

Copyright is owned by the Author of the thesis. Permission is given for a copy to be downloaded by an individual for the purpose of research and private study only. The thesis may not be reproduced elsewhere without the permission of the Author.

**The effect of feeding and management on faecal pH of
the New Zealand Thoroughbred racehorse**

**A thesis presented in partial fulfilment of
the requirement for the degree Master of
Applied Science in Equine Nutrition at
Massey University, Palmerston North, New
Zealand.**

Anita Williamson

2006

Abstract

Quantifying the risk factors for hindgut acidosis is the first step in understanding the problems of poor management and feeding practices of horses in race training. A non-invasive measure of hindgut acidosis can be obtained by measuring faecal pH (Davie *et al.*, 2000; Eastwood, 2002; Rowe *et al.*, 1995; Zeyner *et al.*, 2004; Zeyner *et al.*, 1992; Zeyner, 1993). In this study fourteen 3-year-old Thoroughbreds in regular race training, ten Thoroughbred yearlings aged 13-15 months and 140 Thoroughbred horses of mixed aged in race training were surveyed. Approximately $200\text{g}\pm 5\text{g}$ of faeces per horse was obtained from all faecal masses in the horse's stable or yard at the time of collection. Faecal pH was measured using a commercial pH meter.

The study was divided into three experiments; experiment one consisted of fourteen Thoroughbreds within the same racing stable and identified that subtle change in diet, management, and workload had no effect on mean faecal pH during an 83 day observation period. Experiment two, identified gender, between day or time of faecal collection and the amount of concentrate offered (kg), total feed weight and roughage to concentrate ratio of the diet had no significant effect on the faecal pH of ten Thoroughbred yearlings undergoing sales preparation. However there was considerable variation in faecal pH between horses.

Experiment three surveyed 140 Thoroughbred horses under the management of 16 racehorse trainers. Trainer age, number of years training horses, horse age, horse gender, weeks in race training or racing class had no effect on mean faecal pH. Acidic faecal pH (pH ≤ 6.32) was associated with small stables (1-12 horses). Trainers from small stables offered more concentrate feed than larger stables (13+ horses). Acidic pH was associated with trainers that offered grain as the only form of concentrated feed, or offered $\leq 2.25\text{kg}$ hay/day, and horses that were fast eaters. Horses that displayed stereotypic behaviours had more alkaline faecal pH than horses that never expresses stable vices (6.70 ± 0.35 vs. 6.43 ± 0.29). The total weight of concentrates offered, feed frequency, bedding type, exercise workload and the number of hour's horses were at pasture had no effect on mean faecal pH.

Acknowledgements

I would like to acknowledge my supervisors, Chris Rogers and Elwyn Firth for the opportunity to undertake this research project. The guidance, knowledge and suggestions they have put forward has helped considerably in the completion of this report.

I am most appreciative of the staff at Foxton Racecourse, especially Marty Johnston, for his contribution and commitment to those early morning faecal collections. I would like to thank all the Thoroughbred horse trainers and Sam Williams at Little Avondale Stud to allow me to question their management, feed practice and obtain horse faecal samples

Finally I would like to take this opportunity to acknowledge my family and friends for their kind support and encouragement, with a special thanks going out to Lindy, Gwyn, Sarah, Nikki, Fraser, Clem and Olivia for their constant support and commitment to listen.

Anita Williamson

Table of Contents

ABSTRACT	I
ACKNOWLEDGEMENTS	II
TABLE OF CONTENTS	III
LIST OF FIGURES	IV
LIST OF TABLES	V
LIST OF ABBREVIATIONS	VI
REVIEW OF LITERATURE	1
INTRODUCTION	1
THE NEW ZEALAND THOROUGHBRED INDUSTRY	3
ADAPTATION FROM NATURAL TO SPECIALISED DIET	5
THE STRUCTURE AND FUNCTION OF THE GASTROINTESTINAL TRACT	7
ROLE OF MICROBIAL FERMENTATION	11
WASTAGE IN THE THOROUGHBRED INDUSTRY	15
FEED REQUIREMENTS OF HORSES	19
MANAGEMENT AND TRAINING METHODS	30
FAECAL PH.....	36
FAECAL PH AS A TOOL TO DETERMINE HINDGUT ACIDOSIS	37
AIM OF THE STUDY	38
HYPOTHESES	38
MATERIALS AND METHODS	39
RESULTS	45
DISCUSSION	58
CONCLUSION	65
TERMINOLOGY	66
REFERENCES	70
APPENDIX	85

List of Figures

FIGURE 1. MEAN FAECAL PH OF 14 THOROUGHbred RACEHORSES OVER AN 84 DAYS OBSERVATION PERIOD. -----	45
FIGURE 2. MEAN FAECAL PH OF 14 THOROUGHbred RACEHORSES IN TRAINING OVER AN 84 DAY OBSERVATION PERIOD. -----	46
FIGURE 3. MEAN FAECAL PH OF 14 RACEHORSES IN TRAINING FOR 84 DAY (12 WEEKS) OBSERVATION PERIOD. -----	46
FIGURE 4. THE EFFECT OF EXERCISE INTENSITY ON MEAN FAECAL PH FOR 14 THOROUGHbred RACEHORSES FOR A 78 DAY OBSERVATION PERIOD. -----	48
FIGURE 5. THE EFFECT OF HORSE AGE ON MEAN FAECAL PH FOR 140 THOROUGHbred RACEHORSES SAMPLED FROM 16 TRAINING STABLES. -----	50
FIGURE 6. THE NUMBER OF CONCENTRATED MEALS PER DAY ON MEAN FAECAL PH OF 140 HORSES. -----	52
FIGURE 7. BOX AND WHISKER PLOT OF THE MEAN FAECAL PH AND THE DIFFERENT QUANTITIES OF CONCENTRATE OFFERED PER DAY. -----	53
FIGURE 8. BOX AND WHISKER PLOT OF THE MEAN FAECAL PH AND THE NUMBER OF WEEKS 140 RACEHORSES HAD BEEN IN RACE TRAINING. -----	56

List of Tables

TABLE 1. ENERGY AND CRUDE PROTEIN CONTENT OF COMMON NEW ZEALAND PASTURES ADOPTED FROM HUNT (1994).	6
TABLE 2. ENERGY REQUIREMENTS OF HORSES (NRC 1989).	23
TABLE 3. COMPARISON OF DIFFERENT MAINTENANCE ENERGY EQUATIONS FOR HORSES.	25
TABLE 4. COMPARISON BETWEEN KOHNKE <i>ET AL</i> (1999) AND NRC (1989) FOR ENERGY REQUIREMENTS FOR HORSES AT DIFFERENT WORK LOADS.	26
TABLE 5. COMPARISON OF DIFFERENT MAINTENANCE PROTEIN REQUIREMENTS.	28
TABLE 6. DESCRIPTION OF RACEHORSE GAITS AND VELOCITY (M/SEC).	31
TABLE 7 NEW ZEALAND GAIT DESCRIPTIONS IN VELOCITY (M/S)*	40
TABLE 8. NUTRIENT ANALYSIS OF DIET, NUMBER OF DAYS 14 HORSES WERE ON A DIFFERENT FEED PROGRAMME AND MEAN FAECAL pH A 84 DAY OBSERVATION PERIOD.	47
TABLE 9. SUMMARY DATA OF FEED OFFERED AND MEAN FAECAL pH FOR 10 HORSES WITH INDEPENDENT DIETS, SAME MANAGEMENT BUT NO EXERCISE.	49
TABLE 10. DESCRIPTIVE DATA OF THE RACING GRADES OF THE 140 THOROUGHBRED RACEHORSES SURVEYED.	50
TABLE 11. THE NUMBER OF HORSES AND NUMBER OF HORSES SAMPLED WITHIN THE SURVEYED TRAINING STABLES.	51
TABLE 12. MEAN FAECAL pH AND TOTAL NUMBER OF HORSES IN STABLES GROUPED INTO FOUR CATEGORIES.	51
TABLE 13. THE AMOUNT OF TIME 140 RACEHORSES WERE TURN OUT TO PASTURE ON MEAN FAECAL pH.	52
TABLE 14. A MATRIX OF SIGNIFICANT DIFFERENCE IS PRESENTED BELOW. *=(P<0.05)	53
TABLE 15. MEAN AMOUNT OF CONCENTRATE OFFERED TO HORSES IN RELATION TO CATEGORIZED STABLE NUMBER.	54
TABLE 16. THE EFFECT OF AMOUNT OF HAY OFFERED TO 140 HORSES ON MEAN FAECAL pH.	54
TABLE 17. TIME OF DAY HAY WAS OFFERED TO 140 THOROUGHBRED RACEHORSES.	55

List of Abbreviations

ADF – Acid-detergent fibre
ATP– Adenosine triphosphate
BW – Body weight in kg
Cal – calories
CP – Crude protein
DCP – Digestible crude protein
DE – Digestible energy
DM – Dry matter
FFA – Free fatty acids
GE – Gross energy
GDP– Gross Domestic Product
HCl – Hydrochloric acid
J – Joules
KJ – Kilojoule
Mcal – Megacalories
ME – Metabolisable energy
MJ – Megajoule
NZRB– New Zealand Racing Board
Net – Net energy
NZ – New Zealand
NSC – Non-structural carbohydrates
NDF – Neutral detergent fibre
NRC – National Research Council
R: C– Roughage to concentrate ratio
TAB– Totalisator agency board
TAGs– Triglycerides
VFA–Volatile fatty acids

Review of Literature

Introduction

New Zealand is internationally recognised as a producer of quality Thoroughbred horses. The Thoroughbred industry generated approximately \$NZ1,08 million dollars Gross Domestic Product (GDP) in 2004 (0.95% of New Zealand's total GDP) with 13,567 full time employment positions (NZRB, 2004; Perkins, 2001). However, there is limited information available to describe the training and management practices of New Zealand racing Thoroughbred horses.

Wastage is a term to describe the economic losses, poor performance, lost opportunity and the large turnover of horses exiting the racing industry, that may occur at any stage of a horse's racing career (Bailey, 1998; More, 1999). There may be many reasons why a racehorse fails to perform and exits from the race industry (Mohammed *et al.*, 1991). In recent years there has been extensive research conducted in Australia, United Kingdom, South East Asia, United States and New Zealand to identify possible risk factors for horses exiting the breeding and racing sectors (Bailey *et al.*, 1997, 1999; Jeffcott *et al.*, 1982; Mohammed *et al.*, 1991; Perkins, 1999; Perkins *et al.*, 2005a, 2005b).

Many studies have identified that a low roughage content, a higher proportion of concentrated readily digestible carbohydrates in the diet, restricted feeding times and confinement significantly increases the prevalence of colic, laminitis, gastric ulcers and stereotypic behaviours that may negatively effect the performance of a horse (Davie *et al.*, 2000; Frape, 1998; Hunt, 1994; Nicol, 1999; Pagan, 1997a). In studies of colic, it appears that even subtle changes in feed management can have a direct effect on the performance of a horse in race training (Cohen *et al.*, 1999; Hudson *et al.*, 2001). Yet, wastage due to feeding practice and inducement of metabolic acidosis from excessive fermentation at the hindgut causing loss of training and racing opportunities has not been researched to date.

Feed practices supplying excess soluble carbohydrates to the caecum and colon are known to produce fermentative products such as volatile fatty acids, lactic acid and endotoxins. Products of hindgut fermentation cause the pH in the caecum and colon to fall below 6.3 pH units (Johnston *et al.*, 1998; Willard *et al.*, 1977). Quantifying the risk factors for hindgut acidosis and being able to quantify sub-clinical cases is the first step in understanding the problems of poor management and feeding practices at an industry level. A non-invasive measure of hindgut acidosis can be obtained by measuring faecal pH as demonstrated by Zeyner *et al* (1992; 1993), Rowe *et al* (1994), Davie *et al* (2000) and Eastwood (2002).

Identifying hindgut acidosis by measuring faecal pH has never been conducted on racing Thoroughbred horses in New Zealand. The aim of this project was to identify management variables related to low faecal pH in racing Thoroughbred horses.

The New Zealand Thoroughbred Industry

The New Zealand Thoroughbred industry is internationally renowned for producing top quality bloodstock that is competitive in the world's strongest racing countries. Some believe the international success of New Zealand Thoroughbred horses is attributed to horses being developed on a pasture-based diet (Avery, 1997; Wallace, 1977).

In June 2004, the NZRB reported \$NZ 1, 08 million was generated by the New Zealand Thoroughbred industry. Over 2003/2004, the sale of young un-raced stock realised \$NZ 75 million. \$NZ 47.23 million was realised at the 2003 National Yearling Sale and a further \$NZ 27.8 million realised from "ready to run" or private sales with 50.8% of all purchases made by overseas buyers (NZRB, 2004). Australia was the largest export market followed by Singapore, Malaysia, Korea and Macau (NZRB, 2004).

The NZRB reported 23,000 people actively participate across the various sectors of the Thoroughbred industry, with 59% in full time employment (NZRB, 2004; Perkins, 2001). The actual number of employees involved in the Thoroughbred racing industry is masked by several factors:

1. Employees involved in more than one industry sector.
2. Significant industry reliance upon volunteers.
3. Employees may be remunerated in cash or "kind".
4. Or there is limited information available on the number of track riders, stable hands, handlers, jockeys, farriers, vets and horse transporters throughout the industry (NZRB, 2004), it is assumed there are large seasonal fluctuations.

The true size and scope of the Thoroughbred racing industry in New Zealand is deceiving. Fifty-two racecourses and sixty-nine racing clubs are responsible for annually organising 286 race meetings and 2,810 races, trials and jump outs, while competing for limited/diminishing financial support and patronage (NZRB, 2004). For example, the Auckland Racing Club organises 50 race meetings annually, reflecting the largest number of race meetings held in any one region (NZRB, 2004).

Auckland has the greatest per capita representation and thus commercial support, whereas regional clubs have to rely on fewer race days and mostly local support from sponsors and patrons. All race clubs receive funding from the NZRB and the Totalisator Agency Board (TAB) (NZRB, 2004), but to a level that does not reflect the contribution of the Thoroughbred industry to the New Zealand economy.

Studies indicate trends of a worldwide decline in actual racehorse numbers. Perkins (1999; 2001) reported in the 1991-1992 New Zealand racing season, 6,776 horses raced, but by 1999/2000 numbers decreased to 5,599. Two racing seasons later, (2002/2003) New Zealand statistics indicated 8900 horses were trained for an average of 8 months, but only 5,469 horses made it to the races (NZRB, 2004). The estimated cost of training a racehorse ranges from \$14,000 to \$20,000 per annum. This sum does not include purchase price, veterinary, transport, farrier, breaking-in, gear, registration, nomination and acceptance fees, agistment, business and marketing related costs (NZRB, 2004).

Adaptation from Natural to Specialised Diet

The horse (*Equus Caballus*) is a grazing/browsing monogastric, hindgut fermenting herbivore with a large body, long limbs, specialised teeth, single hoof and a highly organised social structure (Budiansky, 1997; Dossenbach and Dossenbach, 1988; Hoffman, 2003; Jacobs *et al.*, 1999). Horses do not secrete enzymes capable of digesting structural carbohydrates found in cell walls of forage, instead, the horse relies on a symbiotic relationship with micro-organisms for digestion and fermentation in the large intestine (hindgut) to provide the main source of energy (Budiansky, 1997; Duncan *et al.*, 1990; Hoffman, 2003; Langer, 1988).

Ruminants also use microbial fermentation to obtain nutrients from forage. Ruminants have a large capacity fore-stomach with a slower transit time for digesta, this allows more efficient microbial fermentation per nutrient extraction per weight of digesta (Budiansky, 1997).

To compete in a similar ecological niche the horse employs a low nutrient extraction strategy, by increasing feed intake rate and transit time of digesta through the digestive tract (Duncan *et al.*, 1990; Illius and Gordon, 1992; Jackson, 2003) For example, a free ranging horse will graze 10-16 hours per day with a digesta transit time of 21-48 hours. A similar sized ruminant will graze 7-8 hours with a 90 hour transit time if on the same high fibre pasture (Budiansky, 1997; Frape, 1998; Mayes and Duncan, 1986; McDonald *et al.*, 2002; Pearson *et al.*, 2001).

Because plant material contains large amounts of water, soluble proteins, lipids, sugars and structural carbohydrates but little starch, horses must spend 10-16 hours a day grazing in order to meet maintenance energy requirements (Frape, 1998). The New Zealand Thoroughbred breeding industry relies on pasture as the primary basis of feeding (Goold *et al.*, 1988; Kohnke *et al.*, 1999).

The main advantage of pasture is the perceived considerable reduction in overall feed costs, compared to supplementary feeding. Also it is believed horses with access to good pasture rarely suffer digestive or behavioural problems (Avery, 1997; Kohnke *et al.*, 1999).

In New Zealand, well-managed pastures provide most of the nutritional requirements for most horses (Hunt, 1994). Pasture mineral composition may vary regionally because it is influenced by composition, season, soil fertility, soil type and endophyte toxins (Bryden, 1995; Grace *et al.*, 2002; Hunt, 1994). Energy and crude protein content of common New Zealand pastures is presented in Table 1.

Table 1. Energy and crude protein content of common New Zealand pastures adopted from Hunt (1994).

Pasture Type	Energy content (MJDE/kg/DM)	Crude protein (% DM)
Ryegrass/white clover		
Autumn	10.8	25
Winter (leafy)	11.2	25
Spring (leafy)	12.0	22
Summer(leafy)	10.3	15
Summer (stalky)	8.0	10
Red clover		
pre bloom	11.0	23
full bloom	10.0	18
Lucerne		
immature	12.0	25
pre bloom	11.5	22
early bloom	11.0	20
mid bloom	10.5	16

MJDG= Megajoules digestible energy DM= Dry matter

In contrast, horses in intensive management generally have restricted activity, restricted diet, and are fed highly concentrated starch cereals and dried forage to meet energy requirements (Frape, 1998; Ralston, 1986). High fibre forage/hay diets fails to meet energy requirements for horses in intense work, thus the industry relies heavily on feeding large amounts of concentrated feeds (Avery, 1997; Frape, 1998; Hoskins and Gee, 2004; Huntington and Jenkinson, 2003; Kohnke *et al.*, 1999; Milton McClure, 1990).

There is limited information on the management and feeding practices of New Zealand racing Thoroughbred horses. However, anecdotally, horses are confined to a stable or small yard for the majority of the day and may have access to pasture for a only a short time (Huntington and Jenkinson, 2003; Kohnke *et al.*, 1999). A typical racehorse diet consists of a concentrated feed mix high in carbohydrates, only available at specific feeding times during the day. Hay is always available *ad lib* slab in hay manager (Huntington and Jenkinson, 2003; Kohnke *et al.*, 1999).

The Structure and Function of the Gastrointestinal Tract

Mouth

The mouth is suited for the prehension and ingestion of forage (Frape, 2004). The incisors shear, or nip off grass swards enabling close grazing on short pasture (Frape, 1998). The molars use a grinding action to reduce particle size (Budiansky, 1997; Jackson, 2003; Menard *et al.*, 2002). Such efficiency of food gathering and mastication allows equids a greater feed intake and transit time on a low quality diet (Mueller *et al.*, 1998). Mastication stimulates increased secretion of parotid saliva. Saliva lubricates food bolus, has small but not inconsequential α -amylase enzyme action, but significant concentration of bicarbonate (HCO_3^-) 50mEq/l, that assists gastric acid buffering in the stomach (Frape, 2004; Hoffman, 2003; Maskell and Johnson, 1993).

The oesophagus connects the mouth to the stomach (Titora and Grabowski, 2000). Secretion of mucus from cells lining the oesophagus further lubricates and aids the movement of food bolus from the mouth to the stomach (Maskell and Johnson, 1993; Titora and Grabowski, 2000).

Stomach

The volume of the stomach of an adult horse is small and limited, approximately 18 litres or 7.5-10 % of the total capacity of the digestive tract; compared to 30.2% and 61.3% for the small intestine and hindgut (Argenzio, 1993; Frape, 2004; Greet, 2000). The stomach is the first place of digestion and is divided into saccus caecus, oesophageal, cardia, fundus and pyloric regions. There is limited knowledge of the saccus caecus and oesophageal regions, although studies have shown these sites have a high pH environment, saliva flow, limited exposure to gastric acid and anaerobic gram-positive microbial colonization (Frape, 2004; Greet, 2000; McDonald *et al.*, 2002). The cardia region is small. Studies have identified this to be a major site of gastric ulceration (Greet, 2000; McDonald *et al.*, 2002). The fundus (or gastric gland) secretes gastric acids, hydrochloric acid (HCl), pepsinogen and lipase (Frape, 2004; Greet, 2000).

The pyloric region, even during fasting, continuously secretes gastric acid and gastric juice (consisting of intrinsic factor for vitamin B₁₂ absorption, somatostatin, water, inorganic salts, mucus and HCl) (Frape, 2004; Greet, 2000; McDonald *et al.*, 2002; Merrit, 1999).

The main action of gastric acids is to hydrolyse proteins and to a lesser extent, carbohydrates (Hoffman, 2003; McDonald *et al.*, 2002). A relatively small number of *Lactobacillus acidophilus* and *Streptococcus bovis* microbe species can exist in the fundic region of the stomach (10^8 - 10^9 bacteria/g digesta). Such microbes are responsible for the production of lactic acid and could consequently contribute to the decrease in the acidity from a pH of 5.4-5.8 in the fundus to 1.5-2.6 in the pyloric region of the stomach (de Fombelle *et al.*, 2003; Frape, 1998; Jackson, 2003; Merrit, 2003a). Water passes directly through the stomach avoiding dilution with gastric juices and is absorbed in the hindgut (Frape, 2004; Greet, 2000).

