The 2014 Bayer lectures focused on ‘Faster, fitter horses’ and featured three speakers who are currently conducting equine research in New Zealand.

All three speakers have received assistance with their research from the NZ Equine Research Foundation (NZERF), and it was quite special to have them share the knowledge learned from their research with us in this way.

Dr Lee Morris, BVSc DVSc DipACT, is a director of EquiBreed NZ Ltd and specialises in the veterinary treatment of equine infertility, frozen semen technologies and stem cell therapies. Her talk focused on how the use of stem cells facilitates healing in the horse. They have provided new alternatives for treating lameness, wounds, eye injuries, post breeding endometritis and musculoskeletal injuries. Her enthusiasm was obvious, as she showed how stem cell therapy has the potential to reduce wastage in the equine industry, improve the welfare of horses and reduce long term costs associated with rehabilitation.

Dr Ray Cursons, PhD, is a graduate of Massey University and currently a senior lecturer in molecular genetics at the Science Faculty of the University of Waikato. His topic was “Equine Strangles, the disease that will not go away”. Strangles is caused by the bacterium Streptococcus equi subsp. equi. Alarming, it is currently the most frequently diagnosed infectious disease of horses world wide. He outlined how

Dr Michelle Logan, MA VetMB MACVSc Cert AVP, graduated from the University of Cambridge, UK, and has practiced as a veterinarian in both the North and South Islands of NZ. Her topic “Is your horse at risk from Equine Metabolic Syndrome (EMS)” is currently being widely discussed and researched. EMS is a disorder that results in an abnormal response by the body to the energy gained from food and it is this abnormal response that can lead to clinical problems including laminitis. Michelle’s talk covered which horses are at risk of having EMS and how it is diagnosed, treated and managed. Horses with high levels of insulin in the blood are most at risk of developing EMS.

The 2014 Bayer lectures were again held at four venues; Karaka, Matamata, Palmerston North and Christchurch. The NZERF thank Bayer for their ongoing support, the lecturers for their professional presentations and New Zealand Bloodstock for providing their facilities at Karaka. All those lucky enough to have been able to attend would have come away with a greater understanding of each topic and will look forward to hearing the results from ongoing research studies. Copies of the proceedings are available from the NZERF office and will be on the website, www.nzrf.co.nz.
Factors affecting the reproductive performance of Thoroughbred mares in New Zealand

Dave Hanlon, Matamata Veterinary Services

There have been no studies performed to investigate factors that influence the reproductive performance of Thoroughbred mares on NZ stud farms. Most of the advice regarding management and veterinary procedures that are used in this country are extrapolated from studies performed in other countries, ie. under different environmental, nutritional and managerial systems.

As part of a large epidemiological study we aimed to address these deficiencies in the knowledge of New Zealand equine reproductive performance. A study was performed involving five stud farms in the Waikato region of New Zealand during three consecutive breeding seasons (2006-2008). A total of 1482 individual mares contributed 2007 “mare years” and 3402 oestrous cycles over the three seasons. Mares were served by 87 individual stallions.

The aims of the study were:

1. To collect and describe basic measures of reproductive performance to enable comparisons to previous studies from other countries, utilizing similar methodology.
2. To perform appropriate statistical analyses not previously used with equine data to determine those factors that significantly affect reproductive performance on NZ stud farms, and
3. To determine the relative contribution of the mare, stallion and stud farm towards reproductive performance.

In this, the second of a 3-part series of articles on this study, we will outline the findings in terms of the mare-related variables that were found to significantly influence reproductive performance.

The main finding from the study was that the age of the mare is one of the most important factors influencing reproductive performance. Mares that were older than 14 years performed significantly worse in all measured outcomes compared with mares 14 years of age or less (Fig 1). In addition, we found that the performance of maiden mares was no better than the performance of “barren” mares, but that both of these categories (grouped together as “dry mares”) had better reproductive performance than foaling mares (Fig 2).

