

GLUCK EQUINE

Research & Service Report

Research and Development of Fecal Egg Count Methods

Paul Slusarewicz, PhD, adjunct professor at the University of Kentucky Gluck Equine Research Center, and co-founder and chief scientific officer at MEP Equine Solutions LLC, is developing a method to rapidly detect and count the number of parasite eggs in feces. Slusarewicz, who began this work as a visiting scholar at UK, has been collaborating with and working in the lab of Martin Nielsen, DVM, PhD, DEVPC, DACVM, assistant professor in the Department of Veterinary Science at the Gluck Center.

Slusarewicz's previous CEO at a different company, local businessman Eric Hauck, introduced Slusarewicz to Nielsen, and the three co-founded MEP Equine Solutions LLC to research, develop, and commercialize the technology behind the Parasight System, which uses a smartphone to carry out fecal egg diagnostics.



The Parasight Systems fully functional prototype imaging device.

Jessica Scare, PhD candidate in Nielsen's lab, assisted with early validation work. Stefanie Pagano, master's student of Biomedical Engineering at UK, and Chris Mills, a senior in Biosystems Engineering also at UK, also helped in the lab. As part of a UK-wide effort, the egg-binding protein that is central to the technology behind the Parasight System was produced by professors Mike Mendenhall and David Rodgers at Protein Core in the UK Center for Molecular Medicine.

Slusarewicz is a biochemist with experience in pharmaceuticals, biologics, and medical devices. He began his work in the equine field when he was able to transfer his work with proteins in humans to heal tendons in equines.

The idea for an easier method for fecal egg counting came in March 2014 and he began work in Nielsen's lab in June 2014, after Hauck raised research money from investors. The product takes a fecal sample, treats it with various chemicals that make the eggs glow green when illuminated with blue light, and then uses an iPhone to photograph and count the par-

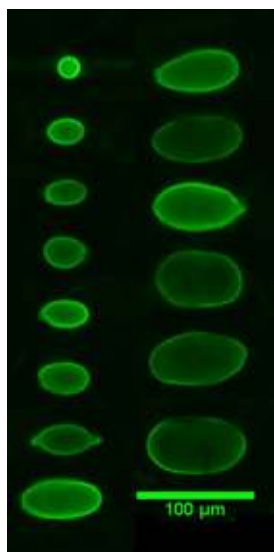
asite eggs. The whole process takes less than five minutes. This technology can identify parasite eggs of different parasite classes such as ascarids and strongyles in horses.

The Parasight System was developed to be simple and precise, while useable both on-site and in any veterinary practice. It is an alternative to the current McMaster and Stoll egg counting methods, which require a lab and lab technician to perform microscopy and manually count each individual egg with a clicker. The product prototype received an overwhelming response at the American Association of Equine Practitioners Annual Convention in Salt Lake City, Utah, Dec. 6-10, 2014.

Slusarewicz said, "If you make the process easier, people will use it."

Slusarewicz's work will impact the equine industry by changing how equines are treated for parasites. Knowing precisely what parasites to target and even whether de-worming is necessary at all will lead to more effective treatment. This means less drug resistance from parasites and fewer chemicals put into equines. He also highlighted that this technology is not limited to equines; it can be used to detect parasites in many species such as household pets, cows, sheep, and goats.

According to Slusarewicz, there are several directions he would like to continue research and development from this point. The current project focuses on creating an ambulatory product, but a sophisticated desktop system with a better camera



A microscope image of a wide variety of parasite eggs and oocysts from a single cow sample stained using the dye developed by Dr. Slusarewicz at the Gluck Center.



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*Interim Executive Director and
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Gluck Equine Research Foundation
108 Gluck Equine Research Center
Lexington, KY 40546-0099

Phone: (859) 218-1089

Fax: (859) 257-8542

E-mail: jenny.evans@uky.edu

Website: <http://ca.uky.edu/gluck>

Layout by Hannah Forte

Cover photo by Erin Morgan

*The mission of the Gluck Equine Research Center is
scientific discovery, education and dissemination of
knowledge for the benefit of the health and
well-being of horses.*

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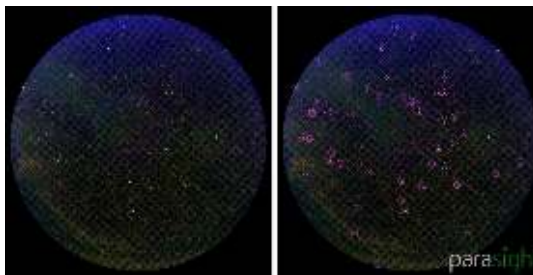
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Fecal Egg Count, continued

that can discriminate a wider variety of eggs and an automated system for use by a commercial mail-in service are other ways this technology could be utilized.

Looking further ahead, Slusarewicz said that he has ideas on how to develop a test using color change in a tube, so animal owners can track treatment progress themselves. Also, the identification of antigens on the eggs of particularly pathogenic parasite species, which would allow their detection in animals, is another area for development.

Slusarewicz said, "I envision a more sensible and precise method for fecal egg counting that is more convenient for veterinarians and animal owners. With this method, a microscope will no longer be needed, and the identification of eggs will not depend on the subjectivity of the person performing the test. This will all make it easy for everyone to do the right thing, by testing instead of treating prophylac-



The Parasight System's app in action. An image captured using an iPhone on the imaging unit before and after counting by The Parasight System's iPhone app.



Fluorescing equine strongyle eggs stained by The Parasight System and photographed using an iPhone 5s attached to The Parasight System's imaging device.

tically, and so prevent the ever-growing problem of parasite drug resistance."

