Deterministic modelling of energy supply and demand for foaling mares managed at pasture on commercial Thoroughbred stud farms.

A thesis presented in fulfilment of requirement for the degree, Master of Science (Animal Science).

Massey University, Palmerston North, New Zealand.

Yin Ying Chin

2018

Abstract

There is an economic incentive to breed and foal mares early in the season within the New Zealand commercial Thoroughbred production industry. This has caused disparity between the natural and commercially imposed breeding season, and possibly, the period of peak pasture energy availability and the mare energy requirements. A deterministic model was developed to model the energy balance, energy intake and energy requirement of Thoroughbred mares managed at pasture under commercial conditions to assess their energy status. The response of energy intake and energy balance to changes in five variables were tested. The variables tested were dry matter intake, bodyweight, foaling date, pasture metabolisable energy, and energy requirement. For all foaling dates modelled, the mare was in energy surplus during pregnancy. Onset of lactation created a rapid and significant decrease in mare energy balance. Delay in foaling increased the magnitude of the post-partum energy deficit, and later foaling mares experienced a prolonged decrease in post-partum energy balance. The size of the deficit would, theoretically, decrease 1 body condition score, decrease circulating leptin concentration and initiate lean body mass mobilization which was previously found to negatively impact reproductive performance in the mares. The duration and size of the post-partum energy deficit could be reduced by shifting foaling date closer to the beginning of breeding season (1st September), thus synchronizing period of peak pasture energy with the mare's theoretical requirements. Energy balance and energy intake were more sensitive to pasture energy, dry matter intake and energy requirement changes with less influence from bodyweight to energy balance, suggesting pasture quality and quantity is an important influencer on nutritional status of the mare kept on pasture. Initiatives should be taken to monitor pasture quality and growth pattern, and optimize energy and dry matter intake in mares at pasture. These efforts could assist in maximizing pasture utilization, minimize energy deficit and achieve cost effective pastoral and nutritional management. When more pasture quality and growth data becomes available for equine stud farms, the model has the potential to achieve farm specific application on a wide range of equine body weights.

Acknowledgement

I would like to thank my supervisors Associate Professor Chris Rogers, Dr. Erica Gee and Dr. Penny Back for their guidance since my undergraduate studies. Their dedication and patience have allowed me to learn and work under a supportive environment. I am grateful for the opportunity to work on my topic of interest and with the teachers who inspired this endeavour. A special thanks to Dr. Javier Roca for going above and beyond in assisting with my thesis writing, constructing the nutritional model, data analysis, and providing helpful advice from time to time. Huge thanks to my family for their support, encouragement and acting as my reminder for being diligent and persistent in the face of challenges. This incredible journey would be impossible without them. Special thanks to Terry, Vicki and Cindy for sharing their knowledge and providing me with the opportunity to acquire more experience and understanding in the equine industry, which expanded my thinking and interpretation of my findings. This is dedicated to my good friend and role model, Adele: "Getting the most out of life is not about how much you keep for yourself, but how much you pour into others."

Table of Contents

List of Tables

List of Figures

Chapter 1: Introduction

The commercial equine population in New Zealand generates profit through the breeding and production of Thoroughbred race horses and has a large export focus with approximately 40% of the foal crop exported either as yearlings or racehorses (Waldron *et al.* 2011). Mares are managed using a pasture based system with varying pasture quality (energy and dead matter content) and quantity (growth rates/ dry matter production) throughout the year. This follows a defined pattern similar to sheep, beef and dairy pastures, having lowest pasture energy content during summer (January - March) and lowest pasture dry matter growth during winter (May - August) (Litherland and Lambert 2007; Hirst 2011). Pasture energy content increases from a nadir in summer and peaks in spring while dead matter follows an opposite pattern (Litherland and Lambert 2007; Hirst 2011).

The mean foaling date of New Zealand Thoroughbred mares was between 9th and 16th of October across eleven breeding seasons (Roca 2018, unpublished data). This aligns the period of increased energy requirement of mares with a period of low pasture dry matter production and pasture energy availability which would limit mare energy intake. Typically, pasture energy content starts to decline between October and November depending on the region (Litherland and Lambert 2007; Hirst 2011) which is the time where energy requirements of a mare would increase dramatically due to the onset of lactation (NRC 2007). During winter (May - August), pasture growth is lowest and dry matter production was reported to drop below animal dry matter requirements (Grace 2005). This period coincides with the last trimester of pregnancy of a mare that is due to foal in October. Together, these may initiate a decline in energy balance during late pregnancy and lactation and possibly subject mares to a prolonged energy deficit.

Under commercial stud farm conditions it is challenging to balance the constantly varying pasture energy supply and demand, and maintain quality pasture (Rogers *et al.* 2007). During the breeding season (September - December), stocking rates on breeding farms increase substantially over recommended rates due to the influx of mares for breeding and retention of young stock (Rogers *et al.* 2007; Hirst 2011). This poses a challenge in maintaining pasture quality due to increased grazing pressure and the selective grazing and faecal/ urine avoidance (“latrine”) behaviour in horses (Rogers *et al.* 2007; Hirst 2011), both which will limit voluntary feed intake and reduce the nutritive value of pasture available (Bruinenberg *et al.* 2002; Hirst 2011). Latrine avoidance leads to the formation of patches of un-grazed “roughs” in the area horses had previously defecated and shorter “lawns” in preferred grazing areas (Bott *et al.* 2013). The quality of “roughs” will decrease as they mature whereas the pasture in the “lawns” will diminish under high grazing pressure encouraging growth of undesirable plant species (i.e. weeds) (Bruinenberg *et al.* 2002; Bott *et al.* 2013). Mares access to pasture can also be limited by frequent removal of mares from paddocks for breeding management during the breeding season (Rogers *et al.* 2007). Therefore, despite general belief that the pasture available can sustain nutrient requirements of all horse classes year round, the nutritional status of the mare is subjected to variables such as fluctuating availability, access and quality of pasture, which are affected by season and commercial management factors.

According to cross-sectional surveys on New Zealand commercial stud farms, stud farms attempt to supplement mares with formulated feed during the last trimester of pregnancy but the main objective of this is to meet requirements for minerals and other specific nutrients instead of energy (Rogers *et al.* 2007; Hirst 2011). Supplementary feeding was least frequent during lactation, as it is assumed that mares energy requirement can be met with increased pasture availability during spring (Hirst 2011). This may not be true for mares that foal later and will be lactating through late spring and the early summer period when pasture growth and quality is declining. The reasons provided by stud managers as to why they chose to utilize supplementary feed were either difficulty of knowing if mares energy requirements are met from pasture or a solely pasture diet did not produce the desired body condition (Hirst 2011). There was also very little use of feed budgeting and estimation of pasture cover by breeding farms (Hirst 2011). Therefore, there is a need to develop tools and collect information on nutritional status of mares to assists in nutritional management on equine stud farms.

As reproductive performance is crucial to the Thoroughbred breeding industry, it would be advantageous to know the energy intake and energy balance changes of mares as dietary energy is the main driver of changes in body condition score (Henneke *et al.* 1984; Kubiak *et al.* 1987; Gentry *et al.* 2002), body weight (Henneke *et al.* 1984; Kubiak *et al.* 1987), leptin (Gentry *et al.* 2002; Salazar-Ortiz *et al.* 2011) and metabolic hormones such as insulin growth factor-1 (IGF-1) and insulin (Sticker *et al.* 1995; Gentry *et al.* 2002; Salazar-Ortiz *et al.* 2014). Decreases in the above parameters in mares were reported to negatively affect reproduction in terms of foal survival (Ransom *et al.* 2013), reproductive efficiency (Henneke *et al.* 1984; Nagy *et al.* 1998; Godoi *et al.* 2002), and ovarian function (Salazar-Ortiz *et al.* 2011; Salazar-Ortiz *et al.* 2014). Mares also had higher risk of embryonic loss if they lost weight or if their energy requirement during early lactation is not met (Newcombe and Wilson 2005). In dairy cows, negative energy balance and prolonged decrease in post-partum energy balance were associated with a longer post-partum to ovulation interval and ovulation failure in the first follicular wave in non-cystic cows (Beam and Butler 1998, 1999). All these evidence suggests decreases in body condition, body weight and energy balance should be avoided, to uphold reproductive performance. However, to date, the daily energy intake and energy balance changes of mares managed under commercial pasture conditions in New Zealand have not been investigated.

Chapter 2: Literature Review

2.1 The commercial Thoroughbred breeding industry

The Thoroughbred industry in New Zealand focuses on commercial breeding and production of racehorses. The breeding sector generates income from local and export sales of yearlings and "ready to run" 2-year-old racehorses (Waldron *et al.* 2011). Every year, approximately 40% of the foal crop is exported (Waldron *et al.* 2011). In 2017, these exports generated a return of $138m NZD (NZTBA 2017).

In New Zealand, the commercial breeding season runs from the 1st of September to the 1st of December (Rogers *et al.* 2007), with 87% of mating occurring before the 30th of November (Hanlon *et al.* 2012b). This condensed breeding window is shaped by the desire from breeders for mares to foal as close as possible to the official birthdate of all Thoroughbreds on the 1st of August (Rogers *et al.* 2007). This is done in an attempt to optimise foal maturity at yearling sales that occurs in February the following year after the foal was born, and due to the aversion of buyers towards foals born later than the last week of November (Waldron *et al.* 2011; Hanlon *et al.* 2012b; Gee *et al.* 2017).

Breeders also attempt to maintain a yearly foaling pattern in the mare to achieve greatest productivity and financial viability (Gee *et al.* 2017). To maintain a yearly foaling pattern, mares have a window of 25 days post-partum to conceive (Osborne 1966). Hence, breeding mares on their first post-partum oestrus (foal heat) will increase the likelihood of a mare getting pregnant within the breeding season (Gee *et al.* 2017). However, only 32% of Thoroughbred mares on commercial stud farms were served on foal heat and had a first cycle pregnancy rate of 54% (Hanlon *et al.* 2012a).

The imposed short breeding season, infrequent use of foal heat breeding and low first cycle pregnancy rate means emphasis should be put on early breeding to improve productivity through early foaling. This can potentially provide more breeding attempts after the foal heat thus could increase the chances of mares becoming pregnant within the commercially imposed breeding season (Hanlon 2012).

2.2 Commercial breeding season versus natural breeding season in the mare

The mare is a seasonally poly-oestrus, long day breeder (Nagy *et al.* 2000). The seasonality of reproduction is primarily influenced by photoperiod which controls the secretion of hypothalamic and pituitary hormones through the hypothalamic pituitary gonadal axis (Nagy *et al.* 2000; Davies Morel 2015). Increase in daylight length, or exposure to light during the photosensitive phase, suppresses melatonin production, which removes inhibition on gonadotropic releasing hormone (GnRH) and stimulates the release of pituitary hormones (follicle stimulating hormone, luteinizing hormone and prolactin) which are essential for folliculo-genesis and cyclic ovarian function (Hsueh *et al.* 1984; Garza *et al.* 1986). The annual reproductive cycle of a mare can be separated into the following stages:

1. Winter anoestrus: Reproductive system is inactive; no ovarian activity and oestrus behaviour.
2. Spring transition: Minimal to sporadic ovarian activity; follicles exhibit growth and regression but fail to ovulate.
3. Cyclicity (spring/summer): Marked by first ovulation of the year; followed by regular ovulatory oestrous cycles.
4. Autumn transition: Minimal to sporadic ovarian activity; transition into winter anoestrus.

To ensure pregnancy is achieved within the commercially imposed breeding season, breeders attempt to advance the onset of the reproductive cycle in the non-pregnant mare by artificially advancing the onset of transition using methods such as artificial lighting programmes and hormone therapy using injection or intra-vaginal implants of GnRH, GnRH agonists, progestin, dopamine antagonists and human chorionic gonadotropin (Aljarrah 2004; McCue *et al.* 2007).

After foaling, breeders may attempt to breed mares at foal heat if possible or may "short-cycle" the foal heat using prostaglandin to shorten the dioestrus interval (Lofstedt 1988). These management techniques aim to shorten the foaling to conception interval so that mares become pregnant again within the short breeding window hence maintaining a yearly foaling pattern, and preserving the artificially established reproductive season (Lofstedt 1988; Gee *et al.* 2017).

The incentive to foal and start breeding early in the breeding season highlights the disparity between the commercially imposed and natural breeding season of the mare. This can be observed when comparing the commercial breeding season to the proportion of mares ovulating during the commercial breeding season in the southern hemisphere (Figure 1, Hanlon (2012)). At the start of commercial breeding season (1st September) in spring, only 20% of non-pregnant mares are ovulating. The highest proportion of mares ovulating is seen in January, which is one month after the commercial breeding season has ended.

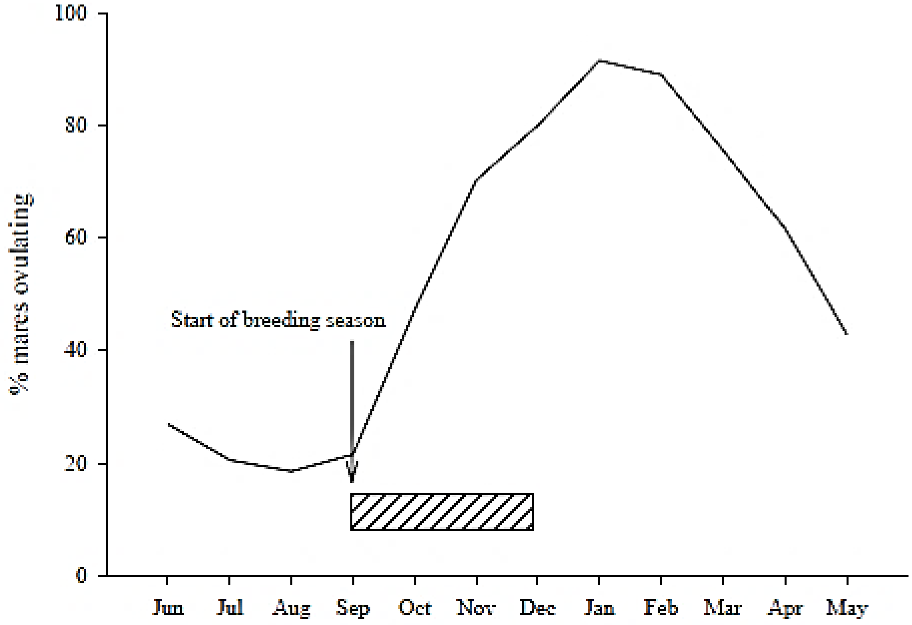


Figure 1. Proportion of mares ovulating each month over a 12 month period in South Eastern Australia. Data obtained from Sydney horse abattoir. Hatched area represents commercial breeding season. Figure from Hanlon (2012).

Under feral conditions with seasonal pasture growth and energy content, foaling occurs during peak pasture availability to maximize offspring survival (Ransom *et al.* 2013). Given that mares have a long gestation length (344 - 352 days, *Gee et al.* 2017), conception needs to occur between late spring and summer (December - March) to ensure foaling occurs during the period with optimal environmental and nutritional conditions (Hanlon 2012), presumably to provide adequate and quality food supply to meet the increased energy demand by the lactating mare. Under the New Zealand commercial pastoral conditions, the annual reproductive cycle of the mare can be displaced from the seasonal pattern of pasture growth and energy content changes, by a disparity of the commercial and the natural breeding seasons. In turn, the pasture and energy supply during the period of increased energy demand may be affected.

2.3 Seasonality of pasture

New Zealand pasture exhibits seasonality in quality, availability and growth rates. The main determinant of pasture quality is the energy concentration of pasture (Litherland and Lambert 2007). The energy content of the pasture is dependent on the ratio of cell wall and cell contents, and the digestibility of cell wall (Litherland and Lambert 2007). Cell contents include readily digestible non-structural carbohydrates, proteins and organic acids (Litherland and Lambert 2007; Longland 2012). Cell wall includes lignin, cellulose and hemicellulose. The digestibility of forage dry matter and cell wall are dependent on, and negatively correlated to the lignin concentration (Mowat *et al.* 1969; Jung *et al.* 1997; Longland 2012). Pasture quality will decrease with age (maturity), ambient temperature rise and dead matter accumulation due to a decrease in energy content and digestibility (Litherland and Lambert 2007).

As pasture matures, the stem to leaf ratio decreases due to elongation of stem (Bruinenberg *et al.* 2002; Longland 2012). The degradability of dry matter will also decrease due to an increase in cell wall content with decreases in their digestibility (Bruinenberg *et al.* 2002), and decreases in the non-structural carbohydrates due to reproductive growth (Waite and Boyd 1953; Mackenzie and Wylam 1957; Bruinenberg *et al.* 2002).

Increase in the ambient temperature accelerates the rate of plant morphological aging for example, the leaf appearance rate, reduction rate in stem to leaf ratio and leaf to sheath ratio, and time to death (Litherland and Lambert 2007). Increase in ambient temperature also accelerates increase in lignin content of individual plant morphological components (Litherland and Lambert 2007).

The proportion of dead matter in the pasture is the major determinant of pasture quality as it explains around 70% of variation in the dry matter digestibility of spring and autumn pasture (Litherland and Lambert 2007). The mean dead matter energy content summarized from the literature was 5.7 MJ ME/kg DM (4 - 8 MJ ME/kg DM) with organic matter digestibility of dead stem and dead leaf ranging from 19 - 25% and 53 - 56%, respectively (Litherland and Lambert 2007). The energy content and organic matter digestibility of dead matter are lower compared to the average pasture dry matter energy content (11 MJ ME/kg DM) and organic matter digestibility (77% DM) (Litherland and Lambert 2007).

Therefore, increases in the percentage of dead matter in pasture, aging of tillers, together with lack of new growth will decrease the pasture quality. Occurrence of these events are influenced by water supply (i.e. from rainfall), temperature (correlated with sunlight), and grazing management with the former two being found to vary throughout the year in different regions of New Zealand (Hirst 2011).

2.3.1 Energy content of pasture

Equine and dairy/sheep pasture data (Table 1) show that energy content rises from autumn (10.7 - 10.9 MJ ME/kg) and over winter (~11 MJ ME/kg DM) and reach peaks in spring. Energy content was highest during early spring (11.5 - 12.5 MJ ME/kg DM) or when plants were in the leafy state in spring (12 MJ ME/kg DM) (Table 1). Energy content then decreases and reaches nadir in summer (ranging from 8 - 10.5 MJ ME/ kg DM between studies, lowest in summer stalky plants).

Overall, equine pasture and dairy/sheep pasture energy patterns follow a similar seasonal trend. However, there was considerable variation in energy content and the pattern of energy content changes in dairy pasture between regions (Litherland and Lambert 2007). In contrast, a study of equine pasture by Hirst (2011) observed less variation in energy content, possibly due to fewer data points compared to the dairy study. Equine pasture was sampled bi-monthly in Hirst (2011) while dairy pasture samples were taken monthly in Litherland and Lambert (2007). Hence, the results provided in Hirst (2011) only demonstrated a seasonal trend instead of a monthly trend. The dairy study had a larger sample size and was representative of both research and commercial dairy/sheep farms (n=6328, years 2002 - 2005) (Litherland and Lambert 2007). Alternatively, the equine pasture study data was representative of 40% of commercial breeding farms in New Zealand with 46% of participating farms clustered within the Waikato region (Hirst 2011).

Table 1. Average pasture energy content for equine and dairy pasture.

Season	[1]Pasture energy (MJ DE/kg DM) in equine pasture	
Autumn	10.8	
Winter	11.4-11.2	
Spring	10.8-11.8	
Spring (leafy)	12.0	
Summer (leafy)	10.3	
Summer (stalky)	8.0	
	Pasture energy (MJ ME/kg DM) in equine vs dairy pasture	
	[2]Equine Pasture	**[3]Dairy Pasture**
Autumn	10.7	10.9
Winter	11.2	11.0
Early Spring	12.5	11.5
Late Spring	12.0	11.4
Summer	9.3	10.5

[1]Hoskin and Gee (2004); [2]Hirst (2011);[3]Litherland and Lambert (2007)

2.3.2 Pasture chemical composition

The chemical composition changes in equine pasture reported by Hirst (2011) are shown in Table 2. The dead matter percentage in the pasture was lowest during spring, reaching its peak in summer. Neutral detergent fibre (NDF) content of pasture follow a similar trend whereas organic matter digestibility (OMD) follows an opposite trend. Soluble carbohydrates were lowest in summer and highest in late spring. These changes in chemical composition agree with the life cycle of perennial pasture. From spring to summer, forage enters a reproductive state from vegetative state (Bruinenberg *et al.* 2002). Cell wall content (NDF) increases with the maturity of the plant which is associated with a decrease in the digestibility of plant (Bruinenberg *et al.* 2002). With a rise in temperature and daylight length during summer, the rate of morphological aging and death of the plant is accelerated hence the drop in soluble carbohydrates and increase in dead matter percentage.

Table 2. Seasonal variation in chemical composition of equine pasture in New Zealand from Hirst (2011).

Mean ±SEM (%DM)	Summer	Autumn	Winter	Early Spring	Late Spring
ADF	28.0±0.4^{a}	25.2±0.3^{be}	24.1±0.3^{c}	21.9±0.2^{d}	24.9±0.2^{be}
NDF	55.8±0.7^{a}	43.4±0.5^{bc}	43.2±0.7^{bc}	39.0±0.4^{d}	46.5±0.4^{e}
Soluble CHO	5.1±0.3^{a}	10.5±0.3^{bcd}	10.9±0.3^{bcd}	11.0±0.4^{bcd}	12.3±0.3^{e}
OMD	63.5±0.8^{a}	74.9±0.7^{b}	76.8±0.8^{c}	86.4±0.5^{d}	80.3±0.4^{e}
Dead Matter	19.8±1.8^{a}	4.1±0.7^{bce}	3.7±1.2^{bcde}	0.6±0.4^{cde}	2.9±0.8^{bcde}

Within a row, averages with different superscripts are significantly different, $P<0.05$.
SEM= standard error of mean
%DM= values expressed in percentage of dry matter
ADF= Acid detergent fibre
NDF= Neutral detergent fibre
CHO= Carbohydrates
OMD= Organic matter digestibility

2.3.3 Pasture availability

There is a wide variation in dry matter yield of perennial pasture between regions (Figure 2, Litherland and Lambert 2007). Data collected from a dairy farm in Manawatu shows year to year variation in seasonal and annual pasture production within a farm (Figure 3, Litherland and Lambert 2007). These results suggest dry matter yield is different between year, region, and farm. However, a consistent trend can be observed. The pasture yield is lowest during winter and then increases during spring. Pasture yield then decreases around October (late spring) and reaches its second nadir of the year in summer, with exceptions where irrigated farms in the Canterbury and all farms in Southland regions maintained pasture yield through spring and summer (Figure 2, Litherland and Lambert 2007).

In summary, there is limited information on pasture energy content, availability, growth and quality on commercial equine stud farms in New Zealand. The information available on pasture energy and chemical content changes on commercial equine stud farms were collected from five Thoroughbred breeding stud farms in the Waikato region by Hirst (2011). Combining information from dairy/ sheep (Litherland and Lambert 2007) and equine (Hoskin and Gee 2004; Hirst 2011) studies, available pasture data showed pasture quality is best during spring. After that, quality starts to decrease and reach the lowest point in summer. The pasture energy content starts to decrease between October and November depending on region with an earlier decrease in warmer regions. The quality will gradually recover over the autumn and winter and peak again in the next spring. Pasture growth is lowest during winter and highest in late spring with the second lowest point occurring in summer. The pasture growth rate (kg DM/ha/day) varies between year and regions.

