a significantly higher number of fourth-year students chose the ears and hind legs to evaluate the horse's behaviour ($p < 0.001$ and $p = 0.012$, respectively).

Table 2.3. Number and percentage of students that selected each of the 12 pre-selected terms to describe the behaviour of the horse in picture.

| Term selected | First-year students Number (%) | Fourth-year students Number (%) |
|----------------------|-----------------------------------|------------------------------------|
| Aggressive (N) | 2 (2%) | 2 (3%) |
| Alarmed (N) | 13 (12%) | 8 (11%) |
| Annoyed* (N) | 56 (52%) | 55 (79%) |
| Apathetic (N) | 3 (3%) | 1 (1%) |
| At ease (P) | 5 (5%) | 0 |
| Curious (P) | 5 (5%) | 0 |
| Fearful (N) | 8 (7%) | 1 (1%) |
| Friendly (P) | 3 (3%) | 0 |
| Happy (P) | 0 | 0 |
| Look for contact (P) | 0 | 0 |
| Pushy (N) | 0 | 0 |
| Relaxed (P) | 13 (12%) | 3 (4%) |

* term evaluated to be the best to describe the horse's behaviour. N = negative term. P = positive term.

Table 2.4. Distribution of first- (n = 115/186) and fourth-year (n = 71/186) veterinary students' responses to the evaluation of the horse's behaviour in picture and knowledge of general equine behaviour principles.

| Variables | First-year students Number (%) | Fourth-year students Number (%) |
|--|-----------------------------------|---------------------------------------|
| Correct evaluation of horse's behaviour | 56 (52%) | 55 (79%) |
| Negative term selected | 86 (77%) | 68 (96%) |
| Positive term selected | 26 (23%) | 3 (4%) |
| Body parts used | | |
| Back | 6 (5%) | 3 (4%) |
| Ears | 76 (67%) | 66 (93%) |
| Eyes | 36 (32%) | 17 (24%) |
| Front legs | 1 (1%) | 1 (1%) |
| Hind legs | 74 (65%) | 59 (83%) |
| Mouth | 6 (5%) | 10 (14%) |
| Neck | 21 (18%) | 18 (25%) |
| Nostrils | 8 (7%) | 9 (13%) |
| Tail | 91 (80%) | 64 (90%) |
| Other | 4 (4%) | 4 (6%) |
| Learning theory principles – perceived knowledge of: | | |
| Learning theory | 39 (35%) | 41 (60%) |
| Positive reinforcement | 97 (86%) | 71 (100%) |
| Negative reinforcement | 90 (80%) | 69 (98%) |
| Positive punishment | 53 (47%) | 67 (94%) |
| Negative punishment | 68 (61%) | 68 (96%) |
| Score \geq 50% (practical examples) | 50 (44%) | 63 (89%) |

Only 35% of first-year students answered positively to the question “Have you heard the term *learning theory* before?” Although most first-year students considered that they understood the principles of positive and negative reinforcement, and negative punishment (86%, 80% and 61%,

respectively), less than half of them obtained a score of 50% or higher for the four practical examples (Table 2.4).

Compared to first-year students, more fourth-year students selected that they were aware of the term learning theory and of the concepts of positive/negative reinforcement/punishment. On a 5-point scale (final score of correct answers), students' results to the practical examples of positive/negative reinforcement/punishment was greater in fourth-year (median (IQR) = 2 (2-3) in first year vs. 4 (3-5) in fourth year; chi-squared test: $p = 4.14 \times 10^{-9}$).

SELF-ASSESSED EQUINE HANDLING SKILLS

Most of the first-year students rated themselves as having poor or sufficient general equine handling skills (Table 2.5), and a considerable number selected that they had never put on a head collar, led, groomed, put on a rug, and lifted front and hind feet of a horse (56/115, 38/115, 51/115, 62/115, 58/115 and 62/115, respectively).

Table 2.5. Number and percentage of first- (n = 115/186) and fourth-year (n = 71/186) veterinary students and their self-assessed level of equine handling skills.

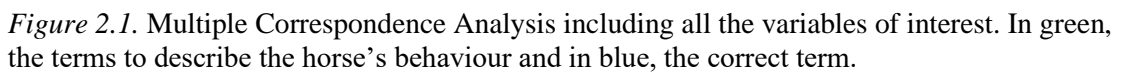
| Variables | First-year students Number (%) | Fourth-year students Number (%) |
|---|-----------------------------------|------------------------------------|
| Self-assessed equine handling skills (good, and above) | | |
| General equine handling skills | 41 (36%) | 46 (65%) |
| Putting a head collar on a horse | 41 (36%) | 60 (85%) |
| Leading a horse | 48 (42%) | 60 (86%) |
| Grooming a horse | 41 (36%) | 51 (73%) |
| Lifting front feet of a horse | 40 (35%) | 55 (77%) |
| Lifting hind feet of a horse | 38 (33%) | 46 (66%) |
| Putting a rug on a horse | 39 (34%) | 50 (71%) |

Sixty-five percent of fourth-year students rated their general equine handling skills good, very good or excellent. On a 5-point scale, students' self-assessment of their equine handling skills

was greater in fourth-year (median (IQR) = 2 (1-4) in first year vs. 3 (2-4) in fourth year; chi-squared test: $p = 0.006$).

INFLUENCE OF ANIMAL EXPOSURE ON EQUINE BEHAVIOUR KNOWLEDGE AND SELF-ASSESSED EQUINE HANDLING SKILLS OF FIRST-YEAR STUDENTS

A multiple correspondence analysis including all the variables of interest showed an average profile in the middle of the plot with two clusters (Figure 2.1). The correct description of the horse's behaviour was clustered with students that were self-rated as very experienced with horses, professional and competitive riders, excellent and very good equine handling skills, a high confidence around horses and small/large animals, and a rural background with horses and large animals on property while growing up (Figure 2.1, shaded green). Shaded in pink on Figure 2.1, a selection of incorrect terms to describe the horse's behaviour was clustered with no equine experience, a beginner horse riding level, an absence of pets, no/poor equine handling skills, and no/low confidence around small/large animals and horses.



The hierarchical cluster dendrogram suggests several close similarities between variables (Figure 2.2):

1. horse riding experience and horse riding level;
2. equine handling skills, level of equine experience, confidence around horses and presence of horses on property;
3. knowledge of learning theory and score obtained from the practical situations;
4. confidence around small and large animals;
5. background and presence of large animals on property.

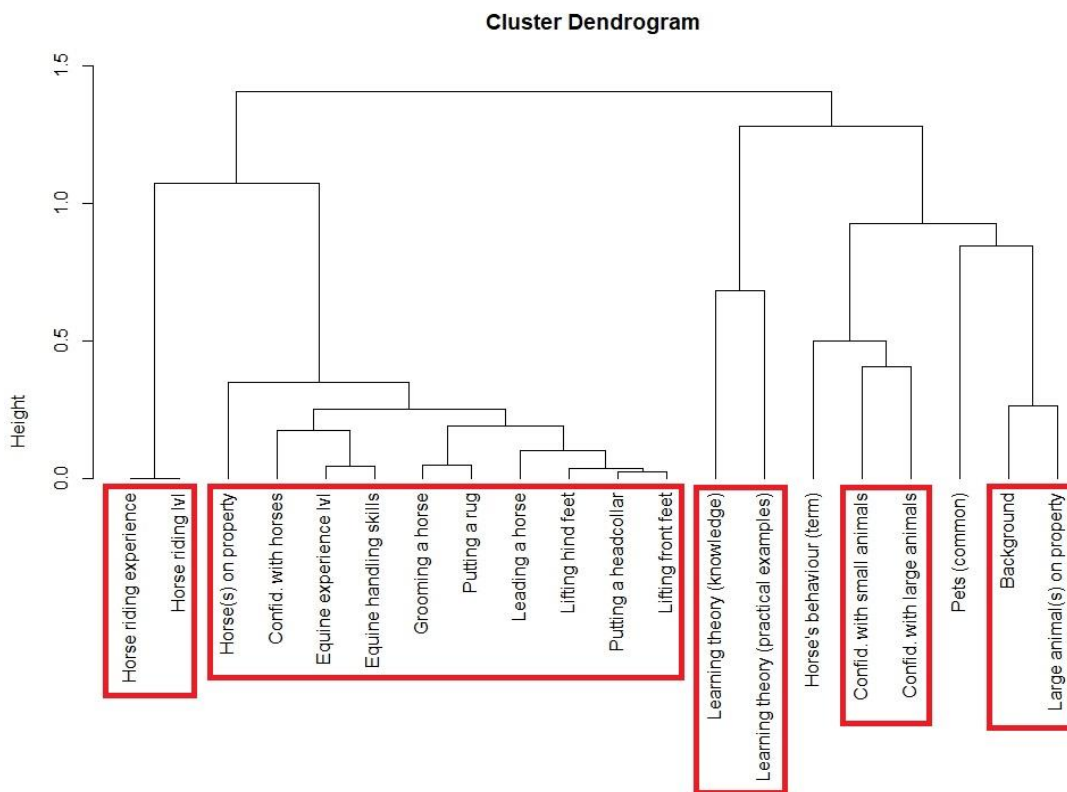


Figure 2.2. Hierarchical cluster dendrogram of the variables of interest used in the Multiple Correspondence Analysis. A maximum of five clusters (in red) was determined with best stability.

Of the students that had horses on their family property when growing up, 81% selected the correct term Annoyed. For students with no horses on their family property, 53% described the horse as Annoyed, 15% as Alarmed, and 11% as Relaxed (Appendix B).

All students that rated their horse riding level as “professional (e.g. instructor, trainer)” selected Annoyed. Students with lower levels of experience selected Annoyed less often: 85% for competitive or show-ring level (e.g. pony club, jumping, eventing, racing, dressage, endurance); 77% for recreational level (e.g. hobby riding) and 56% for trekking level (e.g. organised group rides on hired horses, casual one-off paid experiences). Less than half (44%) of the beginners level (e.g. leading lesson) selected this term.

In the final statistical model including fourth-year students, three variables had a significant effect on the correct interpretation of equine behaviour: (1) the fourth year of the programme, (2) the presence of horses on property, and (3) pet ownership (Table 2.6).

Table 2.6. Best-fitted binomial logistic regression model outcomes and adjusted odds-ratios (AOR), with interpretation of equine behaviour as dependant variable. Before the stepwise regression method, the full model included the following independent variables: presence of pets, confidence around large animals, horse riding level, presence of horses on property, and knowledge of learning theory (score of practical examples).

| | AOR | 95% CI | <i>p</i> -Value |
|---|------|--------------|-----------------|
| Interpretation of equine behaviour (Incorrect/Correct) – AIC = 152.4 | | | |
| Year (First/Fourth) | 3.02 | 1.33 – 7.17 | 0.01 |
| Presence of pets (No/Yes) | 4.89 | 1.3 – 23.78 | 0.027 |
| Horses on property (No/Yes) | 4.46 | 1.86 – 11.62 | 0.001 |

DISCUSSION

Other than basic demographics (gender and age), very few studies have described in detail the background of veterinary students. The present study focused on gaining a better understanding of students’ knowledge of equine behaviour and self-assessed equine handling skills on entry to the undergraduate veterinary programme. It is the first study to evaluate the influence, if any, of prior animal exposure on these two variables.

The overall response rate of 87% and the response rate per question suggest that the results may be representative of the whole group of veterinary students. The data of the present study agree with previous studies that report most students enrolling in undergraduate veterinary programs are females, come from an urban environment and have owned at least one pet (Chapman et al., 2007; Gronqvist et al., 2017; Klupiec et al., 2014; Wake et al., 2006).

Students enter the programme with various levels of knowledge of, confidence of and experience with different animal species. Regarding horses, and similarly to Riley et al. (2015), almost all students had been in contact with and ridden a horse before enrolment. However, the level of horse riding ranged from beginners to professionals and the level of equine experience from no experience to very experienced students. Moreover, the limited number of students who showed a high confidence around horses suggests that a one-time horse experience might not be enough to overcome all fears when interacting with this species. The intercorrelation between confidence and time spent with horses revealed by Chamove et al. (2002) supports this thinking. Therefore, significant experience may be needed to provide veterinary students with the high confidence with large animals and horses found to be associated with an accurate interpretation of horse behaviour and high self-assessed equine handling skills in this study.

In the present study, the horse's ears, hind legs and tail were students' most chosen body parts used to try assessing the horse's behaviour, regardless of a correct or incorrect evaluation. In addition, a significant positive association was found between the selection of the ears, hind legs and tail in the decision-making and the accurate horse behaviour assessment. These are widely used parameters while assessing equine behaviour (Goodwin, 2007; Hall et al., 2014). However, only half of the first-year students correctly interpreted the behaviour of the horse. A wide range of behaviours were selected and 23% of students failed to recognise that the horse was in a negative affective state. Gronqvist et al. (2017) found similar results with students choosing contradictory terms to evaluate equine behaviour and being unable to assess affective states. Overall, these results suggest that most of the students had a good understanding of which of the horse's body parts they should focus on, but many lack competency in the way they should be

interpreting these body signals. This suggests that prior to starting practical classes, teaching should focus on improving student's ability to interpret the relevant body parts to identify the horses' affective states and react accordingly with adequate cues and signals.

Indeed, the correct and well-timed use of cues and signals when interacting with a horse increases calmness and effectiveness of handling (Doherty et al., 2017). On entry to the veterinary programme, a larger proportion of veterinary students assessed themselves to have a good knowledge of the principles that constitute the way horses learn, although little more than a third (35%) had come across the term learning theory before. However, their actual understanding, as measured in the questionnaire, was poor, with only 44% obtaining a score higher or equivalent to 50% when given practical examples. This falls in line with a frequent confusion between terms (Michael, 1975), mainly due to the use of the words “negative” and “punishment” (McLean, 2005). Although horse training mainly focuses on negative reinforcement (Visser & Van Wijk-Jansen, 2012), even equestrian coaches are not always able to accurately articulate the learning theory terms and definitions (Warren-Smith & McGreevy, 2015). Even though the information of this present study was obtained from a questionnaire and was not based on real-life situations, it gives a general idea of the students' level of equine behaviour knowledge before entering the veterinary programme.

First-year students' knowledge of equine behaviour has to be highlighted as many veterinary schools do not include a criterion based on handling skills for selection for entry to the programme. Unsurprisingly, first-year students' own-assessment of their competency in handling horses ranged from low (33%) to high (13%). The hierarchical cluster dendrogram suggested a close similarity between equine handling skills and level of equine experience. Although a majority of students had had at least one riding experience, the multiple correspondence analysis (MCA) also showed one cluster between a good level of horse riding and good handling skills. Similarly, there was a cluster between low level of horse riding and poor handling skills. The results provided by the MCA give a better insight of the variables clustered with an accurate interpretation of horse behaviour and good self-assessed handling competency and start to suggest

a link with previous animal experience. Similarly to the study of Gronqvist et al. (2017), students' level of experience with horses was clustered with interpretation of the horse's behaviour.

First- and fourth-year students shared a similar demographic and background profile, which suggests a group comparison was appropriate. Towards the end of their degree, veterinary students showed a significantly greater competency of self-assessed equine handling. This result was expected as the questions were based on techniques that students are taught during their first year of the Massey University undergraduate veterinary programme. Therefore, this may be the result of experience gained in the programme. Although large animal handling practical classes can sometimes suffer from economical and/or resources pressures (Klupiec et al., 2014; McGreevy et al., 2007), these results suggest the crucial importance of these classes in the veterinary undergraduate degrees. However, students in both years did not differ in confidence around horses and even in fourth year, confidence around small animals was the highest. This is consistent with a previous study (White & Chapman, 2007) and may be due both to the unpredictability of horses and to insufficient (in quality and/or duration) external equine placements. Underlying this result could also be an overconfidence of first-year students, whereas fourth-year students who have a greater understanding of the programme's expectations in regards to their capability, could underestimate their confidence. This thinking is supported by Goldfinch and Hughes (2007) who showed that most students entered university with a high confidence level in their skills, and revealed that previous work experience was not associated with greater confidence in first-year students' skills.

The statistical model revealed a significant effect of the year of study of the students. This suggests that the knowledge veterinary students acquired during their degree is highly valuable for a better interpretation of equine behaviour. The presence of horses on property was also significant and positively associated with accurate interpretation of equine behaviour. This could have been expected as most of these students were also involved in the care and management of the horses on their property. Similarly, there was a positive association of pet ownership with an accurate interpretation of horse behaviour. While this particular result needs to be taken with

caution, as most students had owned a common pet species (162/181), the necessary interactions with an animal in their household potentially prepare students for interpreting horse behaviour. This may indicate that an early exposure to common companion animal species allows students to be more attentive to horses' subtle body and posture changes and interpret more accurately the animal's behaviour.

Being a comparative and not a paired study is one limitation of the present work. The analysis of students' competency in handling horses using self-assessed questions may include a bias in the form of socially desirable responding (SDR). SDR is indeed common in studies using questionnaires and investigating people's competency, health or behaviours (Lajunen & Summala, 2003; van de Mortel, 2008). In order to limit this effect, future studies may want to independently assess students' animal handling skills. Integrating data on equine-related students' injuries prior to and during the veterinary programme would also bring more information in regards to students' equine experience and handling skills.

CONCLUSION

Although most students had been in contact with horses before, the majority entered the veterinary programme with a low (1) level of confidence around horses and (2) self-assessed equine handling skills competency. Their basic understanding of equine behaviour was also low and only half of the students correctly interpreted the horse's behaviour. In order to keep students safe during equine practicals, these results highlight the necessity of teaching basic equine behavioural concepts and observational skills beforehand.

Although their knowledge of equine behaviour and their self-assessed competency in handling equines increased significantly between the first- and fourth year of the programme, a previous exposure to horses and/or companion animals on property seem to confer students a considerable advantage.

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PRELUDE TO CHAPTER 3

Chapter 2 reported a low understanding of equine behaviour concepts, a limited ability to interpret a horse's behaviour and poor self-assessed equine handling skills for 1st-year veterinary students. The same investigation on 4th-year students highlighted their significant increased competency and knowledge compared to students on entry. Two background factors (i.e. history of pet ownership and presence of horses on the students' family property while growing up) were also identified as beneficial to correctly interpret a horse's behaviour.

Because horses are used for students to practice and refine their handling and clinical skills, there is a need to describe the teaching activities the horses are involved in. Chapter 3 is a retrospective and observational study which provides an analysis of the type, frequency and regularity of use for teaching of the horses over a calendar year.

This chapter is prepared in the style format of the scientific journal *Animals*.

CHAPTER 3. HORSES USED FOR EDUCATIONAL PURPOSES IN NEW ZEALAND: A DESCRIPTIVE ANALYSIS OF THEIR USE FOR TEACHING

SIMPLE SUMMARY

Many equine and veterinary science degree programmes use horses during practical teaching classes. The use of horses during teaching was studied over a calendar year. The teaching horses were characterised as older non-reactive mares and geldings that had been used for teaching for a number of years after retirement from (harness) racing or sport. These horses were grouped into and managed as different herds based on suitability for specific practical teaching classes, the frequency of teaching activities per horse was relatively low (1-2 sessions per week). Two broad types of practical classes were identified which were characterised by the restraint method used (yards vs. stocks), duration of the class and number of students per horse. The classes included rectal examinations (in stocks, shorter duration, few students) and general animal handling and husbandry, which included animal handling, lameness evaluation, clinical examination and foot trimming (in yards, longer duration, more students). Although the workload from teaching within this cohort of horses was low, more work to determine additional markers of teaching horses' welfare may be required.

ABSTRACT

Horses are used in practical teaching classes in many equine and veterinary science degree programmes to develop and refine the handling and clinical skills of students. In this study, the activities of 24 teaching horses grouped in three herds were investigated over an entire calendar year. Although also used for research and general husbandry, teaching-related activities were the predominant use of the horses. Herd B was used for a greater number of teaching sessions (median = 28, IQR = 27-29.5 per year) than herds M (median = 21, IQR = 20-21 per year) and T (median = 19.5, IQR = 13.75-25.5 per year), which translates to a relatively low workload (one or two

weekly sessions during the teaching semester). Sedation was used in dentistry classes (in alignment with national best practice standards) but rarely required for other teaching activities. Mare reproductive rectal- and medical rectal examination practical classes (specific to 5th-year veterinary teaching and characterised by more restraint (in stocks)) were significantly shorter and had fewer students per horse than the other practical classes. Although the low workload reported suggests an opportunity to increase students' exposure to horses without compromising the horses' welfare, further investigation to determine specific stressors to the horses in the teaching environment may be required.

INTRODUCTION

Animal handling is a critical skill for veterinarians to master. Handling horses well is likely to minimise their stress and pain, lower their reactivity, and reduce the risk of injury to both horse and handler (Doherty et al., 2017; Hanlon et al., 2007; Hawson et al., 2010; Jaegglin et al., 2005; McGreevy, 2007; Riley et al., 2015). Good animal handling skills depend not only on the knowledge of the animal's behaviour and ethological needs, but on adequate and well-timed reactions to the animal's behaviour changes that require experience and practice to develop (Coleman & Hemsworth, 2014). In conventional horse riding, studies have started to evaluate the amount of experience required to substantially decrease the number of injuries (i.e. approximately 100 hours of riding experience) (Mayberry et al., 2007), but these metrics are currently lacking to assess how much equine exposure is required to develop competency in horse handling. While veterinarians' handling competency is often judged anecdotally by the clients through the horse's behaviour and easiness to handle (Doherty et al., 2017; McGreevy, 2007), there are limited data in the literature quantifying this skill.

Hands-on experiences with animals are therefore emphasised by most universities in their equine and veterinary science degree programmes (McGreevy, 2007) to complement theoretical teaching. The importance of practical classes in the curriculum has grown, as most students that now enter animal-based programmes come from an urban background with limited animal

experience (Guinnefollau et al., 2019; Ostovic et al., 2017; Wake et al., 2006; White & Chapman, 2007). Equine practical teaching classes may well be the first occasion for some students to interact with a horse (Chapman et al., 2007; Old & Spencer, 2011). Successful and positive experiences (e.g. “getting it right”, “being good at it”) in the early stages of their riding careers have been reported to be major drivers in the development of elite equestrian riders (Lamperd et al., 2016). This suggests that allowing students to become confident and competent veterinarians may be influenced by the type of early experiences they have with horses. To enhance students’ training, it would be useful to identify which type of activities should be provided to veterinary students and which are the most suitable horses for this purpose.

In the teaching environment, horses are used for a range of practical classes (e.g. animal handling, lameness investigation, rectal examination, dental examination and treatment) (Austin et al., 2007; Cawdell-Smith et al., 2007; Chapman et al., 2007; Cockram et al., 2007; Hanlon et al., 2007; MacLeay, 2007; McGreevy et al., 2007; Stafford & Erceg, 2007; White & Chapman, 2007) in which students show various levels of equine experience and confidence with horses (Gronqvist et al., 2017; Guinnefollau et al., 2019). Horses are selected for each type of practical teaching class based on their behavioural characteristics and their apparent ability to cope with their use (Austin et al., 2007; Cockram et al., 2007; Stafford & Erceg, 2007; White & Chapman, 2007). Therefore, the type and number of interactions with equine and veterinary science students are likely to vary between animals. These student-horse interactions themselves might also be inconsistent because of variation in the equine behaviour knowledge and equine experience of students (Gronqvist et al., 2017; Guinnefollau et al., 2019). A failure of students to correctly identify horses’ behavioural cues and to give an appropriate response at an appropriate time may lead to confusion for the horse (McCall, 1990; McLean & Christensen, 2017; Overmier & Wielkiewicz, 1983). As a result, horses may develop conflict behaviours that may negatively influence their welfare (McLean & Christensen, 2017; McLean & McGreevy, 2010).

Despite a widespread use of horses for teaching purposes within universities, very little information is available describing the nature and frequency of these activities within the equine

and veterinary science curriculum. The aim of the present study was therefore to describe and quantify the type and frequency of use of teaching horses over an entire educational year in a veterinary school in New Zealand.

MATERIAL AND METHODS

The study population included twenty-four horses, used for undergraduate equine and veterinary science practical teaching classes, at Massey University, New Zealand. The horses included 3 Thoroughbreds, 19 Standardbreds, 1 Stationbred (crossbred) and 1 Kaimanawa (New Zealand feral horse). Ten horses were geldings and 14 were mares, with a mean age of 15 ± 4 years (one horse's age was not officially recorded but thought to be over 15 years). The horses were pasture-based throughout the year and for ease of management were kept in three stable herds of 7 (herd B – mares and geldings), 8 (herd T – mares and geldings) and 9 (herd M – mares only) individuals. The composition of the herd was based on the temperament and perceived tolerance of the horses to the different teaching procedures. Quiet horses tolerant of naïve students comprised one herd (herd T) and were used for 1st-year practical handling classes. A second herd (herd B) was used for medical rectal examination classes and a third herd (herd M) was used for mare reproductive rectal examinations. Pasture was the primary feed source and in the winter months hay was also provided once daily at 2 (1-4) kg of dry matter/horse/day (median, interquartile range). Water was provided *ad libitum* from water troughs in the paddocks. The mean paddock size was of 1.9 ± 0.7 ha and herds were rotated between paddocks based on availability.

The use of the teaching horses was studied over the 2018 calendar year (from 1st January to 31st December). At Massey University in the southern hemisphere, most teaching is concentrated into two semesters which follow the calendar year, but some 5th-year veterinary teaching continues outside the normal semester system. Teaching semesters were 12 weeks long separated by a 2-week mid-semester break. In 2018, semester 1 lectures were given from February 26th (week number 9) to June 1st (week number 22), with a mid-semester break from March 30th to April 13th (week numbers 14-15). Semester 2 lectures were given from July 16th (week number

29) to October 19th (week number 42), with a mid-semester break from August 27th to September 7th (week numbers 35-36).

This study retrospectively analysed data collected and stored electronically within a MS Excel spreadsheet and the weight scale proprietary database (true-test.com) whenever the teaching horses were brought into the teaching facility. The data recorded included the horses' name and weight, herd of animal, date, length and type of event, name of main teaching staff involved, course and animal ethics committee protocol numbers, number of students in practical teaching class, use and type of medication (if administered), and additional comments during use.

Data were extracted and categorised. The date of the event was categorised by semester, while 'calendar year' referred to both semesters 1 and 2 and semester breaks. The type of use of the horse during an event was categorised as 'teaching' (i.e. horses used for practical teaching classes with students), 'general husbandry' (i.e. general husbandry care for the horses provided by staff members or veterinarians), 'research' (i.e. horses used for a research project – no change to management) or 'other' (horses used for neither of the above procedures, such as blood harvesting). Each teaching event was categorised based on the type of practical teaching class (i.e. 'animal handling', 'clinical examination', 'foot trimming', 'lameness evaluation', 'dental training', 'medical rectal examination' and 'mare reproductive rectal examination'). Within an event, the use of each horse was identified as a 'horse session' (i.e. one session = one horse). The number of horse sessions varied between events. In teaching events, some classes required the use of the whole herd (e.g. medical rectal- and mare reproductive rectal examinations) and others only a selected portion of the herd (e.g. a maximum of four horses per animal handling practical class). The duration of each individual horse session was quantified as a 'horse hour' (e.g. 30 minutes = 0.5 horse hour).

If the number of students working with each horse was not recorded, then the number of students per horse was estimated by dividing the number of students in the practical teaching class by the number of horses used for this class. If medication was used, it was classified into three categories: 'sedation', 'anti-inflammatory' and 'antibiotic'.

When used for teaching, the horses were individually kept either in stocks or in teaching yards. Information on the conduct of practical teaching classes, including the location of the horse during the class, as well as the type of restraint used were obtained from class supervisors or technical staff members. Data were categorised according to the horse location during the practical teaching class in relation to conspecifics, the method of restraint, the year of students involved in the class, number of students per horse, and the consistency of routine of the practical teaching class (Table 3.1).

