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August 30, 2018

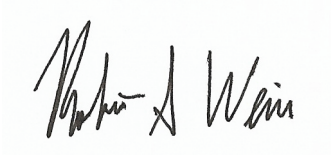
Ms. Tracy Egan
Executive Director NYS
Thoroughbred Development Fund
Schenectady, NY 12305

Dear Ms. Egan,

Enclosed is an electronic copy of the complete annual report for the 2017 Harry M. Zweig Memorial Fund for Equine Research, covering the award period January 1, 2017 through December 31, 2017. The attached version is not for the web, and Laura Mathews will email you a copy specifically for uploading to the web (to include lay summaries vs the full grant information). We apologize for the delay in getting this report to you this year, and we will make every effort to stick with the spring schedule next year.

We hosted a series of Research Presentations highlighting equine research at Cornell funded in part by the Zweig fund on November 29, 2017 at the Cornell College of Veterinary Medicine in Ithaca, New York. The talks were given by various Cornell College of Veterinary Medicine faculty, and covered an array of equine research topics. The Zweig Committee made a special effort to attend by arriving a day ahead of the annual meeting, and other faculty, scientific staff, and administrators attended the event, which was followed by a poster session and reception outside in the new atrium on the first floor of Schurman Hall. Additional information can be found on the Zweig public site at http://www.vet.cornell.edu/research/Zweig/

On behalf of Cornell University, we wish to extend our appreciation for your continued support of equine research.



Robert S. Weiss, Ph.D.

Professor of Molecular Genetics
Associate Dean for Research & Graduate Education

cc: Jill LaBoissiere - Comptroller - NYS Thoroughbred Breeding & Development Fund
Lorin D. Warnick, PhD, Austin O. Hooey Dean of Veterinary Medicine

Cornell University
College of Veterinary Medicine

**and the**

**Harry M. Zweig Memorial Fund for Equine Research**

# Annual Report 2017


# 2017 Harry M. Zweig Memorial Fund for Equine Research Summary Report

The 2017 Annual Report covering the period of January 1, 2017 through December 31, 2017 is provided herein.

For this reporting period, The Harry M. Zweig Memorial Fund for Equine Research Committee awarded funding for 8 of 11 submitted projects. Seven of the projects were new, and one was a renewal. There were also two continuation awards approved for the second year funding period. The total amount allocated for 2017 awards was $449,675. This report includes the complete study, and a separate report consisting of “lay summaries” will be emailed to you for the public website.

Additionally, on Wednesday, November 29, 2017 the Veterinary College hosted the 9th annual poster session and scientific talks, celebrating the collaboration between the Harry M. Zweig Memorial Fund for Equine Research and Cornell University College of Veterinary Medicine. Participants included Cornell faculty, students and scientific staff, showcasing their research to the community and to the Zweig Committee.

***2017 Harry M. Zweig Memorial Fund for Equine Research Awards***

| **CONTINUATION** | | **ANNUAL AWARD** |
|---|---|---|
| Dr. Mohammed | Factors Associated with Musculoskeletal Injuries & Catastrophic Events in Racing Horses (year-2) | $75,676 |
| Dr. Wagner | Effects of Equine Herpesvirus (EHV) Vaccination Frequency on Host Immunity to EHV-1 | $78,993 |
| | Sub-Total: | *$154,669* |
| **NEW/Renewal** | | **ANNUAL AWARD** |
| Dr. Antczak | Cellular Immunity to Equine Herpesvirus Type 1 (EHV-1) (1 year award) | $68,410 |
| Dr. Antczak | Functional Gene Annotation in the Horse (2 year award) | $64,663 |
| Dr. Cheetham | Regenerative Approach to Recurrent laryngeal Neuropathy (2 year award) | $43,130 |
| Dr. Ducharme | Two-Day Tie-Back (injection Laryngoplasty); Proof of Principle (1 year award) | $60,000 |
| Dr. Fortier | Does Intra-Articular Stifle Anesthesia Alleviate Lower Limb Lameness? (1 Year award) | $43,032 |
| Dr. Fubini | The Relationship between obesity and Post-Operative Incisional Infections following Abdominal Surgery in the horse (2 year award) | $18,120 |
| Dr. Reesink | Quantitative Computed Tomography & Bone Quality Assessment for the Prediction of Fetlock Breakdown Injuries in Racehorses (2 year award) | $99,180 |
| Dr. Van de Walle | Microencapsulate stem Cells to Promote Wound Healing (2-Year award) | $53,140 |

Progress and final completed in 2017

- Dr. Doug Antczak's project entitled "Cellular Immunity to Equine Herpesvirus Type 1 (EHV-1) received a no cost extension through 6/30/18. A final report will be included in next year's annual report).
- Dr. Norm Ducharme's project entitled "Two-Day Tie-Back (Injection Laryngoplasty); Proof of Principle" received a no cost extension. A final report will be included in next year's annual report.