At this time, Slusarewicz and his team are working hard towards commercialization and expect that the product will be available to equine veterinarians in less than a year. The Parasight System was recently given an outstanding rating and awarded a \$100,000 Small Business Innovation Research (SBIR) grant by the U.S. Department of Agriculture. For more information, visit <http://www.theparasight-system.com/technology/>.

--Hannah Forte

Survey Finds Farm Managers Practice Outdated Parasite Control Strategies

A recent survey conducted by researchers at the University of Kentucky found a high percentage of Thoroughbred farm managers used rotational deworming without monitoring drug effectiveness.

Martin Nielsen, DVM, PhD, DEVPC, DACVM assistant professor at the UK Gluck Equine Research Center, and Jill Stowe, PhD, director of UK Ag Equine Programs and associate professor in agricultural economics, teamed up for the study.

A survey was sent to registered Thoroughbred farm managers in Kentucky to investigate their current approaches for parasite control, their awareness of drug-resistant parasites, and possible willingness to change their current approaches. The study was conducted to understand how experienced farm managers were approaching parasite control.

The survey showed nearly 70% utilized rotational deworming with no monitoring of drug efficacy even though 80% of those surveyed indicated they take veterinarian advice into consideration for constructing their deworming strategies. The survey also found respondents dewormed horses five to seven times per

year, depending on the age of horses.

"Chances of finding an equine operation without drug-resistant parasites have become very slim in recent years," Nielsen said. "But the study suggests that although managers may be motivated to develop more sustainable deworming strategies, very few have actually done so."

Monitoring of parasite egg shedding and evaluation of treatment efficacy are rarely carried out, and treatment intensities are substantially higher than recommended. The study indicates a need for better dissemination of parasite control recommendations to farm managers and veterinarians.

"As scientists and experts we clearly have not communicated our recommendations effectively to the end users," Nielsen said.

Respondents appeared willing to invest time and/or money in revised or new parasite control programs only if they could be assured success of avoiding resistance and parasitic diseases. Farms closer to the Lexington area were also more likely to consider paying a premium for surveillance-based parasite control strategies. Respondents who have also started us-

ing fecal egg counts in their deworming strategies were also more likely to pay a premium.

"This study shows that in addition to effectiveness of treatment strategies, farm managers also factor efficiency and time costs into their decision-making," Stowe said. "They are willing to adopt more expensive and time-intensive strategies only if those strategies are guaranteed to work exceptionally well."

Nielsen added it is important for farm managers to be aware they might be using drugs which no longer work due to drug resistance.

The paper titled "Attitudes towards implementation of surveillance-based parasite control on Kentucky Thoroughbred farms – current strategies, awareness, and willingness-to-pay" was published in Equine Veterinary Journal.

For more information on parasite control guidelines, see the American Association for Equine Practitioners (AAEP) publication: <http://www.aaep.org/cust-docs/ParasiteControlGuidelinesFinal.pdf?osCsid=mn8molmtif6o0o0rrrksh0eac2>. Nielsen chaired the task force for the publication.

--Jenny Evans

Parasite Control in Young Horses

Martin Nielsen, DVM, PhD, DEVPC, MDACVM, assistant professor in the University of Kentucky department of veterinary science at the Gluck Equine Research Center, lectured on parasite control in young horses at the 6th annual Kentucky Breeders' Short Course Jan. 24 in Lexington, Ky.

Nielsen emphasized the occurrence of anthelmintic resistance during his presentation and highlighted the various dewormers that have the greatest and poorest effect today.

Traditionally, farm managers have opted for year-round treatments applied at regular intervals, often referred to as rotational deworming. This was thought to provide continual protection from parasites. Nielsen referenced a study conducted by the National Animal Health Monitoring System in 1998, which showed United States horse farms using an average of four or more treatments per year. Similar international studies show South African and United Kingdom farms using an average of five to seven treatments per year.

"But change is coming," Nielsen said. "We did not eradicate a single parasite species. We have instead encountered the dreaded 'R' word - resistance. There is no single drug that guarantees full effect."

Strongyles and ascarids have become resistant to many of the dewormers used today. All of the drugs labeled for equine usage today (ivermectin, moxidectin, fenbendazole, and pyrantel) have lost efficacy to at least one type of equine parasite.

"At the same time, all drugs still have efficacy against some parasites," Nielsen said. "But they have lost efficacy against others. Instead of being broad-spectrum, drug classes are now complementary to each other."

Nielsen utilized a jigsaw puzzle as an analogy. To obtain the best parasite control possible, farm managers must employ different drugs that work together to provide a broad range of control. For example, benzimidazoles (fenbendazole and oxbendazole) have strong efficacy against roundworms and large strongyles, but small strongyles have become widely resistant to this drug class.

Instead of using rotational deworming, the latest guidelines recommend performing fecal egg counts and treating according to results. An egg count can be used to evaluate the level of ascarid and strongyle egg shedding in a horse and determine the level of dewormer efficacy against each of these. Knowledge about the relative presence of ascarids and strongyles in a foal can

enable an owner or manager to identify the appropriate dewormer.

During his talk, Nielsen focused on parasite control in foals. Foals experience their highest levels of ascarids between 4 and 5 months old, after which time the strongyles take over. Tapeworms can occur after 6 months old. The first deworming should therefore focus on the ascarids at about 2 to 3 months old. At weaning, both parasite types may be present, and an egg count will reveal if ascarids are still the dominant type. At 8 to 9 months old, a weanling should be treated primarily for strongyles and possibly tapeworms. When a horse is a year old, another treatment should target strongyles and possibly tapeworms.

Nielsen stressed that a foal should receive four to five treatments within its first year of life. Treating less than this is not recommended due to the risk of parasite-associated disease, whereas more frequent treatments would need considerable justification. More information can be found in Nielsen's book, co-authored with Craig Reinemeyer, "Handbook of Equine Parasite Control." The book focuses on the biology of parasites, the general principles of parasite control, and the assessment tools for interpreting laboratory information.