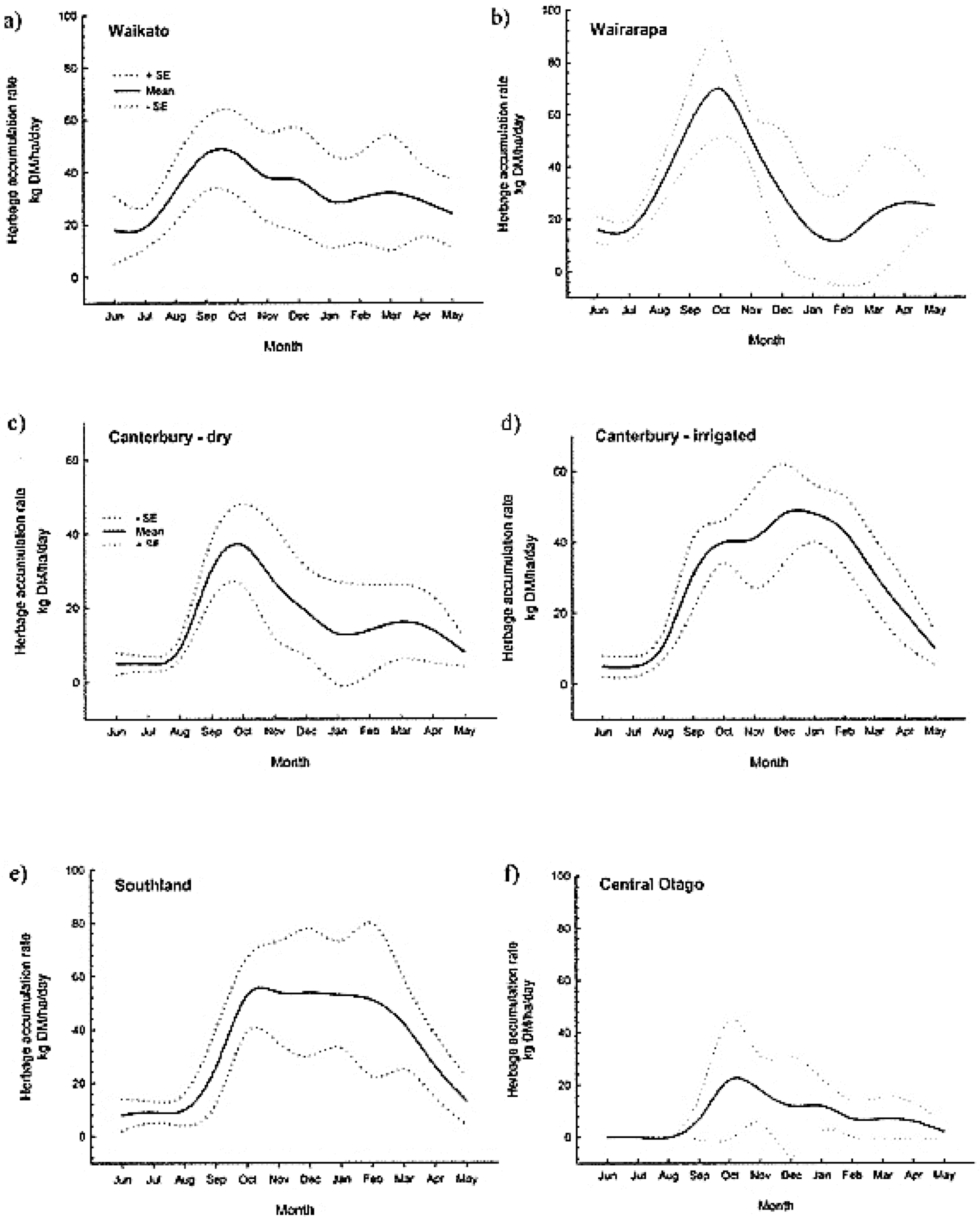


Figure 2. Perennial dairy pasture yield in different regions of New Zealand a) Waikato, b)Wairarapa, c) Canterbury-dry, d) Canterbury-irrigated, e) Southland, f) Central Otago (Valentine and Kemp 2007).

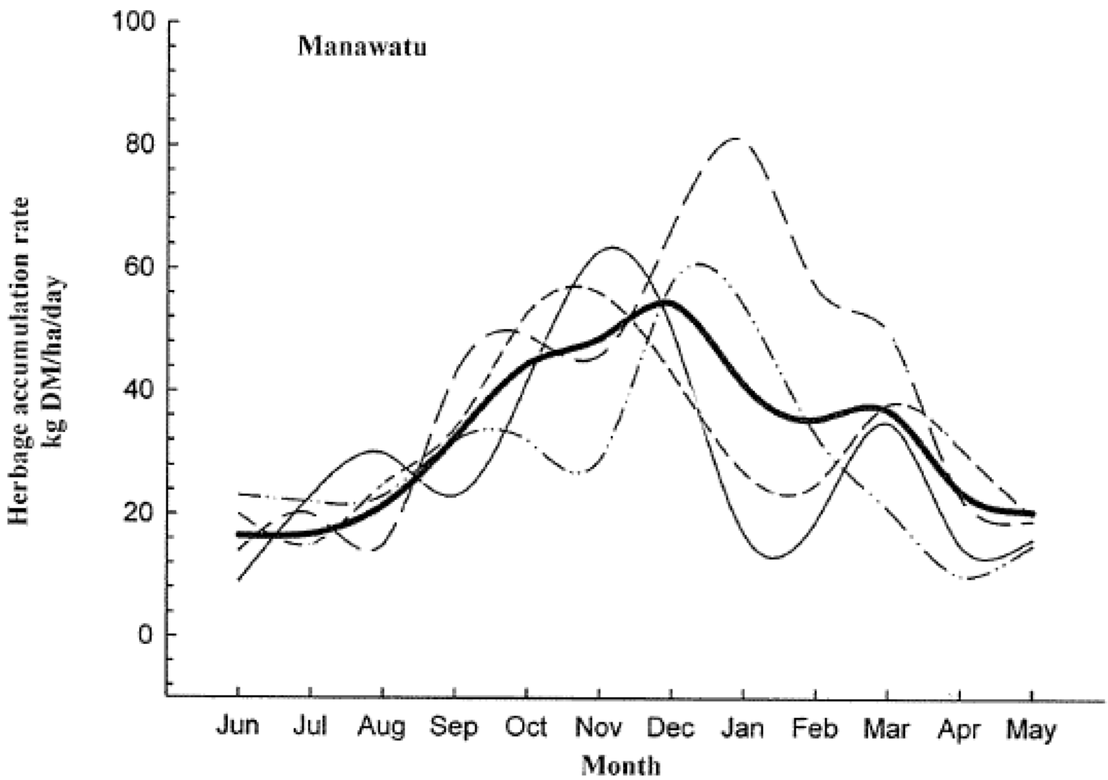


Figure 3. Four year actual pasture yield and seven year mean pasture yield (thick bold line) on a Manawatu dairy farm, showing year to year variation in seasonal and annual pasture production (Litherland and Lambert 2007).

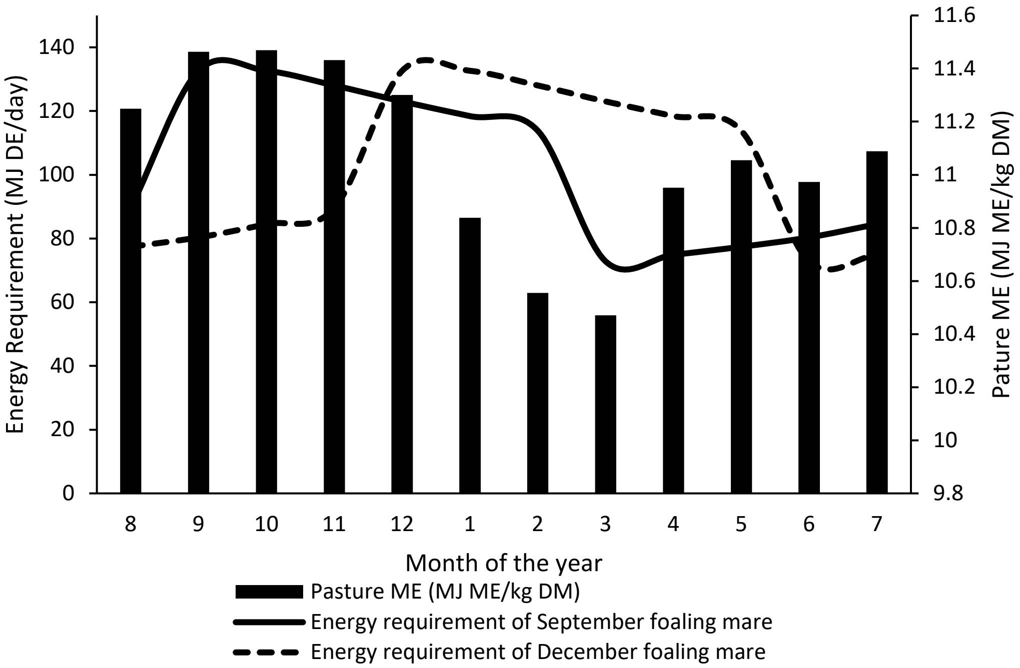
2.4 Energy supply versus energy demand of mares under pastoral system.

The seasonal supply of pasture and how well it is matched to the animal requirements determines the efficiency of pastoral system (Litherland and Lambert 2007). The energy requirements of a pregnant mare starts to increase at five months of pregnancy followed by a further and drastic increase at the onset of lactation immediately after foaling (NRC 2007). Lactation typically continues for five to six months in Thoroughbred broodmares, ending when foals are weaned (Rogers *et al.* 2007). The timing of energy requirement changes is affected by the date of foaling (Figure 4a).

It can be observed in Figure 4a that the period of low pasture energy content coincides with the highest energy requirements of the mare. The degree of overlap between these two periods is dependent on the foaling date. For a December foaling mare, her period of maximum energy requirement will coincide with the lowest pasture energy content of the year whereas, the pasture energy available to a September foaling mare at the same physiological time point is higher. There are limited published information regarding foaling dates of New Zealand Thoroughbred mares. Analysis of unpublished data indicates that the range of the mean foaling date is 9^{th} of October and 16^{th} of October between years 2005 - 2015 (Roca 2018, unpublished data).

This preliminary observation indicates there is an interaction between foaling date and pasture energy availability, and that under commercial conditions the period of high energy requirement of mares is displaced from the period of high pasture energy content. Under a pasture based nutrition scheme the lack of synchronisation may have a negative influence on the energy status of the mare. In the northern hemisphere, mares that foaled in January with limited access to and supply of pasture lost weight, and had lower body weights and lower body condition scores post-foaling than mares that foaled in March, April and May (Pagan *et al.* 2006), supporting an association of foaling date with mare energy status. However, the adequacy of energy supply is also influenced by voluntary feed intake which ultimately determines the energy intake achieved by the mare.

a)



b)

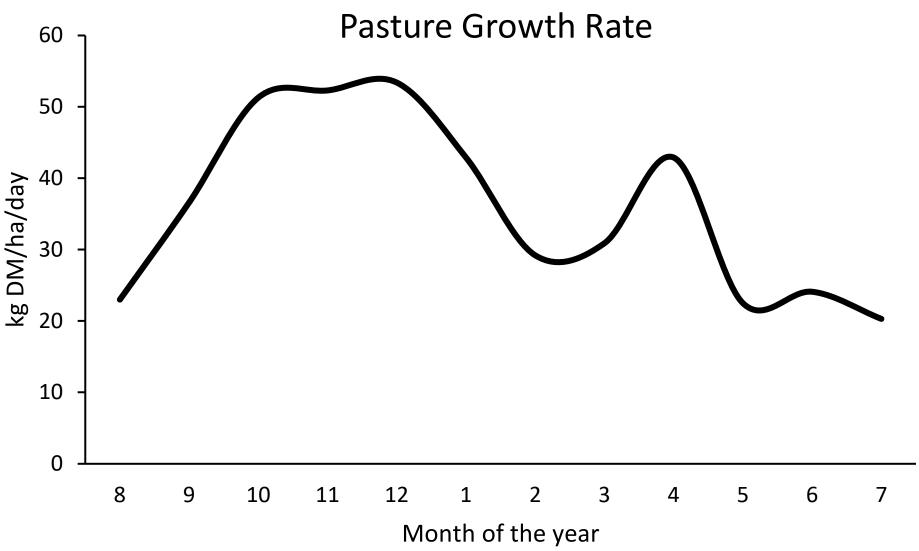


Figure 4. a) Energy requirement of a 500 kg mare foaling at different times of the year (MJ DE/day) versus the pasture metabolisable energy available (MJ ME/kg DM); b) Annual pasture growth rate on Dairy 1, Massey University, Palmerston North in 2015-2016 (Correa 2015 - 2017, unpublished); Energy requirement obtained from NRC (2007); Pasture metabolizable energy obtained from Litherland & Lambert (2007).

Under commercial pasture management conditions, there are several factors that may influence voluntary dry matter intake such as the pasture availability (Collas *et al.* 2015), time available for grazing (Glunk *et al.* 2013) and pasture quality (Hoskin and Gee 2004). Pasture availability is low in winter and summer and may limit voluntary intake (Figure

4b). However, the relationship between herbage allowance and dry matter intake is not well understood in mares. It was previously reported that pasture dry matter production drops below mare dry matter requirements during winter (Grace 2005). This needs validation for commercial equine farms, as there are no published data regarding pasture dry matter production and pasture cover on equine farms in New Zealand.

During the breeding season, stocking rate increases over the recommended density due to influx of mares for breeding and retention of young stock (Hirst 2011). Horses also exhibit selective grazing and faecal/ urine avoidance ("latrine") behaviour where they would avoid grazing paddock areas they previously defecated on and form patches of un-grazed "roughs" and shorter "lawns" (Lamoot *et al.* 2004; Bott *et al.* 2013). Selective grazing and "latrine" behaviour in horses were reported to decrease pasture utilization for up to 52% and are the main factors for decreased pasture quality on equine farms (Ödberg and Francis-Smith 1976; Bott *et al.* 2013).

Hence, pasture on offer and pasture quality can decrease during breeding season due to increased stocking density and limited pasture utilization. Mares are also routinely removed from paddocks to carry out veterinary and health care procedures which may also decrease the time available for grazing (Rogers *et al.* 2007). Therefore, pasture cover and stocking density information alone may not provide an accurate prediction of mare voluntary intake.

At present, there is no published information regarding pasture intake and nutritional status of New Zealand Thoroughbred mares managed commercially on pasture, and their impact on energy intake, energy balance, body weight changes, and body condition score changes. Within the industry, there also appears to be limited use of feed budgeting, monitor of pasture cover and body weight (Hirst 2011). The stock management decisions are not based on pasture cover but on other management aspects (i.e. accessibility) (Hirst 2011).

The studies on body weight and body condition score changes of Thoroughbred mares (Pagan *et al.* 2006), and Quarter horse mares (Lawrence *et al.* 1992) on pasture have been conducted in the northern hemisphere. Body condition score changes of ponies on pasture (Salazar-Ortiz *et al.* 2011) and annual body weight changes in Standardbred mares in Australia have also been reported (Newcombe and Wilson 2005). Many of these studies used body weights and body condition scores to examine the effects of nutritional status of the mare on reproductive performance and foal growth.

As the New Zealand commercial Thoroughbred breeding industry operates on a predominantly pasture based system and reproductive performance of the mare is central to the productivity of the industry, more attention should be directed towards investigating the inter-relationship of the mares nutritional state and their reproductive performance. A greater understanding of the interaction between mare foaling date, energy requirements, pasture quality, and pasture availability changes will allow improvements in the efficiency of pasture utilisation and maximize the use of this cost effective feed source.

2.5 Dry matter intake

Voluntary feed intake on pasture can be estimated by comparing difference in pre-grazing and post-grazing herbage mass and by utilising synthetic markers such as alkanes (Smit *et al.* 2005). However, it is challenging to measure dry matter intake (DMI) of horses on pasture due to their selective grazing and "latrine" behaviour that leads to uneven sward heights (Ödberg and Francis-Smith 1976). Hence, information on DMI of horses on pasture is limited. A few studies have been reported to measure voluntary feed intake of horses on pasture using marker techniques (Cr_20^3 and alkanes) but with limited success (Hoskin and Gee 2004).

The relationship between diet, DMI and the resulting energy intake is not well understood because the substitution of supplement, for pasture, resulted in little or no increase in total energy intake (Collas *et al.* 2014). Increase in pasture quality has been found to both increase and decrease DMI in horses (Hoskin and Gee 2004). Collas *et al.* (2014) reported no difference in energy intake between supplemented and non-supplemented mares due to increased voluntary pasture intake in non-supplemented mares. In contrast, Glunk *et al.* (2013) restricted time of access to pasture in mature horses which led to increased DMI rate (dry matter intake per unit time). Despite the increase in DMI rate, the horses failed to meet their energy requirements from pasture.

There is also limited information regarding the effects of herbage allowance on DMI of horses. According to a review by Hoskin and Gee (2004), McMeniman (2003) suggested forage intake of horses will not increase with increasing sward height and Hughes and

Gallagher (1993) reported that herbage intake rate of horses remained constant with increasing sward height due to increased bite size and decreased bite rate. Increases in herbage allowance may increase bite size, and bite time which limits the intake rate (Fleurance *et al.* 2009). In contrast, Collas *et al.* (2015) found DMI increased linearly with herbage allowance independent of energy supplement due to an increase in grazing time and intake rate. Bite size and bite rate were not measured in this experiment therefore the effects of herbage allowance on DMI remains inconclusive.

In the same study conducted by Collas *et al.* (2015), the digestibility of organic matter decreased between high and low herbage allowance. The authors attributed this observation to an increase in proportion of fibre ingested because the mares grazed closer to the ground. At low herbage allowance, the mares also reduced grazing activity towards the end of experiment possibly due to low post-grazing mass. This indicates that overgrazing due to high stocking density may have a negative effect on digestible energy intake even when intake rate is maintained because of reduced organic matter digestibility. If the mare remains on an overgrazed pasture, her grazing activity will be modified, affecting intake. This information has implications to the grazing management on breeding stud farms during periods of high stocking density.

Studies on voluntary DMI of lactating, non-lactating mares, and mature horses were reviewed and summarized in Table 3. Daily DMI of lactating mares were between 1.95 - 3% body weight. Values reported by Grace *et al.* (2002) for lactating Thoroughbred mares in New Zealand with 24 hours access to pasture were 2.5% which is in agreement with the value provided by the NRC (2007). The daily DMI for non- lactating mares was reported to be 1.5 - 2.2% BWT, and 1.4 - 2.5% for mature horses (Table 3). There is no published information on the DMI of pregnant mares fed pasture. In ruminants, there is data demonstrating a reduction in DMI during late pregnancy with increasing size of the fetus (Forbes 1970; Stanley *et al.* 1993). However, at present there is a lack of robust data on how the horse can alter DMI and transit time to optimise energy absorption (Van Weyenberg *et al.* 2006), and thus understanding from other species cannot be easily applied to the horse in late pregnancy. At present the recommended intake provided by NRC (2007) for pregnant and adult horses was 2% body weight. INRA (2015) provided recommended minimum and maximum dry matter requirements by stage of pregnancy, but these do not represent the minimum or maximum quantity that the mare can consume.

Table 3. Estimated dry matter intake (DMI) of horses fed different types of forages and supplements.

Type of horse	Type of forage	DMI (g DM/kg BW)	DMI of 500 kg horse (kg DM/d)	DMI (%BWT)	n=
Lactating TB	[1]Perennial ryegrass/ white clover mix	24	12	2.5	8
	[2]Orchardgrass / ryegrass (early vegetative)	24-26	12-13	2.3-2.6	3
	[2]Orchardgrass / ryegrass (mid bloom)	26-31	13-15.5	2.6-3.1	3
	-		[3]12.5	2.5	
Lactating Anglo-Arab/ French saddle	[4]Low- high herbage allowance SUP barley	22.4-27	11.2-13.5	2.24-2.7	18
	[4]Low- High Herbage allowance non-SUP	19.5-24.8	9.75-12.4	1.95-2.5	18
Lactating saddle mare	[5]25 plant species	28g	14	2.8	8
	[5]SUP with concentrate	22.6	11.3	2.26	
Non-lactating TB	[2]Orchardgrass (cocksfoot)/ perennial ryegrass	15-22	7.5-11	1.5-2.2	2
	-		[3]10	[3]2	
Pregnant mare	[6]Brome grass hay, alfalfa hay + concentrate	-	-	1.2	12
	-		10	[3]2	
Mature horse	[7]Tall fescue, intake rate*15hrs grazing	-	-	1.43-2.55	8

[1]Grace *et al.* (2002);[2]Marlow *et al.* (1983);[3]NRC (2007);[4]Collas *et al.* (2015);[5]Collas *et al.* (2014);[6]Guay *et al.* (2002);[7]Dowler *et al.* (2012)
SUP: Supplemented feed

2.6 Energy systems

Different types of energy systems can be used to describe energy contents in the feedstuffs, the partitioning of the energy utilised by the animals and predicting energy requirements (NRC 1981) (Figure 5). The following section will be based on these reference sources, NRC (2007), NRC (1981), and INRA (2015).

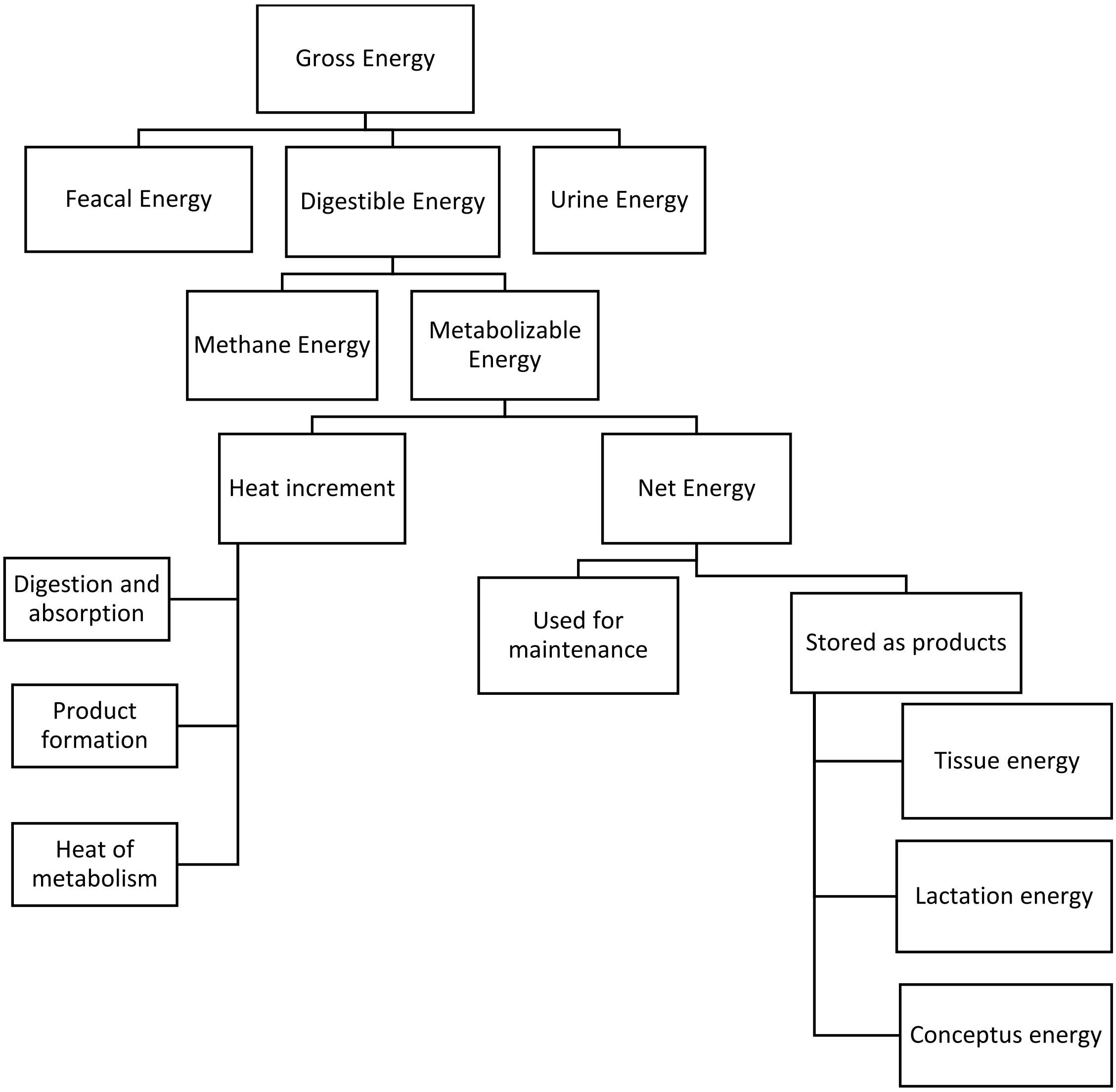


Figure 5. Feed energy partitioning. Information adapted from Waghorn *et al.* (2007) and NRC (2007).

Briefly, gross energy represents the amount of heat produced from the total combustion of the feed in a bomb calorimeter. Food consumed are not completely digested and will be excreted in the faeces. Energy contained in the excreted faeces will be lost and not available to the animal. The difference between energy consumed and energy excreted is termed digestible energy (DE). The DE of a feed can be determined by conducting digestibility trials that involves total faecal collection to determine excreted nutrients in the faeces. Determining the digestible energy value of every feedstuff through digestibility trials is costly and time consuming hence equations were developed based on the chemical composition of feed and their digestibility coefficients to estimate the digestible energy of a feed.