- Dr. Lisa Fortier's project entitled, "Macrophage Regulation Mesenchymal Stem Cell Function" received a no cost extension through. A final report is included herein
- Dr. Hussni Mohammed's project entitled, "Factors Associated with Musculoskeletal Injuries & Catastrophic Events in Racing Horses" received a no cost extension. A final report will be included in next year's annual report.
- Dr. Alan Nixon's project entitled, "Arthritis Control through Dual Axis Lubricin Over-Expression and Catabolic Cytokine" received a no cost extension. A final report will be included in next year's annual report.
- Dr. Alan Nixon's project entitled, "Enhanced Breakdown Screening in Thoroughbred Racehorses through Multimodal Imaging and Serum Biomarker Combinations" received a no cost extension. A final report will be included in the annual report next year.
- Dr. Tracy Stokol's project entitled, Platelets are a Trojan horse that Deliver Equine Herpes Virus to Endothelial Cells" received a no cost extension. A final report is attached herein.

## FURTHER SECURED FUNDING FROM ZWEIG AWARDS IN 2017

The Incentive Program enables the Fund to leverage its investment in Zweig-sponsored research by encouraging Veterinary College faculty to seek either additional or supplementary monies from external sponsors that base their award decisions on a process that involves informed scientific review. The external grant must be closely related to a Zweig project. Eligible sponsors include, but are not limited to, the Grayson Foundation, the NIH, the NSF, and the USDA's National Research Initiative. Recipients provide an annual report on the use of these funds.

*The following external grant award resulted from Zweig funding:*

| ***Principal Investigator*** | ***External Award*** | ***Sponsor*** | ***Project Period*** | ***Awarded Amount*** | ***Incentive Award*** |
|---|---|---|---|---|---|
| Dr. Douglas Antczak | Cytotoxic T-Cell Immunity to Equine Herpes Virus Type 1 | Grayson-Jockey Club | 1/1/17-12/31/18 | $124,746 | $5,000 |
| Dr. Lisa Fortier<br>J. Cassano | Macrophage Regulation of Mesenchymal Stem Cell Function in Tissue Regeneration | Morris Animal Foundation | 3/1/16 - 7/1/16 | $81,334 | $5,000 |
| Dr. Norm Ducharme | Tyro-Hyoid Muscle Training to Treat DDSP | Grayson Jockey Club | 4/1/16 - 12/31/18 | $217,728 | $5,000 |
| Dr. Tracy Stokol | Anticoagulants as Thromboprophylaxis for EHV-I | Grayson jockey Club | 4/1/17-9/30/18 | $116,818 | $5,000 |

# PUBLICATIONS

Can quantitative computed tomography detect bone morphological changes associated with catastrophic proximal sesamoid bone fracture in Thoroughbred racehorses?
**Cresswell E, McDonough S, Palmer S, Hernandez C, Reesink, H**
https://www.ncbi.nlm.nih.gov/pubmed/29758110

Effect of a Histone Demethylase Inhibitor on Equine Herpesvirus-1 Activity *in vitro*
**Tallmadge R, Zygelyte E, van de Walle G, Kristie TM, Felippe MJB**
https://www.frontiersin.org/articles/10.3389/fvets.2018.00034/full

Unfractionated and Low-Molecular-Weight Heparin and the Phosphodiesterase Inhibitors, IBMX and Cilostazol, Block Ex Vivo Equid Herpesvirus Type-1-Induced Platelet Activation
**Stokol T, Serpa P, Zahid M, Brooks M.**
https://www.ncbi.nlm.nih.gov/pmc/articles/PMC5112437/

Morphological changes associated with catastrophic proximal sesamoid bone fracture in Thoroughbred racehorses?
**Cresswell E, Mcdonough S, Palmer S, Hernandez C, Reesink H**
https://onlinelibrary.wiley.com/doi/abs/10.1111/evj.12965

Reduction of Thoroughbred racing fatalities at New York Racing Association racetracks using a multi-disciplinary mortality review process
**Palmer SE, McDonough SP, Mohammed HO**
http://journals.sagepub.com/doi/full/10.1177/1040638717713051

Antigenicity of mesenchymal stem cells in an inflamed joint environment
**Hill J, Cassano J, Goodale M, Fortier L**
https://avmajournals.avma.org/doi/10.2460/ajvr.78.7.867

Peptide-binding motifs of two common equine class I MHC molecules in Thoroughbred horses
**Bergmann T, Lindvall M, Moore E, Moore E, Sidney J, Miller D, Tallmadge R, Myers P, Malaker S, Shabanowitz J, Osterrieder N, Peters B, Hunt D, Antczak D, Sette A.**
https://link.springer.com/article/10.1007%2Fs00251-017-0978-6

Lubricin/proteoglycan 4 increases in both experimental and naturally occurring equine osteoarthritis
**Reesink H, Watts A, Mohammed H, Jay G, Nixon A**
https://www.ncbi.nlm.nih.gov/pubmed/27498214

Secreted factors from equine mesenchymal stromal cells diminish the effects of TGF-β1 on equine dermal fibroblasts and alter the phenotype of dermal fibroblasts isolated from cutaneous fibro proliferative wounds
**Harman R, Ivanna BS, Bihun V, Van de Walle, G**
https://onlinelibrary.wiley.com/doi/abs/10.1111/wrr.12515

First demonstration of equid gammaherpesviruses within the gastric mucosal epithelium of horse
**Pennington M, Cossic B, Perkins G, Duffy C, Duhamel G, Van de Walle G**.
https://www.sciencedirect.com/science/article/pii/S0168170217305099?via%3Dihub

# Gerlinde Van de Walle, DVM, Ph.D.
# Harry M. Zweig Assistant Professor in Equine Health 2017-2019


*Gerlinde Van de Walle and Farrah at the Baker Institute's McConville Barn*

At the November 17, 2016 Harry M. Zweig Memorial Fund for Equine Research annual meeting, in recognition of her success in research related to the health of horses, Dr. Gerlinde Van de Walle was named the third Harry M. Zweig Assistant Professor in Equine Health at the College of Veterinary Medicine, Cornell University. The three-year assistant professorship is effective January 1, 2017 through December 31, 2019.