Table 3.1. Variables chosen to describe the practical teaching classes.

| Variable | Categories |
|---|---|
| Location of the horse | Regular (i.e. same neighbour and location each time) or variable (i.e. different neighbour and/or location) |
| Year of students | 1 st , 2 nd , 3 rd , 4 th or 5 th year |
| Number of students per horse | |
| Length of horse session | |
| Method of restraint | High (i.e. horse in stocks with restriction of movements) or low (i.e. horse in a yard and held by a handler) |
| Routine of the practical teaching class | Regular or variable |

STATISTICAL ANALYSIS

Horse use data (name, herd, date, type of use, type and length of practical teaching class, use of medication, course number and number of students) were examined using pivot tables to generate frequency counts and percentages. The percentage of horse sessions involving the use of sedation for teaching purposes was calculated.

Differences in the use between herds were tested because some practical teaching classes were herd specific. Associations between the herd and the number of horse sessions in each category (i.e. teaching, general husbandry, research, other) were investigated using chi-squared tests. The distribution of the number of horse sessions was nonparametric, so differences between herds were tested using a Kruskal-Wallis analysis of variance. When required, multiple

comparisons between herds were then tested for using a Dunn test (package *FSA*). Between types of practical teaching classes, a Kruskal-Wallis nonparametric analysis of variance (and Dunn test if necessary) was also performed to compare the length of horse sessions, and the number of students per horse. Statistical analyses were performed using R 3.5.3 (R Studio). The threshold used for statistical significance was $p < 0.05$.

The particularities of the practical teaching classes were then investigated using the variables available in Table 3.1.

RESULTS

GENERAL ACTIVITY

From January to December 2018, the horses were used for a total number of 2,091.5 horse hours. This included 1,276.5 horse hours (534/1208 horse sessions – 44.2%) of teaching, 250 horse hours (272/1208 horse sessions – 22.5%) of general husbandry, 515 horse hours (337/1208 horse sessions – 27.9%) of research (i.e. two behavioural and one physiological research projects) and 50 horse hours (65/1208 horse sessions – 5.4%) of other. Teaching represented a total of 479.5 horse hours (herd B), 423.5 horse hours (herd M) and 373.5 horse hours (herd T).

Except for an involvement of herd T in research (7 horse sessions), the distribution of activities between herds in semester 1 did not differ. However, there was a difference in semester 2 between herds (Table 3.2). The proportion of teaching was different across the three herds, with a greater proportion of use for research with herd M (43.4% of horse sessions) than herd B (21.3% of horse sessions). The distributions of general husbandry activities and other were similar between herds.

Table 3.2. Number and percentage of horse sessions (i.e. teaching, general husbandry, research and other) for each of the three herds during the 2018 calendar year, and the 1st and 2nd teaching semesters. P-values were obtained from chi-squared tests when comparing the distribution of variables between herds B, M and T.

| | Herd B (n = 7 horses) | Herd M (n = 9 horses) | Herd T (n = 8 horses) | p-Value |
|----------------------|----------------------------------|----------------------------------|----------------------------------|----------------|
| Calendar year | | | | |
| Teaching | 199 (52.7%) | 188 (41.1%) | 147 (39.4%) | 0.0003 |
| General husbandry | 77 (20.4%) | 93 (20.4%) | 102 (27.4%) | 0.027 |
| Research | 83 (22%) | 147 (32.2%) | 107 (28.7%) | 0.004 |
| Other | 19 (5%) | 29 (6.4%) | 17 (4.6%) | 0.49 |
| Semester 1 | | | | |
| Teaching | 62 (63.9%) | 61 (64.9%) | 50 (53.2%) | 0.19 |
| General husbandry | 35 (36.1%) | 30 (31.9%) | 36 (38.3%) | 0.65 |
| Research | 0 (0%) | 0 (0%) | 7 (7.5%) | 0.0007 |
| Other | 0 (0%) | 3 (3.2%) | 1 (1%) | 0.16 |
| Semester 2 | | | | |
| Teaching | 99 (61.9%) | 76 (41.8%) | 85 (50.9%) | 0.001 |
| General husbandry | 18 (11.3%) | 17 (9.3%) | 27 (16.2%) | 0.14 |
| Research | 34 (21.3%) | 79 (43.4%) | 48 (28.7%) | < 0.001 |
| Other | 9 (5.6%) | 10 (5.5%) | 7 (4.2%) | 0.81 |

TEACHING ACTIVITY

For each herd, Figure 3.1 shows the temporal use over the calendar year as of number of practical teaching classes per week. The majority of the use of horses for teaching occurred during either teaching semester one or two. Seventeen practical teaching classes were scheduled outside of the teaching semesters. Fourteen of these classes referred to 5th-year veterinary teaching and three classes were from courses for which start and finish dates differ from the normal University semester dates (Massey University website).

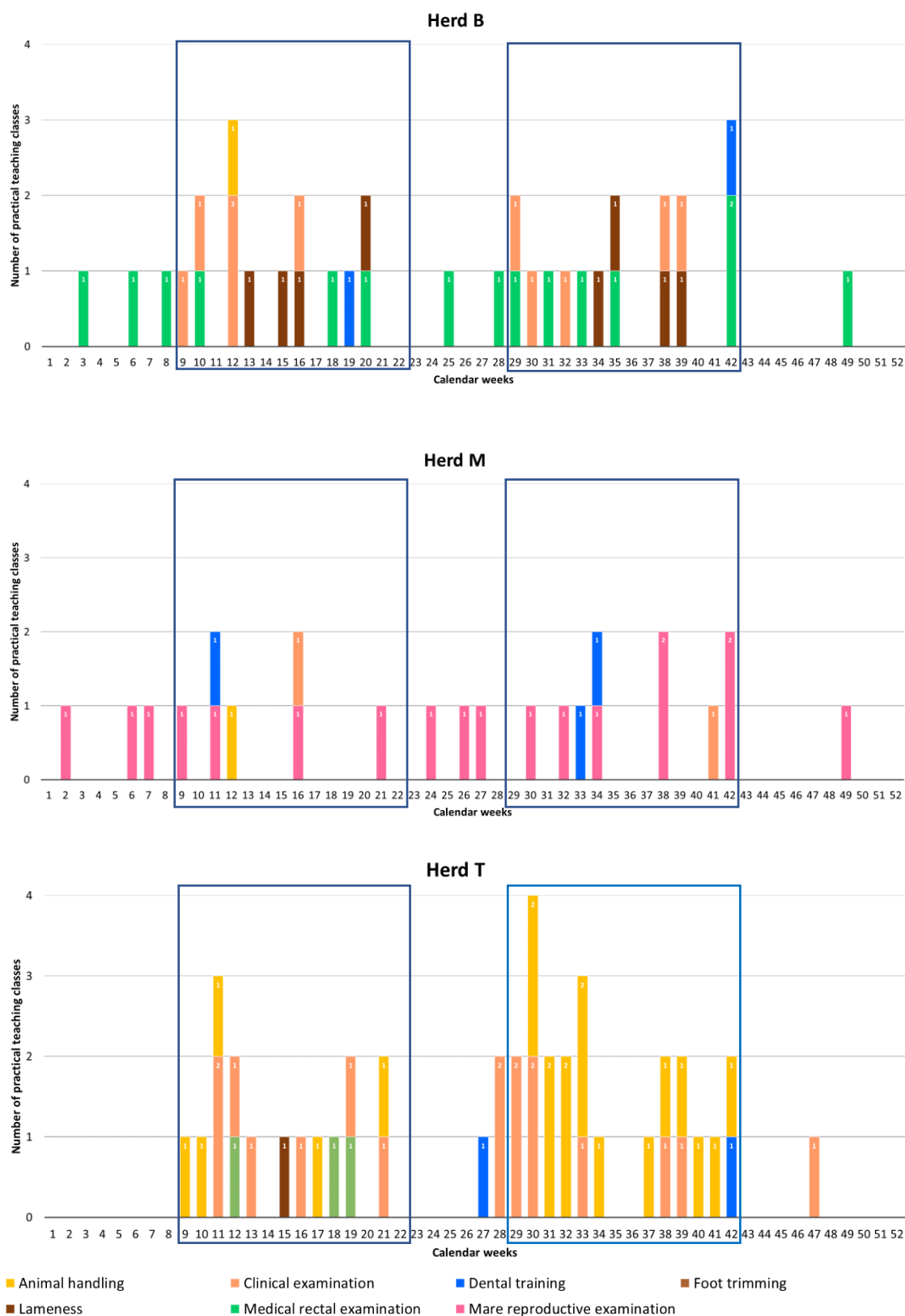


Figure 3.1. Number and type of practical teaching classes per week over the calendar year, i.e. any class a minimum of one horse was used for teaching per week, for each herd (B, M and T). The blue boxes refer to the teaching semesters one and two, i.e. to weeks 9 to 22, and to weeks 29 to 42, respectively.

Over the calendar year, there were more horse sessions for herd B (median = 28, IQR = 27-29.5) than for both herds M and T (median = 21, IQR = 20-21, $z = 3.17$, $p = 0.005$; and median = 19.5, IQR = 13.75-25.5, $z = 2.8$, $p = 0.01$, respectively). Although there was some variation in the number of horse sessions, all horses of herd M were used for the same four types of practical teaching classes (Figure 3.2). In herd B, three horses did not take part in dental training classes, and only two horses were used for animal handling classes. The number of horse sessions was the most variable within herd T, with two horses rarely used. In addition to being used less frequently, these two horses were never involved in foot trimming, dental training and lameness practical teaching classes (Figure 3.2).

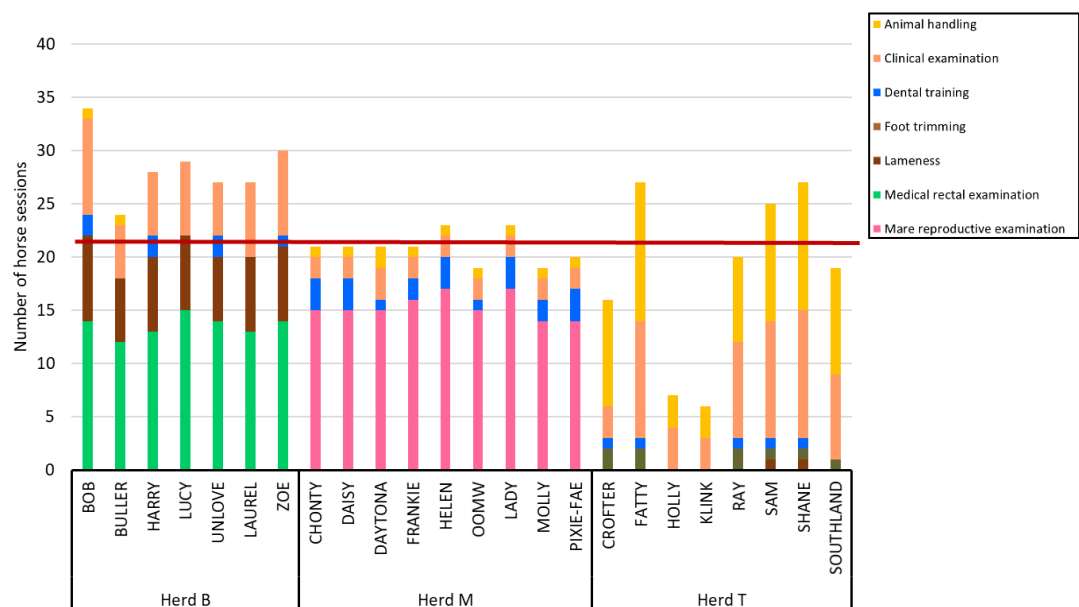


Figure 3.2. Individual number of horse sessions stratified by herd over the calendar year. The red line represents the median (= 22) number of horse sessions of the three herds.

Horses were rarely sedated for teaching purposes (7%, 35/534 horse sessions). Sedation was always used in dental practical classes (33/33 horse sessions), rarely for clinical (1/95 horse sessions) and medical rectal examinations (1/127 horse sessions), and never recorded for the following practical teaching classes: animal handling, foot trimming, lameness evaluation and mare reproductive rectal examination.

In mare reproductive rectal examination and medical rectal examination practical classes, the length of the horse sessions (Kruskal-Wallis test: $\chi^2(6) = 263.6$, $p < 0.001$) and the number of students per horse (Kruskal-Wallis test: $\chi^2(6) = 217.0$, $p < 0.001$) were significantly lower than in animal handling, clinical examination, lameness evaluation, foot trimming and dental practical classes (Table 3.3).

Table 3.3. Characteristics of each type of practical teaching class during the 2018 calendar year. Length of horse session and number of students per horse are presented with median and interquartile range (IQR).

| | Location of the horse | Year of students | Number of students per horse | Method of restraint | Routine of the practical teaching class | Length of horse session (hours) |
|--------------------------------------|-----------------------|---|------------------------------|---------------------|---|---------------------------------|
| Animal handling | Variable | 1 st , 2 nd , 3 rd , 4 th | 3 (2-4) | Low | Variable | 2.5 (2.5-3) |
| Clinical examination | Variable | 1 st , 2 nd , 3 rd , 4 th , 5 th | 5 (4-6) | Low | Regular | 3 (3-3) |
| Dental training | Variable | 3 rd , 5 th | 2 (2-8) | High | Regular | 3 (2.5-4) |
| Foot trimming | Variable | 2 nd , 4 th , 5 th | 4 (2-3) | Low | Regular | 3 (2-3) |
| Lameness evaluation | Variable | 2 nd , 3 rd , 4 th | 4 (3-4) | Low | Regular | 3 (3-3) |
| Medical rectal examination | Variable | 5 th | 1 (1-1) | High | Regular | 2 (1.5-2) |
| Mare reproductive rectal examination | Regular | 5 th | 1 (1-1) | High | Regular | 2 (2-2) |

MAIN PRACTICAL TEACHING CLASSES

Over the calendar year, the four main practical teaching classes in terms of volume (i.e. number of horse sessions and number of horse hours) were mare reproductive rectal examination, clinical examination, medical rectal examination and animal handling (Table 3.4). The rest of the teaching volume was divided between dental training, foot trimming and lameness evaluation.

Table 3.4. Number of horse sessions and horse hours for each type of practical teaching class involving the use of horses during the 2018 calendar year and teaching semesters one and two.

| | Calendar year | | Semester 1 | | Semester 2 | |
|--------------------------------------|-------------------|---------------|-------------------|-------------|-------------------|-------------|
| | Horse sessions | Horse hours | Horse sessions | Horse hours | Horse sessions | Horse hours |
| Animal handling | 82 (15.4%) | 192 | 31 (17.9%) | 63 | 51 (19.6%) | 129 |
| Clinical examination | 127 (23.8%) | 358.5 | 45 (26%) | 100 | 74 (28.5%) | 240.5 |
| Dental training | 33 (6.2%) | 102 | 10 (5.8%) | 22 | 19 (7.3%) | 70 |
| Foot trimming | 9 (1.7%) | 25 | 9 (5.2%) | 25 | 0 (0%) | 0 |
| Lameness evaluation | 50 (9.4%) | 155.5 | 23 (13.3%) | 68.5 | 27 (10.4%) | 87 |
| Medical rectal examination | 95 (17.8%) | 176 | 19 (11%) | 50 | 38 (14.6%) | 62 |
| Mare reproductive rectal examination | 138 (25.8%) | 267.5 | 36 (20.8%) | 67.5 | 51 (19.6%) | 94.5 |
| TOTAL | 534 (100%) | 1276.5 | 173 (100%) | 396 | 260 (100%) | 683 |

The use of the three herds was different between types of practical teaching classes. Herd B, herd M and herd T were used, respectively, mainly for the following practical teaching classes: medical rectal examination (46.8%), mare reproductive rectal examination (73.4%) and animal handling (47.6%).

DISCUSSION

Despite a wide use of horses for educational purposes in most equine and veterinary science degree programmes around the world, there is a current lack of empirical information on the animals' specific use. To the authors' knowledge, this is the first study to quantify the use of horses for educational purposes in the teaching environment. Using a retrospective method over an entire calendar year, the present work documents in detail the management and teaching-related activities of a cohort of teaching horses.

Practical teaching classes are faced with many constraints (e.g. staff, budget, scheduling) in a tight curriculum (Cawdell-Smith et al., 2007). The use of the horses is therefore dictated to a large extent by the programme and the challenge of providing sufficient time for students to practice their skills. Despite a more frequent use during the second teaching semester, the number of weekly teaching-related activities and hours use with these horses was low compared to the frequency and duration of activities (such as riding and training) in other horse populations and equestrian activities. Competition horses (i.e. dressage, show jumping, eventing) are trained for three to six 45-50 minute sessions per week of varying intensity (Bolwell et al., 2015; Lonnell et al., 2014; Rogers et al., 2007; Verhaar et al., 2014; Walters et al., 2008). Similarly, studies reported approximatively one to three hours of work per day and at least one rest day per week for riding school horses (Lonnell et al., 2012; Munsters et al., 2013; Sloet van Oldruitenborgh-Oosterbaan et al., 2006). In these populations, a description of the workload is often used for management purposes and to evaluate the amount of exercise-related physiological stress experienced by the animals. Authors usually report information on the intensity of the session (e.g. speed, duration, heart rate and other physiological measures) in addition to the frequency (Rogers et al., 2007). In this context, practical classes could be considered to provide limited physical challenge to the horses. However, the parameters used to describe the intensity of the workload of ridden horses are not applicable to teaching horses whose movements are generally restrained during practical classes while students practice and improve their handling and clinical skills. Instead, in this population of horses, the frequency and regularity of teaching use may be suitable parameters to assess the workload of teaching horses (Gronqvist et al., 2016).

The teaching horses and the herd composition had had very few changes in the last 10 years and the age of the teaching horses was heavily right skewed with many horses in their teens and a few in early twenties. Given the long-term tenure of the horses, the stability of the herds and the low frequency of use, the authors hypothesise that this population of teaching horses may experience limited physiological or behavioural stress. For these cohorts of horses, given the nature of the teaching classes and the frequency of horse use, any behavioural stressors are likely

to be limited to handling and interactions with inexperienced students. When they enter the veterinary programme, students have low understanding of equine behaviour and knowledge of the principles that constitute the way horses learn (i.e. learning theory) (Gronqvist et al., 2017; Guinnefollau et al., 2019). Associative learning (i.e. classical and operant conditioning) requires the use of at least one stimulus and aims to increase or decrease the frequency, duration or intensity of future occurrences of desirable or undesirable behaviours (Starling et al., 2016). If unable to correctly identify equine body signals, students are therefore unlikely to provide consistent cues and to apply appropriate signals, which may negatively impact on their interactions with the horses. Mistimed and/or inconsistent signals can induce subsequent confusion and conflict behaviours and have been reported to increase arousal and reactivity levels in horses (Doherty et al., 2017; McLean & McGreevy, 2010; Overmier & Wielkiewicz, 1983; Starling et al., 2016).

Students' equine skills competency increased throughout the equine and veterinary science degree programmes. However, many students have very limited equine experience and low confidence in horse handling at enrolment and some still show low self-assessed equine skills and confidence in 4th year (Guinnefollau et al., 2019). Given the current low use reported for the teaching horses in this study, there could be an opportunity to increase student exposure to horses and horse handling. This could be implemented through either formal (syllabus) or informal opportunities. Some limitations with this proposal are the lack of data on the exposure hours required to achieve horse handling competence in naive students, the suitable metrics needed to quantify horse handling competence and how often the horses could be involved in practical teaching classes. Without suitable evaluation criteria and metrics, it is difficult to model the ideal rate of use required to achieve optimal teaching outcomes.

Through the inherent content of practical classes, the advancement of the students in the programme and the class size capacities, different horses will encounter variations in the type and number of interactions with students. To accurately describe their potential impact on the horses' welfare, further research is required on the interactions between horses and students to provide

more robust descriptors of the relative workload these horses experience in the teaching environment. Investigation of other potential stressors during practical work is also warranted. Future studies could include direct measures of stress using behavioural and physiological indicators (e.g. recording of heart rate during practical sessions) of the horses while they are involved in teaching classes to determine additional markers of welfare.

CONCLUSION

This was the first study to describe teaching-related activities of horses kept for educational purposes. Although variable between and within herds, a relatively low frequency of teaching use was reported. Practical classes were different in their frequency and routine, in the number of students per horse and year of students, and in the location and restraint of the horses. Given these findings, there may be an opportunity to increase the use of the horses for teaching to optimise the value of the teaching experience for the students. However, for animal welfare considerations, more research is required to evaluate the optimal number and frequency of equine practical teaching classes.

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PRELUDE TO CHAPTER 4

Chapter 3 provided a detailed description of the teaching-related activities of the teaching horses over the course of a calendar year. Different types of practical classes were identified, which led to variations in the type and number of student interactions between herds and horses. Overall, the relatively low use of the horses for teaching purposes was highlighted.

Because they are used in practical teaching classes involving novice students, the teaching horses are pre-selected and managed in herds of artificial social structures. Given this management at pasture, there is a need to assess the behaviour of the horses within each herd. Through an observational study, Chapter 4 provides information on the horses' time budgets and social interactions over a semester of use for practical teaching.

This chapter is prepared in the style format of the scientific Journal of Veterinary Behavior.

CHAPTER 4. TIME BUDGET AND HERD DYNAMICS OF HORSES KEPT IN GROUPS AT PASTURE AND USED FOR EDUCATIONAL PURPOSES

Highlights:

- Similar time budgets and type and frequency of social interactions to that of free-ranging populations.
- Relatively stable bilateral relationships within the herds.
- Most agonistic dyadic relationships were unidirectional.
- Horses of higher ranks tended to be more frequently chosen as preferred neighbours.
- High behavioural synchronisation between pairs of nearest neighbours.
- Agonistic behaviours were directed towards individuals of similar or lower ranks.

ABSTRACT

Unlike feral and free-ranging populations, whose behaviour and social dynamics are well described, there is a current lack of information on the diurnal behavioural patterns and herd dynamics of horses managed in groups at pasture year-round. Three herds of horses managed at pasture and specifically kept for undergraduate equine and veterinary science teaching at Massey University, New Zealand were observed at three distinct time points across the second semester of teaching. Behavioural activities were video recorded continuously for 1-hour sessions and scored to create time budgets. Social interactions (i.e. affiliative, agonistic and submissive behaviours, and nearest neighbour) were reported to describe bilateral relationships and social networks within each herd. Although different between time points and herds, time budgets were similar to that of previously published information on feral and free-ranging horses with most time (61%) spent feeding (IQR, 41-84%) and then resting for 17% (IQR, 2-37%) of the time.

Social behaviours were infrequent and accounted for a median of 1% (IQR, 0-2%) of the horses' time budget, of which submissive behaviours (55%) were the most frequent. When displayed, agonistic behaviours were of mild intensity (93%). The ranks remained relatively stable across time points. There was some variation in preferred neighbours relationships across the study, but horses of higher position in the herd tended to be more frequently chosen as neighbours. In all three herds, the behaviour of pairs of nearest neighbours was significantly synchronised. Regarding time budgets and social dynamics, these horses at pasture seem to be managed in conditions allowing them to behave and interact similarly to feral and free-ranging populations.

INTRODUCTION

The behaviour of feral and free-ranging horses is well described, with examples available on Camargue horses (Duncan, 1985), Australian feral horses (Berman, 1991), Assateague Island ponies (Keiper & Keenan, 1980), and Przewalski horses (Berger et al., 1999a; Boyd et al., 1988; Souris et al., 2007). Across these populations, horses have been consistently shown to spend the greatest proportion of their time foraging, followed by resting behaviours (standing or lying) and locomotion (Keiper & Keenan, 1980; Salter & Hudson, 1979). The estimated proportion of time spent on each activity is known as a time budget and varies among studies and between horses due to individual (e.g. age, sex) and environmental (e.g. season, temperature, insects) factors (Boy & Duncan, 1979).

Horses are highly social animals and live in herds naturally in a feral environment (Boyd & Keiper, 2005; Keiper, 1986). The need of each individual to access limited resources leads to the creation of a social order (often called dominance hierarchies) (McDonnell, 2003). Once formed, hierarchies – which can be linear or triangular in larger herds (Ingólfssdóttir & Sigurjónsdóttir, 2008; Keiper, 1988; van Dierendonck & Goodwin, 2005) – have been shown to remain relatively stable for up to three years (Keiper & Sambraus, 1986). The social position of members within the group is the result of multiple bilateral dominance relationships and is maintained within each dyad by the display of agonistic interactions, where each horse is either

the initiator or the receiver of the interaction. Agonistic interactions vary from subtle (i.e. displacement) to more violent (i.e. kick) (Keiper, 1986). Because of the associated energetic cost and risk of injury (Waran, 1997), the number and intensity of agonistic interactions seem to decrease in consistent groups in favour of more subtle behaviours (i.e. threats, displacements) (Heitor et al., 2006). The frequency of social interactions between herd members also varies in response to both the environment and population density (Berman, 1991; Franke Stevens, 1990; Ransom & Cade, 2009). In feral horse populations, individuals are frequently seen less than two metres away from a conspecific (Keiper & Sambraus, 1986) and generally do not have more than two social preferred partners (Monard & Duncan, 1996). This spatial proximity in individuals' personal space is usually observed between animals of similar rank and age (Clutton-Brock et al., 1976).

In domestic populations, research has focused on horses kept individually in stables or at pasture in small groups (Benhajali et al., 2008; Flannigan & Stookey, 2002; Heleski et al., 2002; Sweeting et al., 1985; Yarnell et al., 2015). In New Zealand, the temperate climate allows horses to be managed at pasture year-round (Rogers et al., 2017) and an example of this management can be found at Massey University in Palmerston North. As part of the undergraduate equine and veterinary science degree programmes, dedicated teaching horses are kept in consistent groups and used for a variety of procedures during practical classes (Guinnefollau et al., 2020). Teaching horses are pre-selected for teaching purposes based on their calm temperament and tolerance of the teaching procedures. Because they are kept in consistent groups of either all mares or mares and geldings of a similar (older) age, their social structure is different to that of feral and free-ranging herds and they represent a unique population to study.

To the authors' knowledge, there is no study investigating the behaviour and social interactions of herds of horses managed in these conditions at pasture and used for educational purposes. Therefore, the main aim of the present study was to provide information on the time budget and herd dynamics of teaching horses managed at pasture.

MATERIAL AND METHODS

The study population consisted of twenty-four horses used for undergraduate equine and veterinary science practical teaching classes, at Massey University, New Zealand. These horses were kept at pasture in three consistent herds – herd B (n=7), herd M (n=9) and herd T (n=8) – based on their temperament and tolerance of the teaching procedures. The study population included 3 Thoroughbreds, 19 Standardbreds, 1 Stationbred (crossbred) and 1 Kaimanawa (New Zealand feral horse). Ten horses were geldings and 14 were mares, with a mean age of 15 ± 4 years (one horse's age was unknown but thought to be over 15 years). Pasture was the primary feed source and in the winter months (i.e. June to September) hay was also provided daily (between 8-8:30am) at 2 (1-4) kg of dry matter/horse/day (median, interquartile range). Provision of hay was dependent on the horses' body condition and weight, and on the ambient temperature. The hay was provided as one pile of hay per horse with approximatively two horses body lengths (5 metres) between piles. Water was provided ad libitum from water troughs in the paddocks. The mean size of the paddocks the horses were kept on during the study period was of 1.9 ± 0.7 ha and herds were rotated between paddocks based on pasture availability.

A companion paper (Guinnefollau et al., 2020) described the use of teaching horses at Massey University. Briefly, they are used for a variety of practical teaching classes, such as animal handling, foot trimming, dental examination, lameness evaluation, clinical examination, medical rectal examination and mare reproductive rectal examination. Most of the teaching horses' interactions with students at Massey University, New Zealand occurred from February to November (educational year in the Southern Hemisphere) during two, 12-week teaching semesters, but some occurred year-round.

