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EQUINE SCIENCE 2012

"Research for the 21st Century Horse Industry: From Genomics to the Winning Post"

The inaugural Australian Equine Science Symposium arose from discussions that were held at the Magic Millions Thoroughbred sales in January 2006. Following initial support from RIRDC, a committee was formed and Equine Science 2006 planned. Following the success of the initial Symposium it was decided by those at the meeting to hold a biennial conference for equine scientists which:

- Promotes excellence in equine science
- Focuses on science, technology and innovation relevant to the Australian horse industry
- Provides a regular forum for exchange of research findings, ideas and information between Australian & New Zealand equine scientists and with their international colleagues
- Assists young equine scientists with their careers
- Encourages participation by members of the horse industry

At the second Symposium a decision was made by the participants to form an Australasian Equine Science Society and the name of the Symposium changed accordingly.

An additional dimension has been added to AESS 2012, with the incorporation of a satellite meeting of the National Association of Equine Affiliated Academics (NAEAA) into the fourth Symposium.

Proceedings

Papers presented at this Symposium have been peer reviewed. However, the comments and views expressed in the papers are entirely the responsibility of the author or authors concerned.

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CARBOHYDRATES ARE WELL TOLERATED BY MOST HORSES

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The old horse feeding slogan "hay, oats and water" harkens back to a simpler time when horses performed free of feed supplements, drugs and anabolic steroids. It also is a reminder that traditionally cereal grains have been staple feeds for horses. For generations, work horses, racehorses and show horses have been fueled by rations composed mainly of forage and grains. Cereal grains are high in non-structural carbohydrates (NSC). NSC include starch and water-soluble sugars that can be enzymatically digested and absorbed as glucose in the small intestine.

Recently, the use of cereals as a major component of horse feed has been questioned by a small but vocal group of zealots who have used the Internet to effectively voice their opinions. Their concern is based on the somewhat flawed premise that feeding even moderate quantities of NSC will lead to behavior or metabolic disturbances and disease in clinically normal horses. Nonetheless, this has led many feed manufacturers to drastically alter their horse feed formulations to cater to the "low-carb" craze. If horses need a low starch feed, hay is a readily available alternative!

Of course, there are legitimate concerns about feeding excessive NSC to horses. Feeding too much starch in a single meal can overwhelm the small intestine's digestive capacity, resulting in large quantities of starch escaping to the large intestine, where it is rapidly fermented to volatile fatty acids (VFAs) and lactic acid. Changes in the pH of the hindgut due to alterations in the microbial populations and acid profiles may result in hindgut acidosis (HGA). Horses suffering from HGA may develop loose manure, anorexia, behaviour changes, colic or display stereotypical behaviors such as wood chewing and stall weaving. HGA is one of the predisposing factors in laminitis. Furthermore, long-term exposure to pH levels below 5.8 will begin to have deleterious effects on the epithelial lining of the colonic and caecal walls that may affect absorptive capacity. HGA can also result from rapid consumption of grains containing poorly digested starch eg unprocessed wheat, corn or barley or from pastures with high fructan levels. Temperate grasses often have high levels of fructans which are resistant to digestion in the small intestine.

With good feeding management, carbohydrates in grains are well tolerated by most horses. There is, however, a small but important population of horses that do not tolerate carbohydrates well. Knowing how much carbohydrate should be included in a particular horse's daily ration is key to successful feeding.

First some background on carbohydrates in horse feeds. The carbohydrates in equine feeds can be categorised by either their function in the plant or from the way they are digested and utilised by the horse. From a plant perspective, carbohydrates fall into three categories: (1) simple sugars active in plant intermediary metabolism; (2) storage compounds such as sucrose, starch, and fructans; and (3) structural carbohydrates such as pectin, cellulose, and hemicellulose. For the horse, it is more appropriate to classify carbohydrates by where and how quickly they are digested and absorbed. Carbohydrates can either be digested and/or absorbed as monosaccharides (primarily glucose and fructose) in the small intestine, or they can be fermented in the large intestine to produce volatile fatty acids (VFA) or lactic acid. The rate of fermentation and types of end products produced are quite variable and can have significant effects on the health and well-being of the horse.

A physiologically relevant system to categorise carbohydrates in equine diets would be composed of three groups. (1) A hydrolysable group (CHO-H) measured by direct analysis that yields sugars (mainly glucose) for metabolism. This includes simple sugars, sucrose, and some starches that are readily digested in the small intestine and produce fluctuations in blood glucose after feeding. (2) A rapidly fermentable group (CHO-FR) that yields primarily lactate and propionate. This group includes starches that escape digestion in the small intestine as well as galactans, fructans, gums, mucilages, and pectin. (3) A slowly fermentable group (CHO-FS) that yields mostly acetate and butyrate. This group includes the compounds captured in neutral detergent fibre (NDF) such as cellulose, hemicellulose, and lignocellulose. Hydrolysable carbohydrates (CHO-H) are an important component of equine diets, particularly for the performance horse, where blood glucose serves as a major substrate for muscle glycogen synthesis. Too much blood glucose, however, may contribute to or

aggravate certain problems in horses such as equine Cushing's disease (ECD), equine metabolic syndrome (EMS), some developmental orthopedic diseases (DOD), recurrent equine rhabdomyolysis (RER), and polysaccharide storage myopathy (PSSM). It may also adversely affect behavior in certain individuals.

The quantity of blood glucose produced in response to a meal is a useful measure of a feed's CHO-H content. Table 1 contains the glycemic index of several equine feeds measured at Kentucky Equine Research (KER). Glycemic index characterises the rate of carbohydrate absorption after a meal and is defined as the area under the glucose response curve after consumption of a measured amount of a test feed divided by the area under the curve after consumption of a reference meal, in this case oats. Rapidly fermentable carbohydrates (CHO-FR) such as pectin can yield propionate, which is an important gluconeogenic substrate for the horse. However, rapid fermentation can also produce lactic acid, which may lead to a cascade of events culminating in laminitis. Undigested starch from cereals and fructans from pasture are the most likely compounds contributing to lactic acidosis in the hindgut. Slowly fermentable carbohydrates (CHO-FS) from the plant cell wall are absolutely essential to maintain a healthy microbial environment in the horse. These carbohydrates alone, however, may not be able to supply enough energy to fuel a high-performance athlete. Carbohydrates in horse feeds have traditionally been estimated by measuring cell wall components as NDF and calculating the remaining carbohydrate by difference as non fibre carbohydrate (NFC), where NFC = 100 - water - protein - fat - ash - NDF. More recently, laboratories have provided a direct analysis of additional carbohydrates in equine feeds.

Table 1. Glycemic index (GI) of equine feeds and forages.

Feed	Glycemic Index
Sweet feed	129
Whole oats	100
Equine Senior®	100
Beet pulp + molasses	94
Cracked corn	90
Re-Leve®	81
Beet pulp (unrinsed)	72
Orchard grass hay	49
Rice bran	47
Ryegrass hay	47
Alfalfa hay	46
IR Pellet & Orchard grass hay	34
Rinsed beet pulp	34
Bluestem hay	23

Table 2 contains the chemical composition of several common equine feedstuffs as analysed by Equi-Analytical Laboratories in Ithaca, NY. In addition to NDF and the calculated values of NFC, Table 2 contains measured levels of water-soluble sugars (WSC) and starch. The sum of WSC and starch is considered the nonstructural carbohydrate (NSC). WSC in cereal grains and by-products such as beet pulp are composed of simple sugars that produce a pronounced glycemic response and fit into the CHO-H category. By contrast, much of the WSC in temperate grasses are actually fructans, which should be included in the CHO-FR fraction. Therefore, they would have little effect on glycemic response but may contribute to the development of hindgut acidosis and laminitis.

Table 2. Carbohydrate content of some common equine feeds.

	Oats	Corn	Beet pulp	Soy hulls	Legume hay	Grass hay
WSC (%)	3.9	3.5	10.6	3.6	9.0	10.7
Starch (%)	44.3	70.5	1.3	1.7	2.4	2.8
NSC (%)	50.7	73.1	12.1	5.3	11.4	13.3
NFC (%)	50.9	76.4	44.4	19.8	30.8	19.5
NDF (%)	27.9	9.8	41.9	61.7	38.5	63.8

Starch is the predominant carbohydrate fraction in cereal grains. Although all starch is made up of glucose chains, how the starch molecule is constructed varies in different types of grain. These differences in the architecture of individual starches have a large impact on how well they are digested in the horse's small intestine. Of the grains most commonly fed to horses, oats contain the most digestible form of starch, followed by sorghum, corn, and barley. Processing can have a huge effect on precaecal digestibility, particularly in barley, sorghum and corn. In a KER study, steam flaking corn caused a 48% increase in glycemic response compared to coarse cracking. NSC is a mixture of CHO-H and CHO-FR fractions. NSC tends to be higher in CHO-H in processed cereal grains and mixes but may be high in CHO-FR in certain unprocessed cereals or high-fructan forages. NFC represents an even more mixed group of carbohydrates because in addition to the compounds found in NSC, they may also contain significant quantities of pectin and other fermentable compounds not captured in NDF. For instance, beet pulp contains only 12.1% NSC but 44.4% NFC. At present, there is no satisfactory, commercially available analytical method to segment carbohydrates into categories that are physiologically meaningful for the horse.

Performance horses depend on NSC as a major source of dietary energy. Digestion of NSC results in increases in blood glucose. Under the influence of insulin, blood glucose is taken up by the liver, muscle and adipose tissue, and stored as liver glycogen, muscle glycogen or fat. These substrates are later used as fuels for muscle contraction during exercise. Kentucky Equine Research (KER) has evaluated the rations of hundreds of sport horses and racehorses competing successfully at the highest level. The vast majority of these horses consume feeds that contain 30-40% NSC, which supplies 35-50% of the concentrate's digestible energy (DE) content. This is lower than the 44-65% NSC found in straight cereal grain diets since modern performance feeds also derive a significant quantity of DE from fat and fermentable fibre. A typical high-performance ration (forage + concentrate) contains 18-22% NSC, which provides 28-32% of the ration's total DE.

At the other end of the equine spectrum are horses that cannot tolerate even moderate amounts of NSC in their rations. These metabolic disorders may becoming more widely recognized but still only represent a small part of the equine population. Many of these disorders such as ECD, EMS, OCD, RER and PSSM can be managed nutritionally by careful regulation of caloric intake with particular attention paid to the source of energy provided. Although these disorders have very different aetiologies, they are all either triggered or aggravated by excessive starch and sugar intake.

The most prevalent of these disorders is EMS, which results in insulin resistance (IR) and an increased risk of laminitis. Horses and ponies with EMS tend to be obese with cresty necks. These animals often have had prior bouts of laminitis and are easy keepers. Management strategies to reduce the incidence and severity of EMS include exercise, weight loss and a ration that contains no more than 10% NSC.

In between the extremes of the elite equine athlete and the obese, laminitic pony lies the majority of horses in the population. These horses are clinically normal, and while many may be older and sedentary, they have not displayed any signs of EMS. Will a carbohydrate-rich ration make these horses insulin resistant and thus more susceptible to EMS as suggested by the "low-carb" zealots? Are high-fat, low-carbohydrate diets more suitable?

To answer these questions KER recently conducted a study with aged Thoroughbred geldings (21.5 yr ± 3.32 yr; BCS 5.0-6.0) to determine whether an oat-based ration (20% NSC, 31% DE from NSC) or a high-fat ration (12.7% fat, 30% DE from fat) would affect glucose tolerance as compared to an all grass-hay diet (9.4% NSC, 17% DE from NSC). Glucose tolerance was measured using an intravenous glucose tolerance test (IVGTT). In this test, a solution of glucose is infused into the horse and blood samples are taken over a 6-hour period. The amount of time it takes for blood glucose to return to normal indicates how well the horse's liver, muscle and fat cells are able to take up and utilize glucose. Horses with impaired glucose tolerance take longer for blood glucose to return to baseline. Each dietary period of the Latin square design study lasted 28 days. During the IVGTT it took significantly less time (126.6 ± 25.8 min.) for blood glucose to return to baseline in the oat-fed horses compared to either the all-hay (198.4 ± 40 min.) or the high-fat-fed horses (216.7 ± 23.5 min.). These results suggest that feeding normal, non-obese horses a ration with a significant quantity of its calories coming from NSC isn't detrimental and may even improve glucose tolerance.

For obese horses, the level of energy intake is more important than the source of calories. Obesity and lack of exercise are the two predisposing factors for IR and EMS. The main goal of feeding an obese horse is to reduce its caloric intake below its caloric requirement so that it will burn body fat and lose weight. Concentrates containing higher fibre and lower fat and NSC are less calorically dense and can be used to deliver fewer calories without drastically reducing feed intake. Feeding hay and a low-intake balancer pellet is also a good alternative for obese horses. As a management tool for obesity, increasing exercise is just as important as lowering caloric intake since exercise increases caloric expenditure and has been shown to improve insulin sensitivity, even in overweight horses.

In conclusion, horses with specific metabolic issues certainly benefit from low (<10%) NSC diets, but these types of "super low-carb" diets are not necessary for normal sedentary or exercised horses. Concentrates that contain 20-30% NSC and that produce a lower glycemic response than higher NSC performance feeds may be useful for sedentary and lightly exercised horses and for horses that tend to become more excitable on higher glycemic feeds. Heavily exercised performance horses need more NSC in their diets, and cereal grains and grain by-products remain important and cost-effective sources. Inadequate starch and sugar intake in hard working horses fed diets with most energy coming from fat and fibre may lead to delayed replenishment of muscle glycogen stores and slow recovery from competition or training. To paraphrase Mark Twain, "The news of grain's demise in horse feed has been greatly exaggerated."

EQUINE PARENTAGE VERIFICATION: SNiPS or STaRs?

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Independent genetic verification of Thoroughbred parentage is a critical condition for acceptance into the Australian Stud Book, and all Thoroughbred Stud Books recognised by the International Stud Book Committee, and is a prerequisite to naming, sale, racing and breeding of Thoroughbred horses. Similarly, many other horse Breed Societies and Registries require DNA testing to verify foal parentage prior to breed registration.

Currently, equine parentage is verified by testing a minimum of 12 internationally agreed Simple Tandem Repeat DNA markers (STaRs), known as the Equine Primary Panel of STR DNA markers. Each equine STaR DNA marker is a repeating unit of two nucleotides, eg. CACACACACA…. The number of STaR repeat units vary, producing multiple different variants (alleles) of each STaR DNA marker in the horse population. The horse STaR markers have between 4 and 12 different alleles in the horse population. The multi-allele nature of STaR markers means that the parentage of 99% of foals can be verified using the 12 internationally agreed equine STaR DNA markers. Also, the ambiguous parentage of the remaining 1% of foals can be resolved by genotyping an additional 12-15 equine STaR DNA markers.

Completion of the horse genome sequence has characterised thousands of single nucleotide polymorphisms (SNiPs) distributed across the 31 chromosomes, X and Y chromosomes that comprise the horse genome. Serious consideration is being given to using SNiPs, as a source of genetic variation, to confirm parentage in horses. SNiPs exist in one of two alternative states, eg. either T or G, in the horse population. The two-allele nature of SNiPs means that at least 300 SNiPs would have to be genotyped, in the Sire, Dam and Foal, to achieve the same level of confidence in horse parentage verification as is currently achieved using the 12 internationally agreed STaR DNA markers. So genotyping 300 SNiPs would verify the parentage of 99% of foals. A further 300 SNiPs would have to be genotyped to resolve the parentage of the remaining 1% of foals.

Using SNiPS for parentage verification would allow the simultaneous genetic testing for parentage verification and many, but not all, inherited conditions in horses, mostly diseases and coat colour and/or pattern traits. This could be achieved by including the causative mutations in the SNiP parentage panel. This is not currently the case. SNiP-based parentage would also provide more detailed genetic profiles of different horse populations, such as Thoroughbreds compared to Quarter Horses.

Converting to SNiP-based parentage verification for horses would also require substantial investment in: new capital equipment to support high-throughput genotyping of a panel of 300 SNiPs; new genetic database systems to store, analyse and report parentage results based on 300 to 600 SNiP genetic loci (as compared to the 12 to 27 STaR genetic loci analysed now); establishment of new international agreements and horse genotyping comparison tests to ensure international compatibility of horse parentage DNA profiles, essential for the international horse breeding industry; and, not least, re-genotyping of the current breeding population of horses with a new SNiP parentage panel. These, and other factors will be discussed in considering the question of whether horse parentage verification should change from STaRs to SNiPs.

Stud Books, Breed Societies and Owners expect 100% accuracy, 100% of the time in the results of parentage analyses and genetic screening tests. A major change in the genetic basis of horse parentage analysis, such as changing from STaRs to SNiPs, comes with an increased risk of a parentage analysis error. In an industry where the value of a foal is significantly determined by its parentage, do the potential benefits of SNiPs out way the current benefits of STaRs?

DETERMINATION OF THE PHARMACOKINETICS OF EIGHTEEN EQUINE MEDICATIONS BY EQUINE THERAPEUTICS RESEARCH AUSTRALIA (ETRA)

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Equine Therapeutics Research Australia (ETRA) is a consortium of three Australian Universities and the four Australian Racing Laboratories. The primary role of ETRA is to determine reliable excretion times for equine medications, so that the legitimate use of these substances can occur with less risk of positive drug tests. ETRA is also working with international authorities to try to achieve the harmonisation of drug detection limits in Australia, Europe and Asia.

The Australian rules of racing and other equestrian events forbid horses from competing if their blood or urine contains detectable concentrations of any therapeutic substance on the day of competition. Unfortunately, there has been uncertainty about excretion and detection times for many important equine medications. ETRA has sought to address this problem by evaluating 18 medications over a four year period. The medications tested were chosen following consultation with a reference group of peak industry bodies: Equine Veterinarians Australia, The Australian Horse Industry Council, The Australian Racing Board, The Equestrian Federation of Australia, Harness Racing Australia, Queensland Racing, Racing and Wagering Western Australia, Racing New South Wales and Racing Victoria.

The aims and objectives of the project were to:

- identify the most important therapeutic substances used in horses;
- administer these compounds to groups of not fewer than 10 horses, collect plasma and urine samples and analyse these for the presence of the parent drugs and their metabolites;
- model the analytical data, presenting a clear summary of the pharmacokinetics and rate of excretion of each compound for the benefit of veterinarians and racing authorities;
- explore a new approach to the reporting of detection times based on a probability model which takes into account variation among horses.

The test substances (Tables 1 and 2) were selected after widespread consultation with industry stakeholders and veterinarians. The drugs were administered to groups of 10 to 12 horses housed either at Charles Sturt University, NSW, or The University of Queensland, Gatton, Qld. Blood and urine samples were collected at frequent intervals over an appropriate period of time, ranging from 5 days to 100 days depending on the predicted excretion time of the drug. Over all the administrations more than 20,000 blood and urine samples were collected. Eighteen drug administration trials have been performed and Table 1 lists those compounds which have been administered, analysed and have had pharmacokinetics completed. Table 2 lists the compounds which have been administered, but which are still undergoing chemical analysis by the racing laboratories.

Table 1 Twelve therapeutic substances administered to groups of 10 or 12 horses (n) to determine their pharmacokinetics in blood and excretion times in urine

Class	Compound	Route*	Dose	n
Non-steroidal anti-inflammatory drugs	Phenylbutazone	oral	2 g twice daily for 1 day then 1 g twice daily for 4.5 d (1.5 to 2 mg/kg twice daily)	12
	Flunixin	IV	1.1 mg/kg	12
	Ketoprofen	IV	1 g/horse (1.86 to 2.29 mg/kg)	12
Corticosteroids	Hydrocortisone	IV	1 g/horse (1.55 to 2.02 mg/kg)	10
	Prednisolone	oral	1 g/day for 5 days (~2 mg/kg)	12
	Methylprednisolone	IM	200 mg/horse (0.39 to 0.47 mg/kg)	12
Sedatives	Acepremazine	IV	30 mg/horse (0.045 to 0.063 mg/kg)	12
	Detomidine	IV	0.04 mg/kg	12
Antispasmodics	Buscopan	IV	30 mL/horse (120 mg HBB, 0.21 to 0.27 mg/kg and 15 g dipyrone, 26 to 33 mg/kg)	12
Local anaesthetics	Mepivacaine	SC	400 mg/horse (0.68 to 0.99 mg/kg)	12
	Lignocaine	SC	0.8 mg/kg	12
	Prilocaine	SC	400 mg/horse (0.84 to 0.60 mg/kg)	12

*IV – intravenous injection, IM – intramuscular injection, SC – subcutaneous injection.

Table 2 Four therapeutic substances administered alone or in combination to groups of 12 horses (n) to determine their pharmacokinetics in blood and excretion times in urine – currently under investigation

Class	Compound	Route*	Dose	n
Corticosteroids	Dexamethasone	IV	0.06 mg/kg	12
Analgesics	Butorphanol	IV	20 mg/horse	12
Tranquilizers	Xylazine	IV	400 mg/horse	12
	Xylazine + butorphannol	IV	250 mg xylazine/horse 10 mg butorphanol/horse	12
Antibiotics	Procaine penicillin (1d)	IM	12 mg/kg single administration	12
	Procaine penicillin (5d)	IM	12 mg/kg daily for 5 days	12

*IV –intravenous injection, IM – intramuscular injection

All blood and urine samples from the administrations were transported to one of the four official Australian racing forensic laboratories where they were subjected to rigorous chemical analysis. Validated analytical methods and advanced instrumentation (usually LC-MS or GC-MS) were used. The pharmacokinetic data were analysed using WinSAAM software, University of Pennsylvania, PA, USA) and STATA Statistical Software 11.1 (Stata, 2007) . A summary of the results appear in Table 3

Table 3 Peak plasma concentrations and excretion times for 12 equine medications

Compound	Route[a]	Dose	n[b]	T_{max}[c]	E[d]
Phenylbutazone	oral	2 g twice daily for 1 day then I g twice daily for 4.5 d (1.5 to 2 mg/kg twice daily)	12	4.3 h	>9 days
Flunixin	IV	1.1 mg/kg	12	5 min	>10 days
Ketoprofen	IV	1g/horse (1.86 to 2.29 mg/kg)	12	5 min	3 days
Hydrocortisone	IV	1 g/horse (1.55 to 2.05 mg/kg)	10	18 min	36 h
Prednisolone	oral	1 g/day for 5 days (~2 mg/kg)	12	40 min	5 days
Methylprednisolone	IM	200 mg/horse (0.39 to 0.47 mg/kg)	12	36 min	10 weeks
Acepromazine	IV	30 mg/horse (0.045 to 0.063 mg/kg)	12	5 min	>6 days
Detomidine	IV	0.04 mg/kg	12	3 min	3 days
Buscopan	IV	3 mL/horse (120 mg HBB, 0.21 to 0.27 mg/kg and 15 g dipyrone, 26 to 33 mg/kg)	12	5 min	13 days
Mepivacaine	SC	400 mg/horse (0.68 to 0.99 mg/kg)	12	1 h	>4 days
Lignocaine	SC	0.8 mg/kg	12	30 min	5 days
Prilocaine	SC	400 mg/horse (0.84 to 0.60 mg/kg)	12	35 min	3 days

[a]IV – intravenous injection, IM – intramuscular injection, SC – subcutaneous injection; [b]n – number of horses used; [c]T_{max} – time after administration at which plasma concentrations of the parent drug peak; [d]E – time after administration at which no drug or metabolite was detected in blood or urine in any horse (nb – does **not** represent official detection time).

A detailed account of the ETRA pharmacokinetic studies appear in the RIRDC report compiled by Sillence *et al* (2012).The results of this report will allow veterinarians to develop improved therapeutic protocols which will benefit the many thousands of horses across Australia treated each year for illnesses and injuries. In addition, these data will assist the racing authorities in setting more appropriate, accurate and reliable detection and reporting times for therapeutic substances. This information will subsequently benefit the veterinarians, owners and trainers associated with more than 650,000 starters in Thoroughbred racing, harness races and other equestrian pursuits in Australia each year.