***The intent of the Harry M. Zweig Assistant Professorship in Equine Health is to support a junior faculty member showing promise and productivity in promoting equine health. The honor would be granted for a period of up to three years.***

Dr. Van de Walle's equine research program focuses on infectious diseases and wound healing, work that has been funded by the Zweig Memorial Fund, Boehringer Ingelheim, the National Institutes of Health, the United States Department of Agriculture, the Cornell Stem Cell Program, and the American Quarter Horse Foundation. The results promise to not only help improve the health and wellbeing of horses, but of humans and other animal species as well.

In her infectious disease work, Dr. Van de Walle examines the possible roles of viruses in two common conditions in horses: ulcers and hepatitis. Gastric ulcers are particularly widespread among racing horses, and it has long been suspected this is due to the stressful experiences of training, travel, and the track. Dr. Van de Walle's work has uncovered evidence that an infectious cause may be to blame for at least some of these cases, a possibility that offers new treatment options or even a cure, says Dr. Van de Walle. "If we can link ulcers to an infectious cause we can treat these affected horses and heal them," she says.

In the case of hepatitis, Dr. Van de Walle investigates the ways in which nonprimate hepacivirus (NPHV) causes liver inflammation. Given the close relationship between NPHV and hepatitis C virus, which causes disease in humans, these studies could lead to benefits for both species.

When it comes to wounds, horses often heal quite slowly, particularly on their lower legs. Dr. Van de Walle has been testing stem cells to determine whether they could aid the wound healing process in tissue samples in the lab. Their results show that the substances secreted by stem cells can prevent thick, lumpy scarring (called proud flesh) from forming and also reduce the severity of existing scars. Dr. Van de Walle plans to carry the work forward to testing in horses, and to examine how those treatments may be used in wound management for other animal species, including humans.

The Zweig Assistant Professorship presents her and her research team of graduate students and postdoctoral associates with the opportunity to have a greater impact on the health of these animals, which have always had a unique and powerful relationship with humans. *"The support from the Zweig Memorial Fund is a great foundation; it puts us in a strong position to launch our research into the broader scientific community," says Dr. Van de Walle.*

# CORNELL CLINICAL FELLOW IN EQUINE HEALTH

At the 2007 Annual meeting, the Harry M. Zweig Committee approved the allocation of funds to help support a Cornell Clinical Fellow in Equine Health. Dr. Sophy Jesty was selected as Cornell's first Clinical Fellow, followed by Dr. Sarah Pownder, and more recently another individual has been identified as a Clinical Fellow, Dr. Joy Thomlinson and supported in part by Zweig funds, and all have been highly successful. Cornell's College of Veterinary Medicine's two-year Clinical Fellows Program is the first in the country to address a growing shortage of academic veterinarians who conduct research on animal diseases and basic biology.

The program is designed to help students meet the financial and time demands of qualifying for a position in veterinary academic medicine, which has traditionally required students to complete an M.S. or Ph.D. after they finish their doctorate in veterinary medicine (DVM). The two-year program, available to veterinarians who have completed a three-year residency, offers an annual salary of $65,000 plus benefits and an additional $15,000 per year to fund a research project.

Dr. Joy Tomlinson research associate in the lab of Dr. Gerlinde Van de Walle, the 2017 Harry M. Zweig Assistant Professor in Equine Health, is working with colleagues on research that may lead to better treatment and screening for Theiler's disease in horses, and a system for developing a hepatitis C vaccine for humans. https://ecommons.cornell.edu/handle/1813/51542

Patent updates (on-going)

During 2016 patent 9,366,671 "Novel Immunogenic Proteins of Leptospira" patent was issued to Dr. Y-F Chang on 6/14/16 (US). Including; UK (EP244728); Germany (60347502.7); Europe (2447278), Canada (2820949), Germany (60347502.7), France (EP2447278); US (93,366,671) Canada (9,176,133) 11/3/15, and United Kingdom (EP2447278 on 4/8/15. Applicant: College of Veterinary Medicine, Cornell University, Ithaca, NY (2016)

During 2017 patent application No. 15/429802 (US); submitted 2/10/17; Therapeutic Targeting of Mitochondria to Prevent Osteoarthritis" Inventors: M. Delco, L. Fortier. Applicant: College of Veterinary Medicine, Cornell University, Ithaca, NY (2017)

Converted patent application No. 62/216,010 (US); submitted 9/9/2015; "Multiplexing and Enhancing Serological Assays via Cytokine Fusion Proteins." Inventor: B. Wagner; Applicant: College of Veterinary Medicine, Cornell University, Ithaca, NY (2015)

Provisional patent application No. 61/903,619 (US); submitted 11/13/2013; "Stimulation of Neonatal Immunity". Inventors: B. Wagner, G. Perkins; Applicant: College of Veterinary Medicine, Cornell University, Ithaca, NY (2013).