--Jackson Wells

Graduate Student Spotlight: Emily Rubinson

From: Bethesda, Md.

Degrees and institutes where received: Bachelors of Art, Smith College, Massachusetts; Masters of Science, University of Kentucky

Emily Rubinson wanted to pursue a degree at the University of Kentucky Gluck Equine Research Center so she could apply the molecular and microbiological techniques she had used in human medicine to animals.

"And since I am in Kentucky, I wanted to get some experience with horses," Rubinson said. "I have been fortunate to perform my research under the supervision of Dr. Martin Nielsen (DVM, PhD, EVPC, assistant professor at the Gluck Center)."

The main hypothesis for Rubinson's research project was the inflammatory reaction to vaccination, and if the resulting vaccine efficacy will be affected by deworming the horse simultaneously.

Unlike humans, horses are never completely free of parasites, but parasite bur-

dens are largely impacted by the regular deworming treatments often practiced on horse farms. It is also very common to carry out vaccination and deworming on the same day, but Rubinson said the question is, "Is the vaccine response altered by deworming?"

In recent years, research in humans has shown possibly beneficial effects of worm parasites in the gastrointestinal tract, Rubinson said. These worms have been found to reduce and change the inflammatory response and can counteract various allergic conditions, such as asthma.

"We don't know, however, if similar mechanisms might be in play in the horse," Rubinson said.

To test the hypothesis, Rubinson said, "We measured the inflammatory response (acute phase proteins, hematology, and cytokines) in response to a combination of three different vaccinations given to yearling ponies. All ponies were vaccinated with the same three vaccines, but one group was dewormed with ivermectin, another with pyrantel pamoate, and the last group

was not dewormed."

Rubinson's research found that the inflammatory response to vaccination was affected by deworming to some extent, but the vaccine antibody titers were not significantly different between the groups.

"Therefore, our conclusion was that vaccination and deworming can be carried out on the same day without losing vaccination effects," she said.

Rubinson earned her master's degree at UK in December. As for the future, Rubinson hopes to work in the field of veterinary microbiology, preferably parasitology, and then after a few years go back to school to get her doctoral degree.

--Shaila Sigsgard



Emily Rubinson

Study Finds Common Equine Parasite Misidentified in Textbooks

A recent study led by Martin Nielsen, ADVM, PhD, DEVPC, DACVM, assistant professor at the University of Kentucky Gluck Equine Research Center, found that all veterinary medicine textbooks have misidentified a common equine parasite.

The large equine roundworm *Parascaris equorum*, commonly referred to as the ascarid, which is known for infecting foals, is actually a different species—*Parascaris univalens*. The research suggests *P. univalens* is the main species now observed in equines. The broader designation *Parascaris* spp. should be used instead unless cytological characterization (a technique for characterizing chromosomes) has confirmed the species.

“*Parascaris univalens* is really the forgotten parasite,” Nielsen said. “It is almost never mentioned in the textbooks, and most people have only heard about one roundworm species infecting equids.”

P. univalens was discovered more than 130 years ago. The species only possesses one germ line chromosome pair as opposed to two for *P. equorum*, but the two species are otherwise considered structurally identical.

“We really wanted to find specimens of both species to study and find differ-

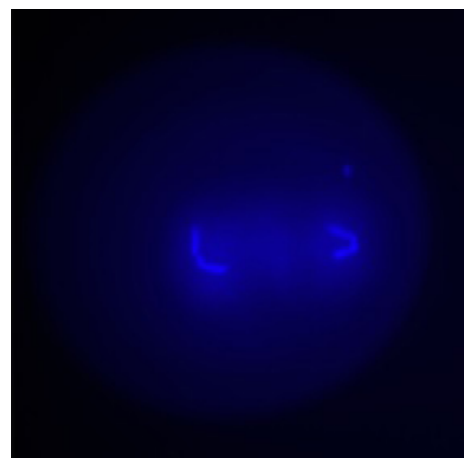
ences in their DNA,” Nielsen said. “The only way to tell them apart is to look at their chromosomes, so we invited a leading expert, Dr. Clara Goday, to the Gluck Equine Research Center to teach us the delicate technique of parasite karyotyping.”

Karyotyping is a technique to study and characterize chromosomes in a sample of cells.

For the study, 30 live worms were obtained and dissected. All of the samples were identified as *P. univalens*. Then, the karyotyping technique was performed on ascarid eggs from foal fecal samples. *P. equorum* was not identified among these, whereas *P. univalens* was found in 17 samples, with the remaining eight being inconclusive.

“We were part of another study analyzing numerous *Parascaris* specimens from several different continents, and the conclusion there was that only one species was found,” Nielsen said. “We compared genetic information obtained for *P. univalens* in our study with gene codes already published as *P. equorum* and found that they were probably mislabeled.”

Others from the UK Gluck Center involved in the study were Jennifer Bellaw, a PhD candidate in veterinary science,



Parascaris univalens was given its name because of the single chromosome pair (univalent). The image shows the two chromosomes labeled with a fluorescent dye.

Eugene Lyons, PhD, professor, and Teri Lear, PhD, associate professor. The group from the UK Gluck Center collaborated with Jianbin Wang, PhD, assistant professor, and Richard Davis, PhD, professor, University of Colorado School of Medicine, and Clara Goday, PhD, at Centro de Investigaciones Biológicas in Spain.

The article was published in the December issue of *Parasitology Research*.