Further analyses were performed on dry mares and foaling mares separately because some of the variables influencing their reproductive performance are mutually exclusive, for example, only foaling mares can be served on foal heat and only dry mares can be maidens.
of mares that had not been under lights were served in September. By the end of the season, significantly more of the light exposed mares were in foal compared with those mares that had not been under lights (96.2% vs 88%).

As already mentioned, there was no effect of dry mare status (maiden or barren) on reproductive performance. In addition there was no effect of barren mare status (missed, slipped or not served) on reproductive performance.

<table>
<thead>
<tr>
<th></th>
<th>Lights¹</th>
<th>No lights²</th>
</tr>
</thead>
<tbody>
<tr>
<td>Mares served in Sept. (%)</td>
<td>98.6⁰</td>
<td>39.5⁰</td>
</tr>
<tr>
<td>Mean days to 1st service</td>
<td>8.8⁰</td>
<td>38.5⁰</td>
</tr>
<tr>
<td>Median days to conception</td>
<td>19⁰</td>
<td>51⁰</td>
</tr>
<tr>
<td>PR to 1st service (%)</td>
<td>69.1⁰</td>
<td>54.2⁰</td>
</tr>
<tr>
<td>Twins at 15 days (%)</td>
<td>14.6⁰</td>
<td>9.6⁰</td>
</tr>
<tr>
<td>Seasonal pregnancy rate (%)</td>
<td>96.2⁰</td>
<td>88.0⁰</td>
</tr>
</tbody>
</table>

Table 1: Reproductive performance of dry Thoroughbred mares either exposed to an artificial lighting programme prior to the start of the breeding season (Lights) or left exposed to natural lighting conditions (No lights). Values within rows with different superscripts are significantly different (P<0.05).

**Foaling mare reproductive performance**

Apart from the mare’s age, the main factors affecting foaling mare performance were whether a mare was served on foal heat and the foaling date of the mare.

In this study, mares were only served on foal heat if they met certain selection criteria: ovulation occurring 10 or more days post-foaling, less than 1cm of intra-uterine fluid present on ultrasonography and no history of retained foetal membranes or difficulty foaling. Using these criteria, 32% of foaling mares were served on foal heat. The reproductive performance of mares served on foal heat was superior to those mares that were not served on foal heat.

Significantly more mares were served earlier in the season and overall more of them had conceived by the end of the season (Fig 3).

Foaling date also had a significant effect on the reproductive performance of foaling mares. Mares that foaled prior to 16th October were 3 times more likely to be in foal by the end of the season compared with mares foaling after 16th October. Interestingly, 35% of mares foaled after 16th October.

**Conclusions**

In this population of Thoroughbred mares, the main factors affecting reproductive performance are the age of the mare and her reproductive status (dry or foaling). Dry mares that are exposed to artificial lights have superior reproductive performance compared to those under natural lighting conditions. Foaling mares that are selected carefully and served on foal heat have better reproductive outcomes compared to those not served on foal heat. The earlier a mare foals in the season, the better her chances of producing another foal in the subsequent breeding season.

In the third and final article of this series we will detail the major factors that affected stallion fertility in this study and how much of the overall reproductive performance on a stud farm is due to the mare, the stallion, and the stud farm.

This study was supported by the New Zealand Racing Board.

Local tissue concentrations of penicillin after administration of beta-lactam-based drugs into the guttural pouches of healthy horses

Anna Kendall*, Joe Mayhew* and Kiro Petrovski*

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Beta-hemolytic Streptococcus spp. consistently demonstrate high susceptibility to penicillin, and minimum inhibitory concentrations (MIC) of 0.012mg penicillin/L for S. zooepidemicus and as low as 0.009mg penicillin/L for S. equi have been shown. Treatment of chronic carriers of S. equi (the bacterium that causes strangles) with a gelatine and penicillin formulation deposited directly into the guttural pouches has been proposed by several authors, and local antimicrobial treatment has many advantages including decreased exposure of resident flora in other parts of the body to the antibiotic, which may reduce the risk of adverse effects such as development of antimicrobial resistance and antibiotic-induced colitis. However, this treatment recommendation is based on case observations alone, and data on tissue concentrations of the antibiotic after local treatment is lacking.