Patent application No. 6465-02PC (submitted 9/6/13) -- "A Method to Induce Protection and Immune Alertness in Neonates". Inventors: B. Wagner, G. Perkins, College of Veterinary Medicine, Cornell University, Ithaca, NY (2014) Patent PCT/US14/651 was issued to Dr. Wagner on November 12, 2014.

Zweig News Capsules

There were two issues of the Zweig News Capsule published in 2017 (64 & 65). Copies of these issues can be found in Appendix (E).

All Zweig News Capsules can be found at the Zweig Website at:
http://www.vet.cornell.edu/zweig/

## SUMMARY OF EXPENDITURES

The 2017 Summary of Allocations was presented and approved at the Zweig Committee Annual Meeting in November 2017 (Appendix B).

## 2018 ZWEIG PROGRAM

Six (6) projects were approved for funding, from a roster of thirteen (13) applications, at the Harry M. Zweig Memorial Fund annual November 2017 meeting. The list of projects funded for 2018 are shown in (Appendix D).

# APPENDIX A
# Progress & Final Reports Resulting from **2017** Funding

| | |
|---|---|
| Dr. Antczak | Cellular Immunity to Equine Herpesvirus Type 1 (EHV-1) |
| Dr. Antczak | Functional Gene Annotation in the Horse |
| Dr. Cheetham | Regenerative Approach to Recurrent laryngeal Neuropathy |
| Dr. Ducharme | Two-Day Tie-Back (injection Laryngoplasty); Proof of Principle |
| Dr. Fortier | Does Intra-Articular Stifle Anesthesia Alleviate Lower Limp Lameness? |
| Dr. Fortier | Macrophage Regulation of Mesenchymal Stem Cell Function in tissue Regeneration |
| Dr. Fubini | The Relationship between Obesity and Post-Operative Incisional Infections following Abdominal surgery in the Horse |
| Dr. Mohammed | Factors Associated with Musculoskeletal Injuries and Catastrophic Events in Racing Horses |
| Dr. Reesink | Quantitative Computed Tomography & Bone Quality Assessment for the Production of Fetlock Breakdown Injuries in Racehorses |
| Dr. Stokol | Platelets are a Trojan Horse that Deliver Equine Herpes Virus to Endothelial Cells |
| Dr. Van de Walle | Microencapsulate Stem Cells to Promote Wound Healing |
| Dr. Van de Walle | Defining the Relationship between Equine Herpesvirus & Development of Equine Gastric Ulcer Syndrome (EGUS) |
| Dr. Wagner | Effects of Equine Herpesvirus (EHV) Vaccination Frequency on Host Immunity to EHV-1 |
| | |

# *Harry M. Zweig Memorial Fund for Equine Research*
# *2017 Annual Progress Report for Zweig Committee*

| | |
|---|---|
| P.I.: | Dr. Douglas Antczak |
| Title: | Functional gene annotation in the horse |
| Project Period: | 1/1/17-12/31/18 |
| Reporting Period | 1/1/17-12/31/17 |

## ***TITLE:*** *Functional gene annotation in the horse*
## ***PRINCIPAL INVESTIGATOR(S):*** *Douglas Antczak*

This project is designed to identify regulatory elements in the equine genome using an exciting new technique developed by Dr. Charles Danko. Chromatin Immunoprecipitation and Sequencing (ChRO-seq) is an adaptation of Global Run-on and Sequencing assays developed here at Cornell. We are testing horse samples using CH-RO-seq and comparing the data with results generated by other members of the Equine Genome research consortium who are using older ENCODE methods under the international Functional Annotation of Animal Genomes (FAANG) collaboration.

*Specific Aims:* The aims shown are essentially the same as originally submitted. We have made a few small changes (see strikethroughs below). These will be explained in the text in the Plans section.

**Aim 1: To identify equine placenta-specific functional elements.** We would define and compare tissue-specific regulatory elements in equine invasive (chorionic girdle) and non-invasive (allantochorion) trophoblast. We would analyze this data in the context of gene expression patterns that we have established previously in the Antczak lab.