Finally, as the information will be available to racing jurisdictions world-wide, this will assist Asian, European and American authorities in avoiding duplication of effort and moving towards a harmonised approach to drug detection.

Sillence M, Noble G, Schneiders F, Bryden W, Cawdell-Smith J, de Laat M, Jarrett M, Young B, McKinney A, Cawley A, Booth J, Vine J, Glowacki L, McGree J, Boston R, Nelis S, Kirkpatrick C, Shaw N, and Smyth B (2012*). **The Pharmacokinetics of Equine Medications***, A Report 11/117. Rural Industries Research and Development Corporation, Canberra, ACT ; 98p

STATUS OF THE HENDRA VIRUS VACCINE: PROTECTION FOR HORSES AND HUMANS

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The *henipaviruses* Hendra virus (HeV) and Nipah virus (NiV) are distinguished from all other *paramyxoviruses* by their broad species tropism and ability to infect and cause fatal disease in humans, horses, monkeys, pigs, cats, dogs, ferrets, hamsters and guinea pigs.

The use of safe efficacious vaccines against several important viral pathogens is a mainstay of human and animal infection prevention strategies. HeV spillover events have occurred with increasing frequency since 1994, and because of high mortality rates in horses and humans, implementation of a strategy to prevent infection in horses by active immunization is important to provide protection to humans, and a level of protection for the entire horse industry.

As with all *paramyxoviruses* the attachment G, and fusion F glycoproteins on the surface of the virion are the principle antigens to which virtually all neutralizing antibodies are directed. Previous immunizing studies in cats and ferrets as experimental models have demonstrated the potential for complete protection from live virus challenge and importantly for the ability of subunit vaccines to prevent viral replication and shedding in challenged individuals. The soluble form of HeV G glycoprotein, HeV-sG, has demonstrated remarkable ability to achieve this and to be cross protective against NiV. In very recent studies HeV-sG vaccination of nonhuman primates followed by intratracheal HeV or NiV challenge has also been conducted, and complete protection from Henipavirus induced disease was achievable with no recoverable virus or evidence of virus shedding being noted.

One of the major clinical recommendations made by human and veterinary health professionals at the 2009 International Henipavirus Workshop held in Queensland, and co-sponsored by the World Health Organisation, was that "Vaccination for horses should be prioritised for Hendra virus infection control" (Reid,2010).

Federal and State Government funding for vaccination experiments with horses at the Australian Animal Health Laboratory (AAHL) was finally achieved in mid-2010 and commenced in October 2010. HeV-sG in a high and low dose antigen formulation was used to immunize horses (prime/boost two dose regime, 3 week interval) and horses subsequently received a lethal virus challenge under BSL-4 laboratory conditions. All vaccinated horses remained clinically disease free and there was no evidence of virus replication or virus shedding in any of the immunized horses.

The HeV-sG as a subunit vaccine is now in commercial development as a livestock vaccine and currently under testing as an equine vaccine. There is a possibility that following successful completion of safety and immunogenicity trials that a form of vaccine may be available under an emergency permit approval in 2012. Final registration approval of the vaccine is unlikely before 2013.

The significant efforts to date have been a result of a collaborative research program between the Uniformed Services University, Bethesda Maryland, USA, the Henry M Jackson, Foundation, AAHL and a corporate pharmaceutical partner.

Reid P A (2010) Working towards a Hendra virus vaccine. *Aust Vet J* ;88(5): N4

EQUINE METABOLIC SYNDROME: CAUSES AND CONSEQUENCES

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Equine Metabolic Syndrome (EMS) is becoming recognized as a common condition, particularly affecting ponies and certain breeds of horses. EMS is a specific phenotype which includes the following:

- Increased adiposity in specific locations (regional adiposity) or generally (obesity).
- Insulin resistance characterized by hyperinsulinemia or abnormal glycemic and insulinemic responses to oral or intravenous glucose and/or insulin challenges.
- A predisposition towards laminitis. Clinical or subclinical laminitis that has developed in the absence of recognized causes such as grain overload, colic, colitis, or retained placenta.

The condition was named after human metabolic syndrome, which is a collection of risk factors (including insulin resistance and increased adiposity) associated with an increased risk of cardiovascular disease. As with human metabolic syndrome, there may be other elements to EMS which play a role in this phenotype; such as dyslipidaemia, hyperleptinaemia, vascular dysfunction and chronic low-grade inflammation associated with obesity.

Causes: pathophysiology

Insulin resistance (IR) involves defects of insulin signalling such as reduced insulin receptor tyrosine kinase activity and reduced post-receptor phosphorylation steps that impinge on the metabolic and vascular effects of insulin. Obesity has been linked to the development of IR in humans, and there are two primary theories linking obesity to IR: 1) the down-regulation of insulin signaling pathways induced by adipokines and cytokines produced in adipose tissue (see above); and 2) the accumulation of intracellular lipids in insulin-sensitive tissue such as skeletal muscle (lipotoxicity). However, development of IR may often be independent of obesity in horses and ponies, and could potentially be associated with frequent and sustained release of high levels of insulin into the pancreas (triggered by lush pasture or feeds with a high glycaemic index in sensitive animals) which causes receptor downregulation at the target organs.

In the context of domesticated equids experiencing a chronic state of overnutrition, natural seasonal changes in body condition and insulin sensitivity may be replaced by progressive obesity and IR with associated adverse health consequences. Adipose tissue is no longer regarded as just an energy storage organ, but an endocrine organ producing many hormones (adipokines or adipocytokines). Production of inflammatory adipokines may lead to a self perpetuating cycle of enhanced adipose tissue inflammation, adipokine synthesis and secondary acute phase protein synthesis by the liver. (Obesity in people is characterized by a state of chronic low-grade inflammation). Few data are available on the pathophysiological effects of obesity or regional adiposity in EMS. A cresty neck score has been developed to assess the expansion of adipose tissues within the neck region, and it has been hypothesized that nuchal crest fat may be a particular source of these cytokines; although recent evidence is providing less support to this theory.

Consequences: linking IR to Laminitis

The link between insulin resistance and laminitis is still unclear. Hyperinsulinemia may be the key factor, since it has been shown that prolonged infusion of insulin can induce laminitis. It is unclear whether insulin may have a direct or indirect effect on the lamellar tissues. However, insulin is structurally related to insulin-like growth factor (IGF-1), and can bind the IGF-1 receptor, albeit at lower affinity than IGF-1. IGF-1 receptors are present on lamellar epidermal epithelial cells, and insulin may have a proliferative effect. This effect would be consistent with some of the changes observed in insulin-induced laminitis.

Other potential mechanisms linking IR and laminitis include endothelial cell dysfunction and vasoconstriction within blood vessels of the foot, impaired glucose uptake by epidermal laminar cells and matrix metalloproteinase activation by glucose deprivation or reactive oxygen species. Whatever the cause, it is becoming evident that ‘endocrinopathic laminitis’ may account for a large number of naturally occurring laminitis cases. Therefore understanding these mechanisms is important for determining strategies to prevent and treat laminitis.

RATE OF GAIN, FEED EFFICIENCY AND GASTRIC ULCERS IN GROWING HORSES ARE AFFECTED BY FEED PROCESSING

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Equine gastric ulcer syndrome (EGUS) has been a difficult challenge to the horse industry. Horse producers and managers attempt to achieve optimal performance in the show ring or race track, and rapid growth and development in young horses without being at odds with general health and well-being. Unfortunately, some of the methods for achieving high-level performance in horses include the feeding of high-concentrate diets, the use of non-steroidal anti-inflammatory drugs, and generally stressful housing and training environments. Each of these practices has been found to lead to an increased occurrence of gastric ulcers in horses.

Nutrition research using growing horses (weanlings and yearlings) has found that feeding total mixed rations (TMR) may improve growth and feed efficiency of these young animals, but the effects on stomach ulcers may be dependent on both the concentration of grains versus forage in the TMRs and the amount of processing of the components (pelleted versus cubed). A completely pelleted TMR diet (CP) with 50% grain and 50% forage appears to be equally bad or slightly worse in causing endoscopically-detected stomach ulceration than a nutritionally identical diet of 50% longer-stemmed forage and 50% grain fed separately (HG). However, a TMR diet of hay and oats in cubed form (C) using only 25% oats was found to cause no endoscopically-detected stomach ulcers, while still providing a growth advantage (P<.05) over a nutritionally identical diet fed as plain hay cubes and grain fed separately (HG).

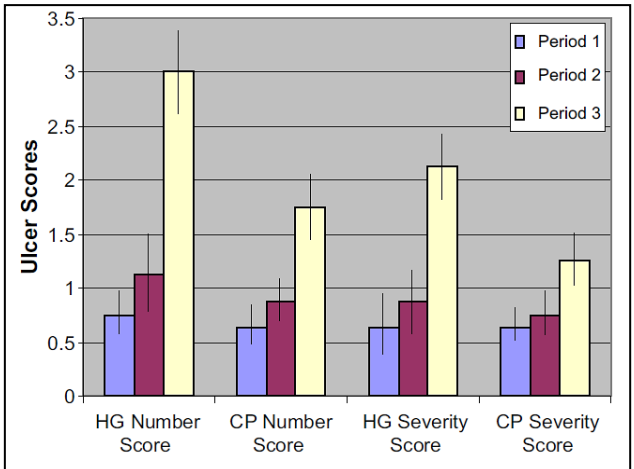
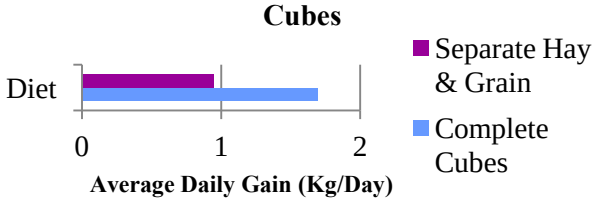


Fig. 1. Number and severity ulcer scores (±SEM) for groups on hay and grain diet (diet HG) and complete pelleted diet (diet CP) in periods 1, 2, and 3. Both groups on HG and CP consumed only alfalfa hay during period 1, and then group HG switched to diet HG in period 2 and diet CP in period 3. Group CP switched to diet CP in period 2 and diet HG in period 3.

Figure 2– Average Daily Gain of Yearlings Consuming Hay Cubes/Grain Separately vs. Complete Cubes



Horses consuming the TMR (diet C) had an ADG (Kg/day ± SEM) of 1.69 ± .79 while horses consuming the separate hay and grain (diet HG) horses had an ADG of 0.95 ± .18 (Figure 2). Diet C horses had a feed efficiency (G/F ± SEM) of .09 ± .04 while diet HG horses had a feed efficiency of .05 ± .01. ADG was found to be significantly greater (P=0.046) for diet C, while G/F between the two diets was not significantly different (P=0.065) but showed a trend in favor of diet C. There were no significant differences between the two diets in DM or CP digestibility. Diet C's DM digestibility was 64.04 ± 6.74 while diet HG′s was 66.80 ± 5.76. Diet C's CP digestibility was 58.29 ± 3.94 while diet HG′s was 57.85 ± 2.54. Endoscopies found only one horse with a single grade 1 ulcer severity score. The results support the additional hypothesis that neither diet would cause significant gastric ulceration. It is concluded that a complete cube diet of 75% hay (80% alfalfa hay, 20% tall fescue) and 25% oats does not cause stomach ulceration while achieving an acceptable growth rate in yearling horses.

THE ROLE OF THE FOAL IN THE EPIDEMIOLOGY OF RHODOCOCCAL DISEASE IN AUSTRALIA.

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Disease caused by *Rhodococcus equi* remains a significant problem for the horse breeding industry in Australia and worldwide. Pyogranulomatous bronchopneumonia and gastrointestinal abscessation are the most common disease manifestations caused by *R. equi* in foals in Australia. Disease management and diagnosis of rhodococcal pneumonia will improve through a better understanding of the age-specific responses to rhodococcal infections in the foal.

The foal plays an important role in the ecology and epidemiology of rhodococcal disease, both as a replicator of virulent *R. equi* within the gastrointestinal tract (GIT) and a disseminator of virulent *R. equi* into the external environment in faeces and exhaled breath. Recently, increased attention has been given to the potential for rhodococcal pneumonia to be not only an infectious but also a contagious disease in foals, with the possibility of foal to foal spread of virulent organisms in exhaled breath. Variation in the immune response of foals, after exposure to environmental virulent *R. equi*, is thought to be an important factor in determining the risk of development of disease in individuals. The effectiveness of the immune response is likely to be influenced by the age of the foal and the associated level of exposure to virulent *R. equi*.

Ecological and epidemiological studies in Australia have demonstrated that high airborne levels of virulent *R. equi* are associated with high disease prevalence on farms. More recent studies have also shown age-related differences in the level of virulent *R. equi* detectable in faeces and exhaled breath of foals. Six week old foals were a significant source of environmental contamination with virulent *R. equi*, shedding higher concentrations of virulent *R. equi* than three week old foals. Neonatal foals had minimal contamination of virulent *R. equi* throughout their GIT at birth but were capable of replicating *R. equi* within the GIT to high levels by one week of age. A significantly lower percentage of neonatal foals had detectable virulent *R. equi* in their breathing zones than when sampled at 6 weeks of age. These findings have important implications for management practices aimed at reducing the prevalence of rhodococcal disease on farms. This includes the separation of mare/foal pairs based on foal age and the removal of faeces in high traffic areas of farms.

Determining age-related differences in the response of foals to virulent *R. equi* exposure will ultimately lead to a better understanding of the specific factors involved in protecting foals against rhodococcal disease. These factors can then be targeted to enhance existing disease management recommendations aimed at dampening this significant disease of foals worldwide.

CHALLENGES IN TEACHING EQUINE SCIENCE

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Equine science has been taught as a distinct subject at Massey University since 2001, initially as some selective papers, and as a major within the AgriScience degree since 2003. The programme has a strong agricultural science and systems approach to teaching with the students required to complete two systems / capstone type papers in their final year. Within the capstone papers students are required to work in groups to solve real world, generally on-farm, problems relating to equine production. Often the capstone group composition includes students from other majors (Agriculture and AgriCommerce) which provides a cross-pollination of ideas and approaches. The programme is taught from within the Institute of Animal, Veterinary and Biomedical Sciences and therefore has a healthy mix of practitioners, animal scientists and veterinarians contributing to the papers. The equine papers and the entire degree programme is taught conventionally on site and also in distance mode throughout New Zealand, which enables industry participants to upskill in conjunction with employment.

The Massey equine programme is also offered as a minor within the Sport and Exercise degree in recognition of the career path for equestrian coaches and elite sport horse production. This programme is recognised by, and affiliated with, the National governing body for equestrian sport Equestrian New Zealand (ESNZ?). In conjunction with ESNZ the university runs a mentoring and elite rider support programme to stimulate the development of future Olympians.

Enrolments within the equine programmes are typically females from an urban environment with an involvement in, or background in equestrian sport. This student profile provides some interesting challenges in progressing from a background based on usually a single equine unit of high marginal utility to a production focus of larger numbers of horses which are produced or trained for a commercial focus and the generation of income.

IT'S NOT ALL HORSING AROUND A ROUND YARD AT CSU

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The Bachelor of Equine Science commenced at Charles Sturt University (CSU) in 2007 with predecessor Applied Science, Associate Diploma and Diploma courses dating back to 1896. Taking a global approach to the equine industry, CSU looks to opportunities within Australia and internationally. Equine graduates of CSU are employed in the industry in an equally broad range of placements, from livery managers in the Royal Household, to research scientists to owner-trainers in central Australia.

With a strong undergraduate program covering many facets of equine science and management, one of the many strengths of the Equine Science degree at CSU is its Honours and Postgraduate programs. Over the last 25 years, much original equine research has been conducted by Equine Science students at CSU Wagga Wagga. The depth and breadth of our Equine Research is vast, ranging from Equine Performance - the heritability of performance traits, TCO_2 and transported horses, using swimming rats as a model for the athletic horse to Equine Health – surveying parasite burdens, determining the need for trained equine therapists, investigating the effects of NSAIDS on GIT integrity and efficacy of NSAIDS in treating synovitis, health status of pony club horses and investigating mechanisms that lead to laminitis to Behaviour and Welfare - the management of stereotypies and objectively measuring reactivity, to Equine Nutrition – determining the digestibility of different diets, manipulating insulin status in insulin-resistant ponies through diet, investigating the effect of meal frequency on plasma glucose and insulin and evaluating rate of intake to Equine Reproduction – population control in brumbies, variation in kisspeptide onto GnRH receptors across mare oestrus, optimal freezing conditions for sperm and effect of seminal IGF-1 on testicular function to Equine Pharmacokinetics – investigating the relationship between exogenous growth hormone on mediator hormones and determining the pharmacokinetics of common equine therapeutic drugs.

With international recognition for Equine Research ongoing at CSU, there are many opportunities for both undergraduate and postgraduate students to be involved in a broad range of "in situ" research and teaching activities. These activities serve as valuable learning experiences, incorporating, apart from the obvious equine management skills, more generic skills such as communication on all levels, personnel management, logistics planning, budgeting and critical thinking.

EQUINE SCIENCE DEGREES AT UQ: A WINNING FORMULA

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The Equine Program at The University of Queensland (UQ) is a major stream within a larger integrated animal science program. The three year **Bachelor of Applied Science (Equine Science)** gives students a sound basis in animal science with a focus on the horse. The extended major in Equine Science has five dedicated compulsory equine courses that build on the foundation courses in chemistry, biochemistry, biology, anatomy, physiology and microbiology and the underpinning animal science disciplines of nutrition, reproduction and genetics. The scientific principles of these courses are applied and expanded in the equine courses outlined below. In addition, students may elect to undertake an international study program and equestrian coach education

The program was designed following discussion with industry. This identified a number of major areas that need to be addressed: the application of behavioural science to training and horse welfare, occupation health and safety; genetics and breeding; exercise physiology; rehabilitation from injury; nutrition, disease and biosecurity. There is an expectation that all students will start this program with basic skills and knowledge of horse husbandry. Industry expects graduates to have a high level of both horse and laboratory-based skills.

ANIM1006 – Equine Husbandry and Equitation: This is an introductory course which provides an overview of the equine industry and its many different sectors. The course provides an introduction to the many aspects of equine production and management including breeds and genetics, nutrition, behaviour, welfare, occupational health and safety, equitation science, the performance disciplines, husbandry and management. It provides the basis for the equine courses that are taken in subsequent years.

ANIM2024 – Equine Behaviour and Performance: All aspects of the interaction between the horse and humans require knowledge of the behaviour of horses of all classes. This course develops an understanding and application of behavioural science as it relates to the horse, and human-horse interaction. Basic learning theory is applied to horses at all levels of training, from birth to performance with particular emphasis on horse welfare and occupational health and safety. The biomechanics of equine locomotion and its relationship to training, exercise and performance are examined.

ANIM2039 – Equine Breeding and Stud Management: The equine breeding industry is diverse due to the large number of horse breeds and their different breed registration requirements. Effective breeding management is key to optimising productivity in the equine industry. This course provides the underpinning knowledge of equine reproductive anatomy, physiology and endocrinology of both the stallion and the mare, and its application to breeding management. It covers all aspects of the equine breeding cycle including stallion fertility and management, the management of the oestrous cycle of the mare and techniques used to optimise breeding efficiency, the physiology of equine pregnancy, placentation, foetal development, parturition, lactation and post-partum mare and foal management. Stallion, mare and foal health are addressed.

ANIM3039 – Equine Exercise and Rehabilitation: The horse is bred for athletic performance. There are many different equine disciplines that require different training techniques to optimise performance. This course is a study of exercise physiology and biochemistry as it applies to the equine athlete, injuries that affect the performance horse and how they are managed and rehabilitated. It builds on the biomechanics of locomotion presented in ANIM2024. Development of training programs and their assessment are

discussed. The pathophysiology of equine injury is investigated and an understanding of the appropriate treatment and rehabilitation of injuries is developed.

ANIM3030 – Equine Nutrition and Health: Equine nutrition is the foundation for athletic and breeding performance of the horse. It has significant implications for horse health. This course covers the unique anatomy of the equine gastrointestinal tract, nutritional physiology as it relates to athletic and reproductive performance and metabolic disease. It applies the principles of nutrition to equine growth and development, performance and reproduction. General aspects of equine health and disease management are covered including biosecurity.

Great emphasis in the program has been placed on the **integration knowledge with practical experience** to enhance the student experience. UQ has excellent facilities for all aspects of student /horse interaction, including breeding and performance (particularly riding) activities. Importantly, the program is offered in distance mode which provides access for students nationally and is currently being offered by the University of New England as the equine component of their Animal Science degree (Brown and Bryden,2012) The program is ideally suited for international student internships . It also provides an excellent basis for students wishing to pursue a higher degree.

The course content in ANIM1006 and ANIM2024 is based on the latest developments in equine science and students gain an appreciation of behaviour and management through direct interaction with horses. In ANIM1006, interaction is limited to educated horses to provide the basis of appropriate and safe handling of horses. In contrast, ANIM2024 provides hands on experience with young, naïve horses which develops higher levels skills relating to horse behaviour and training. Together the courses provide the skills necessary for the use of all classes of horses in performance and research environments. ANIM2039 extends the skills developed in the earlier courses to horses in a reproduction setting. This is complemented with laboratory based skills related to reproduction. In ANIM3039 students monitor responses to exercise in the horse through the use of HR monitors, blood biochemical analysis and GPS technology. This is used to evaluate of fitness and training. Students undertaking ANIM3030 students are required to apply experimental methodology to a nutrition related project. It involves the monitoring of horses, collection and analysis of both equine and nutritional data and the submission of a research report.

The University is currently considering introducing a four year degree; **Bachelor of Equine Science and Management** to complement the three year program. The challenges in maintaining these programs is overcome the perspective that equine programs lack scientific rigour, the cost of maintaining horses in a program, which can be significantly offset (Cawdell-Smith *et al.*, 2011) and job opportunities for graduates, which are many in a global industry. Nevertheless, the key goal of our equine programs is to produce graduates who not only have the necessary knowledge and practical skills, but also are confident and capable equine scientists.

Brown WY and Bryden WL (2012) Collaboration: key for equine science at UNE. *Proc Australasian Equine Sc Symp.* 4:17.

Cawdell-Smith AJ, Hohenhaus MA, Coyle MP, Delzoppo NS, Anderson ST and Bryden WL (2011)The role of horses in the undergraduate equine science program at the University of Queensland *Proceedings of the 2011 NAEAA Conference*, Tennessee, pp27-28.

COLLABORATION: THE KEY FOR EQUINE SCIENCE AT UNE

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Against a backdrop of increased competitiveness for Australian government funding, there are limited examples of true cross-institutionl collaboration in tertiary sector education. This paper demonstrates how a cross-institutional, collaborative approach can enhance a new or existing degree program, improve efficiency through resource sharing and deliver greater outcomes for the student.

Whilst Animal Science degree programs have traditionally focused on production animals, the inclusion of companion animals into the curriculum has proved successful at some institutions (McNamara, 2009), and Equine Science degrees are now well estsblished at universities in both Australia and abroad. The new Bachelor of Animal Science (2011) at the University of New England (UNE) captures this broader focus with the inclusion of three specialty majors in its degree progam: Canine and Equine Science, Wildlife Management, and Livestock Production.

The canine component of the combined Canine and Equine Science major is well supported by existing canine research programs and academic expertise at UNE; however with no existing expertise at UNE in equine science, and no campus facilities to support this area of specialty, alternative options for delivering the equine component were explored. A solution was provided through collaboration with the University of Queensland (UQ) to provide the equine component on a cross-institutional basis, enabling the delivery of a double major in Canine and Equine Science in the UNE Animal Science degree.