The aim of this study was to analyse tissue levels of penicillin after local administration into the guttural pouches of healthy horses. Four horses received local treatment into the guttural pouch and tissue levels of penicillin were analysed either 12 or 24 hours later. In this study, tissue penicillin concentrations achieved were inconsistent between horses (ranging from < 0.01mg/kg – 7.81 mg/kg, mg/kg considered equivalent to mg/L) but in most samples the levels were sufficiently above the MIC for S. equi to be effective. Results suggest that local treatment may be effective, but that more research on optimal drug formulation is warranted.

It should be noted that although these drugs are commonly used for local treatment in the guttural pouch of horses with S. equi infection, their use is off-label. One horse in this study showed evidence of inflammation of the guttural pouches after treatment with the intramammary formulation and the potential adverse reactions need to be determined.

The authors would like to thank the NZERF for their financial support which made it possible to execute this project.
NEW PROJECTS SUPPORTED BY THE NZERF

The identification of foal gastroenteritis using molecular techniques

**Dr Ray Cursons, University of Waikato**

Gastroenteritis (“scours”) is a common disease of foals, especially in young foals and in the post-weaning period. It can have different causes such as nutritional upsets or any one of a number of infectious agents. Although most episodes of gastroenteritis resolve with appropriate rehydration therapy, some persist and require additional antimicrobial treatment to clear the infection. However, effective treatment for infectious cases requires knowledge of the aetiological agent or agents involved, not only for treatment purposes but also for infection control. Traditional microbiological culture methods are often too slow or do not provide a meaningful result, especially in those gastroenteritis cases caused by enteric toxins.

To circumvent many of the problems associated with traditional microbiological investigations, I propose the use of molecular biology (namely Polymerase Chain Reaction) to look for gene-targets that are specific for a number of common gastrointestinal pathogens. The use of molecular diagnostic markers combined with multiplex polymerase chain reaction technology has previously been shown to be very effective and more sensitive than traditional microbiological culture for the diagnosis of diarrhoea. In this study I will investigate the occurrence of the five most common bacterial gastrointestinal pathogens, namely *Lawsonia intracellularis*, *Rhodococcus equi*, *Salmonella* sp., and the toxins A and B for *Clostridium difficile*. I also intend to use a Rotavirus latex agglutination test for Rotaviruses. The end objective of this research is to provide practitioners with a rapid test in order to effectively manage outbreaks of gastroenteritis in foals.

Streptococcus zooepidemicus in New Zealand horses

**Dr. Ray Cursons and Olivia Patty
University of Waikato**

Respiratory and uterine diseases play an important role in the equine industry with large welfare and economic implications. The bacterium *Streptococcus zooepidemicus* is highly diverse and generally a part of the normal microflora of the horse’s mucosal surfaces. Traditionally it is associated with opportunistic infection in the horse, only causing disease in situations of stress such as secondary to virus infection, heat stress or tissue injury. Infection with *S. zooepidemicus* has been linked to respiratory disease, foal pneumonia, endometritis and abortion. It has also been isolated from equine joints, lymph nodes, nasal cavities, lungs and reproductive tracts. Infection in humans can also occur, causing a serious illness that is known as a zoonotic (spreading from animals to humans) infection. Furthermore, it is the cause of severe infection in a number of other animals. Current research has revealed that only certain strains of *S. zooepidemicus* cause disease. Historically these disease-causing (virulent) strains have been impossible to distinguish from the non-disease causing (avirulent) strains that colonise the horse. In virulent strains, the acquisition and expression of different virulence genes is what gives these strains their disease-causing capabilities. Sophisticated molecular methods now allow the differentiation of these different strains using these virulence genes.

This research will provide information on the prevalence of *S. zooepidemicus* in respiratory and uterine infections in New Zealand horses. It will also investigate the strain characteristics associated with disease to broaden the understanding of the different strains and their potential to cause disease.
The following article summarises the main papers on fertility of the mare that were presented at ISER in January 2014.