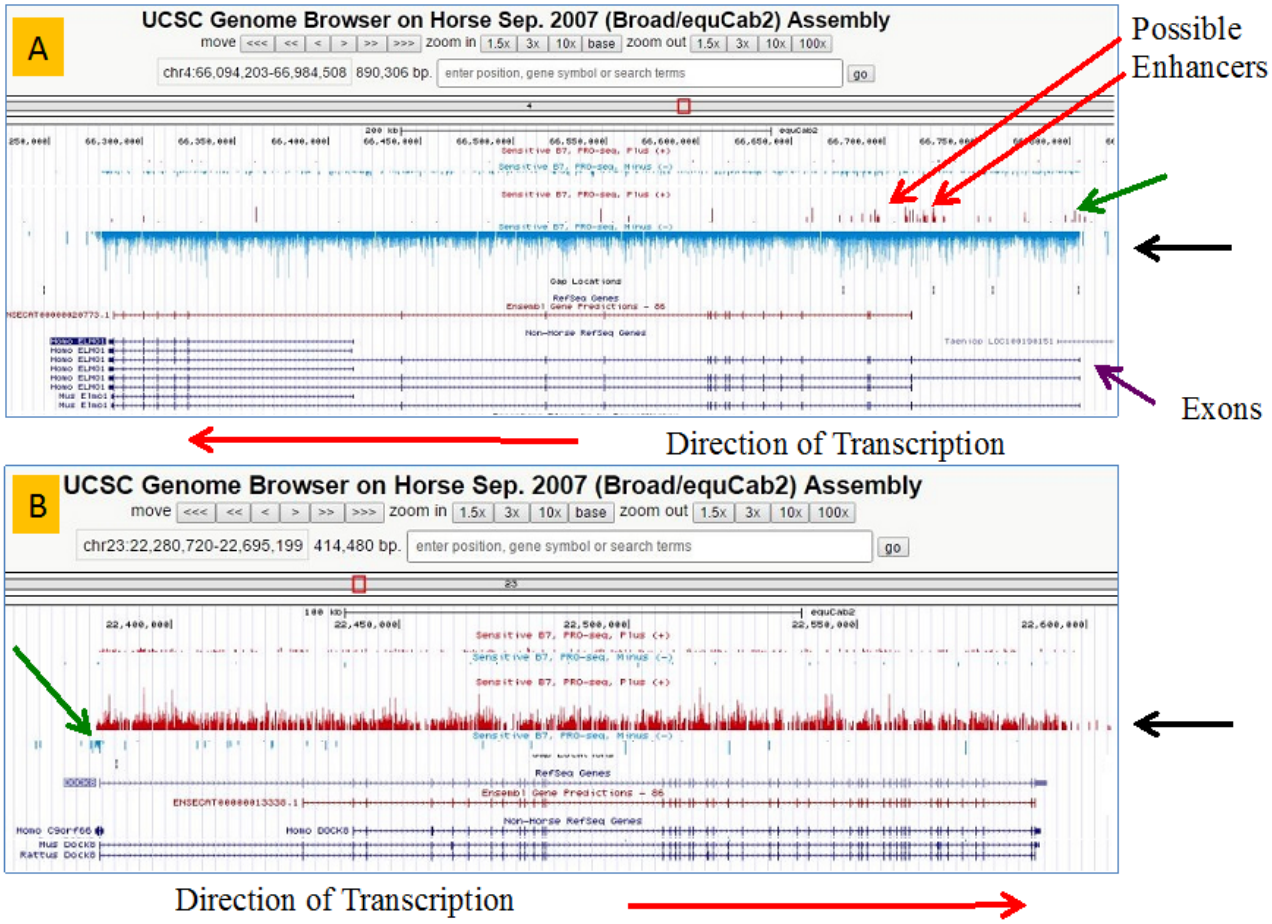
**Aim 2. To identify functional regulatory elements in horse CD4+ T-cells.** We would characterize functional regulatory elements from equine CD4+ T-cells isolated from peripheral blood using horse-specific monoclonal antibodies and FACS sorting magnetic bead separation techniques and compare this data with results from studies of human CD4+ T-cells by Co-PI Danko.

**Aim 3**. **To directly compare the Cornell GRO-seq / bioinformatic approach with established ENCODE techniques.** We would characterize regulatory elements in samples of equine liver unseparated Peripheral Blood Lymphocytes from two horses under study for the equine ENCODE project (called FAANG) at the University of California at Davis.

Our experiments in this project are on track for successful completion. We have produced exciting new data from horse placental samples tested using the ChRO-seq technique. An example is shown in Fig. 1, which depicts results for two genes, ELMO1 and its interaction partner DOCK 8. In previous work we had identified ELMO1 as a highly unregulated gene in horse invasive trophoblast (see Table 1). Our ChRO-seq results are consistent with this earlier work and validate the ChRO-seq approach.

**Table 1. Summary of invading tumor-associated genes expressed in horse invasive trophoblast:** comparison of fold expression increase compared to non-invasive trophoblast is shown. Mean of results from three independent conceptuses. Data obtained by Expression Array hybridization – Antczak lab unpublished.

| Gene | Function | Expression Array results | Q-RT-PCR validation |
|---|---|---|---|
| ADAMTS1 | Cell adhesion | 3x | 6x |
| CEACAM1 | Cell adhesion | 6x | 4x |
| ELMO1 | Effector of cell migration | 61x | 53x |
| LAVERIN | Cell invasion | 48x | 108x |
| PECAM1 | Cell adhesion, Proteolysis, Angiogenesis | 21x | 172x |




We have also applied to external funding agencies for grants to expand this study. One proposal (Kleberg Foundation) was not funded; the second (USDA) is under review.

**Fig. 1. ChRO-seq results for two genes from initial test of invasive and non-invasive horse trophoblast.** A. ELMO1 (from Table 1) B. Dock 8, pathway partner to ELMO1. Red arrows below figures show direction of transcription. Black arrows indicate gene transcription activity in invasive trophoblast. Thin blue (A) or red (B) lines above show non-invasive trophoblast, where no transcriptional activity is detected. Green arrows show polymerase pause sites, indicating a gene under active transcription. Location of possible enhancer elements in ELMO1 introns is shown. Purple arrow points to small vertical lines that indicate exons.

Our tests of the ChRO-seq system hold promise for accelerating the pace of characterization of regulatory elements in the horse genome. The results thus far have met our high expectations. Dr. Danko presented some of our results in an invited Keynote Address at the International Society for Animal Genetics conference in Ireland in July 2017. His presentation was very well received.