--Jenny Evans

Emma Adam Receives AAEP Foundation Past Presidents' Research Fellow

University of Kentucky doctoral candidate Emma Adam, BVetMed, MRCVS, DACVIM, DACVS, received the 2014 AAEP Foundation Past Presidents' Research Fellow for her research into articular cartilage repair of degenerative joint disease, the leading cause of lameness and retirement in all breeds and disciplines of equine athletes.

Dr. Adam was recognized Dec. 8 during the Frank J. Milne State-of-the-Art Lecture at the AAEP's 60th Annual Convention in Salt Lake City, Utah. The \$5,000 grant is awarded annually to a doctoral or residency student who has made significant progress in the field of equine health care research.

Dr. Adam's research seeks to enhance understanding of articular chondrocytes' exact gene expression in order to recapitulate them experimentally as therapy

cells for articular cartilage lesions. Using RNA-sequencing scrutiny of the genes expressed in cartilage at different stages of development, the goal is to obtain new insight and knowledge into what defines a mature, robust articular chondrocyte. Such understanding will advance therapeutic efforts to generate and support fully functional articular cartilage cells during tissue repair.

Dr. Adam received her veterinary degree from the Royal Veterinary College at the University of London in 1993. She completed her large animal internal medicine residency at Texas A&M University in 2004 and large animal surgery residency at the University of Pennsylvania's New Bolton Center in 2007. Her Ph.D. dissertation research is being conducted at the University of Kentucky's Gluck Equine Research Center under the mentorship of

Dr. James MacLeod.

“We are delighted to honor Dr. Adam and thank her for continuing her education and career toward equine research,” said AAEP Foundation Chairman Jeff Berk, VMD. “If our knowledge of horse health is going to continue to advance, we must encourage and financially support more veterinarians to become researchers like Dr. Adam.”

The AAEP Foundation established the Past Presidents' Research Fellow in 2006. The award is made possible through the monetary contributions of AAEP past presidents. For more information about this program and other scholarships offered through the AAEP Foundation, please visit www.aaepfoundation.org and select “Scholarships” in the “Apply for Funding” drop-down menu.

--Edited Press Release

Reproduction in Geriatric Mares

Barry Ball, DVM, PhD, Dipl. ACT, Albert G. Clay endowed chair in equine reproduction and professor in the department of veterinary science at the University of Kentucky Gluck Equine Research Center, gave a talk about reproduction in the geriatric mare at the 6th Annual Kentucky Breeders' Short Course in Lexington, Ky., Jan. 24.

The older mare undergoes several uterine changes. Ball focused on pregnancy and ovarian, oviductal, and uterine factors.

Unlike humans, mares do not undergo menopause, although their eggs do deplete eventually. Ball emphasized that the actual age and reproductive age of a mare are not the same. The reproductive age is dependent on the number of oocytes (eggs) populated in the mare's ovary at birth. Therefore, fertility decreases as the antral follicle count, or ovarian reserve, diminishes. Ovarian senescence refers to an ovary with no antral follicles or a depleted ovarian reserve.

According to Ball, there will eventually be a marker to determine the reproductive age of older mares. This would benefit the equine industry by allowing individuals to predict how fast the mare will

age reproductively.

Anovulatory hemorrhagic follicles (AHF) are follicles that do not ovulate but fill with a blood clot. AHF incidence is directly correlated to increasing age in mares, and is also associated with prolonged length of the estrous cycle, or days between ovulation, if a mare is bred but does not ovulate. This causes low fertility.

In the oviduct, the geriatric mare has a higher risk for oviductal plugs. W.R. Twink Allen, BVSc, PhD, ScD, DESM, MRCVS, director at the Paul Mellon Laboratory of Equine Reproduction, developed technology using a hormone called prostaglandin E (PGE2) to manage these mares. Using PGE2 as a topical treatment for relaxing the smooth muscle of the oviduct looks to be a possible solution for releasing these plugs from the oviduct, Ball said.

The geriatric mare has an increased risk of uterine problems such as endometritis, endometrial cysts, and endometriosis. Endometrial cysts take the form of fluid-filled structures inside of the uterus that are caused by dilated lymphatics. Angiosis of the uterine arterioles is vascular disease associated with

pregnancy. Endometriosis is a degenerative change that occurs in older mares.

A mare older than 10 years of age who has not previously given birth may have issues with cervical changes and adhesions, which can cause fluid accumulation in the uterus and delayed uterine clearance of fluid. Endometritis is the inflammation of the uterine lining. It can result in early embryonic loss. Proteins in the seminal plasma, which may act to reduce inflammation and protect sperm from inflammatory response, could help resolve persistent endometritis.

Fertility declines when the mare is between the ages of 10 and 13. As age increases, the rate of embryonic loss increases as well. Other pregnancy problems include growth retardation of the fetus, placental insufficiency, hemorrhaging, uterine artery rupture, and ruptured prepubic tendon.

Ball concluded by restating that older mares experience a reduced oocyte (egg) reserve and increased early pregnancy loss. They are also more susceptible to uterine diseases such as endometriosis, angiosis, lymphatic cysts, and placental insufficiency.

--*Hannah Forte*

Graduate Student Spotlight: Gabriel Monteiro Davolli



Gabriel Monteiro Davolli

From: Rio Grande do Sul, Brazil

Degrees and institutes where received: Medical Veterinarian (DVM equivalent), Universidade Federal do Rio Grande do Sul, Porto Alegre, Brazil

Gabriel Davolli was involved in equine reproduction in a research

environment during veterinary school. He visited Lexington and the University of Kentucky Gluck Equine Research Center during this time, and after graduation from veterinary school in Brazil, he came back to pursue a research project through the Gluck Center as a master's student.

"I looked forward to being responsible for the different stages of a research project—from planning to execution and analysis, under the guidance of experts in the field," Davolli said.