Horses are hind-gut fermenters. Fibre and undigested water soluble carbohydrates that escaped from the small intestine will undergo microbial fermentation in the large intestine and produce volatile fatty acids as an energy source to the body and methane as a by-product (Geor and Harris 2007; Kienzle and Zeyner 2010). Energy compounds in methane cannot be utilized and therefore are considered as lost energy. Energy can also be lost in the form of urine (Kienzle and Zeyner 2010). Urinary losses increase with greater protein content of the diet (Kienzle and Zeyner 2010). The energy available after methane and urine losses is termed metabolisable energy (ME).

Energy is required for the mastication, digestion, absorption and metabolism of ingested food. These processes utilise energy and generates heat which is referred to as heat increment. After deducting the energy lost in these processes, the energy available to be partitioned to maintenance, work and production can be determined. The energy available for maintenance, work and production is termed net energy (NE). End products of digestion are used at different levels of efficiency for different physiological functions. Each feed has different digestion end products depending on its chemical composition and digestibility. Therefore, multiple net energy values for different purposes have to be assigned to one feed.

2.7 Published recommendation for nutrient requirements for horses.

There are multiple published feeding standards for horses which include the National Research Council (NRC) requirements (NRC 2007), French horse net energy system (INRA) (INRA 2015) and German feeding standards (Coenen *et al.* 2011). This section will be based on the following reference sources: NRC (2007), INRA (2015), Coenen *et al.* (2011) which are the published materials for each system unless cited otherwise.

The NRC requirements (NRC 2007) are based on heat production obtained from horses fed at different levels with either forage, hay, grain or mixture a of hay-grain diet. The digestible energy requirements were then obtained using the estimated efficiency of DE use for ME within each study. The measurements were taken when horses were confined within a metabolic chamber with minimal physical activity. Therefore, requirements will need to be adjusted for husbandry conditions where horses are housed in groups, grazing on large pasture, moved frequently, and housed outdoors under different weather conditions.

Group housing may increase animal maintenance requirement up to 10% (Coenen *et al.* 2011). Foraging may increase energy requirement by 10% (Vermorel *et al.* 1997) and up to 50% for horses grazing large pasture (Coenen *et al.* 2011). Heat and cold will increase energy requirements by 10 - 20% (Coenen *et al.* 2011). Additionally, there is a report on variation in maintenance requirements between breeds, with Thoroughbreds reported to have the highest maintenance requirement compared to other horse breeds such as warmbloods, Quarter horses, and Icelandic horses (Coenen *et al.* 2011). The maintenance requirement of Thoroughbreds is 23%, 28%, and 46% higher than warmbloods, Quarter horses, and Icelandic horses, respectively (Coenen *et al.* 2011). Therefore, the energy requirement of a Thoroughbred mare on pasture may be substantially higher compared to other horse breeds.

The French horse net energy system (INRA 2015) was developed to account for differences in utilisation of ME from different feeds due to differences in proportion of digestion end products and the biochemical pathways used by these end products to produce energy (Harris 2001). This system also uses a horse feed unit which corresponds to the NE available to a horse in one kg of standard barley at maintenance which is not a common feed unit utilised within New Zealand. The NE is obtained based on the DE content of feed, the efficiency of DE use for ME, and the efficiency of ME utilisation for maintenance (k_m) (Martin-Rosset *et al.* 1994). The utilisation efficiency of ME for maintenance was based on limited data from feeding trials in horses fed hay or a mixed diet. The feed energy value used was derived for ruminants which may not reflect the true energy value for horses due to a higher ability to utilise fibre energy in cattle compared to horses (Pearson *et al.* 2006). The same efficiency value is used for ME utilisation for production purposes as the system assumes maintenance energy accounts for the largest part of total energy requirement which may not be true for lactating horses (Martin-Rosset *et al.* 1994; INRA 2015). The lactation energy requirement can contribute 50% of total daily requirements (INRA 2015). In order to practically utilise the French NE system, a NE value for feed is required. A different NE value is also required for different production purposes for each feed. At present, a NE value is not yet available for New Zealand pasture.

The German feeding standards were first published in 1994 and were based on digestible energy. The system was recently revised in 2011 and is moving towards integrating ME into the feeding standards (Coenen *et al.* 2011). The ME is estimated using an equation developed by Kienzle and Zeyner (2010) which included correction factors obtained by measuring methane and urinary losses correlated to crude protein and fibre intake. At present, there is still limited data available for renal and methane losses in equine feedstuffs. Metabolisable energy requirements in the German system were determined based on data obtained from indirect calorimetry in warmblood, heavy warmblood and ponies (Coenen *et al.* 2011). The data were then standardised for breed by setting the warmblood as 100% and assuming the maintenance requirement of Thoroughbreds to be 23% higher than warmblood horses (Coenen *et al.* 2011).

Unlike the NRC system that used body weight to determine maintenance requirements, the maintenance requirements in French horse feeding system and the German feeding standards are based on metabolic body weight (kg $BWT^{0.75}$). This exponent states that body surface is not a linear function of body weight in order to account for differences between animals with widely differing sizes. A previous study concluded that maintenance energy requirements varied linearly with body weight for equids weighing 125 - 856 kg (Pagan and Hintz 1986). It was suggested that use of this linearity poses a disadvantage for horses due to inclusion of ponies in the study (Coenen *et al.* 2011). It is thought that ponies have lower maintenance requirements than other breeds e.g. the Thoroughbred which is higher in lean body mass (Coenen *et al.* 2011). This also underlines the possibility that energy requirement is not entirely dependent on body size. However, there is a lack of published evidence to support benefits of using metabolic scale to estimate energy requirement. Moreover, body composition was found to vary between individuals, fitness status and age as well as breed (INRA 2015). Therefore, without a study demonstrating that using metabolic body weight will improve the estimation of energy requirements for horses differing in body size, body composition and training level, it is unknown which method of estimation is more advantageous.

Considering that other nutrients and energy required for other purposes are estimated based on bodyweight, using a metabolic scale may add to the complexity and limit practicality particularly in terms of modelling.

2.8 Estimating energy requirements in mares

The methods used to determine daily energy requirements of pregnant and lactating mares are uniform across the published systems described earlier (NRC 2007; Coenen *et al.* 2011; INRA 2015). Energy requirements can be determined using a factorial approach for pregnant and lactating mares by assessing requirements for maintenance, pregnancy and lactation separately.

A maintenance requirement is the amount of energy required to prevent a change in total energy contained in the body under thermo-neutral environment (NRC 1981). Total energy required during pregnancy includes additional energy required for the growth of fetus, placental tissue and uterus, and maintenance of these tissues (NRC 2007; Coenen *et al.* 2011; INRA 2015). Therefore, growth of fetus and non-fetal tissue needs to be modelled. The energy required for the growth of fetus, placenta and uterus is based on energy required to deposit the tissue, energy retained in the tissue and the efficiency of energy utilised in these processes (NRC 2007; Coenen *et al.* 2011; INRA 2015). The energy required to maintain the deposited tissue is based on the oxygen consumption of the entire conceptus (NRC 2007; Coenen *et al.* 2011). Energy required to support lactation is the product of milk yield, energy content of the milk and the efficiency of energy utilization for milk production (NRC 2007; Coenen *et al.* 2011; INRA 2015).

2.8.1 Pregnancy

2.8.1.1 The fetal growth models

The German system described in Coenen *et al.* (2011) developed a model for fetal growth by statistically transforming fetal body mass into percent of birthweight using data from aborted fetuses collected by Meyer and Ahlswede (1978). To enable application to all types of horse breed, the growth curve is then normalized to kg birthweight with assumptions that dynamics of prenatal growth and chemical composition of the conceptus are not different between breeds. This model estimates the birthweight of a foal from a 500 kg mare to be 51 kg using an estimated gestation length of 331 - 338 days.

The French system draws data from three studies (INRA 2015). A polynomial model was then developed by comparing and adjusting the analysis of residual between adjusted value and residual value using three models. The body weights predicted in this model are then used in predicting fetal weight based on % dam weight. The estimated birthweight is 55.6 kg or 10.1% of dam weight for light horse breeds (body weight undefined), and gestation length is estimated to be 340 days.

The NRC (2007) uses body weight data from studies on aborted and still born foals between day 150 and term of gestation reported in Meyer and Ahlswede (1978), Platt (1978), and Giussani *et al.* (2005) to model fetal weight as percentage of birthweight. Birthweight was estimated at 9.7% of dams' bodyweight (48.5 kg for 500 kg mare) and gestation length is estimated to be 339 - 342 days (NRC 2007).

Data for fetal-growth *in utero* is limited as all three systems included data from still born and aborted fetuses. The NRC and the French model used three data groups whereas the German system used one data group (NRC 2007; Coenen *et al.* 2011; INRA 2015). The approach of modelling differed between systems but the outcomes were similar. All models used a gestation length of around 340 days with minimal weight gain in the first five months of gestation, and increased growth per unit time in the last five months of gestation (NRC 2007; Coenen *et al.* 2011; INRA 2015). The exception to this was the German model reflecting the majority of the increased growth rate occurring in the last 90 days of gestation (Coenen *et al.* 2011). The gestation lengths used in all models are shorter than the reported mean gestation length of New Zealand Thoroughbred mares which were 350±10 days (Rosales *et al.* 2017) and 352±10 days (Van Rijssen *et al.* 2010) but is within the standard deviation range. The gestation length of New Zealand Thoroughbred mares were found to be longer compared to mares of the same breed in the United Kingdom (344 days) (Davies Morel *et al.* 2002) potentially due to differences in latitude, climatic conditions and feed availability (Gee *et al.* 2017). However, the estimated birthweight was around 10% of maternal weight for all models which is in agreement with reported birthweight of Thoroughbred foals between 50.3±5.9 and 51.2±6.2, which also equivalent to 10% of mare body weight (Platt 1984). Therefore, the fetal growth models from all three systems appear to be representative of fetal growth in Thoroughbreds.

2.8.1.2 Non-fetal tissue accretion

There is limited information regarding the rate of non-fetal tissue accretion. In the German system, the non-fetal tissue accretion is estimated based on the weight of fetus, and the ratio of fetus: adnexa at different points of gestation which is then used to estimate the growth curve of the entire conceptus (Coenen *et al.* 2011). The French system estimated from bibliographic data, that the weight changes of fetal membranes and maternal tissues are equivalent to 20 and 10% of fetal tissue gain respectively, and that the chemical composition of fetal membranes and maternal tissues are considered to be similar to that of the fetus at the same stage of gestation (INRA 2015). The NRC system assumes the rate of non-fetal tissue accretion is linear and that non-fetal accretion is 0.09 g/kg maternal BW/day based on limited measurements of uterine and placenta tissue during second trimester by Ginther (1992).

2.8.1.3 Oxygen consumption of conceptus

In the German system, the oxygen consumption value in non-fetal tissue is measured and standardised to 1 kg fetal bodyweight to represent the entire conceptus (Coenen *et al.* 2011). The value is then converted to the amount of heat production in order to estimate the metabolic requirement of the conceptus. Within the NRC system (NRC 2007), an adapted oxygen consumption value of 0.276 MJ /kg BW (66 kcal/kg BW) for accumulated fetal and non-fetal tissue is used, based on measurements in horses by Fowden *et al.* (2000) and in cattle by Reynolds *et al.* (1986).

2.8.1.4 Chemical composition of conceptus

Coenen *et al.* (2011) stated that the German system developed a model estimating chemical composition (energy, protein) of the fetus depending on day of gestation using data from Meyer and Ahlswede (1978). The French system also draws on values from the same dataset and estimates changes in chemical composition (energy, protein) on a monthly basis (INRA 2015). Whereas, the NRC system assumes each unit of gain to be 20% protein and 3% lipid (NRC 2007).

2.8.1.5 Pregnancy requirements

Pregnancy requirements in the German system are calculated from the energy contained per kg fetus plus the daily heat production of the entire conceptus based on its oxygen consumption (Coenen *et al.* 2011). This method of estimation provides caloric needs for growth of conceptus in ME without assumptions based on the efficiency of DE use because oxygen consumption is the direct parameter for energy metabolism (Coenen *et al.* 2011).

As described earlier, the French system assumes the fetal membranes and maternal tissue depositions are equivalent to 20% and 10% of fetal tissue gain, respectively. Therefore, gestation requirements are calculated from fetal weight gain and changes in chemical composition multiplied by 1.3 (INRA 2015). It is then multiplied by the total energy stored by the conceptus divided by the efficiency of energy utilization (25%). The efficiency of utilization is the mean between cows and sows due to absence of the measurement in mares (Martin-Rosset *et al.* 1994).

The pregnancy requirements in the NRC system are the sum of maintenance DE costs for conceptus tissue and the energy required for tissue deposition (NRC 2007). The energy cost of tissue deposition is the energy required to deposit per unit tissue gain containing 20% protein and 3% lipid with assumption that efficiency of DE use for tissue deposition during pregnancy is 60% (NRC 2007).

The energy requirements estimated by the three systems for a 500 kg mare in her 11th month of gestation are presented in Table 4. The DE value obtained from NRC is converted to ME assuming the renal and gaseous losses is approximately 16% (Kienzle and Zeyner 2010). The NE value obtained from the French system is converted to ME based on k_m value used in the system (78.5%) (INRA 2015). The energy requirements estimated by the German and NRC are in agreement, at 75.9 MJ ME/day and 75.6 MJ ME/day, respectively. Whereas, the energy requirement estimated by the French system is 65.9 MJ ME/day which is lower compared to the other two systems. This lower estimation may be due to, the system not including maintenance energy of conceptus, or the efficiency value for maintenance is not representative of the efficiency for pregnancy (k_p). These speculations cannot be confirmed as there is no available information on k_p in horses. The use of oxygen consumption in the model also requires further critical evaluation. Overall, all three systems here have drawn information from limited sources due to scarcity of data hence some similar data were used across the different systems. However, each system conducted modelling using different approaches and assumptions, and obtained similar modelling outcomes.

Table 4. Daily requirement of a 500 kg mare at her 11th month of pregnancy and 1st month of lactation estimated based on the German, French (INRA) and NRC systems.

Systems	**Pregnancy requirement**		**Lactation Requirement**	
	Default values	**MJ ME/day**	**Default values**	**MJ ME/day**
[1]German	75.9 MJ ME/day	75.9	103.98 MJ ME/day	103.98
[2]French (INRA)	5.5 UFC/day	65.9	8.5 UFC/day	102.01
[3]NRC	89.5 MJ DE/day	75.6	132.63 MJ DE/day	106.11

1UFC= 2250 kcal= 9.42 MJ; $k_m = k_p$ and k_l = 78.5%
Efficiency of DE use for ME = 84% (urinary and methane losses in horses is 16%)
[1]Coenen *et al.* (2011), [2]INRA (2015), [3]NRC (2007)

2.8.2 Lactation

2.8.2.1 Milk production

Data for milk yield is questionable due to the lack of reliable methods for measurements under different experimental conditions such as frequency of suckling, milking conditions (i.e. machine, hand), and methods used to obtain foal intake (Doreau and Martuzzi 2006). Peak lactation is reported to be between 1 - 3 months according to Wood's model (Figure 6) which was originally developed to predict milk yield in dairy cows (Doreau and Martuzzi 2006). However, variation in daily milk yield of mares prevents defining a model that has a high power of predictability for milk yield (Figure 7) (Coenen *et al.* 2011). However, peak milk yield is unlikely to reach peak in the third month of lactation as the daily gain of foal has decreased drastically during the 1st 6 weeks of life (Coenen *et al.* 2011). Other studies reported peak milk yield to be at the 1st or 2nd month of lactation which is closer to the pattern of intake and requirements of the foal (Chavatte 1997). Heavy breed mares produce more milk daily compared to light breed mares (Figure 8) but there were no difference in daily milk yield when expressed in terms of per 100 kg BW (2.5 - 3 kg/100 kg BW) (Doreau *et al.* 1986; Doreau and Boulot 1989; Doreau *et al.* 1990; Doreau *et al.* 1991; Doreau *et al.* 1993).

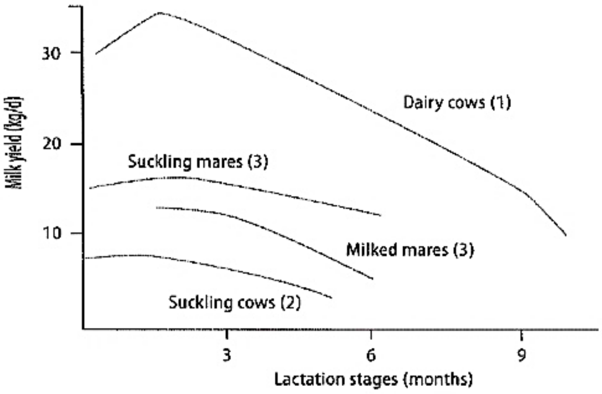


Figure 6. Lactation curves for nursing or milked mares and milked or nursed cows calculated using Wood's model (Doreau and Martuzzi 2006). Figure from INRA (2015).

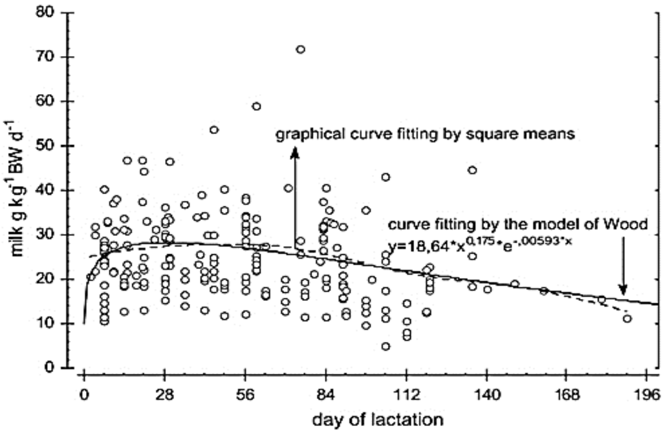


Figure 7. Daily milk yield of mares statistically transformed to gram per kg body weight (x = day of lactation) (Coenen *et al.* 2011).

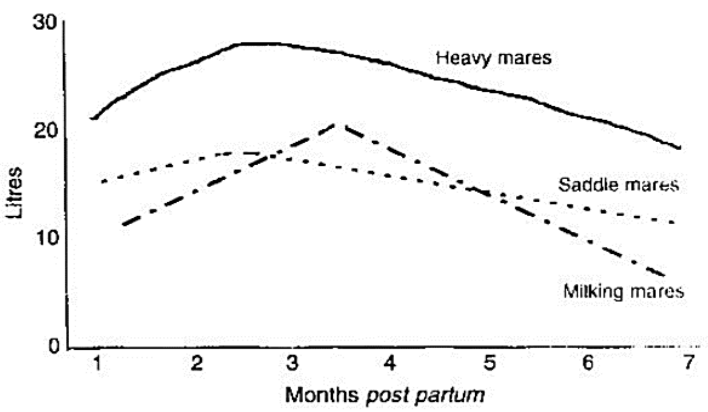


Figure 8. Mean daily milk production in mares according to stage of lactation and breed. Figure from Chavatte (1997).

2.8.2.2 Gross energy of milk

There are variations in gross energy of milk. Gross energy between week 1 to week 8 of lactation was 2.54 MJ/kg - 2.06 MJ/kg (608±57 kcal/kg - 493±41 kcal/kg) milk in French breed mares (Doreau *et al.* 1990) and 2.43 MJ/kg - 1.70 MJ/kg (582 kcal/kg - 407 kcal/kg) milk from week 1 - week 25 in Hafflinger nursing mares (Mariani *et al.* 2001). Non-breed specific mean gross energy of mares' milk were reported to be 1.93 MJ/kg - 2.29 MJ/kg (460 kcal/kg - 549 kcal/kg) (Doreau *et al.* 1988; Chavatte 1997; Malacarne *et al.* 2002). Similar trends can be observed in Figure 9. Other than variation between lactation stages, there are considerable variation in gross energy at the same point of lactation as well (Figure 9). Overall, the variations due to breed and lactation stage are between 1.67 - 2.51 MJ/kg (400 - 600 kcal/kg) milk.

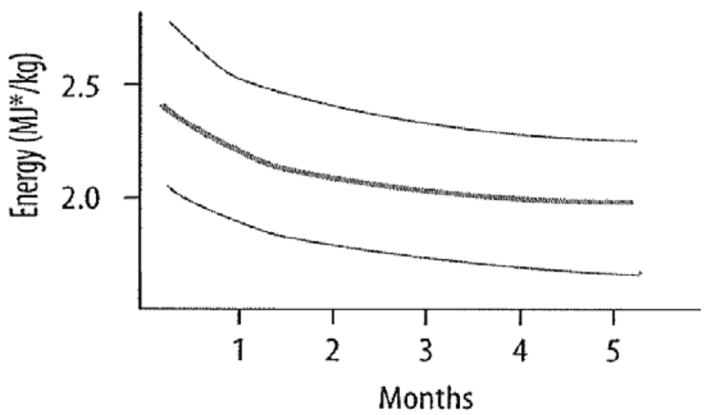


Figure 9. Variation in composition of mare's milk gross energy with stage of lactation. Figure from INRA (2015).

2.8.2.3 Lactation Requirements

The German system predicts daily milk yield using the Wood's model (Coenen *et al.* 2011). However, the predicted peak milk yield does not follow the intake and the requirements of the foal. To create a more biological model, a bodyweight exponent ($BW^{0.82}$) is introduced. This exponent was obtained from an interspecies study on the relationship between milk energy intake by suckling and body weight at peak milk yield that included results from ponies (Coenen *et al.* 2011). Coenen *et al.* (2011) also stated that the analysis of energy requirements in growing foals resulted in the same exponent (unpublished, Kienzle). The energy requirement for lactation is calculated by multiplying the gross energy content of milk which is standardized at 2.14 MJ (511 kcal). The efficiency of energy use for milk production is 60%.

The French system predicts milk yield and gross energy content of milk on a monthly basis up to 6 months of lactation drawing data from published work (Table 5) (INRA 2015). The milk production from the 1st month to 6th month of lactation is 3 kg/100 kg BW and 2 kg/100 kg BW respectively. Whereas the gross energy is 2.28 MJ/kg (545 kcal/kg) milk and 1.80 MJ/kg (431 kcal/kg) milk which are comparable with those reported in Doreau *et al.* (1988), Chavatte (1997), and Malacarne *et al.* (2002). The efficiency of energy use for milk production is 65%.

The NRC (2007) summarised milk production data from available studies and constructed an equation to estimate milk yield by day. Total milk produced per month is then calculated to obtain a daily average. The gross energy in milk is standardized at 2.09 MJ/kg (500 kcal/kg) milk and the efficiency of energy use for milk production is 60%.

The lactation requirements estimated by the three systems are consistent with each other and is presented in Table 4 (page 27). To compare the values, requirements were converted to MJ ME/ day. Digestible energy obtained from (NRC 2007) was converted to ME assuming 16% renal and methane energy loss (Kienzle and Zeyner 2010). The NE value from French system was converted to ME based on the k_m value used in the system (75.8%) (INRA 2015). The lactation requirements were calculated for a 500 kg mare in her 1st month of lactation using the French and the NRC system while requirements were calculated for a mare in 2nd week of lactation using the German system. The requirements estimated by the German system, French system and NRC are 103.98 MJ ME/ day, 102.01 MJ ME/day and 106.11 MJ ME/day respectively (Table 4).

Table 5. Reference value for milk production and composition for estimating lactation requirements. Figure from INRA (2015).

Month	Production[1] (kg/j/100 kg BW)	Milk composition[1]		Minerals[2]			
		Energy (kcal/kg)	Protein (g/kg)	Ca (g/kg)	P (g/kg)	Mg (g/kg)	Na (g/kg)
1st	3.0	545	24	1.20	0.80	0.10	0.16
2nd	3.3	512	21	0.90	0.60	0.07	0.14
3rd	3.2	468	20	0.90	0.60	0.07	0.14
4th	2.9	455	16	0.70	0.50	0.05	0.11
5th	2.2	431	12	0.70	0.50	0.05	0.11
6th	2.0	431	12	0.70	0.50	0.05	0.11

[1] Doreau *et al.* (1990, 1992, 1993) and a literature survey by Doreau and Martin-Rosset (2002).
[2] Literature reviews by Doreau *et al.* (1990), Schryver *et al.* (1986), and Smolders (1990).

2.9 Nutritional regulation of reproduction

2.9.1 Body condition score and energy intake

Body condition score and energy intake have been shown to affect reproductive performance and ovarian activity. At conception, during pregnancy and post-partum, an overall decrease in body condition score that resulted from a reduction in dietary energy intake was found to increase the inter-ovulatory interval (Henneke *et al.* 1984), increase the parturition to ovulation interval (Godoi *et al.* 2002), increase the number of cycles per conception and reduce the pregnancy rate (Henneke *et al.* 1984). A study in Lusitano mares showed mares that were not in energy deficit at conception and lactation had improved fertility in the first two oestrus cycles compared to mares that were in energy deficit at conception and lactation (Fradinho *et al.* 2014).