PROCEDURE

The study took place between June and October 2018 during the second semester of practical teaching. Semester 2 teaching occurred from July 16th to October 19th, with a mid-

semester break from August 27th to September 7th. Therefore, the study occurred at three chosen time points: (1) Start, in week number 24 (semester 1 teaching ended on week number 22); (2) During, in week number 36 (during the mid-semester break); and (3) End, in week number 41 (semester 2 teaching ended on week 42).

At each time point, observations were conducted for each herd over four consecutive days and distributed randomly when possible (i.e. depending on the availability of the herds and their use for teaching or research projects) between three times of day: 0800 to 1000 ('morning'), 1100 to 1300 ('midday') and 1500 to 1700 ('afternoon') New Zealand daylight time (Table 4.1). When hay was provided to the horses (i.e. in the time points described as Start and During), the observed herd was fed at 0800 and morning observations started at 0900. If used for teaching, general husbandry or research projects, the herds were observed at least one hour after being returned to pasture. The behaviour of the teaching horses was recorded continuously for 1-hour sessions using a video digital camera.

Table 4.1. Relative composition of observations per time of day at each time point.

| | Start | During | End |
|---------------|-------|--------|-----|
| Herd B | | | |
| Morning | n=1 | n=2 | n=0 |
| Midday | n=2 | n=1 | n=2 |
| Afternoon | n=1 | n=1 | n=2 |
| Herd M | | | |
| Morning | n=2 | n=1 | n=2 |
| Midday | n=1 | n=1 | n=2 |
| Afternoon | n=1 | n=2 | n=0 |
| Herd T | | | |
| Morning | n=1 | n=1 | n=1 |
| Midday | n=1 | n=2 | n=2 |
| Afternoon | n=2 | n=1 | n=1 |

The researcher (LG) was located in an adjacent pasture 10 metres away from the pasture gate and was in place 10 minutes prior to the start of the video recording to reduce possible changes in behaviour due to arrival of the researcher. The camera was placed on a tripod, but the researcher was present for the entire recording to adjust the angle, focus and zoom. If the position of the horses in the pasture did not allow the entire herd to be within the field of view, the researcher prioritised the video recording of the highest number of horses at once.

Each session included a report of climatic (ambient temperature and wind speed – available from <https://www.metservice.com/towns-cities/locations/palmerston-north> at the time of recording / weather station within 5 kms from the horses) and temporal (time and date) data. Information on estimated pasture mass (in kg of dry matter/ha) and hay quantity if provided (in kg of dry matter offered/horse/day) were available. Estimated pasture mass was measured once at each time point during the week of observation using a rising-plate-meter.

Individual characteristics, such as name, age, body condition score (Henneke et al., 1983) at each time point (BCS – assessed once, by the same person, the week prior to the observations), weight at the time of observations, and sex and breed, were collected for the analysis.

Time budget

Videos were randomly ordered for the footage analysis, which was carried out at the end of the study period. The recognition of the horses was based on physical characteristics (i.e. size, colour of the coat, mane and tail) and/or colour of the neck collars (i.e. plastic strap encircling the neck of the animal and used for identification). Continuous assessment of the horses' behaviours was made following the focal sampling method (Altmann, 1974) to record both behavioural events and states. The ethogram (Table 4.2) was adapted from Ransom and Cade (2009), and included the category 'Other' (i.e. 'human awareness' and 'out of sight' (Ransom & Cade, 2009)) to account for biased observations. The same researcher assessed the behaviour of all horses. The repeatability of the behaviour categorisation was assessed by comparing the coding of the same two assessments of two randomly selected horses per herd (at each time point). Behaviours (e.g. grazing, walking, grooming, standing attentive) were assessed between the minutes 0-2, 29-31 and 58-60 of the selected videos. A Cohen's kappa coefficient was calculated between each repeat (i.e. the same video scored twice) using the R package '*irr*' on the number of occurrences of the selected behaviours. Cohen's kappa coefficient values can vary from 0 (any agreement is totally due to chance) to 1 (complete agreement), with values higher than 0.61 showing substantial agreement. Agreement of score assessment between video repeats was "almost perfect" (Landis & Koch, 1977) with a mean Cohen's kappa coefficient of 0.88 ± 0.09 .

Table 4.2. Ethogram (adapted from Ransom and Cade, 2009) of the complete and mutually exclusive list of behavioural states (duration, D) and events (frequency, F) recorded.

| Category | Subcategory | Behaviour | Description |
|-----------------|------------------|--------------------------------|--|
| Feeding (D) | Drinking | | Lower head; put lips in water trough in posture typical of ingesting water in horses |
| | | | Bites off and ingests grasses and forbs close to the ground. Horses move as they graze; therefore, as long as the horse is feeding while it is moving, it should be considered as feeding rather than locomotion |
| | Feeding | Grazing | |
| | | Feeding hay | Bites off and ingests hay |
| | | Sniffing food | Progressing across the ground while scenting food |
| Resting (D) | Standing resting | | General lack of attention and relaxed state (e.g. eyes half-closed, ears on the side, head lowered, often one rear foot slightly elevated) – standing up |
| | | Resting recumbent | General lack of attention and relaxed state (e.g. eyes half-closed, ears on the side) – laying down, can be sternal or lateral |
| Locomotion (D) | Walking | | Walks at least 10 steps without interruption |
| | | Trotting | Moves forward with a two-beat gait |
| | | Cantering | Moves forward with a three-beat gait |
| | | Galloping | Moves forward with a fast, four-beat gait |
| Maintenance (D) | Grooming | | Rolling, shaking, nibbling or licking on self, rubbing, and periodic stomping to displace flies and biting insects |
| | | Urinating | Elimination of fluid waste |
| | Eliminating | Defecating | Elimination of solid waste |
| | | Standing attentive / vigilance | Rigid body posture with head upright, ears pointed forward, eyes open and directed towards a stimulus (sound or smell) – should not be confused with human awareness |
| Social (D, F) | Affiliative | Allo-grooming | Mutual grooming |
| | | Olfactory investigation | Nose-nose or nose-body contact |
| | Agonistic | Threatening | Laterally pinned back ears, arched neck, and/or movement of the head toward the opposing horse, but with no physical contact |
| | | Pushing | Forceful contact with another horse using its head, neck, or shoulder |
| | | Chasing | Offensive movement to another horse, ears back in order to displace the animal from the immediate area |

| | | | |
|--|------------|---------------------|---|
| | | Threatening to kick | Threatens another horse by showing the rump, ears back with a movement of the rear leg; no physical contact |
| | | Threatening to bite | Threatens another horse by showing the teeth, ears back; no physical contact |
| | | Biting | Bites another horse; involves physical contact |
| | | Kicking | Kicks another horse with the fore- or hind legs; involves physical contact |
| | | Rearing | Lifts its forelegs off the ground and elevates its body into a more vertical position |
| | Submissive | Avoiding | Turns head because of another horse |
| | | Running away | Moves because of another horse |
| | Other (D) | Human awareness | Any behaviour due to observer's movements or any other human activity in the environment – generally standing attentive |
| | | Out of sight | Not visible, not recognisable, or too far to identify behaviours |
| | | | |

Nearest neighbours

Nearest neighbours were identified using a 5-minute scan sampling method (Altmann, 1974) for all recorded data, during which the identity of the nearest neighbour for each horse at the specific time was recorded. The nearest neighbour was the closest horse to any body part of the focal individual outside of agonistic contexts. When at a distance of at least three horse body-lengths away from the nearest horse (Benhajali et al., 2009; Bourjade et al., 2009) and/or when it was the only individual not visible, the focal horse was considered isolated. If several horses were not visible at a focused time, the nearest neighbour data of these individuals were considered missing.

Social interactions

Social interactions were evaluated at the dyad-, group- and individual levels using observed agonistic and submissive behaviours.

For the individual level, the rank of each horse was assessed by observing agonistic interactions and by calculating the rank index (Zeitler-Feicht, 2003). Within each herd, individuals were scored one point for each agonistic behaviour that resulted in a submissive

behaviour or for each spontaneous avoidance (i.e. submission without agonistic behaviour) from the receptor horse. By definition, a rank relationship was considered established only if one individual showed at least twice as many dominant behaviours than the other. The rank index for each horse was calculated as follows:

$$\begin{aligned} \text{Rank index}_{\text{Horse A}} &= \frac{\text{number of subordinate animals}_{\text{Horse A}}}{\text{number of established rank relationships}_{\text{Horse A}}} \\ &= \frac{\text{number of establish rank relationships won}_{\text{Horse A}}}{\text{number of established rank relationships}_{\text{Horse A}}} \end{aligned}$$

As an example, in a herd of six individuals, each horse can be the potential dominant partner in a maximum of five established rank relationships with its conspecifics. If Horse A was the winner of one out of these five rank relationships, its rank index would be $1/5 = 0.2$. On the contrary, if Horse B was the winner of four out of five established rank relationships, its rank index would be $4/5 = 0.8$.

The rank index ranged from 0.0 to 1.0, and individuals were then given the corresponding rank within the herd (1 = highest-ranked horse, n = lowest-ranked horse).

STATISTICAL ANALYSIS

Statistical analyses were performed using R 3.5.3 (R studio). The threshold used for statistical significance was $p < 0.05$. Data were analysed separately for each herd.

Time budget

The behavioural states were grouped into subcategories and categories, as shown in Table 4.2, to allow calculation of the proportion of time spent performing different behaviours. The category ‘Other’ (‘human awareness’ and ‘out of sight’) was not included in the analysis. Therefore, the denominator was the total duration of the video recording during which the horse was neither out of sight nor showing signs of human awareness (subsequently referred to as ‘time visible’). Percentage of time spent in each behaviour category or subcategory was calculated.

These data were nonparametric; therefore, the median (and interquartile range; IQR) percentages of each behavioural category were calculated for each herd for the Start, During and End time points; for each time of day and for each day of observation.

Nearest neighbours

Preferred social partners were identified as the most frequently nearest neighbours. The number of times each pair of horses was seen together was used to build a network of the relationships within the herds (software Microsoft Visio 2016). Only the most frequently closest neighbours at the Start, During and End time points were represented on the network.

A sociability index (SI) was calculated following a method developed to assess individual sociability of sheep (Sibbald et al., 2005), and subsequently used for horses (Hauschildt & Gerken, 2015). The SI indicates the relative proportions of time that each horse was observed as the nearest neighbour of any other animals in the herd. The index is scaled to have an expectation of 1.0 under the null hypothesis of random mixing. Low sociability is therefore defined by a value < 1.0 and high sociability by a value > 1.0. Kendall rank correlation coefficients were calculated between the SI calculated at Start, at During, and at End.

Coefficient of synchrony

To analyse the behavioural synchrony within each herd, the percentage of observations when 100% ($P_{100\%}$) and > 50% ($P_{50\%}$) of individuals performed the same activity at the same time were calculated (Hauschildt & Gerken, 2016; Rook & Penning, 1991).

A Cohen's kappa coefficient of agreement (K) was calculated between pairs of nearest neighbours to determine if behavioural activities (i.e. feeding, resting, maintenance, locomotion and social) were more synchronised than could be expected by chance (Rook & Penning, 1991). The tolerance was set to 0, requiring both behavioural activities to be the same for it to be considered agreement.

Social interactions

Occurrences of agonistic and submissive behaviours (i.e. behavioural events) were analysed using pivot tables to generate frequency counts. Because individual horses were observed for different lengths of time, the frequency of occurrence of agonistic and submissive behaviours was standardised for comparison by calculating their frequency per hour. Medians and IQRs were calculated at the Start, During and End time points; at each time of day and with or without additional provision of hay.

In each herd and at each time point, the dyads were described following the methodology of Val-Laillet et al. (2008) used in cattle, and subsequently used in horses by (Vervaecke et al., 2006):

- Unknown dyads (%): percentage of unknown relationships. The horses of these dyads were not observed to display agonistic behaviours to one another.
- One-way dyads (%): percentage of dyads in which only one horse of a pair displayed agonistic behaviours towards the other (one-way relationship).
- Two-way dyads (%): percentage of dyads in which both horses of a pair displayed agonistic behaviours towards the other at least once (two-way relationship).
- Tied dyads (%): percentage of dyads in which both horses displayed agonistic behaviours towards the other the same number of times (tied two-way relationship).

As another descriptive measure, the Directional Consistency Index (DCI) was calculated across all dyads of a herd (Val-Laillet et al., 2008; van Hooff & Wensing, 1987; Vervaecke et al., 2006) as:

$$DCI_{Dyad\ A-B} = \frac{\text{number of agonistic behav}_{Horse\ A} - \text{number of agonistic behav}_{Horse\ B}}{\text{total number of agonistic behav}_{Dyad\ A-B}}$$

where Horse A is the horse that displayed the most agonistic behaviours in the dyad A-B; and Horse B is the horse that displayed the least agonistic behaviours in the dyad A-B.

The DCI at the herd level represents an average of all the dyads in the herd and ranges from 0 (completely equal exchange) to 1 (completely unidirectional) (van Hooff & Wensing, 1987). For example, a DCI of 1 in a herd means that all dyads are one-way dyads, while a DCI of 0 means that all dyads are tied dyads (Val-Laillet et al., 2008).

Social order

Social orders of each herd were built using the rank index of each horse. To measure the stability of the social structures over time, Kendall rank correlation coefficients (“Kendall’s tau”) were calculated between the ranks attributed at Start, at During, and at End. Kendall’s tau was used to measure the association between two quantitative and ordinal variables. Values obtained range from -1 to 1 (0 = no correlation, 1 = perfect correlation) and the sign of the coefficient shows the direction of the correlation (i.e. a negative coefficient means that the variables are inversely related) (Akoglu, 2018).

The dynamic of each pair of horses was analysed by creating a network of directed agonistic interactions for each herd (Cytoscape 3.7.1). Each network included the ranks of the horses and the number of agonistic interactions directed towards other group members. In order to place the animals that were the most connected to others in the middle, the layout chosen was a Compound Spring Embedder (CoSE) design.

Associations between the rank of an individual and the rank of its nearest neighbour were studied using Kendall rank correlation coefficients. The same correlation coefficients were used to study the association of the SI of an individual and its attributed rank.

RESULTS

Each herd was video recorded for a total of 12 hours and the study resulted in a total of 220 individual horse hours of recording. During each 1-hour video recording, individual horses were each visible for a mean duration of 47 ± 10 minutes (herd B), 44 ± 12 minutes (herd M) and 44 ± 12 minutes (herd T). Ambient temperature ranged from 2-11 °C in Start (mean was 8.8 ± 2.5 °C),

8-14 °C in During (mean was 11.4 ± 1.7 °C), and 7-18 °C in End (mean was 12.8 ± 4.4 °C). Wind speed ranged from 4-33 km/h in Start (mean was 18 ± 11 km/h), 4-18 km/h in During (mean was 12 ± 5 km/h), and 13-35 km/h in End (mean was 21 ± 8 km/h). Average estimated pasture mass was 1190.5 ± 49.1 kg of dry matter/ha in Start, 1062.1 ± 252.7 kg of dry matter/ha in During and 898.5 ± 93.3 kg of dry matter/ha in End. There was no observed variation in the body weight or Henneke's body condition scores of the horses between time points (median weight: 538 kg (IQR, 510-569 kg), median BCS: 6 (IQR, 6-7)).

Time budget – across all observations

Overall, feeding behaviours accounted for a median of 61% (IQR, 41-84%) of the horses' time budget, resting for a median of 17% (IQR, 2-37%), maintenance behaviours for a median of 7% (IQR, 3-12%), locomotion for a median of 5% (IQR, 2-8%) and social behaviours for a median of 1% (IQR, 0-2%). Self-grooming accounted for a median of 14% (IQR, 4-34%) of the maintenance behaviours and standing attentive for a median of 79% (IQR, 53-90%).

Time budget – between time points

Table 4.3 provides in-depth information of each herd's time budget at the Start, During and End time points. For each herd and each timepoint, the largest proportion of the time budget was spent feeding (44.6-88.3%). Time spent on other activities was generally in descending order of resting, maintenance, locomotion, and social behaviours.

The range of the observed behaviour values was wide, suggesting large variation within each time point observation (Table 4.3). In the subsequent analysis of the time budget, only the feeding and resting behavioural activities were investigated as they accounted for the majority of behaviours.

Table 4.3. For each herd, median (interquartile range) percentage time dedicated to the behavioural categories at each time point. Values given are for 7 (herd B), 9 (herd M) and 8 (herd T) horses.

| | Start | During | End |
|---------------|------------------|------------------|------------------|
| Herd B | | | |
| Feeding | 50.8 (24.9-75) | 69.9 (43.6-83.3) | 67.9 (39.2-86.5) |
| Resting | 26.3 (9.4-55) | 9.3 (1.1-16.6) | 14.4 (0.2-42.3) |
| Maintenance | 9.7 (7.1-14.3) | 8.7 (5.2-12.4) | 9.5 (6-13.3) |
| Locomotion | 2.7 (1.7-4.4) | 7.5 (6.1-9.8) | 2.7 (2.4-3.1) |
| Social | 0.2 (0.1-0.6) | 0.9 (0.6-1.5) | 0.9 (0.5-1.6) |
| Herd M | | | |
| Feeding | 44.6 (21.2-81.8) | 74.7 (56.1-88.1) | 54.8 (42.8-73.6) |
| Resting | 28.9 (4.6-62.5) | 9.3 (0-35.3) | 11.2 (1.4-32.2) |
| Maintenance | 5.2 (3.6-11.6) | 4.2 (2-8.6) | 13.1 (7.3-18.6) |
| Locomotion | 4.9 (2-8.5) | 4.5 (2.6-5.8) | 8.8 (6.6-12.3) |
| Social | 1 (0.6-1.3) | 0.5 (0.2-1.2) | 1.2 (0.3-2) |
| Herd T | | | |
| Feeding | 88.3 (57.6-96) | 54.4 (42.2-74.9) | 56.2 (38.3-65.5) |
| Resting | 2.8 (0-19.5) | 26.6 (6.2-39.3) | 30.5 (22.7-35.3) |
| Maintenance | 3.8 (1.4-7.1) | 6.6 (3.4-8.2) | 6.1 (2.6-12.1) |
| Locomotion | 2.2 (1.1-4.8) | 5.1 (3.6-8) | 5.6 (2-13.7) |
| Social | 0.3 (0.1-0.5) | 0.8 (0.2-1.9) | 2.3 (0.7-3) |

Time budget – between times of day

The median percentage of time of each horse within a herd engaged in feeding and resting behaviour activity per time of day are detailed in Table 4.4. The range of the observed behaviour values was wide, suggesting large variation within each time of day observation.

Table 4.4. For each herd, median (interquartile range) percentage of time dedicated to the behavioural categories at each time of day. Values given are for 7 (herd B), 9 (herd M) and 8 (herd T) horses.

| | Morning | Midday | Afternoon |
|---------------|----------------|---------------|------------------|
| Herd B | | | |
| Feeding | 48 (33-82) | 60 (25-80) | 67 (44-88) |
| Resting | 21 (5-32) | 24 (3-64) | 15 (0-33) |
| Herd M | | | |
| Feeding | 42 (25-66) | 75 (54-86) | 78 (58-91) |
| Resting | 31 (11-59) | 6 (1-18) | 6 (0-35) |
| Herd T | | | |
| Feeding | 55 (46-71) | 64 (36-91) | 62 (51-86) |
| Resting | 14 (9-34) | 28 (2-38) | 22 (2-36) |

Time budget – between days of observation

The percentage of time of each herd engaged in feeding and resting behaviour activities per time of day are presented in Appendix C. These data reflect that the behaviour pattern across herd, day and observation period is heterogeneous. In some cases, there was variation within the discrete day and observation time for a herd indicating that within a day of observation, horses within a herd may not all be engaged in the same activity at the same time.

Behavioural synchronisation

All group members in herd M were rarely observed engaged in the same activity at the same time (Table 4.5). In herd T, horses were more frequently observed engaged all in the same activity at the same time in During and End (both 17% of observations) compared to Start (6% of observations), and in morning (14% of observations) and midday (22% of observations) compared to afternoon (2% of observations). Horses in herd B were observed all engaged in the same activity at the same time between 10-30% of observations (Table 4.5).

In Start, During and End, over half of the horses in both herds B and T were engaged in the same activity at the same time in more than 50% of observations. In herd M, over half of the horses were engaged in the same activity at the same time in more than 50% of observations only in Start and During.

In each of the 3 herds, midday and afternoon time periods showed the highest behavioural synchronisation with over half of the horses in each herd engaged in the same activity at the same time in more than 50% of observations.

Feeding was the behaviour showing the most behavioural synchronisation across herds with over half of the horses within each herd engaged in the same activity at the same time in almost 50% of observations. Resting followed with synchronisation in 8-20% of observations ($P_{50\%}$).

These results indicate that within a herd, there may be distinct or consistent groupings of horses (“bands”) that were synchronised in their behavioural activities.

Table 4.5. Behavioural synchronisation per herd between time points or times of day and by behaviour. Percentage of observations during which 100% (P100%) or at least 50% (P50%) of horses were engaged in the same activity at the same time.

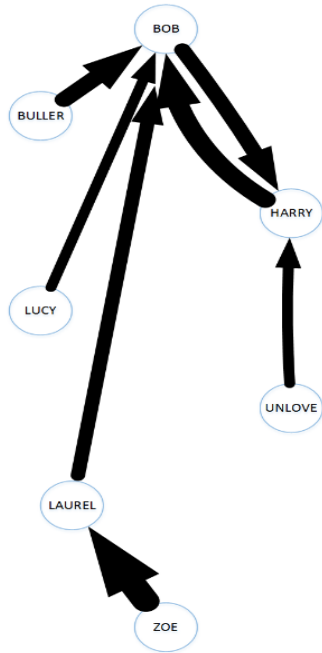
| | Herd B | | Herd M | | Herd T | |
|--------------------|-------------------|------------------|-------------------|------------------|-------------------|------------------|
| | P _{100%} | P _{50%} | P _{100%} | P _{50%} | P _{100%} | P _{50%} |
| Time point | | | | | | |
| Start | 10 | 73 | 2 | 3 | 6 | 54 |
| During | 31 | 63 | 2 | 56 | 17 | 65 |
| End | 19 | 73 | 4 | 40 | 17 | 60 |
| Time of day | | | | | | |
| Morning | 11 | 44 | 3 | 50 | 14 | 47 |
| Midday | 23 | 77 | 0 | 58 | 22 | 63 |
| Afternoon | 23 | 79 | 6 | 64 | 2 | 65 |
| Behaviour | | | | | | |
| Feeding | 19 | 47 | 2 | 46 | 12 | 45 |
| Resting | 1 | 20 | 0 | 8 | 1 | 14 |
| Locomotion | 0 | 2 | 1 | 2 | 0 | 1 |
| Maintenance | 0 | 2 | 0 | 2 | 0 | 1 |
| Social | 0 | 0 | 0 | 0 | 0 | 0 |

Nearest neighbours

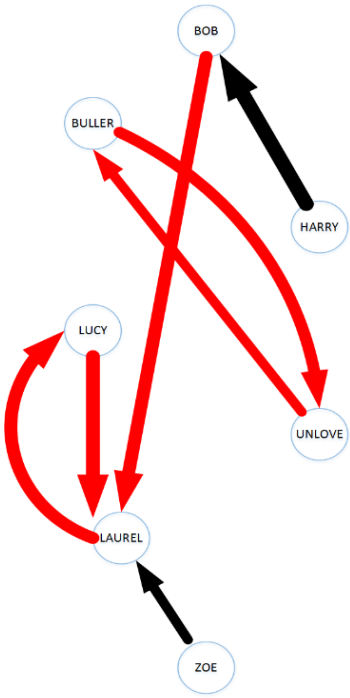
The dynamics of the preferred neighbours within each herd are represented in Figure 4.1. In all three herds, there were some strong and consistent neighbour relationships. Across the study time points, two consistent preferred neighbours relationships were observed in herd B, two in herd M, and one in herd T. Other pairs appeared consistent during an observation period but did not persist across the study time points with the horses forming new or different nearest neighbour relationships.

Herd B

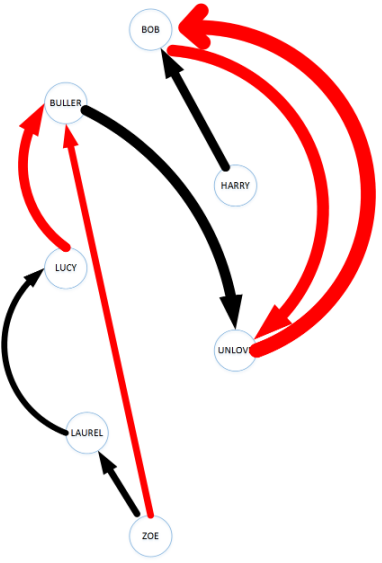
A)



B)

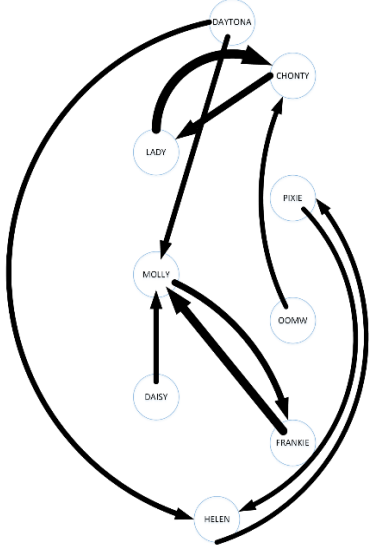


C)

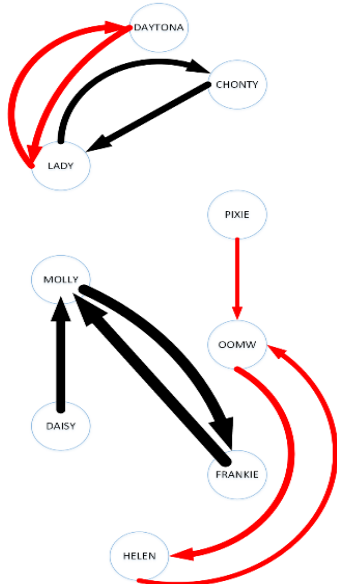


Herd M

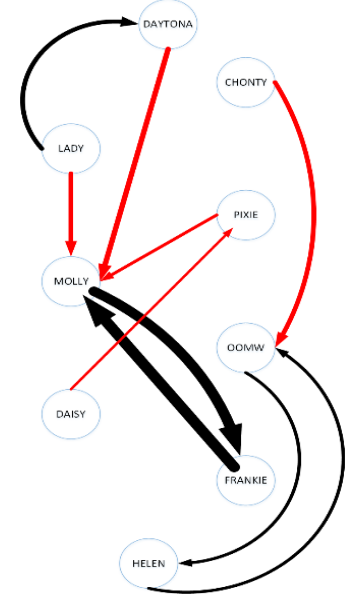
A)



B)

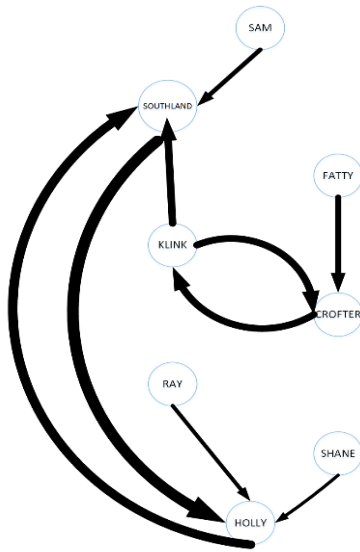


C)

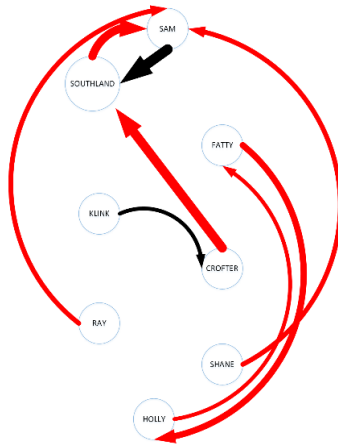


Herd T

A)



B)



C)

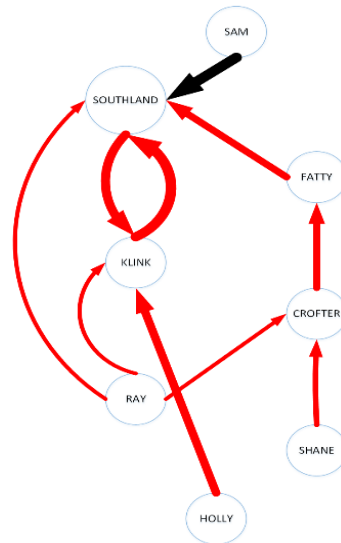


Figure 4.1. Nearest neighbours relationships within each herd (herd B, herd M, herd T) at each study time point (A = Start, B = During, C = End). The black arrows represent consistent preferred neighbours between one time point and the previous one, while red arrows highlight a new preferred neighbour. The arrows are directed and weighted: the thickness of the arrow is correlated with the number of times the horse (the start of the arrow) was seen close to its preferred neighbour (end of the arrow).