SEASONALITY
In both hemispheres the artificial start to the breeding season in Thoroughbreds is out of synchrony with the natural breeding season and this provides challenges to the breeder to get early foals. Furthermore, the New Zealand Thoroughbred breeding season is short, and so it was with interest that we learnt of the efficacy of a light mask (www.Equil.urme.com) to advance cyclicity in mares. Several studies evaluated the roles of dopamine and prolactin in follicle development. One USA study showed that the use of drugs to inhibit prolactin did not effectively advance the onset of cyclicity in mares. More complicated protocols involving oestrogen, dopamine and sulpiride were better at advancing the onset of cyclicity than non-treated controls. Of great interest was the use of recombinant gonadotropins to make mares ovulate and maintain pregnancy during winter and early in the season without exposure to lights. Unfortunately, these hormones are not commercially available.

CYCLING MARES
USA studies have shown that in horses, as in women, antimullerian hormone (AMH) is highly correlated with follicle reserves in old mares (>18yo). Another USA study illustrated the differences in the follicular environment and oocyte (egg) lipid content in obese mares with metabolic syndrome compared to normal mares. Furthermore, short term obesity also affected the quality of the follicular fluid and oocyte lipids. Polish researchers demonstrated that follicles in 6 month-old fillies are capable of producing steroid hormones and that androgens are involved in development of the equine ovary. Then the Brazilians described the differences in protein expression in the follicles during development from 10–40mm; these studies will provide information about mare factors ultimately affecting the quality of the oocyte. More studies in Brazil also revealed that young mares (<12yo) take longer to respond to Deslorelin (43h) than to Chorulon (39h), whereas there was no difference in the time to ovulation after induction with either hormone in old mares (41-44h).

After ovulation, the corpus luteum (CL) forms. Polish and Portuguese researchers described how inflammatory cytokines acted in groups to increase prostaglandin (PGF2α) production and result in luteolysis (demise of the CL). Another collaborative study between Poland and Japan found that a different prostaglandin (PGE) supported maintenance of the CL. Then the Polish and Portuguese demonstrated the detrimental effects of feeding lucerne to cycling mares due to the oestrogenic effects of this feed and its ability to disrupt the balance of PGF2α and PGE in the reproductive tract, adversely affecting cyclicity and pregnancy maintenance. Another collaborative study between the Poles and Portuguese illustrated the fine balancing act of different proteins in controlling PGF2α and PGE. Then the Brazilians described how in horses, only a low dose of PGF2α is required to bring the mare back into season, compared with other species (eg: cattle) because of an auto-amplification system recently identified in horses. The administration of serial prostaglandin injections immediately after ovulation will prevent the rise in progesterone, thus making the next follicle available for breeding. New tests were validated by the Japanese for mare-side testing of progesterone within 25min of sampling. There were contradictory results from studies evaluating the benefits of Chorulon administration during dioestrus to improve progesterone levels or pregnancy rates.

ENDOMETRITIS
The prevention and treatment of endometritis is now a routine part of many breeding programmes, whether it is for old mares or after frozen semen insemination. A USA study of a research herd of Shetland ponies under natural mating conditions in the field found that they also experienced post breeding endometritis. Another study illustrated the beneficial effect of stallion vocalisation to induce uterine contractions and this effect was replicated with recorded stallion sounds. A Swiss study found that there was a lot of variability amongst mares in their oxytocin release patterns in response to teasing by a stallion. Another Swiss study also showed that mares exposed to stallions year-round will cycle earlier in the season than mares not exposed to stallions year-round. The Japanese demonstrated how older mares have reduced expression of lactoferrin in their uterus than younger mares and this may result in an increased susceptibility to infection.
In a USA study using dead sperm to induce endometritis, it was found that supplementation of mares with omega-3 oils for 63 days significantly reduced post breeding endometritis. Another study showed that there were mild differences in the uterine inflammatory response after insemination with live versus dead sperm.

Some of the serious pathogens involved in endometritis are gram negative bacteria (E.coli, Pseudomonas, Klebsiella) that produce biofilms which confer resistance of the bacteria to antibiotics. Unfortunately, a USA study showed that uterine treatments popular in the USA (acetylcysteine and Tris-EDTA) were not effective at reducing biofilms.