Davolli has conducted research regarding hormonal therapy under the supervision of Ed Squires, PhD, Dipl. ACT (hon.), professor at the Gluck Center. Specifically, he has worked with hormones in stallions to suppress behavior.

When asked how this research will contribute to the field of equine science, Davolli said, "It has potential applications as a behavior modulator, which could be used as a substitute to castration in aggressive stallions, and as treatment

of stallions persistently infected with equine arteritis virus (EAV, a virus that causes equine viral arteritis [EVA], an upper respiratory tract and reproductive disease in horses). This virus depends on testosterone to be survive within the reproductive tract of carrier stallions and is cleared after prolonged testosterone depletion. These carrier stallions are known for disseminating EAV through infective semen (fresh, cooled, or frozen), leading to EVA outbreaks across states and overseas."

Davolli has also been closely involved with several research projects conducted by other doctoral students at the Gluck Center. For example, he worked with Anthony Claes, PhD, DVM, who is now an assistant professor at Utrecht University, on ovarian and hormonal changes in geriatric mares. He also worked on several projects with Igor Canisso, PhD, DVM,

Monteiro Davolli, p. 7

Monteiro Davolli, *continued*

who is now an assistant professor at the University of Illinois, involving using colostrum changes as a sign of impending foaling in mares and other projects related to placentitis.

"I was also lucky to collaborate in projects of other students and in preliminary trials of products to be used at the time of breeding and for semen processing," Davolli said. "Finally, I was part of an experiment investigating the effects of the supplementation of DHA (an omega-3

fatty acid) to stallions and its effects on semen quality."

To be involved with investigations of basic biology, as well as applied studies of interest to the industry, has been a valuable learning outcome, Davolli said.

"It is neat to understand that many times one project can accomplish both jobs if there is good planning, execution, and sound interpretation of the data," Davolli said. "On the other hand, it also teaches you to accept the inherent limitations of an experiment and how far the

information can be stretched, which is where a follow-up experiment comes in."

After finishing his master's program, Davolli plans to begin a six-month internship with Park Equine Hospital in Lexington for the foaling season. Looking to the future, Davolli said that a residency involving the holistics of reproduction would be an excellent way to see where the scientific knowledge is lacking and thinking of potential ideas for research.

--Hannah Forte

UK Researchers Study *Lawsonia intracellularis* Infection and Risk

Allen Page, PhD, DVM, a former University of Kentucky Gluck Equine Research Center post-doctoral fellow, gave a talk about *Lawsonia intracellularis* at the 4th Annual UK Equine Showcase in Lexington, Ky. Jan. 23.

Equine Proliferative Enteropathy (EPE) is a disease of foals that causes hyperplasia, or thickening of the walls of the intestinal tract.

L. intracellularis mainly affects weanlings and causes anorexia, rapid weight loss, fever, depression, rough hair coat, throat latch and ventral edema, colic, and diarrhea.

In most cases if caught early, *L. intracellularis* is not fatal; however, affected foals may take a couple months to catch up to others in body condition. In rare instances, however, clinicians have diagnosed EPE cases and treated them appropriately, yet the foals die as a result of cell injury or death of cell tissue in the intestinal tract.

In a recent study by Page and David Horohov, PhD, Jes E. and Clementine M. Schlaikjer Endowed Chair at the Gluck Equine Research Center, Interim Chair of the Department of Veterinary Science, and Interim Director of the Gluck Equine Research Center, it was found that colts were at significantly higher risk than fillies. However, numerous cases were reported in fillies as well. The study also showed that foals who were weaned after August were at lower risk for developing EPE.

"We found that mares were more at risk for exposure to *Lawsonia* during the summer months than their foals," Page said. "Since most foals were still nursing during this time and likely exposed to the same environments and materials as the mares, we interpreted this to mean that the mares were somehow protecting their foals from exposure to *Lawsonia* during the summer."

There are currently three tests available

for *Lawsonia intracellularis*--the serum immunoperoxidase monolayer assay (IPMA), the ELISA test, and a fecal PCR test. The IPMA and ELISA both test for antibodies to *Lawsonia*, whereas the PCR test detects the presence of the bacteria's DNA in feces.

In most cases, antimicrobials with intracellular penetration, including Oxytetracycline, Doxycycline, Erythromycin, and Chloramphenicol are given concurrently with supportive care such as plasma, replacement fluids, antiulcer medications, and parenteral nutrition. However, each treatment depends on the severity of the disease at the time, and in mild cases, some veterinarians skip the supportive care.

According to Page, there is a vaccine that has been approved for use in pigs but currently there is no vaccine labeled for use in horses.

--Alexandra Harper

Graduate Student Spotlight: Igor Canisso

Igor Canisso, DVM, PhD, MSc, DACT, DE-CAR, an assistant professor of equine theriogenology in the department of veterinary clinical medicine at the University of Illinois Urbana Champaign campus, graduated with a PhD in equine reproduction from the UK Maxwell H. Gluck Equine Research Center in 2014.

During his time at the Gluck center, Canisso was advised by Barry Ball, DVM, PhD, Dipl. ACT, professor and Albert G. Clay Endowed Chair in Equine Reproduction at UK's Gluck Equine Research



Canisso

Center. Canisso's primary research project was on equine placentitis, which is a major cause of late-term pregnancy loss in broodmares.

Canisso said he chose UK be-

cause it has the world's leading program in equine research, and the reproduction lab where he earned his doctoral degree is an excellent facility.