Digesta in the stomach is dependent on diet composition. For example, finely chopped, pelleted particles or grains pass rapidly through the stomach and enter the small intestine

with little digestion compared to particles that require extensive mastication such as high fibre/roughage hay (Frape, 2004). Roughage increases saliva secretion, creating a buffering and lubricating effect to digesta. Roughage also has a slower transit time through the digestive tract therefore a more efficient digestion processes (Frape, 2004; Greet, 2000).

Small intestine

The small intestine of the horse is approximately 18-25m. It is the main site of enzymatic digestion and nutrient absorption of soluble carbohydrates, proteins, lipids, minerals and vitamins. The small intestine consists of three regions duodenum, jejunum and ileum (Frape, 1998; Frape, 2004; Hayes, 1987; Jose-Cunilleras and Hinchcliff, 2003; Rowe *et al.*, 2001; Totoro and Grabowski, 2000).

As well as digesta, bile (produced by the liver) and the pancreas secretions (trypsin, sodium, potassium, chloride, and bicarbonate) flow into the duodenum to assist digestion, and resultantly cause a rapid rise of intestinal pH from 2 to 7, due to pancreatic buffering capabilities (Frape, 1998; Hoffman, 2003). Assisting digestion in the jejunum and ileum is a small population of *Lactobacillus acidophilus* and *Streptococcus bovis* anaerobic microbes, 10^8 - 10^9 bacteria/gram digesta, capable of producing nutrients for the horse (Frape, 1998; Frape, 2004).

A diet high in non-structural carbohydrate (NSC) is not well digested in the equine small intestine despite secretion of sufficient and capable enzymes (Cuddeford, 2001; Meyer *et al.*, 1995; Rowe *et al.*, 2001). Incomplete NSC digestion may occur because of:

1. High NSC intake exceeds the capacity of the small intestine
2. Enzyme action is restricted by the microstructure of plant-sourced carbohydrates especially grains
3. Enzyme action is restricted by different starch structures (B or C crystalline)
4. Different food processing techniques such as retrogradation
(Cuddeford, 2001; Rowe *et al.*, 2001)

An outcome of NSC digestion in the small intestine is negligible fermentation which significantly results in the transfer of starch to the caecum and colon which in turn may cause extensive fermentation and accumulation of D-Lactic acid within the hindgut (Bryden, 1995; Frape, 1998; Gustaffson, 2004; Hintz, 1994; Rowe *et al.*, 2001).

The hindgut

The hindgut consists of the caecum and colon and is similar to the rumen and reticulum of ruminants (Janis, 1976). The hindgut provides an adequate environment for microbial fermentation, absorption of volatile fatty acids (VFA's), water and sodium (Bryden, 1995; Frape, 1998; Greet, 2000; Gustaffson, 2004). Water is absorbed and exchanged with H⁺ and bicarbonate for VFA's (Bryden, 1995; Frape, 1998; Gustaffson, 2004). The horse hindgut is similar to a rumen in ruminants, thus the acidity of digesta is dependent on rate of acid production, absorption and total amount of acid produced (Janis, 1976; NRC, 2001). Continued NSC loading in the hindgut is associated with increased VFA's production and increasing hindgut acidity (Frape, 1998) (the consequence of NSC loading in the hindgut is discussed in detail under the heading *Accumulation of Lactate and pH in the Hindgut*).

Role of Microbial Fermentation

Microbiology

The gastrointestinal tract of a foal is initially a sterile environment but colonisation of the tract with microbes occurs immediately after parturition, via sucking/feeding from the udder, pasture and coprophagy (Gustaffson, 2004; Hintz and Cymbaluk, 1994).

The knowledge of intestinal microbes and microbial populations is still limited in horses. It is estimated 500 microbial species (most being gram-positive anaerobic bacteria) occupy the small intestine and hindgut, with approximately 99% of the microbial population being bacterial and the remainder being protozoa and fungi (Frape, 1998; Gustaffson, 2004; Moore and Dehority, 1993). Protozoa are much smaller and metabolically less active than bacteria and do not play a major role in fermentation (Frape, 1998; Rowe *et al.*, 1995). Microbes undergo microbial death and consequential catabolism may contribute to the absorption of amino acids and peptides absorbed across the mucosa (Hintz and Cymbaluk, 1994; McMeniman *et al.*, 1987). Some authors state that the small proportion of microbial derived protein is produced from microbial turnover but is not directly utilised by the horse (Gustaffson, 2004; Mackie and Wilkins, 1988).

Microbial colonisation and fermentation begins in the small intestine at the proximal duodenum with approximately 2.9×10^6 bacteria/gram digest, the population increases in the jejunum and ileum (29×10^6 and 38.4×10^6 respectively). The caecum contains the greatest microbial count with approximately 25.9×10^8 bacteria/gram digest but numbers decline in the colon (6.1×10^8) (Kern *et al.*, 1974; Mackie and Wilkins, 1988).

Different concentrations of microbes in the caecum have been reported by Argenzio and Steven (1984), Garner *et al* (1978), Goodson *et al* (1985; 1988), Willard *et al* (1977) and in colon by Hintz *et al* (1971), Julliant *et al* (2001), Kern *et al* (1974), and Moore and Dehority (1992; 1993). Microbial populations are relatively stable in the hindgut unless

sudden changes in; diet, carbohydrate source, intermittent feeding, quantity administered and plant lignification can significantly influence a different microbial population with different fermentative properties (Argenzio and Stevens, 1984; Clarke *et al.*, 1990; Goodson *et al.*, 1985; Hooper *et al.*, 2002; Kern *et al.*, 1974; Medina *et al.*, 2002).

Fermentation

Microbes ferment carbohydrates and proteins into carbon dioxide, methane and VFA's - acetate, propionate and butyrate (Frape, 1998; Hintz and Cymbaluk, 1994; Jackson, 2003). VFA's are readily absorbed from the hindgut and may contribute to 30% of the dietary energy available for utilisation by the horse (Bergtone, 1990; Hintz *et al.*, 1971; Hintz and Cymbaluk, 1994; Mackie and Wilkins, 1988; Martin-Rosset and Vermorel, 1991). Acetate, propionate and butyrate ratios normally range from 70:20:10 to 75:15:10 but are altered by diet. Higher acetate and lower propionate values are obtained with horses fed forage diets while increasing starch or non-structural carbohydrate component, increases propionate ratios (Ralston, 1986).

Caecal VFA's concentration may control meal size, feeding frequency and feeding behaviour, although receptors have not been identified (Johnston *et al.*, 1998; Ralston, 1986; Willard *et al.*, 1977). Horses fed larger amounts of concentrate feed seem to produce a greater amount of propionate in the hindgut and will spend significantly more time engaged in wood chewing, coprophagy or searching for food than horses fed roughage (Bailey, 2003; Johnston *et al.*, 1998). Studies have identified horses fed diets encouraging higher acetate and lower propionate ratios have produced minimal effect on plasma acid-base status, absorption of water and reabsorption of bicarbonate across intestinal mucosa during fermentation (Frape, 2004; Gill and Lawrence, 2003; Hammond *et al.*, 1986). Studies have used virginiamycin and dietary calcium (in the form of CaHPO_4) to prevent excessive growth of lactic acid-producing bacteria and to control the rapid accumulation of fermentative by-products in the hindgut (Gustaffson, 2004).

Intestinal pH

Frape (1998) proposed that the optimum pH in the hindgut of horses for microbial activity is pH 6.5. Absorption of VFA's across intestinal epithelium occurs when the intestinal pH is close to the pK_a of VFA's. For horses maintained on mixed grass pasture the pH of the duodenum, jejunum, and ileum was respectively 6.32, 7.10, and 7.47 pH units, and the pH decreased in the caecum and colon to 6.70 and 6.67 pH units respectively (Mackie and Wilkins, 1988). Kern *et al* (1974) had similar pH values, ileum (pH 7.4), caecum, (6.6) and colon (6.6) for horses fed meadow hay. These pH values are similar at the same sites along the intestinal tract to ruminant animals fed on hay and pasture (White, 1986).

Difference in pH along the intestinal tract is most influenced by VFA's being exchanged out of the hindgut via cyclic net exchange pattern for water, H^+ , bicarbonate, sodium (Na^+) and chloride (Cl^-) ions (Bryden, 1995; Frape, 1998; Gustaffson, 2004). De-ionised VFA's are transported across the lumen with NaCl and water in exchange for H^+ and bicarbonate, buffering the inorganic acid capacity of the hindgut (Bryden, 1995; Clarke *et al.*, 1990; Frape, 1998). Continued carbohydrate loading increases VFA's production and may influence the acidity of the hindgut (Cuddeford, 2001; Frape, 2004; Hoffman, 2003; Hussein *et al.*, 2004; Richards *et al.*, 2004; Rowe *et al.*, 1995).

Accumulation of Lactate and pH in the Hindgut

When the quantity of concentrated feed is greater than 4g non-structural carbohydrates(NSC)/100kg horse body weight, there is potential for incomplete digestion and absorption of NSC at the small intestine (Bryden, 1995; Frape, 1998; Gustaffson, 2004; Hintz and Cymbaluk, 1994; Rowe *et al.*, 2001). NSC may enter the caecum and colon and cause extensive fermentation and accumulation of lactic acid within the hindgut (Bailey, 2003; Frape, 1998; Garner *et al.*, 1978; Rowe *et al.*, 1995). Lactic acid may continue to accumulate for 24-36 hours after NSC first enter the hindgut (Frape, 2004).

Lactic acid is produced from ten species of gram-positive lactic acid producing bacteria (*Lactobacillus* and *Streptococcus bovis* species). Another contributor to lactic acid is when *Enterbacteriaceae* species, which converts lactic acid to propionate lyses under unfavourable hindgut conditions (Bailey, 2003; Frape, 2004; Garner *et al.*, 1978; Rowe *et al.*, 1995). Rapid fermentation and production of lactic acid drops hindgut to an acidic pH, this causes lyses of gram-negative (VFA producing) bacteria and increase endotoxin released into the systemic circulation. This may initiate metabolic acidosis, neurological abnormalities, digestive disorders such as colic and osmotic diarrhoea or systemic breakdown, circulatory collapse and laminitis (Bailey, 2003; Clarke *et al.*, 1990; Frape, 2004; Harris, 1997; Moore and Allen, 1996; Rowe *et al.*, 1995; Sprouse *et al.*, 1987).

There are ten species of bacteria that produce fermentative D (-) lactate or a mixture of D (-) and L (+) lactate. A few microbes produce fermentative L (+) lactate as it is solely produced in the muscle from anaerobic respiration, after which L (+) lactate is readily broken down to pyruvate by the muscle. Fermented lactic acid is not metabolised quickly and therefore accumulates in circulation, exacerbating the effect of endotoxin (Frape, 2004). The liver is the only effective method to catabolise lactic acid, if the liver is damaged (by disease, infection or poor feed management) it may intensify metabolic acidosis (Bailey, 1998; More, 1999)

Wastage in the Thoroughbred Industry

Causes of poor racing performance

The success of a racehorse is measured by race performance. Any horse failing to train or race is removed from the racing environment. The rationale of a horse failing to perform is deemed to be either lack of competitive ability, injury or illness and may occur at any stage of development and production of a racehorse (Bailey, 1998; More, 1999). In recent years there has been extensive research to identify possible risk factors of Thoroughbred horse wastage (Bailey, 1998; Bailey *et al.*, 1997, 1999; Jeffcott *et al.*, 1982; Mohammed *et al.*, 1991; Perkins, 1999; Perkins *et al.*, 2005a, 2005b; Rogers and Firth, 2005). Musculoskeletal breakdown is the major cause of reported wastage in equine industries. Some musculoskeletal complications occur at the end of training or racing when metabolic limitations and acidosis are greatest (Rossdale *et al.*, 1985).

Nutrition issues and intensive management

Intensive feeding and management of horses has increased the prevalence of abnormal behaviours and digestive or metabolic disorders, while horses with access to pasture rarely suffer such problems (Avery, 1997; Houpt, 1987; Kohnke *et al.*, 1999; McGreevy, 1995; Pell and McGreevy, 1999; Redbo *et al.*, 1998; Rushen *et al.*, 1993). Stabled horses have more behavioural problems (stereotypes- crib biting, weaving, wind sucking) and metabolic disorders (colic, diarrhoea, laminitis and gastric ulcers) which may affect a horse's racing performance than horses on pasture (Frape, 2004; Hunt, 1994; Nicol, 1999). Metabolic disorders and stereotypes in intensively managed horses may be due to (i) limited continuous grazing and (ii) reduced social contact (Johnston *et al.*, 1998).

Stereotypic behaviours

Captivity and concentrated diets are associated with development of stereotypes (McGreevy *et al.*, 1995; Nicol, 1999; Nicol *et al.*, 2002; Redbo *et al.*, 1998). Waters *et al.* (2002) concluded that feeding concentrates soon after weaning was instrumental in causing an increase in duration of foals crib-biting.

Johnston *et al.* (1998) observed similar outcome from a study of nine stabled horses were fed increasing amounts of concentrate and decreasing amounts of forage over a four week time period. In another study, horses fed a high fibre low protein diet (6.2% CP) practised coprophagy which was eliminated when the protein intake increased to 10% CP (Willard *et al.*, 1977).

The methodology of confinement, restricted feeding, concentrated diet and limited access to forage or pasture may alter saliva stimulation and digestive functions. (Houpt, 1995; Johnston *et al.*, 1998) Behavioural stereotypes may be performed in attempt to induce and regulate metabolic and abdominal discomfort caused by intensive management and feeding practices (Houpt, 1995; Nicol, 1999; Wurbel *et al.*, 1998). Studies have demonstrated a decrease in stereotypic behaviours by increasing the availability of forage (and protein) in the diet (McGreevy, 1998; McGreevy *et al.*, 1995; Willard *et al.*, 1977). It is proposed that forage reduces stereotypic behaviours because:

1. Forage increases gut fill and possibly satisfies physiological feedback mechanisms by reducing the time and motivation to perform stereotypic behaviours (crib biting, weaving, wind sucking) (Clarke *et al.*, 1990; Johnston *et al.*, 1998; McGreevy, 1998; McGreevy *et al.*, 1995).
2. Food deprivation or feeding a concentrated diet causes an increased exposure of gastric juices to the stomach mucosa (Murray and Eichorn, 1996; Rowe *et al.*, 1994) and thus the horse may perform stereotypic behaviour to increase the flow of alkaline saliva (Nicol, 1999).
3. Saliva is essentially alkaline and buffers gastric acid and juices through the gastrointestinal tract.
But saliva is only secreted during mastication (Alexander and Hickson, 1970; Frape, 1998). Horses fed restricted meals of high grain diets may not produce enough saliva to buffer stomach contents and thus perform stereotypic behaviours to simulate saliva secretion after meal ingestion (Nicol *et al.*, 2002).

Digestive disorders

Digestive disorders include gastric ulceration, colic, diarrhoea and laminitis, and are associated with confinement, restricted feeding, concentrated diet or limited access to forage or pasture.

Gastric ulcers (lesions in the stomach) are a result of gastric mucosal damage from excessive gastric acid and pepsin (Lewis, 1995). The incidence of ulcers is more prevalent in horses that are highly stressed, in race training or regularly administered anti-inflammatory drugs such as phenylbutazone (Lewis, 1995; Murray, 1999). Gastric secretion is greater in horses fed grain in comparison to those fed only hay (Nadeau *et al.*, 2000; Smyth *et al.*, 1988). Therefore, high grain diets may increase the incidence of ulceration due to continuous secretions of gastrin, limited salivary secretions and buffering effect to gastric acids (Hammond *et al.*, 1986; Lewis, 1995; Nadeau *et al.*, 2000).

Colic is another negative consequence of intensive management and is caused by a number of aetiologies (Frape and Boxall, 1974). Lewis (1995) suggest horses in competition or training are predisposed to colic more significantly if there is a minor changes in dietary routine. Possible reasoning is, any change in diet may cause changes in microbial populations which influences digestibility and digestive efficiency (Argenzio and Stevens, 1984; Goodson *et al.*, 1985; Goodson, 1988) and thus may predispose the horse to colic.

Laminitis is another metabolic disorder associated with intensive management. Laminitis is the inflammation of the sensitive laminae in the sub-mural laminar structures of the foot (Adam's, 1974; Stashak, 2002). This definition is a gross over-simplification of a very complicated ailment associated with a number of systemic disorders with different levels of severity (Baxter, 1996; Hood *et al.*, 1993; Perkins, 2001; Stashak, 2002). A typical clinical sign of laminitis is lameness (Hood, 1999).

There is limited knowledge about the cause of laminitis but it is possible certain management and feeding practices may influence the condition in racehorses. Two theories presumed to cause laminitis are (1) toxic/metabolic accumulation and (2) vascular dysfunctions (Hood, 1999). By provoking and disturbing the structure and function of laminae of the hoof and initiate laminitis (Sloet va Oldenbourgh-Oosterbaan, 1999).

1. Toxic/metabolic dysfunction occurs from feeding horses concentrated diets. The NSC content found in concentrated feed is believed to produce excessive fermentation and increased lactic acid and endotoxins at the hindgut. However, laminitis has not been produced by administration of endotoxin to previously healthy horses, suggesting that endotoxin as a predisposing factor to laminitis remains speculative (Hood, 1999; Moore and Allen, 1996; Stashak, 2002).
2. Vascular dysfunction is thought to originate from concentrated diets and subsequent monoamines released by *Lactobacillus* and *Streptococcus* bacteria. Vascular mechanisms are damaged when there is an increased circulation of monoamines which are potent vasoconstrictor products (Bailey *et al.*, 2000).

Diarrhoea may be associated with lactic acid accumulation in the hindgut, damage to the permeability of intestinal mucosa and significantly increase endotoxin release (Gustaffson, 2004). Diarrhoea can be associated with prolonged clinical signs like anorexia, fever and severe depression (Gustaffson, 2004) which may be detrimental to the horse's well-being and performance.

The consequence of the intensive management and feeding practices currently employed in Thoroughbred racing stables may influence stereotypic behaviours and digestive disorders, thereby affecting horse performance and increasing the incidence of wastage.

Feed Requirements of Horses

Nutrient Sources

All living organisms require nutrients for tissue and cellular maintenance. Extracting nutrients from food is a complex of processes (Harris, 1997; Pagan, 2003). A horse's diet provides dietary nutrients through catabolism of soluble or structural carbohydrates, proteins and fats to form either chemical, mechanical or heat energy to allow bodily functions (Harris, 1997; Kohnke *et al.*, 1999; Pagan, 2003).

Energy

All dietary energy sources catabolise to adenosine triphosphate (ATP) which is the immediate energy commodity for bodily function and muscular movement (Bryden, 1995; Eaton, 1994; Pagan, 2003). Horses can store dietary energy in adipose tissue in the form of triglycerides (TAGs). Liver stores glycogen, and skeletal muscle stores intramuscular glycogen, TAGs, ATP and creatine phosphate, while blood carries glucose and free fatty acids (Harris, 1997; Pagan, 2003).

ATP is formed by anaerobic phosphorylation, without the presence of oxygen, or by aerobic phosphorylation, in the presence of oxygen (Eaton, 1994). Anaerobic phosphorylation can occur in one of three ways to generate energy for body functions:

1. Directly with ATP stored in skeletal muscle (in very small quantities).
2. By splitting creatine phosphate (stored in skeletal muscle) into two ATP molecules.
3. Catabolism of glucose derived from glycogen or blood glucose.

(Eaton, 1994)

Anaerobic metabolism of glycogen or glucose is the fastest method of ATP production but is inefficient. Aerobic metabolism is the slowest energy generation of ATP but twelve times more efficient than anaerobic metabolism. Aerobic glucose (derived from carbohydrate, fat or protein) is converted to 36 ATP molecules, compared to anaerobic glucose where three ATP molecules and two molecules of lactate are produced (Pagan, 2003).

Carbohydrates

Carbohydrates generally have an empirical formula $(CH_2O)_n$ and may contain other chemical compounds like nitrogen, phosphorus, sulphur, alcohol and acids (McDonald *et al.*, 2002). Plant carbohydrates are the main source of dietary energy for horses especially during short term high intensive oxidative exercise (Eaton, 1994; Harris, 1997; Hoffman, 2003; NRC, 1989). Depending on type and amount fed, plant carbohydrates may be divided into two major groups; those that undergo hydrolysis into simple sugars in the small intestine, or undergo microbial fermentation and produce VFA's in the hindgut (Hoffman, 2003). VFA's are important sources of dietary energy for horses grazing on pasture or fed high forage diets, while grains hydrolyse into simple sugars because of predominately higher levels of starch (approximately 60-80% DM) with lower levels of fibre than pasture or forage (Hoffman, 2003; Hoffman *et al.*, 2001; NRC, 1989).

The main difference between the two groups is dependant on the linkage of the sugar molecules, influencing carbohydrate solubility and digestibility. Carbohydrates that undergo fermentation are β -1,4 linked molecules and include structural carbohydrates consisting of cellulose, lignin, hemicellulose, soluble fibre, fructan, and galactan, that is resistant to enzymatic hydrolysis (Rowe *et al.*, 2001).

The methods best considered to determine the extent of structural carbohydrate in feeds is the Van Soest detergent methods.

1. Neutral detergent fibre (NDF) test determines lignin, cellulose and hemicellulose.
2. Acid detergent fibre (ADF) test determines the proportion of internal plant material and is a good measure of plant digestibility.