At 6h after breeding, there is a peak in inflammation with the expression of approximately 300 genes that are involved in this inflammatory response. The use of corticosteroids to reduce the severity of endometritis has been shown to result in differences in protein expression in the uterus compared to non-treated controls, which may explain their beneficial mechanism of action. New treatments such as platelet rich plasma (PRP) have been shown to be effective at reducing inflammatory markers and improve pregnancy rates in mares susceptible to post breeding endometritis. Stem cell injections into the endometrium improved the uterine condition in older mares. Kerosene treatment of the uterus, originally described by our own NZ Dr Charles Roberts, was revisited by the Americans. They found that despite the intense inflammatory response to kerosene treatment seen at 24h after treatment with a reduction in the ciliated cells lining the uterus, by 4 days after treatment the numbers of ciliated cells returned to normal levels.

CERVICAL PROBLEMS
A study in Kentucky of 239 cases of surgical repair of cervical trauma revealed that after repair the per cycle pregnancy rate of the mares was 67% when compared to 42% per cycle pregnancy rate in untreated horses. If left untreated the mares had a higher incidence of embryonic loss or abortion if they did conceive.

The cervix of older maiden mares may fail to relax during oestrus. The Germans demonstrated the efficacy of using either PGE gel or oxytocin treatment to relax the oestrous cervix of these mares.

Overall, these papers revealed a lot of new information, with particular regard to the gene expression for proteins involved in reproduction. They also elaborated on and explained the mechanisms of actions of many aspects of reproduction that we take for granted. This understanding of reproductive biology will enhance our ability to develop and apply new treatments to improve cyclicity and treat endometritis.

Next issue: Part III: The pregnant mare and perinatology.

Copies of the scientific proceedings are available for purchase for a limited time. Please contact Dr Lee Morris, EquiBreed NZ. ph: 07 870 1845
email: lee@equibreed.co.nz

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Chairperson’s Corner

Since its formation in 1976, the NZ Equine Research Foundation has been very fortunate to have the services of a number of highly motivated and skilled Board members, some of whom have given many years to our organization. Over time the Board membership has grown so that currently it has representatives from across the equine industry including the racing sector (NZ Thoroughbred Racing, Harness Racing NZ, NZ Thoroughbred Breeders Association, NZ Standardbred Breeders Association and the NZ Racing Board), the sport horse sector (Equestrian Sport NZ, NZ Pony Clubs Association), veterinarians (NZ Equine Veterinary Association), and Massey University. At our July Board meeting in Palmerston North we were very pleased to have two former members join us for lunch – Dr Brian Goulden, former Chairman (1976-1988) and Dr Ian Anderson, a long standing member who also organized the annual Bomac (now Bayer) Lecture series. As a result of the commitment and foresight brought to the NZERF by such former Board members we now have a robust and effective organization that continues to pursue its original aims of providing education and support to people working in the equine industry in NZ and improving the outcomes for the horses in NZ. So thank you to all former Board members, including Dr Goulden and Dr Anderson!

L to R: Dr John O’Flaherty (former Chairman and current Board member), Dr Brian Goulden (former Chairman), Dr Ian Anderson (former Board member) and Dr Margaret Evans (current Chairman).

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The NZ Thoroughbred Breeders’ Association (Newsletter)
NZ Thoroughbred Racing (NZTR Thoroughbred Racing Monthly)
Harness Racing New Zealand (Harness Racing Weekly)
The NZ Standardbred Breeders’ Association (Newsletter)
Equestrian Sport New Zealand (ESNZ Bulletin)
The NZ Equine Veterinary Association (Equine Veterinary Practitioner)
NZ Farriers Association (Inc) (N.Z.E.A. Newsletter)
NZ Thoroughbred Owners Federation (Bulletin)
Taranaki Miniature Horses (Newsletter)
The Morgan Horse Association of NZ Inc. (Newsletter)
NZ Hanoverian Society (Inc.) (Newsletter)