Only one horse per herd obtained a SI indicating a high sociability: Bob in herd B, Molly in herd M and Southland in herd T. There was no association between SI and time points, indicating variations in the choice of the nearest neighbours throughout the study period.

Behavioural synchrony within pairs of nearest neighbours

Significant behavioural synchronisation between pairs of nearest neighbours was found in each of the three herds (herd B: $K=0.53$, $p < 0.05$; herd M: $K=0.51$, $p < 0.05$; herd T: $K=0.58$, $p < 0.05$).

Social interactions

Overall, social behaviours accounted for a median of 1% (IQR, 0-2%) of the horses' time budget. Social interactions were primarily submissive (1076 occurrences – 55%), followed by agonistic (766 occurrences – 39%) and affiliative interactions (115 occurrences – 6%). Across all observations, the median number of agonistic interactions per hour was 2.2 (IQR, 1.0-5.2), 2.3 (IQR, 0-6.4) and 1.3 (IQR, 0-4.7) in herds B, M and T, respectively (Appendix D).

Threatening behaviour was the most frequent agonistic interaction observed in herd B (238/260 – 92%), herd M (259/271 – 96%) and herd T (216/235 – 92%). Few other agonistic behaviours were observed: pushing (0/766 – 0%), chasing (21/766 – 2.7%), threatening to kick (18/766 – 2.3%), threatening to bite (6/766 – 0.8%), biting (7/766 – 0.9%), kicking (1/766: - 0.1%) and rearing (0/766 – 0%).

Types of dyads and Directional Consistency Index

Table 4.6 shows the types of bilateral relationships for agonistic behaviours within each herd and at each time point. Over half of the dyads were one-way dyads for herds B and M, across all time points, and at During and End for herd T. Unknown dyads were the second most frequent type of observed dyads, except in herd B at During and in herd T at End. Across all time points, less than 15% of dyads were two-way dyads. Tied dyads were only observed in herd B at Start (2/21 dyads). These results are consistent with the very high Directional Consistency Index observed in all three herds.

Table 4.6. Measures of the asymmetry of agonistic behaviours displayed within each herd.

| | Herd B | | | Herd M | | | Herd T | | |
|-------------------|--------|--------|------|--------|--------|------|--------|--------|------|
| | Start | During | End | Start | During | End | Start | During | End |
| Unknown dyads (%) | 19 | 0 | 10 | 14 | 22 | 42 | 46 | 18 | 7 |
| One-way dyads (%) | 62 | 86 | 86 | 78 | 69 | 56 | 46 | 71 | 79 |
| Two-way dyads (%) | 10 | 14 | 5 | 8 | 8 | 3 | 7 | 11 | 14 |
| Tied dyads (%) | 10 | 0 | 0 | 0 | 0 | 0 | 0 | 0 | 0 |
| DCI | 0.85 | 0.92 | 0.97 | 0.97 | 0.96 | 0.97 | 0.93 | 0.94 | 0.94 |

Unknown dyad: dyad where the horses did not display agonistic behaviours towards each other; *One-way dyad*: dyad where only one horse displayed agonistic behaviours towards the other; *Two-way dyad*: dyad where both horses displayed agonistic behaviours towards each other; *Tied dyad*: dyad where both horses displayed as many agonistic behaviours towards the other; *DCI*: Directional Consistency Index ranging from 0 (completely equal exchange between horses) to 1 (completely unidirectional) (Val-Laillet et al., 2008).

Social order

The bilateral dominance relationships within each herd at the Start, During and End time points are shown in Appendix E. Although some movements in the ranks were observed for individuals in the middle positions, in all three herds the top and bottom horses did not vary throughout the study. Comparisons of the ranks throughout the study period indicated stability (i.e. correlation between the measured ranks at different time points) of the group in herd B (Start-During: Kendall's tau = 0.95, $p = 0.004$; During-End: Kendall's tau = 0.98, $p = 0.002$; Start-End: Kendall's tau = 0.88, $p = 0.006$), herd M (Start-During: Kendall's tau = 0.96, $p < 0.001$; During-End: Kendall's tau = 0.91, $p = 0.002$; Start-End: Kendall's tau = 0.93, $p < 0.001$) and herd T (Start-During: Kendall's tau = 0.69, $p = 0.018$; During-End: Kendall's tau = 0.89, $p = 0.003$; Start-End: Kendall's tau = 0.64, $p = 0.03$). The social order was strictly linear in End for herd B, in Start for herd M, and in During for herd T.

From Start to During, two bilateral dominance relationships in each herd were modified (Appendix E). From During to End, one bilateral dominance relationship was modified in herd B, and three in each of herds M and T.

Figure 4.2 shows the position of each horse in the social network of its herd and the directed number of agonistic behaviours towards other group members. In each of the three herds, the total number of agonistic behaviours displayed towards other individuals tended to be higher for horses in the middle-high position (ranks 2 and 3). Horses of lower ranks were the initiators of fewer, but the receivers of more, agonistic interactions. Placed in the centre of the network, the individuals the most connected to others were the horses of the lowest positions.

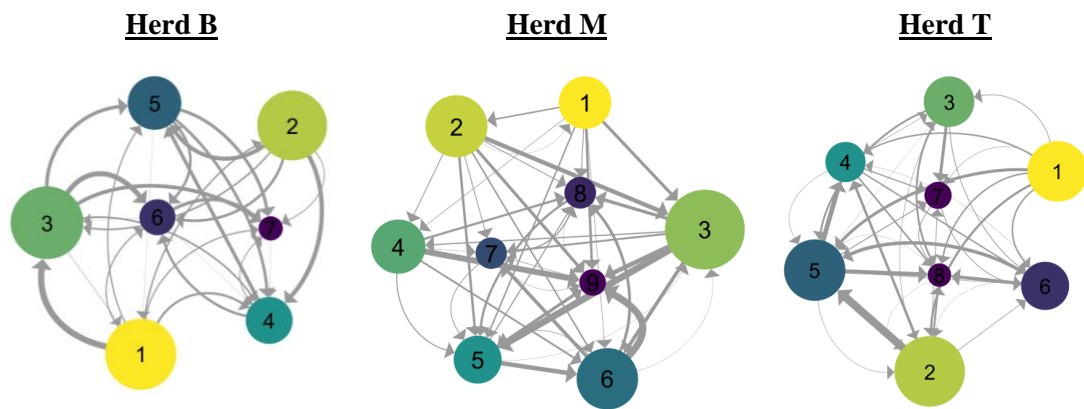


Figure 4.2. Social networks of herd B, herd M, and herd T. The nodes represent the horses and their rank positions (number 1 is the highest rank) in the herd. The size of the nodes represents the total number of agonistic behaviours performed against all other group members. The edges (arrows) represent the number of agonistic behaviours directed towards another group member. They are directed and weighted (i.e. the thicker the edge, the higher the number of agonistic behaviours). The layout chosen is a Compound Spring Embedder (CoSE) design that places in the middle the animals that are the most connected to others.

Link between nearest neighbour and rank order

The ranks of the horses were not correlated with their nearest neighbour's ranks, indicating no preference of proximity between horses of similar ranks. However, the ranks were negatively correlated with the SI in herd B ($p = 0.027$). Horses with a higher position in the herd were more often chosen as nearest neighbours by the rest of the group than horses of lower ranks. Although not significant, a similar tendency was observed in herd M and herd T.

DISCUSSION

Time budgets and social interactions of feral and free-ranging populations are well described. However, less information is currently available for domestic horses managed in groups of artificial social structures and kept at pasture year-round. By following a cohort of horses used for practical classes through a semester of teaching, the present work documents the allocation of their time between behavioural activities and herd dynamics at pasture.

Overall, the horses' activities and distributions of observed time were similar to the time budget observed for both feral or free-ranging horses kept at pasture (Przewalski horses: Boyd et al. (1988), Camargue horses: Duncan (1980), Sorraia horses: Heitor et al. (2006), Shetland pony mares: Hauschildt and Gerken (2015)). Feeding was the predominant activity of the horses in the present study, accounting for over 60% of the day-time time budget, followed by resting behaviours and maintenance and social activities. Significant variations in time budgets across observation days and time periods were observed. In horses, time budgets can be affected by environmental parameters and individual characteristics, such as time of day (Boyd et al., 1988; Ransom et al., 2014; Rubenstein, 1981; Tyler, 1972), climatic conditions (Boyd et al., 1988; Crowell-Davis et al., 1985; Tyler, 1972), season (Berger et al., 1999b; Lamoot & Hoffmann, 2004; Rubenstein, 1981; Tyler, 1972), availability of forage type (Rubenstein, 1981), age (Boy & Duncan, 1979; Salter & Hudson, 1979; Tyler, 1972) and reproductive status (Boyd, 1988). Despite a provision of hay in the morning, time spent feeding in the present study was higher in the rest of the daylight-time periods (i.e. midday and afternoon). This suggests that the horses likely started to consume the hay as soon as it was provided, leading to a resting period mid-morning.

In herds of free-ranging or domestic horses, with or without stallions, social activities account for 0.5 to 2% of the horses' time budget (Boyd & Bandi, 2002; Boyd et al., 1988; Souris et al., 2007; Yarnell et al., 2015). The number of observed agonistic behaviours per hour was slightly higher than that of feral and free-ranging horses which ranges from 0.2 to 2.0 (Boyd et al., 1988; Clutton-Brock et al., 1976; Heitor et al., 2006; Houpt & Keiper, 1982) and may be the

result of a restricted (as opposed to a free-ranging) environment. Indeed, several studies have reported an increased number of agonistic behaviours when the enclosure size decreased, highlighting the effect of density (Flaugar & Krueger, 2013; Houpt & Keiper, 1982; Jørgensen et al., 2009; Keiper & Receveur, 1992; Majecka & Klawe, 2018; Pierard et al., 2019). Resource restriction was mitigated by providing hay to the horses which maintained an adequate weight throughout the study, suggesting that competition was limited, despite an enclosed environment and a lower estimated pasture mass compared to other studies (Bengtsson et al., 2018). Contrary to Clutton-Brock et al. (1976) where food provision increased aggression rates in free-ranging horses possibly due to a disruption of the proximity between individuals, occurrences of agonistic behaviours between horses in the present study tended to be less frequent when additional hay was provided. These results suggest that food was placed sufficiently far apart and in sufficient quantity to attenuate increased aggression.

In stable and long-term groups, spontaneous submissive behaviours can be observed and may reduce the risk of injury and the costs involved with escalating conflict (Heitor et al., 2006; Jørgensen et al., 2009). Therefore, it was not surprising to record a higher frequency of submissive behaviours compared to agonistic behaviours. Indeed, once dominance relationships are established at a group level, the number of agonistic interactions and their intensity decrease in favor of more subtle and lower intensity behaviours (Heitor et al., 2006), whereas there seem to be an increased risk for bite and kick injuries in unstable herds (Knubben et al., 2008). Similarly to the results of Keiper (1988) and Heitor et al. (2006), ‘threatening’ behaviours in the present study accounted for the majority of the agonistic interactions observed, whereas behaviours of higher intensity (e.g. kicks, bites) were rarely or never recorded. The high asymmetry (proportion of one-way dyads and high DCI) of the observed agonistic interactions also supports the presence of clear dominance relationships at the group level. As reported in other groups of domestic horses (Heitor & Vicente, 2009; Vervaecke et al., 2006), most observed bilateral relationships were unidirectional (i.e. only one horse displaced the other). In addition, the rank position of the horses within the herd was correlated with the total number of agonistic behaviours they displayed

towards the other herd members. These behaviours were predominantly displayed towards individuals of lower ranks, which is similar to other findings on free-ranging horses (Clutton-Brock et al., 1976; Heitor et al., 2006; Keiper & Receveur, 1992). Therefore, lower-ranked horses were frequently the receivers but not the initiators of agonistic behaviours. Confirming the idea that interactions in social groups do not occur randomly (Dugatkin & Earley, 2003), more agonistic behaviours were also recorded between group members of similar rank position in the herd, where there might be a higher potential for variations. Contrary to the top- and bottom ranks, the rank index did not always distinguish and attribute clear rank positions to individuals in the middle. This may be due to (1) a number of horses which, in some cases, did not interact with each other directly, or to (2) unstable dominance relationships between pairs of horses (two-way dyads).

Horses typically have one or two preferred social partners (Monard & Duncan, 1996). In the present study, high behavioural synchronisation was reported between pairs of nearest neighbours, reflecting the observations of Shetland pony mares kept at pasture (Hauschildt & Gerken, 2015) which were divided into dyads and small subgroups of 3-4 individuals. Contrary to Clutton-Brock et al. (1976) who observed that neighbours shared similar rank positions, no association between the rank of an individual and its neighbour's rank was reported in the present study. However, horses that were of higher ranks in the herd tended to be more frequently chosen as preferred neighbours by their conspecifics. Similar results between sociability index and rank were reported by Hauschildt and Gerken (2015) and by Clutton-Brock et al. (1976) who described top-ranking horses rarely observed alone.

The present study faces the inherent limitations of observational studies of animal behaviour, such as the potential influence of the observer's presence or the lack of information about other factors that may have driven the changes observed throughout the study. Another limitation is the limited number and length of observations, which did not cover all hours of daytime and prevent reporting a detailed overview of the behavioural patterns. Although the teaching use of the horses was previously described as relatively low in a companion paper

(Guinnefollau et al., 2020), future work might investigate any influence of the disturbance due to teaching-related activities on the social dynamics of the herds when brought back at pasture.

Importantly, the horses of this study were managed in conditions allowing them to display similar time budgets and social interactions to that reported for free-ranging populations. Bilateral relationships within the herds remained relatively stable throughout the study, in spite of clearer rank positions for top- and bottom horses compared to those in the middle, and top-ranking horses were more frequently chosen as preferred neighbour. Keeping social animals in environments which allow them to interact and engage in potentially rewarding behaviours provides “greater opportunities to experience positive welfare states” (Mellor, 2016). Despite necessary caution when comparing feral and captive populations, these results seem to highlight the benefits of the herd centric pasture management of these teaching horses.

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PRELUDE TO CHAPTER 5

In Chapter 4, an analysis of the behavioural activities of the teaching horses was performed to establish the time budgets and social dynamics within each herd. The management in herds at pasture allowed the horses to express natural behaviours and to display similar time budgets and social interactions to that reported for free-ranging populations.

Despite the likely benefits of this management in groups at pasture, discrepancies between horses in type and number of interactions with novice students were reported in Chapter 3. Therefore, there is a need to determine if a semester of practical teaching and interactions with students has an impact on their responses to humans. The behavioural responses of the teaching horses to humans across a semester of teaching are investigated in Chapter 5.

This chapter is prepared in the style format of the scientific journal PloS One.

CHAPTER 5. TEACHING HORSES' RESPONSES TO HUMAN APPROACH OVER THE COURSE OF A SEMESTER

ABSTRACT

Many equine and veterinary science programmes rely on the use of horses for students to develop and enhance practical handling skills. Due to students' limited equine experience, teaching horses may be exposed to inconsistent and inadequate handling signals which would influence their future interactions with students. A forced human approach test was performed on 22 horses at three time points (start, during and end of teaching semester) to identify any changes in the perception horses had of humans and persistence of any change. Each researcher tested each horse once per time point. Parameters characterising the way researchers approached the horses during the test and the horses' responses were evaluated. Horses were more likely to accept human contact when approached with a slow pace, arms straight down at the sides of the person and a gaze directed at the horse's shoulder. The odds of human's acceptance reduced with increasing horse use in the 2 weeks prior to assessment, with no difference observed between the start and end of semester. These results suggest that teaching horses' reactions to humans may be influenced greatest by interactions in previous 2 weeks rather than cumulative exposure over a teaching semester. Changes in the teaching horses' responses were not observed to be long lasting. Future studies are required to understand the effect of the number and type of horse-student interactions during practical teaching classes.

INTRODUCTION

Undergraduate equine and veterinary science degree programmes frequently last for 3 to 6 years and aim to train future graduates for roles in breeding and racing organisations, private industries, veterinary practices, or academia (Austin et al., 2007; Cawdell-Smith et al., 2007; Chapman et al., 2007; Cockram et al., 2007; McGreevy et al., 2007; Rosol et al., 2009; Stafford & Erceg, 2007). Throughout their programme, students are required to gain competency and

improve their skills in various disciplines, such as clinical examination, animal handling and behaviour, animal nutrition and care, enterprise management, and communication with clients (Denniston & Russell, 2007; Lofgren et al., 2015; MacLeay, 2007; Stafford & Erceg, 2007). Teaching these competencies involves providing a combination of learning activities through which both theoretical and practical knowledge can be gained, including using live animals during practical classes to provide hands-on experience (Austin et al., 2007; Baillie et al., 2005; Cawdell-Smith et al., 2007; Chapman et al., 2007; Cockram et al., 2007; Eichel et al., 2013; Gronqvist et al., 2016; Hanlon et al., 2007; Ille et al., 2016; MacLeay, 2007; McGreevy, 2007; McGreevy et al., 2007; Riley et al., 2015; Ryan et al., 2009; Stafford & Erceg, 2007; van Vollenhoven et al., 2017; White & Chapman, 2007).

Many universities teaching equine and veterinary science degree programmes have dedicated group- or single-housed ‘teaching horses’ specifically kept for practical teaching (Austin et al., 2007; Chapman et al., 2007; Hanlon et al., 2007; Stafford & Erceg, 2007). Teaching horses are commonly used in animal handling classes and in clinical and physical examination classes (MacLeay, 2007; Stafford & Erceg, 2007). Teaching horses may be regularly exposed to a variety of approaches (e.g. pace, gaze direction, body posture, body position) and handling signals by potentially high numbers of students. Most students enter animal-based programmes with an urban background, no or little animal experience, and therefore, have limited knowledge of equine behaviour (Gronqvist et al., 2017; Guinnefollau et al., 2019). Therefore, equine practical classes may be the first time these students are interacting with horses (Chapman et al., 2007; Heath, 2008; Old & Spencer, 2011; Ostovic et al., 2017; White & Chapman, 2007).

A combination of lack of experience and lack of knowledge means that the interactions of students with teaching horses may not always be consistent. Students may not identify the subtle behavioural cues used by horses to communicate (Old & Spencer, 2011), which may prevent an anticipation of the horse’s responses and an appropriate reaction by the handler. In addition to poor timing, inconsistency and inadequacy of handling signals are indeed likely to induce confusion in the horse by reinforcing incorrect responses from the animal (McLean &

Christensen, 2017; McLean & McGreevy, 2010). Because previous contacts play a role in the horse's perception of humans (as positive, negative, or neutral), teaching horses may be regularly exposed to situations which would influence their future interactions with humans. However, despite the crucial necessity of the equine practical teaching classes, there is a lack of published data on the type and frequency of student-horse interactions for each individual horse in the educational environment.

In order to measure aversion towards humans and to assess the animals' acceptance of human contact, behavioural tests, such as human-interaction tests, have been developed (Hausberger et al., 2008). Human-interaction tests in horses generally consist of evaluating the horse's responses to a moving or stationary human, to an attempt to touch them, or to handling (Waiblinger et al., 2006). In addition to providing information on the perception the horses have of humans, human-interaction tests have been used to study the impacts of handling types (Fureix et al., 2009), number of handlers (Maros et al., 2010), type of human posture (Smith et al., 2017), housing management (Maros et al., 2010; Søndergaard & Halekoh, 2003), and eye contact (Verrill & McDonnell, 2008).

Using a forced human approach test (FHA test) over a semester of practical teaching, the aims of this study were (1) to assess the behavioural responses of the horses used for teaching to three different persons by studying their acceptance level of human approach and contact; and (2) to identify any changes in these responses in relation to their use as 'teaching horses'.

MATERIAL AND METHODS

The study population included twenty-four horses used for undergraduate equine practical teaching classes at Massey University, New Zealand. The horses included Thoroughbreds (n = 3), Standardbreds (n = 19), Stationbred (n = 1; crossbred) and Kaimanawa (n = 1; New Zealand feral horse). Ten horses were geldings and 14 were mares, with a mean age of 15 ± 4 years (one horse's age was not officially recorded but thought to be over 15 years). These horses were kept at pasture in three stable herds – herd B (n = 7, 4 mares and 3 geldings), herd T (n = 8, 1 mare

and 7 geldings) and herd M (n = 9, mares only) – that are used for a variety of practical teaching classes.

Details of the use of teaching horses at Massey University have been previously described in a companion paper (Guinnefollau et al., 2020). Briefly, the teaching horses are managed in herds, and assigned to herds based on their temperament and tolerance of the teaching procedures the herd is used for. Practical classes include animal handling, medical rectal- and mare reproductive rectal examinations, lameness evaluation, dental training, foot trimming and clinical examination. At Massey University, most of the horses' interactions with students occur from February to November (educational year in the southern hemisphere) during two teaching semesters of 12 weeks each, with an exception for the 5th-year veterinary science teaching which does not operate within the normal semester system and occurs year-round.

Pasture was the primary feed source and in the winter months (i.e. June to September) hay was also provided daily (between 8-8:30am) at 2 kg (1-4 kg) of dry matter/horse/day (median, interquartile range). Provision of hay was dependent on the horses' body condition and weight, and on the ambient temperature.

PROCEDURE

This study was approved by the Massey University Animal Ethics Committee (MUAEC Protocol 18/28) and took place between June and October 2018, during the second semester of practical teaching. Semester 2 lectures were given from July 16th (week number 29) to October 19th (week number 42), with a mid-semester break from August 27th to September 7th (week numbers 35-36). Therefore, and dependent on the availability of the teaching facilities and horses, the study occurred at three chosen time points: (1) Start, in week number 24 (semester 1 lectures ended on week number 22); (2) During, in week number 36 (during the mid-semester break); and (3) End, in week number 41 (semester 2 lectures ended on week 42). The time of day was categorised as 'morning' (0800 to 1000), 'midday' (1100 to 1300) or 'afternoon' (1500 to 1700). Twenty-two horses were used for the behavioural testing. One of two horses not used for testing (herd T) was used as company for the tested horses in order to reduce possible anxiety due to

separation from the herd. The accompanying horse was kept in a holding yard near the test yard (with visual but not physical contact possible).

The forced human-approach (FHA) test was adapted from the methodologies described by Søndergaard and Halekoh (2003) and Burn et al. (2010). Each horse was habituated to the testing yard while being physically separated from its herd (but with visual contact possible and presence of an accompanying horse). The horse was placed in the test yard (a 8.8 m × 5.1 m-indoor yard it was familiar with) each day for 5 minutes, and was considered habituated when it no longer displayed neighing, defecating, snorting or trotting behaviours (Lansade et al., 2008). The herds and the order of testing of horses within each herd were randomly selected for both the habituation period and the tests.

The test occurred over three consecutive days at each of the three time points (Start, During and End). Three researchers were used and performed the tests separately. Each researcher conducted the test for each horse at Start, During and End. Over the three consecutive days, each horse was tested once per day and the order of the researchers was pseudo-random to match with the random order of the herds. The researchers were chosen because of their different level of involvement with the horses in the study population: Researcher A (LG – lead author) had no contact with the horses prior to the start of the study; Researcher B had contact during practical teaching classes; and Researcher C had daily contact for general husbandry and practical teaching classes. For subsequent behavioural analysis, each test was continuously video recorded by two digital video cameras on tripods placed on each side of the testing yard.

At the start of each test the researcher put the horse alone in the test yard for 2 minutes and then entered the yard again using a different gate. Then, when possible and/or safe to do so, the researcher approached the horse's head while looking in the horse's direction. The approach was made from about 20 degrees on its left side, rather than from directly ahead. The researcher stopped at about 2 metres away from the horse (Step 1). If the horse did not show a negative response (e.g. threatening, moving away or turning head away), the researcher walked another step towards the horse and slowly raised one hand (Step 2) to allow the horse to sniff it. If the

horse sniffed the hand, the researcher walked another step forward if necessary and slowly attempted to touch the neck (Step 3). At each step, the decision whether to stop the test or move forward was made by the researcher. The test was concluded when the horse responded in one of a number of ways as summarised in Table 5.1 and a score was assigned. Possible scores ranged from 0 (the horse turned its head away from, moved away from or threatened the researcher at Step 1) to 3 (the horse stood still and allowed physical contact at Step 3).

Table 5.1. Details of the three steps, possible behavioural responses for the horse, and the resulting scores obtained at each step during the human-approach test. The definition of the threatening posture was based on the ethogram of Ransom and Cade (2009).

| Researcher's action | Horse's response | Outcome | Score |
|---|--|-----------|-------|
| <u>Step 1</u> Stops 2 m away | Approaches calmly toward researcher | To step 2 | |
| | Ears forward and potentially extends head and neck toward researcher | To step 2 | |
| | No response (no change in behaviour, no gaze towards the researcher) | To step 2 | |
| | Turns head away from researcher | Test ends | 1 |
| | Moves away from researcher | Test ends | 1 |
| | Displays a threatening posture (ears laid back with potentially head extended and neck arched) | Test ends | 1 |
| <u>Step 2</u> Raises hand | Sniffs the hand | To step 3 | |
| | No response (no change in behaviour, no gaze towards the researcher) | To step 3 | |
| | Turns head away from researcher | Test ends | 2 |
| | Moves away from researcher | Test ends | 2 |
| | Displays a threatening posture (ears laid back with potentially head extended and neck arched) | Test ends | 2 |
| <u>Step 3</u> Touches neck | Stands still | Test ends | 3 |
| | Moves away from researcher | Test ends | 2 |

Along with the horses' behavioural responses, parameters characterising the way researchers approached the horses at each step of the test were recorded (Table 5.2). In order to investigate these parameters, the mannerisms of the approach were not standardised prior to the test, and the researchers were asked to approach the horses as naturally as possible but consistently.

Table 5.2. Researchers and their parameters of approach recorded during the human-approach test. Fluidity, pace and body flexibility were measured using 5-point scales, whereas gaze, body posture and head position were binary variables, and arms position was measured on a 3-point scale.

| Variable | Scale score | Description |
|------------------|-------------|---|
| Fluidity | 1 | Intermittent gait, stops occur at each step during most of the approach |
| | 2 | Hesitant gait, several stops (3-4) during the approach |
| | 3 | A couple of stops possible but mostly continuous gait |
| | 4 | Continuous gait with no stops, but hesitation can be observed |
| | 5 | Very fluid gait, no stops or hesitation |
| Pace | 1 | Very slow/only long steps |
| | 2 | Slow/several long steps |
| | 3 | Medium pace, about 1 m/s |
| | 4 | Fast/several short steps |
| | 5 | Very fast/only short steps |
| Body flexibility | 1 | Very stiff during the whole approach, rigid shoulders and arms |
| | 2 | Stiff with possible relaxing body at one point during the approach |
| | 3 | Some observed tension but mostly relaxed |
| | 4 | Relaxed, shoulders move with the hips |
| | 5 | Very relaxed during the whole approach, shoulders and arms move with the hips |
| Gaze | 1 | Looks at horse's shoulder |
| | 2 | Looks at horse's head |
| Body posture | 1 | Straight/stands tall |
| | 2 | Slightly bent |
| Head position | 1 | Straight |
| | 2 | Bent/head down |
| Arm position | 1 | Arms straight down at the sides of the person, floppy, can move with the gait |
| | 2 | One or two arms on belly or lower body |
| | 3 | One or two arms on upper body |

The assessment of the types of approach made by the researchers during the tests was performed at the end of the study period by LG using the video recordings. From a random selection of 10 approaches (15% of total number of approaches), the first five were used to create an ordinal scale for each parameter of approach, whereas the other five were used to test the consistency of the assessment. All tests were then analysed using the ordinal scales created.