(McDonald *et al.*, 2002)

Carbohydrates that undergo hydrolysis in the small intestine consists of α -1,4 linked molecules, named soluble sugars or non-structural carbohydrate (NSC), and include disaccharides, oligosaccharides (maltotriose) and starch (Hoffman, 2003; McDonald *et al.*, 2002). The ability of horses to convert NSC to glucose is metabolically more efficient than fermentation of VFA's (Hoffman, 2003; Rowe *et al.*, 2001). The best method to determine the amount of NSC in horse feed is by the following calculation

$$\text{NSC} = 1000 - (\text{CP} + \text{ether extract} + \text{NDF} + \text{ash})$$

(McDonald *et al.*, 2002; Pagan *et al.*, 1998)

A small amount of NSC is hydrolyzed by gastric acid in the stomach but the primary site of NSC hydrolysis is the small intestine with α -amylase, α -glucosidases (sucrase, glucoamylase, maltase), and β -galactosidase (lactase) enzymes (Hoffman, 2003). Horses have limited ability to hydrolysis abundant amounts (greater than 4g/100kg body weight) of NSC at the small intestine, due to low and variable concentrations of NSC enzymes. If quantities of NSC exceed this amount there was a greater risk of NSC entering the hindgut and causing hindgut acidosis which could be associated with digestive disorders (Cuddeford, 2001; Hoffman, 2003; Hussein *et al.*, 2004; Richards *et al.*, 2004).

Fats

Fats are triglycerides (TAGs) which are three fatty acid units attached to a single glycerol molecule. Fatty acids can range in length and structure and undergo β -oxidation via aerobic phosphorylation to form ATP (Eaton, 1994). TAGs are regarded among the richest energy source, containing 2.25 times more energy per unit weight than other glucose sources (Bryden, 1995; Eaton, 1994). The addition of fat into a horse's diet is an efficient method to increase intake of dietary energy and lipid soluble vitamins, while reducing metabolic heat production. Fats may also play an important role in exercise performance (Bryden, 1995; Eaton, 1994; Harris, 1997; Hoffman, 2003; Lawrence, 1990; NRC, 1989).

Nutrient Requirements

Nutrient supply should meet horse maintenance, growth and work load levels, and are subsequently related to body weight, activity and environmental temperatures (Hintz and Cymbaluk, 1994; NRC, 1989). Maintenance requirements must match nutrient needs for normal activities of a non-working horse while maintaining a consistent body weight and digestive and metabolic efficiency (Harris, 1997; Kohnke *et al.*, 1999; NRC, 1989). Nutrient requirements for exercise work load are above maintenance requirements and take into consideration exercise intensity, rider weight and tack, degree of fatigue, condition and fitness, diet composition, environment conditions and terrain (Harris, 1997; Hintz and Cymbaluk, 1994).

Nutrient requirements above maintenance for growth are influenced by horse's age or level of maturity but also depends on previous feeding and developmental history (Harris, 1997; Hintz and Cymbaluk, 1994). The New Zealand Thoroughbred usually begins race training at 18 months to 24 months of age, therefore nutrients must meet both the growth and work load requirements (Rogers, personal communication, 2005).

Calculating Energy Requirements

The dietary energy potential of food can be described in four ways, gross energy (GE), net energy (NE), metabolisable energy (ME) and digestible energy (DE). DE is the most practical method to describe the amount of dietary energy available from food sources but not the most accurate (Harris, 1997). DE is calculated from the gross amount of energy of the food source (GE) minus the remaining energy in faecal output including endogenous losses, undigested food and gastrointestinal microbes (Grace, 1997; Pagan, 2003). DE is described in joules (J) which is the international standard unit for measuring energy. Calories (cal) is the old imperial measurement for energy (where 1 cal = 4.18 J) (Harris, 1997).

However, the energy requirements for a horse are commonly described in Megacalories (Mcal) or Kilojoules (KJ) of DE per kg of body weight (BW) (Bryden, 1995; Pagan, 2003). DE requirements are based on physiological conditions of maintenance, growth, pregnancy, lactation, breeding and exercise work load (Harris, 1997; NRC, 1989; Pagan, 2003). The NRC (1989) has produced guidelines for these possible physiological states and is the most common requirement equations applied, refer to Table 2

Table 2. Energy Requirements of Horses (NRC 1989).

Estimations of digestible energy (DE) requirements (Mcal of DE/day)		
	Specifications	DE
A. Maintenance		
1	200-600 kg Bwt	1.4+ 0.03 BW
2	>600 kg BW	1.82 + 0.0383 BW- 0.000015 BW ²
B. Pregnant mares		
1	9 months	1.11 (maintenance DE)
2	10 months	1.13 (maintenance DE)
3	11 months	1.20 (maintenance DE)
C. Lactating mares		
		<u>Foaling- 3months</u>
1	a. 200-299kg BW	(maintenance DE)+(0.04BW*0.792)
	b. 300-900 kg BW	(maintenance DE)+(0.04BW*0.792)
		<u>3 months- weaning</u>
2	a. 200-299kg BW	(maintenance DE)+(0.03BW*0.792)
	b. 300-900 kg BW	(maintenance DE)+(0.03BW*0.792)
D. Working horses		
1	Light work ^a	1.25 (maintenance DE)
2	Moderate work ^b	1.50 (maintenance DE)
3	Intense work ^c	2.00 (maintenance DE)
F. Growing horses		
1	Not in training	(maintenance DE)+(4.81+1.17x-0.023x ²)(ADG)
2	In training	
	Light work ^a	1.25 (maintenance DE)+(4.81+1.17x-0.023 x ²)(ADG)
	Moderate work ^b	1.50 (maintenance DE)+(4.81+1.17x-0.023 x ²)(ADG)
	Intense work ^c	2.00 (maintenance DE)+(4.81+1.17x-0.023 x ²)(ADG)
		BW = body weight in kg
		x=age in months
		ADG= average daily gain (kg/day)

^a light work example = walking, trotting, pleasure riding/hacking

^b medium work example = ranch work and jumping

^c intense work example = racing and polo

Maintenance energy requirements is the amount of nutrients (described in DE) to sufficiently fuel normal activity of a non-working horse, maintaining a consistent body weight (Harris, 1997; Kohnke *et al.*, 1999; NRC, 1989). American and European countries use to calculate maintenance requirements by BW (kg) * ^{0.75} DE. However, this equation overestimates maintenance needs for small ponies and large horses (Harris, 1997).

The French use a different system called UFC, l'unité fouragire cheval (French horse feed unit) (Martin-Rosset *et al.*, 1994). Comparisons of the two maintenance energy equations used today are presented in Table 3.

Table 3. Comparison of different maintenance energy equations for horses.

Maintenance Requirements	Equation
NRC (1989)	(Mcal of DE/day)
200-600 kg BW	1.4+ 0.03 BW
>600 kg BW	1.82 + 0.0383 BW- 0.000015BW ²
Martin-Rosset <i>et al</i> (1989)	(Kcal of DE/day)
	140 BW ^{0.75}

Energy requirements increase proportionally to exercise type, intensity, duration, work effort, fitness, body condition, and environmental temperature for a horse in regular training or exercise (Harris, 1997; Hintz, 1994; Hintz and Cymbaluk, 1994; Kohnke *et al.*, 1999; Southwood *et al.*, 1993a). The NRC (1989) work load DE requirements (Mcal/day) increase 25, 50 and 100% respectively above maintenance requirements. Other authors believe that these recommendations are not specific enough and have formulated energy requirements for various activities.

Pagan and Hintz (1986) suggested the energy expenditure with work was better estimated by energy cost described by the following equation.

$$\text{Energy expenditure (cal/kg/min)} = e^{3.02 + 0.065x}$$

x= velocity (m/min)

Table 4 presents the different calculations for work load energy requirements between NRC (1989) and Kohnke *et al* (1999).

Table 4. Comparison between Kohnke *et al* (1999) and NRC (1989) for energy requirements for horses at different work loads.

Example		Estimate DE	Estimate total feed amount/day
NRC (1989)		(Mcal of DE/day)	
Light work	walking, trotting, pleasure riding/hacking	1.25 (maintenance DE)*	
Moderate work	ranch work and jumping	1.50(maintenance DE)*	2% BW
Intense work	racing and polo	2.00(maintenance DE)*	
Kohnke <i>et al</i> (1999)		(MJ/DE/100kg BW/day)	
Light work ^a	basic dressage, showing	17.5 (17-18)	
Moderate work ^b	advanced dressage, showjumping, polocross, eventing, racing (early-mid training)	22 (21-23)	2% BW
Intense work ^c	racing (full training), polo, endurance horses	29 (28-30)	

^a 30-60 minutes at walk, trotting or cantering

^b 30-120 min walk, trotting, cantering and some galloping

^c 30-60 min trot, canter, gallop and short high intensive galloping

Example: A 500 kg horse in race training (intense work) nutrient requirements would be

$$\begin{aligned} \text{NRC (1989)} &= 2\% \text{ Body weight (500*2\%)} = 10\text{kg total amount of feed/day} \\ &= (1.4 + (0.03*500))*2=32.8\text{Mcal/day/10kg (total feed weight)} = 3.28\text{Mcal/kg fed} \\ &= 13.7 \text{ MJ/kg fed} \end{aligned}$$

$$\begin{aligned} \text{Kohnke } et \text{ al (1999)} &= 0.29 *500 = 145 \text{ MJ/day} & (145\text{MJ/kg}/4.18 =34.7 \text{ Mcal/day}) \\ &=34.7\text{Mcal/day /10kg (total feed weight)} & = 3.47 \text{ Mcal/kg fed} \\ & & =14.5\text{MJ/kg fed} \end{aligned}$$

Energy intake and performance

Racehorses have an energy requirement that can not be provided from a natural forage diet. To obtain the necessary dietary energy the racing industry relies heavily on large amounts of concentrated feed (Bryden, 1995; Harris, 1997; Kohnke *et al.*, 1999). Trainers try to increase dietary energy by reducing forage intake and feeding more concentrated high energy grain meals, due to a perception that forage increases gut fill that negatively affecting racing ability (Frape, 1988; Hoskins and Gee, 2004). Despite this, Pagan and Harris (1999) state feeding forage before exercise does not affect performance and instead, small quantities of hay should be offered before exercise to ensure proper gastrointestinal function.

Protein

Proteins make up a large portion of body tissue and fat free body mass of a mature horse (NRC, 1989). Twenty-two amino acids form protein constituents of muscle, blood, enzymes and hormones (Bryden, 1995; NRC, 1989). Amino acids are divided into two types, non-essential and essential amino acids. Ten amino acids (lysine, leucine, methionine, arginine, threonine, tryptophan, isoleucine, histidine, phenylalanine and valine) are considered essential amino acids and must be provided in the diet (Boisen *et al.*, 2000; Bryden, 1995). The remainder are non-essential amino acids and are diet derived or produced by the animal (Bryden, 1995). Dietary protein is plant sourced and provides amino acids for tissue synthesis but is an insufficient source for consistent dietary energy (Bryden, 1995; Hoskins and Gee, 2004; Lawrence, 1994; Pagan, 1997a).

Protein digestion begins at the stomach and is further digested and absorbed at the small intestine (Frape, 2004). Microbes at the hindgut hydrolyse proteins, ammonia (NH₃) and urea to form microbial protein or synthesis of amino acids (Bryden, 1995; Frape, 2004). The ability of the horse to absorb and utilise microbial protein and non-protein nitrogen is supported by Argenzio and Stevens (1984), Eaton (1994) and Frape (2004). But other authors state that the small proportion of microbial derived protein is not directly utilised by the horse and is excreted in urine and faeces (Bryden, 1995; Hintz and Cymbaluk, 1994; McMeniman *et al.*, 1987). It has also been suggested that excessive protein intake may initiate microbial proliferation, and increase fermentation, which may predispose the horse to laminitis.

Chopping or pelleting reduces food particle size and results in reduction of mastication and saliva secretion, increasing gastrointestinal transit time, reducing gastric juice and acid emulsification, and therefore reducing protein digestion (Bryden, 1995; Frape, 2004). Interestingly, quality forage (roughage) with high protein content does not seem to cause such problems in the gastrointestinal tract, thus it is suggested concentrated feeds should be fed following a small amount of roughage (Bryden, 1995).

Crude protein requirements

Protein requirements are expressed as digestible crude protein (CP) which is based on apparent faecal digestibility of nitrogen, calculated by multiplying nitrogen by a factor of 6.25. This is not the measure of actual protein in the diet (Kohnke *et al.*, 1999; Olsman *et al.*, 2003; Pagan, 1997a, 1997b). Several studies have presented different maintenance CP recommendations (Table 5).

Table 5. Comparison of different maintenance protein requirements.

Author	Maintenance protein requirements for 500 kg horse
NRC (1989)	60 g/kg day**
Bryden (1995)	85 g /kg DM* day
Martin-Rosset <i>et al</i> (1994)	2.8 g/kg ^{0.75*} day
Kohnke <i>et al</i> (1999)	1.3-1.5 g/kg** .day

* Per feed weight

** Per horse body weight

Bryden (1995) believes horse maintenance protein requirements are low and can be met by good quality forage and grain. NRC (1989) estimates CP levels should be at 11.4% DM for intensive work. However most New Zealand pastures are 12-25% DM CP (Hoskins and Gee, 2004; Waghorn and Barry, 1987) and thus, if pasture is the only component of the diet, it will exceed NRC (1989) protein requirements for horses in intensive work if horses are on pasture long enough to get this daily intake. Excess protein above the horse's requirement has a metabolic cost, as it requires catabolism and excretion and may be detrimental to athletic performance (Graham-Thiers *et al.*, 2001). Previous studies have determined amino acid requirements and CP for optimum growth in young horses, but there are few studies for mature horses (Hintz and Cymbaluk, 1994; Staniar *et al.*, 2001).

Protein intake and performance

Amino acids have been given to racehorses to improve their speed (Hintz and Cymbaluk, 1994). There is much debate as to whether there is need for increased protein and amino acid requirements for muscular repair and turnover following heavy exercise (Bryden, 1995). For horses beginning race training there may be an extra requirement of protein for muscle development. However, restriction of dietary protein may be beneficial during strenuous exercise by diminishing production of heat and lactic acid (Graham-Thiers *et al.*, 2001).

Mineral and Vitamins

Minerals and vitamins are not energy-producing nutrients but are metabolites that aid metabolism and homeostasis. The horse requires twenty-one different minerals that are broken down into major and trace minerals (Frape, 2004; Hayes, 1987; NRC, 1989). Of all major minerals, calcium, phosphorus, magnesium, potassium, sodium, chloride and trace minerals; selenium, copper, manganese, iodine, zinc, iron, fluoride and cobalt are considered by NRC (1989) worthy of having a daily requirement. In New Zealand pasture mineral composition is largely influenced by botanical composition, season, soil type and fertiliser history (Bryden, 1995; Grace *et al.*, 2002; Hunt, 1994). Since the 1930's, New Zealand soils have identified selenium, copper, cobalt, iodine and zinc to cause trace element deficiencies highlighted with animal health issues (Morton *et al.*, 1999).

Vitamins are complex organic structures (Frape, 2004). There are two categories of vitamins, lipid soluble (vitamin D, E, K, A) and water soluble (vitamin C and eight B vitamins) all have important roles in the horse (NRC, 1989). New Zealand pastures have a good source of vitamins (exception vitamin D, which is produced by the skin and ultra violet light) therefore it is considered unnecessary to supplement horses with vitamins if on pasture-fed system (Hunt, 1994).

Management and Training Methods

In New Zealand the racing season runs from August 1 to July 31 in the following year, the Thoroughbred horse official birth date is 1 August, and thus foals born in August to October and can start their racing careers at 18 months (Perkins, 1999; Perkins *et al.*, 2004b).

Stable management

The majority of racehorses in New Zealand are kept in stables. When bedding material of either straw or fine wood chips (Kidd, 2002). Horses are normally turned out to paddocks for a short time each day or fed cut grass in their stables, when out of training, racehorses are generally spelled to pasture (Bourke, 1968; Huntington and Jenkinson, 2003; NRC, 1989; Pearce, 1975).

Training methods

Training programmes have been regarded as an art rather than science, with skills learnt through experience or handed down from previous generations, apprenticeships with other horse trainers, or trial and error approach (Milton McClure, 1990; Rose, 1993). Rose (1993) observed training routines at racing stables in Australia and New Zealand and reported that most horses undertook small amounts of exercise as a daily fitness routine. Training from pre-training to first trial or race start took on average 11 ± 3 weeks (with a range of 5-23 weeks) (Bailey *et al.*, 1999; Perkins *et al.*, 2005a). The duration of training may be for a further 3-4 months (depending upon performance) before the horse is turned out for a spell or retirement (Bourke, 1968).

Trainers rely on subjective methods to assess a horse's performance (Rose, 1993). Appearance of the horse's recovery after exercise, track times or information from the jockey, track conditions, failure to train or race, ability or soundness all influence the continuation or retirement from training (Bourke, 1968, 1995; Rose, 1993).

Training schedules are described by horse gaits. But it must be highlighted that countries have differences in describing horse gaits and expectations of gait velocity. In New Zealand, gaits and gait velocity descriptions are very similar to Australia and the UK. Terminology for the respective countries is represented in Table 6.

Table 6. Description of racehorse gaits and velocity (m/sec).

	Trot	Canter	$\frac{1}{4}$ pace	$\frac{1}{2}$ pace	Gallop
New Zealand ^a	3-4.5 m/sec	5-8 m/sec	9-11 m/sec	12.5-14.2 m/sec	15-16 m/sec
Australia ^b	4 m/sec	7 m/sec	Slow gallop	13 m/sec	Race speed
United Kingdom ^b	3-4.5 m/sec	5-8 m/sec	-	13.3-15.3 m/sec	-
France ^c	5 m/sec	5.8 m/sec	-	-	10 m/sec
United States ^d	Trot 4.2 m/sec	Slow gallop 6.5-7.5 m/sec	Gallop 7.3-8 m/sec	Gallop 8-10 m/sec	Breeze/sprint 15-16 m/sec

^a Rogers and Firth (2005)

^b Evans (1994)

^c Martin- Rosset *et al* (1994) (based on a 560kg horse carrying 100kg load- rider & tack)

^d Foreman *et al* (1990)

Horse trainers can be successful without formal knowledge of exercise or training science (Evans, 1994). Typically, racehorses are trained using a three-phase strategy which progressively increases work intensity. Phase one is pre training or slow-speed training, phase two is aerobic and anaerobic training and phase three is anaerobic or speed training (Evans, 1994). Typically, Australian Thoroughbred racehorses will partake phase-one training for 4-5 weeks at distances of 3000-5000m at a trot and canter (Southwood *et al.*, 1993a).

After the pre-training period, Australian racehorses usually alternate between fast and slow training sessions (Southwood *et al.*, 1993a) namely phase two. Horses on slow sessions are exercised at trot or canter averaging 5500m at 4-7m/s. Fast sessions start with a 1000m warm up, average 2000m at 12-16 m/s, and finish with 1000m of trotting/cantering (Southwood *et al.*, 1993a). In North America phase two is referred to as breeze working, horses work at 75% maximum speed on the seventh to tenth day with the remaining days walking, trotting or swimming (Harkins *et al.*, 1990).

Central American Thoroughbred trainers typically gallop racehorses at near maximum speed for 600-1000m every fifth day, the remaining days are slow gait work; walking, trotting and cantering averaging a total distance of 5000m (McCutcheon *et al.*, 1987).

Phase three (anaerobic training or speed training) traditionally has not been factored into Thoroughbred training regimes, but focuses on speed and acceleration. For example horses are worked at pace for 600-1600m at 14-16 s/200m then galloped over 200-600m at 95-100% top speed and it is probably unwise to train horses near 100% gallop for longer distances because of increased likelihood of overtraining syndromes (Rose, 1993).

Scientific research has tended to focus on physiological responses to fitness levels, overtraining responses (Tyler-McGowan *et al.*, 1999), or evaluation of poor training/race performance (Bourke, 1995; Divers and Dreyfuss, 1990; Perkins, 1999) instead of race training programmes. Training methods for Thoroughbred horses, optimal number of training days/week, intensity and frequency of exercise for different distances, and methods to maintain fitness, all require further investigation (Rose, 1993), and have not really been investigated in detail in the last 12 years.

Feed management

Horses at racing stables are not fed scientifically but by the criteria of racing success (Milton McClure, 1990; Southwood *et al.*, 1993a). On any given day, observation at different stables found variation in feed ingredients, different number of feedings per day with mixed amounts fed, as rations per horse are determined by general appearance and performance of individual horses (Milton McClure, 1990; Southwood *et al.*, 1993a). Southwood *et al.* (1993b) performed a survey determining feeding practices in Australia of Thoroughbred and Standardbred stables, which drew similar findings with observed feeding practices for New Zealand Thoroughbred stables (Huntington and Jenkinson, 2003)

A concentrated feed is a term given to horse feeds that have high energy content and are highly digestible. There are two types of concentrated feed regimes, cereals and commercially prepared feeds. Oats, wheat, barley, maize or corn are usually crushed, rolled, ground or heated to increase digestibility. Huntington and Jenkinson (1992) concluded that oats were the most popular cereal fed to racehorses in Australia and New Zealand. Southwood *et al* (1993a) surveyed feed practices at 25 Thoroughbred stables in Sydney and found 84% of trainers fed 3.6 ± 0.4 kg oats/day which is comparable with Gallagher *et al* (1992) reporting 3.8kg oats/day fed to Thoroughbreds in Detroit, U.S.A.

Prepared feeds are a combination of grains, termed premixed sweetfeeds or hard feeds are specifically designed and marketed for a particular class of horse or equine discipline and usually consist of cereals that are further processed into a textured, extruded or pelleted form. In Sydney, 4% of stables fed commercially prepared feeds as primary sources of concentrates, while 44% of all the trainers fed a combination of commercially prepared concentrates with grains (1992). This was similar for Thoroughbred trainers surveyed in Detroit (Gallagher *et al.*, 1992). 60% of New Zealand Thoroughbred trainers fed on average 2kg per day concentrates consisting of commercially prepared feed with oats, chaff and supplements (Huntington and Jenkinson, 2003).