In the remainder of 2017 and through 2018 we will test the remaining samples from this project. Based on our preliminary studies to enrich horse PBL for CD4+ T cells, we have decided to use FACS sorting with a positive gate using anti-horse CD4 and CD3 monoclonal antibodies and a negative gate using anti-horse CD8. We have secured directly labeled antibodies with 3 different conjugates for this enrichment. The FAANG horse PBMC samples from our collaborators in California were found to be of low quality, so we decided to switch to liver. The liver samples have already been used in ChIP-seq and methylation sequencing by other members of the FAANG group, and so should provide a good comparison with the ChRO-seq system. We will also test additional samples of horse invasive and non-invasive trophoblast placental samples, as described in the initial proposal.

*Harry M. Zweig Memorial Fund for Equine Research*
*2017 Annual Progress Report for Zweig Committee*

| | |
|---|---|
| P.I.: | Dr. Jonathan Cheetham |
| Title: | Regenerative approach to recurrent laryngeal neuropathy |
| Project Period: | 1/1/17-12/31/18 |
| Reporting Period | 1/1/17-12/31/17 |

***TITLE:*** *Regenerative approach to recurrent laryngeal neuropathy*
***PRINCIPAL INVESTIGATOR(S):*** *Jonathan Cheetham*

**Aim 1. Determine the optimal *in vivo* dose of IL10 released from an agarose hydrogel.** We will achieve this aim by loading an inert agarose hydrogel with IL10 at three doses (0.5, 1 and 3 µg IL10/ml 0.7% agarose) which bracket the anticipated optimal dose. A non-critical (8mm) gap will be created in in the sciatic nerve of rats expressing green florescent protein in their axons (Thy1_GFP). Rats will be euthanized at day 21 and axon extension determined. Negative controls will be 0.7% agarose only and an empty conduit. We will then repeat this experiment with a delayed TIB-CP graft and a 10mm conduit to determine the optimal dose in delayed repair.

**Aim 2. Determination of effects of immunomodulation on macrophage phenotype, peak tetanic force and neuromuscular junction formation.** Using the same experimental approach and groups as Aim 1, the regenerative bridge within the conduit will be isolated at day 7, the peak of macrophage infiltration, and immunolabeled using direct immunofluorescence to determine the phenotype of the host immune cells which participate in the immune response. Ten weeks after immediate and delayed nerve graft we will determine peak tetanic force, muscle fiber diameter and the number of reinnervated neuromuscular junctions. Neuromuscular junction formation is important as it closely correlated to force generation of any given muscle, including the CAD muscle in the equine larynx. Force generation by the CAD muscle is important to maintain a patent airway

We have completed Aim 1 and demonstrated that immunomodulation with exogenous IL10 produces a profound dose-dependent improvement in motor neuron regeneration (figure 1).

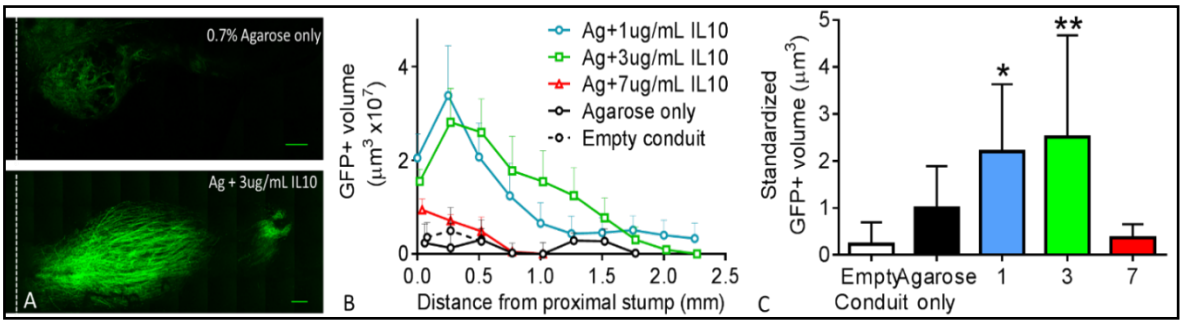



**Figure 1**. (A) Representative regenerative bridges from transgenic rats expressing green fluorescent protein (GFP) under the Thy-1 promotor in their peripheral axons. Flattened z-stack obtained using confocal microscopy 21 days after placement of a 10mm conduit after sciatic transection. Conduit filled with agarose only or agarose or 3 ug/mL IL10. White dotted line denotes proximal stump. (B) Dose dependent effect of addition of exogenous IL10 at three doses (n=6-8/group) showing volume of GFP+ axons extending from proximal stump. GFP expression depicted as volume of GFP (y axis) per 250µm region of interest extending from proximal stump (x axis) for each group. (C) Total volume of GFP+ axons within regenerative bridge. Data are mean and standard error (se) ** denotes significantly different from agarose only control (Dunnet's test, * $p<0.05$, ** $p<0.01$ ).

We have also confirmed this result in mice (Figure 2) and begun to investigate the mechanism of this effects. The data generate in figure two was supported by additional laboratory funds outside the Zweig support. We felt it was important to verify this result in another species as we determine the mechanism of this effect.