STATISTICAL ANALYSIS

Statistical analyses were performed using R 3.5.3 (R studio). The threshold used for statistical significance was $p < 0.05$. Frequency counts and percentages were generated to summarise data of the researchers' parameters of approach. Fisher's exact tests were performed to compare the distribution of these variables between researchers and a multiple correspondence analysis (MCA) (package '*factoextra*') was generated to visualise potential clusters between researchers. Treatment of missing values was performed using the '*BaylorEdPsych*' package for Little's test (Beaujean & Beaujean, 2012). Little's test null hypothesis states that missing values are likely to be missing completely at random (MCAR) (Little, 1988).

In order to investigate the effect of use for teaching on the horses' behavioural responses to human approach, four parameters were created based on the horses' use (i.e. number and length of sessions) (Guinnefollau et al., 2020). These parameters referred to the cumulative (1) number of practical sessions (hereafter named "sessions – 2 weeks" and "sessions – 3 weeks") and (2) number of practical session hours (hereafter named "hours – 2 weeks" and "hours – 3 weeks") in the two and three weeks prior to each study time point (Start, During and End) for each individual horse. As the measures of teaching use were nonparametric, the differences between time points were tested using a Friedman one-way repeated measure analysis of variance.

In order to study the effects of different variables on the behavioural responses of horses to the researchers, the score each horse obtained during the test (Table 5.1) was set as the dependent ordinal variable (Score) for further analysis. Multi-collinearity among independent variables was explored using the variance inflation factor (VIF) from the package '*car*', with variables kept for

further analysis when $VIF < 10$. Univariable ordinal logistic regression models were run between the dependent variable Score and each independent variable of interest (i.e. day and time of day, if the horses had been fed hay before the test, order of turn for each horse, herd, researcher, study time point, researcher's pace, gaze direction, body posture, head and arms position, and fluidity of gait and measure of teaching use) using the 'polr' function from the package 'MASS'.

Multivariable ordinal logistic regression models were built using the 'clm' function of the package 'ordinal' and variables were included when $p < 0.2$. Due to collinearity between the study time points (Start, During and End) and the teaching use measures for each horse ($\chi^2(2) = 77.9, p < 0.001$ – see Table 5.4 for an example of this measure), two separate multivariable models were built with Score as the dependent variable. Model 1 was used to investigate the study time points (Start, During and End) and researchers' parameters of approach as independent variables, with the variables 'horse' and 'herd' included as random effects. Model 2 included the horses' teaching use measures and researchers' parameters of approach as independent variables. As teaching use was measured at the horse-level, neither horse nor herd were included as random effects in Model 2. For both models, the best-fitted model was obtained from the full model after a stepwise regression method based on AIC (Akaike Information Criterion). Using a likelihood ratio test, the best-fitted model was retained as such if it did not significantly differ from the full model.

RESULTS

Description

A minimum of one day and a maximum of three consecutive days were needed for the horses to be habituated to the testing yard. Results of univariable ordinal logistic regression models indicated that the day and time of day, if hay had been provided to the herd prior to the test and the order of turn for each horse had no influence ($p < 0.2$) on the score of approach obtained during the FHA test and were subsequently not considered in the following analysis. A

summary of the researchers' scores for each parameter of approach is available in Table 5.3 and clusters between researchers are presented on Figure 5.1.

Table 5.3. Number and percentage of the researchers' scores for each parameter of approach during the human-approach test. p-Values were obtained from Fisher's exact tests when comparing the distribution of variables between researchers.

| | | Researcher A | Researcher B | Researcher C | <i>p</i> -Value |
|------------------|----------------------|--------------|--------------|--------------|-----------------|
| Fluidity | | | | | |
| | 1 | 3 (4.8%) | 7 (11.9%) | 17 (28.3%) | < 0.001 |
| | 2 | 18 (28.6%) | 29 (49.2%) | 35 (58.3%) | |
| | 3 | 37 (58.7%) | 21 (35.6%) | 8 (13.3%) | |
| | 4 | 5 (7.9%) | 2 (3.4%) | 0 (0%) | |
| Pace | | | | | |
| | 2 | 14 (22.2%) | 3 (5.1%) | 8 (13.3%) | < 0.001 |
| | 3 | 33 (52.4%) | 30 (50.8%) | 21 (35%) | |
| | 4 | 15 (23.8%) | 15 (25.4%) | 15 (25%) | |
| | 5 | 1 (1.6%) | 11 (18.6%) | 16 (26.7%) | |
| Body flexibility | | | | | |
| | 1 | 2 (3.2%) | 0 (0%) | 0 (0%) | 0.1 |
| | 2 | 16 (25.4%) | 15 (25.4%) | 22 (36.7%) | |
| | 3 | 35 (55.6%) | 32 (54.2%) | 35 (58.3%) | |
| | 4 | 10 (15.9%) | 11 (18.6%) | 3 (5%) | |
| | 5 | 0 (0%) | 1 (1.7%) | 0 (0%) | |
| Gaze | | | | | |
| | To horse's head | 63 (100%) | 8 (13.6%) | 49 (80.3%) | < 0.001 |
| | To horse's shoulder | 0 (0%) | 51 (86.4%) | 12 (19.7%) | |
| Body posture | | | | | |
| | Straight/stands tall | 42 (66.7%) | 4 (6.8%) | 2 (3.3%) | < 0.001 |
| | Slightly bent | 21 (33.3%) | 55 (93.2%) | 59 (96.7%) | |

Head position

| | | | | |
|----------|------------|------------|------------|---------|
| Straight | 7 (11.1%) | 53 (89.8%) | 26 (42.6%) | < 0.001 |
| Bent | 56 (88.9%) | 6 (10.2%) | 35 (57.4%) | |

Arms position

| | | | | |
|---------------|------------|------------|------------|---------|
| Straight | 62 (98.4%) | 58 (98.3%) | 12 (19.7%) | < 0.001 |
| On belly | 0 (0%) | 0 (0%) | 32 (52.5%) | |
| On upper body | 1 (1.6%) | 1 (1.7%) | 17 (27.9%) | |

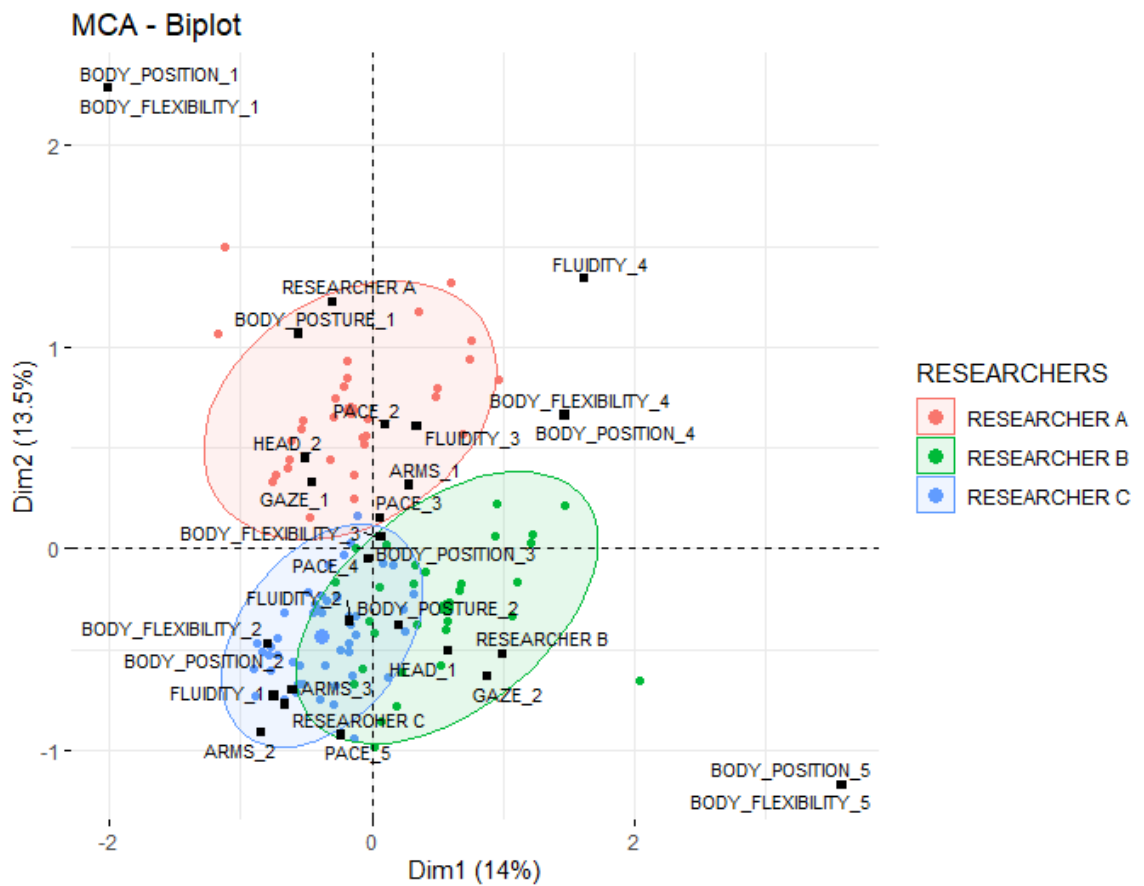


Figure 5.1. Multiple correspondence analysis (MCA) of the researchers' parameters of approach. Includes fluidity, pace, body flexibility, gaze direction, body posture, and head and arms positions. The three clusters provided by the MCA represent the researchers (researchers A, B and C).

Teaching use measurement

Table 5.4 shows an example by herd of the chosen parameters as a measure of the horses' teaching use. In Start, there was no use of the horses for practical teaching classes in the previous 2-week period and no differences between herds. However, differences in use between herds were observed in During and End.

Table 5.4. Measure of horse's teaching use by herd across the three time points of the study. Teaching use is reported as the cumulative number of practical sessions in the previous 2 weeks. Time points are the start of -, during and end of the semester of practical teaching of interest. Data are presented as median and lower and upper interquartile ranges (IQRs).

| | Start | During | End |
|--------|--------------|--------------|--------------|
| Herd B | 0 (IQR 0, 0) | 3 (IQR 3, 3) | 2 (IQR 2, 2) |
| Herd M | 0 (IQR 0, 0) | 2 (IQR 2, 2) | 0 (IQR 0, 0) |
| Herd T | 0 (IQR 0, 0) | 0 (IQR 0, 1) | 2 (IQR 2, 3) |

Variation of horses' responses to the human-approach test by time point

The number of horses that obtained each score is shown on Figure 5.2. No significant difference was observed between herds (Table 5.5). However, the researcher had a significant effect on the scores, with researchers B and C having lower odds of obtaining higher human acceptance from the horses, compared to researcher A (Table 5.5). Due to multi-collinearity between the variable 'researcher' (VIF = 15) and the researchers' parameters of approach, only the latter were used for modelling. Multi-collinearity was also observed within the researchers' parameters of approach, and body flexibility (VIF = 11) was therefore excluded from the rest of the analysis.

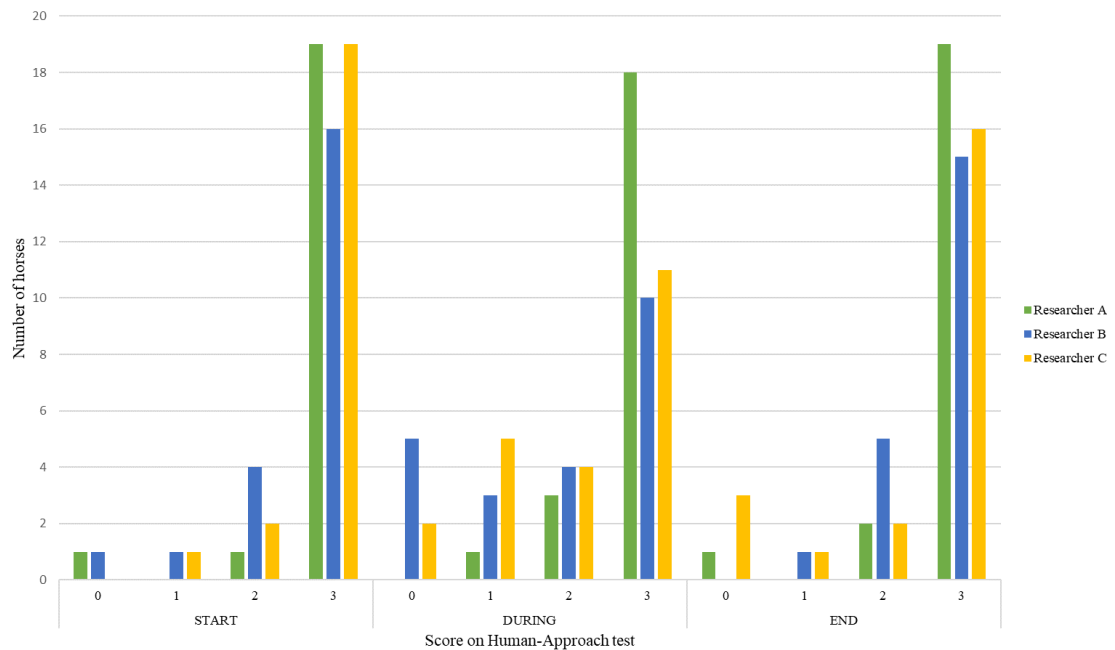


Figure 5.2. Scores obtained during the Human-Approach test. Number of horses who obtained a human approach test score from 0 to 3 across the three time points of the study (start of -, during and end of semester of practical teaching) with each researcher. 0 = lowest score of approachability (least approachable), 3 = highest score of approachability (most approachable).

As represented on Figure 5.2, more horses obtained lower scores at During compared to Start, but human acceptance from the horses did not differ between Start and End (Table 5.5). Four out of the six researchers' parameters significantly decreased the odds of having a higher human acceptance from the horses: high fluidity, high pace, slightly bent posture and arms on upper body. On the contrary, gaze directed to the horse's shoulder and head down increased the odds of having a higher score (being more approachable). The odds of having a higher human acceptance from the horses decreased with the teaching use measures (cumulative number of hours or practical sessions used for practical teaching in the 2- or 3-week period prior to each time point) (Table 5.5).

Table 5.5. Outcomes of univariable ordinal logistic regression models. The horse score of approach (score = 0 as the reference) was set as the dependent variable and the predictors as independent variables. The teaching use refers to the cumulative numbers of hours or practical sessions used for practical teaching in the 2- or 3-week period prior to each time point.

| Predictors | Univariable ordinal logistic regression model | | | |
|------------|---|---------------------|-----------|-----------------|
| | | OR | 95% CI | <i>p</i> -Value |
| Herd | M | Reference | - | - |
| | B | 0.61 | 0.3-1.24 | 0.17 |
| | T | 1.37 | 0.61-3.21 | 0.45 |
| Researcher | A | Reference | - | - |
| | B | 0.28 | 0.11-0.64 | 0.003 |
| | C | 0.35 | 0.14-0.82 | 0.019 |
| Time point | Start | Reference | - | - |
| | During | 0.28 | 0.12-0.62 | 0.002 |
| | End | 0.67 | 0.28-1.59 | 0.37 |
| Fluidity | 1 | Reference | - | - |
| | 2 | 0.86 | 0.04-1.0 | 0.77 |
| | 3 | 0.77 | 0.03-0.8 | 0.63 |
| | 4 | 0.7×10 ⁵ | 0.03-0.85 | < 0.001 |
| Pace | 2 | Reference | - | - |
| | 3 | 0.27 | 0.04-1.0 | 0.088 |
| | 4 | 0.2 | 0.03-0.8 | 0.044 |
| | 5 | 0.2 | 0.03-0.85 | 0.048 |

| | | | | |
|-----------------------------------|----------------------|-----------|-----------|--------|
| Gaze | | | | |
| | To horse's head | Reference | - | - |
| | To horse's shoulder | 2.89 | 1.48-5.7 | 0.002 |
| Body posture | | | | |
| | Straight/stands tall | Reference | - | - |
| | Slightly bent | 0.33 | 0.12-0.79 | 0.02 |
| Head | | | | |
| | Straight | Reference | - | - |
| | Bent | 2.1 | 1.08-4.16 | 0.03 |
| Arms | | | | |
| | Straight | Reference | - | - |
| | On belly | 1.45 | 0.58-4.14 | 0.45 |
| | On upper body | 0.29 | 0.11-0.78 | 0.01 |
| Teaching use (hours – 2 weeks) | | 0.85 | 0.78-0.94 | 0.0007 |
| Teaching use (sessions – 2 weeks) | | 0.63 | 0.48-0.83 | 0.0009 |
| Teaching use (hours – 3 weeks) | | 0.9 | 0.85-0.96 | 0.002 |
| Teaching use (sessions – 3 weeks) | | 0.75 | 0.6-0.92 | 0.008 |

Before the stepwise regression method, the full model 1 included the following independent variables: time point within the semester of teaching, pace, gaze, body posture, head and arms position, and fluidity of gait of the researchers; along with herd and horse as random effects (Table 5.6).

The variables 'body posture', 'head position' and 'fluidity of gait' were not significantly associated with Score in the multivariable model 1. The best-fitted model 1 revealed a significant effect of the time point, pace, gaze direction and arms position of the researcher. When the gaze of the researcher was directed at the horse's shoulder, the odds of obtaining a higher score of approach were 3.08 times that of when the gaze was directed at the head of the horse. The odds of having a higher score significantly decreased in During compared to Start, with a faster pace

of approach (4 out of 5) compared to a slow approach (2 out of 5), and when the arms of the researcher were on the upper body.

Table 5.6. Best-fitted multivariable ordinal logistic regression model 1 outcomes and adjusted odds-ratios (AOR). The score obtained by the horses during the human-approach test was set as the dependent variable. Best-fitted model was chosen using lowest Akaike Information Criterion (AIC).

| Horse's score to human-approach test (Reference = 0) – AIC = 294.5 | | AOR | 95% CI | p-Value |
|---|---------------------|-----------|-----------|---------|
| Time point | | | | |
| | Start | Reference | - | - |
| | During | 0.27 | 0.1-0.66 | 0.006 |
| | End | 0.64 | 0.23-1.7 | 0.37 |
| Pace | | | | |
| | 2 | Reference | - | - |
| | 3 | 0.35 | 0.05-1.4 | 0.2 |
| | 4 | 0.17 | 0.02-0.72 | 0.03 |
| | 5 | 0.3 | 0.04-1.47 | 0.17 |
| Gaze | | | | |
| | To horse's head | Reference | - | - |
| | To horse's shoulder | 3.08 | 1.47-6.6 | 0.003 |
| Arms | | | | |
| | Straight | Reference | - | - |
| | On belly | 0.86 | 0.31-2.66 | 0.8 |
| | On upper body | 0.26 | 0.09-0.79 | 0.01 |

Variation of horses' responses to the human-approach test in relation to their use for teaching

Before the stepwise regression method, the full model 2 included the following independent variables: pace, gaze direction, body posture, head and arms position, and fluidity of gait of the researchers, and measure of teaching use for each individual (Table 5.7). The variables 'body

posture', 'head position' and 'fluidity of gait' were not significantly associated with Score in the multivariable model 2. The best-fitted model 2 revealed a significant effect of the teaching use measure, pace, gaze direction and arms position. Compared to a low teaching use, the odds of obtaining higher scores significantly decreased with a higher use for teaching (Table 5.7).

Table 5.7. Best-fitted multivariable ordinal logistic regression model 2 outcomes and adjusted odds-ratios (AOR). The score obtained by the horses during the human-approach test was set as the dependent variable. Best-fitted model was chosen using lowest Akaike Information Criterion (AIC).

| Horse's score to human-approach test (Reference = 0) – AIC = 290.7 | | AOR | 95% CI | p-Value |
|---|---------------------|------------|---------------|----------------|
| Teaching use (sessions – 2 weeks) | | 0.58 | 0.42-0.81 | 0.001 |
| Pace | | | | |
| | 2 | Reference | - | - |
| | 3 | 0.3 | 0.04-1.23 | 0.14 |
| | 4 | 0.14 | 0.02-0.61 | 0.02 |
| | 5 | 0.29 | 0.04-1.43 | 0.16 |
| Gaze | | | | |
| | To horse's head | Reference | - | - |
| | To horse's shoulder | 3.3 | 1.56-7.16 | 0.002 |
| Arms | | | | |
| | Straight | Reference | - | - |
| | On belly | 0.96 | 0.35-2.97 | 0.94 |
| | On upper body | 0.25 | 0.09-0.76 | 0.01 |

DISCUSSION

Very few studies have described the use of animals for practical teaching in animal and veterinary science degree programmes, and there is a current lack of scientific literature on the impact of these practical teaching classes on the animals. By focusing on equine practical teaching classes, the present study is, to the authors' knowledge, the first study to assess the behavioural

responses of teaching horses over a semester of teaching to identify changes in these responses in relation to their use for teaching. Most studies assessing horse's behavioural responses to humans using forced-approach tests (FA tests) have failed to detail the human parameters during the approach (e.g. pace, body and arms position, gaze direction). Because a lack of description of these human parameters limits any reproduction of these studies and confounds the interpretation of results, we have attempted to investigate these parameters to limit their potential bias.

The researcher, or person who approached the horse, influenced the outcome of the behavioural test in the present study. The two researchers familiar to the horses were similar in the responses they obtained, whereas the unfamiliar researcher was able to approach and initiate contact with the horses more frequently. While the ability of horses to discriminate between familiar and unfamiliar persons has been demonstrated in several studies (Ijichi et al., 2018; Sankey et al., 2011), this cannot be investigated further here due to a distinct behavioural pattern in the unfamiliar researcher's way of approach compared to the two others. The odds of positive responses from the horses may therefore have been altered. As suggested by Ijichi et al. (2018), the temperament and handling skills of an unfamiliar but experienced handler can positively affect horses' responses during a handling procedure. While this study's aim was to assess horses' potential behavioural changes over time and researchers were asked to approach consistently, this first observation highlights again the necessity of (1) providing maximum details of the human parameters of approach, and (2) having homogenous testing procedures both within and between researchers to acquire comparable results.

A slow pace, a gaze directed at the horse's shoulder and arms kept straight down at the sides were more likely to be associated with being able to approach a horse more easily. Speed of approach to horses has been linked to a successful (slower pace) or failed (faster pace) approach and several studies have reported a flight response triggered from 2.5 meters (Austin & Rogers, 2007; Birke et al., 2011), once inside what appears to be the horses innate safety limit and tolerance zone only preferred conspecifics can enter (Benhajali et al., 2008; van Dierendonck et al., 2004). A fast speed of approach has been shown to increase both the tendency of the flight

response and the flight distance in horses (Birke et al., 2011). The literature is however more divided on the impact of the person's eye direction on horses' responses. While Birke et al. (2011) reported increased flight distance when looking away from the horse, Verrill and McDonnell (2008) observed no difference between avoidance and maintenance of eye contact, and Sankey et al. (2011) obtained better responses from the horses when the person giving an order was looking at the horses. As with the posture of the whole body, these parameters (pace, gaze and arm position) are likely to be affected by confidence and stress when approaching a horse. Human characteristics of approach are, therefore, important parameters to take into consideration when investigating horses' responses to humans.

It has been suggested that horses are sensitive to peoples' emotions, stress and body signals and may react in different ways based on their perception of these signals (Smith et al., 2016; Smith et al., 2017). In fact, a correlation was reported between increased human heart rate and increased horse heart rate while handling or riding, in a study investigating the effect of a nervous human (Keeling et al., 2009). When touched by people who were confident with horses and had a positive attitude towards animals, Hama et al. (1996) found lower heart rates in horses. In addition to physiological parameters, a person's general attitude when handling a horse is often associated with the horse's behaviour. In fact, Chamove et al. (2002) observed strong correlations between handler's confidence and attitude towards horses, reduction in the horse's ear movement, more frequent forward ear position and low lead tension and horse's resistance; all of which suggest lower stress and a relaxed horse (Chamove et al., 2002). In the equine and veterinary teaching environment, where animals are being handled by many students of various confidence and equine experience levels (Gronqvist et al., 2017; Guinnefollau et al., 2019), potential mistimed or inconsistent signals are likely to be frequent during practical teaching classes. Whether it be using one stimulus for two distinct responses, or two opposite stimuli simultaneously, disassociations between stimuli and responses can induce subsequent confusion in horses (McLean & McGreevy, 2010). By increasing reactivity and arousal levels (Doherty et

al., 2017; Overmier & Wielkiewicz, 1983; Starling et al., 2016), these mistimed and inconsistent signals may negatively impact the student-horse interactions during practical teaching classes.

Although fundamental for practical teaching and development of skills, the use of horses should aim to limit the animals' welfare cost. As a reflection of the human-horse relationship, the present study investigated the evolution of horses' responses at different time points across the semester of teaching and revealed a variation in approachability. While the first and third time points (Start and End of semester 2) did not show a significant difference in the teaching horses' responses, the middle of the teaching semester was associated with significant lower scores, i.e. less acceptance of human approach and/or contact. Further analysis revealed a significant impact of an increased use of the horses for teaching on the animals' acceptance of human approach and contact. The higher the number of practical sessions or hours used for during the 2 or 3 weeks prior to the test, the less likely the horses were to accept the researchers' approach and/or contact, even though they were used for a relatively low number of practical teaching sessions or hours (Guinnefollau et al., 2020). Scores increased again at the end of the semester in line with a reduction in practical teaching sessions, suggesting that any impact on the horses' responses during the test was not long lasting.

While the human-animal interaction in itself may have an indirect impact on the animal's welfare (Hausberger et al., 2008), the animal's perception of humans has been shown to be dependent on the nature and quantity of previous contacts (Søndergaard & Halekoh, 2003). In the context of the present study, most of these previous contacts refer to the inevitable interactions between horses and students during practical teaching classes. For this population of horses, other contacts with humans may involve interactions with experienced caretakers or lecturers, who likely handle the horses effectively and consistently. Although the horses used for different teaching purposes did not appear to react differently to humans in the present study, future work is needed to investigate the effect of the predictability of practical teaching classes (i.e. type and differences in students' respective horse handling skills) on horses' behavioural responses to humans. The quantity (i.e. number of students per horse) of horse-student interactions should also

be studied further as the time taken to approach a human by riding school horses with more handlers was found to be significantly longer than for horses with one handler (e.g. privately-owned horses) (Maros et al., 2010).

Controlling for every environmental parameter regarding the horses during the study period was not possible due to the design of the present study; however, there were no significant changes reported of management or within herds. Although horses were physically separated from their herd during the behavioural tests, they had been habituated to the test yard and were allowed the presence of an accompanying horse. The order of testing was random, and no effect of the day was observed, suggesting habituation to the test was unlikely. Although the analysis was performed by one of the three researchers involved in the testing, it was conducted at the end of the study and results could not have influenced the way of approach. The authors' ability to influence results while scoring each horse was also minimal based on the binary outcome following the horses' responses (i.e. test ended with negative response). This study revealed a possible effect of a specific measure of the use of the horses for teaching. However, because the use of the horses could reflect multiple factors, the authors did not assume that the actual impact of students on the horses' behavioural responses across the teaching semester was studied. In order to fully understand the impact of students on horses' arousal levels, future studies may want to design human-approach tests utilising students with different equine experience and/or confidence levels.