It is common for racehorse diets to be supplemented with additional protein, vegetable oil, minerals and vitamins (Huntington and Jenkinson, 2003; Southwood *et al.*, 1993a). Huntington and Jenkinson (2003) reported that horse trainers in Australia and New Zealand over-supplement with protein. Excessive protein supplementation may be detrimental to athletic performance (Graham-Thiers *et al.*, 2001) and could predispose horses to laminitis via the release of monoamines in the hindgut (Bailey *et al.*, 2000). Common protein supplements used in horse feeds are soyabean, canola and linseed meal, copra, cottonseed, sunflower and lupin seeds, peas, broad beans, milk powder and brewers yeast (Kohnke *et al.*, 1999). It has become popular for trainers to supplement diet with vegetable oils to increase energy intake and supply of fat soluble vitamins (Lawrence, 1990).

Roughage

New Zealand and Australian racehorses are fed roughage in the form of pasture, hay or chaff (Huntington and Jenkinson, 2003). In Australia, Southwood *et al* (1993a) found the average daily roughage intake was 2.0 ± 0.3 kg which contrasted with Bourke (1968), who found horses were fed up to 5kg roughage per day in Australia. While Huntington and Jenkinson (2003) reported approximately 2.9kg roughage was fed per day in New Zealand and Australia. Interestingly, Gallagher *et al* (1992) reported 6.6 ± 1.7 kg roughage was feed in the USA. The main source of roughage for Thoroughbred horses is hay and for Standardbred horses is chaff (Huntington and Jenkinson, 2003; Southwood *et al.*, 1993a).

NRC (1989) requirements suggest that horses in light work should consume a diet containing 80% roughage and 20% concentrate whereas horses in heavy work might consume 33% roughage and 67% concentrate (Lawrence, 1990). Consequently, concentrate to roughage ratios recommended for horses in race training are lower than NRC guidelines (Davie *et al.*, 2000; Gallagher *et al.*, 1992; Glade, 1983; NRC, 1989; Southwood *et al.*, 1993a).

Some trainers' believe horses can not be maintained on pasture throughout race training, because pasture fails to meet energy requirements and causes excessive gut fill and these horses may not eat there concentrated meals (Frape, 2004; Hoskins and Gee, 2004; Huntington and Jenkinson, 2003; Milton McClure, 1990). However, good quality pasture can be an excellent source of vitamins, fibre (roughage), and protein that positively influences gut health and function (Hoskins and Gee, 2004). Huntington and Jenkinson (2003) suggest most racing stables in Australia and New Zealand have access to pasture but trainers would prefer to cut pasture and feed horses in stable or turn out horse to pasture for up to 2.5 hours a day.

Feeding and water regime

It is common for stabled horses to be offered concentrated feeds more than twice daily, (Houpt, 1987). Most New Zealand trainers feed three times a day with some trainers feeding horses four times a day (Huntington and Jenkinson, 2003). Milton McClure (1990) reported the most common feeding practice is three feeds a day with a light grain ration early morning prior to training, another after training (late morning) with the final and largest ration late afternoon (approximately 4pm) with fresh water available at all times.

Faecal pH

Intensive management and feeding of Thoroughbred racehorses may satisfy energy demands and short-term performance goals, but may negatively affect future performance because of developmental, metabolic, or stereotypic abnormalities. Identifying hindgut acidosis for horses in intensive management and feeding is a potential tool in rectifying mismanagement and feeding practices.

Authors have attempted to measure hindgut acidosis and quantifying its prevalence. Alexander and Donald (1949), Lowe *et al* (1970), Willard *et al* (1977) and Garner *et al* (1978), have used cecal or colonic fistulation of the hindgut to sample contents for analysis of microbial action and fermentation. However, this technique is invasive, expensive and impracticable. Rowe *et al* (1995; 1994) and Johnston *et al* (1998) have collected blood D-lactate to determine hindgut acidosis. This sampling method is less invasive but still expensive, impracticable and not a good representation of hindgut acidosis at lower concentration of blood D-Lactate (Rowe *et al.*, 1995).

Measuring faecal pH in horses has been reported by Johnston *et al* (1998) and Zeyner *et al* (2004; 1992; 1993) who performed studies that identified a proportional drop in horse faecal pH when dietary concentrates were increased compared to roughage content. Rowe *et al* (1994) concluded a decrease in faecal pH from 7.10 to 6.20 in 12 horses after feeding a concentrated diet. Davie *et al* (2000) also measured faecal pH and concluded 16 intensively managed racehorses receiving a diet high in concentrates had an average faecal pH of 6.30 (stable A: n=8, pH $6.30 \pm 0.09_{SD}$ and stable B: n=8, pH $6.30 \pm 0.07_{SD}$) compared to 12 horses on pasture (pH $6.9 \pm 0.22_{SD}$). These respective studies support the relationship of high lactic acid production from excessive hindgut fermentation of concentrates.

Faecal pH as a Tool to Determine Hindgut Acidosis

Many studies have used faecal pH as an indirect, non-invasive, economic and practical methodology to determine the extent of fermentation at the hindgut (de Fombelle *et al.*, 2003; Eastwood, 2002; Hussein *et al.*, 2004; Johnston *et al.*, 1998; Nicol *et al.*, 2002; Olsman *et al.*, 2004; Richards *et al.*, 2004; Rowe *et al.*, 2001; Rowe *et al.*, 1994; Zeyner *et al.*, 2004). Many authors have used varying methodologies to measure horse faecal pH. Richards *et al* (2004), Zeyner *et al* (2004), Eastwood (2002) and Rowe *et al* (2001) used 1:1 ratio of faecal mass (g) to distilled water (ml). Nicol *et al* (2002) used 1:4 ratio of faecal mass to distilled water (5g : 20 ml). Johnston *et al* (1998) added 1ml distilled water to faecal mass once removed from rectum. While de Formbelle *et al* (2003), Hussein *et al* (2004) and Olsman *et al* (2004) measured horse faecal pH straight from faecal mass.

Finn and Rogers (2001 unpublished data) measured the validity of Zeyner's (1992; 1993) faecal pH methodology in pasture-maintained horses, and concluded that the methodology is practical, as time between collection, analysis and freezing of the samples had no significant effect on pH. The effect of different feeding and management strategies on faecal pH of New Zealand horses maintained at pasture while supplemented with grain were studied by Eastwood (2002); there was no significant decrease in faecal pH and it was proposed that grazing pasture may have a potential to buffer the effects of acidosis caused by grain overload in the hindgut.

Identifying hindgut acidosis by measuring faecal pH has never been conducted on racing Thoroughbred horses in New Zealand. The benefit of determining risk factors associated with hindgut acidosis and being able to quantify sub clinical cases is the first step in understanding problems of poor management and feeding practices that are largely associated with high wastage rates of intensively managed Thoroughbred horses. If hindgut acidosis is addressed as a priority it may be a step forward in improving performance, health and welfare of horses with the possibly of reducing wastage and associated costs within the racing Thoroughbred industry.

Aim of the study

In the review of relevant literature there was no information to show a relationship between management of racing New Zealand Thoroughbreds and influence on faecal pH.

The aim of this study was to identify the effect of different feeding and management strategies of racing New Zealand Thoroughbred horses on faecal pH. The objectives of the project were; (i) examine faecal pH of horses managed under one stable with the same feeding and similar training programmes; (ii) examine sources of variation in faecal pH of horses with different feed programmes and no exercise, and. (iii) examine the effect of faecal pH from horses from different stables with different feed and training programmes

The research presented in this thesis was obtaining data by survey. The survey interviewed New Zealand racing Thoroughbred trainers on their current racehorses. A single horse faecal sample was collected and pH tested for every horse identified.

Hypotheses

1. There is a significant difference of mean faecal pH between days and week of sampling.
2. There is a significant difference of mean faecal pH between horse, gender and horse age categories.
3. There is a significant difference of mean faecal pH between trainers.
4. There is a significant difference of mean faecal pH between stables.
5. There is a significant difference of mean faecal pH between exercise workload categories.
6. There is a significant difference of mean faecal pH between diets.
7. There is a significant relationship of mean faecal pH to horse behaviour.

Materials and Methods

EXPERIMENT ONE: TESTING SIMILAR DIETS AND MANAGEMENT

Animals

Faecal material was collected from fourteen 3-year-old Thoroughbreds in regular race training.

Management of horses

The horses were trained by a licensed trainer using a typical racing programme at Foxton Racecourse, Foxton, New Zealand, for an 84 day experimental period. The horses started race training on day one of this trial. Each horse was exercised six days per week and during the day horses were confined to a stable or sand yard (4x4m).

Feed management

The horses were fed twice daily a combination of oats, lucerne chaff, commercial mix (NRM Sweetfeed[®]) bran and red clover hay (details of the feeding programme are presented in Appendix B). The horses had no access to pasture during the trial. Water was available *ad Libitum*. The trainer maintained daily records of the feed offered to each horse throughout the observation period. Feed samples were collected and subjected to analysis (Grace *et al.*, 2003). Diet parameters, Digestible energy (DE) (Mcal/kg), dry matter percentage (DM), crude protein (CP) (g/kg DM), Ash (g/kg DM), Acid detergent fibre (ADF) (g/kg DM), and Neutral-detergent fibre (NDF) (g/kg DM) were experimental analysed.

Training intensity

The horses were exercised at varying intensities for six days a week, unless injured. Each horse had daily training distance and times recorded by the trainer into a logbook for 78 days. From this data, it was possible to calculate mean velocity (m/s) of each horse's exercise session.

Days were categorised as exercise or days of no work. Gait codes (no work, walk, trot, canter, $\frac{1}{4}$ pace, $\frac{1}{2}$ pace and gallop) were translated from mean velocity (m/s) into typical New Zealand trainer gait descriptions (see Table 7). Exercise intensity was divided into no work, medium (trot & canter) and high intensity (quarter pace, half pace and gallop) translated from Table 7.

Table 7 New Zealand gait descriptions in velocity (m/s)*

New Zealand gait descriptions*	Velocity (m/s)
No work	0 (no exercise)
Walk	≤ 2.9 m/s
Trot	3-4.5 m/s
Canter	4.6-8.9m/s
$\frac{1}{4}$ pace	9-12.4 m/s
$\frac{1}{2}$ pace or pace work (PW)	12.5-14.2 m/s
Gallop	14.3 ⁺ m/s

* Adopted from Rogers and Firth (2005).

Data collection

Faecal sampling started when all horses entered the stable to start their three-year-old race training campaign. Faeces were collected daily at 0530 hours prior to the days training. Approximately 200g \pm 5g of faeces per horse was obtained from all faecal deposits in the stable or yard at time of collection. The faecal samples were placed in a sterile plastic bag (Harvey's plastic singlet bags, 255x155x529, 15 μ m), labelled with horse ID, date and stored at 4°C before being transported to the laboratory for analysis. The horses were weighed four times at 21 day intervals throughout experimental period.

Analytical methods

The technique for faecal pH measurement was adapted from Eastwood (2002). 50g \pm 0.2g of faecal matter (Metler toedo college B502 scale) was mixed with 50mls of distilled water until it formed a homogenous suspension. Faecal pH was measured using a pH probe (Denver Instrument Company, Basic pH meter) that was accurate to \pm 0.01pH units and was standardized in buffer pH 4, 6, 10 before faecal measurements were undertaken.

Three faecal measurements were read and daily mean faecal pH taken from these variables. The probe was rinsed with distilled water between measurements.

The weight of feed products (oats, NRM Sweetfeed[®], bran, lucerne chaff and hay) were estimated by measuring the weight of one “typical” scoop or slab of hay (Metler toedo college B502 scale). Average daily roughage to concentrate was calculated by total daily concentrate offered (kg) divided by the total roughage offered (kg). Diet parameters (DE, DM, CP, R: C, Ash, ADF and NDF) were experimental analysis and each parameter was tested against mean faecal pH. Training intensity were categorised and analysis as exercise or days of no work, gait and exercise intensity against mean faecal pH.

Statistical analysis

Data was entered into MS Excel (Microsoft Corporation) and rounded to two decimal places. All analysis is based on the horse daily mean faecal pH value. Data is presented as mean ± standard error. All statistical analyses were performed using SPSS Version 12.0 (SPSS Inc, Chicago, IL, USA).

A General Linear Model (GLM) was used to test the effect of week, diet parameters, training intensity on mean faecal pH. A least significant difference post- hoc test (LSD) was used when a significant difference was identified, for all analysis the significance level was set at $P < 0.05$.

EXPERIMENT TWO: INDEPENDENT DIETS, SAME MANAGEMENT BUT NO EXERCISE

Animals

Faecal samples were collected from ten Thoroughbred yearlings (6 colts and 4 fillies) aged 13-15 months.

Management of horses

The trial was conducted at a commercial Thoroughbred stud, Masterton, New Zealand. Faecal samples were collected for a 12 hour period over four consecutive days. All horses were confined to a stable (4x4m) for approximately 12 hours during the day, and were turned out to pasture for the night. The horses were brought in from paddocks at approximately 0600 hours and turned out to pasture at approximately 1800 hours. During the 12 hour confinement to the stable the horses were fed, groomed, and handled as part of the typical Thoroughbred yearling preparation programme.

Feed management

When confined to the stable the horses were fed three daily feeds containing, lucerne chaff, commercial mixes (NRM Evolve[®], NRM Prepare[®] and NRM Equijewel[®]) and meadow hay. A feed record was maintained throughout trial period (Refer to Appendix C for feed programme). Only concentrate feed samples were collected and DE (Mcal/kg) analysed. A typical scoop of feed or slab of hay was weighed (EKS, 49:01F MA portable scale ($\pm 0.5\text{kg}$)) as basis for the estimate of amount of feed offered (kg) to each horse. Amount of pasture eaten during the period 1800 hours-0600 hours was not included in total feed weight during analysis of feed management.

Data collection

Faecal samples were collected at 0730, 1200, 1500, 1730 hours. Approximately 200g±5g of faeces per horse was obtained from all faecal deposits in the stable at the time of collection. The faecal samples were placed into a sterile plastic bag (Harvey's plastic singlet bags, 255x155x529, 15µm) and labelled with horse ID. Faecal samples were analysed within an hour of collection using the analytical methods described previously.

Statistical analysis

The weight of feed offered was estimated from the weight of a typical measured scoop for each of the feed products offered. All data was entered in MS Excel (Microsoft Corporation) and rounded to two decimal places. All analysis is based on the horse daily mean faecal pH value.

A GLM tested the effect of day, time of faecal collection, gender, DE, and R: C on mean faecal pH. A least significant difference post-hoc test was used when a significant difference was identified, for all analysis the significance level was set at $P < 0.05$.

EXPERIMENT THREE: SURVEY OF HORSES, WITH DIFFERENT MANAGEMENT, DIET AND EXERCISE PROGRAMME

Trainers

Sixteen trainers were surveyed from Auckland (n=9), Taranaki/Wanganui (n=3) and Palmerston North (n=4). A letter advertising the survey was sent to all licensed trainers in the respective regions. Participants were then chosen at random from the list of licensed trainers published in the 2004 New Zealand Thoroughbred Monthly magazine.

Animals

Information was obtained about each horse's diet, workload and behaviour. Approximately 200g±5g of faeces per horse was obtained from all faecal masses in the horse's stable or yard at the time of collection. The faecal samples were placed in a sterile plastic bag (Harvey's plastic singlet bags, 255x155x529, 15µm) and labelled with a unique horse ID. Faecal samples were analysed within an hour of collection using the analytical methods described previously.

Survey questions

Trainers were categorised according to age, the number of years training horses and the number of horses at the stable. For each horse that had a faecal sample collected the trainer was asked horse statistics (sex, age, number of weeks in work and racing class), stable management, feed management, training intensity and horse behaviour (eating and stereotypic) (Refer to Appendix D for survey questionnaire). All data parameters were grouped into logical categories for analysis.

Statistical analysis

A GLM was used to test the effect of horse, trainer, stable management, feed management, training intensity, and horse behaviour. A least significant difference post-hoc test was used when a significant difference was identified, for all analysis the significance level was set at $P < 0.05$.

Results

EXPERIMENT ONE: TESTING SAME DIET AND MANAGEMENT

Horse

The mean faecal pH of the horses was $\text{pH } 6.40 \pm 0.01$. There was a significant effect of day, weeks and horse on mean faecal pH ($P < 0.05$). There was large between-horse variation (Figure 1) and variation in mean faecal pH between days (Figure 2). A post-hoc test (LSD) identified that horses in Week two had a more acidic mean faecal pH than any other week except Week three ($P = 0.134$) (Figure 3).

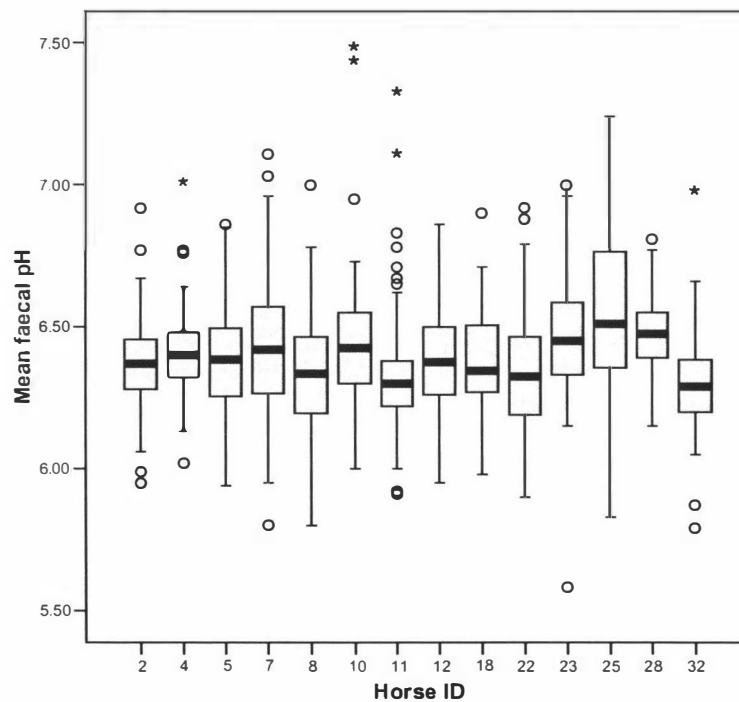


Figure 1. Mean faecal pH of 14 Thoroughbred racehorses over an 84 days observation period.

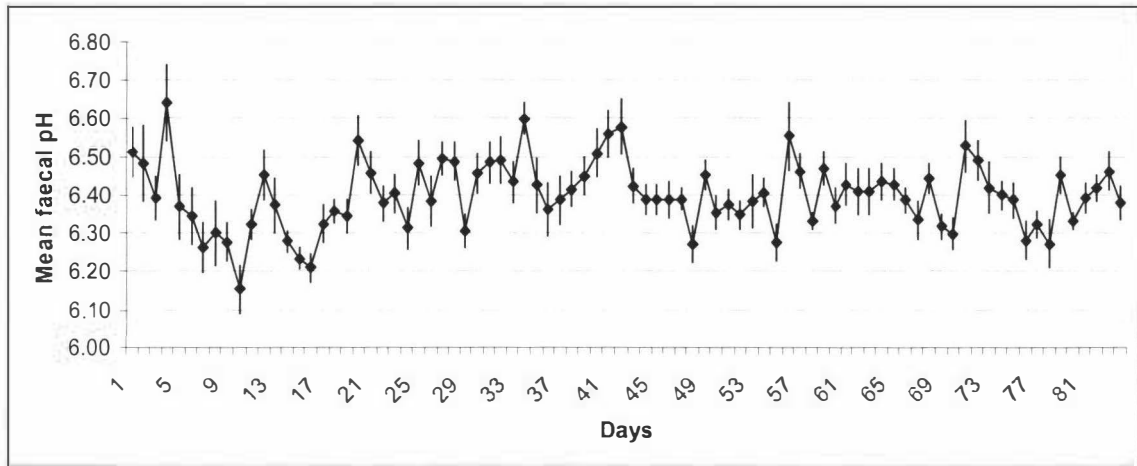


Figure 2. Mean faecal pH of 14 Thoroughbred racehorses in training over an 84 day observation period.

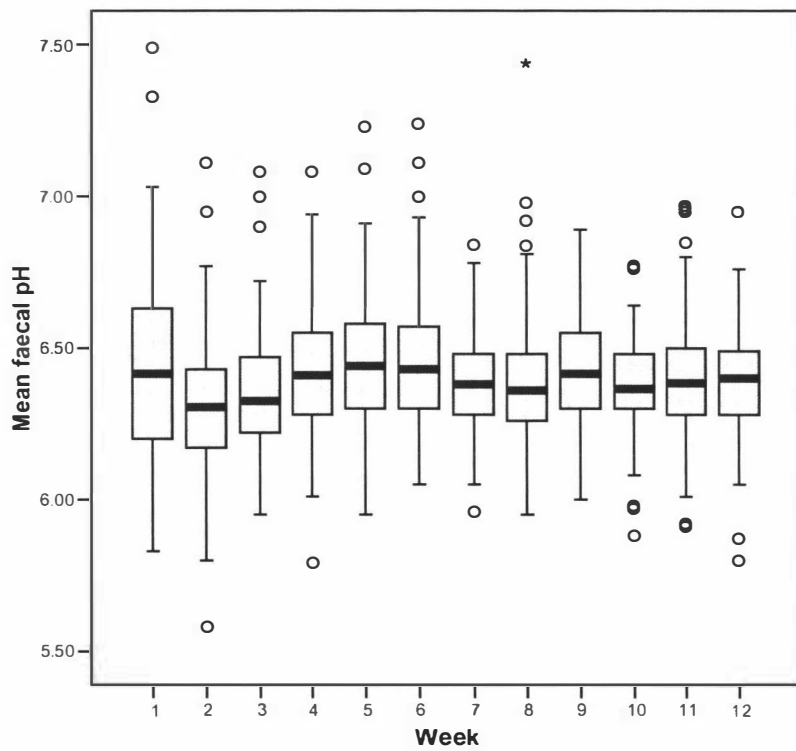


Figure 3. Mean faecal pH of 14 racehorses in training for 84 day (12 weeks) observation period.