**Figure 2**. Immunomodulation with exogenous IL10 produces a profound dose-dependent improvement in motor neuron regeneration. The sciatic nerve was transected in WT mice and repaired immediately with a 5mm conduit filled with 0.7% agarose and loaded with IL10 at 0.5,1,3 or 15 µg/mL. Repair was assessed using retroDISCO at eight weeks, n=6 / group [39]

We have also shown that the addition of exogenous IL10 promote the formation of neuromuscular junctions 7 weeks after repair (figure 3).

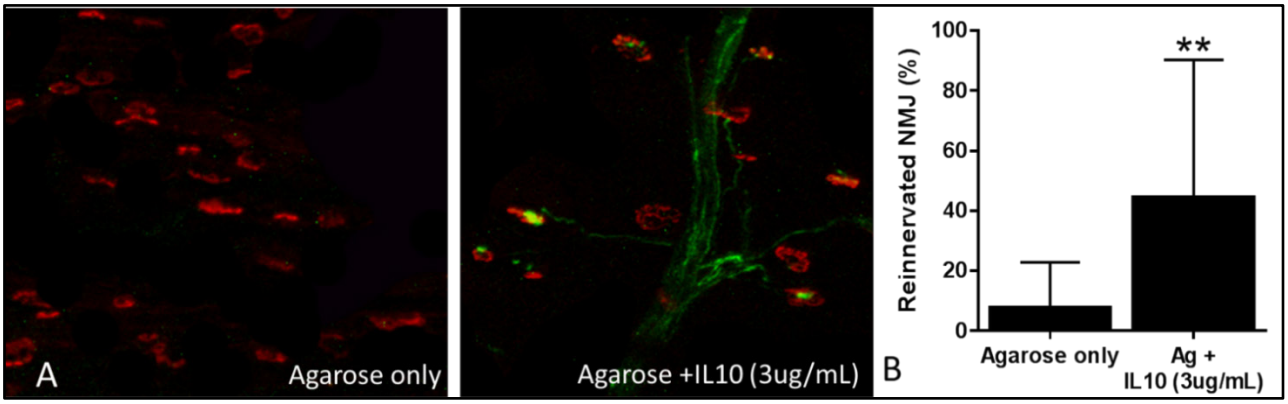



**Figure 3**. Modulation of macrophage response using IL10 promotes formation of neuromuscular junctions (NMJ) in the extensor digitorum longus (EDL) muscle of Thy1-GFP rats 8 weeks after sciatic nerve transection and conduit repair. A. Representative images of terminal axons (green) innervating post synaptic Acetylcholine receptors (AChRs), labelled with rhodamine-α-bungarotoxin (red) conjugated to Alexa Fluor-594 [47]. B. Proportion of innervated NMJ standardized to contralateral control.

We have now begun obtaining data on peak tetanic force as described in Aim 2. We are also using kinematic gait analysis and walking track analysis to determine the effect of exogenous IL10 on the time to recovery after nerve injury. This is an additional outcome measure to determine function and allow observation of improvement over time.

Dr Elaine Claffey, a surgical resident and trainee on the award presented these results in a poster at the recent American College of Veterinary Surgeons meeting and won first place (Immunomodulation for Nerve Injury. Elaine F. Claffey, Tim Moore, Jonathan Cheetham).

We have used these data to support two submissions to the NIH in October of 2017 (RO1 - Manipulating Macrophage Phenotype to Accelerate Recurrent Laryngeal Nerve Repair, Cheetham PI; Innovator Award- Macrophages: Conductors of the Orchestra for Nerve Repair, Cheetham PI).

***Harry M. Zweig Memorial Fund for Equine Research***
***2017 Annual Progress Report for Zweig Committee***

| | |
|---|---|
| P.I.: | Dr. Norm Ducharme |
| Title: | Two-Day Tie Back (Injection Laryngoplasty):Proof of Principle |
| Project Period: | 1/1/17-12/31/18 |
| Reporting Period | 1/1/17-12/31/17 |

Dr. Ducharme was granted a no cost extension through 12/31/18.

## ***TITLE:*** Two-day Tie-back (Injection Laryngoplasty): Proof of Principle

***PRINCIPAL INVESTIGATOR(S):*** *Norm Ducharme*

**BACKGROUND:** The research entitled "Two-day Tie-back (Injection Laryngoplasty): Proof of Principle" aims to determine a new treatment option for horses affected by laryngeal hemiplegia as a consequence to recurrent laryngeal neuropathy (RLN). Laryngeal collapse associated with RLN is a common cause of upper airway obstruction yielding abnormal respiratory noise and poor performance. Most racehorses affected by this pathology, are treated with laryngoplasty in combination with ventriculocordectomy. Laryngoplasty works by suspending the arytenoid in an open position by fastening the muscular process of the arytenoid cartilage to the caudal cricoid cartilage with sutures, restoring airway mechanics toward normal[1-6]. Ventriculocordectomy, removing the vocal cord and the ventricule, reduces or eliminates abnormal respiratory noise and obstruction produced by their fluttering during exercise[7]. Main drawbacks of these procedures are length of post-operative recovery time, laryngoplasty failure rate up to 20-30% of treated horses[2,3,8-10], arytenoid chondritis[11] and high prevalence of tracheal aspiration leading to cough, recurrent pneumonia and post-operative DDSP[12-14].