CONCLUSION

This study highlighted that the way humans approach horses influences the teaching horses' behavioural responses. Human approach and contact were more likely to be accepted with a gaze directed at the animal's shoulder, a slow pace and straight arms. The animals' acceptance of human approach and contact was negatively associated with an increased use of the horses for teaching in the few weeks prior to the test. Changes in the teaching horses' behavioural responses, as a result of higher teaching volume, were not observed to be long lasting.

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PRELUDE TO CHAPTER 6

Chapter 5 reported that a slow pace of the human during the approach, straight arms and a gaze directed at the horse's shoulder increased horses' acceptance of human contact. The horses' responses were moderately negatively associated with their use for teaching in the previous weeks, but this effect did not appear to be long-lasting.

Because the teaching horses interact with students during practical teaching classes, the potential impact of the student-horse interactions on the teaching horses is investigated in Chapter 6. The teaching horses' physiological and behavioural responses are evaluated before, during and after three types of practical classes.

This chapter is prepared in the style format of the scientific journal *Applied Animal Behaviour Science*.

CHAPTER 6. HORSES' PHYSIOLOGICAL AND BEHAVIOURAL RESPONSES DURING UNDERGRADUATE VETERINARY PRACTICAL TEACHING CLASSES

Highlights:

- Heart rates during practical teaching classes were in the range of a resting horse.
- Horses had low stress behavioural and physiological responses to teaching activities.
- Horses appeared to be accustomed to their use for teaching.

ABSTRACT

Horses are used in many equine and veterinary science degree programmes during practical teaching classes for students to develop and refine their handling and clinical skills. There are few studies investigating the stress responses of these horses within the teaching environment. The aim of this study was to evaluate the physiological and behavioural responses of teaching horses during three types of practical teaching classes: equine handling (EH), medical rectal examination (MR) and mare reproductive rectal examination (RR). The study population included 20 teaching horses from three herds (mean time in herd of 7 ± 3 years). The horses' heart rate (HR) was assessed before (horses in holding yard and entering the stocks/teaching yard), during (horses in stocks/teaching yard, with or without interaction with students or lecturer) and after (horses in holding yard and leaving the stocks/teaching yard) a practical class. In addition, behaviour (pawing, eating hay, vocalisation, head surveying, tail swishing, head movement, shifting weight backwards and ears orientation) of the horses used during rectal examination classes was recorded. Median class duration was 99.50 min (IQR: 95.75-112.25 min) for EH, 37.50 min (IQR: 32.25-40 min) for MR, and 47 min (IQR: 47-48 min) for RR classes. Horses interacted with students for 26% (RR) to 40% (EH and MR) of the classes' duration. There was no change in median HR (35 bpm, interquartile range [IQR]: 33-39 bpm) between activities during EH classes.

Median HR was greater when horses entered the stocks compared to the holding yard before the class in MR (entering stocks: 52.5 bpm, IQR: 43-58 bpm; yard-before: 39 bpm, IQR: 36-40 bpm; $p = 0.047$) and RR (entering stocks: 46 bpm, IQR = 41-49 bpm; yard-before: 37 bpm, IQR: 33-41 bpm; $p = 0.031$) classes. For both rectal examination classes, horses spent most of their time eating hay (MR: 94%, IQR: 73-100%; RR: 61%, IQR: 46-79%). In RR classes, horses spent less time eating hay (36%) and had their ears pointed backwards more frequently (1.39 occurrences/min) during an interaction with a student compared to no interaction. Limited physiological and behavioural stress responses were observed in response to the three types of practical classes, suggesting this population of teaching horses may be habituated to both their use and environment for teaching.

INTRODUCTION

Given the complexity of equine behaviour, live horses are used in many equine and veterinary science degree programmes during practical teaching classes. These classes vary in many ways, including the type of procedure(s) performed on horses, the class duration, the number and year of study of the students, the method of restraint of the horse and the routine adopted (Guinnefollau et al., 2020). In the University teaching environment, the aim is to balance the potential benefits for teaching with the animal welfare cost. Practice is guided by a code of ethics for the use of animals in teaching (Anonymous, 1999), coupled with internal review by an institutional animal ethics committee, with attention focused on the types of procedures and the frequency of use of the horses in order to limit any negative impact on the horses. However, there is limited research around the use of horses for teaching purposes and the events that could be perceived by horses as negative during teaching procedures and potentially elicit stress responses (i.e. a stressor).

For relevant results, the quantification of stress responses relies preferably on a combination of non-invasive methods that don't themselves cause stress. In the literature, salivary endocrine levels (Erber et al., 2013; Ille et al., 2016; Schmidt et al., 2010; Yarnell et al., 2015), eye

temperature (Yarnell et al., 2015), heart rate (HR) and heart rate variability (HRV) (Erber et al., 2013; Ille et al., 2016; Leiner & Fendt, 2011; Schmidt et al., 2010; Strand et al., 2002), and behaviour (Erber et al., 2013; Leiner & Fendt, 2011; Strand et al., 2002; Yarnell et al., 2015) are established parameters to assess short-term stress responses in horses. Nevertheless, caution is required in the interpretation of the results of each individual assessment method as they may be influenced by physical activity, other stressors, pain or arousal levels (Dawkins, 2003; Hockenhull & Whay, 2014). Adequate techniques measuring stress responses during practical classes should also aim to avoid disrupting the course of the teaching.

Some research suggests that short (5 minutes maximum) reproductive transrectal palpation and ultrasound examinations performed by experienced veterinarians or researchers do not elicit major stress responses in mares (Ille et al., 2016; Schonbom et al., 2015). Even when differences between mean heart rates before and during the procedures were reported, HR values were not greater than 60 beats per minute (bpm), which lies within the range of 25-60 bpm described for a resting horse (Clayton, 1991).

Other research suggests that the teaching horses experience of the procedures they are used for may attenuate their stress responses. In comparison to mares without previous experience of transrectal palpation and ultrasound examination, teaching mares that were familiar with the procedures had lower and stable stress-related endocrine parameters (Berghold et al., 2007). The authors concluded that the rectal reproductive examination was not a major disturbance to the teaching horses' welfare.

In the teaching environment however, the impact of procedures such as transrectal palpations on the horses may be questioned as they are performed by students of various experience levels (Berghold et al., 2007). The duration of the procedure may also be longer during teaching compared with routine and short transrectal palpation and ultrasound examinations performed by veterinarians. So far, however, very few studies have investigated horses' stress responses to transrectal palpation in the veterinary teaching environment.

It was recently reported that a 20-minute transrectal palpation of the reproductive tract performed by veterinary students elicited a stress response in experienced teaching mares (van Vollenhoven et al., 2017). A significant increase in mean heart rate and variations in HRV parameters were observed during the first five minutes of the procedure compared to baseline values (at pasture and/or in stocks) before palpation. Heart rate values were the highest at 65 minutes post procedure and were explained by a disruption of the routine of the horses that were usually led out of stocks directly after the procedure. This study was a first step in the investigation of horses' physiological stress responses in the veterinary teaching environment. However, the procedure was standardised and performed by one student per horse for the whole duration, which is different from a routine practical teaching class where interactions with various veterinary students may occur. Therefore, it is uncertain to what extent these findings can be applied to routine practical classes.

To the authors' knowledge, there is currently no published literature on the responses of teaching horses to veterinary teaching procedures other than transrectal reproductive examination. Therefore, the aim of this study was to investigate the stress responses of teaching horses during three types of routine veterinary practical teaching classes (i.e. equine handling, medical rectal examination and mare reproductive examination) in field conditions by measuring physiological (i.e. heart rate) and behavioural parameters.

MATERIAL AND METHODS

HORSES

At Massey University, New Zealand, twenty-four horses are routinely used for undergraduate equine practical teaching. Their use for teaching has been described in detail in a companion paper (Guinnefollau et al., 2020). Briefly, the horses are kept at pasture and managed in three herds (herd B, herd M and herd T) based on their temperament and tolerance of the teaching procedures. They are used for a variety of practical teaching classes (i.e. equine handling,

lameness, dental, foot trimming and clinical examination, and medical rectal and mare reproductive examinations).

In this study, data were collected on 20 horses (herd T = 6 horses – geldings only, herd B = 7 horses – 4 mares and 3 geldings, herd M = 7 horses – mares only). The study population included Thoroughbreds (n = 3), Standardbreds (n = 16) and Stationbred (n = 1; crossbred) horses. Nine horses were geldings and eleven were mares, with a mean age of 16 ± 4 years. The horses were in the teaching herds for a mean of 7 ± 3 years.

PROCEDURE

This study was approved by the Massey University Animal Ethics Committee (MUAEC Protocol 18/37). Because students were also being observed in the video recordings, the study was peer-reviewed by the Massey University Human Ethics Committee (4000019531) and judged to be low risk.

The study took place between August and October 2018, during the second semester of practical teaching. Three types of practical teaching class were chosen for this study: 1st-year equine handling (EH – which uses herd T and occurs once a week), 5th-year medical rectal examination (MR – which uses herd B and occurs once a fortnight) and 5th-year mare reproductive rectal examination (RR – which uses herd M and occurs once a fortnight). Each horse was monitored twice (i.e. during two practical teaching class scheduled on different days) with different and unfamiliar students each time.

General description of the normal practical classes:

Annual approval from the Massey University Animal Ethics Committee is required for the use of the horses in all practical teaching classes, and details of this use are stored electronically.

Equine handling classes: The horses were brought into the holding yard of the teaching facility in their usual herd 30 minutes before the start of the class. At the start of the class, the three horses

chosen by the teaching technicians for the class were walked in-hand into three yards of the indoor teaching facility, with the rest of the herd remaining outside in the holding yard. Each horse had access to a hay net in its yard. Each week's practical training involved different cohorts of students who were divided into small working groups of three to four students per horse. The students performed three activities (A: safely approach, catch, halter and lead the horse; B: lift the horse's feet safely – from once foot up to all four feet depending on the students' confidence – in no particular order; and C: apply and remove a rug safely on the horse). At the end of these activities, time was allocated for the students to groom the horse before they walked the horse in-hand into the outdoor holding yard.

Mare reproductive- and medical rectal examination classes: The horses were brought into the holding yard of the teaching facility in their usual herd 30 minutes before the start of the class. At the start of the class, the entire herd was walked in-hand from the holding yard into stocks adjacent one another in the indoor teaching facility, where the horses were individually offered hay in hay nets. Each week of classes involved different students. After a talk describing the manipulation, the students were supervised while they prepared the horses with tail protection and performed the rectal examinations. In MR classes, students were taught to palpate the accessible parts of the intestinal tract and associated organs, and in RR classes, they were taught to transrectally palpate the mares' reproductive tract. At the end of the class, the students removed the tail protection and cleaned the perineal region of the horses to remove lubricant. The horses were then walked in-hand back to the outside holding yard.

Heart rate measurement

Five Polar Equine devices (Polar Electro OY, Kempele, Finland) were used to measure the heart rate of up to five horses during a practical teaching class. The measurement equipment consisted of two electrodes and a transmitter attached to the horse's thorax by a surcingle. Electrocardiogram gel was applied on the electrodes and the horse's coat was lightly wetted to promote signal transmission. Data were collected and recorded using a wristwatch receiver/data

logger. At completion of the practical teaching class, the heart rate data were extracted and stored in a Microsoft Excel spreadsheet.

Each horse chosen for monitoring during the practical teaching class was equipped with a Polar Heart rate monitor, attached via a surcingle, and returned to the herd in the holding yard. The recording of the heart rate data started 5 minutes after the horse was equipped with the heart rate monitor, and data were collected continuously from 15 minutes before the start of the practical (baseline), during the practical, and for 15 minutes after the end of the practical. The heart rate equipment was then removed and the herd was returned to the pasture as per usual management.

Behavioural observations

The horses were video recorded for the entire duration of the class (i.e. when in the teaching yards or stocks) using a digital video camera (Sony Handycam HDR-PJ230E). After time synchronisation with the heart rate monitors, the cameras were fixed, either using tripods (for the EH classes) or on a customised wall mount 1.5 meters above and in front of the horse's head (for the RR and the MR classes). The videos were subsequently used to describe each activity during the classes, and to continuously assess the behaviour (i.e. recording of all occurrences or duration of the behaviours) of each monitored horse in MR and RR classes. In EH classes, horse behaviour was not assessed due to (1) the number of students (3-4) around each horse; (2) the possibility of the horse to move freely in the yard; and (3) the interaction of the students with the horse (e.g. petting the head). The ethogram used to determine horse behaviour is presented in Table 6.1.

Table 6.1. Behavioural indicators used in the medical and mare reproductive rectal examination practical teaching classes. F = frequency; D = duration.

| Behaviour | Description |
|---|--|
| Eats hay (D) | grasping hay followed by jaw movements and swallowing (Stewart et al., 2003) |
| * Paws (F) | raising a front leg and dragging it forwards and backwards across the floor (Stewart et al., 2003) |
| * Vocalises (F) | includes neighs, whinnies, snorts and squeals (Stewart et al., 2003) |
| Shifts weight backwards (F) | in stocks – putting weight backwards and pressing the rump on the gate (at the rear of the horse) (adapted from Stewart et al. (2003)) |
| Ears oriented forward (F) | both ears pricked up pointing forwards and stationary for 3 seconds or more (Young et al., 2012) |
| Ears scanning (F) | both ears moving back and forth at varying speeds (Young et al., 2012) |
| Ears oriented backwards (F) | both ears gently back and stationary for 3 seconds or more (Young et al., 2012) |
| * Ears laid back (F) | both ears pressed caudally against the head and neck (McDonnell & Haviland, 1995) |
| Head surveying (F) | head scanning through 45 degrees or more (Young et al., 2012). The movement starts and ends with the head aligned with the body. |
| * Head movement (F) | movements of the head such as headshaking, nodding, bobbing and circling (Young et al., 2012) |
| * Tail swishing (F) | flicking tail to one side and/or the other of the hindquarters (Young et al., 2012) |
| * behaviours believed to be indicative of stress (Young et al., 2012) | |

DATA ANALYSIS

Data collected for each horse included the horse's name, herd of animal, date and type of class. The duration of the video and heart rate recordings were divided into different time periods and categorised as the following mutually exclusive activities: holding yard before the class (i.e. baseline of 15 minutes; from the time the heart rate monitor was fitted until the horses were caught and fitted a halter by a student); enters teaching yard/stocks (i.e. from the time the horses were

caught and fitted a halter by a student in the holding yard to the time the horses were inside the teaching yard/stocks); in teaching yard/stocks (i.e. from the time the gate was closed to the time it was opened again before leaving); cleaning (i.e. putting on or removing tail protection; cleaning the horse (in MR and RR classes only – from the time the student started touching the horse until they stopped); student (i.e. x students working with the horse – from the time the student started touching the horse until they stopped); lecturer (i.e. demonstration by a lecturer – from the time they started touching the horse until they stopped); leaves teaching yard/stocks (i.e. from the time the gate was opened until the halter was removed in the holding yard); and holding yard after the class (i.e. recovery period of 15 minutes – from the removal of the halter until the heart rate monitor was stopped). In EH classes only, the type of manipulation by the students was categorised as: safely approach, catch, halter and lead the horse (i.e. activity A); lift the horse's feet safely (i.e. activity B); and apply on and remove a rug safely from the horse (i.e. activity C).

The class duration referred to the time in the teaching yard or stocks (i.e. after entering and before leaving). The time in the teaching yard, or in stocks, referred to the horse being in the teaching yard or stocks with no student or lecturer manipulating (i.e. handling, rectal examination or cleaning). The total time a horse was manipulated by students was obtained by adding together all student manipulations' time for an individual horse.

For MR and RR classes, manipulation by a student was considered from the time the student first touched the croup of the horse until the time the student stopped touching the croup. Each activity (A, B and C) in EH classes started when the student approached the horse (from approximately 1 metre away) and stopped when the student walked away from the horse (to approximately 1 metre away). Manipulation by the lecturer was categorised following the same procedure.

In conjunction with the heart rate data, the frequency and duration of selected behavioural indicators (Table 6.1) were analysed for the horses used in MR and RR classes. Due to differences in the duration of each activity, behaviours were either expressed as number of occurrences per minute or as percentage of time.

STATISTICAL ANALYSIS

Statistical analyses were performed using R 3.5.3 (R studio). The threshold for significance was $p < 0.05$. All data were non-parametric (class duration, time per student, total time with students, number of students, time in yards or stocks, cleaning time, time with lecturer, HR values and behavioural data) and were reported as medians and interquartile ranges (IQRs) throughout.

Differences in median HR, behaviour frequencies and behaviour proportions between activities (e.g. yard – before, entering stocks, student, lecturer, leaving stocks) were tested using Friedman's test for repeated measures. Post hoc differences were tested using a Wilcoxon test and the Bonferroni adjustment method.

RESULTS

CLASS CHARACTERISTICS

Characteristics of each type of practical teaching class are presented in Table 6.2. EH classes were the longest. Horse total interaction time with students represented 40% of the class in EH (IQR: 39-40%; 39.5/99.5 min) and MR (IQR: 30-42.5%; 15/37.5 min), and 26% (IQR: 23-26%; 12/47 min) in RR. When horses interacted with the lecturer in EH classes, the time of interaction was greater compared to MR and RR classes. The lecturer in RR classes interacted in each class with all horses observed, whereas with only 3 out of the 14 horses in MR classes.

Table 6.2. Characteristics of the three types of equine practical teaching classes. The number of students and the durations of the classes (mins) are presented in median and IQR (interquartile range). For equine handling classes: activity A = safely approach, catch, halter and lead the horse; activity B = lift the horse's feet safely; activity C = apply and remove a rug safely on the horse. Equine handling (n = 6 horses), medical rectal examination (n = 7 horses), mare reproductive rectal examination (n = 7 horses).

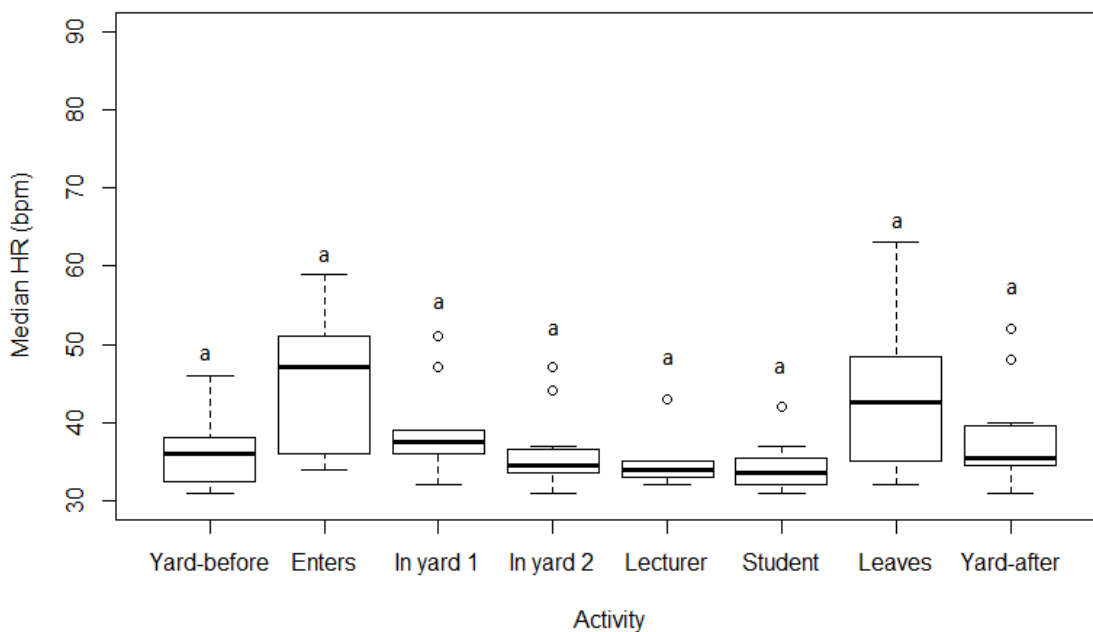
| | Equine handling | Medical rectal examination | Mare reproductive rectal examination |
|---|------------------------|-----------------------------------|---|
| Class duration (min) | 99.50 (95.75-112.25) | 37.50 (32.25-40) | 47 (47-48) |
| Horse interaction time per student (min) | 11 (7.25-13) | 7.00 (5.50-9.00) | 2 (1-3.25) |
| Activity A | 3 (1.75-4) | - | - |
| Activity B | 1.5 (1-2.25) | - | - |
| Activity C | 6.5 (5-8.75) | - | - |
| Horse total interaction time with students (min) | 39.5 (37.75-45.25) | 15.00 (11.25-17.00) | 12 (11-12) |
| Activity A | 10.5 (9.5-12.5) | - | - |
| Activity B | 5 (4.5-8) | - | - |
| Activity C | 24 (20.5-26) | - | - |
| Number of students interacting with each horse | 4 (3-4) | 2 (2-2) | 4 (4-4) |
| Time spent in teaching yards or stocks by the horses without interaction (min) | 55.5 (48.5-62) | 17.50 (15.25-19.75) | 30 (30-31) |
| Cleaning duration (min) | - | 3 (2-3) | 2 (2-2) |
| Proportion of horses the lecturer interacted with | 5/12 | 3/14 | 14/14 |
| Horse interaction time with lecturer | 22 (21-27) min | 1.00 (0.50-1.50) min | 44 (37-71) secs |

HEART RATE

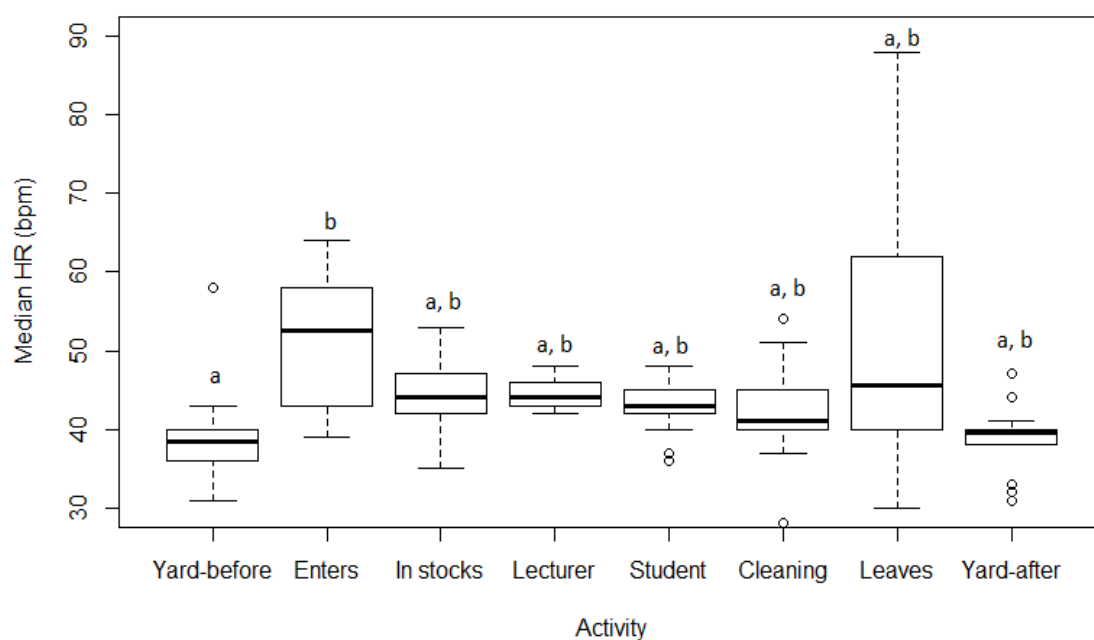
Examples of the horses' heart rates before, during and after the practical teaching classes are available in Appendix F.

The HR values are represented in Figure 6.1 below. No significant differences in median HR were observed between activities during EH classes. Entering stocks was associated with higher HR values compared to before the class in the holding yard in both MR (entering stocks: 52.5 bpm, IQR: 43-58 bpm; yard-before: 39 bpm, IQR: 36-40 bpm; $p = 0.047$) and RR classes (entering stocks: 46 bpm, IQR = 41-49 bpm; yard-before: 37 bpm, IQR: 33-41 bpm; $p = 0.031$). A similar tendency of increased HR between holding yard and leaving stocks was observed in RR classes ($p = 0.063$).

Equine handling:



Medical rectal examination:



Mare reproductive rectal examination:

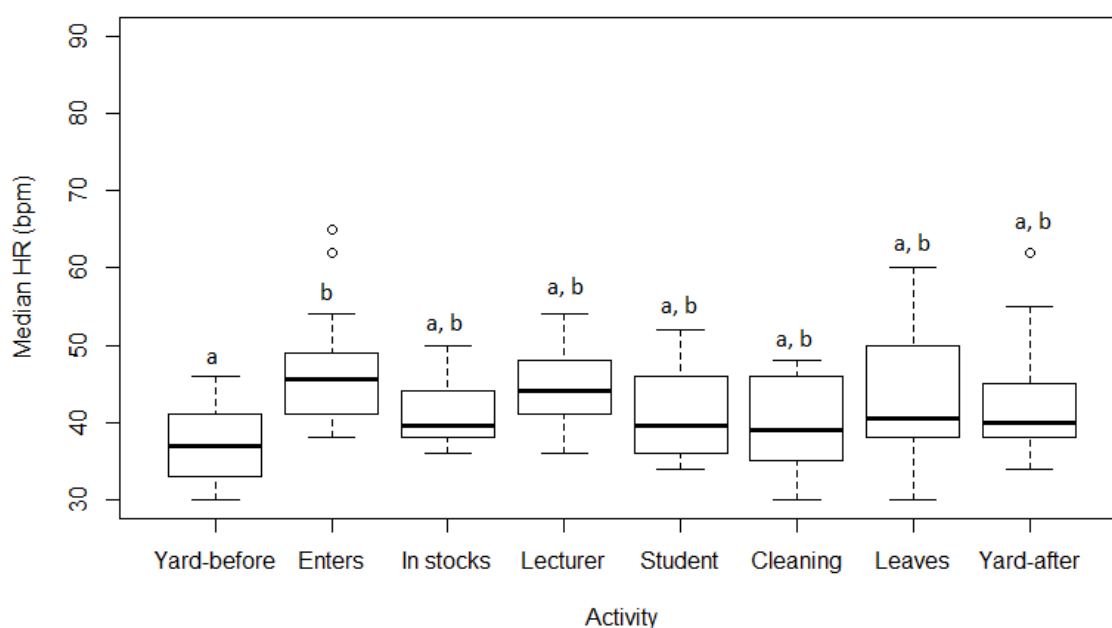


Figure 6.1. Boxplots of heart rate (HR) values between activities during the three practical teaching classes. Only for the equine handling classes: “In yard 1” = in teaching yard – without student; “In yard 2” = in teaching yard – with students. Equine handling (n = 6 horses), medical rectal examination (n = 7 horses), mare reproductive rectal examination (n = 7 horses). Different superscript letters indicate significant difference ($p < 0.05$) among activities.

BEHAVIOUR

The horses spent a median 94% (IQR, 73%-100%) of time eating hay in MR classes and 61% (IQR, 46%-79%) in RR classes. Head surveying and orientation of the ears were the most frequent behavioural events recorded (Table 6.3).

There was no difference in behaviour duration and frequency between activities during MR classes. In RR classes, the horses spent less time eating hay when students performed a rectal examination compared to no interaction with the students. Shifting weight backwards in stocks was observed more frequently during a rectal examination by a student compared to both horses being cleaned and left in stocks without interaction with students. Being in stocks without interacting with students was associated with less ears pointing backwards compared to students performing a rectal examination and cleaning the horses (Table 6.3).