2.9.2 Leptin

Reproductive and ovarian activity are also related to leptin levels because leptin acts on the neuroendocrine system to modulate the reproductive activity by reflecting the adequacy of nutrition and metabolic reserve (Barash *et al.* 1996; Gentry *et al.* 2002; Ferreira-Dias *et al.* 2005; Morley and Murray 2014). Leptin is a hormone secreted from adipocytes and its plasma concentration is related to body fat stores (Buff *et al.* 2002; Buff *et al.* 2005). An increase in leptin levels and high body condition score (>5) were associated with ovarian activity and cyclicity during the anovulatory period whereas low body condition (<5) and low leptin concentrations were associated with profound seasonal anoestrus and longer duration of transition to ovulation (Gentry *et al.* 2002).

Ferreira-Dias *et al.* (2005) reported changes in leptin concentrations that influenced reproductive and ovarian activity but without changes in body weight and body condition indicating other factors may affect leptin secretion. It was reported that leptin expression and secretion are regulated by glucose transport and metabolism, and is stimulated by insulin in cultured rat adipocytes (Mueller *et al.* 1998). This has been observed in horses, where mares with constantly elevated insulin had elevated leptin compared to mares with lower insulin (Storer *et al.* 2007). Indeed, a study in ponies found plasma leptin significantly correlated with plasma concentration of glucose and insulin (Salazar-Ortiz *et al.* 2011). Therefore, it appears that energy status may not only modify body condition score, it may also modify leptin without initiating changes in body condition score through pathways such as influencing glucose and insulin levels. Leptin was also found to affect the *in vitro* equine oocyte maturation and fertilization rate which may have implication on the mares fertility (Consiglio *et al.* 2009).

2.9.3 Plasma insulin growth factor (IGF-1)

Abrupt energy and protein restriction creates prompt reduction in plasma IGF-1 within 24 – 48 hours and altered plasma growth hormone (Sticker *et al.* 1995). Changes in plasma IGF-1 were reported to affect ovarian function in the mares (Salazar-Ortiz *et al.* 2014). Two months energy intake at 60% of energy requirements modified the systemic concentration of IGF-1 and bioavailability of IGF-1 in ovaries through increase in IGFBP-3 concentrations (Salazar-Ortiz *et al.* 2014). Effects of poor nutrition on recruitment and selection of follicles were also observed (Salazar-Ortiz *et al.* 2014). The effect of long term energy deficit on plasma IGF-1 changes and ovarian function has been demonstrated, but the effect of acute energy deficit on ovarian activity remains to be elucidated. However, the potential effect of acute energy deficit should not be overlooked.

Similarly, studies in dairy cows suggest a positive relationship between energy balance, plasma IGF-1, function of dominant follicles, and shorter post-partum to ovulation interval (Beam and Butler 1999). Specifically during early post-partum period, a more positive or an improvement in energy balance before day 25 post-partum enhanced recruitment of follicles (Lucy *et al.* 1991). In addition, the number of days to the lowest negative energy balance post-partum were found to be highly correlated with days to first ovulation in dairy cattle due to suppression of pulsatile luteinising hormone secretion (Canfield and Butler 1990). Ovulation is not stimulated until luteinising hormone pulse frequency increases with recovery of energy balance in cattle (Canfield and Butler 1990). Energy balance in mares has not been studied, however findings from cattle indicate possible effects of the pattern in daily energy balance on post-partum ovarian activity (Lucy *et al.* 1991), luteinising hormone secretion and post-partum to ovulation interval (Canfield and Butler 1990).

In summary, the pre and post-partum energy status and potentially the daily energy balance pattern plays an important role in determining the reproductive performance in the mare. However, it is not fully understood how body condition, leptin and other blood metabolites would response to changes in energy status of different magnitude and duration due to the scarcity of empirical data for energy status. Research on the full mechanism behind neuro-endocrine regulation of the reproductive system by leptin in horses is scarce. Though it has been recently proposed that nutrition and photoperiod have their respective roles in reproduction in mares with photoperiod determining the annual rhythm of ovarian activity whereas nutrition, primarily the energy level available, may determine the length of the phase of ovulatory activity (Salazar-Ortiz *et al.* 2011). This further supports the concept of energy status as a major mediator to reproductive function besides photoperiod and it was proposed that reproductive function is regulated by energy levels through the leptin-GH-IGF-1 system (Salazar-Ortiz *et al.* 2011).

2.10 Reproductive performance of Thoroughbred mares in New Zealand

At present, there are only three reports regarding reproductive performance of Thoroughbred mares in New Zealand by Van Rijssen *et al.* (2010) and Hanlon *et al* (2012a,b). It is difficult to compare the reproductive performance between countries and studies as there are no standard measures of reproductive performance in the equine industry and there is lack of consistency in use of outcome measures amongst investigators (Hanlon 2012).

Foaling to conception interval of Thoroughbred broodmare on one commercial Thoroughbred stud farm in New Zealand was reported to be 32±6 days (Van Rijssen *et al.* 2010). October and November breeding mares had shorter foaling to conception interval compared to December breeding mares (30±15 days vs 37±20 days) (Van Rijssen *et al.* 2010). The reproductive performance measures of five New Zealand commercial

Thoroughbred stud farm reported by Hanlon *et al* (2012a,b) were first cycle pregnancy rate, end of season pregnancy rate, foaling rate, pregnancy loss (day 14-16 and day 42 of gestation), and mating to conception interval (Table 6 and 7) (Hanlon *et al* 2012a,b). After controlling for effects of stallion, stud farm, and year of study, the variables that were most important to reproductive performance were mare-related, especially mare age and reproductive status (dry or foaling) (Hanlon *et al.* 2012a). Increase in mare age significantly reduced reproductive performance independent of reproductive status and foaling mares had significantly poorer reproductive performance than dry mares when controlled for age (Hanlon *et al.* 2012a).

Table 6. First cycle pregnancy rate (FCPR), end-of-season pregnancy rate (SPR) and foaling rate of five Thoroughbred stud farms in the Waikato region of New Zealand over three consecutive breeding season (2006-2008) from Hanlon *et al.* (2012b).

Variable	**ID**	**FCPR**	**SPR**	**Foaling Rate**
Stud	1	58.6	86.1	78.1
	2	54.7	92.6	82.3
	3	68.2	90.9	90.1
	4	60.9	90.1	81.2
	5	47.3	81.4	77.8
Mean		53.6	85.3	80.2

Table 7. Pregnancy loss rates and multiple pregnancy rate by reproductive status for Thoroughbred mares from the Waikato region of New Zealand over three consecutive breeding season (2006-2008) from Hanlon *et al.* (2012b).

	Maiden			Barren			Foaling		
	%	No.	95% CI	%	No.	95% CI	%	No.	95% CI
Pregnancy losses									
14 - 42 d	3.7^a	10/260	1.9-7.0	7.9^b	39/494	5.7-10.6	4.8^a	49/1021	3.6-6.3
42 d - term	2.8	7/253	1.1-5.6	3.0	14/468	1.6-5.0	3.1	30/983	2.1-4.3
Multiple preg. at 15d	15.8^a	41/260	11.6-20.8	19.0^a	94/494	15.7-22.8	9.2^b	94/1021	7.5-11.1

^{a,b} Values within the same row with different superscripts are significantly different ($P < 0.05$)

There is also significant effect of foaling date to end of season pregnancy rate and start of mating to conception interval (Hanlon *et al.* 2012a). After controlling for the effect of serving stallion, stud farm, mare age and foal heat service, the daily probability of conception for mares foaling after 22nd October was 0.76 times less than mares foaling before 11th September (Hanlon *et al.* 2012a). With each additional day that a mare foaled later in the season, the end of season pregnancy rate reduced by a factor of 0.97 (Hanlon *et al.* 2012a). In other words, mares that foaled later in the season will have a longer start of mating to conception interval and are less likely to achieve pregnancy by the end of the season compared to the mares that foaled early in the season. The cause behind the effect of foaling date on mating to conception interval was unidentified.

The start of mating to conception interval in this study was defined as number of days from 1st September to the last service date of mares diagnosed pregnant at 42 days of gestation hence does not represent ovulation to conception interval (Hanlon *et al.* 2012a). Therefore, the reason for this longer mating to conception interval may be the longer parturition to ovulation interval or more oestrous cycles required for conception. Reduction in energy intake and body condition during pregnancy, post-partum and at conception were reported to increase parturition to ovulation interval and number of oestrus cycle required per conception (Henneke *et al.* 1984; Godoi *et al.* 2002). Energy deficit at conception and lactation negatively affected the fertility of mares during the first two oestrous cycle compared to mares that were not in energy deficit (Fradinho *et al.* 2014). This evidence warrants investigation on the energy intake and energy balance of mares that foaled on different foaling dates because foaling date may affect the balance between energy supply and demand under a pastoral system hence creating a variation in energy status between early and late foaling mares. More information on energy status and reproductive performance of New Zealand Thoroughbred mares will help validate if energy status can provide further explanation to the variation in reproductive performance not related to mare-level factors.

2.11 Concepts of modelling

To conduct a study on energy balance that represents the whole New Zealand Thoroughbred population would require multiple long term studies in a variety of areas, with large sample sizes incurring huge expense in time and costs. Therefore, a more effective approach is to conduct nutritional modelling with readily available data and knowledge. It can serve as a great tool to predict the observations before conducting field studies to pin point areas that require research thereby providing guidance in research design and methodology in the long term.

2.11.1 Definition of modelling

Modelling is the process of developing a set of equations that represents the behaviour of a system (Wastney 1999). Biological models can belong to, and consist of sub-models that belong to different levels of organizational hierarchy in biology, depending on the purpose of the model and availability of data on the system. In animal science, the typical scheme proposed by (Thornley and France 2007) is a presented in Table 8. A model with animal level integration may also contain sub-models at lower integration levels (i-1, i-2...), and the output of the model would be the result of interaction between the sub-models.

Table 8. The organisational hierarchy of biology as described by Thornley and France (2007).

Level	**Description of level**	
i +1	Group of animals	Estimating feed requirement for a group of animals
i	Animal	Estimating energy requirement of an animal
i-1	Organs	Estimating pre-caecal absorption of nutrients
i-2	Tissue	
…	Cells, organelles …	

2.11.2 Types of model

All models and sub-models can also be categorized based on mathematical forms (Wastney 1999; Thornley and France 2007). Models can be:

Deterministic vs stochastic: Deterministic models make predictions for quantities without associating probability of distribution whereas stochastic models include element of randomness and are based on probability.

Dynamic vs static: Dynamic models predict changes that occur with time whereas static models make predictions for quantities that only represent one point in time.

Empirical vs Mechanistic: Empirical models aim to describe the responses of a system using mathematical equations generated from analysis of experimental data. Mechanistic models aim to provide explanation of a phenomena being modelled or understanding underlying biology.

2.11.3 Steps in building a model

The aim of modelling is to create a mathematical "likeness" of a system to ensure the model behaves in a same way as the system (Wastney 1999). To ensure modelling fulfils this aim, the components, the interaction of components and relevant variables in the system needs to be defined. Then, purpose of the model needs to be clarified as it dictates the output measured, the form of the model and the data required to develop it (Wastney 1999). Then, action can be taken to locate and examine existing models and information about the system. A graphic representation of steps in building a model is presented in Figure 10 using the energy balance system for pregnant and lactating Thoroughbred mares managed on pasture as an example.

2.11.3.1 Purpose of modelling

Models can be developed to serve as a research tool or as a field-applied model (Tylutki 2011). Regardless of its primary objective, models are also developed with specific purposes such as: determining the "structure" of a system, to calculate parameters of interest, integrate information on a system, predict response to a perturbation, derive mechanistic principle underlying the behaviour of a system, and identify differences under different conditions (Wastney 1999).

All models contains variables and they can be categorized as state, rate and driving variables (Thornley and France 2007). State variables define the state of the system being modelled at a given time. Rate variables define the occurrence rate of a process (Thornley and France 2007). Driving variables are natural or imposed external inputs that affects the dynamic of the organism (Thornley and France 2007). Driving variables can become state variables depending on the model operation (Thornley and France 2007). For example, the state variables in an energy balance model for mares under a pasture system would be mare bodyweight and foaling date. Whereas, pasture energy and DMI can be considered as both state and driving variables i.e. pasture energy can be controlled by model operation (entered as model input) but the variable itself changes with time within the model.

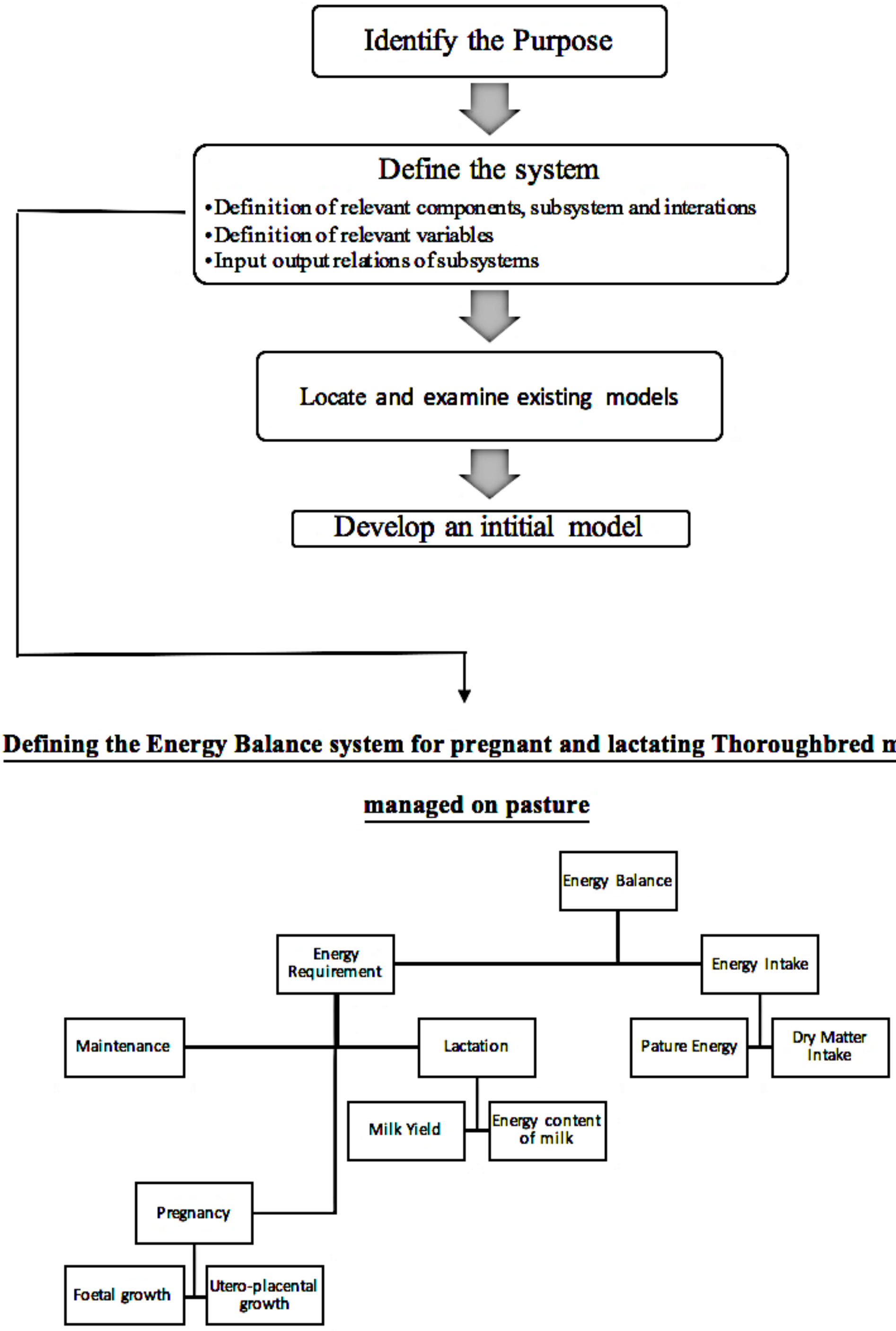


Figure 10. Steps in building a biological model using the Energy Balance system for pregnant and lactating Thoroughbred mares as an example. Information adapted from Wastney (1999), Gold (1977).

The purpose of developing an energy balance model for Thoroughbred mares in this study would be to predict field observations, and serve as guide for further research and research designs with potential to eventually become a field-applied model as its robustness increase with available data. The purpose of the energy balance model would be to (1) make quantitative predictions on the energy balance of the mare and its changes that occur with time throughout the modelled period (late pregnancy and first 5 months of lactation) by integrating parameters that simulate scenarios of a typical Thoroughbred broodmare managed on pasture in New Zealand, (2) to determine the response/ behaviour of the model and sub-models to changes in variables using modelled data.

2.11.3.2 Defining the energy balance system for pregnant and lactating Thoroughbred mares managed on pasture (Figure 10)

After the purpose of the model is defined, the steps to defining the system becomes clear. The energy balance is a reduction of energy requirement from energy intake which can be ranked as the sub-models. Energy requirements can be modelled with empirical data available for maintenance requirement, energy required during pregnancy, and energy required for lactation. Subsequently, energy requirement during pregnancy and lactation can be modelled using dynamic empirical models available for feto-placental development, milk yield and energy content of milk. Energy intake can be modelled with the unanimously accepted mathematical equation for energy intake which is a multiplication of DMI in kilograms and energy content of feed per kg DM. Therefore, the model will include a mixture of static, dynamic, empirical and mechanistic models. The model does not account for probability of distribution, and the output of the model changes with time hence it is a deterministic model that is also dynamic and mechanistic.

2.11.4 Testing for robustness

It is necessary to test the robustness of the model to assess the field applicability of the data and accuracy of the predictions. Robustness of a model can be assessed in several ways through analysis of sensitivity, identifiability, and stability (Thornley and France 2007). The common procedure in a sensitivity analysis is to modify the inputs (variables) to compute a change in output in order to rank relative influence of an input (variables) to an outcome (Thornley and France 2007). Identifiability refers to whether the features of the model also exist and can be obtained from the system or proposed experiment conditions (Thornley and France 2007). Lack of identifiability will make validating the model or further hypothesis testing impossible. Stability refers to the response of a system to perturbation (Thornley and France 2007). Stability of a model can be tested only if its steady state is defined.

The energy balance is generated by the model through integrating multiple inputs simultaneously. As a result, the impact of inaccurate assumptions and the order of importance of a parameter to the outcome is unknown. Therefore, the most suitable procedure to test for robustness of this model would be the sensitivity analysis. Knowing the influence of each variables, the area within the commercial pasture system that requires attention can be identified, the conditions where the model can be applied can also be determined (i.e. the range of equine bodyweight that this model is applicable to). A sensitivity analysis at the sub-model level can describe their relationship with the variables and reveal the interaction between different parts of the model.

2.12 Conclusion

The New Zealand Thoroughbred breeding industry operates predominantly with mares grazing on pasture with highly seasonal growth, availability, and energy content. Energy requirement of the mare changes with physiological stage with increases during late pregnancy and lactation. There is an overlap of high energy requirements and low pasture energy availability which may influence the energy intake of mare and the efficiency of pasture energy utilisation. The degree of overlap between these periods is dependent on foaling date. The commercial stud farm conditions and the seasonal pasture availability may potentially facilitate a limitation on DMI and influence mare energy intake. All of the above will interact with each other and influence the nutritional status and daily energy balance changes. Evidence reported within literature suggests pre and post-partum energy status and potentially the daily energy balance pattern would influence the reproductive performance in the mares.

However, there are no published information regarding energy intake, energy balance, bodyweight changes, and body condition score changes of New Zealand Thoroughbred mares managed commercially on pasture. Therefore, the nutritional status of mares is not known under these conditions. This lack of empirical data is also hindering the progression to fully understand how body condition, leptin and other blood metabolites would response to changes in energy status of different magnitude or duration, and subsequently, the respective influence of nutrition on regulating the reproductive function in the mare compared to seasonal and climatic factors under pastoral conditions.

Within the industry, there is little use of feed budgeting, monitoring of pasture cover and pasture intake, and monitoring of bodyweight and body condition score changes in mares. Therefore, there is also difficulty in knowing if mares' energy requirements are met from pasture. This is also one of the reasons for using supplementary feed by stud managers (Hirst 2011). There is also a report that a solely pasture diet failed to produce desired improvements in body condition (Hirst 2011). Hence there is a need to develop tools that would assist in nutritional management of mares, and also improve pasture utilization. This can be achieved by developing a robust model to predict energy requirement, energy intake and energy balance of the mares at different DMI levels and pasture energy availability.

In order to address the need from industry and the gaps in published literature, the objectives are to (1) develop a precise nutritional model to estimate daily energy intake, energy requirements and energy balance of mares managed commercially on pasture, (2) to quantify and characterize the pattern of energy balance changes during late pregnancy and lactation using the model data, (3) to test the robustness and application of the model by identifying the response and sensitivity of energy balance towards changes in mare and pasture variables such as mare body weight, foaling date, dry matter intake, and pasture energy.

Chapter 3: Deterministic modelling of energy supply and demand for foaling mares managed at pasture on commercial Thoroughbred stud farms.

3.1 Methods

3.1.1 The model

A deterministic model was developed to assess energy intake, energy requirements and energy balance of mares managed commercially on pasture during the last 5 months of pregnancy and first 150 days of lactation. The variables included in this model were dry matter intake (DMI) (kg DM/day), pasture metabolisable energy (MJ ME/kg DM), foaling date (days from 1st of September) and mare body weight (kg). Energy balance was calculated by deducting energy intake from published energy requirements (NRC 2007). All calculations are as described in Table 9.

Briefly, daily energy requirements were determined using a factorial approach including maintenance, pregnancy and lactation requirements, and extra requirements needed under pasture based husbandry conditions (i.e. foraging, group housing, heat and cold, and walking). Energy required for pregnancy was determined based on fetal-utero-placental growth, whereas, energy required for lactation was based on milk yield.

To estimate pregnancy requirements, growth of fetus and non-fetal tissue were modelled using equation 1 - 4 in Table 9. Fetal weight was calculated based on % birthweight relative to days of gestation and birthweight. Percentage birthweight was modelled using equation 2 with birthweight estimated to be 9.7% of dam weight which is close to birthweight of Thoroughbred foals from other studies (10%) (Platt 1978; Elliott *et al.* 2009). Digestible energy required for pregnancy ($\mathrm{DE}_{\mathrm{preg}^{\mathrm{T}}}$) is the sum of (1) energy required for maintenance($\mathrm{DE}_{\mathrm{maint}}$), (2) maintenance requirement of fetal-utero-placental tissue ($\mathrm{DE}_{\mathrm{preg}^{\mathrm{m}}}$), and (3) energy required for utero-placental tissue deposition and fetal growth ($\mathrm{DE}_{\mathrm{preg}^{\mathrm{G}}}$) (equation 6-9).

For lactation requirements, daily milk yield is estimated using equation 5. Digestible energy required for lactation ($\mathrm{DE}_{\mathrm{lact}}$) was calculated using equation 10. Equation 5 estimates milk yield by day and the pattern approximates the lactation curve of nursing mares (Chavatte 1997; Doreau and Martuzzi 2006) and the trend of foal intake (Oftedal *et al.* 1983; Doreau *et al.* 1990; Chavatte 1997). The values for gross energy in milk reported varied between 400 - 600 kcal/kg milk solid (Doreau *et al.* 1988; Doreau *et al.* 1990; Chavatte 1997; Mariani *et al.* 2001; Malacarne *et al.* 2002). Therefore, an average value of 0.279 MJ/kg (500 kcal/kg) milk was used for gross energy of milk (NRC 2007). Efficiency of DE for milk production is 60% and maintenance DE requirement ($DE_{maint(lact)}$) for lactating mare is 36.3 kcal/kg BW (0.154 MJ/kg BW) (NRC 2007).

Extra costs for eating was 10% above maintenance requirement in stabled horses fed a forage diet (Vermorel *et al.* 1997). Walking (2km/day, flatland) increases the energy requirement by 10% in dairy cattle (Nicol and Brookes 2007). Group housing may increase energy requirements by 10% (Coenen *et al.* 2011). Heat and cold will increase requirements by 10 - 20% (Coenen *et al.* 2011). Therefore, keeping a mare outdoor on pasture in groups and under intensive management may increase energy expenditure up to 40% - 50%. Cymbaluk and Christison (1990) estimated energy costs for mares grazing on pasture in a cold climate without thermal stress is 35% above maintenance requirement. However, the maintenance requirement of Thoroughbred horses were reported to be 20% and 30% higher than warmblood and Quarter horses respectively (Coenen *et al.* 2011). To estimate energy requirements specific to Thoroughbred mares, the value used in the main analysis to account for extra energy expenditure due to husbandry, foraging and housing is 45%.