This arrangement has not been without some administrative challenges; but overall it has proved to be a cost-effective option for the host university (UNE) to incorporate an additional area of specialty; and the provider university (UQ) benefits from additional student enrollments into its existing course offerings. Students enrolled in the UNE Animal Science degree have the option to include up to five equine subjects in their degree, which they undertake by distance education through UQ (Cawdell-Smith *et al* 2012). A recent development has been the inclusion of a 1st year subject (ANIM1006) with a practical horse component, to provide both internal and external UNE Animal Science students the opportunity for hands-on experience early in their degree. A further positive outcome of this has been increased local community involvement with UNE through enagaging a local secondary school, its riding instructors and facilities, for the delivery of some practical horse components.

Cawdell-Smith AJ, Hohenhaus MA and Bryden WL (2012) Equine science degrees at UQ: A winning formula *Proc Australasian Equine Sc Symp.* 4:15-16.

McNamara JP (2009) ASAS Centennial Paper: The future of teaching and research in companion animal biology in departments of animal sciences. *J Anim Sci*. 87:447-454

COMMERCIAL PRODUCTION OF HORSES AT PASTURE IN NEW ZEALAND

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In New Zealand the production of horses is pasture based due to the temperate climate that allows pasture growth throughout the year. The largest studbooks in New Zealand are the Thoroughbred and Standardbred, bred for flat and harness racing, respectively. Many New Zealand-bred horses go on to compete internationally, and the sustainability of the local breeding industry can be attributed, in part, to the income generated from such exports. For the past 20 years the numbers of mares, stallions and foals have been in decline for both Thoroughbreds and Standardbred. It is predicted that the reduced numbers of horses available for racing will have a significant negative effect on the domestic Thoroughbred racing market in the very near future.

Both Thoroughbred and Standardbred studbooks are closed, but they have different regulations regarding the use of reproductive technologies. In the Thoroughbred industry all breeding must be by natural service (live cover), while the Standardbred industry allows the use of artificial insemination (AI) using fresh, cooled or frozen semen, and embryo transfer.
A major constraint on reproduction is the condensed commercial breeding season, especially for Thoroughbreds, which is effectively 01 September to 01 December. This is significantly shorter than the Northern Hemisphere breeding season of around 150 days. The difficulties imposed by the short breeding season are further confounded by the mismatch between the imposed and physiologic breeding seasons for Thoroughbreds, the highly variable gestation length of mares, and the large book sizes of popular stallions (especially in the Thoroughbred industry). It spite of these difficulties, end-of season pregnancy rates and foaling rates are similar to Northern Hemisphere data.

Nutrition is a key factor in reproductive success, however, pasture quality and availability may be limited on studs during the breeding season in association with high stocking densities. When sufficient high quality pasture is available it can meet all the nutritional requirements of lactating and dry mares, with the exception of selenium. Young horses fed pasture based diets under New Zealand conditions have similar growth rates to those reported for their overseas counterparts that are fed grain-based diets. However, despite the availability of cheap, high nutritive value pasture, studmasters in New Zealand feed grain-based concentrates to all classes of horses on studs, and feed large quantities to weanlings and yearlings.

Historically, much of the data captured on horse production has focused on the Thoroughbred industry. However, the use of assisted reproductive technologies, and the effect of these on the production process, means direct translation of results from the Thoroughbred industry to the Standardbred industry is not optimal or suitable. Further research is therefore required to characterise the management and reproductive performance of Standardbred horses in New Zealand

EFFECT OF INCREASING ADIPOSITY, INDUCED BY A HIGH FAT LOW GLYCAEMIC DIET OR SIMILAR DIET WITH ONCE DAILY GLUCOSE, ON INSULIN SENSITIVITY IN HORSES AND PONIES

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Obesity is associated with insulin resistance (IR), both of which constitute major risk factors for laminitis in horses and ponies. Regarding the major causes of IR, the relative importance of obesity vs. the high glycaemic index of diets (causing hyperinsulinaemia) is not yet fully understood. Diets high in non-structural carbohydrates (NSC) may apparently cause IR in horses independent of obesity (Hoffman *et al*, 2003). The aim of this study was to investigate the changes in insulin sensitivity when horses and ponies became obese, either in the absence of a glycaemic/insulinaemic stimulus or with a once daily glucose/insulin peak.

Six adult Standardbreds, mixed-breed ponies and Andalusian-cross horses, all in moderate Body Condition Score (BCS; 4-5/9), were assigned to one of three diet groups (comprising two from each breed). An insulin-modified frequently sampled intravenous glucose tolerance test (FSIGT) was undertaken before and after a period of increasing adiposity. Minimal model analysis of the glucose and insulin curves derived values for insulin sensitivity (SI). Percentage increase in body fat was determined by deuterium dilution (Dugdale *et al*, 2011).

All horses had *ad libitum* access to pasture hay which was low in NSC. In addition, all horses received twice daily a basal ration of soaked soyahull pellets, lucerne chaff and a vitamin and mineral supplement. One group (FAT; n=6) received caloric supplementation in the form of granulated vegetable fat and canola oil (increasing up to 200g/100kg BW), to provide up to ~210% of their estimated maintenance energy requirements. Another group (FAS; n=6) received added glucose (1.5g/kg BW) in the morning feed to cause a single daily glycaemic (and insulinaemic) response. This group also received some vegetable fat in the evening, to ensure their diet was isocaloric with the FAT group. A control group (CON; n=6) received hay plus the basal ration with no added fat or glucose (diet sufficient to meet maintenance requirements and maintain a moderate BCS). The study period lasted for 21 weeks and weekly monitoring included BW, BCS, ultrasound fat measurements and blood samples.

In the weight gain groups, % body fat increased from 9.3 ±1.5 to 15.1 ±1.6 % in the FAT group (paired *t*-test; P=0.05) and from 7.8 ±2.3 to 14.9 ±1.7 % in the FAS group (P=0.003). BCS also increased significantly, from 4.9 ±0.4 to 7.3 ±0.5; and from 4.8 ±0.2 to 7.7 ±0.7 (P=0.0004 and 0.0001; FAT and FAS groups, respectively). BCS and % body fat did not change significantly in the CON group. Surprisingly, SI did not change significantly in the FAT group (2.6 ±0.5 to 3.0 ±0.8). Furthermore, there was a significant improvement in SI in the FAS group, from 2.6 ±0.8 to 4.7 ±0.9 (*P*=0.009). There were no significant changes in SI within the control group.

These data indicate that increasing adiposity alone did not cause IR in the horses and ponies which gained weight on a high fat, low glycaemic diet. Additionally, the single daily glucose meal appeared to improve insulin sensitivity despite increased adiposity; thus the short term glucose/insulin peaks may have slightly improved insulin effectiveness. With careful dietary modification it may be possible to reduce the likelihood of IR in animals with increased adiposity, thereby reducing one potential risk factor for laminitis (although obesity may remain a risk factor independent of IR).

Dugdale AH, Curtis GC, Milne E, Harris PA, Argo CM. (2011). *Equine Vet J*: 43(5):562-70.
Hoffman RM, Boston RC, Stefanovski D, Kronfeld DS, Harris PA. (2003). *J Anim Sci* 81(9):2333-42.

MODERATE DIETARY CARBOHYDRATE IMPROVES AND HIGH DIETARY FAT IMPAIRS GLUCOSE CLEARANCE IN AGED THOROUGHBRED GELDINGS

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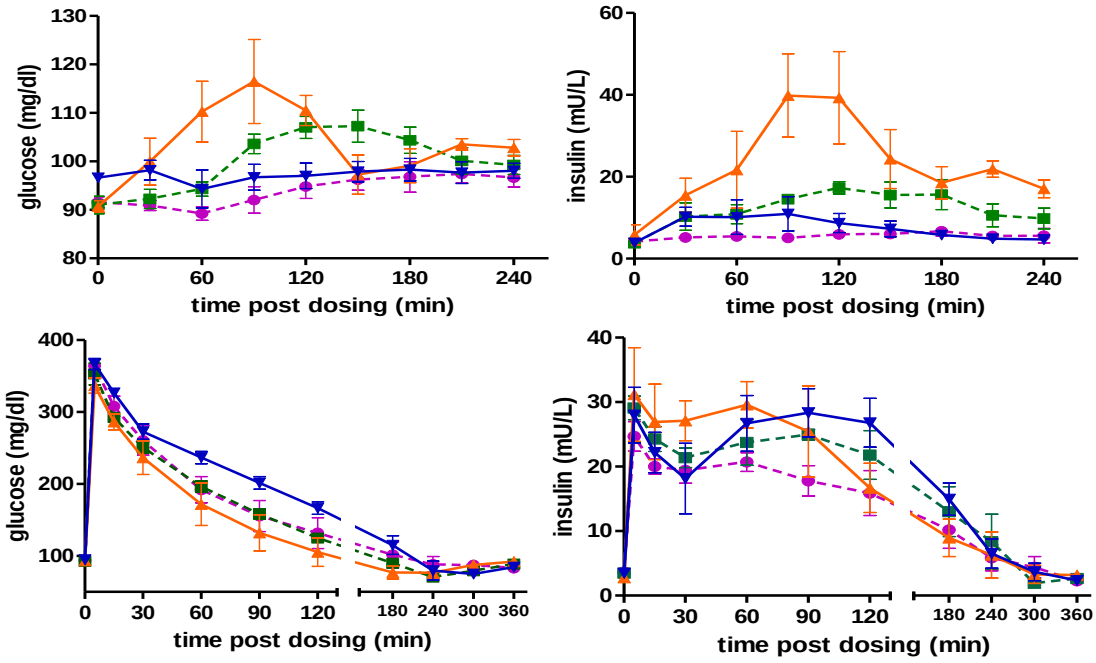
Low carbohydrate diets are often recommended for horses with metabolic syndrome and high fat diets are useful for managing horses suffering from RER and PSSM. It is questionable whether these types of diets are appropriate for normal, non-obese horses. This study was conducted to determine whether a moderate daily intake of carbohydrate from oats or a high level of fat intake from vegetable oil would affect glucose clearance as measured by an intravenous glucose tolerance test (IVGTT).

Four aged Thoroughbred geldings (21.5 yrs ± 3.32 yrs; Weight 572.16 Kg ± 50.53 Kg; BCS 5.0-6.0) were used in a 4 X 4 Latin square design study to assess the effect of energy source on glucose, insulin, NEFA and triglycerides during a glycemic response test (GRT) and an IVGTT. Each period lasted 4 weeks. The treatments were isocaloric and consisted of ~11 Mcal DE/d from grass hay and ~7.5 Mcal DE/d from either 1. Additional grass hay (GRASS), 2. Lucerne/grass blend pellet (ALF), 3. Whole oats (OATS) or 4. Lucerne cubes + soybean oil (OIL). GRASS and ALF supplied 53-54% of DE from fiber, the OAT treatment supplied 31% of DE from NSC and the OIL treatment supplied 30% of DE from fat. On day 14 of each period the GRT was conducted where horses were fed ½ of their regular daily intake (3.75 Mcal DE) of their respective treatment feed, Blood samples were taken before (0 m) and at 30 m, 60 m, 90 m, 120 m, 150 m, 180 m, 210 m and 240 m post feeding. On day 28 of each period the IVGTT was conducted where a 50% dextrose solution was administered IV at a rate of 0.5g glucose/kg BW over 10 min and blood samples were collected immediately before (0 min) and at 5, 15, 30, 60, 90, 120, 180, 240, 300 and 360 min post administration. Data were analysed using repeated measures ANOVA with Fisher's post-hoc test. Significant differences were accepted at the 5% level.

A moderate intake of NSC (31% of DE) improved glucose clearance during IVGTT compared to an all hay diet, while a high dietary fat (30% of DE) impaired glucose clearance during IVGTT compared to an all hay diet. Blood glucose returned to baseline in 126.6 ± 25.8 min in OATS compared to 216.7 ± 23.5 min in OIL ($p<0.05$).

Figure 1. Plasma glucose and insulin during GRT (top) and IVGTT (bottom) in horses fed GRASS (●), ALF (■), OAT (▲), and OIL (▼).



The results of this study suggest that feeding a diet containing a moderate quantity of carbohydrate (31% of DE) improves glucose tolerance in non-obese aged horses compared to an all hay diet. Conversely, feeding a high fat diet (30% of DE) impairs glucose clearance. Moderate carbohydrate equates to 2.0-2.5 kg/d of grain and a high fat diet equates to about 500g/d of oil (for a 500kg horse). More research is needed to determine the effect of different energy sources on glucose clearance in horses with metabolic syndrome, obese horses and in non-obese exercised horses.

METABOLIC CHAOS: CONSEQUENCE OF THE MODERN EQUINE LIFESTYLE

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Horses have evolved as herbivores over the past 3 million years and have adapted to grazing low quality roughages. Horses have a small stomach and usually graze for about 14 - 18 hours a day, and where possible, select a wide range of plant species and grasses from the environment to provide a balanced nutrient intake (Janis, 1976). For many horses their "modern" life is very different.

Today, many horses have restricted or no access to pasture, and are supplemented with a range of feedstuffs to provide the nutrients required for growth, development and performance. Often, the pastures that are offered to horses have been developed as feeds for ruminants, which have a totally different digestive system. Some of these pastures are unsuitable for horses, because they supply the wrong types and balance of nutrients (*www.safergrass.org*). Grain and grain by-products are the lowest cost, most convenient and readily available feeds to supply the additional digestible energy (DE) required by horses. Cereal grains form the basis of concentrate feeds and are rich in non-structural carbohydrates (NSC), especially readily digestible sugars and starch. Plant breeders have been effective in developing grains for human consumption with higher and more digestible starch and sugar content. In the past, horses never consumed these types of grains. As a result of our lifestyle, it is convenient for horse owners to feed these concentrate feeds twice daily . Further, many horses are not worked to the same extent as they were before mechanisation. Consequently, horses are often grazing pastures designed for ruminants, are supplemented twice daily with large amounts of high NSC feeds, and are often overfed and underworked.

There is an increasing incidence of metabolic related ailments in horses including obesity, colic, gastric ulcers, laminitis and associated lameness, polysaccharide storage myopathy (PSSM), insulin resistance (IR), equine metabolic syndrome (EMS), Cushings disease, and Equine Systemic Proteoglycan Accumulation (ESPA) (Frank, 2009, Firshman and Valberg, 2007, Pollitt and Visser 2010). Many of these conditions are treated by a range of veterinary medications. The increasing incidence of these metabolic diseases raises the obvious question, is there an underlying cause?. It is inconceivable that the horse and its metabolism have suddenly changed over the past 50 -100 years.

Why is there an increase in metabolic disease?

It is proposed that the increasing incidence of metabolic disease reflects an underlying cause of which feeds and feeding practices are central, and that the changes in the way we feed our horses is causing metabolic chaos. The result in many horses is one or some of the metabolic diseases listed above. Not all horses are affected equally, and some breeds are more susceptible to some syndromes. Therefore the outcome of metabolic chaos will be tempered by breed, age, activity and fitness and dietary history.

In humans, despite all efforts over the past two decades, obesity and coronary heart disease (CHD) are now an epidemic in western societies. Lifestyle changes and low fat diets have not worked, and focus has turned to high glycaemic index (GI), high sugar diets as a possible cause. The evidence that refined carbohydrates, and specifically sugar is a major causal agent in obesity and CHD humans is compelling. This invites the proposition that one of the major dietary factors contributing to metabolic chaos in horses is the amount and frequency of sugar and starch intake. It is now well recognised that the amount and type of sugar and starch in equine feeds has changed significantly. Feeding high NSC feeds twice daily will cause spikes in circulating levels of insulin and glucose (Richards and Kempton, 2012) making maintenance of glucose homeostasis difficult. The accompanying loss of insulin sensitivity may lead to insulin resistance (IR), and consequently higher circulating levels of insulin and glucose.

How does the horse dispose of the increased circulating glucose? It can be stored as

- body fat causing obesity. Insulin causes partitioning of glucose into fatty acids, and accumulation in fat cells
- polysaccharides in muscles (PSSM) in some breeds such as Quarter horses, draft breeds, Appaloosas, warm bloods, and some racing horses.
- proteoglycan in connective tissue causing swollen legs and tendons (ESPA).

A consequence of insulin IR is increased circulating levels of insulin and there is increasing evidence that this is pivotal to the development of many of the metabolic diseases listed above. There are however differences between animals in their ability to cope with IR and hence development of the disease.

It is common practice to feed pregnant mares during the last trimester of pregnancy and there is some evidence that this can result in metabolic chaos in the foetus and subsequent foal. It is well recognised that continued feeding of high GI diets may cause gestational diabetes is in women. Mothers who suffer Type II diabetes during the last trimester of pregnancy have higher circulating levels of insulin, which cross the placenta and affect the foetus. Babies born from mothers with gestational diabetes have increased probability of becoming a Type I diabetic by the age of 10, especially if they consume high sugar diets.

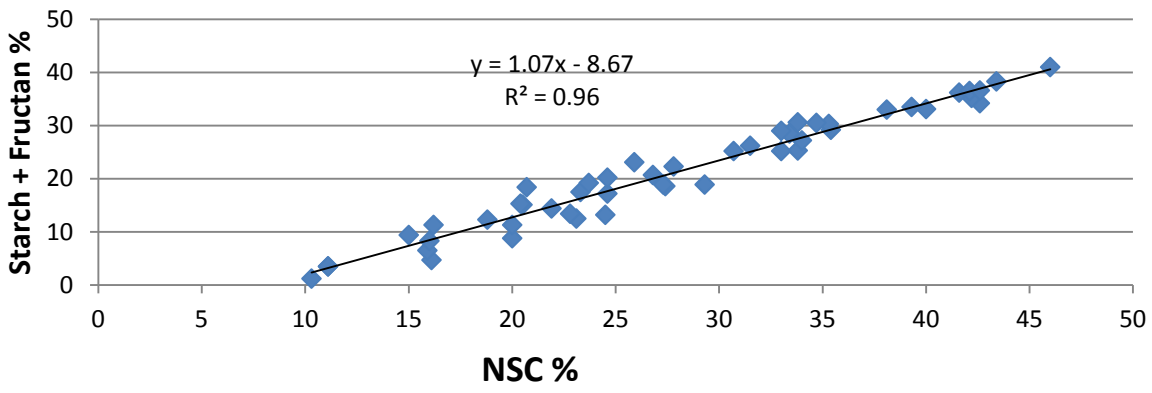
It would be of great interest to know what influence the maternal diet and subsequent feeding management of the weanling have the metabolic state of the yearling. In this regard it is common to feed foals and yearlings high NSC feeds to increase growth rate and achieve better body condition scores for the sale ring, or for two year old racing. It is to note that the metabolic disease "old age Cushing's" is now appearing in horses as young at 7 years old.

Dietary NSC and metabolic chaos

There are a number of carbohydrates that comprise the NSC fraction of plants (see Equi Analytical (*www.equi-analytical.com*). These include starch, sugars and fructan. Fructan, determined by difference between WSC (water soluble carbohydrate) and ESC (ethanol soluble carbohydrate), is a major storage carbohydrate in cool temperature (C3) grasses. Fructans are resistant to enzymatic digestion in the small intestines, however they are readily fermented in the equine hindgut. Fructan content in some pastures are reported to be 15- 30%.

We have submitted over 50 commercially available equine feeds from Australia, UK and USA to Equi Analytical. The NSC content of the feeds varied from 70g/kg to 470g/kg and the fructan content varied from <10g/kg to 80 g/kg. Of interest is the close correlation between NSC and starch plus fructan content (Figure 1). This relationship indicates that in feeds containing more than 30% NSC, over 80% of the NSC is as starch plus fructan. Fructan has received much attention as a possible trigger for laminitis.

Figure 1. Starch + fructan as % of NSC in commrcial horse feeds from Australia, UK and USA



The form of carbohydrates in a diet is the major determinant of site of digestion. High intake of readily digested sugars can increase gastric ulceration. It is contended that on high sugar concentrations to the small intestine may induce leaky gut syndrome (dysbiosis), allowing passage of sugars into the bloodstream, and increased levels of circulating glucose. Leaky gut syndrome is recognised in humans, and may also occur in horses overloaded with dietary sugars. High sugar content may also cause changes to the stomach and intestinal microflora.

Horses have an intestinal starch digestion capacity of approximately 200g/100kg bodyweight, but no enzymatic capacity to digest fructans in the small intestines. Undigested starch and fructans pass to the hindgut, where they are fermented to volatile fatty acids (VFA's), mainly acetate, propionate and butyrate. In horses where the hindgut microbiota have not been adapted, these substrates are rapidly fermented causing hindgut acidosis, which can lead to rapid onset of laminitis. In the hindgut, high fibre diets produce higher levels of acetic acid, whereas high starch diets produce higher levels of the glucogenic substrate propionate (Table 1), which is directly converted to glucose in the liver.

Table 1. Volataile fatty acid ratios (%) in ponies fed roughage and grain (Hinz et al 1971)

Roughage/ grain	Acetic	Propionic	Butyric	Total VFA mM/l
1:0	73.5	17.1	10.2	49.1
1:4	59.4	25.1	12.5	42.5

Processing of feeds by pelleting, micronisation, extrusion, steam flaking can shift the site of digestion between the small intestines, and the hindgut, by altering the intestinal digestibility of starch. Notwithstanding, high starch diets will increase circulating glucose, and therefore insulin release.

Conclusions

Long term feeding high NSC feeds can increase circulating levels of glucose in horses either directly following digestion in the small intestine, or from the fermentation of starch and fructans in the hindgut. The substrates starch and fructan produce higher molar ratios of the VFA propionate, which is converted to glucose in the liver. Feeding high NSC feeds twice daily will cause spikes in insulin and glucose. Horses in active work can metabolise much of this glucose.

Many horses however are overfed high (>12-15%) NSC diets, underworked and cannot utilise this additional glucose. It is proposed that feeding high NSC feeds causes these horses to produce high levels of glucose, secrete increased insulin and in some cases become insulin resistant. It appears that a nutritional imbalance with respect to the carbohydrate content of the diet precipitates metabolic chaos, which potentially can result in a variety of equine metabolic diseases.

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THE INCIDENCE OF OCD IN THOROUGHBREDS IN THE HUNTER VALLEY, NSW, AUSTRALIA

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During growth, the skeleton of young horses undergoes a process of endochondral ossification, whereby cartilage is converted to bone. Osteochondrosis occurs when this process is disrupted. Osteochondritis dissecans (OCD) is the name given to this abnormal process when it occurs at the joint surface, affecting the subchondral bone (Gaughan 2012). OCD is a multifactoral disease, with genetics, caloric intake, growth rate and mineral nutrition playing a role (McIlwraith, 2011). Despite knowing these high risk factors for OCD, it is a difficult disease to prevent in young Thoroughbreds. A Thoroughbred with OCD often requires expensive surgery and loses value as buyers are wary of purchasing animals with this skeletal abnormality, making it a high priority as a condition to prevent for Thoroughbred breeders. This paper examines the incidence of OCD in Thoroughbreds born in the 2010 season and raised in the Hunter Valley region of NSW, Australia.

As part of a larger study, complete radiograph reports were collected from 250 weanlings, foaled in 2010 from 9 Thoroughbred breeding farms located throughout the Hunter Valley region. Some farms contributed every radiograph report taken on farm for that year while other farms provided a smaller sub-section of weanlings with selection based primarily on those weanlings that would be radiographed at some point prior to sale. Radiograph reports were assessed by the farms veterinarian (who varied for each farm) and the number of OCD lesions tallied along with the occurrence of lucencies, cysts and fragments. The results are presented in Table 1.