Table 6.3. Percentage of time and numbers of occurrences per minute (median, IQRs) of behaviours for horses during medical and mare reproductive rectal examination practical teaching classes. Medical rectal examination (n = 7 horses), mare reproductive rectal examination (n = 7 horses).

| Medical rectal examination | | | | | |
|-----------------------------------|------------------|------------------|------------------|-----------------|-----------------|
| Activity | In stocks | Lecturer | Student | Cleaning | <i>p</i> -Value |
| Eating hay (%) | 91 (69-100) | 100 (64-100) | 100 (90-100) | 94 (50-100) | 0.18 |
| Pawing (n/min) | 0 (0-0.18) | 0 (0-0) | 0 (0-0.06) | 0 (0-0) | 0.39 |
| Vocalisation (n/min) | 0.05 (0-0.12) | 0 (0-0) | 0 (0-0.12) | 0 (0-0) | 0.3 |
| Head surveying (n/min) | 1.9 (1.74-2.81) | 1.5 (1.31-2.25) | 1.42 (0.92-2.11) | 1.5 (1.08-2.75) | 0.24 |
| Shifting weight backwards (n/min) | 0 (0-0) | 0.5 (0.25-0.75) | 0.58 (0.29-0.76) | 0 (0-0) | 0.1 |
| Ears oriented forward (n/min) | 2.03 (1.77-2.39) | 2 (1-3.5) | 1.39 (1.11-1.94) | 1.9 (1-2.25) | 0.97 |
| Ears scanning (n/min) | 1.66 (1.37-2.48) | 1.11 (0.81-4.06) | 1.64 (1.35-1.98) | 2.58 (1.85-3) | 0.33 |
| Ears oriented backwards (n/min) | 1.8 (1.4-2.05) | 1.11 (0.56-1.81) | 1.89 (1.57-2.11) | 3.33 (2.08-4) | 0.62 |

| Mare reproductive rectal examination | | | | | |
|--------------------------------------|-------------------------------|--------------------------------|-------------------------------|--------------------------|-----------------|
| Activity | In stocks | Lecturer | Student | Cleaning | <i>p</i> -Value |
| Eating hay (%) | 71 (62-89) ^a | 13 (0-83) ^{ab} | 36 (16-55) ^b | 44 (25-66) ^{ab} | 0.02 |
| Pawing (n/min) | 0 (0-0.03) | 0 (0-0) | 0 (0-0) | 0 (0-0) | 0.01 |
| Vocalisation (n/min) | 0 (0-0.04) | 0 (0-0) | 0 (0-0) | 0 (0-0) | 0.02 |
| Head surveying (n/min) | 1.13 (0.87-1.62) | 1.11 (0-2.22) | 1.11 (0.59-1.41) | 1.83 (0.88-2.38) | 0.65 |
| Shifting weight backwards (n/min) | 0 (0-0) ^a | 0.5 (0-1.08) ^{ab} | 0.35 (0.1-0.74) ^b | 0 (0-0) ^a | < 0.01 |
| Tail swishing (n/min) | 0.01 (0-0.03) | 0 (0-0) | 0 (0-0) | 0 (0-0) | < 0.01 |
| Ears oriented forward (n/min) | 1.26 (1.12-1.49) | 1.86 (1.03-3.33) | 1.22 (1-1.72) | 2 (1-2.75) | 0.43 |
| Ears scanning (n/min) | 1.3 (1.12-1.65) | 1.56 (1.03-2.22) | 1.35 (1-1.77) | 1.67 (0.86-2.92) | 0.65 |
| Ears oriented backwards (n/min) | 1.13 (1.11-1.25) ^a | 2.11 (1.11-3.25) ^{ab} | 1.39 (1.32-1.95) ^b | 2 (2-2.25) ^b | 0.02 |

^{a, b, c}: Different superscripts indicate significant differences among activities within a row, based on a Bonferroni post hoc test ($p < 0.05$). Head movement (n/min) was never observed; tail swishing (n/min) was only observed during mare reproductive rectal examination classes.

DISCUSSION

Despite a wide use of horses for educational purposes in most equine and veterinary science degree programmes, empirical information is currently lacking on the horses' responses to practical teaching classes. To the authors' knowledge, this is the first study to describe teaching horses' physiological and behavioural responses during teaching and within three types of practical classes.

The teaching horses in this study had low heart rates and showed behavioural responses that did not appear to reflect stress or anxiety. Many of the parameters measured during and across the practical classes reflected data published for horses at rest, and the baseline HR (at rest) data collected previously on a subset of these horses by Reid et al. (2017). The horses spent most of

their time in the practical class eating the hay provided upon arrival, which suggests a low-stress environment as the presence of stressors may lead to reduced or suppressed feeding behaviours (Kay & Hall, 2009).

Based on the conceptual model proposed by Gronqvist et al. (2016), horses' stress responses during teaching can be minimised with greater consistency and repeatability of events. Indeed, horses can adapt (habituate) to stimuli in their environment through repetitive exposure (McLean & Christensen, 2017). If performed in similar environment and conditions, veterinary procedures such as rectal examination may therefore – through the process of habituation – not elicit significant stress responses in the teaching horses (Berghold et al., 2007). Within each type of practical class in the present study, the time the horses were manipulated and the number of students per horse were highly repeatable, and behavioural or physiological stress responses of the horses were not observed.

The only brief and minor increase in HR was measured when the horses were brought from their usual holding yard into stocks for the practical class, and a similar tendency was observed at the end of the class. In addition to the effect of walking on heart rate, this change in HR may reflect a change in the environment or an anticipatory response of the horses to a forthcoming situation, as suggested by Peters et al. (2012). This was further confirmed by the results of a study of van Vollenhoven et al. (2017), which suggested that an unexpected time in stocks after the rectal examination compared to usual management practices elicited equal or greater increase in the horses' HR compared to the rectal examination procedure itself.

Further evidence for the predictability and repeatability of events was highlighted in the horses' behaviour during the class. Behaviours such as pawing, tail swishing, vocalisation or head movements were rarely observed, which suggests a lack of behavioural stress responses during the class (Young et al., 2012). Compared to no interaction, the proportion of time eating hay was lower and horses showed a greater frequency of ears oriented backwards during an interaction with veterinary students. This likely search of environmental (e.g. visual and/or auditory) cues

during an interaction with students placed behind them may reflect awareness of students approaching and interacting with them.

Contrary to a recent investigation of mares' responses to a standardised transrectal palpation of the reproductive tract procedure (van Vollenhoven et al., 2017), the present study was conducted in real-life conditions of a veterinary practical class. The behavioural and physiological responses of both teaching mares and geldings were therefore evaluated. Despite the presence of several students in the class and of students' various levels of experience and confidence, activities during the three types of practical classes investigated were not associated with increased stress responses of the horses. These results suggest that these teaching horses seem accustomed to being used in sessions with unfamiliar students.

Each horse had been pre-selected and allocated to a herd based on its perceived tolerance of the teaching procedures. Failure to fit in with the teaching activities would result in removal of horses from the teaching herds. There were very few changes in the composition of the teaching herds during the last 10 years (Guinnefollau et al., 2020). The long-term tenure of the horses and the stability of the herds supports the hypothesis that the horses have habituated to the teaching activities and environment. This may explain the lack of stress responses observed in this population of horses during classes. The horses were also always used in their same herds, which probably limited any stressor caused by social isolation (Reid et al., 2017), and were provided with hay, which can be used both as an occupation and distraction (Doherty et al., 2017).

Although necessary to consider, the frequency of use and type of procedure may not be the sole parameters to include in an ethical evaluation of the teaching horses' use. Given the different types of practical classes, combining these parameters with others, such as the duration of the class or the number of students interacting with each horse, may be of importance to assess the horses' responses to their use. Although the horses of the present study were involved in practical teaching classes of different type, duration, number of students, etc. (Guinnefollau et al., 2020), limited stress responses of the horses were reported. Animal Ethics Committees may therefore require having these data available to include in their ethical evaluation.

In this study, horse age was not controlled for, however this population had a very narrow distribution of age. Similarly, controlling for gender in the analysis was not possible due to two types of classes using single-sex herds. The absence of a HRV assessment and the investigation of a limited number of behavioural indicators may appear as a limitation to this study. However, their value may be questionable as the behavioural data and low and consistent heart rates reported here suggest that the teaching activities investigated do not seem to create any additional stressor to these horses.

CONCLUSION

This study reported a low and consistent heart rate of the teaching horses across and during the practical classes. The behavioural assessments indicated limited stress responses and suggest that this population of teaching horses may be habituated to both their use and environment for teaching. These findings of limited, or no quantifiable stress, support the use of horses within practical teaching classes to enhance the education of Veterinary and Animal Science students.

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CHAPTER 7. GENERAL DISCUSSION

STUDENTS' KNOWLEDGE AND SKILLS DURING THE DEGREE

Chapter 2 revealed 1st-year students' relatively limited knowledge of equine behaviour, understanding of learning theory and self-assessed equine handling skills. Given the complexity of equine behaviour and the fact that horses learn from each experience (McCall, 1990; McGreevy et al., 2018), this lack of competency may impact on the student-horse interaction, and subsequently influence both students' safety and horse welfare during practical teaching classes. Early exposure to animals was reported in Chapter 2 as a significant factor for students to correctly interpret a horse's behaviour. However, and as indicated in previous studies, 65% of first-year veterinary students had an urban background, compared to 35% for rural (Chapman et al., 2007; Gronqvist et al., 2017; Klupiec et al., 2014; Wake et al., 2006). Although most students had owned a pet, the presence of large animals and horses on their family's property was less frequent and reflected students' level of confidence and experience with these animals. To improve students' interactions with horses, exposure to horses and teaching of basic handling skills therefore seems essential. Evidence for this is the self-reported increased equine handling competency and knowledge of equine behaviour of 4th-year veterinary students, which highlight the significant benefit of combined theoretical and practical teaching during the programme. Nevertheless, it should be noted that the two populations of students were different and therefore the study presented in Chapter 2 did not evaluate students' progression throughout the programme. Therefore, there may be an opportunity to monitor the same cohort of students before and after a year in the programme, or from start to finish through a prospective study. Within the programme, research such as this would be valuable as part of internal auditing of efficacy of teaching. To avoid an over- or under- assessment of their skills by the students, it is also acknowledged that an objective, rather than perceived and self-assessed, evaluation of students' skills and knowledge is required.

Assessing the level of understanding equestrians and veterinarians have of the concept and terminology of learning theory has recently been the focus of several studies. These studies reported a frequent confusion between the terms positive/negative punishment/reinforcement when equestrians or veterinarians were asked to define them using their own words (Brown & Connor, 2017; Warren-Smith & McGreevy, 2015). However, whether or not a definition is formulated correctly does not necessarily reflect whether or not principles are correctly applied in practice. Therefore, a change of the terms, or the provision of a clear definition of terms, may have to be considered for further studies. Indeed, there is no data reported within the literature to indicate the association between the correct application in practice of the learning theory methods and the theoretical understanding of the said methods. In Chapter 2, an attempt to overcome this misunderstanding of the terms was made by providing examples of practical scenarios to students. However, an objective evaluation of the learning theory principles used in practice by students or professionals would provide more robust information about their handling skills.

A POTENTIAL TO INCREASE THE USE OF THE HORSES FOR TEACHING?

In Chapter 3, a relatively low use of Massey University's population of teaching horses was reported for practical teaching classes, compared to other populations of working horses (Bolwell et al., 2015; Lonnell et al., 2014; Lonnell et al., 2012; Munsters et al., 2013; Rogers et al., 2007; Sloet van Oldruitenborgh-Oosterbaan et al., 2006; Verhaar et al., 2014; Walters et al., 2008). The three herds of horses were managed at pasture year-round and presented similar time budgets and social dynamics (Chapter 4) as those reported for free-ranging populations (Boyd et al., 1988; Duncan, 1980; Hauschildt & Gerken, 2015; Heitor et al., 2006). Keeping horses in groups allows these social animals to express natural and social behaviours (Yarnell et al., 2015) and is associated with "greater opportunities to experience positive welfare states" (Mellor, 2016).

A positive horse-human relationship refers to the reaction of the horse to humans and to the human's perception of horses and has been associated with better horse welfare (Hausberger et al., 2008). Horses in the present study were easy to approach, despite the absence of restraint and

possible voluntary avoidance (Chapter 5). There was no difference in the horses' responses between herds. Given that each herd was used for specific, and sometimes quite disparate, types of practical classes (Chapter 3), this suggests that the type of use for teaching did not influence the human-horse relationship. In addition, the horses' heart rates remained low and consistent during practical teaching classes, and behavioural indicators of stress (Young et al., 2012) were rarely (low or medium stress) or never (high stress) observed (Chapter 6). Based on the scale developed by Young and others (2012), the horses displayed behaviours indicative of no or low stress level during the classes. These low behavioural and physiological stress responses reported suggest that these teaching horses may be habituated to their use (teaching activities and environment).

In New Zealand, when using live animals for the purposes of research, testing or teaching, each procedure must be declared and approved by an animal ethics committee (AEC), which evaluates the severity of the procedure, the steps taken to minimise the severity, and the benefit of the procedure. As indicated by Mellor and Reid (1994), animal welfare in a research or teaching environment should focus on how much the welfare of the animals may be compromised, instead of how good it may be. Despite the necessity for additional robust descriptors than the ones used in this thesis to evaluate the horses' use for teaching and the welfare cost to the horses, the present work was a first step in the assessment of teaching horses' behavioural responses to their use. Based on the indicators used in the present work and on the evaluation of the horses' use for teaching, this population of teaching horses is likely to experience limited physical, physiological and behavioural stress. Therefore, the impact of practical teaching classes on these teaching horses is likely to be low and of little compromise to their welfare. In this situation, there may be a potential to increase the use of this population of horses for teaching in order to enhance veterinary students' learning experience, while still maintaining good animal welfare standards and low welfare compromise.

A greater exposure to horses during practical teaching classes is indeed likely to have a positive effect on students' understanding of equine behaviour and on their ability to handle

horses safely and efficiently. While acknowledging the time and budget constraints of the veterinary programme (Cawdell-Smith et al., 2007), increasing students' experience and exposure to horses during their study degree may be of great value to new graduates in order to remain safe when they practice. Through active learning in practical teaching classes, students are likely to better retain, retrieve and apply their knowledge (Balcombe, 2000). Providing more opportunities to students to interact with horses during practical classes, may also help to overcome the existing discrepancies (in both type of enterprise and quality) between external equine placements (Gronqvist et al., 2016; White & Chapman, 2007).

RECOMMENDATIONS

To achieve optimal teaching outcomes, a balance, i.e. trade-off, must be determined between the use of the horses for teaching and students' learning experience. This trade-off requires the identification of the least amount of use for the horses required to provide students with sufficient exposure from the start of their programme.

First-year students' levels of equine experience and confidence were low, and they had limited knowledge of equine behaviour. Given the background of most students when they enrol, the provision of online or digital material prior to any equine practical teaching class may be beneficial. The recommendation of using online resources prior to engaging in hands-on experiences during classes is supported by Old and Spencer (2011). These authors reported increased knowledge of animal behaviour after learning activities, and importantly, these activities eliminated the difference between students with or without qualification and perceived level of knowledge of ethology. By providing digital learning materials specific to equine behaviour and learning, institutions could aim to homogenise first-year students' knowledge of equine behaviour prior to any practical class and interaction with live horses. Increasing students' knowledge and confidence through formal opportunities such as these may be beneficial to create safe and positive student-horse interactions during practical classes. In addition, informal opportunities such as external equine placements, should be offered more frequently to students

to increase their exposure to horses and subsequently improve the student-horse interactions, through increased equine behaviour knowledge and handling confidence.

During the equine and veterinary science degree programmes, students may benefit from increased exposure to horses. Indeed, 4th-year veterinary students indicated that they still had limited equine handling skills competency, equine behaviour knowledge and confidence around horses (Chapter 2). Given that the said handling skills are basic and necessary to master for students' safety, this is especially concerning from a health and safety point of view. In line with the limited welfare compromise reported for the teaching horses, a suggestion to increase the horses' use may therefore be proposed. Results of Chapter 5 suggest any increased use of the horses may need to be spread more equally throughout the semester for increased regularity. The impact of teaching on the horses can only be addressed if detailed information about their use is available. In the present study, these precise data regarding the use of each horse (type of practical teaching class, procedure performed, medication, sedation, duration of the class) were extremely valuable. These data allowed the difference of use for teaching (type and frequency) between horses and the impact of this use on the human-horse relationship to be determined. Therefore, for institutions using horses for educational purposes where these data are not currently recorded, it is recommended to consider having these data available for Animal Ethics Committees to evaluate the ethical impact of use for teaching.

The consistency and repeatability of the horses' use might be a more significant factor in determining welfare outcomes for teaching horses than their frequency or type of use. This may be achieved through standardisation of some of the classes' characteristics, such as the location of the horse or routine of the practical class, which were identified in Chapter 3 as having a potential for variation between two classes of the same type (e.g. animal handling). Chapter 6 reported limited adverse behavioural and physiological responses of the horses to predictable and repeatable rectal examination classes, further supporting the benefits of consistent events. Through the process of habituation, horses' stress responses to repeatable classes may therefore be minimised. This research also demonstrated that pre-selecting horses based on their perceived

tolerance of the teaching procedures is valuable in terms of animal welfare outcomes. This pre-selection and the management of the horses in herds of long-term stability further support the hypothesis that this population of horses have habituated to their use. Considering these results, educational institutions should aim to pre-select horses for teaching and use them in specific and standardised class events/activities.

Improving the student-horse interaction during a class is the last parameter which warrants consideration to increase the predictability of a practical teaching class. For classes where the horses are not restrained in stocks, Chapter 5 identified the best approach to elicit the least avoidance or negative behavioural responses from the horses: a slow pace, a gaze directed at the horse's shoulder and arms straight along the body. By teaching these skills (i.e. use of consistent, adequate and correctly-timed signals) and equine behaviour early in the equine and veterinary science curricula, students may therefore be better equipped to handle horses with increased consistency, improving safety and minimising stress responses of the horses.

FUTURE WORK

During practical teaching classes, interactions between students and horses are inevitable. Because many first-year students are unskilled horse handlers, these student-horse interactions are likely to generate negative affects to the teaching horses (Mellor et al., 2020). The impact of these interactions on the horses was investigated in Chapter 6. The horses' heart rate was not significantly higher during a student interaction in rectal examination classes or in the animal handling classes. Nevertheless, future studies may wish to evaluate horse behaviour in other practical classes such as animal handling, where the horses are exposed to more variable interactions (less restraint of the horse, more naïve students, more students per horse, less consistent routine) with students (Chapter 3). Although behavioural assessments (type, frequency, duration) are established indicators of stress in horses, the ethograms used need to be carefully defined to include behavioural parameters that are adapted to each type of situation (type of restraint, locomotion in lameness investigation class, amount of student interaction).

Evaluating horse behaviour in classes such as animal handling may provide an opportunity to investigate the impact of the student-horse interaction. Indeed, positive human attributes and behaviours (e.g. attitude, voice, aptitude and handling skills) are factors that may positively influence the teaching horses' experiences (Mellor et al., 2020) and are therefore worth investigating. Similar research on the behavioural and physiological responses of the horses are required for the other types of equine practical classes (e.g. dental training, clinical examination).

Appropriate criteria and metrics are needed to investigate the impact of teaching on the horses' behaviour, physiology and subsequent welfare. The indicators of stress chosen need to be non-invasive, sensitive enough, not interfere with the teaching activities and adequate to be used during a practical teaching class, which involves the presence of students, staff members and many environmental stimuli. For a short-term stress assessment, investigating levels of endocrinological parameters (i.e. salivary cortisol) or eye temperature may be of interest (see König von Borstel et al. (2017) for a review of indicators of stress in horses). The present work assessed teaching horses' responses to their use over a semester of practical teaching. Therefore, the long-term impact of the use for teaching on the horses requires additional investigation. This may be addressed through the evaluation of the horses' behaviour and mental state across a year of use, and include the use of fear or preference tests (AWIN, 2015; König von Borstel et al., 2017). However, the number of studies in the literature aiming to assess horses' affective states emphasises its complexity and challenge; and welfare indicators need to be valid, reliable and feasible.

How to efficiently teach equine handling skills to students to improve their confidence and predictability when interacting with a horse, should be investigated. Although these studies reported limited stress responses of the horses to their use for teaching, identifying the most efficient practice would optimise the horses' use in return. Optimising students' interaction time with the horses during practical classes may also be a possibility for institutions with limited economic resources or for which increasing the horses' use may not be an option. This would have to be done in the first few years of the undergraduate equine practical programme, when

students' confidence, experience and knowledge are the lowest. Lastly, the return on investment of an increased use of the horses for practical teaching on students' knowledge of equine behaviour and equine handling skills competency still warrants investigation. To provide optimal (i.e. a positive and safe experience for both students and horses) teaching, there is a need to evaluate how much equine exposure is required to improve students' competency in the duration of their degree. However, these metrics are currently lacking and prevent a better optimisation of teaching for both students and horses.

CONCLUSION

The research presented in this thesis provided what is believed to be the first published description and evaluation of the use for teaching of horses in the undergraduate equine and veterinary science environment. The presence of horses in equine practical teaching classes is fundamental to provide students with sufficient exposure to remain safe when they graduate and are required to handle horses that have compromised health and may be in pain. This thesis demonstrated that the current management and use of teaching horses at Massey University, New Zealand elicits limited stress and changes in behaviour from the horses during and outside practical classes. In order to improve students' safety during (i.e. in equine practical classes and placements) and after the programme, and to enhance their learning experience, these findings demonstrate the opportunity to increase the use of this population of teaching horses. This may be done after further investigation of their use and its impact of the horses, and after further research on the most efficient equine teaching practice.

The results of this thesis are particularly relevant to the studied teaching horse population. However, there are also highly relevant to any other institution using horses for educational purposes and to animal ethics committees, which may use these results as additional indicators to guide their decisions regarding the use of horses for equine and veterinary science teaching purposes.

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APPENDICES

APPENDIX A: QUESTIONNAIRE PROVIDED TO FIRST- AND FOURTH-YEAR VETERINARY STUDENTS



SCHOOL OF
VETERINARY
SCIENCE

Interpretation of horse behaviour, evolution of animal handling skills and background of veterinary students

General respondent information

| | |
|--------------------|--|
| Student ID number: | |
|--------------------|--|

What gender do you identify with?

| | | | |
|-------------------------------|---------------------------------|--------------------------------|---|
| <input type="checkbox"/> Male | <input type="checkbox"/> Female | <input type="checkbox"/> Other | <input type="checkbox"/> Prefer not to disclose |
|-------------------------------|---------------------------------|--------------------------------|---|

Growing up, where did you spend the majority of your time?

| | |
|-------------------------------------|-------------------------------------|
| <input type="checkbox"/> Urban area | <input type="checkbox"/> Rural area |
|-------------------------------------|-------------------------------------|

Growing up, did you spend the majority of your time on a property where large animals were kept?

| | Yes | Yes, and you were actively involved in the care and management of: | No |
|---------------|--------------------------|--|--------------------------|
| Horses | <input type="checkbox"/> | <input type="checkbox"/> | <input type="checkbox"/> |
| Sheep | <input type="checkbox"/> | <input type="checkbox"/> | <input type="checkbox"/> |
| Alpacas/Lamas | <input type="checkbox"/> | <input type="checkbox"/> | <input type="checkbox"/> |
| Cows | <input type="checkbox"/> | <input type="checkbox"/> | <input type="checkbox"/> |
| Goats | <input type="checkbox"/> | <input type="checkbox"/> | <input type="checkbox"/> |
| Pigs | <input type="checkbox"/> | <input type="checkbox"/> | <input type="checkbox"/> |
| Deer | <input type="checkbox"/> | <input type="checkbox"/> | <input type="checkbox"/> |

Animal experience

Growing up, did you spend the majority of your time on a property with pet(s)?

| |
|--|
| <input type="checkbox"/> Yes, please list the type(s): |
| <input type="checkbox"/> No |

Have you had a previous work experience with animals (paid, volunteer)?