Week two was significantly different to all weeks except Week three ($P < 0.05$).

Diet

Mean horse weight over the observation period was 430 ± 4.16 kgs; this calculation was used to compare NRC horse feed requirements to actual feed offered. Diet parameters were calculated according to total amount of DE, DM, CP, Roughage to concentrate ratio, ash content, ADF and NDF offered in the different feed programmes throughout the experimental period, and presented in Table 8. All diet parameters during the different feed programme were tested against mean faecal pH, all had a significant effect on mean faecal pH ($P \leq 0.05$), but only explained 0.3 -2.1% of the variance.

A nutrient analysis resulted in the horses offered on average; 8.76kg total feed, this was divided into 2.08 ± 0.02 kg hay and 5.23 ± 0.01 kg concentrate per day (35:65%, mean roughage: concentrate percentage) and a mean of 2.56 Mcal/kg DE over the experimental period. Indicating, that the NRC (1989) horse nutrient requirements for three-year old horses with a mean body weight of 430 ± 4.16 kgs in intensive race training was under nutrient requirements for total feed weight, R:C and DE, while CP intakes were higher than requirement.

Table 8. Nutrient analysis of diet, number of days 14 horses were on a different feed programme and mean faecal pH a 84 day observation period.

Number of days on different diet programme	Total Feed weight (kg)	DE (MJ/kg DM)	DM (%DM/kg)	CP (g/kgDM)	R: C (%)	Ash (g/kgDM)	ADF (g/kgDM)	NDF (g/kgDM)	Mean faecal pH \pm SE
19	8.13	8.40	84.55	15.07	35:65	43.07	137.37	222.05	6.49 \pm 0.02
1	7.46	11.16	84.80	15.25	38:62	39.33	130.33	209.66	6.17 \pm 0.22
5	8.01	11.29	84.66	15.26	35:65	42.07	136.17	220.25	6.31 \pm 0.02
38	8.95	11.29	84.68	15.33	38:68	45.92	150.08	238.02	6.41 \pm 0.01
21	7.79	11.29	84.80	15.64	35:65	40.21	131.84	214.47	6.37 \pm 0.01
Mean	8.76	10.70	84.70	15.31	35:65	42.12	137.16	220.89	6.35 \pm 0.06
NRC requirements	9*	13.46		2.86**	40:60				

* At 2% body weight (kg) for a horse working at high intensity above maintenance averaging 430 ± 4.16 kg

** At 60g/430kg horse body weight=25.8g. For g/kg= total requirement/total feed weight (25.8/9) = 2.86g/kg.

Training

During the monitoring period the horses were exercised for 78 days following a typical New Zealand Thoroughbred training regime. There was no difference in mean faecal pH between days of no exercise and days of exercise ($P=0.461$), the pace of exercise (no work, trot, canter, $\frac{1}{4}$ pace and $\frac{1}{2}$ pace) ($P=0.720$), or exercise intensity per day (rested, medium and high intensity) ($P=0.531$). There was a trend for mean faecal pH to be more acidic with increasing exercise intensity but large variation in mean faecal pH was observed in the horses working at high intensity (Figure 4).

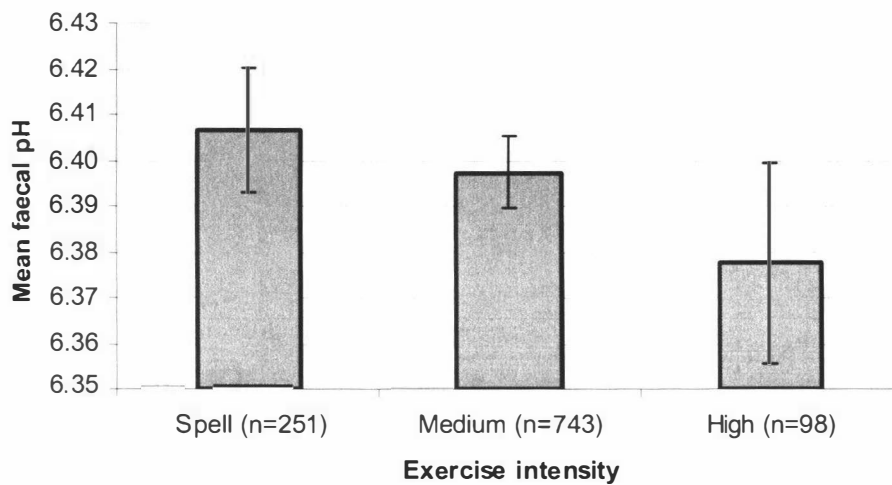


Figure 4. The effect of exercise intensity on mean faecal pH for 14 Thoroughbred racehorses for a 78 day observation period.

EXPERIMENT TWO: INDEPENDANT DIETS, NO EXERCISE BUT THE SAME MANAGEMENT

The mean faecal pH of the ten horses was pH 6.61 ± 0.05 with large between-horse variation. There was no significant difference in faecal pH between the six colts (pH 6.58 ± 0.07) and four fillies (pH 6.65 ± 0.07), between day of collection or time of faecal collection. The amount of concentrate offered (kg), total weight and R: C of the diet offered to the horses is presented in

Table 9. There was no difference in total weight of feed offered, while R: C ratio and weight of concentrates offered explained respectively 4% and 9% of the mean faecal pH variance.

Table 9. Summary data of feed offered and mean faecal pH for 10 horses with independent diets, same management but no exercise.

Horse ID	Total conc. (kg)	Total feed weight (kg)	R: C (%)	Mean faecal pH \pm SE
1	4.89	10.27	52:48	6.75 \pm 0.11
2	6.19	11.56	46:54	6.49 \pm 0.12
3	5.61	10.99	49:51	6.73 \pm 0.22
4	7.96	13.34	40:60	6.24 \pm 0.04
5	4.29	9.67	56:44	6.94 \pm 0.20
6	7.77	13.14	40:60	6.25 \pm 0.08
7	4.15	9.53	56:44	6.84 \pm 0.08
8	2.38	7.75	69:31	6.57 \pm 0.08
9	7.62	12.99	41:59	6.95 \pm 0.19
10	5.34	10.71	50:50	6.32 \pm 0.11

EXPERIMENT THREE: SURVEY OF RACEHORSES, WITH DIFFERENT MANAGEMENT, DIET AND EXERCISE PROGRAMME

Trainer Effect

The trainer's age and the number of years training horses had no effect on horse mean faecal pH ($P > 0.05$).

Horse Effect

A total of 140 horses were surveyed, 71 were colts or geldings (pH 6.41 ± 0.04), and 69 were mares or fillies (pH 6.46 ± 0.04). There was no effect of horse age ($P = 0.196$) or gender ($P = 0.432$) to mean faecal pH (Figure 5). There was no significant difference in mean faecal pH due to racing grade ($P = 0.77$) Table 10.

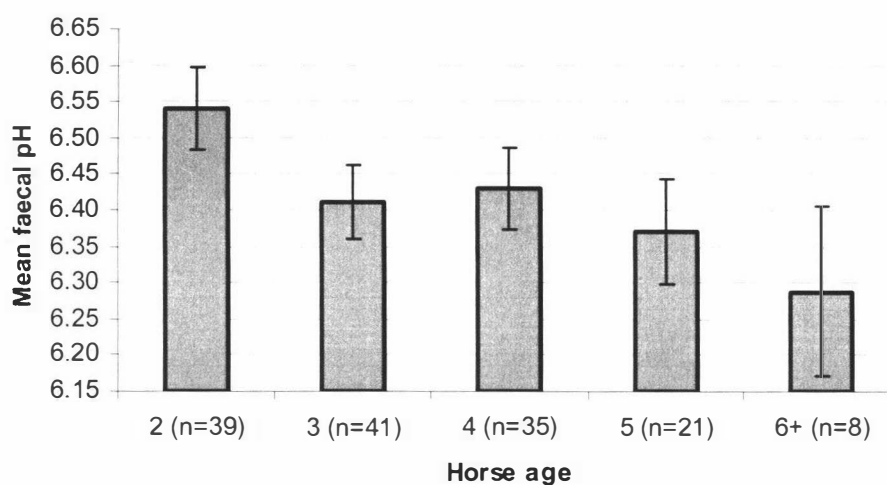


Figure 5. The effect of horse age on mean faecal pH for 140 Thoroughbred racehorses sampled from 16 training stables.

Table 10. Descriptive data of the racing grades of the 140 Thoroughbred racehorses surveyed.

Horse racing grade	N	Mean faecal pH \pm SE
Unraced	62	6.51 \pm 0.05
Maiden	55	6.40 \pm 0.04
Progressive	15	6.36 \pm 0.08
Open	7	6.19 \pm 0.11
Hurdler	1	6.30 \pm 0.00

Stable Effect

The number of horses at each stable varied from 8-41 (mean 24) horses. The number of horses at the stable and number of horses sampled within the stable are shown in Table 11. Horse faecal pH measurements were made from an average of 9 horses, range 4-14 horses in each stable.

Table 11. The number of horses and number of horses sampled within the surveyed training stables.

Trainer ID	1	2	3	4	5	6	7	8	9	10	11	12	13	14	15	16
N horses at stable	9	30	25	14	9	17	40	41	25	40	12	24	12	8	30	36
N horses sampled	4	7	13	8	6	9	12	13	8	12	4	14	7	8	10	6
% sampled	44	23	52	57	67	53	30	32	32	30	33	58	58	100	33	17

Stables with ≤ 20 horses ($n=46$) had significantly more acidic mean faecal pH (pH 6.34 ± 0.59) than stables with >20 horses ($n=94$: pH 6.48 ± 0.59 ; $P=0.017$). When stables were grouped into four categories based on horse numbers (Table 12) there were large differences in mean faecal pH between groups ($P=0.001$). A post-hoc test identified significant difference between all categories except stables with 25-36 and 37⁺ ($P=0.683$). Stables with 1-12 horses appeared to have more acidic mean faecal pH than horses from other stables. Bedding type (straw, saw dust, sand and pasture) had no significant effect on mean faecal pH ($P=0.487$).

Table 12. Mean faecal pH and total number of horses in stables grouped into four categories.

Number of horses in stable	N	Mean Faecal pH \pm SE
1-12	29	6.15 \pm 0.05
13-24	30	6.59 \pm 0.05
25-36	44	6.42 \pm 0.04
37+	37	6.55 \pm 0.05

Ninety-seven percent of the horses surveyed, were confined to an area less than 4x4m for greater than 12 hours per day. There was no difference in mean faecal pH between horses turned out daily to a paddock (n=70, pH 6.46±0.04) or remaining in a stable or small yard with no access to pasture (n=70, pH 6.41±0.04; P=0.445). There was a significant effect of the number of hours (0, 4, 5, 6 or 9⁺ hours) horses were turned out daily to a pastured paddock (P= 0.001). The post- hoc test identified horses turned out for four hours had significantly less acidic mean faecal pH (P= 0.001) than any other time horses were turned out (Table 13).

Table 13. The amount of time 140 racehorses were turn out to pasture on mean faecal pH.

Time at pasture (hours)	N	Mean faecal pH ± SE
0	70	6.41±0.04 ^a
4	13	6.84 ±0.05 ^b
5	25	6.44±0.05 ^a
6	24	6.32 ±0.08 ^a
9 ⁺	8	6.29 ±0.09 ^a

Different superscript are significantly different (P=0.001)

Feed Management

The type of concentrated feed offered to horses, commercially prepared (n=46, pH 6.41±0.05), grains (n=19, pH 6.28±0.07) or mixed feed (mixture of both commercially prepared and grains) (n=75, pH 6.49± 0.04) altered mean faecal pH (P=0.03). A post- hoc test identified only a significant difference in mean faecal pH between mixed and grains diets (P= 0.011). The frequency of feeding two, three, four or five meals a day had no significant effect on a mean faecal pH (Figure 6).

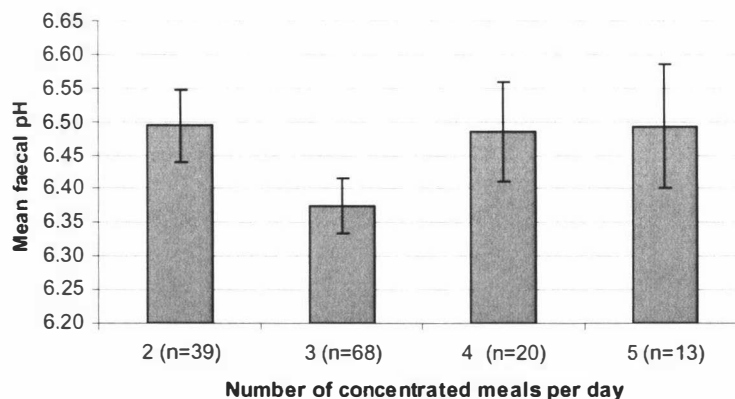


Figure 6. The number of concentrated meals per day on mean faecal pH of 140 horses.

The trainers offered on average 5.55 ± 0.14 kg concentrates per day. Mean faecal pH was significantly altered ($P=0.001$) by the quantity of concentrate offered per day but only explained 1.7% of the variance (Figure 7). Horses categorised by stable size were offered less concentrate feed in relation to increasing stable horse numbers (Table 14).

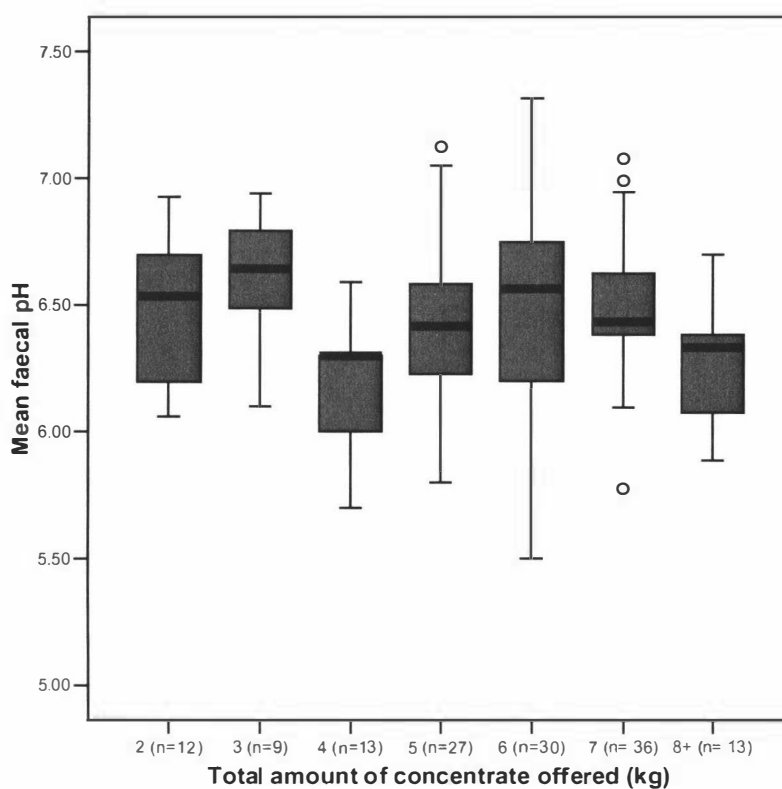


Figure 7. Box and whisker plot of the mean faecal pH and the different quantities of concentrate offered per day.

Table 14. A matrix of significant difference is presented below. * = ($P < 0.05$)

Amount of concentrate offered (kg)	2	3	4	5	6	7
3	-					
4	*	*				
5	-	-	*			
6	-	-	*	-		
7	-	-	*	-	-	
8+	-	*	-	-	*	*

Table 15. Mean amount of concentrate offered to horses in relation to categorized stable number.

Number of horses in stable	Number	Mean concentrate offered ±SE (kg/day)
1-12	29	6.57±0.11
13-24	30	5.99±0.36
25-36	44	5.34±0.23
37 ⁺	37	4.64±0.24

On average a typical slab of hay weighed 2.25±0.07kg and all trainers surveyed offered hay daily to every horse. There was a significant difference in mean faecal pH due to the amount of hay offered to the horse per day (P=0.003) (Table 16). Mean faecal pH was significantly more acidic (P=0.001) for horses given less than 2.25 kg of hay per day compared to horses given more than 2.25 kg.

Table 16. The effect of amount of hay offered to 140 horses on mean faecal pH.

Amount of hay (Offered daily) (kg)	N	Mean faecal pH ± SE
1.69 (0.75 slab)	7	6.00 ^a ±0.12
2.25 (1 slab)	84	6.46 ^b ±0.03
3.38 (1.5 slabs)	12	6.37 ^b ±0.09
4.50 (2 slabs)	37	6.49 ^b ±0.05

Different superscripts are significantly different (P=0.001). Assumes on average 1 slab of hay is approximately 2.25 kg

There was no significant difference in mean faecal pH if horses were offered hay once-a-day (n=76 pH 6.42 ± 0.04) or twice daily (n=51 pH 6.46 ± 0.40) (P=0.555). However the time when the hay was offered had a significant effect on mean faecal pH (P=0.002). A post- hoc test identified no consistent pattern between the times hay was offered to horses and mean faecal pH (Table 17) There was no significant effect on mean faecal pH to concentrate: roughage ratio.

Table 17. Time of day hay was offered to 140 Thoroughbred racehorses.

A matrix of significant difference is presented below. *=(P<0.05)

Time hay offered	Hay offered daily	N	Mean faecal pH +SE
Morning (AM)	1	23	6.25 ±0.07
Mid morning (MM)	1	13	6.49 ±0.09
Lunch (L)	1	30	6.58 ±0.06
Mid afternoon (MA)	1	19	6.33 ±0.07
Evening (PM)	1	4	6.42 ±0.16
Morning and evening (AM/PM)	2	33	6.50 ±0.06
Mid morning & mid afternoon (MM/MA)	2	10	6.54 ±0.10
Mid morning & evening (MM/PM)	2	8	6.19 ±0.11

Time hay offered	AM	MM	L	MA	PM	AM/PM	MM/MA
MM	*						
L	*	-					
MA	-	-	*				
PM	-	-	-				
AM/PM	*	-	-	*	-		
MM/MA	*	-	-	-	-	-	
MM/PM	-	*	*	-	-	*	*

All trainers surveyed used supplements and electrolytes. Fourteen trainers used a scoop to measure feed. One trainer weighed out each feed, and one poured an approximate quantity into feed bins.

Training

There was no difference in mean faecal pH ($P=0.480$) due to the number of weeks the horses had been in race training (Figure 8). Galloping two or more days a week, or horses performing high intensity exercise (pace work, jumping and galloping) for the greater than three days a week, did not alter mean faecal pH.

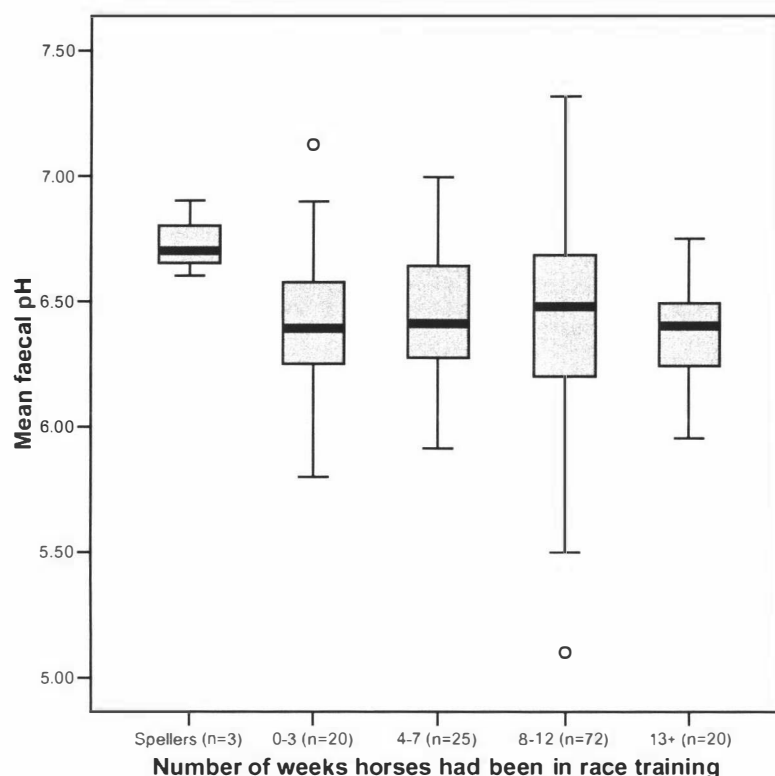


Figure 8. Box and whisker plot of the mean faecal pH and the number of weeks 140 racehorses had been in race training.

Horse Behaviour

There was no significant difference in mean faecal pH due to horse eating behaviours, normal ($n=114$; pH 6.45 ± 0.03), fast ($n=16$; pH 6.32 ± 0.11), or slow ($n=10$; pH 6.46 ± 0.35 $P=0.344$). However, six horses that expressed stereotypes (crib biting, weaving, wind sucking or box walking) had a mean faecal pH (pH 6.70 ± 0.35) significantly less acidic than horses ($n=134$) that were observed by trainers to never express stable vices (pH 6.43 ± 0.29 $P=0.043$).

Discussion

The aim of this study was to identify variables that contribute to the variation in faecal pH within a population of intensively managed Thoroughbred horses.