Energy intake is the product of pasture intake and pasture energy (equation 11). Daily values for pasture metabolisable energy (ME) in New Zealand derived for ruminants were included in this model (Figure 11) (Litherland and Lambert 2007). Dry matter intake of horses of different breeds, in different physiological state and on different diets were summarized in Table 10 (Marlow *et al.* 1983; Grace *et al.* 2002; Guay *et al.* 2002; NRC 2007; Collas *et al.* 2014; Collas *et al.* 2015). Dry matter intake of pregnant mares was estimated to be 2% BWT whereas DMI of lactating mare was 2.5% BWT.

Energy balance was obtained by deducting energy intake from energy requirements (equation 12). However, energy intake is available in the form of ME (MJ ME/day) hence energy requirement was converted from DE (MJ DE/day) into ME (MJ ME/day) prior to calculating energy balance. The ME is energy available after accounting for energy loss through urine and methane (NRC 2007). The urinary and methane energy losses in horses of different breeds fed different diets were summarised in Table 11. The assumption for urine and methane loss in the model is 16%.

Table 9. Equations used to model pregnancy tissue accretion, milk yield, digestible energy (DE) requirements, energy intake and energy balance for Thoroughbred mares during pregnancy and lactation.

[1]Fetal Weight (kg)	% Birthweight*birthweight (kg)	[eq:1]
[1]% Birthweight	$1*10^{-7}X^{3.5512}$ (R^2= 0.929) X= days of gestation	[eq:2]
[1]Birthweight (kg)	0.097*maternal bodyweight (kg) 97% maternal bodyweight	[eq:3]
[1]Placental tissue | Uterine Development (g/day)	0.09 g/kg BW/d	[eq:4]
[1]Milk yield (kg/day)	$Y= a *d^{0.0953}*e^{-0.043d}$ Y= kg milk/day a= 0.0274287*mature weight (kg) d= day of lactation	[eq:5]
Total energy requirement for pregnancy (DE_{preg^T}) (MJ/day)	$DE_{maint} + DE_{preg^m} + DE_{preg^G}$	[eq:6]
[1]Maintenance requirement of pregnant mare (DE_{maint}) (MJ/day)	33.3 kcal/kg BW= 0.139 MJ/kg BW	[eq:7]
[1]Maintenance requirement for accrued feto- placental tissue (DE_{preg^m}) (MJ/day)	66.6 kcal/kg tissue = 0.279 MJ/kg tissue	[eq:8]
[1]Energy required for feto- placental tissue gain (DE_{preg^G}) (MJ/day)	[($FP_{(lipid)}$*ADG (kg)*$GE_{(lipid)}$ (Mcal)) + [($FP_{(protein)}$ tissue*ADG (kg)*0.0056 Mcal)]/0.6 FP: 1 unit feto-placental tissue= 20% protein, 3% lipid (0.2 and 0.03) GE content (protein)= 5.6 kcal/g = 0.0234 MJ/g GE content (lipid)= 9.4 kcal/g = 0.0393 MJ/g Efficiency of DE for tissue deposition= 0.6 (60%)	[eq:9]
[1]Total Energy Required for lactating mare (DE_{lact}) (MJ/day)	DE_{maint} + [(*milk yield** $GE_{(milk)}$)/0.6] Gross energy(milk): 500 kcal/kg milk= 0.279 MJ/kg milk Efficiency of DE for milk production: 0.6 (60%)	[eq:10]

[1]Maintenance requirement for lactating mare (DE_{maint}) (MJ/day)	**36.3 kcal/kg BW= 0.152 MJ/kg BW**	
Energy Intake (MJ ME/day)	Pasture ME (MJ ME/kgDM)* dry matter intake (kg DM/day)	[eq:11]
	ME: Metabolizable energy Dry matter intake: Pregnant mare (2% BWT), Lactating mare (2.5% BWT)	
Energy Balance (MJ ME/day)	Energy Intake (MJ ME/day) - Energy Requirement (MJ ME/day)	[eq:12]

[1] NRC (2007)
1 kcal = 0.00418 MJ
eq = equation

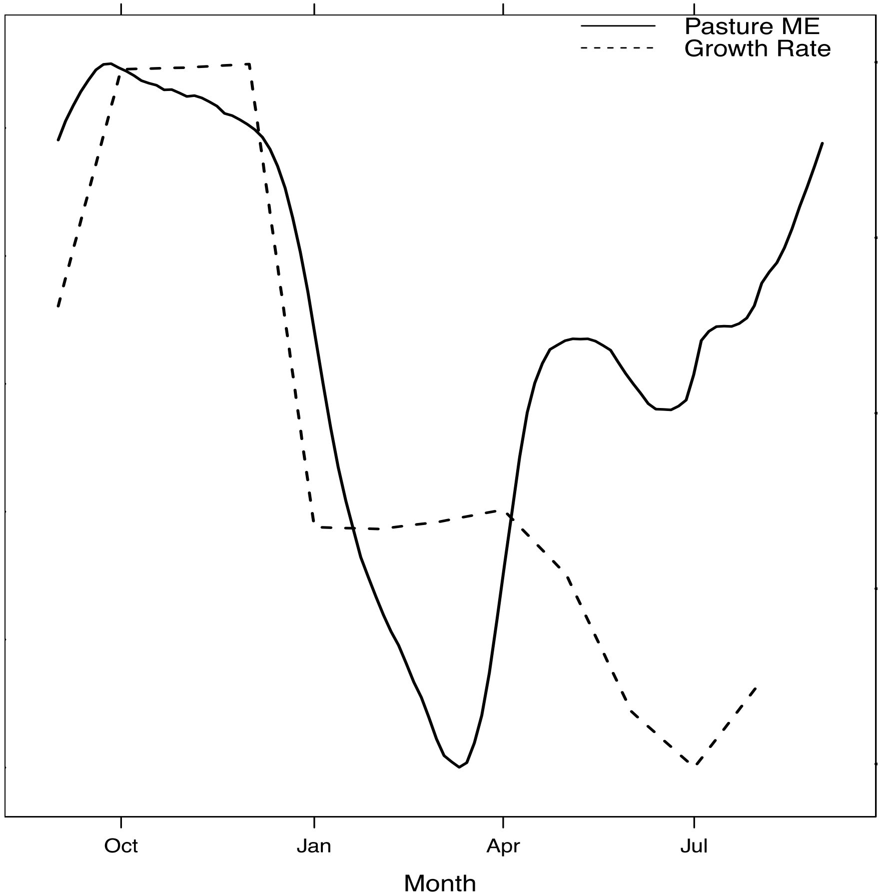


Figure 11. Pasture metabolisable energy (ME) of typical New Zealand pasture used in the energy balance model adapted from (Litherland and Lambert 2007), and growth rate of pasture collected from Dairy 1, Massey University, Palmerston North (Correa 2015 - 2017, unpublished).

Table 10. Estimated voluntary dry matter intake (DMI) of horses fed different types of forages and supplements.

Type of horse	Type of forage	DMI (g DM/kg BW)	DMI of 500kg horse (kg DM/d)	DMI (%BWT)	n =
Lactating TB	[1]Perennial ryegrass/ white clover mix	24	12	2.5	8
	[2]Orchardgrass / ryegrass (early vegetative)	24-26	12-13	2.3-2.6	3
	[2]Orchardgrass / ryegrass (mid bloom)	26-31	13-15.5	2.6-3.1	3
	-	-	[3]12.5	2.5	-
Lactating Anglo-Arab/ French saddle	[4]Low- high herbage allowance SUP barley	22.4-27	11.2-13.5	2.24-2.7	18
	[4]Low- High Herbage allowance non- SUP	19.5-24.8	9.75-12.4	1.95-2.5	18
Lactating saddle mare	[5]25 plant species	28g	14	2.8	8
	[5]SUP with concentrate	22.6	11.3	2.26	-
Non- lactating TB	[2]Orchardgrass(cocksfoot)/ perennial ryegrass	15-22	7.5-11	1.5-2.2	2
	-	-	[3]10	[3]2	-
Pregnant mare	[6]Bromegrass hay, alfalfa hay +concentrate	-	-	1.2	12
	-	-	[3]10	[3]2	-
Mature horse	[7]Tall fescue, intake rate*15hrs grazing	-	-	1.43-2.55	8

[1]Grace *et al.* (2002);[2]Marlow *et al.* (1983);[3]NRC (2007);[4]Collas *et al.* (2015);[5]Collas *et al.* (2014);[6]Guay *et al.* (2002); [7]Dowler *et al.* (2012)
SUP: supplement feed

Table 11. Efficiency of digestible energy (MJ DE) utilization in pregnant and lactating mares, and effects of diet.

Physiological state	DE (MJ/day)	ME (MJ/day)	Energy loss (%)
[1]Pregnancy (Last 8 weeks)	118.36	99.03	16.33
[1]Lactation (17 weeks)	182.48	153.54	15.85
[2,3]Diet	**DE (MJ/kg DM)**	**ME (MJ/kg DM**	**Energy loss (%)**
Grass hay, late 1st cut	7.5	6.3	16
Grass hay, 2nd cut	8.6	7.1	17.44
Alfalfa hay, late cut	7.8	5.9	24.36
Spring pasture	9.8	8.3	15.31
Oats	13.8	12.6	8.7
Barley	14.3	13.2	7.69
Maize	15.5	14.6	5.81

[1]Burlacu *et al.* (1993); [2]Kienzle and Zeyner (2010); [3]NRC (2007)

3.1.2 Model simulation

In the main modelling procedure, energy intake, energy requirement and energy balance was calculated for a mare with extra energy expenditure of 45% above maintenance that foaled on 0 to 90 days from 1st of September in every simulation. Analysis was carried out for mare with body weight of 450 kg, 500 kg, 550 kg and 600 kg based on the weight range reported for Thoroughbred mares (Slade *et al.* 1970; Grace *et al.* 2002; Pagan *et al.* 2006). The energy intake and energy balance of foaling date 0, 15, 50 and 90 days from 1st of September was compared for each body weight. Modelling of energy balance was repeated on a 500kg mare with extra energy expenditure at 25% above maintenance to represent the energy balance of a mare that has lower extra energy expenditure, or non-Thoroughbred breeds that have lower maintenance energy requirements. A sensitivity analysis was then carried out based on the main modelling procedure to investigate the response of energy intake and energy balance to changes in pasture ME, DMI, body weight, foaling date and energy requirement. Only one factor was adjusted in each analysis while other factors were kept at default values (Table 12).

Table 12. Default values for dry matter intake, pasture metabolizable energy (ME) and mare body weight (BWT) in the sensitivity analysis of the energy balance model for foaling and lactating Thoroughbred mares.

Factors	Default Values
Dry Matter Intake	Pregnant mare (2% BWT) Lactating Mare (2.5% BWT)
Pasture ME	1 (100% of given value in Figure 1.)
BWT	500

3.2 Results

3.2.1 Modelling of energy balance pattern (Main analysis)

The changes in energy balance of a mare with an initial bodyweight of 500 kg, with extra energy expenditure of 45% above maintenance (REQ+45), foaling either at 0, 15, 50 and 90 days after the 1st of September (FD0, FD15, FD50, and FD90) are presented in Figure 12. The energy balance was positive during mid to late gestation with energy balance starting at 17 MJ ME/day (FD0) and 19 MJ ME/day (FD90) at 110 days pre-partum. The energy balance decreased at a different rate as pregnancy progressed. For all foaling date scenarios, mares had a positive energy balance between 1 - 2 MJ ME/day at foaling. Overall, the energy balance decreased at a greater rate in later foaling mares (0.1MJ ME/day for FD0 vs 0.2 MJ ME/day for FD90).

There was an acute decrease in energy balance due to foaling and the onset of lactation (Figure 12). Energy balance decreased from 1 MJ ME/day at foaling to an average of -16 MJ ME/day on day 5 of lactation, for all foaling dates modelled. After day 10 of lactation, the influence of foaling date on energy balance started to show different patterns. There was no further negative change in energy balance in mares foaling on FD0 and FD15, whereas, energy balance of mares foaling on FD50 and FD90 continued to decrease and reach nadir at day 30 and day 50 of lactation, respectively. At day 100 of lactation, the daily energy deficit of FD90 mare was 12 MJ ME (34%) greater than the energy deficit of FD0 mare. Overall, the energy balance changes modelled for FD0 and FD15 were similar up to day 80 and started to diverge after that.

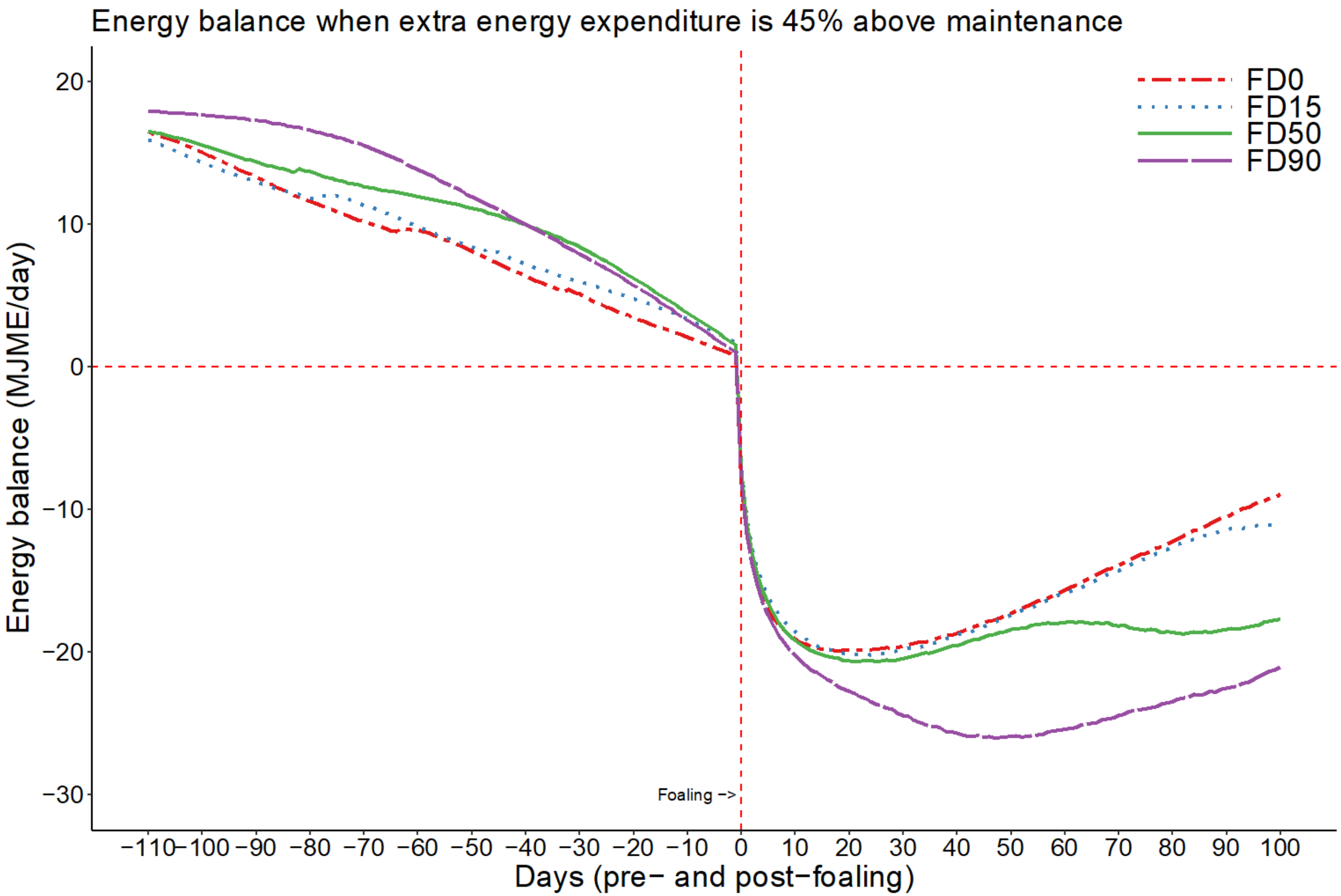


Figure 12. Pre and post-partum changes in energy balance (MJME/day) of a Thoroughbred mare (BWT=500kg) managed at pasture under commercial conditions, foaling at 0 days (FD0), 15 days (FD15), 50 days (FD50), 90 days (FD90) after 1st September when extra energy expenditure is 45% above maintenance.

3.2.2 Modelling of energy balance pattern using lower extra energy expenditure at 25% above maintenance.

The changes in energy balance of a mare with an initial bodyweight of 500 kg, an extra energy expenditure of 25% above maintenance (REQ+25), foaling either at 0, 15, 50 and 90 days after the 1st of September, is presented in Figure 13. The energy balance presented in Figure 13 is intended to represent horses with lower energy expenditure level, or non-Thoroughbred breeds with lower maintenance requirements. The pattern of energy balance changes is identical irrespective of extra energy expenditure level however the magnitude of energy balance is different. During pregnancy, the REQ+25 mare is in a greater energy surplus compared to REQ+45 mare. At 110 days pre-partum, the energy balance is 29 MJ ME/day (FD0) and 30 MJ ME/day (FD90). The energy balance of REQ+25 mare is 12 MJ ME (58%) and 11 MJ ME (63%) higher than the energy balance of a REQ+45 mare foaling on the same foaling dates. At foaling, the energy balance was between 16 - 17 MJ ME for all foaling date scenario, and is 16% higher than energy balance of a REQ+45 mare for all foaling date scenarios.

Post-partum, REQ+25 mares that foaled on FD0, FD15 and FD50 were in a slight energy surplus throughout 150 days of lactation, in contrast to REQ+45 mares that foaled on the same foaling dates that were in energy deficit. A REQ+25 mare that foaled on FD90 entered negative energy balance at day 20 post-partum and reached nadir on day 53 at -3.8 MJME/day (2.8% of daily requirement). By day 100 of lactation, REQ+25 mare returned to zero energy balance. Overall, REQ+25 mare had a higher energy balance compared to REQ+45 mares throughout the entire period modelled

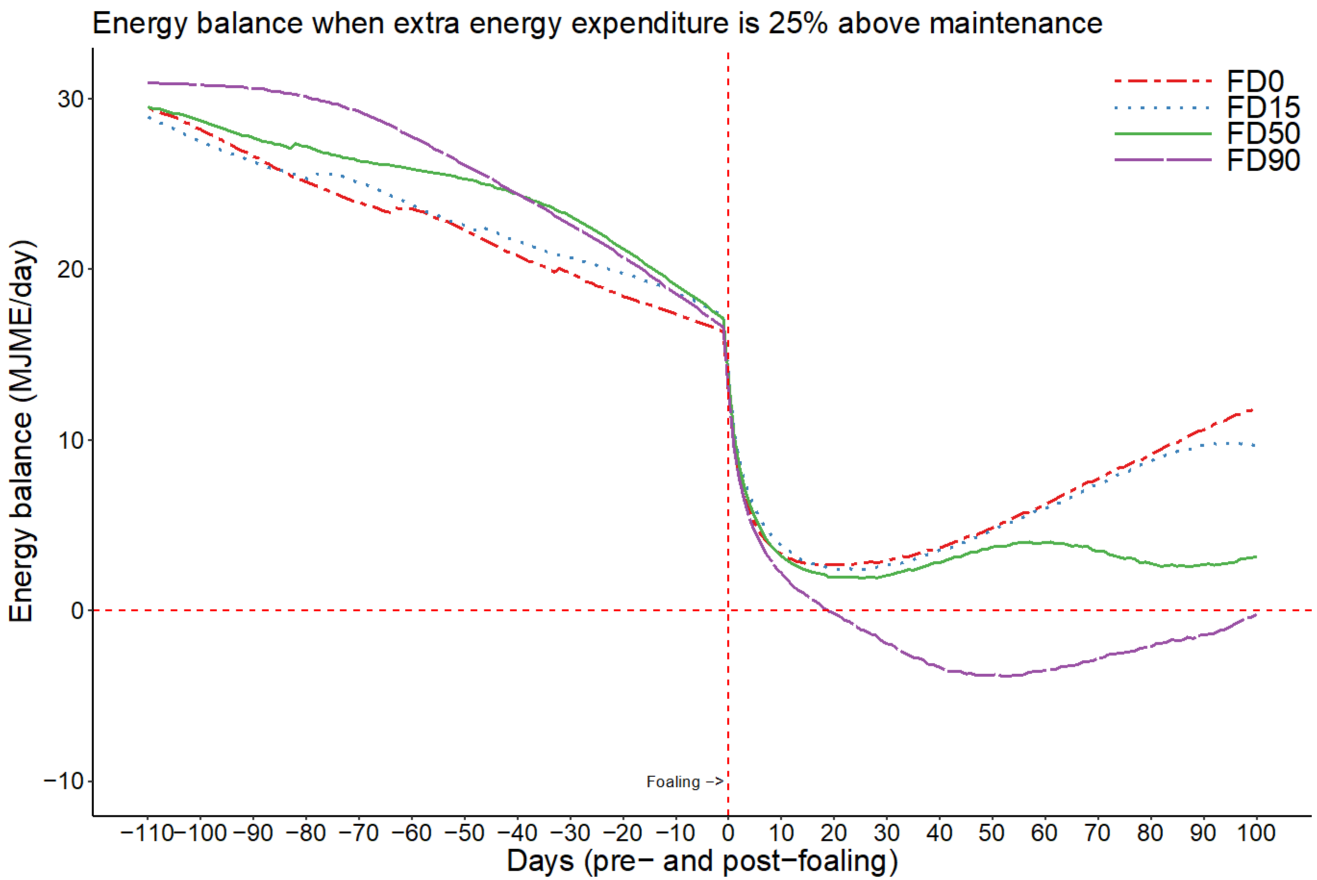


Figure 13 Pre and post-partum changes in energy balance (MJME/day) of a Thoroughbred mare (BWT=500kg) managed at pasture under commercial conditions, foaling at 0 days (FD0), 15 days (FD15), 50 days (FD50), 90 days (FD90) after 1st September when extra energy expenditure is 25% above maintenance

Table 13. Total energy balance (MJ ME), total energy requirements (MJ ME) and total energy balance expressed as percentage of requirements (%) of Thoroughbred mares managed at pasture under commercial conditions with different initial bodyweight (BWT) (kg) and relative foaling date (FD) during last three months of pregnancy and during the first 150 days of lactation.

Pregnancy				
FD	BWT(kg)	Total Energy Balance (MJ ME)	Total Requirements (MJ ME)	% of Requirements
FD0	450	845.8	13780.7	6.1
	500	939.8	15311.9	6.1
	550	1033.8	16843.1	6.1
	600	1127.8	18727.7	6.1
FD15	450	898.6	13780.7	6.5
	500	998.4	15311.9	6.5
	550	1098.3	16843.1	6.5
	600	1198.1	18374.7	6.5
FD50	450	1057.2	13780.7	7.7
	500	1174.6	15311.9	7.7
	550	1292.1	16843.1	7.7
	600	1409.5	18374.7	7.7
FD90	450	1172.2	13780.7	8.5
	500	1302.4	15311.9	8.5
	550	1432.7	16843.1	8.5
	600	1562.9	18374.7	8.5
Lactation				
FD	BWT(kg)	Total deficit (MJ ME)	Total Requirements (MJ ME)	% of Requirements
FD0	450	-1814.7	21133.4	-8.6
	500	-2016.3	23482.6	-8.6
	550	-2218.0	25829.7	-8.6
	600	-2419.6	28177.9	-8.6
FD15	450	-1954.2	21133.4	-9.2
	500	-2171.3	23482.6	-9.2
	550	-2388.5	25829.7	-9.2
	600	-2605.6	28177.9	-9.2
FD50	450	-2360.0	21133.4	-11.2
	500	-2622.2	23482.6	-11.2
	550	-2884.4	25829.7	-11.2
	600	-3146.6	28177.9	-11.2
FD90	450	-2621.5	21133.4	-12.4
	500	-2912.8	23482.6	-12.4
	550	-3204.1	25829.7	-12.4
	600	-3495.4	28177.9	-12.4

3.2.3 Effect of mare bodyweight on modelled energy balance

At the same foaling date, changes in energy balance followed a similar pattern irrespective of mare body weight (Figure 14 and 15). However, for every 50 kg increase in mare body weight, there was a 10% change in absolute value of energy balance. However, when energy balance was expressed in percentage of mare requirements, mare bodyweight had no effect on the magnitude of energy balance changes (Table 13). The energy surplus was 6.1% to 8.5% of total energy requirement in the last three months of pregnancy. The energy deficit was -8.6% to -12.4% of total energy requirements throughout lactation (150 days modelled) depending on relative expected foaling date (Table 13).