Small animals = dogs, cats, rabbits, ferrets, rodents, fish, reptiles, birds

Large animals = horses, sheep, cows, deer, goats, pigs, alpacas/lamas

| |
|---|
| <input type="checkbox"/> Yes, please specify: <input type="checkbox"/> Small animals, please list the type(s) of animals: <input type="checkbox"/> Large animals, please list the type(s) of animals: |
| <input type="checkbox"/> No |

Please rate your confidence around both small and large animals:

| | Not at all confident | Somewhat confident | Confident | Very confident | Don't know |
|----------------|--------------------------|--------------------------|--------------------------|--------------------------|--------------------------|
| Small animals: | <input type="checkbox"/> | <input type="checkbox"/> | <input type="checkbox"/> | <input type="checkbox"/> | <input type="checkbox"/> |
| Large animals: | <input type="checkbox"/> | <input type="checkbox"/> | <input type="checkbox"/> | <input type="checkbox"/> | <input type="checkbox"/> |

Between school and entry to this veterinary programme, have you done any formal qualification related to animals?

| |
|---|
| <input type="checkbox"/> Yes, I have completed at least one qualification - <i>please specify the area:</i> |
| <input type="checkbox"/> Yes, but my qualification(s) is(are) incomplete - <i>please specify the area:</i> |
| <input type="checkbox"/> No |

Horse experience

Have you ever had contact with horses?

| |
|------------------------------|
| <input type="checkbox"/> Yes |
| <input type="checkbox"/> No |

Prior to today, have you been actively involved in the care and management of horses?

| |
|--|
| <input type="checkbox"/> Yes - <i>please give a time estimation:</i> |
| <input type="checkbox"/> No |

Please rate your confidence around horses:

| | | | | |
|--------------------------|--------------------------|--------------------------|--------------------------|--------------------------|
| Not confident at all | Somewhat confident | Confident | Very confident | Don't know |
| <input type="checkbox"/> | <input type="checkbox"/> | <input type="checkbox"/> | <input type="checkbox"/> | <input type="checkbox"/> |

Please rate your level of experience with horses:

| | | | | |
|--------------------------|--------------------------|--------------------------|--------------------------|--------------------------|
| Not experienced at all | Somewhat experienced | Experienced | Very experienced | Don't know |
| <input type="checkbox"/> | <input type="checkbox"/> | <input type="checkbox"/> | <input type="checkbox"/> | <input type="checkbox"/> |

Have you ever ridden a horse?

| |
|--|
| <input type="checkbox"/> Yes, <i>please rate your riding ability:</i> <ul style="list-style-type: none"> <input type="checkbox"/> Beginner-leading lesson <input type="checkbox"/> Trekking (e.g. organised group rides on hired horses, casual one-off paid experiences) <input type="checkbox"/> Recreational (e.g. hobby riding) <input type="checkbox"/> Competitive or Show-ring (e.g. pony club, jumping, eventing, racing, dressage, endurance) <input type="checkbox"/> Professional (e.g. instructor, trainer) |
| <input type="checkbox"/> No |

Handling skills

Please rate your horse handling skills:

| | | | | | |
|-------------------------------|-------------------------------------|-------------------------------|------------------------------------|------------------------------------|-------------------------------------|
| <input type="checkbox"/> Poor | <input type="checkbox"/> Sufficient | <input type="checkbox"/> Good | <input type="checkbox"/> Very good | <input type="checkbox"/> Excellent | <input type="checkbox"/> Don't know |
|-------------------------------|-------------------------------------|-------------------------------|------------------------------------|------------------------------------|-------------------------------------|

Please rate your ability to:

| | Put a head collar on a horse: | Lead a horse: | Groom a horse: |
|------------|-------------------------------|--------------------------|--------------------------|
| Never done | <input type="checkbox"/> | <input type="checkbox"/> | <input type="checkbox"/> |
| Poor | <input type="checkbox"/> | <input type="checkbox"/> | <input type="checkbox"/> |
| Sufficient | <input type="checkbox"/> | <input type="checkbox"/> | <input type="checkbox"/> |
| Good | <input type="checkbox"/> | <input type="checkbox"/> | <input type="checkbox"/> |
| Very good | <input type="checkbox"/> | <input type="checkbox"/> | <input type="checkbox"/> |
| Excellent | <input type="checkbox"/> | <input type="checkbox"/> | <input type="checkbox"/> |

| | Lift front foot of a horse: | Lift hind foot of a horse: | Put a rug on a horse: |
|------------|-----------------------------|----------------------------|--------------------------|
| Never done | <input type="checkbox"/> | <input type="checkbox"/> | <input type="checkbox"/> |
| Poor | <input type="checkbox"/> | <input type="checkbox"/> | <input type="checkbox"/> |
| Sufficient | <input type="checkbox"/> | <input type="checkbox"/> | <input type="checkbox"/> |
| Good | <input type="checkbox"/> | <input type="checkbox"/> | <input type="checkbox"/> |
| Very good | <input type="checkbox"/> | <input type="checkbox"/> | <input type="checkbox"/> |
| Excellent | <input type="checkbox"/> | <input type="checkbox"/> | <input type="checkbox"/> |

Track choice

Which type of practice do you most likely see yourself in?

| | | | |
|--|---|---|---|
| <input type="checkbox"/> Small animal | <input type="checkbox"/> Equine | <input type="checkbox"/> Production animal | <input type="checkbox"/> Small animal + Production animal |
| <input type="checkbox"/> Small animal + Equine | <input type="checkbox"/> Special track (zoo animals/research) | <input type="checkbox"/> Production animal + Equine | <input type="checkbox"/> Don't know |

Animal behaviour



Picture obtained from:

https://sielearning.tafensw.edu.au/toolboxes/toolbox801/site/harness/1_catch_restrain/2_approach/10_body_language/1_act_photos/activity_nf.htm

Please tick **the term that best describes** the horse in picture:

| | Definition |
|---|--|
| <input type="checkbox"/> Aggressive | Hostile, defensive aggression, intention to harm |
| <input type="checkbox"/> Alarmed | Worried/tense, apprehensive, nervous, on guard against a possible threat |
| <input type="checkbox"/> Annoyed | Irritated, displeased, exasperated, bothered by something, upset, troubled |
| <input type="checkbox"/> Apathetic | Having or showing little or no emotion, disinterested, indifferent, unresponsive |
| <input type="checkbox"/> At ease | Calm, carefree, peaceful |
| <input type="checkbox"/> Curious | Inquisitive, desire to investigate |
| <input type="checkbox"/> Fearful | Afraid, hesitant, timid, not confident |
| <input type="checkbox"/> Friendly | Affectionate, kind, not hostile, receptive, confident |
| <input type="checkbox"/> Happy | Feeling, showing or expressing joy, pleased, playful, satisfied |
| <input type="checkbox"/> Look for contact | Actively looking for interaction, interested, eager to approach |
| <input type="checkbox"/> Relaxed | Not tense or rigid, easy-going, tranquil |
| <input type="checkbox"/> Pushy | Assertive or forceful |

Which body part(s) of the horse led you to this conclusion? *Please tick all*

| | |
|-------------------------------------|-----------------------------------|
| <input type="checkbox"/> Back | <input type="checkbox"/> Mouth |
| <input type="checkbox"/> Ears | <input type="checkbox"/> Neck |
| <input type="checkbox"/> Eye | <input type="checkbox"/> Nostrils |
| <input type="checkbox"/> Front legs | <input type="checkbox"/> Tail |
| <input type="checkbox"/> Hind legs | <input type="checkbox"/> Other |

Have you heard of learning theory before?

| |
|------------------------------|
| <input type="checkbox"/> Yes |
| <input type="checkbox"/> No |

Do you know what:

| | Yes | No |
|------------------------------|--------------------------|--------------------------|
| - positive reinforcement is? | <input type="checkbox"/> | <input type="checkbox"/> |
| - negative reinforcement is? | <input type="checkbox"/> | <input type="checkbox"/> |
| - positive punishment is? | <input type="checkbox"/> | <input type="checkbox"/> |
| - negative punishment is? | <input type="checkbox"/> | <input type="checkbox"/> |

Leg pressure is applied to the horse's sides; he moves forward and the pressure is released. This is an example of:

| | |
|---|---|
| <input type="checkbox"/> Positive reinforcement | <input type="checkbox"/> Negative reinforcement |
| <input type="checkbox"/> Positive punishment | <input type="checkbox"/> Negative punishment |
| <input type="checkbox"/> I don't know | |

The horse resists moving forward off the leg and is tapped with the crop. This is an example of:

| | |
|---|---|
| <input type="checkbox"/> Positive reinforcement | <input type="checkbox"/> Negative reinforcement |
| <input type="checkbox"/> Positive punishment | <input type="checkbox"/> Negative punishment |
| <input type="checkbox"/> I don't know | |

The horse smells treats in your pocket and noses you. You move away and the attention he is seeking is withdrawn. This is an example of:

| | |
|---|---|
| <input type="checkbox"/> Positive reinforcement | <input type="checkbox"/> Negative reinforcement |
| <input type="checkbox"/> Positive punishment | <input type="checkbox"/> Negative punishment |
| <input type="checkbox"/> I don't know | |

The horse stands quietly and receives a food reward. This is an example of:

| | |
|---|---|
| <input type="checkbox"/> Positive reinforcement | <input type="checkbox"/> Negative reinforcement |
| <input type="checkbox"/> Positive punishment | <input type="checkbox"/> Negative punishment |
| <input type="checkbox"/> I don't know | |

Thank you.

APPENDIX B: PERCENTAGE OF VETERINARY STUDENTS THAT SELECTED THE 12 PRE-SELECTED TERMS TO DESCRIBE THE BEHAVIOUR OF THE HORSE IN PICTURE, DEPENDING ON THE PRESENCE OR ABSENCE OF HORSES ON THEIR OWN PROPERTY

| Term selected | Horses on property: NO | Horses on property: YES |
|----------------------|-----------------------------------|------------------------------------|
| Aggressive (N) | 3 (2.54%) | 1 (1.69%) |
| Alarmed (N) | 18 (15.25%) | 3 (5.08%) |
| Annoyed* (N) | 63 (53.39%) | 48 (81.36%) |
| Apathetic (N) | 3 (2.54%) | 1 (1.69%) |
| At ease (P) | 4 (3.39%) | 1 (1.69%) |
| Curious (P) | 5 (4.24%) | 0 (0%) |
| Fearful (N) | 8 (6.78%) | 1 (1.69%) |
| Friendly (P) | 1 (0.85%) | 1 (1.69%) |
| Happy (P) | 0 (0%) | 0 (0%) |
| Look for contact (P) | 0 (0%) | 0 (0%) |
| Pushy (N) | 0 (0%) | 0 (0%) |
| Relaxed (P) | 13 (11.02%) | 3 (5.08%) |

* term evaluated to be the best to describe the horse's behaviour. N = negative term. P = positive term.

APPENDIX C: PERCENTAGE OF TIME OF EACH HERD ENGAGED IN FEEDING AND RESTING BEHAVIOUR ACTIVITIES PER TIME OF DAY

The table shows the median (interquartile range) percentage of time dedicated to the behavioural categories during each day of observation, by herd. Values given are for 7 (herd B), 9 (herd M) and 8 (herd T) horses. (a) refers to morning observations, (b) refers to midday observations, and (c) refers to afternoon observations.

| | Start | | During | | End | |
|---------------|-------------------------|--------------------------|-------------------|--------------------------|-------------------|--------------------------|
| Herd B | Feeding | Resting | Feeding | Resting | Feeding | Resting |
| Day 1 | 14 (7-22) (b) | 73 (71-82) (b) | 32 (22-46) (a) | 31 (21-32) (a) | 44 (41-44) (c) | 27 (22-38) (c) |
| Day 2 | 60 (49-61) (b) | 25 (18-29) (b) | 82 (80-85) (b) | 1 (0-2) ^(b) | 59 (29-76) (b) | 33 (10-57) (b) |
| Day 3 | 44 (39-48) (a) | 30 (23-46) (a) | 90 (78-91) (a) | 1 (0-5) ^(a) | 89 (86-91) (c) | 0 (0-0) ^(c) |
| Day 4 | 88 (80-90) (c) | 1 (0-8) ^(c) | 49 (44-51) (c) | 32 (24-36) (c) | 64 (27-77) (b) | 21 (10-64) (b) |
| Herd M | | | | | | |
| Day 1 | 25 (24-28) (a) | 54 (39-62) (a) | 77 (69-84) (b) | 6 (3-10) ^(b) | 65 (55-69) (a) | 9 (5-14) ^(a) |
| Day 2 | 1 (0-13) ^(a) | 84 (81-89) (a) | 72 (66-86) (a) | 19 (1-27) ^(a) | 58 (26-88) (b) | 17 (0-40) (b) |
| Day 3 | 89 (76-91) (b) | 3 (0-6) ^(b) | 74 (47-90) (c) | 19 (0-49) ^(c) | 45 (40-49) (a) | 30 (13-35) (a) |
| Day 4 | 78 (68-83) (c) | 6 (0-11) ^(c) | 91 (61-97) (c) | 0 (0-35) ^(c) | 55 (52-75) (b) | 11 (0-21) (b) |
| Herd T | | | | | | |
| Day 1 | 97 (95-99) (b) | 0 (0-0) ^(b) | 52 (47-70) (c) | 28 (4-34) ^(c) | 56 (53-58) (c) | 34 (30-38) (c) |
| Day 2 | 88 (73-96) (c) | 4 (0-12) ^(c) | 38 (16-49) (b) | 54 (47-80) (b) | 66 (64-66) (b) | 29 (29-31) (b) |
| Day 3 | 82 (55-89) (c) | 8 (2-35) ^(c) | 84 (76-89) (b) | 4 (2-8) ^(b) | 30 (25-35) (b) | 35 (29-42) (b) |
| Day 4 | 56 (43-72) (a) | 20 (7-38) ^(a) | 52 (45-72) (a) | 22 (13-37) (a) | 56 (52-67) (a) | 10 (8-17) ^(a) |

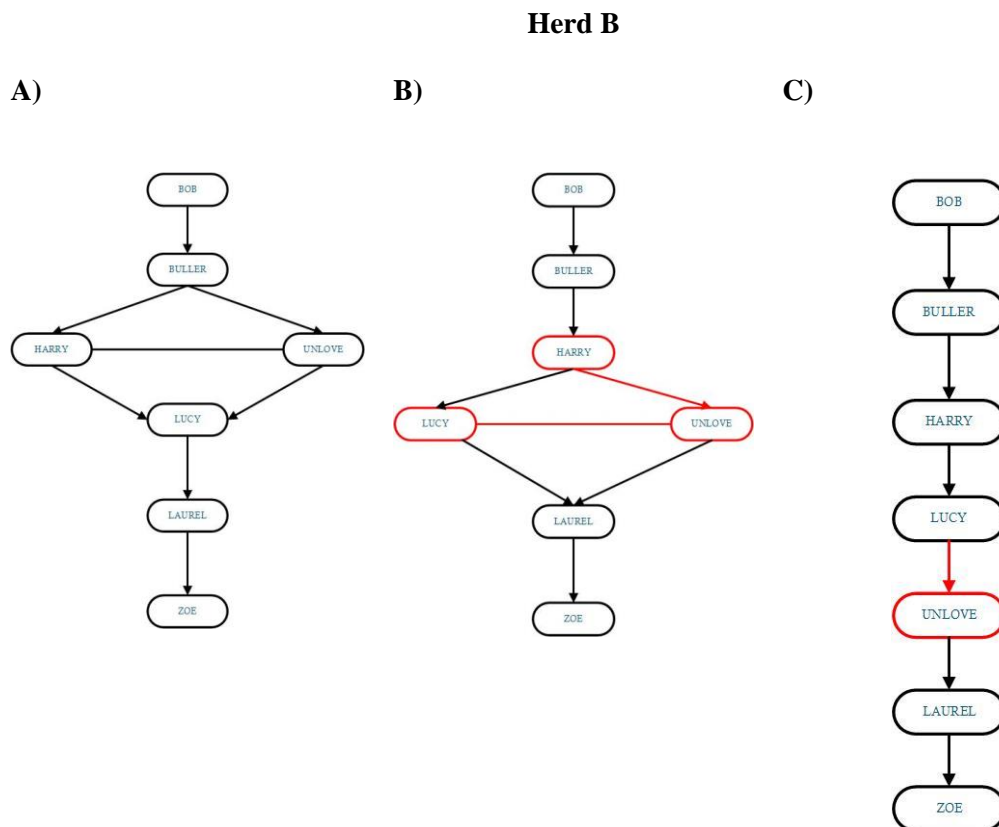
APPENDIX D: ESTIMATED NUMBER OF AGONISTIC AND SUBMISSIVE BEHAVIOURS PER HOUR AT EACH TIME POINT, EACH TIME OF DAY AND WITH OR WITHOUT ADDITIONAL PROVISION OF HAY

The table shows the median (interquartile range) number of social interactions per hour, by herd. Values given are for 7 (herd B), 9 (herd M) and 8 (herd T) horses.

| | Herd B | | Herd M | | Herd T | |
|----------------------|-----------|------------|-----------|------------|-----------|------------|
| | Agonistic | Submissive | Agonistic | Submissive | Agonistic | Submissive |
| Time point | | | | | | |
| Start | 1 (0-3) | 1 (0-3) | 4 (0-8) | 4 (1-10) | 1 (0-2) | 1 (0-3) |
| During | 4 (2-8) | 6 (2-11) | 1 (0-3) | 2 (0-6) | 1 (0-6) | 4 (1-7) |
| End | 2 (1-6) | 4 (0-10) | 2 (0-5) | 5 (0-9) | 3 (1-7) | 5 (2-11) |
| Time of day | | | | | | |
| Morning | 2 (0-7) | 3 (1-8) | 2 (0-6) | 3 (0-8) | 2 (0-4) | 3 (1-6) |
| Midday | 2 (1-5) | 4 (1-7) | 1 (0-3) | 2 (1-4) | 1 (0-4) | 2 (0-6) |
| Afternoon | 2 (1-3) | 1 (0-5) | 1 (0-3) | 1 (0-3) | 1 (0-3) | 1 (0-4) |
| Hay provision | | | | | | |
| Yes | 2 (0-4) | 3 (0-6) | 2 (0-5) | 2 (0-5) | 1 (0-3) | 1 (0-4) |
| No | 2 (1-6) | 4 (0-10) | 2 (0-5) | 5 (0-9) | 3 (1-7) | 5 (2-11) |

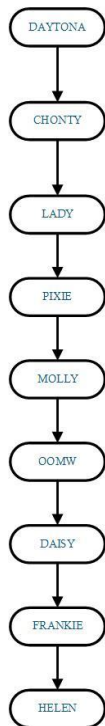
APPENDIX E: HIERARCHICAL RELATIONSHIPS WITHIN EACH HERD (HERD B, HERD M, HERD T) AT EACH STUDY TIME POINT (A = START, B = DURING, C = END)

The black arrows represent consistent bilateral dominance relationships between one time point and the previous one, while red arrows highlight a new bilateral dominance relationship. Within a herd, horses at the same horizontal level had the same rank index and, therefore, were attributed the same rank position.

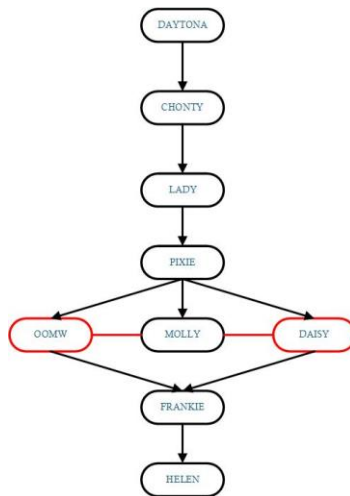


Herd M

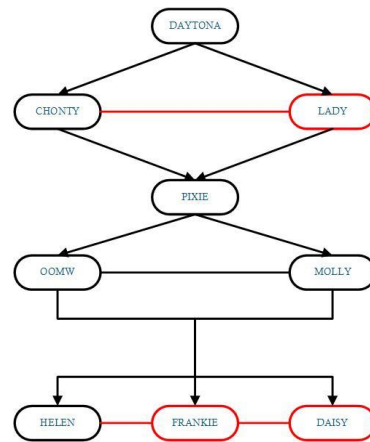
A)



B)

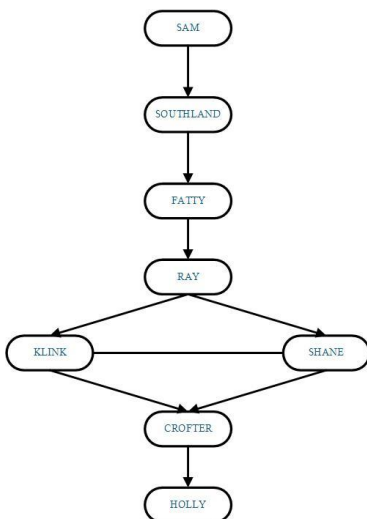


C)

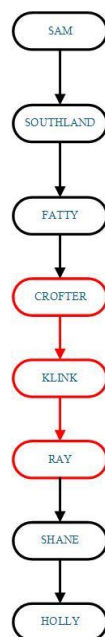


Herd T

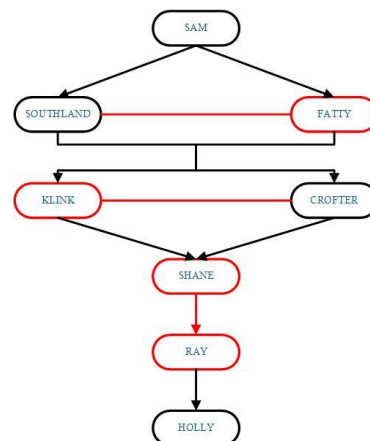
A)



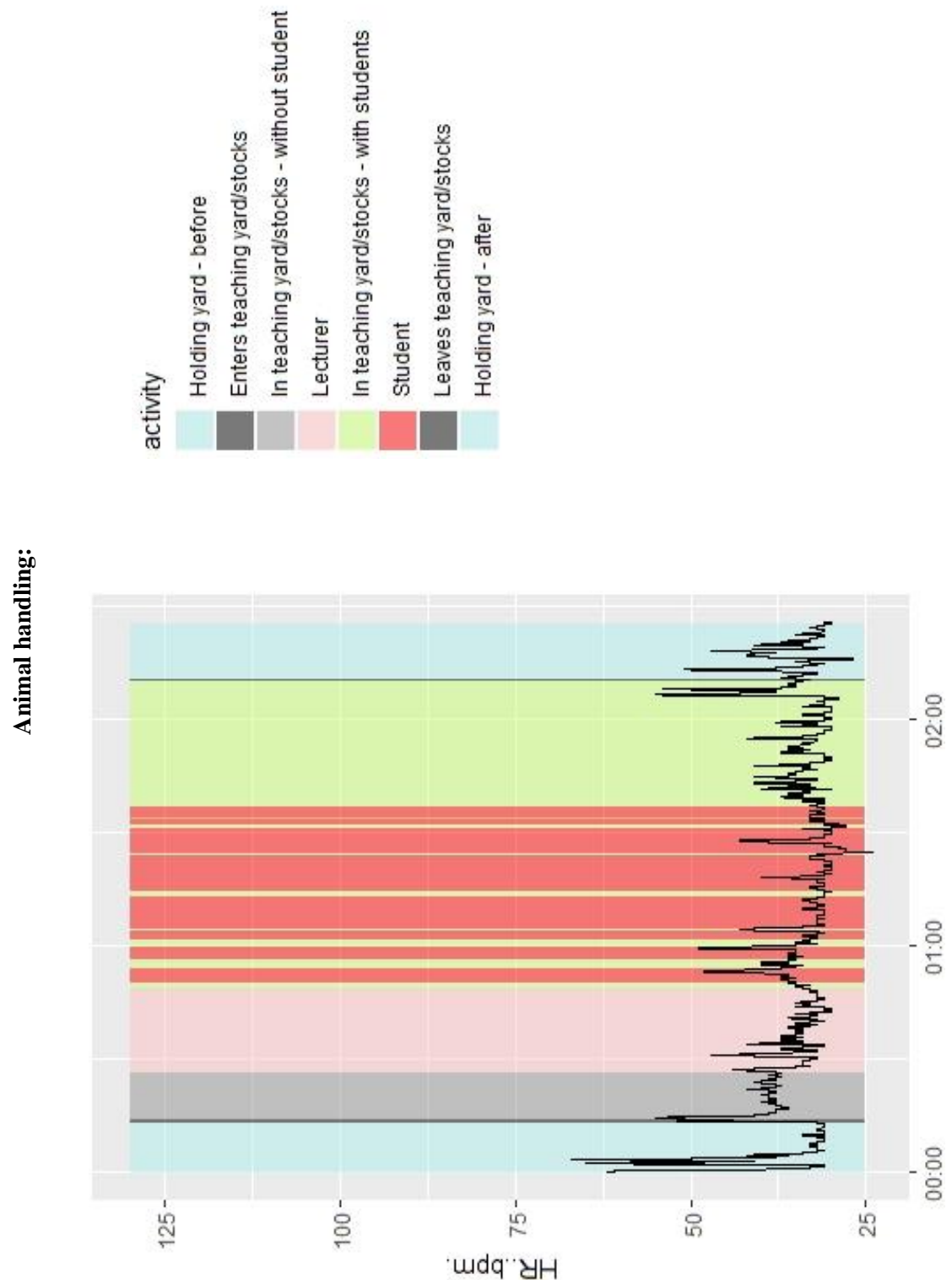
B)



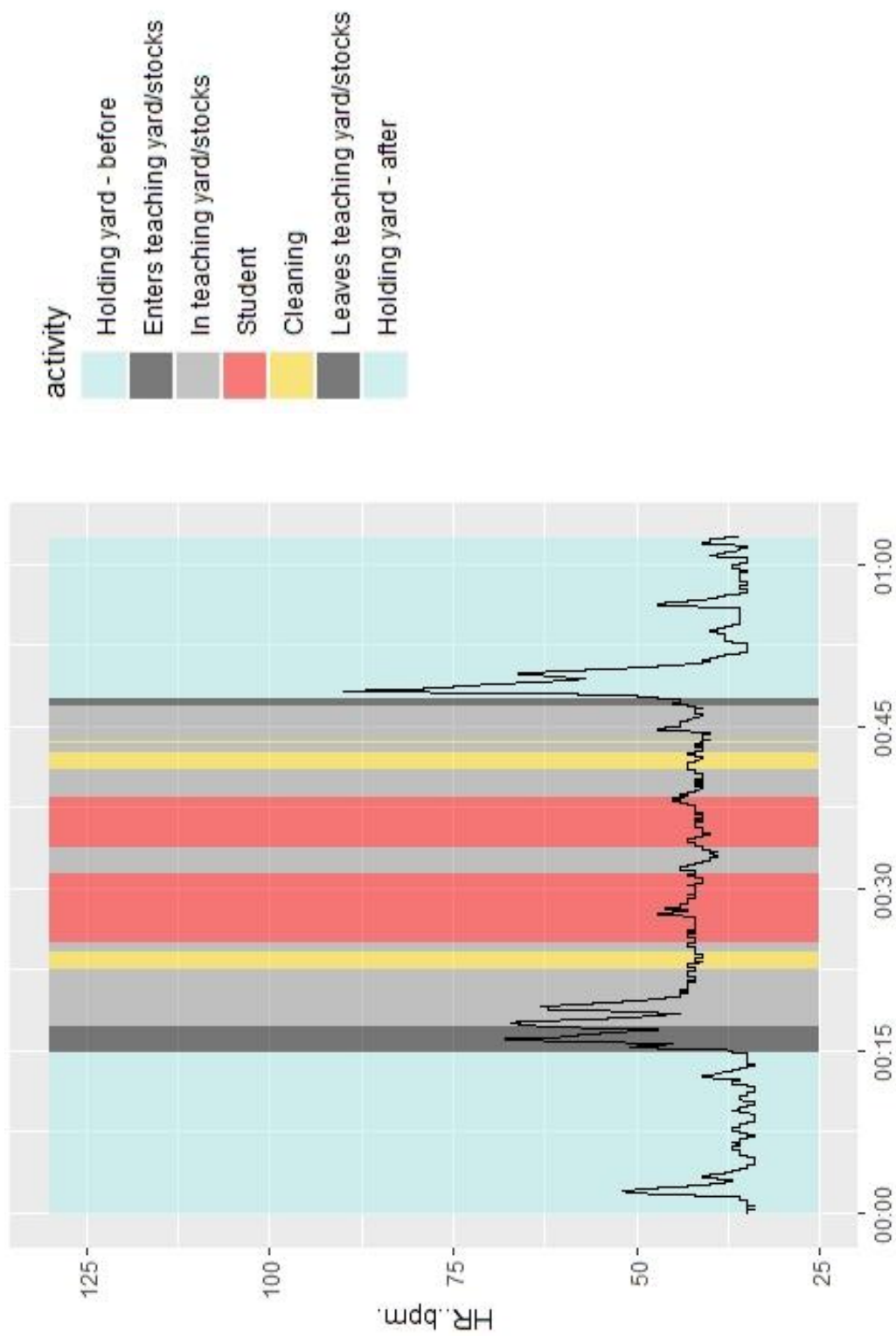
C)



APPENDIX F: EXAMPLES OF THE HORSES' HEART RATES BEFORE, DURING AND AFTER THE PRACTICAL TEACHING CLASSES



Medical rectal examination:



APPENDIX G: AUTHOR CONTRIBUTIONS

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| <p>Please select one of the following three options:</p> <p><input checked="" type="radio"/> The manuscript/published work is published or in press</p> <ul style="list-style-type: none"> Please provide the full reference of the Research Output: Guinnefollau, L., Gee, E. K., Bolwell, C. F., Norman, E. J. and Rogers, C. W. (2019) Benefits of Animal Exposure on Veterinary Students' Understanding of Equine Behaviour and Self-Assessed Equine Handling Skills, <i>Animals</i>, 9(9). <p><input type="radio"/> The manuscript is currently under review for publication – please indicate:</p> <ul style="list-style-type: none"> The name of the journal: The percentage of the manuscript/published work that was contributed by the candidate: Describe the contribution that the candidate has made to the manuscript/published work: <p><input type="radio"/> It is intended that the manuscript will be published, but it has not yet been submitted to a journal</p> | |
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| Date: | 02-Nov-2020 |
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| In which chapter is the manuscript /published work: CHAPTER 4 | |
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| Candidate's Signature: | Laureline Guinefollau <small>Digitally signed by Laureline Guinefollau DN: cn=Laureline Guinefollau, o=MZ, email=lg@massey.ac.nz Date: 2020.11.05 06:39:10 +1300</small> |
| Date: | 05-Nov-2020 |
| Primary Supervisor's Signature: | Charlotte Bolwell <small>Digitally signed by Charlotte Bolwell DN: cn=Charlotte Bolwell, o=MZ, email=cb@massey.ac.nz Date: 2020.11.20 09:51:13 +1300</small> |
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|---|------------------------|---|
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| <input type="radio"/> The manuscript is currently under review for publication – please indicate: <ul style="list-style-type: none"> The name of the journal: Applied Animal Behaviour Science The percentage of the manuscript/published work that was contributed by the candidate: Describe the contribution that the candidate has made to the manuscript/published work: | | |
| <input type="radio"/> It is intended that the manuscript will be published, but it has not yet been submitted to a journal | | |
| Candidate's Signature: | Laureline Guinefollau | <small>Digitally signed by Laureline Guinefollau DN: cn=Laureline Guinefollau, o=MZ, email=laureline.guinefollau@massey.ac.nz Date: 2020.11.05 09:41:12 +1300</small> |
| Date: | 10-Jun-2021 | |
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