Sample population- trainers

The sixteen trainers sampled represented a typical cross section of trainers in the New Zealand Thoroughbred racehorse industry. In the 2003/2004 season there were 1436 racehorse trainers in New Zealand, 80.5% were resident in the North Island (NZRB, 2004). All of the trainers in the current survey were from the North Island of New Zealand. South Island trainers had the opportunity to participate in the survey but none responded to letters, faxes or phone calls. All of the trainers surveyed were licensed with New Zealand Thoroughbred Racing, eleven were registered as public trainers and five were registered as owner/trainers for the 2003/2004 season.

During the same racing season 25% of all trainers in New Zealand were licensed as public trainers, 48.5% as owner trainers and 26.5% as 'permits to train' holders. It is highly unlikely that the over-representation of public licence trainers in the survey would have had a biasing effect on the results of the survey. Within New Zealand, the public trainers train the majority of the horses, as the permit to train and owner trainers are restricted by racing regulations as to the number of horses they can train. There was also very subtle variation between trainers in horse management and it is highly probably that differences in mean faecal pH of horses between trainers is more attributed to individual horse variation than the type of training licence held.

The trainer's age and number of years training horses were used as indicators of training experience, neither of which had an effect on faecal pH. This survey found racehorse training as a profession had an aged population. Only two trainers surveyed were younger than 30 years of age. A possible explanation for an older population could be that younger people are in other employment (such as jockeys) within the industry and are gaining valuable industry experience before taking over training responsibilities.

Sample population- horses

Thoroughbred racing is predominantly based in the North Island. The NZRB (2004) reported that 80% of the 8982 Thoroughbreds in race training during the 2003/2004 racing season were with a trainer based in the North Island. Of the current study, 14 Thoroughbred horses in race training were sampled repeatedly from one stable and 140 Thoroughbred horses sampled once were all Thoroughbred horse trainers throughout the North Island of New Zealand. It was unfortunate that trainers from the South Island were not willing to participate in the survey. However, as the majority of the racing and the horses are based in the North Island the lack of South Island participation does not compromise generalisation of the survey results to the wider New Zealand racing community.

There is limited national and international data on stable horse numbers in Thoroughbred racing establishments. This study surveyed 16 racing Thoroughbred trainers throughout New Zealand and the number of horses at each stable ranged from 8-41 (median 25). The number of horses in a trainer's stable may represent the financial position, racing success or marketing ability. The latter two being more relevant for trainers registered as public trainers as they may rely on racing success and marketing ability to encourage new clients. This survey used the number of horses in a stable as an indicator of trainer management. Mean faecal pH was more acidic in stables consisting of equal to and less than 20 horses than stables with greater than 20 horses. Interestingly, stables with ≤ 2 horses were more acidic in relation to mean faecal pH than stables with increasing number of horses. This study also identified that horses in smaller stables were offered increasing amounts of concentrate per day than horses within larger stables. A possible anecdotal explanation is trainers of a larger number of horses may offer a less individualised feeding programme and have greater financial constraints, both which would discourage the over-feeding of horses in their care.

Experiment one and two identified significant between-horse variation in mean faecal pH. Aitchison *et al* (1987) concluded that hindgut acidosis does not occur consistently in all animals given an equal quantity of carbohydrate, and it is common to see between-horse fluctuations such as those seen in the study. Between-horse variation could be explained by the differing ability to digest NSC due to variation in microbial populations at the hindgut (Mackie and Wilkins (2002), Medina *et al* (1993), Meyer *et al* (1995) and Rowe *et al* (1995)). Between horses, there may be a range of microbial species producing variations in digestion and fermentation of NSC and resulting in varied levels of faecal pH.

Feed management

The variation in the diet of the racehorses followed in Experiment 1 and 2 had no significant effect on mean faecal pH. This is supported by Richards *et al* (2004) who also found diet had no significant effect on faecal pH. Thus, the diet fed did not appear to alter microbial environment significantly to challenge horse hindgut acidity during the experimental period.

New Zealand trainers tended to feed a greater quantity of oats over barley and maize (corn) as the main form of grain offered, similar to racehorses in Sydney and Detroit (Gallagher *et al.*, 1992; Southwood *et al.*, 1993a). Feeding grains (pH 6.28±0.07) as the main form of concentrate was associated with more acidic faecal pH than offering commercially prepared or mixture of both grains and commercially prepared feeds. Willard *et al* (1977), Radicke *et al* (1991), McLean *et al* (2000) and de Formbelle *et al* (2001; 2003) have found hindgut acidosis can occur by overloading horses with grains such as oats, barley and maize. A possible explanation is that the transit of grains through the gastrointestinal tract is faster due to smaller particle size. The quantity of grain fed may have exceeded pre-caecal digestion and undergone rapid fermentation in the hindgut resulting in faecal samples that was more acidic (Frape, 2004).

Commercially prepared feed ingredients are further mechanically processed to achieve a feed mix that binds together to make a feed that is convenient for the horse owner, palatable for the horse, meets horse nutrient requirements but does not exceed pre-caecal digestion (Frape, 2004; Lewis, 1995). The less acidic faecal pH observed with horses offered commercially prepared and mixed diets may not have exceeded pre-caecal digestion due to slower transit time through the gastrointestinal tract.

The typical amount of concentrates offered to Thoroughbred racehorses was 5.55 kg \pm 1.62 SD, and is comparable to the amount offered to Thoroughbred racehorses surveyed in Detroit (5.6 kg \pm 0.6SD) by Gallagher *et al* (1993a). Greater quantities of concentrate were offered to Thoroughbred racehorses in Sydney (7.8 kg \pm 1.6 SD) (Southwood *et al.*, 1993a). Many studies have used a range of concentrate weights to induce excessive hindgut fermentation. It is reported if concentrates are fed at greater than 4g NSC/kg horse body weight per meal (Potter *et al* (1992) in Hoffman *et al* (2001)) or even as low as 2g NSC/kg horse body weight/meal the amount of NSC overwhelms the digestive capabilities of the small intestine and causes excessive fermentation at the hindgut (Cuddeford, 2001). In this study, faecal pH was significantly altered by amount of concentrate offered but had inconclusive results as to how much concentrate (kg) induced significant faecal acidosis (pH \leq 3.2). In hindsight, a more appropriate approximate measure would have been to determine the amount of NSC per meal/horse body weight to enlighten in the effect on faecal pH. However, in the use of NSC per meal/horse body weight, obtaining accurate horse weights from each stable would be a limitation.

Hay is believed to play an essential role in buffering and preserving the digestive functions of the hindgut and horses fed a 100 % hay diet produce microbial lactic acid in a uniform fashion throughout the caecum and colon, decreasing hindgut acidity (Waller *et al.*, 2003). A possible explanation is, hay has a high dry matter content (85-95% DM) (McDonald *et al.*, 2002; NRC, 1989) which increases saliva secretion influenced by forage mastication, which buffers the acidity produced by VFA's and lactic acid (Alexander and Hickson, 1970; Nicol *et al.*, 2002). Secondly, forage influences a more favourable VFA production (higher acetate and lower propionate ratios) which positively

influences plasma acid-base status, absorption of water, other fermentative products and reabsorption of bicarbonate across intestinal mucosa during fermentation (Frape, 2004; Gill and Lawrence, 2003; Hammond *et al.*, 1986).

All the trainers provided varying quantities of hay, which significantly influenced mean faecal pH. It appears that providing ≥ 2.25 kgs (assuming one slab of hay is 2.25kg) of hay may be a threshold to minimise faecal acidity. This view is supported by no relationship between R: C ratio and faecal pH. Possible explanations could be due to fibre content or gastrointestinal capacity being the first limitation. Also in the current study, mean faecal pH was significantly influenced by intermittent times hay was offered through out the day. There was no clear trend to identify the best time of day to offer hay, thus indicating that it is the feeding of the hay and not the time of the hay feeding that is important.

Increasing the frequency of feeding a concentrated diet is believed to increase nutrient digestion, reduce time between fasting, reduce transit time of digesta and reduce stereotypic behaviour or gastric ulceration (Frape, 2004; Gill and Lawrence, 2003; Hammond *et al.*, 1986). However Gill and Lawrence (2003) found feeding frequency did not affect digestion of starch, CP, DM, CF, ADF or ash. The feeding frequency of concentrates had no effect on faecal pH. Horses appear to be more tolerant to feed frequency than indicated by the literature. A level of tolerance may be due to horse adaptation to restricted feeding regimes and hay available *ad libitum* (Ralston and Baile, 1983). Another possibility is that the horses are pre-conditioned to few but large meals. Most Thoroughbred horses have been raised at commercial stud farms where during weaning and yearling preparation many would have been exposed to restricted feeding regimes.

Horse management

There is little variation in Thoroughbred racing stable management. Ninety four percent of Thoroughbred horses surveyed were stabled (in an area $\leq 4 \times 4$ m) for twelve or more hours per day and 50% of the horses surveyed had no access to pasture and free exercise. This is similar to other countries where the majority of racehorses are maintained in a

stable (Haupt, 1995). It has been suggested that straw may serve as source of forage for horses over other bedding types (Haupt, 1995). However bedding type did not influence mean faecal pH, which would indicate that bedding did not serve as a source of forage for horse in the current study.

The attribute to the success of the New Zealand Thoroughbred racehorse is thought to be due to the access to pasture (Wallace, 1977). The current study showed trainers limited horse access to pasture from 4 to 9 hours. This was in agreement with Huntington and Jenkinson (2003) that trainers have access to pasture but prefer to limit horses time at pasture to prevent excessive gut fill. Confinement and limited access to pasture is similar to other racing countries but contrasts the popular opinion that New Zealand racehorses have greater access to pasture. In recent years the cost of land has increased and many racing clubs have reduced the pasture land available to racehorse trainers. Thoroughbred racing is an international activity and it appears that many aspects of the management of the racehorse are similar across the different countries, some trainers may have consciously removed access to pasture in an attempt to make management of horses at home as similar to the possible management conditions experienced when racing in Asia and Australia.

Davies *et al* (2000), L. Paul unpublished data in Cuddeford (2001) and Eastwood (2002) reported a less acidic faecal pH with horses kept on pasture compared to horses not kept on pasture. It is proposed that access to pasture and grazing may increase oral stimulation of saliva and healthier digestive function of digesta through the gastrointestinal tract and neutralising gastric acidity and excessive fermentation (Alexander and Hickson, 1970; Frape, 2004; Nicol *et al.*, 2002). In contrast, horses in this study turned out to pasture did not show significant benefits in relation to mean faecal pH compared to horses not turned out to pasture daily. However, the horses turned out for four hours had significantly higher mean faecal pH, but were from the same stable and the observation may be due to access to pasture or other management practises associated with that stable.

Workload

Workload appears to have no effect on mean faecal pH. Biologically the direct influence of skeletal L (+) lactate on faecal pH is limited, as it would be directly utilised by skeletal muscle for ATP production. This study highlighted there was no difference in mean faecal pH related to the number of weeks horses had been in race training.

Horse behaviour

Both Johnston *et al* (1977) and Willard *et al* (1998) found if faecal pH falls below 6.3 units behavioural abnormalities have a higher prevalence. This study reported a higher faecal pH (pH 6.70 ± 0.35) for horses associated with stereotypic behaviours despite a small number of horses sampled. Johnston *et al* (1977) and Willard *et al* (1977) proposed that horses perform stereotypic behaviours to overcome acidosis or abdominal discomfort. Horses may initially perform stereotypic behaviour to overcome abdominal discomfort but may continue to perform a particular behaviour as a stable stress mechanism, as a method for endorphin release or relieve boredom (Nicol, 2002; Pell and McGreevy, 1999). In context to this study it is highly probable these factors were driving the more alkaline faecal pH in the stereotypic horses.

In the current study there was no effect of horse feeding behaviour in relation to mean faecal pH, but interestingly horses that were considered fast/greedy eaters had a faecal pH of 6.32 ± 0.11 that was more acidic than slow/picky and normal eaters (pH 6.46 ± 0.35 and 6.45 ± 0.03). There is no other evidence in the literature relating eating behaviour and faecal pH. However, fast/greedy eaters may be associated with poor mastication and incomplete pre-caecal digestion. Thus larger quantities of complex carbohydrate reaches the hindgut and causes excessive fermentation resulting in more acidic faecal pH than horses that are picky/slower or normal eaters.

Conclusion

In contrast to common opinion, New Zealand Thoroughbred racehorse management is similar to practises used in other racing countries with long periods of confinement and limited or no access to pasture. Feeding practices in New Zealand were similar to Australian and the U.S.A indicating that racehorse management is similar between the major racing countries.

This study identified a more acidic mean faecal pH (pH \leq 6.32) was associated with stables that were small (1-12 horses), horses offered grains as main form of concentrated and offered <2.25 kg hay/day. Mean faecal pH was significantly more alkaline when horses with known stereotypic behaviours were identified despite sample numbers being small. Large between-horse variations in mean faecal pH may have limited the capacity of the study to significantly identify subtle relationships between some management parameters and faecal pH. Mean faecal pH was not affected by trainer age and experience, diet variables (DE, DM, CP, Ash, C: R, ADF and NDF), feeding frequency, bedding type, number of hours horses were turned out to pasture and exercise workload.

The lack of relationship between dietary variables and total amount of concentrate (kg) offered in relation to faecal pH may be better represented by the amount of NSC per meal/horse body weight. However, more comprehensive data is required to accurately determine if mean faecal pH is affected by amount of NSC per meal/horse body weight. Further research is required to identify and understand management of racehorses in smaller stables, and the influence of forage, especially hay quantity, type and digestibility in relation to faecal pH to identify a threshold of hay that will buffer hindgut acidosis. An interesting observation was horse feeding behaviour and stereotypic vices on faecal pH. Further research is required to identify if horses with unusual eating patterns and stereotypic behaviours are trying to positively influence hindgut acidosis.

Terminology

Acidic– When pH values are less than pH 7, but in context to this study, acidic is deemed when a pH equal to or lesser than pH 5 than pH 7

ADF – Acid-detergent fibre, proximate analysis of the components of cellulose, lignin, silica and lignified nitrogen in feed samples

Agistment– The payment for grazing and keeping of a horse out of training or racing

Anabolism– Energy requiring metabolic reaction where metabolites are built from small metabolic molecules to larger molecules (McGreevy, 1995; Nicol, 2002)

Alkaline– When pH values are greater than pH 7.

ATP– Adenosine triphosphate

BW – Body weight in kg

Breaking in – To accustom a young horse to racing equipment, methods and to carry a rider

Catabolism– Metabolic reaction where larger complex organic molecules are broken down to smaller molecules with a release of energy (Pagan, 1997)

Calorie – 1 cal = 4.184 joules of energy or 1 Mcal = 4.18 MJ

Colic – A blockage or distension of the intestine

Colt – Entire male horse less than 4-years of age

Commercial feed – A purchased feed type that is a prepared mix of grains, mineral and vitamins also named a sweetfeed or hardfeed. A commercial feed is specifically advertised for a particular equine use.

Concentrate feed – Is a general term given to horse feeds that has high energy content and is highly digestible. With the majority of ingredients being commercial prepared and /or grains, that may be further processed (rolled, crushed, bruised) to enhance digestion in the horse.

Confined/confinement – No immediate access to pasture.

Coprophygy– Consuming faecal material

CP – Crude protein, standard protein contain 16g nitrogen per 100g (crude protein percentage can be estimated from nitrogen percentage x 6.25) (Mason, 1991)

Crib-biting – The horse seizes a fixed object with its teeth, pulls back and draws air usually producing a characteristic grunt noise. The behaviour is very similar for any given horse. Crib-biting does not involve aerophagia, as inhaled air is expelled again via the mouth (McDonald *et al.*, 2002).

DCP – Digestible crude protein, another name for CP

DE – Digestible energy is the amount of energy in the diet that is absorbed by the horse (minus energy utilised for digestion, fermentation, faeces and gas production) (Nicol, 2002)

Deaminated– amino acid nitrogen group removed from protein

DM – Dry matter percentage of the feed sample

Easy work rate– Horses training that is slow and non intensive. A horse working more than 3 days a week trotting, walking, and swimming

FFA– Free fatty acids

Faecal deposition –Faecal droppings

Faecal mass – Accumulation of all faecal depositions

Faecal sample–50g of faecal matter gathered from a faecal mass

Farrier– person who trims hooves or puts horse shoes on horses

Filly– Entire female horse less than 3-years of age

Gelding – Castrated male horse

GDP– Gross Domestic Product

Grains– A horse feed consisting of harvested grains in natural form, that may be processed (rolled, crushed, bruised) to enhance digestion in the horse

Hard work rate– Horses training/racing at fast, long or short and intensive pace work. A horse working more than 3 days a week at cantering, $\frac{1}{4}$, $\frac{1}{2}$ pace, jumping and gallop work

High intensive work– Horse race training at 9-16⁺ m/s velocity for 3 and more days a week Jump out races– Work out races- training/practices races

High pH– pH of faecal material to be closer or equal to pH 7

Kg– Kilogram (1kg= 1000grams (g))

KJ – Kilojoules (1 KJ=1000 joules (J)) (KJ*1000=MJ)

Km= Kilometre (1 km=1000 meters (m))

Lb– 1lb= 2.2 Kg

Light work– Horse in race training at a speed of 0- 4.5 m/sec for 3 and more days a week

Low faecal pH – pH of faecal material to be less than or moving closer to pH 5.5

m/ s= Meters per second

Mare – Entire female over 3 years of age.

Mcal – Megacalories (4184 joules of energy)

Medium intensive work– Horse race training at 5-8 m/sec velocity for 3 and more days a week

Mixed feed –A term given for this report to identify horses fed a diet composing of a commercial feed and grains

MJ – Megajoule (1 MJ= 1000 KJ)

NZRB– New Zealand Racing Board

Non TAB races– No betting and prize money- trial races

NSC– non-structural carbohydrates (NSC= 100- (CP + ether extract + NDF + ash)

Pace work – ¼ and ½ pace (9-14.2 m/s)

Racing groups – New Zealand racing system, to give every horse a fair chance to race horses of the same ability...

Maiden: horse that has not won a race

Intermediate: horse that has won one race

Progressive: horse that has won at least two but no more than four races

Open: horse that has won five for more races

Ruminant – an animal where the stomach is divided into four compartments (Nicol, 2002)

Scoop – An implement that gathers and measures horse feed, otherwise known as a dipper

Slab– A portion of hay that comes off the hay bale freely as a clump

Spell– A term for a period of time a horse is away from training or racing

Stable – Or yard is a confined area no greater than 5 by 5 meters

Stereotypes – Abnormal horse behaviour that becomes repetitive and fixed in form and orientation (Frape, 1998).

Stud – The property where horses are breed

TAB– Totalisator agency board

TAB races– Races that provide prize money and on-course and off course betting

Turn out – A term for a horse being put out into a pastured paddock daily and brought back into stable at a later time.

VFA–Volatile fatty acids

Wastage – Poor performance or unsuccessful racing ability

Weaving – The horse sways its head laterally, sometimes involving the neck, forequarters and hindquarters. Weaving usually occurs while the horse is standing with the head over the stable door or a gate (1999).

Wind-sucking – Similar physical behaviour to crib-biting, the horse achieves the same posture and tension of the neck muscles but without grasping a fixed object (1995).

Yearling – A horse that is one year of age

References

- Adam's, O. R. (1974). *Adam's Lameness in Horses*. Philadelphia, USA: Williams & Wilkins.
- Aitchison, E. M., McDonald, D. L., Casson, P., & Rowe, J. B. (1987). Feed antibiotics and buffers to control rumen lactic acid. 2. Predosing. *Proceeding of the Nutrition Society*, 46(41), Abstract.
- Alexander, F., & Donald, D. E. (1949). Caecostomy in the horse. *Journal of Comparative Pathology*, 59, 127-132.
- Alexander, F., & Hickson, J. C. D. (1970). The salivary and pancreatic secretion of the horse. In A. T. Phillipson (Ed.), *In Physiology of Digestion and Metabolism in the Ruminant*. (pp. 357-389). Newcastle: Oriel Press.
- Argenzio, R. A. (1993). General functions of the gastrointestinal tract and their control and integration. In M. J. Swenson, and Reece, W.O. (Ed.), *Duke's Physiology of Domesticated Animals* (11th ed., pp. 325-335). Ithaca: Comstock Publishing.
- Argenzio, R. A., & Stevens, C. E. (1984). The large bowel- a supplementary rumen. *Proceeding of the Nutrition Society*, 43, 13-23.
- Avery, A. (1997). *Pasture for Horses; a winning resource*. Adelaide, South Australia: Gillingham Printers Ltd.
- Bailey, C. J. (1998, April 16th- 17th 1998). *Wastage in the racing industry- approached to study*. Paper presented at the Epidemiology Workshop for Equine Research Workers, University of Sydney-Veterinary Conference Centre.
- Bailey, C. J., Reid, S. W. J., Hodgson, D. R., & Rose, R. J. (1997). Wastage in the Australian Thoroughbred racing industry: a survey of Sydney trainers. *Australian Veterinary Journal*, 75(1), 64-66.
- Bailey, C. J., Reid, S. W. J., Hodgson, D. R., & Rose, R. J. (1999). Factors associated with time until first race and career duration for Thoroughbred racehorses. *American Journal of Veterinary Research*, 60, 1196-1200.
- Bailey, S. R., Baillon, M.L., Ryocroft, A.N., & Elliott, J. (2003). Identification of equine cecal bacteria producing amines in an in vitro model of carbohydrate overload. *Applied and Environmental Microbiology*, 69(4), 2087-2093.