"I obtained my PhD education from the world's best equine reproduction scientists, so it was an easy decision to

From: Brazil

Degrees and institutes where received: 2003 DVM, Federal University of Parana, Brazil; 2008 MSc, Federal University of Vicosa, Brazil; 2011 Diplomate, American College of Theriogenologists, Cornell University College of Veterinary Medicine; 2012 Diplomate, European College of Animal Reproduction, Cornell University College of Veterinary Medicine; 2014 PhD, Maxwell H. Gluck Equine Research Center, University of Kentucky

join the group at the Maxwell H. Gluck Equine Research Center. Being awarded with a fellowship by the Geoffrey Hughes Foundation through the Department of Veterinary Science certainly gave me the peace of mind that I could complete my

Igor Canisso, p. 8

Igor Canisso, *continued*

graduate work without worrying about stipends,” Canisso said.

Canisso said his time studying equine placentitis was rewarding because of the advances that his group has made in the breeding segment of the “horse capitol of the world.” During his time at the Gluck Center, Canisso had the opportunity to collaborate on research with colleagues in and out of the reproduction laboratory. “I learned how to become an indepen-

dent researcher and obtained my PhD training in the center of the horse industry. [It] certainly gave me a differentiated perspective into my career as an equine reproduction scientist and clinician,” Canisso said.

Just before his PhD defense, Canisso started as an equine theriogenologist and tenure track faculty member at the University of Illinois. He expressed gratitude for the advancements made in equine science, which could not have happened

without funding from the Kentucky Thoroughbred Association and Kentucky Thoroughbred Owners and Breeders Association.

“The experiences, education and connections with the horse industry acquired while working in the bluegrass has certainly helped me in my new endeavors and journey as a clinician and scientist,” Canisso said.

--Hannah Forte

Is HIP an Effective Treatment for *R. equi*?

Fernanda Cesar, DVM, MS, DACVIM, and PhD candidate at the University of Kentucky Gluck Equine Research Center, lectured on the use of Hyperimmune Plasma (HIP) against *Rhodococcus equi* at the 6th Annual Kentucky Breeders' Short Course Jan. 24 in Lexington, Ky. Other authors on this paper included Drs. Macarena Sanz and David Horohov, also of the Gluck Center.

Rhodococcus equi (*R. equi*), a bacterium that is found worldwide, is the leading cause of pneumonia in foals 1 to 6 months old, and has a mortality rate ranging from 1% to 20%. Current antibiotic treatments for infected foals are administered orally, are prolonged and expensive.

There is no approved vaccine to prevent *R. equi*. However, farms with a history of *R. equi* infections often use ultrasound screening and aggressive antimicrobial treatments to detect and treat infected foals. However, this approach has contributed to the appearance of antimicrobial resistance in *R. equi*.

Another approach is to administer HIP to foals at birth – with a second dose at 4 to 8 weeks old – as a preventative measure against *R. equi*. The HIP contains antibodies to *R. equi* which are thought to provide protection to the foal. While the exact mechanism of this protection is unknown, Cesar focused on those antibodies directed against virulence-associated protein A (VapA), an important pathogenic factor of *R. equi*.

Prior field studies of HIP produced conflicting results. Studies from the 1990s indicated HIP administration was an

effective preventative method, yet more current studies from the 2000s showed HIP as being ineffective.



Hyperimmune Plasma against R. equi

“There are a couple of potential reasons for this [conflicting result],” Cesar said. “The age at which the foals were administered HIP varied, and each study had no definitive diagnosis.” Variations between

The take home message:

The administration of HIP seems to have a positive effect on limiting R. equi infections. If a foal did become infected, the duration and severity of illness was decreased. HIP is a safe treatment for foals, but it does not have 100% efficacy in preventing infection.

“Further work is needed, in the field, and in controlled conditions,” Cesar said, “to determine which components of HIP were involved in this protection.”

products and individual foals could also play a role.

In the first study, foals on Thoroughbred horse farms in Lexington that were routinely administered HIP were compared to those foals on control farms where HIP

was not used. Serum samples were collected and evaluated for the presence of antibodies to VapA in the foals and their mares. Four different commercial HIP products were compared; EquiplasREA, PneumomuneRE, ReSolution, and ImmunoGlo -. As the study was not funded by any of the companies that produced the HIP products, there was no conflict of interest.

The results showed that all four products exhibited variation in the amount of antibodies to VapA they contained. Further, there was also significant variability in the amount of VapA antibodies in each of the foals that received the HIP. In fact, some treated foals had fewer antibodies to VapA than did the untreated foals. This may be due to the product variation in antibody composition and/or natural variations found within foals.

In a second study to determine if HIP administration could prevent infection, a group of foals were given HIP and a second group received no treatment. Both groups of foals were exposed to *R. equi* and their clinical response to the disease was monitored.

There was no adverse reaction to the HIP in the treated foals. While the administration of the HIP did not prevent foals from becoming infected with *R. equi*, those foals receiving HIP had less severe disease and more rapidly resolved their infections.

“[It] appears that while infection was not prevented, natural regression was enhanced,” Cesar said.

--Jackson Wells

32 and My Horse

Ernie Bailey, PhD, a professor at the University of Kentucky Gluck Equine Research Center, recently gave a presentation titled “32 and My Horse” on the equine genome and genetics at a UK Equine Forum meeting. Bailey based the lecture on his personal experience with a commercial company called “23andMe” that provides genetic information for recreational related to people purposes. The numbers 23 and 32 reference the number of chromosomes humans and horses have, respectively.

Commercial companies such as “23andMe” provide what Bailey called con-

sumer genetics. Consumer genetics allows those who are not scientists to utilize genetics to identify distant relatives, recognize predilection for disease, and identify the origins of ancestors. Bailey suggested there could be a potential value of consumer genetics companies for the horse industry.