Table 1: The percent of horses with OCD lesions, lucencies, cysts and fragments and percent of lesions found in the fore/hind fetlocks, hocks and stifles in 250 thoroughbreds foaled in 2010 in the Hunter Valley, NSW.

	% horses affected	% of lesions found in specific joints			
		Fore Fetlocks	Hind Fetlocks	Hocks	Stifles
OCD Lesions	23.2	14.4	7.2	33.7	44.6
Lucency	18	50.7	8.5	5.6	35.2
Cysts	4.8	10	30	0	60
Fragments*	16.4	25	67.3	1.9	5.8

*excludes fragments noted as 'chips'.

The 23.2% incidence of OCD lesions is in accord with the 26% occurrence reported in Australasian Thoroughbreds (Eckman et al, 2009) but is much lower than the 50.7% reported in Norwegian Standardbreds (Lykkjen et al, 2012). The incidence of OCD in this study varied between farms (0% from a farm with 8 horses in the study to 37% from a farm with 35 horses in the study). This could reflect genetic, nutritional and/or management causation.

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THE PREVALENCE OF LIMB DEFORMITIES IN STANDARDBRED AND THOROUGHBRED FOALS IN NEW ZEALAND: A PRELIMINARY STUDY

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The aim of the study was to quantify the prevalence of angular and flexural limb deformities in foals during their first week of birth, on two Standardbred and two Thoroughbred stud farms in New Zealand. Further, the study aimed to trial and evaluate a method for prospectively recording on-farm data on limb deformities.

Data on limb deformities were collected on all foals that were on the farms during the 2010/2011 breeding season. Observational assessments were used to score foals from their first week after birth to fifteen months of age. A standardised recording sheet was developed to allow stud masters to record data prospectively for each foal enrolled in the study. Data were recorded on foal information such as, sire, dam, date of birth, gender, conformation, size, condition score and details on the types and severity of the limb deformities. Descriptive statistics were used to describe the population of foals, in terms of the number and type of limb deformities recorded.

The majority of the foals (253/313, 80.83%) had one or more limb deformities recorded in their first week after birth. Of these, 196 had more than one deformity recorded (196/253, 77.47%). Carpal valgus was recorded in the majority of the foals (162/313, 51.8%), with 84/162 (51.85%) being unilateral and 78/162 (48.15%) being bilateral. Most of the deformities recorded (628/807, 77.82%) in this study were slight deformities, believed to be self correcting that required no veterinary attention.

This study provides baseline data for future studies that aim to record detailed prospective data on the incidence of limb deformities in New Zealand. Furthermore, it could provide an opportunity to examine if angular and flexural deformities are associated with racing and sale performances.

THE RELATIONSHIP BETWEEN INSULIN STATUS AND OSTEOCHONDROSIS IN THOROUGHBRED YEARLINGS

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Insulin resistance is a complex pathophysiological condition that appears to underlie a number of chronic conditions often called 'equine metabolic syndrome', including laminitis, obesity, and osteochondrosis (OC). In the horse, the effects of diet and exercise on insulin resistance have been examined, but few studies have investigated insulin resistance and bone development. OC is a developmental disease caused by a defect in the normal process of bone formation resulting in the thickening, cracking and tearing of the joint cartilage of growing horses. In the most advanced stages the condition is termed Osteochondrosis Dissecans (OCD). This condition is known to be associated with dietary deficiencies and/or nutrient imbalances, biomechanical stress or trauma, rapid growth rates, and genetic influences. In regards to insulin resistance and OCD, there is a current hypothesis that elevated post-feeding insulin levels may predispose growing horses to develop the condition (Ralston, 1996).

Our current study investigated the relationship between insulin status and the occurrence of OC in Thoroughbred yearlings. Briefly, yearlings (*n=191*) born in '2007' and '2008' were recruited from four NSW Thoroughbred stud farms and fasting blood samples were obtained following stabling overnight. Plasma insulin and glucose concentrations were determined by radioimmunoassay and Hexokinase methods respectively. Skeletal abnormalities were determined by radiography analysed by experienced veterinarians, and the data were retrospectively classified as either osteochondrosis related lesions (OC), other bone abnormalities, or no abnormality (NA).

Across all farms 27% of yearlings examined exhibited OC lesions whilst 48% of yearlings had NA. A significantly ($P<0.01$) higher proportion of male compared to female yearlings presented with OC lesions. Analysis of insulin revealed lower fasting insulin concentrations in yearlings with OC compared to those with NA (2.3 ± 0.2 vs. 3.4 ± 0.2 mIU/L respectively). In contrast plasma glucose concentrations were not different (100.8 ± 1.1 vs. 101.0 ± 1.4 mg/dL).

Analysis was then performed on blood samples from the 2008 yearling cohort that had been obtained earlier in postnatal life, as foals (2 month) and weanlings (5 months), and also from their mothers during late gestation. In foals, insulin levels were again significantly (P<0.01) lower in horses that developed OC as yearlings (n=13) compared to those classified with NA as yearlings (n=*52*). Overall mean plasma insulin concentrations from 2 to 14 months were 1.5 ± 0.1 vs. 2.8 ± 0.2 mIU/L for OC and NA groups respectively, with no differences in glucose concentrations noted at any age. Interestingly, a moderate inverse relationship ($r = -0.4$, $P<0.05$) was noted between insulin levels in the foals in postnatal life and mare glucose levels during late gestation. Indeed mares that gave birth to foals that developed OC as yearlings had significantly ($P<0.01$) higher plasma glucose levels (93.2 ± 2.0 vs. 86.7 ± 1.1 mg/dL) and body condition scores (4.1 ± 0.1 vs. 3.8 ± 0.1; out of 5.0) during gestation compared to mares that had foals that developed normally.

Taken together our results suggest that hypoinsulinaemia during early postnatal growth of foals may be linked to the development of osteochondrosis. Furthermore the influence of maternal nutrition on metabolism during pregnancy may have consequential effects upon postnatal metabolism and normal skeletal development. Further work is in progress to confirm and extend these findings.

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A PRELIMINARY EXAMINIATION OF THE FORELIMB AND HOOF CONFORMATION IN A POPULATION OF MONGOLIAN HORSES

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Data were collected from a convenience sample of 120 Mongolian horses used in the 2011 Mongolian Derby. Digital images of the hooves were obtained and the lower limb conformation was assessed by four veterinarians involved in the screening of the horses offered for the derby. The horses were predominantly geldings (96%, 100/104), approximately 8.6±2.5 years old and 137±8cm at the withers. Based on a seven point linear score, lower limb conformation was normal with a trend (1 linear score deviation) towards slightly knock kneed, mildly offset cannon and valgus at the fetlock. Hoof measurements were within the norm for horses of this size. Fetlock valgus was associated with a smaller hoof width:length ratio (P=0.016). None of the other hoof measurements were significantly associated with abnormal conformation scores. None of the horses were subjected to routine hoof trimming. Overall, few conformation abnormalities were observed and hoof shape and size was within the normal expected range for horses of this size. The hoof conformation in this population of Mongolian horses represented the natural interaction of the environment with the hoof.

BONE BIOMARKERS IN MARE-FOAL PAIRS THROUGH THE FIRST FOUR MONTHS OF FOAL LIFE.

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The University of Queensland [A] School of Veterinary Science, Gatton, [B] School of Biomedical Sciences, St Lucia and [C]Equine Research Unit, School of Agriculture and Food Sciences, Gatton

Bone quality is an important attribute for performance horses. Bone accrual begins in utero and early life, which may be a critical period for programming metabolic systems. Foals depend on maternal calcium supplies in foetal and early neonatal life, but little is known about the relationship between maternal bone mobilisation and neonatal and foal calcium accretion. We hypothesised that in foals up to four months of age bone accrual would predominate over bone resorption, and that accrual and resorption would be associated with maternal bone turnover.

Serum osteocalcin (OC), a marker of bone accrual, and serum pyridinoline (PYD), a marker of bone resorption, were measured in mare foal pairs once monthly from shortly after birth until the foal was four months of age. OC and PYD were measured using Quidel MicroVue Osteocalcin and PYD assay kits (www.quidel.com).

In foals, the pattern of serum level of OC was non-linear, increasing rapidly after birth to a mean maximum of 122.1(95% CI:97.0 – 147.3) ng/ml at approximately two months, decreasing by 3 – 4 months. Foal OC was weakly correlated with mare OC, r=0.28, p=0.01. Foal PYD was high in the first 24 hours of life, mean 38.9 (95% CI:25.3-52.5) nmol/L, falling to 12.4 (95%CI: 5.5 – 19.2) nmol/L by approximately 3 weeks of age. There was no correlation between mare and foal levels of PYD in the four months after birth.

The rise in serum level of OC after birth suggests a period of rapid bone accretion in the first two months of life. The corresponding fall in PYD suggests little bone mobilisation in this neonatal period, providing evidence that bone accretion predominates in the first month of life, which is a critical period for bone accrual and skeletal mineralisation in foals.

GENE AND PROTEIN EXPRESSION FOLLOWING INJUREY TO THE SUPERFICIAL DIGITAL FLEXOR TENDON (SDFT)

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Tendon injury is a major source of loss in performance horses. The aim of this study was to compare connexin (Cx) and collagen gene and protein expression in spontaneously injured,and contralateral (opposite SDFT to injured SDFT) tendons with normal equine SDFT from horses matched for age-, sex- and performance..

Horses were clinically assessed and expression of 11 Cx isoforms in injured, contralateral and normal SDFT was investigated using PCR; products were sequenced to confirm identity. Gene expression of identified Cx isoforms and collagens I and III were measured using real time RT-qPCR. SDFT cryosections were labeled for Cx43 and 32 gap junction (GJ) proteins and procollagen amino--propeptides I (PINP) and III (PIIINP). Confocal laser scanning microscopy (CLSM) was used to obtain *z*-stacks of labeled SDFT and immunofluorescence was quantified using OBCOL, a custom ImageJ automated macro. Collagen content and solubility were determined by dye-binding assay. Linear regression analyses of log-transformed data were used

PCR identified Cx 26, 30, 31, 31.1, 32, 40, 43 and 50 in SDFT; no Cx was specific to injured SDFT. Cx43 gene expression was significantly upregulated in central ($p=0.006$), peripheral ($p=0.005$) and distant ($p<0.001$) regions of injured SDFT. Cx32 gene expression was significantly upregulated in central ($p=0.005$) and peripheral ($p=0.015$) regions of injured SDFT. Col1A1 gene expression was significantly increased in central ($p=0.001$) and distant ($p=0.019$) regions of injured SDFT. Col3A1 gene expression was significantly increased in central ($p<0.001$), peripheral ($p=<0.001$) and distant ($p=0.001$) regions of injured SDFT. Cx43 protein expression, quantified as number ratio (number of GJ *per* tenocyte nucleus) and average dot volume *per* nucleus (volume of GJ in pixels *per* tenocyte nucleus) was significantly ($p=0.009$; $p=0.002$) upregulated in injured SDFT. Areas peripheral to site of SDFT injury showed significant ($p=0.032$; 0.011) upregulation. Central regions of contralateral SDFT showed a trend ($p=0.099$; 0.096) towards significantly increased Cx43 protein expression. Cx32 protein expression was significantly ($p=0.007$; <0.001) increased in injured SDFT and significantly ($p=0.001$; 0.026) increased in central regions of contralateral SDFT. PINP expression was significantly ($p=0.012$; 0.001) increased in central regions of injured SDFT. PIIINP showed markedly ($p=0.002$) increased average dot volume *per* nucleus in injured central regions of SDFT.

Upregulation of connexin (Cx43 and 32) and collagen (Col1A1/PINP and Col3A1/PIIINP) gene and protein expression were demonstrated. These changes were not restricted to ultrasonographically-detected SDFT pathology indicating diffuse homeostatic alterations in injured SDFT. Trends towards significance and significant changes in Cx and collagen gene and protein expression were also measured in contralateral SDFT. These homeostatic alterations are indicative of increased loading and support clinical observations of increased incidence of injury in contralateral SDFT following initial SDFT injury.

EVIDENCE FOR THE MANAGEMENT OF BACK PAIN IN HORSES BY MANUAL THERAPY

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Back pain is often an underlying cause of poor equine performance. The structure and stressors placed on the thoracolumbar spine predispose it to injury (Jeffcott, 1979). It is difficult to clinically assess back pain resulting in it sometimes being overlooked. There is much debate regarding its aetiology and pathogenesis. Manual therapy, including manipulation and mobilisation, is used by equine physiotherapists and chiropractors to manage thoracolumbar pain in the horse (Gomez-Alvarez et al 2008; Haussler et al, 2010). This paper highlights contemporary research that demonstrates the efficacy of manual therapy for treatment of thoracolumbar pain in the horse and discusses the proposed mechanisms of action of this approach to pain management

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THE IMPORTANCE OF EARLY EXERCISE IN THE HORSE

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Across all equestrian disciplines the single largest reason for wastage is musculoskeletal injury. It is therefore of importance that management and competition structures are in place to optimise the development of the equine musculoskeletal system to minimise wastage.

Data from other species and humans in particular, has demonstrated the benefit of early exercise and the dire consequences of inactivity. The horse has evolved as a cursorial animal capable of covering up to 10 km per day within 9 days of birth. Yet modern equine management systems restrict rather than promote early exercise. Foals had a positive response to early pre weaning paddock exercise (greater cartilage health) and more recent work has demonstrated that exercise over and above that normally occurring with pasture reared foals, introduced as early as 3-weeks-old, had positive effects on the equine musculoskeletal system. The response of juvenile horses to additional exercise is due to the tissue being responsive to priming. Epidemiological data indicates that the window for tissue modification may still be open when the horse is a yearling and even as a 2-year-old.

However, the method in which the exercise is applied may be of as much importance as the timing of the stimuli. A recent prospective study of both Thoroughbred and Standardbred horses demonstrated that the horses that entered training as 2-year-olds had longer and more successful racing careers than those that entered training later in life. It would appear that even the initial stages of training are enough to provide a positive stimulus, as horses first registered with a trainer at two years old had the same advantages as those that raced as 2-year-olds.

The physiological, clinical and epidemiological data indicates that, rather than restrict exercise and the use of horses at a young age, we should realign expectations with the capability of the horses' musculoskeletal system and evolutionary template to maximise orthopaedic health.

VARIABLES INFLUENCING THE AUCTION SALES PRICE OF NEW ZEALAND THOROUGHBRED YEARLINGS

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Data were obtained from the New Zealand Thoroughbred studbook and the online sales results for the 2004 National Yearling Sales Series. Across the three sales categories, 477, 523, and 354 yearlings were offered at the Premier, Select and Festival sales, respectively. There were significant differences in the representation of yearlings across the three sales categories based on: sire service fee, vendor category, purchaser country of origin, and dam age. There were fewer fillies catalogued in the Festival sale compared to Select or Premier ($P<0.05$). Within the general linear model, auction price (log10) was significantly influenced by sales category: Premier ($71,285 95%CI $61,801 -$82,224), Select ($24,831, 95%CI $22,233 -$27,733) & Festival ($9,462, 95%CI $8,072 -$11,091, $P<0.001$). Sales price (log10) was also significantly influences by sire service fee (<$5,000, $5001-$15,000, $15,001-$30,000, & >$30,000)), vendor category (number of yearlings in Premier sale, 0-1, 2-5, 6-11, >13), purchaser country of origin (Australia, New Zealand, Rest of the World), and mare age (<5, 5-12, 13-17, >17 yrs). Colts sold for more than fillies ($29,040, 95%CI $26,668-$31,695 vs. $22,542, 95%CI $20,370-$24,945, $P<0.001$) across all sales categories. There was an interaction of sales category and gender ($P=0.003$), with the premium paid for a colt increasing from Premier through to Select sale. These results indicate that in order to optimise gross returns, vendors require colts that are marketed through the Premier sale, from older established broodmares, with a pedigree that appeals to Australian buyers.

DOES PASTURE AVAILABILITY INFLUENCE HENDRA VIRUS INFECTION OF GRAZING HORSES?

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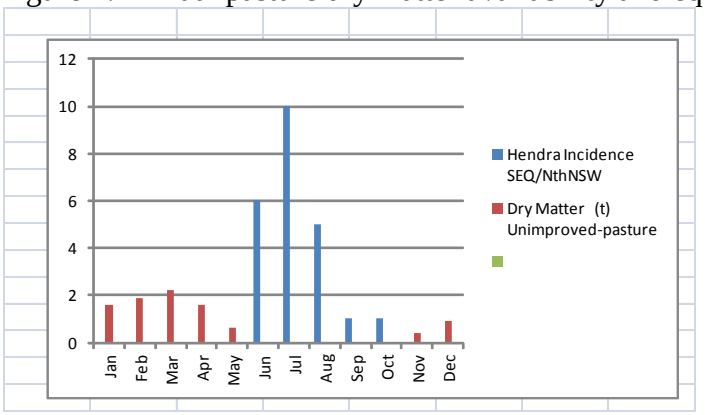
Hendra virus (HeV) is a serious infectious disease of horses and a fatal zoonosis for humans and although the incidence is low, when infection does occur it has very high mortality rates in excess of 70% and 50%, respectively (see Daszak *et al.*, 2006) Since the discovery of HeV some 15 years ago, considerable information on the virus *per se* has been obtained and its reservoir host, all four species (*Pteropus* spp.) of flying fox or fruit bats, has been established. (Plowright *et al.*,2008; McFarlane *et al.*, 2011).

Despite the significant research achievements, the dynamics of horse infection from flying foxes is poorly understood. There has not been a detailed characterisation of horses and their management prior to and up to disease occurrence. In conjunction with the gathering of this information, there is also a need for data that describes the environment in which the HeV spill-overs occur, including property aspect, horse facilities and design, topography, landscape features, pasture cover, vegetation, water supply, herd dynamics and other key horse behavior influencing factors. The year 2011 provided an unprecedented opportunity to undertake a comparative analysis of this data on Hendra infected properties. With this in mind, we have established the location of the HeV infection sites interviewed attending veterinarians, collated local climatic conditions prior to infections and assessed likely forage availability and pasture dynamics in 2011. Initial results are shown in Figure 1.

Figure 1: Annual pasture dry matter availability and equine cases of HeV infection in 2011.



From this preliminary data there appears to be a strong association between pasture availability and infection of horses with HeV. During 2011 climatic factors that influenced forage growth and quality were severe with long frost events at 3 sites and an extended rain event at 2 sites. Poor pasture quality and availability and changes to the pasture species mix due to climatic conditions would significantly reduce nutrient intake and horse grazing patterns. In these circumstances, horse behaviour is significantly influenced and the wellbeing, including the immune system, of the horse would be compromised. This is supported by information presented at industry seminars conducted by the Queensland Government that on most infected properties horses were not receiving adequate supplementary feeding (Dr Nina Kung, November, 2011). In addition a NSW DPI communiqué to industry on HeV infections speculated that horses are likely have been hungry at infection. A hungry horse is more likely to consume bat related material found on pastures setting the scene for a HeV spill-over. Moreover, the management of horses and horse property circumstances has a significant effect on the behaviour of horses, including foraging and feeding behaviour.

The results highlight the possibility that the equine management environment is likely to be a significant factor in the infection of a horse by HeV. There is a need for horse and horse related data to overcome a primary deficiency to further understanding the ecology of Hendra transmission and the risk factors involved. In particular, it will allow improved modeling of HeV risk, provide insights into the flying fox horse interaction, and facilitate development of strategies to prevent HeV transmission to horses.

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McFarlane R, Becker N, Field H (2011) Investigation of the climatic and environmental context of Hendra Virus spillover events 1994–2010. *PLoS ONE* 6(12): e28374. doi:10.1371/journal.pone.0028374

Plowright RK, Foley P, Field HE, Dobson AP, Foley JE, *et al*. (2008) Urban habituation, ecological connectivity and epidemic dampening: the emergence of Hendra virus from flying foxes (*Pteropus* spp.). *Proceedings of the Royal Society B*,275: 861-869.

COMPARISON OF BIOELECTRICAL IMPEDANCE WITH OTHER METHODS FOR ASSESSING ADIPOSITY IN HORSES AND PONIES CHANGING FROM MODERATE TO OBESE BODY CONDITION

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Equine obesity is becoming increasingly common in the domestic horse population worldwide (Stephenson et al, 2011). The main concern with obesity in horses is its strong association with increased risk of disease, particularly insulin resistance and laminitis. Most current methods for assessing obesity / adiposity in horses and ponies tend to be subjective, consider mainly subcutaneous fat, and may vary in accuracy between breeds and types of animal. Bioelectrical impedance analysis (BIA) is a non-invasive method of assessing percentage body fat and has been previously evaluated in horses (Van der Aa Kuhle et al, 2008). The objectives of this study were 1: to further investigate the utility of bioelectrical impedance analysis (BIA) to estimate total body fat (using the deuterium dilution method of Dugdale et al. as the gold standard); 2: to use BIA to estimate body fat content in horses and ponies as they increase in adiposity; and 3: to compare it with other methods of assessing body condition and adiposity.

Eighteen horses and ponies were assigned to either a weight gain group (4 Standardbreds, 4 Andalusians and 4 ponies) or weight control group (2 Standardbreds, 2 Andalusians and 2 ponies). At the start of the study, BIA (Impedimed SBF7 BIA device) was compared against body fat percentage as determined by deuterium dilution technique (previously validated in ponies; Dugdale et al. 2011). Over 22 weeks the horses and ponies in the weight gain group were fed a diet containing increasing amounts of granulated vegetable fat and canola oil (increasing up to 200g/100kg BW), in order to provide ~210% of estimated maintenance energy requirements. Body weight, body condition score (BCS), cresty neck score (CNS), fat depth over ribs and rump (measured by ultrasound), heart and belly girth (Dugdale et al, 2011) were measured each week in addition to BIA.

For BIA measurement, electrode placement at the poll and tail head gave reproducible results, however the use of needle electrodes was poorly tolerated by the subjects. Therefore ECG electrodes (Ag/AgCl) were found to be a viable and useful alternative. The intra-assay coefficient of variation (n=100 measurements) was 9.4%. BIA was shown to correlate moderately well (r^2=0.65; P=0.001) with deuterium dilution determination of total body fat. Body condition score (modified Henneke system) showed a slightly weaker correlation (r^2=0.54; P=0.003) and there was no significant correlation with cresty neck score.

BIA shows potential for being developed as a non-invasive, practical and accurate tool for veterinarians and nutritional consultants for assessing adiposity in horses and ponies. However, a lot more work is required before it can be used routinely in practice; for example the optimum values for the variables used in the impedance equations for calculating % body fat may differ between types of animals.

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Stephenson HM, Green MJ, Freeman SL. (2011). Prevalence of obesity in a population of horses in the UK. *Vet Record* 168(5):131.

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MARES OF LOWER BODY CONDITION SCORE EXHIBIT MARKED INSULIN RESISTANCE AND REDUCED INSULIN SECRETION IN LATE GESTATION

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In pregnancy dramatic changes in maternal metabolism are necessary to ensure normal development of the foetus. Partitioning of nutrients from maternal tissues to the feto-placental unit is associated with decreased insulin sensitivity, increased basal insulin secretion, enhanced β cell responsiveness, decreased insulin clearance, and higher post-prandial glucose and insulin responses. Such metabolic changes can be influenced by diet. Previously (Dobbs *et al.* 2012), we noted that feeding a high energy diet to mares in the last trimester of pregnancy increased basal insulin and glucose concentrations, but did not affect insulin sensitivity, glucose effectiveness, or β cell responsiveness. However in that study, only small differences in bodyweight and body condition were obtained. Therefore, in this follow-up study, we examined insulin and glucose responses in pregnant mares with disparate body condition.