3.2.4 Effect of foaling date on modelled energy balance

Foaling later in the season increased the magnitude of positive energy balance during pregnancy (8.5% in FD90 mare, 6.1% in FD0 mare) (Table 13). Foaling later in the season increased the magnitude of post-partum negative energy balance changes. For example, total post-partum negative energy balance decreased by 32.7% and 38.7% in mares foaling on the 1st of September (FD0) and 1st of December (FD90), respectively (Table 14). Foaling later in the season also increased the magnitude of negative energy balance and prolonged the decrease in energy balance post-partum. Energy balance decreased for 10 days in mares foaling on 1st September versus 50 days in mares foaling on 1st of December (Figure 15). The energy balance nadir of mare foaling on 1st September was 12.2% of requirement versus 16.2% of requirement for a mare foaling on 1st December (Table 14). Overall, there was a positive linear relationship between foaling date and pre-partum energy balance, and a negative curvilinear relationship between foaling date and post-partum energy balance (Figure 16a and 16b).

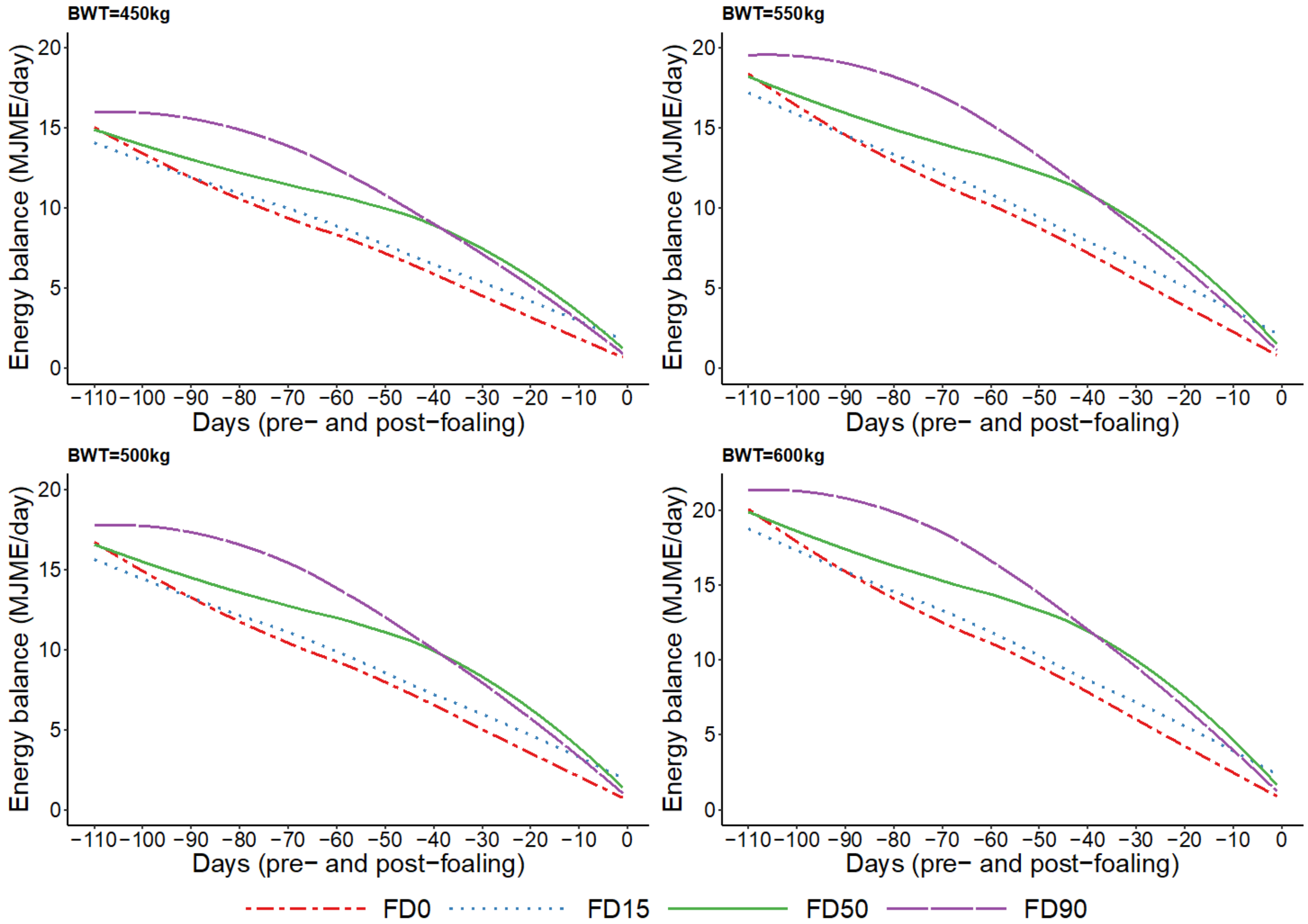


Figure 14. Effect of bodyweight (kg) on modelled pre-partum energy balance (MJME/day) in a Thoroughbred mare (at differing bodyweights) managed at pasture under commercial conditions and foaling at 0 days (FD0), 15 days (FD15), 50 days (FD50), 90 days (90) after 1st September.

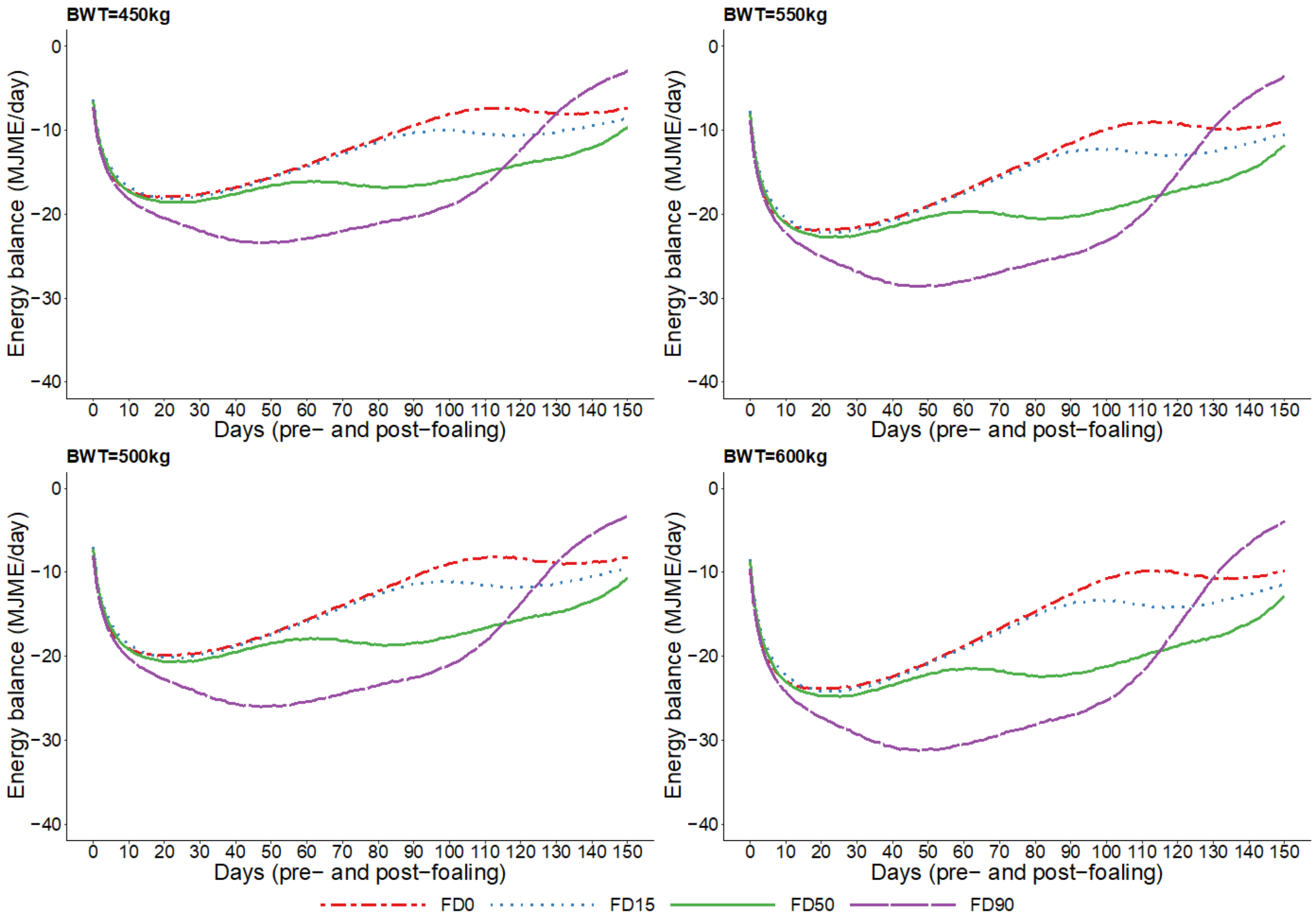


Figure 15. Effect of bodyweight (kg) on modelled post-partum energy balance (MJME/day) in a Thoroughbred mare (at differing bodyweights) managed at pasture under commercial conditions and foaling at 0 days (FD0), 15 days (FD15), 50 days (FD50), 90 days (FD90) after 1st September.

Table 14. Maximum positive energy balance, maximum negative energy balance at any point during the pre and post-foaling period modelled (day -90 to day 150) for Thoroughbred mares managed at pasture under commercial conditions with different initial bodyweight (BWT) (kg) and relative expected foaling date (FD), presented in percentage of requirement and total relative change between two points.

FD	BWT(kg)	Max. positive energy balance (%)	Max. negative energy balance (%)	Total relative change (%)
FD0	450	20.5	-12.2	-32.7
	500	20.5	-12.2	-32.7
	550	20.5	-12.2	-32.7
	600	20.5	-12.2	-32.7
FD15	450	22.2	-12.4	-34.6
	500	22.2	-12.4	-34.6
	550	22.2	-12.4	-34.6
	600	22.2	-12.4	-34.6
FD50	450	22.9	-12.6	-35.5
	500	22.9	-12.6	-35.5
	550	22.9	-12.6	-35.5
	600	22.9	-12.6	-35.5
FD90	450	22.5	-16.2	-38.7
	500	22.5	-16.2	-38.7
	550	22.5	-16.2	-38.7
	600	22.5	-16.2	-38.7

3.2.5 Effects of pasture metabolizable energy (ME) and dry matter intake (DMI).

Daily pasture ME and daily energy intake followed a similar pattern depending on the foaling date modelled (Figure 17). When DMI was kept constant, energy intake was driven by changes in pasture ME, in conjunction with shifts of foaling date. Energy intake obtained through the pasture ME and DMI values used within the model failed to satisfy the energy requirements of mares during lactation (Figure 18a and 18b).

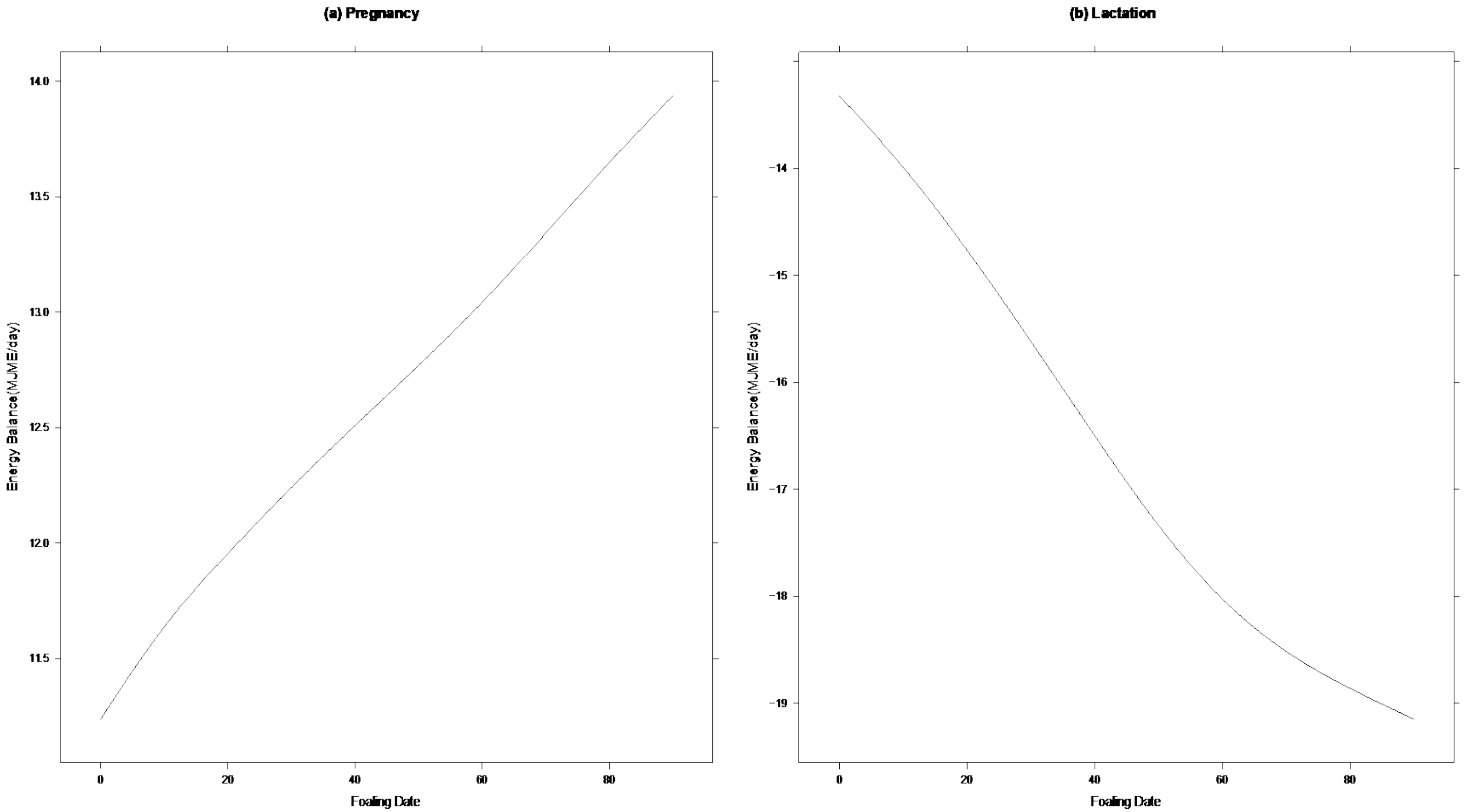


Figure 16. Relationship between average energy balance (MJME/day) and relative expected foaling date (0 to 90 days after 1st September) during (a) pregnancy and (b) lactation in Thoroughbred mares (500 kg BWT).

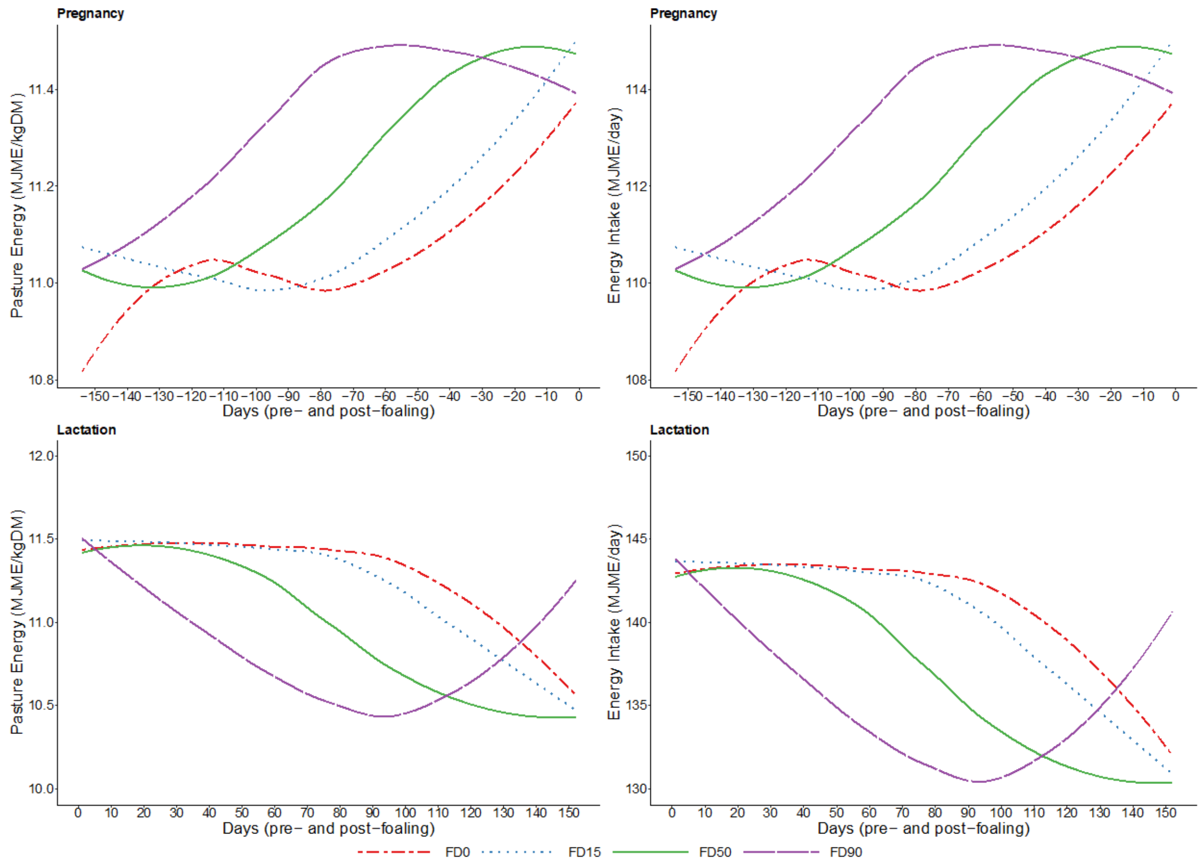
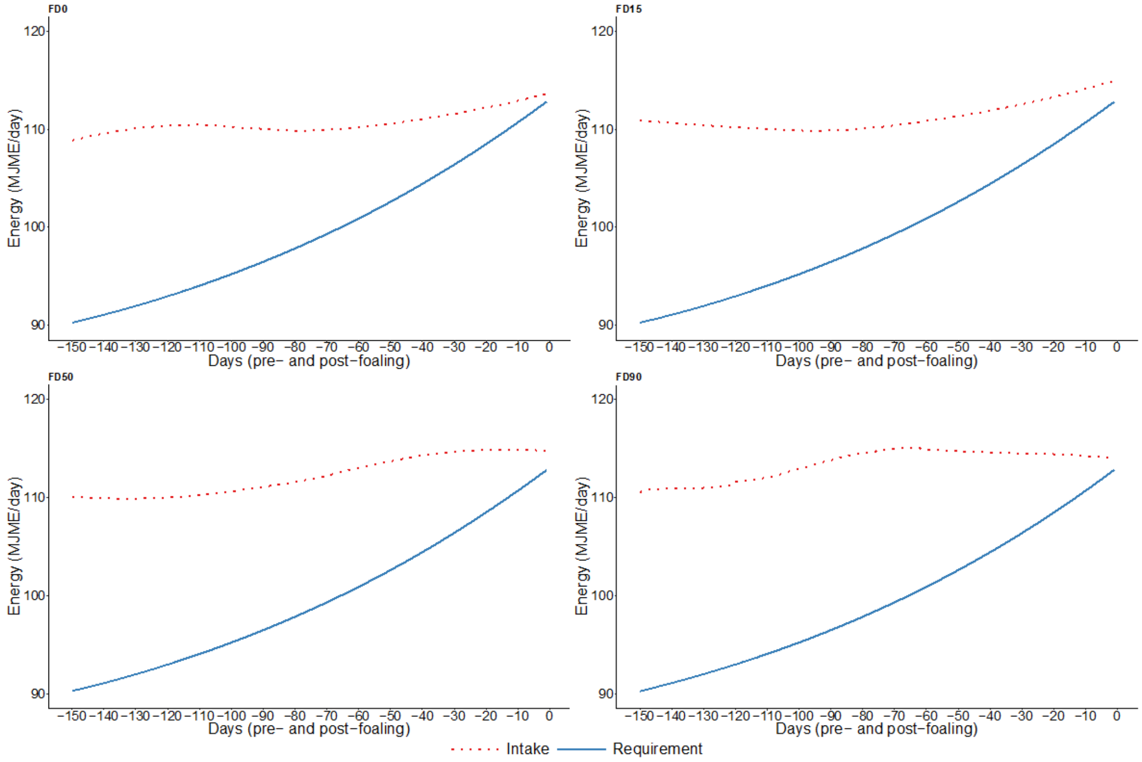


Figure 17. Pasture energy available (MJME/kg DM) and pasture energy intake (MJME/day) of a Thoroughbred mare (500 kg BWT) managed at pasture under commercial conditions, foaling on different relative expected foaling date (FD) (days after 1st September), with dry matter intake at 2% during pregnancy and 2.5% body weight during lactation.

18(a)



18(b)

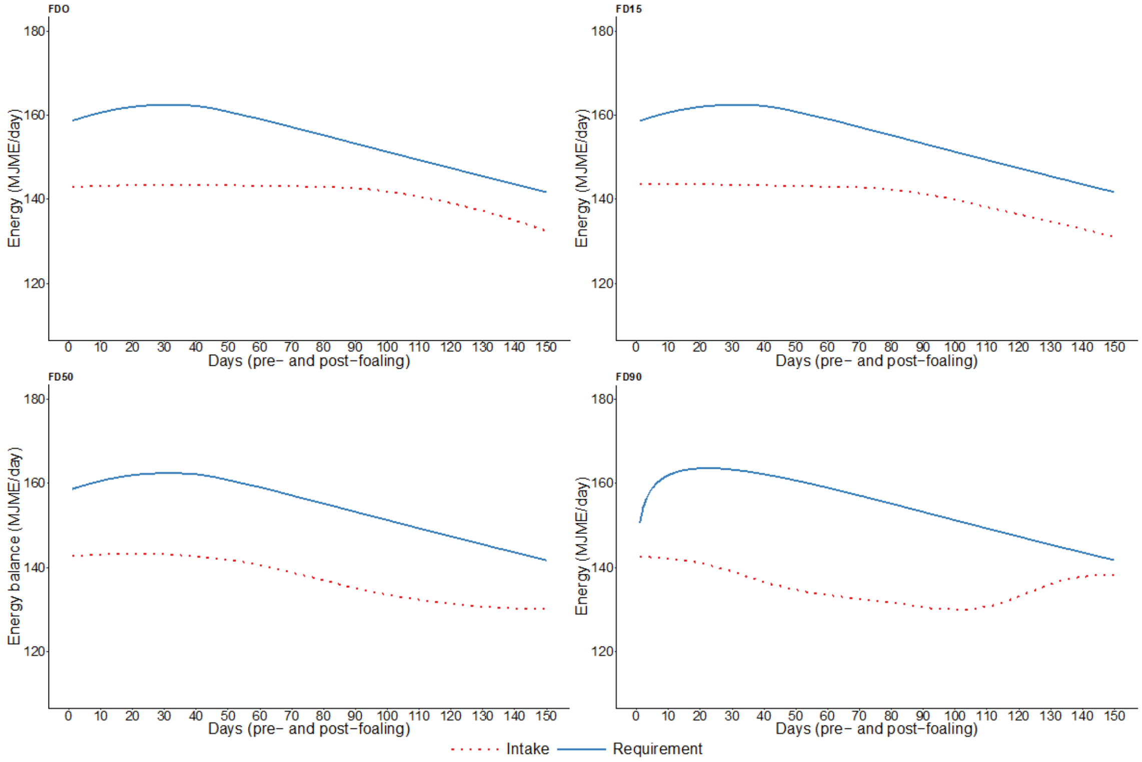


Figure 18. Energy intake and energy requirements of Thoroughbred mare (500 kg BWT) managed at pasture under commercial conditions, foaling on different relative expected foaling date (FD) (days after 1st September), consuming dry matter at (a) 2% body weight during pregnancy and (b) 2.5% body weight during lactation.