- Bailey, S. R., Cunningham, F. M., & Elliot, J. (2000). Endotoxin and dietary amines may increase plasma 5-hydroxytryptamine in the horse. *Equine Veterinary Journal*, 32(6), 497-504.
- Baxter, G. M. (1996). Diagnosing and treating acute laminitis: Symposium on Acute Laminitis. *Veterinary Medicine*, 91, 940-952.
- Bergtore, E. N. (1990). Energy contribution of volatile fatty acids from the gastrointestinal tract in various species. *Physiology Review*, 70, 567-590.
- Boisen, S., Hvelplund, T., & Weisbjerg, M. R. (2000). Ideal amino acid profiles as basis for feed protein evaluation. *Livestock Production Science*, 64(2-3), 239-351.
- Bourke, J. M. (1968). Feeding of Thoroughbred horses. *Australian Veterinary Journal*, 44, 241-245.
- Bourke, J. M. (1995). *Wastage in Thoroughbreds*. Paper presented at the Proceedings of the Annual Seminar of the Equine Branch of the New Zealand Veterinary Association.
- Bryden, W. L. (1995, 14-16th February 1995). *Nutrition of performance horses*. Paper presented at the Proceedings of Equine Nutrition and Pastures for Horses Workshop, Richmond NSW, Australia.
- Budiansky, S. (1997). *The Nature of Horses- Their Evolution, Intelligence and Behaviour*. London: Phoenix Illustrated Orion Publishing Group.
- Clarke, L. L., Roberts, M. C., & Argenzio, R. A. (1990). Feeding and digestive problems in the horses- physiologic responses to a concentrated meal. *Veterinary Clinics of North America: Equine Practice*, 6(2), 433-450.
- Cohen, N. D., Gibbs, P. G., & Woods, A. M. (1999). Dietary and other management factors associated with colic in horses. *Journal of the American Veterinary Medical Association*, 215(1), 53-60.
- Cuddeford, D. (2001). Starch digestion in the horse. In J. D. Pagan, & Geor, R.J. (Ed.), *Advances in Equine Nutrition II* (pp. 95-104). Versailles, Kentucky, USA: Kentucky Equine Research Inc.
- Davie, A., Steward, J., & Skinne, R. (2000). *Faecal pH measurements in thoroughbred horses*. Sydney, Australia.

- de Fombelle, A., Julliand, V., Drogoul, C., & Jacotot, E. (2001). Feeding and microbial disorders in horses: I-Effects of an abrupt incorporation of two levels of barle in a hay diet on microbial profile and activities. *Journal of Equine Veterinary Science*, 21(9), 439-445.
- de Fombelle, A., Varloud, M., Goachet, A. G., Jacotot, E., Philippeau, C., Drogoul, C., et al. (2003). Characterisation of the microbial and biochemical profile of the different segments of the digestive tract of horses fed two distinctive diets. *Animal Science*, 77(2), 293-304.
- Divers, T. J., & Dreyfuss, D. (1990). Evaluating the horse with poor racing performance. *Veterinary Medicine*, 522-529.
- Dossenbach, M., & Dossenbach, H. D. (1988). *The Nobel Horse*. Glenfeld, Auckland, New Zealand: D. Bateman Ltd in association with Hallwag, Switzerland.
- Duncan, P., Foose, T. J., Gordon, I. J., Gakahu, C. G., & Lloyd, M. (1990). Comparative nutrient extraction from forages by grazing bovids and equids a test of the nutritional model of equid- bovid competition and co-existence. *Oecologia*, 84(3), 411-418.
- Eastwood, B. R. (2002). *The Effects of Grain Supplementation on the Faecal pH of Horses Maintained on Pasture*. Massey University, Palmerston North, New Zealand.
- Eaton, M. D. (1994). Energetics and performance. In U. W. B. S. C. David R. Hodgson and Reuben J. Rose Principles and Practice of Equine Sports Medicine: The Athletic Horse Philadelphia (Ed.), *Principles and Practice of Equine Sports Medicine: The Athletic Horse* (First ed., pp. 49-61). Philadelphia, USA: W.B. Saunders Company.
- Evans, D. L. (1994). Training thoroughbred racehorses. In D. R. H. a. R. J. Rose (Ed.), *Principles and Practice of Equine Sports Medicine: The Athletic Horse* (First ed., pp. 393-398). Philadelphia, USA: W.B. Saunders Company.
- Finn, K., & Rogers, C. W. (2001). *Faecal pH trial -unpublished report*. Palmerston North, New Zealand: Massey University.
- Foreman, J. H., Bayley, W. M., Grant, B. D., & Gollnick, P. D. (1990). Standardized exercise test and daily heart rate responses of Thoroughbreds undergoing conventional race training and detraining. *American Journal of Veterinary Research*, 51, 914-920.

- Frape, D. L. (1988). Dietary requirements and athletic performance of horses. *Equine Veterinary Journal*, 20, 163-.
- Frape, D. L. (1998). *Equine Nutrition and Feeding* (2nd Edition ed.). Cornwall, Great Britain: Blackwell Science Ltd, MPG Books Ltd Bodium.
- Frape, D. L. (2004). *Equine Nutrition and Feeding* (Third ed.). Oxford, UK: Blackwell Publishing Ltd.
- Frape, D. L., & Boxall, R. C. (1974). Some nutritional problems of the horse and their possible relationship to those of other herbivores. *Equine Veterinary Journal*, 6(2), 59-68.
- Gallagher, K., Leech, J., & Stowe, H. (1992). Protein, energy and dry matter consumption by racing thoroughbreds: a field study. *Equine Veterinary Science*, 12(1), 43-48.
- Garner, H. E., Moore, J. N., Johnson, J. H., Clark, L., Amend, J. F., Tritschler, L. G., et al. (1978). Changes in the caecal flora associated with the onset of laminitis. *Equine Veterinary Journal*, 10(4), 249-252.
- Gill, A. M., & Lawrence, L. M. (2003). *Effect of feeding frequency on digestion in ponies*. Versailles, Kentucky, USA: Kentucky Equine Research Inc.
- Glade, M. J. (1983). A comparative study of interval and conventional training on Thoroughbred racehorses. *Equine Veterinary Journal*, 15, 31-36.
- Goodson, J., Tyznik, W. J., & Dehority, B. A. (1985). *Effects of an abrupt change from all hay to all concentrate on anaerobic bacteria numbers (grown on selected media), protozoa numbers and pH of the caecum*. Paper presented at the Proceedings of the 9th Equine Nutrition and Physiology Symposium, East Lansing, Michigan.
- Goodson, J., Tyznik, W.J., Cline, J.H., & Dehority, B.A. (1988). Effects of an abrupt diet change from hay to concentrate on microbial numbers and physical environment in the caecum of the pony. *Applied Environmental Microbiology*, 54, 1946- 1950.
- Goold, G. J., J.A. B., & Rollo, M. D. (1988). Management of thoroughbred studs in the Waikato. *Proceedings of the New Zealand Grasslands Association*, 49, 33-36.
- Grace, N. D. (1997). *Energy metabolism of horses*. Paper presented at the Proceedings of the Annual Seminar of the Equine Branch of the New Zealand Veterinary Association, Hamilton, May.

- Grace, N. D., Rogers, C. W., Firth, E. C., Faram, T. L., & Shaw, H. L. (2003). Digestible energy intake, dry matter digestibility and effect of increased calcium intake on bone parameters of grazing Thoroughbred weanlings in New Zealand. *New Zealand Veterinary Journal*, *51*(4), 165-173.
- Grace, N. D., Shaw, H. L., Gee, E. K., & Firth, E. C. (2002). Determination of digestible energy intake and apparent absorption of macroelements of grazing lactating Thoroughbred mares. *New Zealand Veterinary Journal*, *50*, 182-185.
- Graham-Thiers, P. M., Kronfeld, D. S., Kline, K. A., & Sklan, D. J. (2001). Dietary protein restriction and fat supplementation diminish the acidogenic effect of exercise during repeated sprints in horses. *Journal of Nutrition*, *131*, 1959-1964.
- Greet, T. R. C. (2000). *Functional Anatomy and Physiology of the Gastrointestinal Tract*. Paper presented at the Proceedings of the Annual Seminar of the Equine Branch of the New Zealand Veterinary Association.
- Gustaffson, A. (2004). *Doctoral Dissertation in Antibiotic Associated Diarrhea in Horses -with special reference to Clostridium difficile*. Swedish University of Agriculture Science, Uppsala, Sweden.
- Hammond, C. J., Mason, D. K., & Watkins, K. L. (1986). Gastric ulceration in mature Thoroughbred horses. *Equine Veterinary Journal*, *18*, 284-287.
- Harkins, J. D., Kammerling, S. G., Bagwell, C. A., & Karns, P. A. (1990). A comparative study of interval and conventional training in thoroughbred racehorses. *Equine Veterinary Journal*, *9*, 14- 19(S).
- Harris, P. A. (1997). Energy sources and requirements of the exercising horse. *Annual Review of Nutrition*, *17*, 185-210.
- Hayes, M. H. (1987). The Nutrition and Feeding of Horses. In R. D. Rossdale (Ed.), *Veterinary Notes for Horse Owners* (pp. 582-601). London: Stanley Paul.
- Hintz, H. F. (1994). Nutrition and equine performance. *The Journal of Nutrition*, *12* (Supplement), 2723-2729.
- Hintz, H. F., Arenzio, R. A., & Schryver, H. F. (1971). Digestion coefficients, blood glucose levels and molar percentage of volatile acids in intestinal fluid of ponies fed varying forage-grain ratios. *Journal of Animal Science*, *33*, 992-995.
- Hintz, H. F., & Cymbaluk, N. F. (1994). Nutrition of the horse. *Annual Review of Nutrition*, *14*, 243-267.

- Hoffman, R. M. (2003). *Carbohydrate metabolism in horses*. Ithaca, New York, USA.: Department of Animal & Poultry Sciences.
- Hoffman, R. M., Wilson, J. A., Kronfeld, D. S., Cooper, W. L., Lawrance, L. M., Sklan, D. J., et al. (2001). Hydrolyzable carbohydrates in pasture, hay, and horse feeds: Direct assay and season variation. *Journal of Animal Science*, 79, 500-506.
- Hood, D. M. (1999). Laminitis in the horse. *Veterinary Clinics of North America: Equine Practice*, 15(2), 287-295.
- Hood, D. M., Grosenbaugh, D. A., Mostafa, M. B., Morgan, S. J., & Thomas, B. C. (1993). The role of vascular mechanisms in the development of acute laminitis. *Journal of Veterinary Internal Medicine*, 7, 228-234.
- Hooper, L. V., Midtvedt, T., & Gordon, J. I. (2002). How host microbial interactions shape the nutrient environment of the mammalian intestine. *Annual Review of Nutrition*, 22, 283-307.
- Hoskins, S. O., & Gee, E. K. (2004). Feeding value of pastures for horses. *New Zealand Veterinary Journal*, 56(6), 332-341.
- Houpt, K. A. (1987). Abnormal behaviour. *Veterinary Clinics of North America: Food Animal Practice*, 3, 357-367.
- Houpt, K. A. (1995). New perspective on equine stereotypic behaviour. *Equine Veterinary Journal*, 27(2), 82-83.
- Hudson, J. M., Cohen, N. D., Gibbs, P. G., & Thompson, J. A. (2001). Feeding practices associated with colic in horses. *Journal of the American Veterinary Medical Association*, 219(10), 1419-1425.
- Hunt, W. D. (1994). *Pasture for Horses, Commissioned by the New Zealand Equine Research Foundation*. Palmerston North: AgResearch.
- Huntington, P. J., & Jenkinson, G. J. (2003). *Feeding practices in Australia and New Zealand*. Versailles, Kentucky, USA: Kentucky Equine Research Inc.
- Hussein, H. S., Vogedes, L. A., Fernandez, G. C. J., & Frankeny, R. L. (2004). Effects of cereal grain supplementation on apparent digestibility of nutrients and concentrations of fermentation end-products in the feces and serum of horses consuming alfalfa cubes. *Journal of Animal Science*, 82, 1986-1996.

- Illius, A. W., & Gordon, I. J. (1992). Modelling the nutritional ecology of ungulate herbivores; evolution of body size and competitive interactions. *Oecologia*, *89*, 428-434.
- Jackson, S. G. (2003). *The digestive tract of the horse- practical consideration*. Versailles Kentucky: Kentucky Equine Research Inc.
- Jacobs, B. F., Kingston, J. D., & L.L., J. (1999). The origin of grass-dominated ecosystems. *Annals of the Missouri Botanical Gardens*, *86*(2), 590-643.
- Janis, C. (1976). The evolutionary strategy of Equidae and the origin of rumen and caecal digestion. *Evolution*, *30*, 757-774.
- Jeffcott, L. B., Rosedale, P. D., Freestone, J., Frank, C. J., & Tower-Clark, P. F. (1982). An assessment of wastage in Thoroughbred racing from conception to 4 years of age. *Equine Veterinary Journal*, *14*(3), 185-198.
- Johnston, K. G., Tyrrell, J., Rowe, J. B., & Pethick, D. W. (1998). Behavioural changes in stabled horses given nontherapeutic levels of viriniamycin. *Equine Veterinary Journal*, *30*(2), 139-149.
- Jose-Cunilleras, E., & Hinchcliff, K. W. (2003). Carbohydrate metabolism in exercising horses. *Equine and Comparative Exercise Physiology*, *1*(1), 23-32.
- Julliand, V., de Fombelle, A., Drogoul, C., & Jacotot, E. (2001). Feeding and microbial disorders in horses: 3-Effects of three hay: grain ratios on microbial profile and activities. *Journal of Equine Veterinary Science*, *21*, 543-546.
- Kern, D. L., Slyter, L. L., Leffel, E. C., Weaver, J. M., & Oltjen, R. R. (1974). Ponies vs. Steers: Microbial and chemical characteristics of intestinal microflora. *Journal of Animal Science*, *38*, 559-564.
- Kidd, L. (2002). *Surgical Colic - a New Zealand Perspective*. Paper presented at the Proceedings of the Annual Seminar of the Equine Branch of the New Zealand Veterinary Association.
- Kohnke, J. R., Kelleher, F., & Trevor-Jones, P. (1999). *Feeding Horses in Australia -A Guide for Horse Owners and Managers* (No. RIRDC Publication No. 99/49). Canberra, New South Wales, Australia: Rural Industries Research and Development Corporation.
- Langer, P. (1988). *The Mammalian Herbivore Stomach*. New York: Gustav Fischer Stuttgart.

- Lawrence, L. M. (1990). Nutrition and fuel utilization in the athletic horse. *Veterinary Clinics of North America Equine Practice*, 6(2), 393-418.
- Lawrence, L. M. (1994). Nutrition and the athletic horse. In D. R. H. a. R. J. Rose (Ed.), *Principles and Practice of Equine Sports Medicine: The Athletic Horse*. Philadelphia, USA: W.B. Saunders Company.
- Lewis, L. D. (1995). *Equine Clinical Nutrition: Feeding and Care*. USA: Williams & Wilkins.
- Lowe, J. E., Hintz, H. F., & Schryver, D. S. (1970). A new technique for the long term caecal fistulation in ponies. *American Journal of Veterinary Research*, 31, 1109-1111.
- Mackie, R. I., & Wilkins, C. A. (1988). Enumeration of anerobic microflora of the equine gastrointestinal tract. *Enviromental Microbiology*, 54, 2155-2160.
- Martin-Rosset, W., & Vermorel, M. (1991). Maintenance energy requirements variations determined by indirect calorimetry and feeding trials in high horses. *Journal of Equine Veterinary Science*, 11, 42-45.
- Martin-Rosset, W., Vermorel, M., Doreau, M., Tisserand, J. L., & Andrieu, J. L. (1994). The French horse feed evaluation systems and recommended allowances for energy and protein. *Livestock Production Science*, 40, 37-56.
- Maskell, I. E., & Johnson, J. V. (1993). Digestion and absorption. In I. Burger (Ed.), *The Waltman Book of Companion Animal Nutrition* (pp. 25-44). Oxford: Pergammon Press.
- Mason, G. J. (1991). Stereotypies: a critical review. *Animal Behaviour*, 41, 1015-1037.
- Mayes, E., & Duncan, P. (1986). Temporal patterns of feeding in free ranging horses. *Behaviour Proceedings*, 96, 105-129.
- McCutcheon, L. J., Kelso, T. B., Bertocci, L. A., Hodgson, D. R. J., & Bayly, W. M. (1987). Buffering and aerobic capacity in equine muscle and effect of training. In J. R. G. a. N. E. Robison (Ed.), *Equine Exercise Physiology 2* (pp. 348). California: ICEEP Publications.
- McDonald, P., Edwards, R. A., Greenhalgh, J. F. D., & Morgan, C. A. (2002). *Animal Nutrition* (Six ed.). Essex, UK: Pearson Education Limited.

- McGreevy, P. D., & Nicol, C.J. (1998). The effect of short term prevention on the subsequent rate of crib biting in Thoroughbred horses. *Equine Veterinary Journal Supplement*, 27, 30-34.
- McGreevy, P. D., Crippa, P. J., French, N. P., Green, L. E., & Nicol, C. J. (1995). Management factors associated with stereotypic and redirected behaviour in the Thoroughbred horse. *Equine Veterinary Journal*(21), 86-91.
- McGreevy, P. D., Richardson, J.D., Nicol, C.J., & Lane, J.G. (1995). Radiographic and endoscopic study of horses performing an oral based stereotypy. *Equine Veterinary Journal*, 27(2), 92-95.
- McLean, B. M. L., Hyslop, J. J., Longland, A. C., Cuddeford, D., & Hollands, T. (2000). Physical processing of barley and its effects on intra-caecal fermentation parameters in ponies. *Animal Feed Science and Technology*, 85, 79-87.
- McMeniman, N. P., Elliot, R., Groenendyk, S., & Dowsett, K. F. (1987). Synthesis and absorption of cystine from the hindgut of the horse. *Equine Veterinary Journal*, 19, 192-194.
- Medina, B., Girard, I. D., Jacotot, E., & Julliard, V. (2002). Effect of a Preparation of *Saccharomyces Cerevisiae* on Microbial Profiles and Fermentation Patterns in the Large Intestine of Horses Fed a High Fibre or a High Starch Diet. *Journal of Animal Science*, 80(10), 2600-2609.
- Menard, C., Duncan, P., Fleurance, G., George, J. Y., & Lila, M. (2002). Comparative foraging and nutrition of horses and cattle in European wetlands. *Journal of Applied Ecology*, 39, 120-133.
- Merrit, A. M. (1999). Normal equine gastroduodenal secretions and motility. *Equine Veterinary Journal Supplement*, 29, 7-13 (S).
- Merrit, A. M. (2003a). *The Equine Stomach: A Personal Perspective (1963-2003)*. Paper presented at the 49th Annual Convention of the American Association of Equine Practitioners, New Orleans, Louisiana, USA.
- Meyer, H., Radicke, S., Kiengle, E., Wilke, S., & Kleffken, D. (1993). *Investigations of preileal digestion of oats, corn, and barley starch in relation to grain processing*. Paper presented at the Proceeding of the 13th Equine Nutrition and Physiological Symposium, Florida, USA.

- Meyer, H., Radicke, S., Kienzle, E., Wilke, S., Kleffken, D., & Illenseer, M. (1995). Investigations on preileal digestion of starch from grain, potato and manioc in horses. *Journal of American Veterinary Medical Association*, 42, 371-2381.
- Milton McClure, J. (1990). General health care and miscellaneous conditions of the racehorse. *Veterinary Clinics of North America Equine Practice*, 6(1), 223-237.
- Mohammed, H. O., Hill, T., & Lowe, J. (1991). Risk factors associated with injuries in Thoroughbred horses. *Equine Veterinary Journal*, 23(6), 445-448.
- Moore, B. E., & Dehority, B. A. (1992). Effects of diet and protozoa on total and cellulotic bacteria and fungal concentrations in the caecum and colon. *Journal of Animal Science*, 70(1), 240-251.
- Moore, B. E., & Dehority, B. A. (1993). Effects of diet and hindgut defaunation on diet digestibility and microbial concentrations in the caecum and colon of the horse. *Journal of Animal Science*, 71(12), 3350-3358.
- Moore, J. N., & Allen, J. D. (1996). The pathophysiology of acute lameness. *Veterinary Medicine*, 91, 936- 939.
- More, S. J. (1999). A longitudinal study of racing thoroughbreds: performance during the first years of racing. *Australian Veterinary Journal*, 77(2), 105-112.
- Morton, J. D., Grace, N. D., & O'Connor, M. B. (Eds.). (1999). *Use of Trace Elements in New Zealand Pastoral Farming* (Vol. 1). Auckland: New Zealand Fertiliser Manufacturers' Research Association.
- Mueller, P. J., Protos, P., Houpt, K. A., & Van Soest, P. J. (1998). Chewing behaviour in the domestic donkey (*Equus asinus*) fed fibrous forage. *Applied Animal Behaviour Science*, 60, 241-251.
- Murray, M. J. (1999). Pathphysiology of peptic disorders in foals and horses: a review. *Equine Veterinary Journal*, 29, 14-18.
- Murray, M. J., & Eichorn, E. S. (1996). Effects of intermittent feed deprivation, intermittent feed deprivation with ranitidine administration, and stall confinement with ad libitum access to hay on gastric ulceration in horses. *American Journal of Veterinary Research*, 11, 1599-1603.
- Nadeau, J. A., Andrews, F. M., Mathew, A. G., Argenzio, R. A., Blackford, J. T., Sohtell, M., et al. (2000). Evaluation of diet as a cause of gastric ulcers in horses. *American Journal of Veterinary Research*, 61(7), 784-970.