Pregnant (*n=13)* mares were used in the study and given either a high (HE: *n=6*) or low energy (LE: *n=7*) diet for the last trimester of pregnancy. Body condition score (BCS) was assessed on a scale of 1-9. Insulin-modified frequently sampled intravenous glucose tolerance (FSIGT) tests were performed on the pregnant mares on Day 290 and Day 320 of gestation. Minimal model analysis was used to determine insulin sensitivity (Si), glucose effectiveness (Sg), acute insulin response to glucose (AIRg), and disposition index (DI).

There were no significant differences in bodyweights between HE and LE fed mares in late gestation. However BCSs in HE fed mares were significantly ($P<0.001$) higher than LE fed mares on Days 290 and 320. Further BCS significantly ($P<0.05$) increased in HE fed mares between Days 290 (6.3 ± 0.4) and 320 (7.2 ± 0.5), whereas in LE fed mares BCS significantly ($P<0.01$) decreased over these days (4.1 ± 0.3 vs. 3.3 ± 0.3). Basal insulin and glucose concentrations were not significantly between HE and LE fed mares on either Day 290 or Day 320. In HE mares, minimal model parameters did not significantly change from Day 290 to Day 320. On Day 320 values in HE fed mares were Si (1.1 ± 0.4, $x10^{-4}L.mU^{-1}.min^{-1}$), Sg (2.3 ± 0.4 x 10^{2}. min^{-1}), AIRg (347.5 ± 60.8 min.mIU/L) and DI (282.9 ± 79.7). In comparison, insulin sensitivity and the disposition index in LE fed mares were significantly ($P<0.01$) lower on both Day 290 (Si 0.2 ± 0.2, $x10^{-4}L.mU^{-1}.min^{-1}$; DI 33.9 ± 28.8) and Day 320 (Si 0.04 ± 0.01, $x10^{-4}L.mU^{-1}.min^{-1}$; DI 4.3 ± 2.7) compared to HE fed mares. Further the acute insulin response to glucose was similar between the mare groups on Day 290, but significantly ($P<0.01$) lower in LE mares compared to HE mares on Day 320. In contrast, glucose effectiveness was not different at any time.

Overall the results indicate that LE fed mares became insulin resistant in late pregnancy and exhibit diminished β cell responsiveness. With increasing insulin resistance in pregnancy, the expected homeostatic response is enhanced β cell secretion to compensate for decreased insulin-mediated glucose uptake. However we observed inadequate acute β cell secretion in LE fed mares. Such a defect in pancreatic β cell function, accompanying severe insulin resistance, is a prime characteristic of human gestational diabetes mellitus (GDM). Lean women with GDM exhibit pronounced insulin resistance and inadequate insulin secretion (Kautzky-Wller *et al*. 1997), similar to our LE fed mares with low BCS. These novel results highlight the importance of BCS in metabolic responses during pregnancy.

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Kautzky-Wller A. *et al*. (1997). *Diabetes Care* 20: 1717-1723

FISH OIL SUPPLEMENTATION ATTENUATES ABNORMAL GLUCOSE CLEARANCE CAUSED BY HIGH DIETARY FAT INTAKE IN AGED THOROUGHBRED GELDINGS

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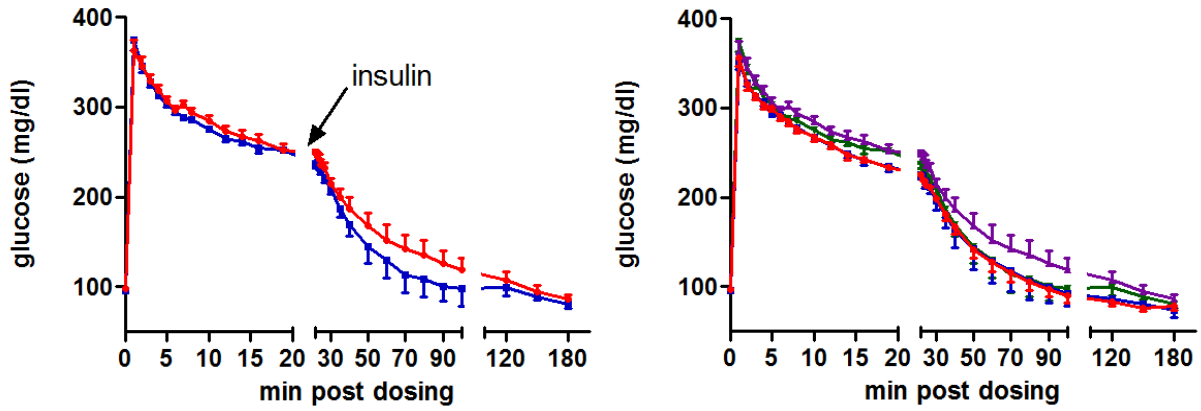
Fats and oils are a good source of energy for horses, but there is a link in some species between high fat diets and insulin resistance (Frank, 2009). Long chain omega 3 fatty acid supplementation (DHA and EPA) prevented the development of insulin resistance and improved insulin sensitivity in some species (Kalupahana et al, 2010) and previous work in horses, indicated fish oil (as a source of DHA and EPA) supplementation moderated glucose response to a grain meal, but did not affect insulin sensitivity (Hoffman et al, 2011). The aim of this study was to investigate the glucose dynamics in horses fed a high fat or moderate carbohydrate diet and to investigate if fish oil moderated these effects.

Four aged, non-obese Thoroughbred geldings (21.5 yrs ± 3.32 yrs; Weight 572.16 Kg ± 50.53 Kg; BCS 5.0-6.0) were used in a 4x4 Latin square design study. The treatments consisted of 1.5% BW grass hay and 120g/d vitamin/mineral supplement and either 1. Oats (4.46g/kg BW) + 60g corn oil (OAT+CO) 2. Oats (4.46g/kg BW) + 60g fish oil (KERx EO3) (OAT+FO) 3. Hay cubes (2.59 g/kg BW) + soya oil (0.86 g/kg BW) + 60g corn oil (FAT+CO) 4. Hay cubes (2.59 g/kg BW) + soya oil (0.86 g/kg BW) + 60g fish oil[1] (FAT+FO). At the end of each 4 week treatment period, a frequently sampled glucose insulin test (FSGIT) was performed with 300 mg/kg glucose solution (50% dextrose) administered IV and frequent blood samples drawn for 180 minutes. At 20 minutes post glucose infusion, 20 mU/kg insulin (Humulin R) was administered. Plasma samples were tested for triglycerides, insulin and glucose. Data for the two OAT treatments, the two FAT treatments, and the differences between these were analysed using ANOVA and results presented at the 5% significance level.

Horses fed a high fat diet (FAT+CO) had decreased glucose clearance following a FSGIT compared with horses fed a moderate carbohydrate diet (OAT+CO) ($p<0.05$). Fish oil significantly affected glucose clearance in the high fat diet ($p<0.05$) (Figure 1a), but had no effect on glucose clearance in the moderate carbohydrate diet. Glucose clearance in the FAT+FO diet was not significantly different to either the OAT+FO or OAT+CO diets (Figure 1b). Moderate carbohydrate equates to 2.0-2.5 kg/d of grain and a high fat diet equates to about 500g/d of oil per day (for a 500kg horse).

Figure 1a Plasma glucose concentrations (mean ± SD) after a FSGIT in horses fed a high fat diet supplemented with either 60g of corn oil (FAT+CO) (■) or 60g of fish oil (FAT+FO) (■). **Figure 1b** Plasma glucose concentrations (mean ± SD) after a FSGIT in horses fed FAT+FO (■), FAT+CO (■), OAT+CO (■), and OAT+FO (■).



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OSTEOCHONDROSIS IN YEARLING STANDARDBRED HORSES AND SUBSEQUENT RACE EARNINGS AND NUMBER OF STARTS AT TWO AND THREE YEARS OF AGE

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Osteochondrosis (OC) is an orthopedic disease involving failure of endochondral ossification of the cartilage precursor to bone. The current study tests the hypothesis that yearling Standardbred horses that were found to have radiographic abnormalities related to OC will have reduced two and three year old earnings and starts. This study also looks at the relationship between the horses with osteochondrosis that had surgery compared to the horses that did not, to determine the longevity of their careers. This study also examined the relationship between the sire of the yearlings and the occurrence of OCD.

Eight hundred nineteen yearling Standardbreds, 354 born in 2005 and 465 born in 2006 were used. Each horse had fluoroscopic radiograph examinations of stifles, hocks, front and rear fetlocks, and knees performed by the same Veterinarian, trained in the detection of OC with the fluoroscope. For each horse the number of starts at 2, 3, and 4 years of age, earnings, and OC status were recorded. Determination of association between OC status and starts was done using chi-square. The statistics were computed using PROC FREQ in SAS Version 9.2. Comparison of distribution of earnings by OC status were computed using PROC TTEST in SAS Version 9.2 (2008, SAS Institute Inc., Cary NC). In the second part of this study, 364 questionnaires were mailed to owners of horses found to have OCD, to determine the rate of surgical removal of OCD fragments so that we might compare differences in starts and earnings between OCD positive horses with or without surgical intervention during their racing careers to-date.

Of the 819 Standardbred horses that were examined fluoroscopically, 462 horses were determined to be free of OC and 357 were determined to have some form of an OC lesion affecting at least one limb. There were 597 horses that made at least one start at 2 and/or 3 years of age. Of the horses not affected with lesions, 75% made starts compared to 70% that were affected with lesions. Based on the chi-square there was not a significant association (P=0.10) between OC status and starts. Horses without lesions had a higher average number of starts (14.74) compared to horses with lesions (13.52) but again this was not significantly different (P=0.19). Horses without lesions had a higher average two-year old earnings ($9,695) compared to horses with lesions ($6,259) but this difference was not statistically significant (P=0.17). Horses without lesions had a lower average three-year old earning ($12,515) whereas horses with lesions had average three-year old earning of ($14,396). These earnings differences between 3 year-old groups were also not significantly different (P=0.68). Horses without lesions had average total 2 and 3 year-old earnings of $22,210, compared to horses with lesions having average 2 and 3 year-old earnings of 20,655 (P=0.79). The average number of starts from the 4 year old racing year to date was 16.78 and the average number of earnings from 4 year old year to date was $16,245. Just 81 of the 364 surveys were returned; however, only 8 horses out of 81 did not have surgery. Therefore, it appears that the vast majority of horses in the population sampled had surgical intervention, making a comparison of surgical or non-surgical OCD management impractical using our current data set. An analysis of OCD incidence by using sires with more than three observations resulted in p-values that ranged from 0.82 to 0.98. No evidence was found that that sire was related to OCD in this study.

The results of this study found that there was no difference in the number of starts or earnings between horses that had OC lesions and horses that did not, and that the sires included in this study were not different in the OCD rates of foals. Most owners of OCD afflicted yearlings choose to remove OCD fragments surgically, making it difficult to compare surgical and non-surgical OCD management.

MICRONISED AND STEAM FLAKED GRAIN STARCH DIGESTIBILITY: VARIATION BETWEEN PROCESS AND MANUFACTURER

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The digestibility of the starch fraction of cereal grains fed to horses is an important consideration when developing feed rations for equines. Grains that have been 'cooked' in some way using thermal or hydrothermal processes allow more starch to be digested in the small intestine (Julliand et al 2006). Digestion in the small intestine is better for two reasons. First, starch that is digested in the small intestine is energetically more efficient for the horse than starch fermented in the hindgut. Second, it reduces the amount of starch flowing to the hindgut and causing hindgut disturbances through the rapid fermentation of starch by amylolytic bacteria (Richards 2003).

Starch digestibility is a difficult parameter to measure with post mortem collection, fistulated animals, the mobile bag technique and glycaemic response all being employed in an attempt to measure starch digestibility (Julliand et al 2006). An *in vitro* starch digestibility assay was developed that showed a positive correlation with starch digestibility measured indirectly via the glycaemic response (Richards 2003). This assay can be used as a tool to compare the digestibility of grains intended for horses. Recently, a small stockfeed manufacturer located in central Victoria employed this method to compare micronised and steam flaked barley and corn from five different manufacturers. The results are presented in the table below.

Table 1: The total starch and percent of that starch digested in 15 minutes *in vitro* when incubated at 37°C with amylase and amyloglucosidase for corn and barley from five different manufacturers (M1 – M5).

Manufacturer	Grain	Processing Method	% Total Starch	% Starch Digested
M1	Barley	Steam Rolled	46.0	34.2
M2		Steam Rolled	44.9	35.2
M3		Micronised	47.9	59.5
M4		Micronised	52.9	72.7
M5		Micronised	55.7	64.1
M2	Corn	Steam Rolled	63.8	53.6
M4		Micronised	69.0	83.6
M5		Micronised	66.4	56.2

The *in vitro* digestibility of micronised barley and corn varied by 13% and 27% respectively between manufacturers. Micronised grains were an average 26.2% more digestible than steam rolled grains. This variation in starch digestibility between processing methods and manufacturers will have implications for feed use efficiency and hindgut health in horses.

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TAGASASTE VERSUS GOLDEN BAMBOO: WHICH DO HORSES PREFER?

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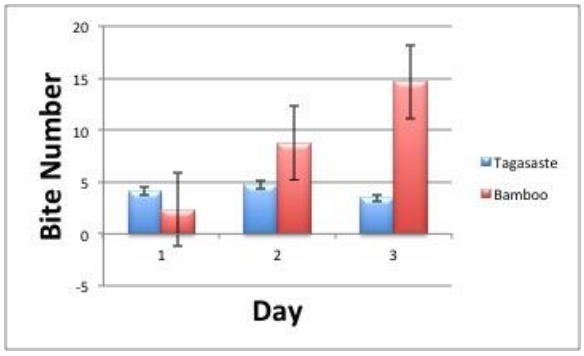
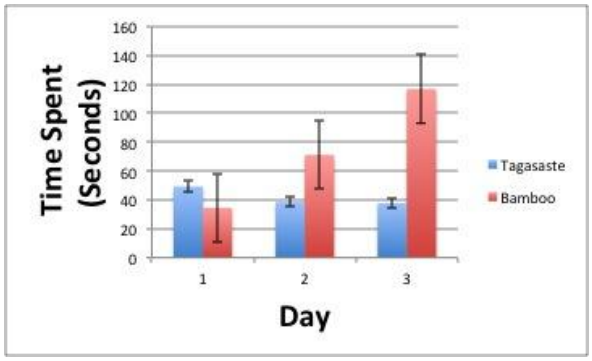
Preference testing is an important tool for evaluating novel feeds for their acceptance and relative palatability in horses. In this study we examined 2 novel forages with the aim of determining the preferred forage and to examine the variability in preferences exhibited between individual horses as measured by the methodology used in this study.

Twenty horses from 12 properties were subjected to a 2-choice preference test for 3 consecutive days. Forages tested were Tagasaste (*Chamaecytisus palmensis*) and Golden Bamboo (*Phyllostachys aurea)*. Horses were tested at their usual residence in pairs or singly. Fresh forages (200 - 400 g) were presented 2 metres apart and positions alternated daily (left or right) to remove chance of side preference (Arave, 1996). Horses' interactions with the forages were observed and videoed for 5 minutes following first approach. Animal preferences were determined from the average time spent eating each forage and the number of bites taken (Figures 1 and 2). Our results indicate an initial preference for Tagasaste on day 1 followed by a greater preference for Bamboo by day 3. The horses tested in pairs showed a dominance hierarchy, with the lowest ranked member of the two horses hanging back in most cases until the dominant horse had moved away. Spatial recognition and memory were also observed with a number of horses going straight to the spot where their preferred forage had been presented the day before.

Very little research has been reported with regards to forage preferences in horses and therefore a lack of information for horse owners wanting to utilise forages to supplement pastures in drought and for behavioural enrichment. Further research is needed into more types of fresh forages that can be fed to horses, and their palatability.

Figures 1 and 2. Average number of bites and time spent eating different forages offered to 20 horses in a 2-choice preference test.





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A CASE OF LEUCAENA LEUCOCEPHALA POISONING IN A THOROUGHBRED GELDING.

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Leucaena leucocephala is a legume native to Central America (Belize and Guatemala). It was introduced into Australia in the 1960's as a fodder plant, primarily for cattle. *Leucaena* has spread widely in Queensland. *Leucaena* contains a toxic, non-protein amino acid, mimosine, an analogue of tyrosine (Shelton and Dalzell, 2007). Although toxicity is rare in ruminants, monogastric species cannot degrade mimosine. Mimosine causes depressed T3 and T4 levels resulting in goitre formation and disrupts DNA synthesis and repair thus targeting tissues with high mitotic rates such as continuously growing hair and hoof tissue. Other reported clinical signs include stomatitis, haemorrhagic enteritis, and proctatitis, oedema of the hind limbs and genitals and 'laminitis' (Kraneveld and Djaenoedin, (1950), Mullenax, CH (1963), Owens, (1958)) Reports of symptoms of toxicity in horses are rare and reports are anecdotal and incomplete. The main reported symptoms are hair loss and 'laminitis'.

An aged thoroughbred gelding gained access to a number of branches of *Leucaena leucocephala*, heavily laden with pods, which had been cut several days earlier. Within a five days of presumed ingestion, the horse developed lameness with a stance characteristic of laminitis. The lameness progressed and about a week later the coronary bands sloughed and the horse was severely lame. At this time, the horse's mane and tail fell out. While the coronary bands reformed within weeks, the disruption to hoof wall production resulted in a complete separation between new hoof growth and the old hoof resulting in wall instability. Approximately one month after ingestion, it was apparent that the soles of the fore hooves had separated. The soles were easily removed revealing a new layer of thin, soft corium beneath. At no stage of the disease was rotation or sinking of the pedal bone, characteristic of laminitis, demonstrated on progressive radiographs. At three months and 5 months, venograms revealed compromised perfusion of the tissue beneath the distal phalanges of the fore hooves on weight bearing. This was associated with extreme lameness and development of sole abscesses.

Many attempts were made to stabilise and protect the hoof. Pain management of this horse throughout 5 months of recovery was the greatest challenge. Phenylbutazone was administered at 2 gm twice daily for much of this time. The horse grew essentially normal hooves and returned to light work approximately ten months after ingestion.

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Mullenax, C.H., (1963) *Australian Veterinary Journal*; 39, 88-91.
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EFFECT OF DEXTROSE SUPPLEMENTATION ON ELECTROLYTE AND WATER ABSORPTION IN RESTING THOROUGHBREDS

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Electrolytes are a critical component of a performance horse's feeding program since they play an important role in maintaining osmotic pressure, fluid balance, and nerve and muscle activity. During exercise, sodium (Na^+), potassium (K^+), and chloride (Cl^-) are lost in large quantities through sweating. Loss of these electrolytes causes fatigue and muscle weakness and decreases the thirst response to dehydration. Unfortified hay and grain rations tend to be deficient in sodium and contain variable quantities of chloride. If forage intake is restricted (less than 1% BW on a dry matter basis) and/or high sweat losses occur then additional potassium supplementation may also be necessary.

Commercial electrolyte products often contain sugar (dextrose), which is purported to improve electrolyte uptake in horses. Two studies were conducted by KER to evaluate if sugar inclusion affects electrolyte and water uptake and retention in idle horses. In study 1, four Thoroughbreds (Age 6.25 yrs ± 2.25 yrs; Weight 574.4 kg ± 82.4 kg) were used in a 4 X 4 Latin square design trial. The horses were dosed with 92 grams of electrolyte (72 g NaCl, 20 g KCl) either alone (elect), with 10 g of dextrose or 100 g dextrose. The electrolyte mixes were dissolved in 1 liter of water and administered via nasogastric tube. A fourth treatment of 1 liter water with no added electrolytes or dextrose served as a control. Plasma samples were taken before and for 4 hrs post dosing and Na^+, K^+, Cl-, BUN, and glucose were measured. The horses were offered water free choice and hourly water intake was measured for 4 hrs dost dosing. Plasma Na^+ and osmolality were significantly elevated post-dosing in all three electrolyte treatments compared to the control ($p<.05$), but dextrose did not affect the rate or duration of increase. All electrolyte treatments increased voluntary water intake for the first 4 hr post dosing compared to the control ($p<.05$). Water intake equaled 0.3 ± .4 l, 5.3± 3.6 l, 5.4 ± 2.3 l, and 4.7 ± 2.5 l in the control, elect, 10 g dex and 100 g dex treatments, respectively.

A second 4X4 Latin square trial was conducted with 4 Thoroughbreds (Age 6.25 yrs ± 2.25 yrs; Weight 546.6 kg ± 35.2 kg). The horses were administered 1mL distilled H_2O/100g BW + 0.15g/Kg BW D_2O via nasogastric tube either 1) alone (control), 2) with 70 g NaCl + 30 g KCl (elect), 3) electrolyte + 10 g dextrose (dex) or 4) electrolyte + 10 g starch (starch). Blood samples were taken immediately before and .5, 1, 2, 3 and 4 hrs post dosing and D_2O, Na^+, K^+, Cl^-, BUN, and glucose were measured. Plasma D_2O at 2 hrs post dosing was used to calculate total body water. Total urine and faecal excretion was measured for 24 hrs before dosing and at 12 hr intervals for 72 hrs post dosing. Plasma Na^+ and osmolality were significantly elevated post-dosing in all three electrolyte treatments compared to the control ($p<.05$), but neither dextrose nor starch affected the rate or duration of increase. Plasma D_2O was elevated to a greater extent ($p<.05$) in the control compared to the 3 electrolyte treatments at 30 and 60 min post dosing suggesting that isotonic electrolyte solutions delay water uptake compared to pure water. Total body water was unaffected by treatment and equaled 59.1 ± 6.3, 60.3 ± 5.3, 62.6 ± 7.7 and 58.4 ± 4.7 ml/kg BW for the control, ELECT, dex and starch treatments, respectively. Urinary and faecal electrolyte excretion was not different between the 3 electrolyte treatments. These studies suggest that adding dextrose or starch to electrolyte mixes does not increase rate of absorption or retention of electrolytes. Dextrose may still have some value in improving palatability of electrolyte mixes, but the higher the dextrose content, the lower the electrolyte content of a product. This means high dextrose products supply lower amounts of electrolytes per kg, and may be less effective as a result.

EFFECT OF WATER DISPERSIBLE NATURAL VITAMIN E ON SERUM AND MUSCLE α -TOCOPHEROL IN VITAMIN E DEFICIENT QUARTER HORSES

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Vitamin E functions as a biological antioxidant, preventing the oxidation of unsaturated lipids within cellular and subcellular membranes by neutralising production of free radicals and serves to maintain normal neuromuscular function. Several specific equine diseases develop in the face of vitamin E deficiency including nutritional myodegeneration, neuroaxonal dystrophy and equine degenerative myeloencephalopathy in young animals. Adult horses deficient in vitamin E may develop a vitamin E deficient myopathy or equine motor neuron disease. Treatment with vitamin E is usually instituted in an attempt to reverse clinical signs. In addition vitamin E is often supplemented in horses with various forms of tying up and nervous disease. However, selecting the type and amount of vitamin E to supplement can be challenging because the bioavailability and potency varies widely among commercial supplements. Water dispersible natural vitamin E sources have previously been shown to have much greater bioavailability than synthetic forms based on dose response studies (Fig 1) and water dispersible vitamin E levels have been shown to increase CSF vitamin E levels to a greater extent than synthetic sources. No studies have evaluated the effect of water dispersible vitamin E supplementation on serum and muscle α-tocopherol in vitamin E deficient horses.