3.2.6 Sensitivity Analysis

Results from the sensitivity analysis are presented in Table 15. Overall, energy balance values in the model is more sensitive to changes in pasture ME, DMI, and energy requirement compared to body weight and foaling date. An increase or decrease in less than 1% in pasture ME, DMI and energy requirement resulted in a change of 1MJ ME/day in energy balance during pregnancy and lactation. Whereas, to create a similar amount of change in energy balance during pregnancy, an 8% change in body weight and a shift of ±33 days on foaling date was required. For 1 MJ ME change in energy balance during lactation, a 5.8% change in BWT or a shift of -12 and +20 days was needed.

Energy intake values in the model were also sensitive to changes in pasture ME, DMI and body weight. Less than 1% change in either variable will increase energy intake by 1 MJ ME during pregnancy and lactation. For foaling date, a shift of -34 and +37 days was required to change energy intake by 1 MJ ME during pregnancy. To create a similar change during lactation, a foaling date shift of -12 and +16 days was required. This suggests that only a small change in pasture ME (e.g. 0.09 MJ when pasture energy is 10 MJ ME), DMI (e.g. 0.09 kg, when DMI is 10kg), and energy requirement (e.g. 1 MJ when energy requirement is 100 MJ ME) was necessary to change the results of the energy balance modelled in the main analysis. Less than 1% increase in body weight increased energy requirement by 1 MJ ME during pregnancy and lactation. For a 500 kg mare, a 5 kg and 3.5 kg change in bodyweight will change the energy requirement and energy intake by 1 MJ ME during pregnancy and lactation respectively.

Table 15. Sensitivity of energy balance (MJME/day), energy intake (MJME/day) and energy requirements (MJME/day) of Thoroughbred mares managed at pasture under commercial conditions to changes in pasture metabolizable energy (ME), body weight (kg), relative expected foaling date, and dry matter intake (% initial body weight) from default values[2].

	Pregnancy	Lactation
Factors	[1]Change required to increase/decrease energy balance by 1 MJME	
Pasture ME	0.9 -0.9	0.7 -0.7
Dry matter intake	0.9 -0.9	0.7 -0.7
Body weight	8 -8	-5.8 5.8
Foaling date	33 -33	-12 20
Energy Requirement	-1% 1%	-0.7% 0.7%
Factors	[1]Change required to increase/decrease energy intake by 1 MJME	
ME	0.9 -0.9	0.7 -0.7
DMI	0.9 -0.9	0.7 -0.7
Body weight	0.9 -0.9	0.7 -0.7
Foaling date	34 -37	-12 16
Factors	[1]Change required to increase/decrease energy requirement by 1 MJME	
Body weight	1 -1	0.7 -0.7

[1] Refers to the % change of Pasture ME, dry matter intake, bodyweight, and energy requirement from default values required to achieve 1 MJ ME change in either energy balance or energy intake. For foaling date, change refers to number of days.

[2] Default values: Pasture ME (100% of values given in Litherland and Lambert (2007)), body weight (500 kg), DMI of pregnant mare (2% body weight), DMI of lactating mare (2.5% body weight), energy requirement (the modelled energy requirement based on default body weight), foaling date (50 days from 1st September; FD50)

3.3 Discussion

Energy balance was positive during pregnancy for all foaling date and body weight scenarios. The energy balance decreased as pregnancy progressed and reached zero energy balance at 1 day before foaling. Foaling and onset of lactation initiated an acute and prolonged period of negative energy balance throughout the lactation period modelled. Foaling date affected the magnitude of energy balance and the pattern of post-partum energy balance changes. Foaling closer to 1st September reduced the magnitude of post-partum energy balance. Foaling earlier in the season (FD0 - 15) resulted in an earlier recovery in post-partum energy balance compared to foaling later in the season (FD50 - 90). The level of energy requirement affected the magnitude of energy balance. Mares in the main analysis with extra energy expenditure at 45% above maintenance requirement, had a more negative energy balance compared to mares with extra energy expenditure at 25% above maintenance requirement. Level of extra energy expenditure had no effect on the energy balance pattern and its changes.

3.3.1 Pre-partum energy balance

The main model analysis described a positive energy balance during early pregnancy. The magnitude of the positive energy balance decreased during the second trimester and approached zero energy balance 1 day before foaling. The findings from our main analysis were consistent with those of Lawrence *et al.* (1992), both indicate a decrease in energy balance as pregnancy progressed which will affect body weight and body fat changes in the mare. During pregnancy, it was reported that Quarter horse mares increased in body weight, body condition score and rump fat thickness from the onset of the 2nd trimester (starting day 90 of gestation). During the third trimester, there was less weight gain (30% of that observed during the second trimester), and a plateau in rump fat thickness and body condition score starting day 240 of gestation. The body condition score and rump fat thickness of the Quarter horse mare described by Lawrence *et al.* (1992) started to decrease from the last 60 days and last 30 days of gestation respectively. Whereas, the Thoroughbred model in this study estimated that negative energy balance was initiated at foaling and onset of lactation. Therefore, the mares managed under commercial conditions at pasture in this study had a later onset of negative energy balance compared to those in the study by Lawrence *et al.* (1992).

3.3.2 Post-partum energy balance

Within this Thoroughbred energy balance model, parturition and the onset of lactation provided an acute and prolonged period of negative energy balance. This coincided with the period of decreased rump fat thickness and reduced body condition score of the lactating Quarter horse mares reported by Lawrence *et al.* (1992). Interestingly, the greatest change in body condition score reported in the Quarter horse mares throughout the study was never more than 1 score of body condition (equivalent to 16 – 20 kg body weight (NRC 2007)). The greatest reduction in rump fat thickness reported (0.4 cm), indicated that the mobilization of subcutaneous body reserves during lactation was moderate despite the prolonged period of energy deficit. Similar levels of body weight loss (0.25kg/d or 6kg) during the 1st month of lactation and a decrease in body condition score (0.2 point on 6- point scale) were reported for early foaling (January foaling) Thoroughbred mares on pasture (Pagan *et al.* 2006). These data imply that the negative energy balance associated with foaling and lactation may be consistent across breeds when managed at pasture.

3.3.3 Energy balance and mobilization of body composition

According to Cordero (2011), a 500 kg mare with body condition score of 6 and body fat of >10% would require a deficit of 4.85 Mcal DE/day (equivalent to 17 MJ ME/d) to lose 1 body condition score over a 30 day period. The final model of Cordero (2011) was unable to estimate actual time required for 1 body condition score loss because the model design did not account for individual variation in metabolic efficiency. In the current study, a 500 kg mare (FD50) had an average daily energy deficit of 18.7 MJME/ day during first 60 days of lactation, which theoretically would be sufficient to cause a loss in up to 1 body condition score. However, Lawrence *et al.* (1992) and Pagan *et al.* (2006) observed limited body condition score and subcutaneous fat loss in mares on pasture. This can be attributed to horses preferentially utilizing their substantial internal (visceral) fat storage. In Thoroughbred foals, approximately 30% of total body fat is carried viscerally (Gee *et al.* 2003). Assessment of body composition in ponies identified visceral fat to account for approximately 47% of total body fat (Dugdale *et al.* 2011). There are no published data on visceral and subcutaneous fat distribution of Thoroughbred mares. In the study by Dugdale *et al.* (2011), there are evidence suggesting that internal fat is always preferentially mobilized when BCS is >4 . External fat starts to be mobilized only when the body condition score drops below 4 (Dugdale *et al.* 2011). This may in part explain the anecdotal reports of limited change in body condition score during lactation with mares managed under commercial pastoral conditions in New Zealand.

The lack of obvious change in body condition score in mares may also be attributed to the mobilization of lean body mass. Lean body mass of well-fed mares was reported to decrease by 1.5% (equivalent to -7.5 kg for a 500 kg mare) during lactation without change in rump fat thickness (Manso Filho *et al.* 2008). Plasma glutamate and plasma glucose decreased for 6 and 8 weeks after parturition respectively (Manso Filho *et al.* 2008). Hence, this lean body mass mobilization may take place to provide amino acid precursors for milk protein and gluconeogenesis for milk lactose. Mares milk is rich in glutamine and the plasma glucose turn over during lactation was reported to be 40% with the majority of glucose turnover (90%) contributed to milk lactose (Anwer *et al.* 1975). This has practical implications when using body condition scoring to assess dietary energy adequacy. It only subjectively assesses subcutaneous fat and may not accurately reflect the relative proportion of body composition mobilised. Thus, making it difficult to detect a nutritional deficit under commercial stud farm conditions without monitoring body weight along with body composition changes.

At present, the empirical data to understand the relative utilisation of sub-cutaneous and visceral body fat with changes in body condition score, and lean body mass mobilization relative to body weight loss for Thoroughbred broodmares managed at pasture is lacking. Therefore, the model findings should be interpreted with caution and further studies are required to fully understand the individual, breed, and age effect on equine body fat distribution, and how body condition score and dietary energy changes can affect body composition.

3.3.4 Foaling date and modelled energy balance

Shifting foaling date closer to 1st of September reduced the post-partum energy deficit by reducing the disparity between energy supply and demand supporting our hypothesis that there is an overlap of high energy requirement and low pasture energy. The modelled results presented in Figure 17 shows the energy intake and pasture ME follow a similar pattern when DMI was kept constant. The patterns are distinctive depending on the foaling date. These findings support the hypothesis that at a given DMI, foaling date would influence pasture energy available and subsequently the energy intake. These results indicate that strategies are needed to manage the fluctuation of energy supply and demand within a pastoral system that is due to the influence of foaling date and seasonality of pasture. A greater understanding on energy requirement of the mares and the pasture quality changes on the farm would be beneficial.

3.3.5 Post-partum energy balance and reproductive performance

Foaling date affected the post-partum energy balance and its pattern. Mares that foaled later in the season (FD50 - 90, after 22nd October) had greater post-partum negative energy balance and showed a prolonged (30 - 50 days) decrease in post-partum energy balance. On the contrary, mares that foaled earlier in the season (FD0 - 15, before 15 Sep) experienced a smaller post-partum negative energy balance, a less dramatic change in energy balance when transitioning from pregnancy to lactation, and showed earlier recovery in energy balance within 20 days post-partum. Similar energy balance changes in dairy cows were reported to have implications on reproductive performance. A positive relationship was reported between post-partum energy balance changes, plasma IGF-1 concentration, function of dominant follicles, and shorter post-partum to ovulation interval (Beam and Butler 1999). An earlier (before day 25 post-partum) recovery in energy balance was reported to improve ovarian function in dairy cows (Lucy *et al.* 1991).

Fluctuation of the modelled energy balance may in part explain the report by Hanlon *et al.* (2012a) which described mares that foaled later in the season (after 22nd October), had longer start of mating to conception intervals and were less likely to be pregnant at the end of the season than mares that foaled earlier (before 11th September). It is possible that early foaling mares in the study of Hanlon *et al.* (2012a) had smaller post-partum negative energy balance and earlier recovery of energy balance which may have contributed to the better reproductive efficiency however this theory requires validation with further research. In the current study, mares had post-partum energy deficit of 8.6% and 12.4% of maintenance requirements which are comparable to findings reported by (Fradinho *et al.* 2014). Mares in the study of Fradinho *et al.* (2014) that had energy deficits of -6.4% and -3.9% of maintenance requirements at conception and during the first 3 months of lactation, respectively, had reduced fertility during the first two oestrous cycle compared to mares that were not in an energy deficit. At present, evidence from this study and others support that the effect of post-partum energy balance on reproductive performance in the mare should be investigated further.

3.3.6 Modelled energy balance and leptin levels

In this Thoroughbred model, a mare weighing 500 kg and foaling at FD50 (22nd October) had energy deficit of -11.2% of daily requirement during lactation. In Lusitano mares, energy deficit of -6.4% and -3.9% of maintenance requirements at conception and during the first 3 months of lactation initiated a 2 - 3 ng/mL decrease in plasma leptin (Fradinho *et al.* 2014). Therefore, the energy deficit estimated by the model may be sufficient to initiate a decrease in leptin levels. Leptin is produced by adipocytes and acts as a signal to the central nervous system on adiposity and regulates the reproductive system.

A recent *in vitro* study reported that addition of 10 ng/mL of leptin to the *in vitro* medium increased fertilization rate by 33% compared to the control group after intracytoplasmic sperm injection (Consiglio *et al.* 2009). The 2 – 3 ng/mL change in leptin levels observed in Lusitano mares by Fradinho *et al.* (2014) would, theoretically, be sufficient to create a 6 - 9% decrease in fertilization rate, and partly explains the increase in number of oestrus cycles required to conceive that was observed in mares that were in energy deficit. Within this Thoroughbred model, the difference in total energy deficit during lactation between mares foaling before 15th September and after 22nd October was 2.6%. This may be sufficient to create a variation in oocyte fertilization rate of mares foaled on different dates. Interpretation of evidence from this study and others provide theoretical indication that the estimated energy deficit in the model may initiate changes in leptin levels that would potentially affect oocyte fertilisation rate, and a small variation in energy balance may be sufficient to create a difference in fertility hence a variation in reproductive performance.

3.3.7 Energy expenditure level and modelled energy balance

In the current study, energy requirement affected the magnitude of energy balance. Increase in energy expenditure increased energy requirement and lowered the energy balance throughout the entire modelled period. As a result, REQ+25 mares had a greater energy surplus during pregnancy, and the early foaling (FD0 - 50) mares avoided post-partum energy deficit when compared to REQ+45 mares. The substantial difference in the modelled energy balance between two energy requirement levels emphasize that the application of the model data to other horse breeds or management conditions are limited. The model data in this study is highly specific to Thoroughbred broodmares managed commercially on pasture, as the environment and housing conditions may affect the type and intensity of activity and hence the energy requirement. There were also reports that maintenance requirements of Thoroughbreds are higher than other horse breeds (Coenen *et al.* 2011). It is also reported that the Thoroughbred is primarily bred for the racing performance trait and little consideration is given for feed conversion efficiency, which results in (anecdotal) observations of large variation in feed efficiency within this breed (Gu *et al.* 2009). Therefore, the energy requirement of the Thoroughbred mares on pasture may be elevated compared to other horse breeds or different from horses under different management and housing conditions. Hence, future improvements in energy requirement models should focus on effect of horse breeds, energy expenditure under different housing, environment and management conditions.

3.3.8 Limitations

In the current study, DMI was held constant throughout the modelled period (2% and 2.5% body weight during pregnancy and lactation, respectively). The assumptions for DMI of lactating mares were based on limited number of reports (Table 10). The effects of pasture growth rate, herbage allowance, pasture energy and their interactions on DMI were not included because they are not fully understood (Marlow *et al.* 1983; Fleurance *et al.* 2009; Dowler *et al.* 2012; Glunk *et al.* 2013; McGowan *et al.* 2013; Collas *et al.* 2014; Collas *et al.* 2015). Therefore, the intake values that were used in the current model reflects a consistent DMI under minimal restricting factors. This value may not be correct to the mare DMI under commercial management conditions and may influence the modelled results. Under commercial conditions, DMI can vary due to changes in herbage allowance or opportunity to access pasture. The stocking density on commercial breeding farms increases dramatically during the breeding season and may reduce the effective pasture DM (Kg DM / ha) on offer (Rogers *et al.* 2007; Hirst 2011). The movement of mares and the need to bring the mare cohort up to the yards for scanning post-partum will also reduce the time available for grazing hence may limit voluntary feed intake of the mare (Rogers *et al.* 2007). Therefore, use of constant DMI limited the sensitivity of our model towards the energy balance and energy intake changes driven by DMI variation.

The application of the model findings on stud farm level is limited. The pasture ME used in this model was derived for ruminants due to lack of information on seasonal changes in pasture energy on New Zealand stud farms. The pasture ME in the model may not reflect the pasture energy value available to horses considering ruminants are more efficient in digesting fibre compared to horses (Pearson *et al.* 2006). The pasture ME used in this model represents the average annual and seasonal pasture energy pattern observed on New Zealand diary and sheep farms (Litherland and Lambert 2007). This pasture ME data cannot be generalised for all dairy, sheep and equine farms due to the amount of pasture ME variation (Litherland and Lambert 2007).

To precisely simulate the commercial stud farm management conditions and estimate energy requirement specific to Thoroughbred horses, the energy requirement in the model accounted for extra energy required for activities on commercial stud farms such as walking (Nicol and Brookes 2007), foraging (Vermorel *et al.* 1997), and group housing (Coenen *et al.* 2011), and increased maintenance requirement in Thoroughbred mares compared to other breeds (Coenen *et al.* 2011). However, there are limited data available on energy costs for these activities for Thoroughbreds. There are also limited reports on difference in metabolic rate and maintenance requirements between different horse breeds. Therefore, the assumptions for extra energy requirement other than maintenance, pregnancy and lactation were based on small number of reports in non-Thoroughbred studies.

3.3.9 Sensitivity analysis

To quantify the effects of over or under-estimation of variables on the results of the main analysis, sensitivity analysis was conducted on DMI, pasture energy, foaling date, body weight, and energy requirement to investigate their effect on energy balance and energy intake changes. Based on the sensitivity analysis results, only a small change (~1%) in pasture ME, DMI and energy requirement may cause a 1 MJ ME change in energy balance and energy intake. A variation in 1 MJ pasture ME and energy requirement or 1 kg DMI, for example, will result in more than 10 MJ ME change in energy intake and energy balance (5.8% of lactation requirement). For a 500 kg mare, a 1% change in DMI will result in 5 kg change in DMI and at least 50 MJ ME (29% of lactation requirement) change in energy intake and energy balance. The sensitivity of energy balance to pasture energy and DMI emphasize that nutritional status of the mare is greatly influenced by pasture quality and quantity therefore it is important to provide quality pasture, and optimize dry matter and energy intake. The significance of energy requirement to the modelled results further support the need for greater understanding in energy requirement specific to breed and field conditions.

Energy balance was less sensitive to body weight because both the DMI and energy requirement increased with body weight in the model. A 3.5kg to 5kg change in body weight is required to initiate a 1 MJ ME change in either energy intake, energy requirement or energy balance. This model included a wide body weight range (450 – 600 kg) thus it can be applied to majority of Thoroughbred body weights (500 – 600 kg) (Slade *et al.* 1970; Grace *et al.* 2002; Pagan *et al.* 2006). It should be noted that, the changes in sensitivity analysis do not fully reflect changes under realistic conditions because only one variable was adjusted in each analysis, where realistically, all the variables may change concurrently. In summary, the results from sensitivity analysis show that data used in the modelling process, and the model design imposed limitations on the model application to specific stud farms, other horse breeds and horses under different management conditions.

3.4 Conclusion

The modelled results from the main modelling procedure are in agreement with the trend for subcutaneous fat thickness, body condition and body weight changes in mares kept on pasture reported in other studies. The present study found that Thoroughbred mares managed under New Zealand commercial grazing conditions, initiate a large and prolonged energy deficit soon after foaling and throughout 150 days of lactation in all foaling dates modelled. The onset of negative energy deficit modelled in this study was quick and may be sufficient to initiate mobilization of body fat and lean body mass, along with a decrease in leptin levels which may negatively affect the reproductive performance of Thoroughbred mares managed at pasture. Further studies are needed to verify the modelled data, along with the resulting changes in body reserves, body weight and leptin levels and the consequences these changes may have on the mare's reproductive performance. The energy balance of the mare is influenced by the balance between energy supply and demand that fluctuates with foaling date and seasonal pasture energy. The sensitivity of energy balance and energy intake to pasture energy and DMI changes emphasize the importance of pasture quality and quantity in nutritional management of the mare. At present, initiative should be taken to optimise dry matter and pasture intake to improve nutritional status of mares, and utilise available pasture effectively to boost industry management and production efficiency. This can be achieved through increasing the use of nutritional model as a tool to estimate energy requirement, closer monitoring of pasture quality and quantity on stud farm, and monitoring body weight, body fat and body composition changes in the mares.

Chapter 4: General discussion

4.1 Summary of findings

In this Thoroughbred model, a mare was in an energy surplus during pregnancy. Foaling and onset of lactation initiated energy deficit throughout lactation for all foaling dates and body weights. The magnitude and duration of energy deficit is theoretically sufficient to cause a decrease in leptin concentration and body condition score. Delay in foaling date increased the magnitude and duration of post-partum energy deficit. No effect was observed in mare body weight on magnitude of energy balance changes and energy deficit. Foaling early (FD0 - 15) can reduce post-partum energy deficit and prevent prolonged decrease in post-partum energy balance by synchronizing peak pasture energy available with the mare's theoretical lactation requirements. Energy intake is driven by changes in available pasture energy in conjunction with shifts of foaling date. These findings support our hypothesis that there is a disparity between pasture energy availability and mare lactation requirements. Increase in energy requirement due to increase in extra energy expenditure reduced the energy balance throughout the modelling period but did not affect the pattern of energy balance changes.

The main drivers of energy balance changes are pasture ME, DMI and energy requirement. Energy intake is more sensitive to pasture ME, DMI and body weight changes. The energy intake and energy requirement vary with similar sensitivity to changes in body weight thereby resulted in less effect of body weight on energy balance compared to pasture ME, DMI and energy requirement.

4.2 Limitations

Within this Thoroughbred model, DMI was held constant throughout the modelled period at 2% and 2.5% of body weight during pregnancy and lactation. This has restricted the ability of the model to include changes in energy balance and energy intake that would be caused by variation in DMI due to changes in herbage allowance or access to pasture. The DMI value is an average estimate and based on limited number of reports. This may cause an over or underestimation of energy balance depending on herbage availability and management conditions. The pasture energy value used in this model represents an average pattern specific to New Zealand dairy pasture. Therefore, farm and geographical specific application of the model is limited.

With extended period of over or undernutrition, body weight and metabolic efficiency may change. As a result, energy requirement may alter throughout the modelled period. These variations were not included in the model due to lack of empirical data in body weight changes, body fat mobilization and metabolic efficiency in Thoroughbred mares. Therefore, this model reflects majority but not all variables originating from commercial management and physiological aspects.

With reports of variation in maintenance requirement between horse breeds and potential variation in energy expenditure under different housing and management conditions, the model data is specific to Thoroughbred mares managed on pasture under commercial conditions. Furthermore, the energy expenditure for activities on commercial stud farms such as walking, foraging, and group housing were based on non-Thoroughbred studies due to lack of Thoroughbred data. Therefore, the modelled results should be applied with caution especially on other horse breeds and management conditions, and requires further verification.

4.3 Future studies

To overcome the limitations above, more accurate estimation of DMI is required. To obtain this information, study on quality (chemical composition) and quantity (herbage allowance) of equine pasture and their effects on DMI is needed. Body weight and body composition changes in mares at pasture is required to understand the pattern of body fat and lean body mass mobilization so that model accuracy for energy requirement changes can be improved. Farm and region specific pasture energy and growth patterns are required to expand the model application. Combining these field data would also help verify the model data.

The impact of energy requirement to energy balance warrants further improvements in current energy requirement models by focusing on effect of horse breeds and energy expenditure differences. More studies on the breed variation on maintenance requirement and metabolic efficiency is required. Studies that measure energy expenditure of horses for specific activities, under different housing, environment and management conditions would be beneficial.

To fully understand the nutritional effects on reproductive performance, the consequence of body weight and body condition changes due to the modelled energy deficit and post-partum energy balance pattern to performance measures (i.e. post-partum to ovulation interval, number of oestrous cycle per conception) should be investigated. Studies to monitor the modification in metabolic indicators such as leptin, insulin and IGF-1 are required as well to contribute towards the full understanding in the nutrition-reproduction relationship and regulation. At present, all information above is limited for New Zealand Thoroughbred broodmares hence preliminary measurements on large sample sizes will be needed to establish a baseline for the population.

4.4 Implications and conclusion

With our findings and knowledge from other studies, evidence suggests in order to accurately assess nutritional state of the mare requires monitoring of body weight and body composition changes. However, weighing mares and monitoring body composition changes are not a common practice in New Zealand stud farms. The subjectivity of body condition scoring may also fail to reflect visceral fat and lean body mass mobilization. Hence, undernutrition may go unnoticed.

The model findings support that there is an overlap of high energy requirement and low pasture energy which may affect the efficiency in utilisation of available pasture energy. At a given DMI, foaling date would influence pasture energy available and subsequently the energy intake. The sensitivity of energy balance to energy requirement indicate that nutrition of mares should be managed differently based on her energy demand which may differ depending on the environment, housing, and physiological state. The sensitivity of energy balance to pasture energy and DMI emphasise that nutritional status of the mare is greatly influenced by pasture quality and quantity. Overall, stud farms would require a strategy to manage the energy supply and demand of mares, and improve pasture energy utilisation. This can be achieved by regular monitor of pasture growth and quality, dry matter and energy intake, and increase the use of nutritional models to understand energy requirement of the mares.