- Nicol, C. J. (1999). Understanding equine stereotypes. In *The Role of the Horse in Europe- Equine Veterinary Journal Supplement* (Vol. 28, pp. 20-25).
- Nicol, C. J. (2002). Equine Stereotypes. *Recent Advances in Companion Animal Behaviour Problems*, 1-5.
- Nicol, C. J., Davidson, H. P. D., Harris, P. A., Waters, A. J., & Wilson, A. D. (2002). Study of crib-biting and gastric inflammation and ulceration in young horses. *The Veterinary Record*, 151, 658-662.
- NRC. (1989). *Nutrient Requirements of Horses* (5th Ed ed.). Washington DC: National Academy Press.
- NRC. (2001). *Nutrient Requirements of Dairy Cattle* (7th ed.). Washington DC: National Academy Press.
- NZRB. (2004). *Size and Scope of the New Zealand Racing Board*. Wellington.
- Olsman, A. F. S., Huurdeman, C. M., Jansen, W. L., Haaksma, J., Sloet van Oldruitenborgh-Oosterbaan, M. M., & Beynen, A. C. (2004). Macronutrient digestibility, nitrogen balance, plasma indicators of protein metabolism and mineral absorption in horses fed a ration rich in sugar beet pulp. *Journal of Animal Physiology and Animal Nutrition*, 88, 321-331.
- Olsman, A. F. S., Jansen, W. L., Sloet van Oldruitenborgh-Oosterbaan, M. M., & Beynen, A. C. (2003). Assessment of the minimum protein requirement of adult ponies. *Journal of Animal Physiology and Animal Nutrition*, 87, 205-212.
- Pagan, J. D. (1997). *Energy and the performance horse*. Paper presented at the Proceedings of the Annual Seminar of the Equine Branch of the NZVA, Hamilton, May.
- Pagan, J. D. (1997a). *Recent advances in equine nutrition research*. Paper presented at the Proceedings of the Annual Seminar of the Equine Branch of the New Zealand Veterinary Association.
- Pagan, J. D. (1997b). *Forages for horses: more than just filler*. Paper presented at the Proceedings of the Annual Seminar of the Equine Branch of the New Zealand Veterinary Association.
- Pagan, J. D. (2003). *Energy and the performance horse*. Versailles, Kentucky, USA: Kentucky Equine Research Inc.

- Pagan, J. D., Harris, P., Brewster-Barnes, T., Duren, S. E., & Jackson, S. G. (1998). Exercise affects digestibility and rate of passage of all-forage and mixed diets in Thoroughbred horses. *Journal of Nutrition*, 128 (S), 2704-2707S.
- Pagan, J. D., & Harris, P. A. (1999). The effects of timing and amount of forage and grain on exercise response in thoroughbred horses. *Equine Veterinary Journal*, 30, 451-457 (S).
- Pagan, J. D., & Hintz, H. F. (1986). Equine energetics: II. Energy expenditure in horses during submaximal exercise. *Journal of Animal Science*, 63, 822-830.
- Pearce, G. R. (1975). The nutrition of racehorses: a review. *Australian Veterinary Journal*, 51, 14-21.
- Pearson, R. A., Archibald, R. F., & Muirhead, R. H. (2001). The effect of forage quality and level of feeding on digestibility and gastrointestinal transit time of oat straw and alfalfa given to ponies and donkeys. *British Journal of Nutrition*, 85, 599-606.
- Pell, S. M., & McGreevy, P. D. (1999). Prevalence of stereotypic and other problem behaviours in Thoroughbred horses. *Australian Veterinary Journal*, 77, 678-679.
- Perkins, N. (1999). *Wastage in the NZ thoroughbred racing industry: An epidemiological investigation*. Paper presented at the Proceedings of the Annual Seminar of the Equine Branch of the New Zealand Veterinary Association.
- Perkins, N. R. (2001). *Findings from a study of the causes of wastage in New Zealand Thoroughbreds*. Paper presented at the Bomac Lecture Series.
- Perkins, N. R., Reid, S. W. J., & Morris, R. S. (2004b). Effect of training location and time period on racehorse performance in New Zealand. 2. Multivariable analysis. *New Zealand Veterinary Journal*, 52(5), 243-249.
- Perkins, N. R., Reid, S. W. J., & Morris, R. S. (2005a). Profiling the New Zealand Thoroughbred racing industry. 1. Training, racing and general health patterns. *New Zealand Veterinary Journal*, 53(1), 59-68.
- Perkins, N. R., Reid, S. W. J., & Morris, R. S. (2005b). Profiling the New Zealand Thoroughbred racing industry. 2. Conditions interfering with training and racing. *New Zealand Veterinary Journal*, 53(1), 69-76.
- Radicke, S., Kienzle, E., & Meyer, H. (1991). *Preileal apparent digestibility of oats and corn starch and consequences for cecal metabolism*. Paper presented at the

- Proceedings of the 12th Equine Nutrition and Physiology Symposium, Calgary, Canada.
- Ralston, S. L. (1986). Feeding behaviour. *Veterinary Clinics of North America: Equine Practice*, 2(3), 609-621.
- Ralston, S. L., & Baile, C. A. (1983). Factors in the control of feed intake of horses and ponies. *Neuroscience and Biobehavioural Review*, 7 (Abstract)(4), 465-470.
- Redbo, I., Redbo-Torstensson, P., Odberg, F. O., Hedeahl, A., & Holm, J. (1998). Factors affecting behavioural disturbances in race-horses. *Animal Science*, 66, 475-481.
- Richards, N., Choct, M., Hinch, G. N., & Rowe, J. B. (2004). Examination of the use of exogenous α -amylase and amyloglucosidase to enhance starch digestion in the small intestine of the horse. *Animal Feed Science and Technology*, 114, 295-305.
- Rogers, C. W., & Firth, E. C. (2005). Musculoskeletal responses of 2-year-old Thoroughbred horses to early training. 2. Measurement error and effect of training stage on the relationship between objective and subjective criteria of training workload. *New Zealand Veterinary Journal*, 52(5), 275-279.
- Rose, R. J. (1993). *Training programmes and the assessment of fitness in the racehorse*. Paper presented at the Proceedings of the Annual Seminar of the Equine Branch of the New Zealand Veterinary Association.
- Rossdale, P. D., Hopes, R., Wingfield Digby, N. J., & Offord, K. (1985). Epidemiological study of wastage among racehorses 1982 and 1983. *Veterinary Record*, 116, 66-69.
- Rowe, J., Brown, W., & Bird, S. (2001). *Safe and Effective Grain Feeding for Horses* (No. RIRDC Publication No. 01/148). Barton, Canberra, New South Wales, Australia: Rural Industries Research and Development Corporation.
- Rowe, J., Pethick, D. W., & Johnston, K. G. (1995). *Controlling acidosis in the equine hindgut*. Paper presented at the Recent Advances in Animal Nutrition in Australia, University of New England, Armidale, NSW, Australia.
- Rowe, J. B., Lees, M. J., & Pethick, D. W. (1994). Prevention of acidosis and laminitis associated with grain feeding in horses. *Journal of Nutrition Supplement*, 124, 2742-2744.

- Rushen, J., Lawrence, A. B., & Terlouw, E. M. C. (1993). *The motivational basis of stereotypes*. Wallingford, Oxford: CAB International.
- Sloet va Oldenbourgh-Oosterbaan, M. M. (1999). Laminitis in the horse: a review. *Veterinarian Quarterly*, *21*, 121-127.
- Smyth, G. B., Young, D. W., & Hammond, L. S. (1988). Effects of diet and feeding on postprandial serum gastrin and insulin concentrations in adult horses. *Equine Veterinary Journal Supplement*, *7 (S)*, 56-59.
- Southwood, L. L., Evans, D. L., Bryden, W. L., & Rose, R. J. (1993a). Nutrient intake of horses in Thoroughbred and Standardbred stables. *Australian Veterinary Journal*, *70(5)*, 164-168.
- Southwood, L. L., Evans, D. L., Bryden, W. L., & Rose, R. J. (1993b). Feeding practices in Thoroughbred and Standardbred stables. *Australian Veterinary Journal*, *70*, 184-185.
- Sprouse, R. F., Garner, H. E., & Green, E. M. (1987). Plasma endotoxin levels in horses subjected to carbohydrate induced laminitis. *Equine Veterinary Journal*, *19*, 25-28.
- Staniar, W. B., Kronfeld, D. S., Wilson, J. A., Lawrence, L. A., Cooper, W. L., & Harris, P. A. (2001). Growth of Thoroughbreds fed a low-protein supplement fortified with lysine and threonine. *Journal of Animal Science*, *79*, 2143-2151.
- Stashak, T. S. (2002). Chapter 8: Lameness. In *Adam's Lameness in Horses* (pp. 645-663). Philadelphia, USA: Williams & Wilkins.
- Totora, G. J., & Grabowski, S. R. (2000). *Principles of Anatomy and Physiology*. New York, USA: John Wiley and Sons Inc.
- Tyler-McGowan, C. M., Golland, L. C., Evans, D. L., Hodgson, D. R., & Rose, R. J. (1999). Haematological and biochemical responses to training and overtraining. *Equine Veterinary Journal Supplement*, *30*, 621-625.
- Waghorn, G. C., & Barry, T. N. (1987). Pasture as a nutrient source. In A. M. Nicol (Ed.), *Feeding Livestock on Pasture* (pp. 21-38). Hamilton, New Zealand: New Zealand Society of Animal Production.
- Wallace, T. (1977). *Pasture management on Waikato equine studs*. Paper presented at the Proceedings of Equine Sessions of the Annual Conference of the New Zealand Veterinary Association.

- Waller, A., Armstrong, S., Smithhurst, K. J., & Lindinger, M. I. (2003). Effects of diet, feeding and daily variation on acid-base balance in horses. *Equine and Comparative Exercise Physiology*, 1(3), 153-165.
- Waters, A. J., Nicol, C. J., & French, N. P. (2002). Factors influencing the development of stereotypic and redirected behaviours in young horses: findings of a four year prospective epidemiological study. *Equine Veterinary Journal*, 34(6), 572-579.
- White, N. (1986). What's next in equine colic research. *Equine Veterinary Journal*, 18, 429-431.
- Willard, J. G., Willard, J. C., Wolfram, S. A., & Barker, J. P. (1977). Effect on diet on caecal pH and feeding behaviour of horses. *Journal of Animal Science*, 45, 87-93.
- Wurbel, H., Freire, R., & Nicol, C. J. (1998). Prevention of stereotypic wire gnawing in laboratory mice, effects on behaviour and implication for sterotypy as a coping response. *Behaviour Proceedings*, 42, 61-72.
- Zeyner, A., Geibler, C., & Dittrich, A. (2004). Effects of hay intake and feeding sequence on variables in faeces and faecal water (dry matter, pH value, organic acids ammonia, buffering capacity) of horses. *Journal of Animal Physiology and Animal Nutrition*, 88, 7-19.
- Zeyner, A., Geisler, C., Kaske, H., & Fuchs, R. (1992). Evaluation of horse feeds via faecal water analysis (water, pH, organic acids). *Pferdeheilkunde Sonderausgabe*, 88-91.
- Zeyner, A., Grass, S., Geissler, C., Kaske, H., & Dittrich, A. (1993). *Effect of diet composition and feeding time on pH value, buffering capacity, content of organic acids and ammonia in equine faeces and studies on establishing reference values for practical application*. VDLUFA- verlag, Darmstadt, Germany.

Appendix

Appendix A: Experiment One's feed programme- (all feed units are as per scoop basis)

SF= NRM sweetfeed™

RHC=Red Clover hay

Day	Week	Date	AM			PM			Bran	RCH (slab)
			oats	SF	chaff	oats	SF	chaff		
1	1	26/07/2003	0.5	0.5	0.5	1.25	1.25	1.25	0	1
2	1	27/07/2003	0.5	0.5	0.5	1.25	1.25	1.25	0	1
3	1	28/07/2003	0.5	0.5	0.5	1.25	1.25	1.25	0	1
4	1	29/07/2003	0.5	0.5	0.5	1.25	1.25	1.25	0	1
5	1	30/07/2003	0.5	0.5	0.5	1.25	1.25	1.25	0	1
6	1	31/07/2003	0.5	0.5	0.5	1.25	1.25	1.25	0	1
7	1	1/08/2003	0.5	0.5	0.5	1.25	1.25	1.25	0	1
8	2	2/08/2003	0.5	0.5	0.5	1.25	1.25	1.25	0	1
9	2	3/08/2003	0.5	0.5	0.5	1.25	1.25	1.25	0	1
10	2	4/08/2003	0.5	0.5	0.5	1.25	1.25	1.25	0	1
11	2	5/08/2003	0.5	0.5	0.5	1.25	1.25	1.25	0	1
12	2	6/08/2003	0.5	0.5	0.5	1.25	1.25	1.25	0	1
13	2	7/08/2003	0.5	0.5	0.5	1.25	1.25	1.25	0	1
14	2	8/08/2003	0.5	0.5	0.5	1.25	1.25	1.25	0	1
15	3	9/08/2003	0.5	0.5	0.5	1.25	1.25	1.25	0	1
16	3	10/08/2003	0.5	0.5	0.5	1.25	1.25	1.25	0	1
17	3	11/08/2003	0.5	0.5	0.5	1.25	1.25	1.25	0	1
18	3	12/08/2003	0.5	0.5	0.5	1.25	1.25	1.25	0	1
19	3	13/08/2003	0.5	0.5	0.5	1.75	0.5	1.25	0	1
20	3	14/08/2003	0.5	0.5	0.5	1.5	1	1.25	0	1
21	3	15/08/2003	0.5	0.5	0.5	1.5	1	1.25	0	1
22	4	16/08/2003	0.5	0.5	0.5	1.5	1	1.25	0	1
23	4	17/08/2003	0.5	0.5	0.5	1.5	1	1.25	0	1
24	4	18/08/2003	0.5	0.5	0.5	1.5	1	1.25	0	1
25	4	19/08/2003	0.5	0.5	0.5	1.5	1	1.25	0	1
26	4	20/08/2003	0.5	0.5	0.5	1.5	1	1.25	1	1
27	4	21/08/2003	0.5	0.5	0.5	1.5	1	1.25	1	1
28	4	22/08/2003	0.5	0.5	0.5	1.5	1	1.25	1	1
29	5	23/08/2003	0.5	0.5	0.5	1.5	1	1.25	1	1
30	5	24/08/2003	0.5	0.5	0.5	1.5	1	1.25	1	1
31	5	25/08/2003	0.5	0.5	0.5	1.5	1	1.25	1	1
32	5	26/08/2003	0.5	0.5	0.5	1.5	1	1.25	1	1
33	5	27/08/2003	0.5	0.5	0.5	1.5	1	1.25	1	1
34	5	28/08/2003	0.5	0.5	0.5	1.5	1	1.25	1	1
35	5	29/08/2003	0.5	0.5	0.5	1.5	1	1.25	1	1
36	6	30/08/2003	0.5	0.5	0.5	1.5	1	1.25	1	1
37	6	31/08/2003	0.5	0.5	0.5	1.5	1	1.25	1	1
38	6	1/09/2003	0.5	0.5	0.5	1.5	1	1.25	1	1
39	6	2/09/2003	0.5	0.5	0.5	1.5	1	1.25	1	1
40	6	3/09/2003	0.5	0.5	0.5	1.5	1	1.25	1	1
41	6	4/09/2003	0.5	0.5	0.5	1.5	1	1.25	1	1
42	6	5/09/2003	0.5	0.5	0.5	1.5	1	1.25	1	1

43	7	6/09/2003	0.5	0.5	0.5	1.5	1	1.25	1	1
44	7	7/09/2003	0.5	0.5	0.5	1.5	1	1.25	1	1
45	7	8/09/2003	0.5	0.5	0.5	1.5	1	1.25	1	1
46	7	9/09/2003	0.5	0.5	0.5	1.5	1	1.25	1	1
47	7	10/09/2003	0.5	0.5	0.5	1.5	1	1.25	1	1
48	7	11/09/2003	0.5	0.5	0.5	1.5	1	1.25	1	1
49	7	12/09/2003	0.5	0.5	0.5	1.5	1	1.25	1	1
50	8	13/09/2003	0.5	0.5	0.5	1.5	1	1.25	1	1
51	8	14/09/2003	0.5	0.5	0.5	1.5	1	1.25	1	1
52	8	15/09/2003	0.5	0.5	0.5	1.5	1	1.25	1	1
53	8	16/09/2003	0.5	0.5	0.5	1.5	1	1.25	1	1
54	8	17/09/2003	0.5	0.5	0.5	1.5	1	1.25	1	1
55	8	18/09/2003	0.5	0.5	0.5	1.5	1	1.25	1	1
56	8	19/09/2003	0.5	0.5	0.5	1.5	1	1.25	1	1
57	9	20/09/2003	0.5	0.5	0.5	1.5	1	1.25	1	1
58	9	21/09/2003	0.5	0.5	0.5	1.5	1	1.25	1	1
59	9	22/09/2003	0.5	0.5	0.5	1.5	1	1.25	1	1
60	9	23/09/2003	0.5	0.5	0.5	1.5	1	1.25	1	1
61	9	24/09/2003	0.5	0.5	0.5	1.5	1	1.25	1	1
62	9	25/09/2003	0.5	0.5	0.5	1.5	1	1.25	1	1
63	9	26/09/2003	0.5	0.5	0.5	1.5	1	1.25	1	1
64	10	27/09/2003	0.5	0.5	0.5	1.5	1	1.25	1	1
65	10	28/09/2003	0.5	0.5	0.5	1.75	0.75	1	0	1
66	10	29/09/2003	0.5	0.5	0.5	1.75	0.75	1	0	1
67	10	30/09/2003	0.5	0.5	0.5	1.75	0.75	1	0	1
68	10	1/10/2003	0.5	0.5	0.5	1.75	0.75	1	0	1
69	10	2/10/2003	0.5	0.5	0.5	1.75	0.75	1	0	1
70	10	3/10/2003	0.5	0.5	0.5	1.75	0.75	1	0	1
71	11	4/10/2003	0.5	0.5	0.5	1.75	0.75	1	0	1
72	11	5/10/2003	0.5	0.5	0.5	1.75	0.75	1	0	1
73	11	6/10/2003	0.5	0.5	0.5	1.75	0.75	1	0	1
74	11	7/10/2003	0.5	0.5	0.5	1.75	0.75	1	0	1
75	11	8/10/2003	0.5	0.5	0.5	1.75	0.75	1	0	1
76	11	9/10/2003	0.5	0.5	0.5	1.75	0.75	1	0	1
77	11	10/10/2003	0.5	0.5	0.5	1.75	0.75	1	0	1
78	12	11/10/2003	0.5	0.5	0.5	1.75	0.75	1	0	1
79	12	12/10/2003	0.5	0.5	0.5	1.75	0.75	1	0	1
80	12	13/10/2003	0.5	0.5	0.5	1.75	0.75	1	0	1
81	12	14/10/2003	0.5	0.5	0.5	1.75	0.75	1	0	1
82	12	15/10/2003	0.5	0.5	0.5	1.75	0.75	1	0	1
83	12	16/10/2003	0.5	0.5	0.5	1.75	0.75	1	0	1
84	12	17/10/2003	0.5	0.5	0.5	1.75	0.75	1	0	1

Feed Analysis	DE	DM%	CP	Ash	ADF	NDF	Scoop weights
	MJ/kg DM		g/kg DM	g/kg DM	g/kg DM	g/kg DM	1 scoop
Red Clover Hay	9.4	82.9	17.6	9.5	28.3	40.1	2.08
Lucerne Chaff	8.8	82.9	23.2	7.7	28	35.7	0.45
Oats	12.2	87	12	2.3	10.8	22	1.27
Bran	11.5	84.9	18.9	4.1	14.8	34.3	0.94
NRM Sweetfeed®	12.6	84.3	14.2	4	10.7	20.3	1.73

Appendix B: Experiment two's feed programme

Horse ID	AM			Lunch		PM		
	Evolve	Prep	E.Jew	Evolve	Prep	Evolve	Prep	E.Jew
1	3/4	1	-	3/4	1/2	-	-	-
2	3/4	1	1	3/4	1/2	-	1/2	1
3	1	3/4	1	3/4	1/2	-	-	-
4	1	1	1	1	1	1/2	1/2	1
5	1	1	-	1	1/2	-	-	-
6	3/4	3/4	1	3/4	1/2	1/2	1/2	1
7	1/2	1/2	-	1/2	1/2	1/2	1/2	-
8	1/2	1/2	-	-	1/2	-	1/2	-
9	3/4	1	1	3/4	1/2	3/4	1/2	-
10	3/4	1/2	-	1/2	3/4	1/2	3/4	-

All horses had 1 scoop chaff in am and pm feed
 All feed units measured as per scoop

Evolve - NRM Evolve™

Prep - NRM Prepare™

E.Jew- NRM Equijewel™

Feed name	Average weight of one scoop (kg)
NRM Prepare	1.49
NRM Equijewel	0.65
Meadow Chaff	0.18
NRM Evolve	1.8
Meadow hay (per slab)	2.40

*Appendix C: Survey Questions***Outline of Questions Asked in Faecal pH Survey****TRAINER**

Trainer age
Full time /part time
Number of years training horses
Number of horses being trained
The trainer was asked to nominate a horse/s so the following questions could be asked.

NOMINATED HORSE/S

Sex of horse
Age
Classification of horse- sprinter, stayer, hurdler etc
Numbers of weeks in work

STABLING

Stabling type
Stable bedding material
Number of hours confined to stable or yard
How many hrs at pasture

FEEDING

Feed composition
Number of feeds/day
Amount given
Timing of feeds
What is feed measured with- scoop, ice cream container etc
Weight of a single feed measure
Any supplements used
Is hay given?
Amount of hay/day
Time given
Type of water feeding system
Feeding behaviour

TRAINING

Training programme for the week of interview
Where the horse is spelled

STABLE BEHAVIOUR- i.e. shows stereotypic vices.