A supplementation study was conducted in 5 vitamin E deficient Quarter Horses aged 4-10 yrs. Horses were selected based on serum vitamin E < 2 µg/ml with no clinical or histopathologic evidence of neuromuscular disease. For four weeks prior to the study and during the 6 week period, horses were fed approximately 1.5% of body weight in grass hay and 2 kg of sweet feed. They were exercised 20 min per day at a walk, trot and canter 5 days per week on a treadmill. Supplementation of 5,000 IU of nano dispersed natural α-tocopherol (KERx Nano•E[a]) was provided in the morning feed 5 days a week. Prior to and on the last day of the period, serum and muscle samples were obtained for α-tocopherol measurement. The gluteus medius muscle was sampled at a standard site. At least 250 mg of muscle was immediately frozen in liquid nitrogen and submitted for α-tocopherol analysis by HPLC. Serum concentrations of vitamin E increased significantly by two fold (p= 0.0037) and muscle concentrations increased by over 3 fold (p=0.05) (Figure 2). The results of this study show that high doses of water dispersible natural vitamin E can increase α-tocopherol levels in muscle of deficient horses.

Fig 1. Change in serum vitamin E level in normal horses after supplementation with 5000 IU of synthetic (●) or nano dispersed natural vitamin E (▲)

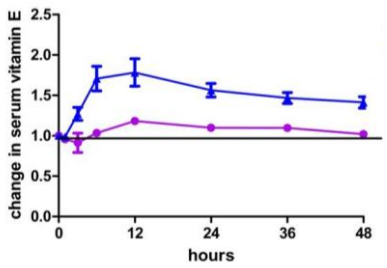
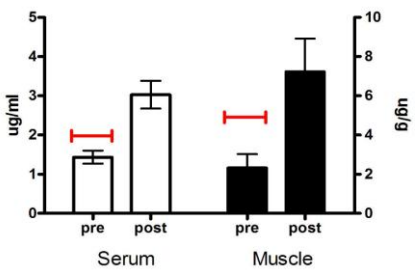


Fig 2. Serum and muscle vitamin E levels in vitamin E deficit horses before and after 6 weeks of supplementation with 5000 IU nano dispersed natural vitamin E (— normal levels).



THE USE OF BITS IN THE GENERAL HORSE POPULATION IN NEW ZEALAND

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The aim was to describe the type of bits used by riders in the general horse population in New Zealand. Data were collected via an online survey from 566 riders and 971 horses. Most respondents were females, from the Northern Districts and riding for >20 years. The 'Common Competitive' was the most frequently reported rider discipline, with most respondents riding at a novice/lower level. Most respondents were riding 1 or 2 horses aged between 7-12 years. Overall, 75% (727/971) of horses were Sport breeds, and 50% of horses were used for 'Common Competitive' discipline and 31% (298/971) for recreation. The most common bit used was a straight bar snaffle bit (85%) of medium thickness used in conjunction with a Cavesson (60%568/1113) or Flash (32% 355/1113) noseband. The most common reason for bit choice cited was "horse worked best in this bit" (60% 677/1120). Over half the respondents (59% 244/412) thought there should be greater flexibility in the type of bit and bridle permitted for competition use.

Bitless bridles were used on 212/1,120 (19%) respondents horses with the most common reason for use being "horse worked best in this bit" (41%) and 28% cited the choice was due to welfare reasons.

STRESS RESPONSES IN YOUNG, STABLED HORSES CAN BE MODIFIED BY MUSIC

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Horses are subjected to stress when they are weaned and first stabled. We investigated the possibility of mitigating such stress by the use of music therapy. Twelve weanlings were divided into two groups and subjected to alternate weekly periods of calming music played in the stable complex between 0930 and 1530 h or no music, in a cross-over design. Music did not affect median heart rate or heart interval, but it significantly reduced heart rate variability. Behavioural measurements indicated that stabled weanlings were more relaxed, spent more time eating, and less time walking in the stable with music compared to no music. Almost two thirds of horses received the lowest score in a stress measure when music was played, compared with one third of those without music. Music also attenuated peak heart rate and the duration of increased heart rate when weanlings were exposed twice a week to a stressful event, the presence of nearby stallions.

Stress management is frequently a problem for young weaned horses particularly when they are stabled. The results of this study suggest that stress responses in weanlings can be modified by playing music whilst in stables. The application of music therapy to enrich the equine environment is an area that shows promise and requires further study

THE IDEOLOGY OF BREEDING AND ITS IMPACT ON BEHAVIOUR IN THE THOROUGHBRED RACING INDUSTRY

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Despite strong evidence to contrary, the thoroughbred racing industry holds that the potential racing abilities of untried yearlings can be predicted by the racing performances of close relatives. As a consequence, vast sums of money are paid for stallion service fees and well-related yearlings in the belief that breeding is a reliable way of identifying talented racehorses. This paper argues that this belief is an ideology and that it shapes industry behaviour in ways that rob the vast majority of its participants from realizing their horses' potential and as such, should be seen as a source of wastage because it impedes the delivery of value.

The efficacy of the belief that breeding is a reliable predictor of performance was questioned by Gaffney and Cunningham (1988), who found that less than 35% of the variance in racing ability can be explained by hereditary factors. More recently, Wilson and Rambaut (2008) argued that whilst breeding contributes to success because there is some genetic basis to performance, stallion fees do not predict racing success. As such, breeding is not a reliable predictor of performance and therefore can be consigned to the status of being an ideology in that it describes an illusory form of thought which departs from the criterion of objectivity (Macey, 2000). Marx argues that ideologies are not benign but provide legitimacy to actions and beliefs that disguise an exploitative relationship, suggesting that there are invariably winners and losers from such arrangements.

Large top-end commercial stud farms are the major beneficiaries of the breeding ideology because they supply of stallions which must accessed to gain entry into the select yearling sales where the median can be 10 times that of non-select sales. Sales companies also benefit from their commission for selling stock whose value is exorbitantly over-stated. Boutique stud farms may benefit in times of high demand, but a lack of numbers heightens their risk should one of their yearlings be less than ideally conformed or suffers an injury prior to the sale. A majority of yearlings sold at select sales in 2011 failed to meet their costs of production and this figure increases substantially for vendors at non-select sales because their "lesser-bred" stock are assumed to have limited potential at best.

The vast majority of trainers also fail to benefit financially from the breeding ideology. With the value pendulum swung firmly in favour of breeders, trainers can only charge a subsistence fee for their services. Also, trainers tend to have lower expectations of lesser-bred horses and this influences the opportunities afforded to them relative to horses that are regarded as being well-bred. Further, competent trainers, who are deemed to have options with regard to the horses they train, kow-tow to owners who are able to provide "well-bred" horses and readily sack lesser-bred horses should they fail to demonstrate considerable ability in their first preparation. Lesser-bred horses, if they are put in work at all, are consigned to less-than-competent trainers whose limited skills, low expectations and substandard operating environments diminish their horses' chances of ever racing at the highest level.

Owners also fail to benefit from the breeding ideology because they feel obliged to pay exorbitant sums of money to have a chance of racing a good horse and the low expectations of trainers of "lesser-bred" stock means that their horses are less likely to be given the opportunity afforded to well-bred horses.

It is found that the breeding ideology has a negative impact on all participants with the exception of the sales companies and the owners of the stallions whose services are required to access select sales. Unmasking this ideology is an important step in engaging more owners and enabling more participants to gain financial returns that reflect their investment of time and money. Research in devising more effective, scientific measures of predicting performance need to be conducted to ensure more value can be delivered to the vast majority of racing participants who are disadvantaged by the breeding ideology.

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PREDICTION OF PASSIVELY ACQUIRED IMMUNOGLOBULIN A CONCENTRATIONS IN THOROUGHBRED FOALS

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Ingestion and absorption of maternal immunoglobulin (Ig) via colostrum is important to protect foals against microbial infection. Methods of measuring Ig concentrations in foals generally rely upon a blood test taken up to 12-72 h post foaling, delaying intervention strategies to prevent the consequences of deficient Ig absorption. A proactive alternative would be the peripartum collection and testing of mares for Ig concentrations to anticipate concentrations in the foal, enabling earlier intervention. Temporal relationships among serum and milk IgG concentrations have been well studied. We chose to explore IgA concentrations in Thoroughbred mare-foal pairs to identify if periparturient foal or mare-associated factors were predictors of foal IgA concentrations. Complete veterinary records, blood and milk samples were collected from 57 Thoroughbred mare-foal pairs from one month before to two months post parturition (PP). Samples were tested using enzyme-linked immunosorbent assay (ELISA) for concentrations of IgA.

Median foal serum IgA concentration from 12 h to 60 days PP showed a pattern consistent with the response observed in previously published studies on foal IgG, peaking at 12 h then progressively declining to its lowest concentration at 24 days. Subsequent concentrations increased exponentially from 24 to 60 days PP. A decrease in median foal IgA concentration at 24 days was positively associated with the concentration found in the foal at 12 h. All foals reached a similar baseline concentration approximately 19-24 days PP. The gradual increase in foal IgA concentration after 24 days was independent of the earlier concentration in the milk, foal and mare serum. Temporal patterns for both milk and foal serum were similar, but there was no correlation present between these two samples beyond 12 h.

Mare serum IgA concentration up to 28 days before parturition was associated with foal serum at 12 h and with colostrum, with concentration then declining slightly from 28 to 60 days PP. Mare serum and milk IgA concentrations were significantly correlated from 19 to 60 days PP. Results of this study suggest that IgA concentrations in mare serum before parturition is a potential predictor of foal serum IgA concentration PP. The prepartum forecast of IgA concentrations in the foal may enable earlier intervention to reduce potential risks of sepsis via mucosal infection.

A LONGITUDINAL STUDY OF CLITORAL FOSSA MICROBIAL FLORA IN THOROUGHBRED BROODMARES

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Most studies of endometritis in mares have, understandably, concentrated on bacteria isolated from uterine swabs. Nevertheless, the same pathogens that cause endometritis are also found as commensals in vulval flora and may be associated with serious ascending and systemic infection in the mare and serious infection of foals born to the mare. Analysis of commensal flora in easily sampled sites such as clitoral fossae can give important information on carriage of bacterial strains with features such as virulence factors and antibiotic resistance as well as providing insights into the dynamics of the commensal microbial flora as a whole.

The study population was 12 thoroughbred mares of varying ages, parity, gravidity and in-foal status, all resident on the same farm for the study period. Clitoral fossa swabs were collected monthly each year from the study mares. Two swabs are collected at each encounter. One in standard Stuarts Transport medium (Copan) for bacterial culture; the other stored at -20 C for metagenomics testing at a later date. For standard bacterial culture, swabs are inoculated onto Blood, Maconkey, Colistin Nalidixic Acid (CNA) and Chocolate Agars. Culture conditions are aerobic; anerobic flora will be detected by metagenomics on stored frozen swabs. Identification and susceptibility testing is by agar dilution (replicator method) utilizing biochemical tests as well as Vitek and MALDI-TOF methods for identification.

Analysis of data from standard bacterial swabs for the first 12 months indicates: the most common organism isolated is Group C Streptococcus (GCS). Conventional methods identified these as *S.equisimilis* or *S.zooepidemicus*. MALDI-TOF identified some of these isolates as *S.equi*, although conventional methods and pulse field gel electrophoresis suggested this was an incorrect identification. Mucoid strains of GCS appeared in the herd, more commonly in mares with history of slipping or failure to get in foal. Only tetracycline and occasional erythromycin resistance was detected in these isolates. The second most common isolate was *E.coli*. Ampicillin and/or gentamicin resistance was present in isolates from 5 of the 12 mares.

It is concluded that Group C streptococci & *E.coli* are the most common organisms detected by conventional methods. MALDI-TOF appears to have trouble in correctly identifying GCS. Metagenomic testing is pending and should shed more light, on the longitudinal dynamics of the microbial flora.

BACTERIOPHAGE THERAPY FOR THE TREATMENT OF CHRONIC BACTERIAL INFECTIONS ON THE STUD FARM - CASE STUDIES

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Interest in the use of lytic bacteriophages (phages) as a therapeutic tool for the treatment of bacterial infections has been rekindled over the past decade. Phages are naturally occurring viruses that infect and kill bacteria with very high specificity, leaving the normal flora intact. This unique specificity conveys an obvious therapeutic advantage in situations where treatment with antibiotics has proven to be unsuccessful or the maintenance of the normal flora is vital to the recovery of the animal.

Bacterial infections of the genital tract are linked to reproductive dysfunction and economic loss in the equine industry. Treatment of infection caused by opportunistic pathogens such as *Pseudomomas aeruginosa* and *Escherichia coli* may be difficult as they are generally very resistant to common disinfectants. Genital decontamination and uterine infusions with antibiotics can sometimes be helpful when the bacteria are susceptible but systemic treatment with antibiotics is generally avoided since it has proved unsatisfactory in most cases. A more targeted antibacterial treatment would be advantageous to the equine industry and therefore the potential of bacteriophages for the treatment of infections in stud farms was investigated.

Preliminary *in vitro* studies showed that phage preparations developed in-house were highly effective in lysing a representative panel of 25 *P. aeruginosa* and 39 *E. coli* isolates obtained from infected broodmares and stallions during the Australian mating season in 2010. To test this concept *in vivo* a total of seven horses with chronic infections were treated with bacteriophages, either alone or in combination with antibiotics. Five of these were treated for *P. aeruginosa*, three broodmares with clitoral infection, one with a uterine infection and a 5-year-old stallion with reduced seminal quality. The remaining two horses, both mares suffering from endometritis caused by *E. coli*, were treated by intra-uterine lavage.

Local and oral antibiotic treatment alone had repeatedly failed to clear these infections despite the bacteria being sensitive to gentamicin or amikacin. After 3 to 7 days of bacteriophage treatment (alone or in combination with antibiotic), the six mares and the stud returned negative cultures. Both bacteria and bacteriophages numbers were monitored throughout the treatment. The results demonstrated a rapid increase in bacteriophage numbers as the counts of pathogenic bacteria declined. This was followed by a reduction of phages, which coincided with the disappearance of the target bacteria. The mares were subsequently mated while the stallion was able to return to service. One out of five treatments followed-up for over one year reported recurrence of the infection.

This study highlights the potential of bacteriophage therapy for the treatment of genital bacterial infection on the stud farm, but also raises the intriguing possibility of applying this technology to other more severe equine infections.

COMPARISON OF THE EFFECTS OF TREATMENT WITH PENTOSAN GOLD AND A STANDARD PENTOSAN PRODUCT, ON THE SYMPTOMS OF JOINT DISEASE IN HORSES.

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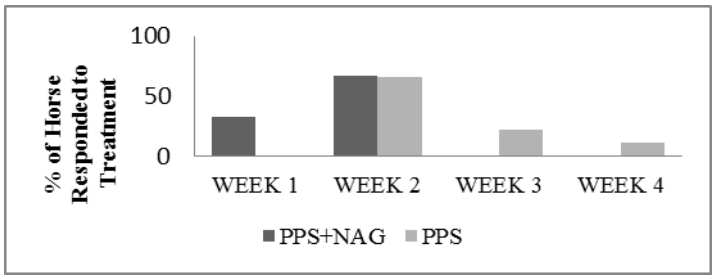
The efficacy of a new combination product for management of inflammatory joint disease in horses Pentosan Gold® injection (PG), which contains as active ingredients Sodium Pentosan Polysulfate (PPS) and N'acetyl glucosamine (NAG), was compared with that of a standard product containing only PPS. Twenty one field cases, presented to veterinarians for joint disease were used in the trial. Horses were examined and treated with either PG or PPS, with random allocation to the treatment group. The veterinarians treating the horses were asked to report on a number of variables: overall response, lameness (AAEP scale), effusion score, flexion score and number of weeks to respond to treatment. Final data was available in 11 horses in the PG group and 9 horses in the PPS group. Mean scores and t-tests were used to compare overall response to treatment and lameness score between the two groups and the number of weeks to response. Mann-Whitney tests were also used to compare outcomes as a non-parametric alternative for non-normally distributed data.

There were statistically significant differences in the overall response and the response time in weeks (Table 1). 91 % of horses treated with PG were reported to have good or very good outcomes compared with 33% for PPS. Horses treated with PG took only 1.7 weeks to respond compared to 2.4 weeks for PPS (Figure 1). There was a tendency for a greater improvement in the flexion response with PG, with 66.6% horses showing an improvement compared to only 20% with PPS. Also there were a larger number of horses which showed reduced joint effusion for PG, 89% compared with 37.5% for PE, but this difference was not statistically significant (Table 1). There were a relatively small number of lame horses in each group, only 4 in PG group and 1 in PPS group, and all showed improvement with no difference between the groups.

Table 1. Results for overall response, response time (weeks), reduction in effusion score (number responded/number reported) and improvement in flexion score (number responded/number reported). Differing superscripts indicate significant differences (p<0.05).

	Mild	%	Good	%	Very good	%	Response time (wks)	Reduced effusion	%	Improved flexion	%
PG	1	9	5	46	5	46	1.7	8/9	89	6/9	67
PPS	6	66	3	33	0	0	2.4	3/8	38	1/5	20

Figure 1. The response time in weeks for horses to respond to the treatment



Overall horses treated with Pentosan Gold (PPS + NG) showed greater improvements in symptoms of joint disease than horses treated with PPS alone.

MACROCYCLIC LACTONE RESISTANCE IN AUSTRALIAN HORSES

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Small strongyles, or cyathostomins, are important to the horse industry both as agents of disease (such as colic and clinical larval cyathostominosis) and because of widespread anthelmintic resistance. Resistance to benzimidazoles within the cyathostomins is widespread, both overseas and in Australia. Consequently, the horse industry has become heavily reliant on macrocyclic lactone (ML) products, which are often used in an "interval" treatment regime (every 8-14 weeks). Not surprisingly, a few reports suggest that we are on the cusp of emerging resistance to these products too.

The ascarid worm, *Parascaris equorum*, is common in young horses and it too is associated with disease but more commonly impacts on growth of young animals. ML resistance in *P. equorum* has been observed overseas for the last decade, but the current extent of anthelmintic resistance in Australian horse worms is unknown due to a lack of surveillance. It is timely that a reliable survey be conducted to assess the issue.

A weak link in the fight against anthelmintic resistance is the lack of cost-effective and reliable methods for its early detection. We currently rely on the "gold standard" technique, the faecal egg count reduction test (FECRT), which is of limited value in equine parasitology because of the small numbers of horses on many properties (decreasing statistical power), mixed cyathostomin infections being the norm, and lack of validation of the technique with ascarid infections. There is a need for more sensitive, in vitro tools to be developed.

The overarching goal of this study is two-fold. Firstly, an extensive survey of ML-resistance is proposed for the southeast QLD, Hunter Valley and North QLD regions to document ML-efficacy and prevalence of resistance. Secondly, the project aims to develop and/or evaluate a number of *in vitro* assays to identify differences in susceptibility of equine worm populations to ML anthelmintics. Of primary importance is the development of an assay for use in determining ML-resistance within *P. equorum* isolates. Two assays which have been previously described as applicable to Trichostrongylid parasites of sheep; the larval development assay and the larval migration assay, will be evaluated for their ability to detect variation in ML-susceptibility between cyathostomin populations.

The benefits of surveillance and/or early detection of ML-resistance include the ability to appropriately adapt and improve worm control strategies on individual farms, and ultimately avoid disease and negative impacts on growth, health and performance caused by worms. In addition, the development and validation of in vitro techniques applicable to both the cyathostomins and *P. equorum* will have the broader impact of providing useful tools for veterinarians and researchers to monitor the efficacy of anthelmintic products. The outcomes of the project will have far-reaching positive impacts on the horse industry and will satisfy a void in information regarding current ML-efficacy in Australian horse worms.

USE OF FUNGAL SPORES (*Duddingtonia flagrans*) FOR BIOLOGICAL CONTROL OF INFECTIVE EQUINE INTESTINAL NEMATODE LARVAE

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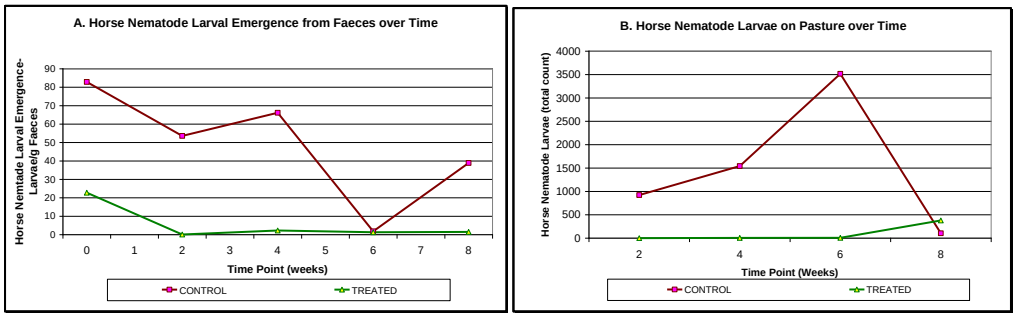
The natural fungus *Duddingtonia flagrans* belongs to a group of nematophagous fungi that physically entrap nematode larvae by means of adhesive hyphal nets before paralysing and consuming them. When the chlamydospores of *D. flagrans* are fed to grazing animals they pass through the digestive tract without germinating and are subsequently deposited in the faeces along with the eggs shed by adult intestinal parasites. The spores germinate within the faecal pats to produce mycelia with hyphal traps which act to substantially reduce the number of emergent parasite larvae able to migrate to the pasture and re-infect grazing animals (Waller and Faedo, 1996). It should be noted that *D. flagrans* is a common soil organism found throughout the world. Studies have shown that spores deposited in faecal pats remain localised and have no significant impact on non-target nematodes and other soil fauna (Knox et al., 2002).

Controlled Australian field trials with grazing animals (sheep, goats, cattle and horses) have shown that use of prototype products containing *D. flagrans* spores can substantially reduce the infectivity of pasture (e.g. Knox and Faedo, 2001). In recently conducted series of equine field studies, a native Australian strain isolated by CSIRO was used. In the studies, faecal pats from both *D. flagrans*-supplemented and control horses were monitored and it was found (Figure 1) that the use of spores reduced (A) the emergence of nematode larvae within the fecal pats and (B) the infectivity of the pasture surrounding the faeces by 70-100% . These cross-location studies have shown that *D.flagrans* is effective against resistant and non resistant nematode larvae when seasonal conditions favour egg hatch and larval transmission.

Figure 1 Example of the effect of supplementation with *D. flagrans* spores on (A) larval emergence from faeces and (B) infectivity of pasture



Importantly, the spores can be administered by mixing into any palatable feed, premix or supplement. Ideally, the horses would be wormed with an effective anthelmintic and moved onto rested pasture where they would be fed the spores during periods of high worm transmission.

Knox, MR and Faedo, M (2001) Biological control of field infections of nematode parasites of young sheep with *Duddingtonia flagrans* and effects of spore intake on efficacy. *Veterinary Parasitology;* 101: 155-160.

Knox, MR, Josh, PF and Anderson, LJ (2002) Deployment of *Duddingtonia flagrans* in an improved pasture system: dispersal, persistence and effects on free-living soil nematodes and microarthropods. *Biological Control* 24, 176-182.

Waller, PJ and Faedo, M (1996) The prospects for biological control of the free-living stages of nematode parasites of livestock. *International Journal for Parasitology* 26, 915-925.

The RIRDC Horse RD&E Program - Update

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RIRDC is a statutory authority established by the Australian Government. RIRDC manages investment in research, development and extension activities (RD&E) for the benefit of varied rural industries including horses (see www.rirdc.gov.au).

The RIRDC Horse Program is guided by a five year plan developed in consultation with industry stakeholders, and managed with input from a Horse Advisory Committee. Each year the RIRDC invests funds in a range of activities for the benefit of Australian horses. Activities include: research projects; publication of monographs written by experts on topics of interest to Australian horse owners; scholarship funding for young horse researchers; sponsorship of conferences; and a quarterly newsletter on horse related matters.

The RIRDC Horse Program has been supported by voluntary contributions from horse owners and organisations and, for a number of years, by matching government funds under the direction of the RIRDC Board. Recently the RIRDC Board made a strategic decision to progressively withdraw matching funds and from 1 July 2013 the program will be reliant only on industry contributions for support. This decision does not indicate a lack of RIRDC support for the Horse Program but rather reflects a strategic decision based on two big picture issues.