According to Bailey, an enormous part of horse breeding is looking at patterns. A common example is coat color genetics. He proposed that breeders may further benefit from looking at DNA patterns. Another example given by Bailey was about Chromosome 18, the gene associ-

ated with sprinting ability. Aside from coat color and athleticism, genetics can also be used to look at gene mutations that cause a variety of health problems.

Bailey is currently researching the genetics of Equine Arteritis Virus (EAV), the cause of Equine viral arteritis (EVA), which is a viral disease of equids, in collaboration with Gluck Center researchers Udeni Balasuriya, PhD, BVSc, MS, PhD, an associate professor of virology, and Peter Timoney, MVB, PhD, FRCVS, Frederick Van Lennep Chair in Equine Veterinary Science.

--Hannah Forte

Horse Farm Disaster Preparedness

Roberta Dwyer, DVM, MS, DACVPM, professor in the University of Kentucky Department of Veterinary Science recently spoke at a UK Equine Forum meeting and discussed horse farm disaster preparedness.

The primary point in Dwyer’s presentation was that human health and safety come first. Although she focused on natural and accidental disasters, she emphasized that it is important to have a plan for every type of disaster.

Plans should account for the unusual and unexpected, including: loss of communications, loss of electricity, lack of personnel, and limitations on transportation due to blocked roads. First start with a personal disaster plan, then create family, farm, communications, and evacuations plans. Situations to plan for include having no electricity, no movement on or off the farm, and no communication for seven or more days. “The loss of electricity can also mean no water supply for farms dependent upon well water, as well as frozen pipes in

winter,” reminded Dwyer. Also consider what would happen if there was a 24 hour notice of evacuation or downed fencing and loose animals.



Have a plan before disaster comes.

Dwyer said that people often debate over whether they should leave horses in or out during different types of storms (high winds, tornadoes, flooding, etc.). While there is no straight answer, the barn structure, age, location, and available pastures should be taken into consideration and educated decisions should be made before weather strikes. “Every

home and barn should have a NOAA weather radio for advanced warning of dangerous weather conditions” recommended Dwyer.

Dwyer explained the types of disasters, how they are declared, and what resources are available at each level. A locally declared disaster is coordinated through the county Emergency Operations Center (EOC), and a governor declared disaster is coordinated through the state EOC with state resources made available. The governor can also request federal disaster declaration from the president, which makes federal resources available. There are numerous emergency support functions ranging from firefighting to long-term community recovery.

Dwyer is also a lead instructor for the nationally funded and recognized Extension Disaster Education Network “Strengthening Community Agrosecurity Preparedness” program, a two-day workshop.

--Hannah Forte

Balasuriya Receives Bobby Pass Excellence in Grantsmanship Award

Udeni Balasuriya, BVSc, MS, PhD, a professor at the University of Kentucky Gluck Equine Research Center, received the Bobby Pass Excellence in Grantsmanship Award from Nancy Cox, PhD, dean of the College of Agriculture, Food and Environment, at the 2014 Celebration of Land-Grant Research on Nov. 18. Balasuriya received the award for leading a grant-funded project titled “Identification of Genetic Factors Responsible for Establishment of Equine Arteritis Virus Carrier State in Stallions,” which received \$2.9 million over five years from the U.S. Department of Agriculture’s National Institute of Food and Agriculture. The Bobby Pass Excellence in Grantsmanship Award is annually given in memory of former UK Department of Entomology chair Bobby Pass.



Dr. Udeni Balasuriya and Dean Nancy Cox

Lloyd's of London and UK Partnership Continues

The well-established and successful partnership between Lloyd's of London and the University of Kentucky College of Agriculture, Food and Environment continued when representatives from Lloyd's recently presented a check to UK.

"We are very pleased to announce an increase in Lloyd's annual support from \$45,000 to \$50,000," said Julian Lloyd, chair of Lloyd's Livestock Committee and bloodstock underwriter at the Am-lin Syndicate. "For 22 years, the Lloyd's Equine Disease Quarterly has served as the benchmark publication for support of equine health. Lloyd's long-standing financial commitment to the University of Kentucky exceeded \$1 million a few years ago, and this increase will help to ensure continued global distribution of this leading-edge publication."

The Lloyd's Equine Disease Quarterly, a research-based publication dedicated to equine health, is produced by the UK Department of Veterinary Science.

"The College of Agriculture, Food and Environment is grateful to Lloyd's of London for this long-standing and

unique support of the veterinary science department in general since 1985, and of the Equine Disease Quarterly since 1992," said Nancy Cox, UKAg dean and director. "This year's support brings their total investment to more than \$1.1 million. We appreciate Lloyd's long-standing



Lloyd's increases annual support of UK.

confidence in the impact of our Equine Disease Quarterly."

The award-winning publication includes articles written by prominent researchers from around the world and provides timely and authoritative reports on some of the most important issues facing the equine industry. The Quarterly reaches more than 18,000 readers in 102

countries. Available in paper and online, its articles are regularly reprinted in numerous scientific and lay equine publications worldwide.

Lloyd's Equine Disease Quarterly is available to subscribers at no charge. It is co-edited by Roberta Dwyer, Peter Timoney and Alan Loynachan from the Department of Veterinary Science.

The most recent Quarterly is online at http://www2.ca.uky.edu/gluck/q_oct14.asp. For more information about the Department of Veterinary Science and the Maxwell H. Gluck Equine Research Center visit <http://www2.ca.uky.edu/gluck/>.

Lloyd's of London is a 325-year-old insurance market whose members underwrite risk on a direct and reinsurance basis in more than 200 countries. As a global leader in specialty insurance, Lloyd's remains committed to supporting equine research and providing the insurance coverage essential to the well-being and prosperity of bloodstock interests worldwide.