Under commercial pastoral management, mares are subjected to undernutrition during lactation. Delay in foaling will increase the post-partum energy deficit and prolong the decrease in energy balance. Interpretation of results in this study and others indicate that due to this, there may be negative consequences on mare reproduction. However, there is a gap in literature between energy balance, body composition, body weight and metabolic hormone changes which is preventing an affirming conclusion regarding the effects of energy balance and reproductive performance. It warrants greater attention and research into this area.

In conclusion, the model findings suggests nutritional and farm operational benefits for stud farms to actively manage the energy supply and demand. The energy deficit and decrease in post-partum energy balance should be avoided due to the prospective effects on reproductive performance. Initiative should be taken to monitor body weight and body composition changes, pasture quality and growth pattern, and optimize energy and dry matter intake in the mares. These efforts can assist in maximizing pasture utilization, minimize energy deficit and achieve costs effective pastoral and nutritional management.

References

Aljarrah AH (2004) Methods to induce early onset of cyclicity in transitional mare. Master thesis, Louisiana State University.

Anwer MS, Gronwall R, Chapman TE, Klentz RD (1975) Glucose utilization and contribution to milk components in lactating ponies. *Journal of Animal Science* **41**, 568-571.

Barash IA, Cheung CC, Weigle DS, Ren H, Kabigting EB, Kuijper JL, Clifton DK, Steiner RA (1996) Leptin is a metabolic signal to the reproductive system. *Endocrinology* **137**, 3144-3147.

Beam SW, Butler WR (1998) Energy balance, metabolic hormones, and early postpartum follicular development in dairy cows fed prilled lipid. *Journal of Dairy Science* **81**, 121-131.

Beam SW, Butler WR (1999) Effects of energy balance on follicular development and first ovulation in postpartum dairy cows. *Journal of Reproduction and Fertility Supplement* **54**, 411-424.

Bott RC, Greene EA, Koch K, Martinson KL, Siciliano PD, Williams C, Trottier NL, Burk A, Swinker A (2013) Production and environmental implications of equine grazing. *Journal of Equine Veterinary Science* **33**, 1031-1043.

Bruinenberg MH, Valk H, Korevaar H, Struik PC (2002) Factors affecting digestibility of temperate forages from seminatural grasslands: a review. *Grass and Forage Science* **57**, 292-301.

Buff P, Dodds A, Morrison C, Whitley NC, McFadin E, Daniel J, Djiane J, Keisler D (2002) Leptin in horses: tissue localization and relationship between peripheral concentrations of leptin and body condition. *Journal of Animal Science* **80**, 2942-2948.

Buff P, Morrison C, Ganjam V, Keisler D (2005) Effects of short-term feed deprivation and melatonin implants on circadian patterns of leptin in the horse. *Journal of Animal Science* **83**, 1023-1032.

Burlacu G, Voicu D, Voicu I, Nicolae M, Petrache E, Georgescu G, Balan S (1993) Study on the energy and protein metabolism in horses. *Archives of Animal Nutrition* **45**, 173-185.

Canfield RW, Butler WR (1990) Energy balance and pulsatile LH secretion in early postpartum dairy cattle. *Domestic Animal Endocrinology* **7**, 323-330.

Chavatte P (1997) Lactation in the mare. *Equine Veterinary Education* **9**, 62-67.

Coenen M, Kienzle E, Vervuert I, Zeyner A (2011) Recent German developments in the formulation of energy and nutrient requirements in horses and the resulting feeding recommendations. *Journal of Equine Veterinary Science* **31**, 219-229.

Collas C, Fleurance G, Cabaret J, Martin-Rosset W, Wimel L, Cortet J, Dumont B (2014) How does the suppression of energy supplementation affect herbage intake, performance and parasitism in lactating saddle mares? *Animal* **8**, 1290-1297.

Collas C, Dumont B, Delagarde R, Martin-Rosset W, Fleurance G (2015) Energy supplementation and herbage allowance effects on daily intake in lactating mares. *Journal of Animal Science* **93**, 2520-2529.

Consiglio AL, Dell'Aquila ME, Fiandanese N, Ambruosi B, Cho YS, Bosi G, Arrighi S, Lacalandra GM, Cremonesi F (2009) Effects of leptin on in vitro maturation, fertilization and embryonic cleavage after ICSI and early developmental expression of leptin (Ob) and leptin receptor (ObR) proteins in the horse. *Reproductive Biology and Endocrinology* **7**, 113.

Cordero V (2011) Development of a nutritional model to predict digestible energy requirements of broodmares based on body condition changes. Master thesis, Texas A&M University.

Cymbaluk NF, Christison GI (1990) Environmental effects on thermoregulation and nutrition of horses. *Veterinary Clinics of North America: Equine Practice* **6**, 355-372.

Davies Morel MCG, Newcombe JR, Holland SJ (2002) Factors affecting gestation length in the Thoroughbred mare. *Animal Reproduction Science* **74**, 175-185.

Davies Morel MCG (2015) 'Equine reproductive physiology, breeding and stud management.' (CABI: Wallingword, UK)

Doreau M, Boulot S, Martin-Rosset W, Robelin J, Dubroeucq H, Lefaivre R (1986) Relationship between nutrient intake, growth and body composition of the nursing foal. *Reproduction Nutrition Développement* **26**, 683-690.

Doreau M, Martin-Rosset W, Boulot S (1988) Energy requirements and the feeding of mares during lactation: A review. *Livestock Production Science* **20**, 53-68.

Doreau M, Boulot S (1989) Recent knowledge on mare milk production: A review. *Livestock Production Science* **22**, 213-235.

Doreau M, Boulot S, Barlet J-P, Patureau-Mirand P (1990) Yield and composition of milk from lactating mares: effect of lactation stage and individual differences. *Journal of Dairy Research* **57**, 449-454.

Doreau M, Boulot S, Martin-Rosset W (1991) Effect of parity and physiological state on intake, milk production and blood parameters in lactating mares differing in body size. *Animal Production* **53**, 111-118.

Doreau M, Boulot S, Chilliard Y (1993) Yield and composition of milk from lactating mares: effect of body condition at foaling. *Journal of Dairy Research* **60**, 457-466.

Doreau M, Martuzzi F (2006) Milk yield of nursing and dairy mares. *European Association for Animal Production* **120**, 57.

Dowler LE, Siciliano PD, Pratt-Phillips SE, Poore M (2012) Determination of pasture dry matter intake rates in different seasons and their application in grazing management. *Journal of Equine Veterinary Science* **32**, 85-92.

Dugdale AHA, Curtis GC, Harris PA, Argo C (2011) Assessment of body fat in the pony: Part I. Relationships between the anatomical distribution of adipose tissue, body composition and body condition. *Equine Veterinary Journal* **43**, 552-561.

Elliott C, Morton J, Chopin J (2009) Factors affecting foal birth weight in Thoroughbred horses. *Theriogenology* **71**, 683-689.

Ferreira-Dias G, Claudino F, Carvalho H, Agrícola R, Alpoim-Moreira J, Robalo Silva J (2005) Seasonal reproduction in the mare: possible role of plasma leptin, body weight and immune status. *Domestic Animal Endocrinology* **29**, 203-213.

Fleurance G, Fritz H, Duncan P, Gordon IJ, Edouard N, Vial C (2009) Instantaneous intake rate in horses of different body sizes: influence of sward biomass and fibrousness. *Applied Animal Behaviour Science* **117**, 84-92.

Forbes JM (1970) The Voluntary Food Intake of Pregnant and Lactating Ruminants: A Review. *British Veterinary Journal* **126**, 1-11.

Fowden AL, Forhead AJ, White KL, Taylor PM (2000) Equine uteroplacental metabolism at mid and late gestation. *Experimental Physiology* **85**, 539-545.

Fradinho MJ, Correia MJ, Grácio V, Bliebernicht M, Farrim A, Mateus L, Martin-Rosset W, Bessa RJB, Caldeira RM, Ferreira-Dias G (2014) Effects of body condition and leptin on the reproductive performance of Lusitano mares on extensive systems. *Theriogenology* **81**, 1214-1222.

Garza F, Thompson DL, French DD, Wiest JJ, St George RL, Ashley KB, Jones LS, Mitchell PS, McNeill DR (1986) Active immunization of intact mares against gonadotropin-releasing hormone: differential effects on secretion of luteinizing hormone and follicle-stimulating hormone. *Biology of Reproduction* **35**, 347-352.

Gee EK, Fennessy PF, Morel PCH, Grace ND, Firth EC, Mogg TD (2003) Chemical body composition of 20 Thoroughbred foals at 160 days of age, and preliminary investigation of techniques used to predict body fatness. *New Zealand Veterinary Journal* **51**, 125-131.

Gee EK, Rogers CW, Bolwell CF (2017) Commercial equine production in New Zealand. 1. Reproduction and breeding. *Animal Production Science* **online early**, -.

Gentry LR, Thompson DL, Gentry GT, Davis KA, Godke RA, Cartmill JA (2002) The relationship between body condition, leptin, and reproductive and hormonal characteristics of mares during the seasonal anovulatory period. *Journal of Animal Science* **80**, 2695-2703.

Geor RJ, Harris PA (2007) 'How to minimize gastrointestinal disease associated with carbohydrate nutrition in horses, Proceedings of the Annual Convention of the AAEP.'

Ginther OJ (1992) 'Reproductive biology of the mare: basic and applied aspects.' (Equiservices Wisconsin, US)

Giussani DA, Forhead AJ, Fowden AL (2005) Development of cardiovascular function in the horse fetus. *The Journal of Physiology* **565**, 1019-1030.

Glunk EC, Pratt-Phillips SE, Siciliano PD (2013) Effect of restricted pasture access on pasture dry matter intake rate, dietary energy intake, and fecal pH in horses. *Journal of Equine Veterinary Science* **33**, 421-426.

Godoi DB, Gastal EL, Gasta MO (2002) A comparative study of follicular dynamics between lactating and non-lactating mares: effect of the body condition. *Theriogenology* **58**, 553-556.

Gold HJ (1977) 'Mathematical modeling of biological systems: An introductory guidebook.' (Wiley: New York)

Grace N (2005) Pasture counts: the contribution of pasture to the diets of horses. *Advances in Equine Nutrition III* **3**, 11.

Grace ND, Shaw HL, Gee EK, Firth EC (2002) Determination of the digestible energy intake and apparent absorption of macroelements in pasture-fed lactating Thoroughbred mares. *New Zealand Veterinary Journal* **50**, 182-185.

Gu J, Orr N, Park SD, Katz LM, Sulimova G, MacHugh DE, Hill EW (2009) A genome scan for positive selection in Thoroughbred horses. *PLoS ONE* **4**, e5767.

Guay KA, Brady HA, Allen VG, Pond KR, Wester DB, Janecka LA, Heninger NL (2002) Matua bromegrass hay for mares in gestation and lactation. *Journal of Animal Science* **80**, 2960-2966.

Hanlon DW (2012) Reproductive performance and the transition period of Thoroughbred mares in New Zealand: Evidence and implications for future alternative management strategies: A thesis presented in partial fulfilment of the requirements for the degree of Doctor of Philosophy in Veterinary Science at Massey University, Palmerston North, New Zealand.

Hanlon DW, Stevenson M, Evans MJ, Firth EC (2012a) Reproductive performance of Thoroughbred mares in the Waikato region of New Zealand: 2. Multivariable analyses and sources of variation at the mare, stallion and stud farm level. *New Zealand Veterinary Journal* **60**, 335-343.

Hanlon DW, Stevenson M, Evans MJ, Firth EC (2012b) Reproductive performance of Thoroughbred mares in the Waikato region of New Zealand: 1. Descriptive analyses. *New Zealand Veterinary Journal* **60**, 329-334.

Harris P (2001) Comparison of digestible energy (DE) and net energy (NE) systems for the horse. *Advances in Equine Nutrition. Nottingham University Press, United Kingdom* 199-215.

Henneke D, Potter G, Kreider J (1984) Body condition during pregnancy and lactation and reproductive efficiency of mares. *Theriogenology* **21**, 897-909.

Hirst RL (2011) Seasonal variation of pasture quality on commercial equine farms in New Zealand : A thesis in partial fulfilment of the requirements for the degree of Master of AgriScience (Equine Studies) at Massey University, Palmerston North, New Zealand.

Hoskin SO, Gee EK (2004) Feeding value of pastures for horses. *New Zealand Veterinary Journal* **52**, 332-341.

Hsueh AJW, Adashi EY, Jones PBC, Welsh JTH (1984) Hormonal regulation of the differentiation of cultured ovarian granulosa cells. *Endocrine Reviews* **5**, 76-127.

Hughes TP, Gallagher JR (1993) 'Influence of sward height on the mechanics of grazing and intake by racehorses, International Grassland Congress.'

INRA (Ed. W Martin-Rosset (2015) 'Equine nutrition : INRA nutrition requirements, recommended allowances and feed tables.' (Wageningen Academic Publishers: Wageningen, Netherlands)

Jung HG, Mertens DR, Payne AJ (1997) Correlation of acid detergent lignin and Klason lignin with digestibility of forage dry matter and neutral detergent fiber. *Journal of Dairy Science* **80**, 1622-1628.

Kienzle E, Zeyner A (2010) The development of a metabolizable energy system for horses. *Journal of Animal Physiology and Animal Nutrition* **94**, e231-e240.

Kubiak JR, Crawford BH, Squires EL, Wrigley RH, Ward GM (1987) The influence of energy intake and percentage of body fat on the reproductive performance of non pregnant mares. *Theriogenology* **28**, 587-598.

Lamoot I, Callebaut J, Degezelle T, Demeulenaere E, Laquière J, Vandenberghe C, Hoffmann M (2004) Eliminative behaviour of free-ranging horses: Do they show latrine behaviour or do they defecate where they graze? *Applied Animal Behaviour Science* **86**, 105-121.

Lawrence LM, DiPietro J, Ewert K, Parrett D, Moser L, Powell D (1992) Changes in body weight and condition of gestating mares. *Journal of Equine Veterinary Science* **12**, 355-358.

Litherland AJ, Lambert MG (2007) Factors affecting the quality of pastures and supplements produced on farms. In 'Pasture and supplements for grazing animals ' (Ed. PV Rattray.) Vol. 14 pp. 81-96. (New Zealand Society of Animal Production, Occasional Publication New Zealand)

Lofstedt RM (1988) Control of the estrous cycle in the mare. *Veterinary Clinics of North America: Equine Practice* **4**, 177-196.

Longland A (2012) Nutritional assessment of forage quality. In 'Forages and grazing in horse nutrition.' pp. 65-82. (Wageningen Academic Publisher: Wageningen)

Lucy MC, Staples CR, Michel FM, Thatcher WW (1991) Energy balance and size and number of ovarian follicles detected by ultrasonography in early postpartum dairy cows. *Journal of Dairy Science* **74**, 473-482.

Mackenzie DJ, Wylam CB (1957) Analytical studies on the carbohydrates of grasses and clovers. VIII. Changes in carbohydrate composition during the growth of perennial rye-grass. *Journal of the Science of Food and Agriculture* **8**, 38-45.

Malacarne M, Martuzzi F, Summer A, Mariani P (2002) Protein and fat composition of mare's milk: some nutritional remarks with reference to human and cow's milk. *International Dairy Journal* **12**, 869-877.
Manso Filho HC, McKeever KH, Gordon ME, Costa HEC, Lagakos WS, Watford M (2008) Changes in glutamine metabolism indicate a mild catabolic state in the transition mare. *Journal of Animal Science* **86**, 3424-3431.
Mariani P, Summer A, Martuzzi F, Formaggioni P, Sabbioni A, Catalano AL (2001) Physicochemical properties, gross composition, energy value and nitrogen fractions of Haflinger nursing mare milk throughout 6 lactation months. *Animal Research* **50**, 415-425.
Marlow CHB, Van Tonder EM, Hayward FC, Van der Merwe SS, Price LEG (1983) A report on the consumption, composition and nutritional adequacy of a mixture of lush green perennial ryegrass (Lolium perenne) and cocksfoot (Dactylis glomerata) fed ad libitum to Thoroughbred mares. *Journal of the South African Veterinary Association* **54**, 155-157.
Martin-Rosset W, Vermorel M, Doreau M, Tisserand JL, Andrieu J (1994) The French horse feed evaluation systems and recommended allowances for energy and protein. *Livestock Production Science* **40**, 37-56.
McCue PM, Logan NL, Magee C (2007) Management of the transition period: hormone therapy. *Equine Veterinary Education* **19**, 215-221.
McGowan CM, Dugdale AH, Pinchbeck GL, Argo CM (2013) Dietary restriction in combination with a nutraceutical supplement for the management of equine metabolic syndrome in horses. *The Veterinary Journal* **196**, 153-159.
McMeniman NP, 2003. Pasture intake by young horses. Rural Industries Research and Development Corporation, Canberra, Australia.
Meyer H, Ahlswede L (1978) The intra-uterine growth and body composition of foals and the nutrient requirements of pregnant mares. *Animal Research and Development* **8**, 86-112.
Morley SA, Murray J (2014) Effects of body condition score on the reproductive physiology of the broodmare: A review. *Journal of Equine Veterinary Science* **34**, 842-853.
Mowat DN, Kwain ML, Winch JE (1969) Lignification and in vitro cell wall digestibility of plant parts. *Canadian Journal of Plant Science* **49**, 499-504.
Mueller WM, Gregoire FM, Stanhope KL, Mobbs CV, Mizuno TM, Warden CH, Stern JS, Havel PJ (1998) Evidence that glucose metabolism regulates leptin secretion from cultured rat adipocytes. *Endocrinology* **139**, 551-558.
Nagy P, Huszenicza G, Juhasz J, Kulcsar M, Solti L, Reiczigel J, Abavary K (1998) Factors influencing ovarian activity and sexual behavior of postpartum mares under farm conditions. *Theriogenology* **50**, 1109-1119.
Nagy P, Guillaume D, Daels P (2000) Seasonality in mares. *Animal Reproduction Science* **60**, 245-262.
Newcombe JR, Wilson MC (2005) Age, body weight, and pregnancy loss. *Journal of Equine Veterinary Science* **25**, 188-194.
Nicol AM, Brookes IM (2007) The metabolisable energy requirements of grazing livestock. In 'Pasture and supplements for grazing animals ' (Ed. PV Rattray.) Vol. 14 pp. 151-172. (New Zealand Society of Animal Production, Occasional Publication: New Zealand)
NRC (1981) 'Nutritional Energetics of Domestic Animals and Glossary of Energy Terms.' (The National Academies Press: Washington, DC)

NRC (2007) 'Nutrient requirement of horses.' (The National Academy Press: Wahington, DC)
NZTBA (2017) 'NZ Breeding, Sales & Export Statistics 1995-2017.' Available at https://www.nzthoroughbred.co.nz/page/nz-breeding-stats/
Ödberg FO, Francis-Smith K (1976) A study on eliminative and grazing behaviour - The use of the field by captive horses. *Equine Veterinary Journal* **8**, 147-149.
Oftedal O, Hintz H, Schryver H (1983) Lactation in the horse: milk composition and intake by foals. *Journal of Nutrition* **113**, 2196-2206.
Osborne VE (1966) An analysis of the pattern of ovulation as it occurs in the annual reproductive cycle of the mare in Australia. *Australian Veterinary Journal* **42**, 149-154.
Pagan JD, Hintz HF (1986) Equine Energetics. I. Relationship between body weight and energy requirements in horses. *Journal of Animal Science* **63**, 815-821.
Pagan JD, Brown-Douglas CG, Caddel S (2006) Body weight and condition of Kentucky Thoroughbred mares and their foals as influenced by month of foaling, season, and gender. *European Association for Animal Production* **120**, 245.
Pearson RA, Archibald RF, Muirhead RH (2006) A comparison of the effect of forage type and level of feeding on the digestibility and gastrointestinal mean retention time of dry forages given to cattle, sheep, ponies and donkeys. *British Journal of Nutrition* **95**, 88-98.
Platt H (1978) Growth and maturity in the equine fetus. *Journal of the Royal Society of Medicine* **71**, 658-661.
Platt H (1984) Growth of the equine foetus. *Equine Veterinary Journal* **16**, 247-252.
Ransom JI, Hobbs NT, Bruemmer J (2013) Contraception can lead to trophic asynchrony between birth pulse and resources. *PLoS ONE* **8**, e54972.
Reynolds LP, Ferrell CL, Robertson DA, Ford SP (1986) Metabolism of the gravid uterus, foetus and utero-placenta at several stages of gestation in cows. *The Journal of Agricultural Science* **106**, 437-444.
Rogers CW, Gee EK, Firth EC (2007) A cross-sectional survey of Thoroughbred stud farm management in the North Island of New Zealand. *New Zealand Veterinary Journal* **55**, 302-307.
Rosales C, Krekeler N, Tennent-Brown B, Stevenson MA, Hanlon D (2017) Periparturient characteristics of mares and their foals on a New Zealand Thoroughbred stud farm. *New Zealand Veterinary Journal* **65**, 24-29.
Salazar-Ortiz J, Camous S, Briant C, Lardic L, Chesneau D, Guillaume D (2011) Effects of nutritional cues on the duration of the winter anovulatory phase and on associated hormone levels in adult female Welsh pony horses (Equus caballus). *Reproductive Biology and Endocrinology* **9**, 130.
Salazar-Ortiz J, Monget P, Guillaume D (2014) The influence of nutrition on the insulin-like growth factor system and the concentrations of growth hormone, glucose, insulin, gonadotropins and progesterone in ovarian follicular fluid and plasma from adult female horses (Equus caballus). *Reproductive Biology and Endocrinology* **12**, 72.
Slade LM, Robinson D, Casey KE (1970) Nitrogen metabolism in nonruminant herbivores. I. The influence of nonprotein nitrogen and protein quality on the nitrogen retention of adult mares. *Journal of Animal Science* **30**, 753-760.
Smit HJ, Taweel HZ, Tas BM, Tamminga S, Elgersma A (2005) Comparison of techniques for estimating herbage intake of grazing dairy cows. *Journal of Dairy Science* **88**, 1827-1836.

Stanley T, Cochran R, Vanzant E, Harmon D, Corah L (1993) Periparturient changes in intake, ruminal capacity, and digestive characteristics in beef cows consuming alfalfa hay. *Journal of animal science* **71**, 788-795.

Sticker LS, Thompson DL, Fernandez JM, Bunting LD, DePew CL (1995) Dietary protein and (or) energy restriction in mares: Plasma growth hormone, IGF-I, prolactin, cortisol, and thyroid hormone responses to feeding, glucose, and epinephrine. *Journal of Animal Science* **73**, 1424-1432.

Storer WA, Thompson DL, Waller CA, Cartmill JA (2007) Hormonal patterns in normal and hyperleptinemic mares in response to three common feeding-housing regimens. *Journal of Animal Science* **85**, 2873-2881.

Thornley JHM, France J (2007) 'Mathematical models in agriculture : Quantitative methods for the plant, animal and ecological sciences.' (CABI: Wallingford, England)

Tylutki TP (2011) Fancy: An equine nutrition and management model. *Journal of Equine Veterinary Science* **31**, 211-218.

Valentine I, Kemp PD (2007) Pasture and supplement resources In 'Pasture and supplements for grazing animals, Ocassional Publication.' (Eds PV Rattray, IM Brookes, AM Nicol.) Vol. 14 pp. 3-11. (The New Zealand Society of Animal Production New Zealand)

Van Rijssen JD, Gee EK, Rogers CW, O'Brien JR (2010) 'Factors influencing gestation length and interval from foaling to conception in mares managed on a commercial Thoroughbred studfarm, Proceedings of the New Zealand Society of Animal Production.'

Van Weyenberg S, Sales J, Janssens GPJ (2006) Passage rate of digesta through the equine gastrointestinal tract: A review. *Livestock Science* **99**, 3-12.

Vermorel M, Martin-Rosset W, Vernet J (1997) Energy utilization of twelve forages or mixed diets for maintenance by sport horses. *Livestock Production Science* **47**, 157-167.

Waghorn GC, Burke JL, Kolver ES (2007) Principles of feeding value. In 'Pasture and supplements for grazing animals. .' (Ed. PV Rattray.) Vol. 14 pp. 35-59. (New Zealand Society of Animal Production, Occasional Publication: New Zealand)

Waite R, Boyd J (1953) The water-soluble carbohydrates of grasses. I.Changes occurring during the normal life-cycle. *Journal of the Science of Food and Agriculture* **4**, 197-204.

Waldron K, Rogers C, Gee E, Bolwell C (2011) 'Production variables influencing the auction sales price of New Zealand Thoroughbred yearlings, Proceedings of the New Zealand Society of Animal Production.'

Wastney ME (1999) 'Investigating biological systems using modeling: Strategies and software.' (Academic Press: San Diego)