The first is that the most appropriate pathway for a mature industry that sees the value of RD&E to direct funds to RIRDC is through a statutory levy. Voluntary contributions as a mechanism for funding these activities are considered to be more appropriate for new and emerging activities or industries.

The second is that the matching dollars for those programs within RIRDC that are dependent on voluntary contributions, are drawn from core Government funding to RIRDC that is intended to fund a wide range of strategic activities. Strategic imperatives to invest in new and emerging industries as well as cross-cutting issues, such as health and safety on farms and climate change, mean that the RIRDC core funds is being directed to strategic areas other than mature industries such as the horse industry.

If Australian horse owners and organisations were to choose to support a levy mechanism for funding research, development and extension activities (the sorts of activities that the RIRDC Horse Program has been funding), then implementation of a levy would secure Australian Government matching specifically for the Horse Program and would provide a solid funding base for continuation of the program. Alternatives where a statutory levy is not implemented to support these investments, are likely to be associated with reduced funding and a decline in the value of these activities.

The Horse Advisory Committee is currently engaging with horse industry bodies to seek views on future funding of horse RD&E in Australia including options that do not involve a statutory levy and options that do involve a statutory levy.

LACTATION IS ASSOCIATED WITH INCREASED CIRCULATING OESTRADIOL LEVELS IN MARES AT OVULATION AND THROUGHOUT EARLY PREGNANCY

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In the mare, following the maternal recognition of pregnancy, the primary corpus luteum (CL) continues to secrete progesterone to maintain pregnancy. However, progesterone levels slowly decline until resurgence of the primary CL following implantation around day 35 of pregnancy. At this time increasing luteotrophic support from equine chorionic gonadotrophin (eCG) stimulates progesterone secretion from the primary CL and further causes luteinization of follicles to form secondary CLs. In comparison, oestradiol 17ß levels during early pregnancy are reported to be quite low up to Day 35, despite the primary CL being capable of oestradiol 17ß synthesis via aromatization of androgens. Nevertheless, following eCG secretion around Day 35 oestradiol levels increase in conjunction with progesterone.

Previously we have reported a serendipitous observation that during early pregnancy (Days 20-40) lactating, multiparous mares had plasma oestradiol 17ß concentrations that were approximately 4-fold higher than levels observed in maiden pregnant mares (Anderson *et al*. 2010). In contrast there was no difference in progesterone levels between multiparous, lactating versus maiden mares. Here we present preliminary data that confirms our original observation.

Four groups of mares were used for this study; multigravid and lactating (n=4); multigravid and non-lactating (n=4); primigravid and lactating (n=4) and maiden (n=4). Blood samples were obtained each morning from the jugular vein from ovulation (day 0) until Day 50 of pregnancy. Plasma was stored at -20°C until assayed for oestradiol 17ß using a commercial RIA.

Overall during early pregnancy, lactating (multigravid and primigravid) mares had significantly ($P<0.001$) higher plasma oestradiol 17ß concentrations than non-lactating (maiden and primigravid) mares. At the time of ovulation the difference in levels between the groups was most marked (271.9 ± 23.3 vs. 17.9 ± 3.4 pg/ml respectively). In non-lactating mares, oestradiol levels did not significantly change after ovulation until a small, but significant, increase on day 35 of pregnancy (21.0 ± 0.6 pg/ml). Thereafter oestradiol in non-lactating mares did not further increase (23.0 ± 2.9 pg/ml on Day 50). In contrast, oestradiol concentrations in lactating mares steadily declined from ovulation (271.9 ± 23.3 pg/ml) across early pregnancy to reach a minimum on Day 50 of pregnancy (89.9 ± 5.5 pg/ml).

The results suggest that lactation in the mare is associated with marked increases in the levels of free circulating oestradiol 17ß. This intriguing observation could be explained by lactation directly stimulating aromatase expression in the primary CL, increasing oestradiol secretion. However oestradiol concentrations in our study were highest in lactating mares at the time of ovulation when the CL is just being formed. This is in accord with an earlier study (Allen *et al*. 1995) that showed similar high oestradiol levels in lactating mares around the time of ovulation The mechanistic explanation of our intriguing observation and its biological significance will the subject of further research

Allen W.R. *et al*. (1995) *Equine Vet. J*. 27, 460-464
Anderson S.T. *et al.* (2010) *Proc. Australasian Equine Sc Symp.* 3: 27.

PASTURE MANAGEMENT ON COMMERCIAL THOROUGHBRED AND STANDARDBRED BREEDING FARMS IN NEW ZEALAND

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Production of horses is pasture-based in New Zealand, however, there is little information on pasture management practices on commercial stud-farms in New Zealand. A face-to-face survey was carried out with 26 stud-masters or stud-owners (22 Thoroughbred farms and 4 Standardbred farms, representing approximately 52% and 38% of the total number of registered Thoroughbred and Standardbred broodmares, respectively). Respondents were asked a total of 34 open, closed and multiple choice questions related to pasture management.

Most farms (12/26) had effective grazing areas of between 100 and 200 ha, with 5/26 <100 ha, and 9/26 >200 ha. Paddock sizes ranged from 1-3 ha on farms <100 ha, 2-5ha on farms 110-200 ha, and 2-8 ha on farms >200ha. Stocking density was greatest on farms <100 ha, with stocking rates of nearly 2 mares/ha in spring and summer and 1 mare/ha in winter, with larger farms reporting about ½ the stocking rates. Most respondents reported that horses were set stocked during the breeding season.

Cross-grazing of pasture was common with cattle (22/26 farms) and/or sheep (13/26 farms). Typically pastures were cross-grazed after horses has been moved from the paddock (24/26 farms). Harrowing was regularly carried out on 24/26 farms, typically in spring, summer and autumn.

Many farms (10/26) planned a pasture renewal programme of every 3-5 years, with others aiming for every 5-10 years (9/26). Most farms (19/26) sowed perennial (or annual) ryegrass in an 80:20 mixture with white clover. Most farms (15/26) chose to resow with low endophyte ryegrass over high endophyte ryegrass (6/26 farms).

All respondents reported that fertiliser was applied to paddocks, annually for 24/26 farms. Soil mineral analysis was carried out by 24/26 farms in the last 2 years. Advice was often sought from fertiliser representatives (23/26 farms) and seed company representatives (18/26), but nutritionists, farm consultants and agronomists were seldom used (2/26, 3/26 and 2/26 farms, respectively).

All farms provided supplementary feed for horses, especially for lactating mares and youngstock. Commercial feeds designed to meet the mineral requirements of mares in late gestation were offered on 17/26 farms. Grass haylage offered to mares during late gestation on 19/26 farms, to lactating mares on 7/26 farms, and to weanlings on 12/26 farms.

This survey provides new information on the pasture management practices of stud-farms in New Zealand. Management strategies were not optimised to maximise pasture dry matter production, or to reduce the numbers of infective parasitic larvae present on pasture.

GLUCOSE AND INSULIN DYNAMICS IN MARES AND THEIR FOALS

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In animals pregnancy is characterized by a progressive decline in insulin sensitivity, a natural adaptation that parallels growth of the feto-placental unit ensuring sufficient glucose supply to the fetus. Ample evidence shows an adverse intrauterine environment can have deleterious consequences for health later in life. In particular, perturbations in glucose and insulin metabolism during gestation in mares can result in changes in metabolism in the resulting neonatal foal potentially predisposing the foal to metabolic disorders associated with insulin resistance later in life. The current study investigates the relationship between glucose and insulin dynamics in pregnant mares and their subsequent foal.

Pregnant (*n=12*) mares were used in the study and offered either a high (HE: *n=6*) or low energy (LE: *n=6*) diet during the last trimester of pregnancy and throughout lactation. Insulin-modified frequently sampled intravenous glucose tolerance (FSIGT) tests were performed the pregnant mares in early gestation prior to nutritional manipulation, a few days prior to parturition, and in lactation (Day 120 post-foaling). After birth, FSIGT tests were conducted in foals at 2 weeks, 4 months and 10 months of age. Minimal model analysis was used to determine insulin sensitivity (Si), glucose effectiveness (Sg), acute insulin response to glucose (AIRg), and disposition index (DI).

In mares diet had a significant ($P<0.01$) effect in late pregnancy, with higher basal glucose (93.0 ± 2.7 vs. 85.0 ± 1.5 mg/dL) and insulin concentrations (5.0 ± 1.3 vs. 3.4 ± 0.9 mIU/L) being observed in HE fed mares compared to LE fed mares respectively. However no significant difference in any minimal model parameter was observed from the FSIGT test immediately prior to parturition, or later in lactation.

In foals, at 2 weeks of age basal insulin concentrations were significantly ($P<0.01$) lower in foals from HE fed mares than foals from LE fed mares (5.2 ± 0.8 vs. 13.4 ± 2.8 mIU/L, respectively), but no differences in glucose levels were observed. Further at this time, both Si (9.2 ± 1.2 vs. 3.9 ± 0.6 $x10^{-4}L.mU^{-1}.min^{-1}$) and Sg (6.6 ± 0.4 vs. 4.1 ± 0.6 $x10^{2}$/min) were significantly ($P<0.01$) higher in HE foals than LE foals. Prior to weaning, basal insulin was significantly ($P<0.05$) lower in HE foals than LE foals, but there were no significant differences in minimal model parameters at 4 months of age. Similarly, at 10 months of age, basal insulin concentrations (0.7 ± 0.1 vs. 2.0 ± 0.2 mIU/L) remained lower in HE foals, but again no differences in minimal model parameters were observed from an FSIGT test.

The results indicate that maternal diet during late gestation and lactation can influence insulin sensitivity and glucose dynamics in the foal during early post-natal life. The enhanced nutritional plane of high energy fed mares resulted in lower basal insulin concentrations in foals during the first year of life. Whether such dynamic changes predispose foals to metabolic problems later in life remains to be determined.

MODULATION OF STALLION FERTILITY WITH DIETARY OIL SUPPLEMENTATION

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Dietary supplementation of omega-3 fatty acids has been shown to improve sperm quality in various domestic species. Specific ratios of polyunsaturated fats within the sperm plasma membrane are vital for normal sperm function and viability. In stallions, where the spermatogenic cycle is approximately 57 days, there is some evidence of improvement in semen quality with optimized dietary supplementation of antioxidants and minerals after a 60 day feeding period.

The aim of this pilot study was to investigate the effect of an omega-3-enriched diet on the stallion sperm quality. Australian Stockhorse stallions (n=2) were fed a standard concentrate diet (control diet) prior to the commencement of this study. Stallions were then fed standard concentrate feed supplemented with: 1) corn oil (control oil supplementation) and 2) commercially available omega-3-enriched nutriceutical (experimental oil supplementation; EO3, Kerx, Brighton, USA) over a period of 75 days. Both stallions received both treatments as a cross-over study. Semen samples were collected every three weeks for assessment of semen volume, concentration and a variety of sperm function tests by flow cytometric analysis: viability, mitochondrial membrane potential, membrane fluidity, reactive oxygen species production, and ratio of X-/Y-bearing spermatozoa.

The addition of oil to the diet, regardless of type, significantly improved sperm viability in both stallions ($P < 0.05$). No significant effects of oil type were found on any of the other assessed parameters in this pilot study.

In conclusion, oil supplementation improves *in vitro* viability of semen in stallion, however, further extension of this study is required to assess full efficacy of omega-3-enrichment of stallion diet on spermatogenesis and semen quality of stallions.

HISTOPATHOLOGY OF THE FETO-PLACENTAL UNIT FOLLOWING EXPERIMENTAL INDUCTION OF EAFL

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A previously unrecognised form of equine abortion was reported in 2004 from the Hunter Valley region of New South Wales. The cases were related by specific histological and microbiological findings in the aborted foetuses. Equine Amnionitis and Foetal Loss (EAFL) was the term adopted to describe this syndrome (Todhunter et al., 2009). Systematic investigations were undertaken to determine a cause and it has been shown that experimental ingestion of whole Processionary caterpillar (*Ochrogaster lunifer*) or processionary caterpillar exoskeleton by pregnant mares induces foetal loss (Cawdell-Smith et al 2012). The objective of this study was to assess histologically fetuses and fetal membranes of pregnant mares experimentally exposed to processionary caterpillars to further explore the pathogenesis of EAFL.

Pregnant mares were experimentally exposed to whole caterpillar or exoskeleton of the Processionary caterpillar via gavage. Specimens were collected from resulting abortions and full term pregnancies consisting of 13 aborted fetuses, 3 fetuses from treated euthanized mares, membranes of 5 full term foals and organs from 3 full term foals. Four control membranes and one control fetus were examined. All three fetuses from the euthanized mares and eleven out of twelve aborted fetuses (92%) had setal fragments embedded in the allantochorion ranging from the villi of the chorion through to the allantois including within allantoic vessels. Placental locations of fragments ranged from the cervical pole region to the body encompassing the umbilical insertion and pregnant horn. Numbers in each fetus ranged from one fragment to a maximum of seven in various locations. Setae were present in the allantochorion from 2 to 22days after the initial treatment. A wide range of inflammation from subtle acute to chronic active was present in all aborted fetuses, all euthanized fetuses, and within at least one tissue level (chorion, allantois, umbilical cord, or amnion) of the membranes from full term foals. Amnionitis, funisitis, and allantoitis were present in 95% of the examined membranes. Pneumonia was present in 89% of the specimens and bacteria were present histologically in 90% of the specimens with or without accompanying inflammation.

The rapid migration of setae within 2 days after mare exposure suggests that direct setae migration into the fetal membranes is the likely initiating factor for EAFL.

Cawdell-Smith, A.J., Todhunter, K.H., Anderson, S.T., Perkins, N.R. and Bryden, W.L. (2012). Equine Amnionitis and Foetal Loss: Induction of the syndrome in mares following exposure to the Processionary caterpillar (*Ochragaster lunifer*).*Equine Vet. J.* 44: 282-288..

Todhunter, K.H., Perkins, N.R., Wylie, R.M., Chicken, C., Blishen, A.J., Racklyeft, D.J., Muscatello,G., Bryden, W.L. and Begg, A.P. (2009). Equine Amnionitis and Foetal Loss: the case definition for an unrecognized cause of abortion in mares. *Aust. Vet. J.* 87: 35-38.

PRESENCE OF CATERPILLAR SETAE IN THE PLACENTA FROM A FIELD CASE OF EAFL

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Equine amnionitis and fetal loss (EAFL) is the term applied to abortions found to be caused by the Processionary caterpillar (*Ochrogaster lunifer*). The case definition stipulates that all other causes of abortion must be excluded, atypical bacteria isolated from lung and/or stomach contents, along with the presence of typical lesions of inflammation within the amnion and umbilical cord (Todhunter et al., 2009; Cawdell-Smith et al., 2012) The pathogenesis of the condition is obscure but our recent experimental findings (Todhunter et al., 2010; 2012a) suggest that rapid migration of caterpillar setae (hairs) from the maternal gut to the uterus facilitates bacterial infection of the placenta and foetus.. Here we describe a case of EAFL with both typical and atypical lesions and for the first time the identification of caterpillar setae in the placenta from a field case.

A pregnant Thoroughbred mare aborted at 206 days gestation without signs of impending parturition and retaining the pregnant horn of the placenta. At post-mortem examination, the foetus showed signs of poor development for gestational age and congestion of the subcutaneous tissues. The chorionic surface of the placenta appeared thickened and hyperplastic around the umbilical insertion and body with two side by side full thickness holes approximately 15cm from the umbilical insertion. The holes were approximately 2-3cm in diameter, irregular in shape and of variable thickness, separated by a thin connective tissue septum. The surrounding allantois was thickened with multifocal to coalescing nodules in a 5 cm radius with perivascular oedema and haemorrhage extending a further 21 cm around the lesions. The cervical pole appeared pale with moderate oedema. The amnion was oedematous with vascular engorgement, and perivascular haemorrhage in an irregular 30cm radius from the umbilical insertion. Histologically, there was severe diffuse chronic active allantoitis, amnionitis, funisitis, and foetal pneumonia characterized by a mixed inflammatory infiltrate dominated by mononuclear cells. Allantoic epithelial hyperplasia was present in all areas of the allantois ranging from Stage I in the cervical pole region to Stage III in the area of the lesions. One small cross section of a caterpillar setal fragment approximately 8 μm in diameter was present embedded a villous within the cervical pole with minimal reaction. Special stains revealed Gram-positive bacilli within a focal area of squamous accumulation and necrosis on the surface of the umbilical cord and rarely within the lungs. An *Actinomyces* sp. was isolated from the lung and stomach contents identified on the basis of colony morphology and staining characteristics.

Chronic active placentitis as found in this case, often manifests as a focal lesion frequently associated with Nocardioform organisms. Chronic active changes within the placenta as seen here have recently been described with focal mucoid lesions in both experimental and field EAFL cases involving a variety of bacteria (Cawdell-Smith *et al.*, 2010; Todhunter *et al.*, 2012b) It is possible the holes themselves are sequelae to setae migration and chronic inflammation however; the diffuse inflammatory reactions are likely associated with their presence allowing communication between the chorionic surface and the allantoic cavity. Importantly, this is the first report of a caterpillar setal fragment being found in placental tissues in a field case of EAFL. It further supports a pivotal role for setal fragment migration in the pathogenesis of EAFL.

Cawdell-Smith, A.J., Todhunter, K.H., Perkins, N.R., Begg, A.P. and Bryden, W.L (2010) Nocardioform like focal mucoid placentitis as sequelae to caterpillar exposure in mares. *Proc. Australasian Equine Sc. Symp.* 3:15

Cawdell-Smith, A.J.; Todhunter, K.H.; Anderson, S.T.; Perkins, N.R.; Bryden, W.L: (2012) Equine amnionitis and fetal loss: Mare abortion following experimental exposure to Processionary caterpillars (*Ochrogaster lunifer*). *Equine vet. J*. 44: 282-288.

Todhunter, K.H., Perkins, N.R., Wylie, R.M., Chicken, C., Blishen, A.J., Racklyeft, D.J., Muscatello, G., Gilkerson, J.R., Bryden, W.L. and Begg, A.P. (2009). Equine Amnionitis and Foetal Loss: the case definition for an unrecognized cause of abortion in mares. *Aust Vet J*. 87::35-8.

Todhunter, K.H., Cawdell-Smith, A.J., Perkins, N.R., Begg, A.P. and Bryden, W.L (2010) Pathology of mares aborting during equine amnionitis and foetal loss. *Proc. Australasian Equine Sc. Symp.* 3:14

Todhunter KT, Cawdell-Smith AJ, Bryden WL, Perkins NR, Begg AP,(2012a). Histopathology of the feto-placental unit following experimental induction of EAFL *Proc. Australasian Equine Sc Symp.* 4:58.

Todhunter KT, Cawdell-Smith AJ, Perkins NR, Begg AP, Bryden WL, (2012b). Focal chronic active placentitis in the mare coexisting with lesions of EAFL-chronic sequelae to caterpillar exposure early in gestation? *Equine vet. J*. Submitted

THE GUINEA PIG AS A MODEL FOR EQUINE AMNIONITIS AND FOETAL LOSS

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A previously unrecognized form of abortion in mid- to late gestation mares was identified in Australia in 2004 and was characterized and termed Equine Amnionitis and Foetal Loss (EAFL) (Todhunter et al., 2009). Research has shown that the Processionary Caterpillar (PC; *Ochrogaster lunifer*) has a role in the aetiology of EAFL (Cawdell-Smith et al., 2012). The use of the pregnant mare to investigate the pathophysiology of this condition is expensive both in the acquisition of mares and their maintenance until the challenge experiments are undertaken. The objective of the present study was to establish a laboratory animal model to investigate the pathogenesis of EAFL.

The guinea pig was chosen as a model because it is a monogastric and a hind-gut fermenter, similar to the horse. Pregnant guinea pigs (n=8) were administered capsules containing exoskeleton, dissected from whole PC, for 5 days from Gestation Day (GD) 25. Pregnancies were monitored using transabdominal ultrasonography. The guinea pigs were autopsied either when foetal death was detected by ultrasonography (n=2; GD 31, 40), or at a set time point from the first day of treatment (n=6; GD 26, 40, 45, 50, 55, 60). In addition, control guinea pigs not administered PC exoskeleton were euthanased (n=4; GD 29, 40, 50, 60). All guinea pigs were anaesthetized, heart blood collected and then euthanized. The fetuses were removed and samples for microbiology and histopathology collected from the peritoneum, uterus, chorionic plate, amnionic, foetal stomach fluid and foetal lungs.

Samples for microbiology were cultured on Horse Blood, McConkey's and Chocolate agar at 37°C both aerobically and anaerobically. In addition, samples were placed in brain-heart infusion broth and further sub-cultured onto plates as described above. Bacteria were isolated from all treated guinea pigs expect one (GD 26) which was euthanased 24 hours after receiving a single dose of PC exoskeleton. The bacteria isolated were environmental or enteric bacteria similar to those isolated from mares experimentally exposed to PC exoskeleton or cases of EAFL. These findings support the use of a pregnant guinea pig model in further investigations of EAFL.

Cawdell-Smith, A.J., Todhunter, K.H., Anderson, S.T., Perkins, N.R. and Bryden, W.L. (2012). Equine Amnionitis and Foetal Loss: Induction of the syndrome in mares following exposure to the Processionary caterpillar (*Ochragaster lunifer*). *Equine Vet. J.* 44: 282-288

Todhunter, K.H., Perkins, N.R., Wylie, R.M., Chicken, C., Blishen, A.J., Racklyeft, D.J., Muscatello,G., Bryden, W.L. and Begg, A.P. (2009). Equine Amnionitis and Foetal Loss: the case definition for an unrecognized cause of abortion in mares. *Aust. Vet. J.* 87: 35-38.

EAFL RISK AND CATERPILLAR ECOLOGY

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Processionary caterpillars can cause mares to abort in a condition called Equine Amnionitis and Foetal Loss (EAFL) (Cawdell-Smith *et al*., 2012). The condition was first described in 2004 (Todhunter *et al*, 2009) and has continued to be a significant cause of abortion in eastern Australia. This abstract summarises recent attempts to better understand caterpillar ecology in order to reduce exposure risk for broodmares on Australian pastures.

Processionary caterpillar moths emerge in late spring (October-November). They are short-lived and mate, lay eggs and then die. There appear to be different types of nest structures including bag-type nests that are dew-drop shaped and appear high in eucalypt trees; bag-type nests that are flattened and adherent to tree trunks at varying heights off the ground, and; ground-based nests that seem more common in wattles (acacia species). It is unclear if these differences reflect the presence of related caterpillar species/subspecies or the use of different habitats. Clearly, more information is required on the biology and ecology of the Processionary caterpillar (*Ochrogaster lunifer*) and related species.

Inspections were made of properties in Queensland and New South Wales in 2011-2012 in an attempt to determine whether it might be feasible to identify and map caterpillar nest locations and then develop strategies for minimising mare exposure to caterpillar material.In spring and early summer of 2011, nests were identified mainly as small, fluffy, white, circular structures (2 to 5 cm in diameter) that were mainly adherent to the base of wattles. Occasionally, nests were found in other tree types and in elevated locations. Over time (through the summer), nests developed into larger accumulations of silk-like material and organic matter (leaves, sticks etc) and caterpillar frass (faeces) and discarded exoskeleton. Some nests remained at the same location while others were abandoned with caterpillars either failing to survive or moving to a new nest location (ground or elevated nest) in different trees. By late autumn, large nests were identified that were empty of caterpillars, consistent with mature caterpillars moving away from nests to pupation sites.