--Aimee Nielson

International Research Team to Map Disease Genes in Horses

The Morris Animal Foundation has awarded a three-year, \$155,000 grant to a team of researchers from Kentucky and Denmark to build a new reference genome sequence for the domestic horse. The sequence will be a much needed tool for animal researchers worldwide and the equine industry in particular because it should significantly improve the ability to understand the role of genetics in animal health and well being.

Ted Kalbfleisch, PhD, of the University of Louisville Department of Biochemistry and Molecular Biology, is the principal investigator on the grant. He will be joined in the research with Ludovic Orlando, PhD, of the Centre for GeoGenetics at the University of Copenhagen's National History Museum, and James MacLeod, VMD, PhD, of the University of Kentucky's Gluck Equine Research Center, in Lexington.

Genome sequencing allows researchers to read and decipher genetic information found in DNA and is especially important in mapping disease genes—discov-

ering the diseases a horse might be genetically predisposed to developing.

"In 2009, Morris Animal Foundation helped fund the first genome reference sequence for the domestic horse," Kalbfleisch said. "We intend to build on this earlier work. In the past five years, there have been dramatic improvements in sequencing technology as well as the computational hardware and algorithms required to analyze the data generated by the technology. Therefore, we now have the tools necessary to vastly improve the reference genome for the horse."

The current reference genome for the horse, known as "EquCab2," has been beneficial in studying horses and their genetic predisposition to disease, but it is not without its shortcomings, Kalbfleisch said.

"The horse research community is working to understand the relationship among genomic structure, variation found within it and complex diseases and traits in the domestic horse," he said. "The EquCab2 reference genome was de-

veloped prior to the development of today's highly sophisticated technology.

"With the application of new high-throughput technologies we have available today, we will map the genome with a focus on what is known as the 'GC-rich regulatory regions.'"

These GC-rich regulatory regions control how genes are expressed (turned on) in order to participate in normal cellular processes. This work will enable scientists to better catalog genetic variation in these regions and understand how it affects health and performance.

"We expect our research to have substantial impact because the horse research community has actively moved to the translational application of genomics in examining important questions in equine science," Kalbfleisch said. "The improved reference genome we will map will directly improve both the quality and productivity of research being carried out in the equine industry."

--Edited Press Release

A New Breed of Funding

Until about a year ago, I had heard the terms “crowdfunding” and “crowdsourcing” several times without fully understanding what they meant. When my wife made a small donation toward a musician’s efforts to raise enough money to release her first album, I started to get an idea of what this was all about. The musician had posted a project description on a crowdfunding website called Kickstarter and invited people to make donations online. Word then spread through social media. The campaign was successful, and the musician reached her goal.

Soon thereafter, I was listening to NPR while driving to work one morning. The station

was broadcasting a feature about scientists using crowdfunding to raise funding for their research. The concept was the same as for the musician, but instead of an album the final product would be a funded research project. The key to success was still effective communication by means of social media. I immediately thought this would be worth trying.

Through the years I have been studying equine parasitology, I have always enjoyed how horse owners appreciate science. The Horse’s magazine, website, and newsletters serve as an excellent illustration of this; people genuinely like science and they want updated information. I figured this would provide a good foundation for a crowdfunding project. Furthermore, I felt my research topic, parasitology, would be a common denominator across various horse breeds, uses, and geographic locations. No matter what, horses will always have worms, and owners will always have opinions about and experiences with controlling them.

No one from the University of Kentucky had tried crowdfunding for research projects

before. But there was a lot of interest in it, and a crowdfunding task force had already been established. Before I knew it, I had become the university’s first pilot research crowdfunding project, examining a promising alternative treatment



Nielsen was UK’s first research crowdfunding project.

modality to reduce our reliance on existing dewormers to which parasites are developing resistance. My team is working with a strain of naturally occurring bacteria capable of killing worms; hence, the

“No matter what, horses will always have worms, and owners will always have opinions about and experiences with controlling them.”

name of our campaign is “Let the germs get the worms.”

We decided to attempt to develop our own website infrastructure for crowdfunding. By doing this, we had the freedom to design the site as we wanted it, we would avoid paying the fees associated with using a commercial crowdfunding website, and American residents could obtain tax credit for their donations. With help from many excellent people at the University of Kentucky, we were able to leverage the already existing online donation system to develop a website as well as three promotional videos. We set up a Twitter account and made use of already existing Facebook and LinkedIn accounts to promote the project. We used an on-

line group emailing service to communicate with donors and supporters.

The crowdfunding campaign launched in the beginning of January 2014 and ran for two months. During this time our page was shared and retweeted well over 400 times, the videos have been viewed more than 2,000 times, about 300 unique users signed up on the website, and we raised more than \$8,500. Further, the campaign has greatly promoted our research program, as we have appeared in several news articles and were hosted on radio shows. Interestingly, we have had some substantial donations come in after we stopped campaigning, and they don’t show signs of stopping. We intend to keep

the site open and provide regular updates about our project and future campaigns.

Our ultimate goal with this project is to develop a probiotic-type product for horses, but the first step is to test it against equine parasites in the laboratory. This is what we will use the crowdfunding money for, and we have already started the work. Preliminary data show a very good

effect of our test bacteria against equine small strongyles, and we are continuing to evaluate this further. You can learn more about the project and follow our progress at <http://equineparasitology.ca.uky.edu>.

--Martin Nielsen



University of Kentucky Gluck Equine Research Center

The Gluck Equine Research Center encompasses faculty members that conduct equine research full time in the areas of infectious diseases and immunology, genetics and genomics, musculoskeletal science, parasitology, pharmacology/toxicology and reproductive health.

Gifts to the Gluck Equine Research Center are used to support research, build facilities, purchase equipment, provide scholarships for graduate students, create educational material and provide continuing education programs for Veterinarians and Horsemen.

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