Caterpillar nests were considered to represent potential exposure risk to pregnant mares if they were located either within the same paddock or within a distance of up to 200-400 metres of the boundary of a paddock containing pregnant mares. Exposure risk was considered likely to increase as caterpillars reached later stages of development in association with increased amounts of shed exoskeleton, and increased likelihood of caterpillars moving from one location to another, either to reach a new food source or to begin pupation. Exposure risk was considered likely to remain low for the first few months of development – up to about February to March. Options for reducing exposure risk in these areas include detection and removal of caterpillar nests between November and March, before the likely period of elevated exposure risk. Identification of high density nest material in one area could result in a decision to move mares to a different location. In the longer term monitoring of which trees might appear more or less likely to attract nests may inform decisions about management of tree species and planting of new species in areas close to paddocks likely to house pregnant mares.

Processionary caterpillars are urticarial and all caterpillar nest material is capable of causing severe itching or allergic reactions in people on contact with skin, and may cause severe irritation if material gets into an eye or into the nose or mouth (Battisti et al., 2011). Nest material is very light and can easily be dispersed on the wind once the nest starts to breakdown. Protocols need to be developed for safe handling of nest material & management

of inadvertent human exposure if individuals do develop clinical signs such as skin reactions, respiratory conditions or eye conditions.

Battisti, A., Holm, G., Fagrell, B. and Larsson, S. (2011) Urticating hairs in arthropods: Their nature and medical significance. *Annual Review of Entomology* **56**, 203-220.

Cawdell-Smith, A.J., Todhunter, K.H., Anderson, S.T., Perkins, N.R. and Bryden, W.L. (2012) Equine amnionitis and fetal loss: Mare abortion following experimental exposure to Processionary caterpillars (*Ochrogaster lunifer*). *Equine Veterinary Journal*, 44; 282-288.

Todhunter, K.H., Perkins, N.R., Wylie, R.M., Chicken, C., Blishen, A.J., Racklyeft, D.J., Muscatello,G., Bryden, W.L. and Begg, A.P. (2009). Equine Amnionitis and Foetal Loss: the case definition for an unrecognized cause of abortion in mares. *Australian. Veterinary. Journal.* 87: 35-38.

HORMONAL PHENOTYPES IN THOROUGHBRED YEARLINGS WITH OSTEOCHONDROSIS

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Osteochondrosis (OC) is an important developmental orthopedic disease in horses. The primary lesion is a defect in endochondral ossification, within the growing cartilage. The pathogenesis of OC is complex, but generally thought to involve abnormal chondrocyte maturation or failure of differentiation. The resultant changes in extracellular matrix composition (dyschondroplasia) can lead to subchondral fractures and cysts, cartilage flaps, presence of loose cartilage fragment (osteochondrosis dissecans), and synovitis. Much research has examined possible causative factors, including biomechanical influences, vascularization, nutritional factors (mineral imbalances and excess energy), and genetic predisposition, but the relative contribution of these appears to vary. However a key role for the endocrine system in metabolic signaling to bone appears important in the pathogenesis of OC. During the past decade a major hypothesis has been proposed; that high energy diets fed to rapidly growing animals, induces post-feeding hyperinsulinaemia and this is associated with OC. However OC lesions are observed in many horses fed low to moderate energy diets. Therefore in this study we sought to define the association of insulin and other metabolic hormones with OC.

Thoroughbred yearlings (*n=191*) born in '2007' and '2008' were recruited from four NSW stud farms and fasting blood samples were obtained following stabling overnight. Plasma concentrations of metabolic hormones (insulin, IGF1, thyroxine, leptin and adiponectin) were determined by standard immunoradiometric and radioimmunoassays Skeletal abnormalities were determined by radiography analysed by experienced veterinarians, and the data were retrospectively classified as either osteochondrosis related lesions (OC), other bone abnormalities, or no abnormality (NA).

Hormone analysis revealed significantly ($P<0.05$) lower fasting insulin concentrations in yearlings with OC compared to those with NA (2.3 ± 0.2 vs. 3.4 ± 0.2 mIU/L, respectively). Also significantly ($P<0.05$) higher IGF1 levels were found in yearlings with OC than NA (264.1 ± 40.9 vs 192.1 ± 13.1 ng/ml, respectively). In contrast there was no significant difference in total thyroxine (1.8 ± 0.2 vs. 1.8 ± 0.1 µg/dL), leptin (2.0 ± 0.2 vs. 2.0 ± 0.1 ng/ml) or adiponectin (2.5 ± 0.1 vs. 2.4 ± 0.1 ng/ml) concentrations between OC and NA groups respectively. Correlation analysis across all animals revealed significant moderate correlations between insulin and glucose ($r = +0.30$), adiponectin and glucose ($r = +0.24$) and IGF1 and total T4 ($r = +0.21$). However in OC animals, significant relationships between insulin and total T4 ($r = -0.32$), and IGF1 and total T4 ($r = +0.37$) were only found.

Overall our results suggest that low insulin and high IGF1 concentrations in fasted yearlings are associated with OC. Whether these hormonal alterations are involved in the pathogenesis of OC remains to be determined. Our observation of higher IGF1 levels in yearlings with OC may be indicative of joint repair occurring in these animals with lesions, rather than being related to the development of OC. In regards to insulin, we also report these yearlings have lower resting insulin concentrations throughout early postnatal life (Dobbs *et al.* 2012), which provides more evidence that insulin is involved in the pathogenesis of OC.

Dobbs, T.N. *et al.* (2012) *Proc. Australasian Equine Sc Symp.* 4:26.

ENDOCRINE RESPONSES IN PROCESSIONARY CATERPILLAR-INDUCED EAFL

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Equine Amnionitis and Foetal Loss (EAFL) was described in Australia in 2004. It is similar to Mare Reproductive Loss Syndrome (MLRS) reported in Kentucky in 2001 and 2002, where the Eastern Tent Caterpillar was identified as the cause of MRLS (Sebastion *et al*. 2008). This report details the endocrine responses in experiments (Cawdell-Smith *et al.*, 2012 that demonstrated the pivotal role of Processionary caterpillars (PC) in the aetiology of EAFL.

Briefly, two experiments were conducted to examine whether PC could induce EAFL in mid-gestation (Experiment 1. 100g whole PC to mares day 194 to 205 of pregnancy; Experiment 2. either 0, 1, 2, or 5g of shed PC exoskeleton to mares days 197 to 237 of pregnancy). Another experiment was conducted earlier in gestation (Experiment 3. 5g exoskeleton to mares days 45-60 of pregnancy). In each experiment mares were given caterpillar treatment daily for 5 days with Day 1 being designated the day of first treatment. Untreated mares of gestational age matching those that were treated were also monitored as controls. Blood samples were collected daily and plasma progesterone, oestradiol 17beta and oestrone sulphate concentrations determined by RIA.

Three mares treated with whole PC (Experiment 1) aborted on Days 5, 10 and 12. The other treated mare was euthanased on Day 10 due to ongoing colic. The foetus from this mare was still alive, but showed gross pathology similar to the other 3 aborted foetuses. In experiment 2 treatment groups receiving 1, 2 and 5g of caterpillar exoskeleton had 1, 1 and 2 abortions respectively. With foetal death being detected on Days 4 and 7 (5g group), Day 7 (2g group) and one mare aborted on Day 67 (1g group). All other mares delivered foals at term. In mares that aborted in both Experiments, plasma progesterone and oestrone sulphate concentrations significantly ($P<0.05$) declined around the time of foetal death. In treated mares that did not abort or control mares there was no decline in steroid hormone levels.

Treatment of mares with PC exoskeleton in the early-placentation stage of pregnancy (Days 45-60) had no significant effect on mean progesterone levels. However, foetal death did occur in 2 out of the 6 treated mares on Days 6 and 7. Progesterone concentrations in these mares declined markedly 24h after foetal death (14.3 to 6.7 ng/ml in one mare, and 5.9 to 3.3 ng/ml in the other mare). However the cervix in these mares remained closed for a further 15 and 8 days respectively, until progesterone concentrations reached 2.0 and 0.9 ng/ml respectively.

Endocrine results indicate that PC have a direct effect on the feto-placental unit with steroid levels (progesterone and oestrogens) declining as a consequence of this action, often within 24h. The results suggest no disruption to the endocrine support for pregnancy prior to foetal death.

Cawdell-Smith A.J. *et al*. (2011) *Equine Vet J*. 44: 282-288.
Sebastion M.M. *et al*. (2008) *Vet Pathol* 45: 710-722.

INSULINAEMIC RESPONSES TO ORAL GLUCOSE IN STANDARDBREDS, PONIES AND ANDALUSIAN-CROSS HORSES

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There appear to be breed-related differences in the prevalence of obesity, insulin resistance and laminitis in horses and ponies (Frank, 2011). Insulin sensitivity (SI) may be influenced by adiposity and dietary non-structural carbohydrate (NSC) content, whilst genetics and epigenetics may contribute to the innate SI of individual horses and ponies. The aim of this study was to compare the insulinaemic response of different breeds of horses and ponies to a meal containing added glucose, whilst animals were in moderate body condition.

Six Standardbred horses (BCS 4.5 ± 0.2 out of 9), six mixed-breed ponies (BCS 4.7 ± 0.2) and five Andalusian-cross horses (BCS 5.1 ± 0.3) were maintained on *ad libitum* pasture hay prior to undergoing a meal glucose tolerance test (MGTT). All horses and ponies consumed a meal containing soyahull pellets (1 g/kg BW; soaked), oaten chaff (1 g/kg BW) and dextrose (1.5 g/kg BW). Serial blood samples were collected for 5 hours after the meal was offered. Blood glucose was determined using a hexokinase enzymatic assay (Glucose Assay Kit, Cayman) and plasma insulin was measured using a radioimmunoassay (Coat-A-Count Insulin RIA, Siemens). Peak glucose and insulin concentrations plus area under the curve (AUC) were compared between breeds using a one-way ANOVA with Bonferroni *post hoc* test.

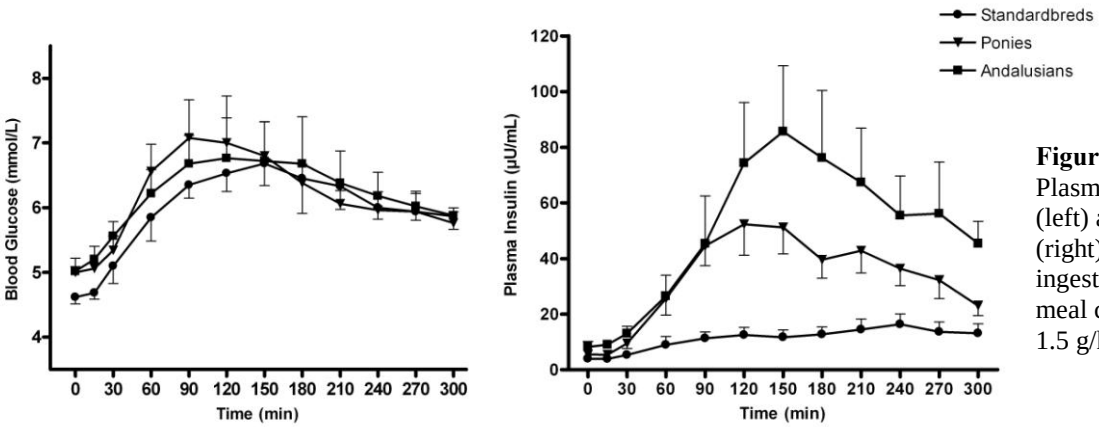

Figure 1: Plasma glucose (left) and insulin (right) following ingestion of a meal containing 1.5 g/kg glucose

The peak insulin concentration was significantly lower in Standardbreds (18.6 ± 3.0 µIU/ml) compared with ponies (65.5 ± 8.1; P=0.0003) and Andalusians (89.2 ± 22.4; P=0.007). The insulin AUC was also significantly smaller in Standardbreds compared with ponies (P=0.005) and Andalusians (P=0.009). There were no differences in the peak glucose concentration or glucose AUC between groups (Fig 1). The time taken to consume the meal was similar between groups.

There are clear differences in the insulinaemic response of Standardbreds, ponies and Andalusian-cross horses to a meal containing 1.5g/kg BW glucose. The exaggerated insulinaemic response of ponies and Andalusian-cross horses to the MGTT may reflect lower SI, even at moderate BCS. These results may help to explain the apparent increase in susceptibility to laminitis in these breeds, especially when they become obese or are allowed to consume a diet high in NSC.

Frank, N. (2011) Equine Metabolic Syndrome. *Vet Clin North Am Equine Prac* 27(1):73-92.

REPRODUCTIVE EFFICIENCY IN A COHORT OF THOROUGHBRED BROODMARES IN NEW ZEALAND

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Reproductive summaries of active Thoroughbred stallions in the 2005/06 breeding season in New Zealand were extracted. Sires were grouped into four categories based on the service fee or if the stallion was listed as a stallion shuttling between hemispheres for the breeding season: <$5,000, $5,001-10,000, >$10,000 and shuttle. From each sire category a sample of 6 sires were randomly selected. For mares bred by each of the selected stallions in 2005/06 reproductive records were extracted for the 2001/02–2010/11 breeding seasons and mare status was assigned as maiden, not served, live foal, missed, slipped or mare/foal dead for each breeding season.

In 2005/06 the selected 24 stallions covered a total of 1637 mares; representing nearly 30% of all mares covered in that season. Most of the selected mares were aged between 7-9 years old (455/1,637; 27.7%) and were served by a stallion in the category of >$10,000. The status of most of these mares was live foal (928/1337, 56.6%), with 14.8% missed (243/1637), 12.8% maidens (211/1637), 3.8% had dead foals (62/1637), and 2.1% slipped (35/1637).

In the following 2006/07 season 73.6% mares had a status of live foal (1206/1637), 15.8% missed (259/1637), 7.2% had dead foals (119/1637), and 3.2% slipped (53/1637). Overall, between 2001/02 and 2005/06 33.4% (218/1,637) of mares had a miss on at least one occasion, 13.3% (548/1,637) had a dead foal, and 7.3% (120/1,637) of mares slipped.

Multivariable logistic regression for the outcome of miss in 2006/07 showed that mares older than 10 years were more likely to miss than younger mares; the status of missed or not served in 2005 reduced the risk of missing in 2006/07. No variables were associated with the outcome of slip in 2006/07 at the multivariable level except previously having dead foal.

Survival analysis revealed that ½ of these of mares had officially retired by 11 years of age. The median age of our selected mares at the start and end of the breeding career was 5 (IQR 4-6) and 14 (IQR 11-17), respectively.

In summary, most reproductive loss was associated with missing and around 1/3 of all mares missed at least once, with older mares were more likely to miss than younger mares. The average reproductive career of mares is quite short, with ½ of all mares retired by 11 years of age. Stochastic modelling by Bosh et al (2009) indicated that mares that miss twice over a 7 year investment period were not profitable, and further work is needed to evaluate this under New Zealand conditions. However, this active removal of aging mares from the breeding population is likely to enhance overall reproductive efficiency.

THE EFFECTS OF THREE OMEPRAZOLE PASTES OMOGUARD, GASTROSHIELD AND GASTROZOL ON GASTRIC pH IN FASTED HORSES ARE COMPARED.

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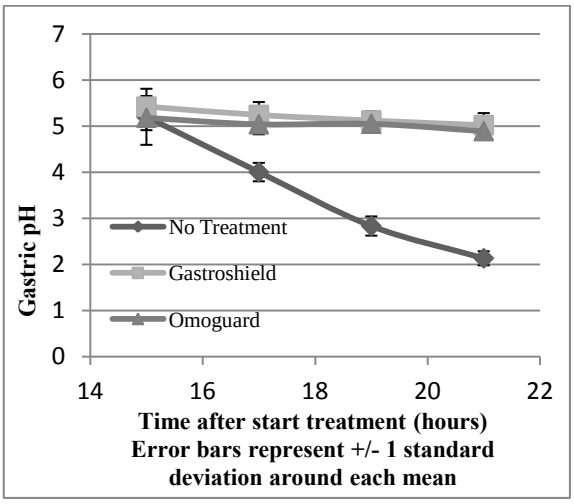
The efficacy of a generic omeprazole paste, Omoguard®, was compared with that of Gastroshield® paste. Gastroshield paste is the only omeprazole paste clinically proven to have consistent therapeutic efficacy in the treatment and prevention of gastric ulcer disease in horses. A formal bioequivalence study to compare the efficacy of Omoguard and Gastroshield in inhibiting gastric acidity in horses was conducted. An additional small study in three horses was performed to measure the efficacy of Gastrozol® in this model. The two pastes, Omoguard and Gastroshield were administered in a randomised cross-over design. Five horses were administered each product for 5 days, samples of gastric fluid were then collected and analysed for pH. Following a ten day wash out the horses were treated with the same protocol for the other product. All the pH results for gastric fluid samples were within the range 4.5 – 5.9 and statistical analysis of the data confirmed that the two products were equally effective (Figure 1).

Figure 1: Gastric pH following withholding feed after no treatment in 3 horses and Gastroshield and Omoguard administration (4 mg/kg) in 10 horses.



In this study Gastrozol at 1mg/kg did not result in an equivalent increase in gastric pH to Omoguard at 4mg/kg. When Gastrozol was administered at 4 mg/kg, it was equally effective as Omoguard at 4mg/kg in increasing gastric pH in horses (Table 1).

Table 1. Gastric pH results following administration of Gastrozol at 1mg/kg and 4mg/kg and Omoguard at 4mg/kg. Differing superscripts within columns indicate significant differences.

Time point	Gastrozol 1mg/kg	Gastrozol 4mg/kg	Omoguard 4mg/kg
15	2.6 ± 0.15^{a}	4.7 ± 0.15^{b}	5.0 ± 0.15^{b}
17	2.7 ± 0.05^{a}	4.5 ± 0.2^{b}	5.2 ± 0.15^{b}
19	2.9 ± 0^{a}	4.7 ± 0^{b}	5.1 ± 0.25^{b}
21	2.7 ± 0.25^{a}	4 ± 0.1^{b}	5.1 ± 0.1^{c}

In cases of equine gastric ulcer syndrome where treatment at 1mg/kg is ineffective it may be important to increase the dose to 4mg/kg.

IN VIVO APPARENT FEED DIGESTIBILITY MEASUREMENTS IN PONIES, STANDARDBRED AND ANDALUSIAN CROSS HORSES

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The prevalence of obesity in the equine population is becoming quite considerable, and obesity has been associated with insulin resistance and the increased risk of laminitis. Ponies and some breeds of horses, such as Andalusians, tend to put on weight easily (developing the 'cresty neck' appearance) and may have an increased incidence of laminitis, compared with breeds such as Standardbreds and Thoroughbreds (Frank, 2011). They may also be less insulin sensitive than the latter two breeds (Bamford et al, 2012). The aim of this study was to compare the rate of increase in adiposity between ponies, Standardbreds and Andalusian cross horses, and to determine whether any differences may be due to differences in feed digestibility.

Eighteen horses and ponies were assigned to either a weight gain group (increasing adiposity; 4 Standardbreds, 4 Andalusians and 4 ponies) or control group (2 Standardbreds, 2 Andalusians and 2 ponies). Body fat percentage was determined by the deuterium dilution technique (Dugdale et al. 2011) at the beginning and end of the weight gain phase. Over 22 weeks (in a dry lot environment) the horses and ponies in the weight gain group were fed a diet based on ad lib hay plus soyabean hull pellets, chaff and granulated vegetable fat and canola oil, to provide up to ~210% of estimated maintenance energy requirements. At the end of the weight gain phase, horses were kept in individual dry lot pens for a 24 hour period during which time they were fed a standardised meal (based on their BW) and their hay intake was recorded. Manure was collected, weighed and subsampled, and the feed and manure was analysed for energy, dry matter, protein, NDF, starch and fat. Percentage digestibility for each of these parameters was calculated and compared between the groups.

In the weight gain groups, percentage body fat increased from 9.6 ±1.4 to 18.3 ±2.2 % in ponies, from 6.7 ±3.6 to 14.7 ±1.7 % in the Andalusian cross group, and from 7.4 ±2.2 to 12.2 ±1.4 % in the Standardbred group. Body condition score increased less in the Standardbred group (from mean 4.5 ±0.3 to 7.1 ±0.1 out of 9) compared with the other groups (from mean 4.6 ±0.2 to 7.6 ±0.3 in ponies and mean 4.8 ±0.2 – 7.6 ±0.2 in the Andalusian cross group). There were no significant differences in apparent feed digestibility between any of the three breed groups. Fat digestibility was significantly greater in those weight gain animals adapted to significant amounts of fat in the diet compared with control animals (66.8 ±3.0 % digestibility vs 19.7 ±5.6 % for control group; $P<0.0001$).

These data show no differences in the apparent feed digestibility between ponies and different breeds of horses when provided with the same diet, environment and adaptation period. The increase in body condition was greater in the pony and Andalusian cross horses than in the Standardbreds; however this is likely to be due to metabolic differences between these breeds, rather than differences in feed digestibility. Differences in the amount of unstructured exercise within the dry lot may be another factor that might differ between breeds and this influence cannot yet be excluded.

Bamford N, Potter SJ, Harris PA & Bailey SR. (2012). Insulinaemic responses to oral glucose in Standardbreds, ponies and Andalusian cross horses. *Proc Aus Equine Science Symp*.

Frank N. (2011) Equine Metabolic Syndrome. *Vet Clin North Am Equine Prac* 27(1):73-92.

BROWSING – AN OVERLOOKED ASPECT OF FEEDING MANAGEMENT IN HORSES?

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Domesticated horses are routinely maintained in conditions with restricted access to forages, which contrasts with free-roaming horses that spend up to 17 h/day grazing and browsing. Horses evolved primarily as grazing herbivores typically consuming an estimated graze-to-browse ratio of 9:1. However, in some situations horses have been observed to consume a diet of up to 50% of browse (trees, shrubs and forbs), depending on the feed availability (*1*). We postulate that limiting foraging behaviour may have negative impacts on digestive health and animal welfare of confined horses.

Horses were introduced to Australia with the first fleet in 1788. Since that time a wild horse population (brumbies) has established and their diet has been reported to consist of a substantial portion of forbs, shrubs and tree foliage (*2*). Additionally, brumbies and pastured horses have been observed to chew bark of select eucalypt species. Some reasons for this have been preposed such as for medicinal purposes or for dietary supplementation, however, the reason why horses chew bark remains unanswered, as investigations into the digestibility and macronutrient content are not conclusive.

Browsing may be an important but generally overlooked aspect of feeding management in horses. Some of the potential benefits of browsing include providing nutrients, reducing stereotypic behaviour, buffering acids in the gastro intestinal tract or chelation of dietary trace minerals to excrete excess amounts. In our studies, we will look at the influence of browsing on time budgets of domesticated horses as well as the nutritive values of alternative fodder sources. Browsing enables horses to increase the time they spend on natural foraging which may reduce boredom that can lead to development of stereotypies. There is currently limited information about the nutrient value and palatability of edible tree and shrub foliages (fodder) for horses. Fodder trees and shrubs can provide an alternative forage source during times of drought and may have beneficial compounds that are not presently well understood.

As a first step to address the abovementioned project objectives, a pilot study will evaluate preference testing methodology in horses for use in later experimental work. Concurrently, a horse industry survey will be conducted to collect information from horse owners about observed incidences of browsing and types of foliages browsed.

The overall aim of this project is to investigate the role of browsing on behaviour and nutrition in domesticated horses.

Putman, R. J., Pratt, R. M., Ekins, J. R., and Edwards, P. J. (1987) Food and Feeding-Behavior of Cattle and Ponies in the New Forest, Hampshire, *Journal of Applied Ecology 24*, 369-380.

Hampson, B. A., Owens, E., Watts, K. A., Mills, P. C., Pollitt, C. C., and de Laat, M. A. (2011) Nutritional analysis of gastric contents and body condition score at a single time point in feral horses in Australia, *American Journal of Veterinary Research 72*, 1226-1233.