Vocal cord augmentation in humans is used to restore voice and treat glottal insufficiency[15-18]. We used vocal cord injection in horses that aspirate after laryngoplasty, to create a barrier thus preventing food contamination into the trachea (Ducharme et al, unpublished studies). Polymethyl methacrylate (PMMA) bone cement was selected as injectable material because it is a solid, durable, inert substance that rarely results in inflammatory tissue reaction. Based on these clinical experiences we hypothesized that vocal cord injection with PMMA would allow support of the arytenoid cartilage and vocal cord against the collapsing pressure during inhalation and swallowing, creating an alternative approach to the traditional laryngoplasty as treatment for laryngeal hemiplegia. A ventral support of the arytenoid cartilage instead of suspension by the muscular process (laryngoplasty) would avoid the failures associated with sutures pulling through cartilages.

**EXPERIMENTAL DESIGN:** This study was divided into 3 phases. Phase 1 and 2 are conducted to: a) develop on cadaver specimens the most effective vocal cord injection technique, b) determine the most appropriate PMMA preparation method, and c) to measure in-vitro performance (i.e., respiratory mechanics) of the "injection laryngoplasty". Phase 3 would evaluate the in-vivo injection safety and efficacy.

The three specific aims to be fulfilled on Phase 1 and 2 were reclassified as:

- Aim 1 (Phase 1): optimize surgical technique of injection laryngoplasty.
- Aim 2 (Phase 2): Measure PMMA injection and setting times, and peak temperature at various dilutions used for delivering the substance to the ventral aspect of the arytenoid.
- Aim 3 (Phase 2): Measurement of trans-laryngeal pressures, airflow, and arytenoid angles in 20 cadaver larynges under control, standard "suspension laryngoplasty" at 80% glottis opening, and "injection laryngoplasty" also at 80% glottis opening.

The project was delayed for 5 months because of import permit issues related to purchasing cadavers larynges from Canada. This has been resolved and Aim 1 and Aim 2 steps have been completed and we are currently working on Aim 3.

**COMPLETED RESULTS TO DATE (Phase 1 and 2)**

*AIM 1*: Six larynges were used to determine the most effective injection site and instruments. Different insertion approaches along the cricothyroid membrane were evaluated, as well as different leverage points ventrally to the arytenoid cartilages. A blunt insertion tool resulted a better option over sharp instruments to maintain the mucosal integrity. Different sizes were assessed, and finally a 6G firm plastic tube resulted in the least traumatic and most effective in delivering the PMMA.

A more caudal insertion approach was found to allowed sliding the injection tool within the vocal cord and creating a cement column just ventrally to the vocal process of the arytenoid. This resulted in a wide vocal cord bulging medially (i.e. potential obstruction) reducing the glottis area, for this reason a more rostral approach was attempted. Inserting the instrument just caudally to the thyroid notch allowed us to slide the instrument along the ventricular fold, reaching the ventral corniculate process. Injection of the PMMA along the ventricular fold allowed a more lateral distribution of the bone cement column, preventing obstructive bulging into the glottis opening. The last technique proved to be more functional with a lesser impact on the laryngeal lumen and was chosen for the upcoming experiments.

*AIM 2*: Determine the setting time, working time, and peak temperature of varying concentrations of powder to reconstitute PMMA has been considered a necessary step of this study because the chemical reaction occurring from mixing the polymethyl methacrylate polymer with the liquid monomer is quite exothermic, yielding temperatures reportedly greater than 70 degrees Celsius [20-21]. Additionally, bone cement is traditionally formulated for use in hip replacements to fasten the implant to the bone. It is used while it is in a dough-like state, while its use in the larynx requires a viscous state to facilitate injection through a needle. It was postulated that using a lower concentration of powder with the same volume of liquid would yield a substrate that could be injected and offer a longer working period before fully setting.
Stryker Surgical Simplex® P polymethyl methacrylate radiopaque bone cement was used. It is packaged into two components: a 20 ml liquid ampule (97.4% v/v methyl methacrylate monomer, 2.6% v/v N. N-dimethyl-p-toluidine, and 75+/- 15 ppm Hydroquinone) and a 40 gram packet of fine white powder (15% w/w Polymethyl methacrylate, 75% w/w Methyl methacrylate – styrene – copolymer, and 10% w/w Barium Sulfate U.P.S.). Four different concentrations of powder were investigated: 5, 6.66, 8.32, and 10 grams of the powder polymer, mixed with a volume of 10 ml liquid, which yields a final paste volume of 12 ml.

Temperature data was collected using a 4-channel Omega data logger thermometer with 20-gauge TT insulated type K-wire thermocouples. The designated volume of bone cement was weighed and hand mixed using a bowl exposed to ambient atmosphere. Once the powder was fully dissolved, the formulation was transferred to syringes with a total volume of 4 ml in each of 3 syringes. The K-wire thermocouples were inserted into the injection port of the syringes for temperature recording from the center of the volume of cement. Data acquisition was started at 2 minutes after initial combination to allow for complete mixing and transferring of bone cement. Temperature readings were recorded at 5 second intervals for approximately 30 minutes, or until the bone cement returned to room temperature after reaching the peak. The fourth probe was used to measure ambient temperature. The readings from the three syringes were averaged to create a single value per experiment. Peak temperature and set time were determined in accordance with the specifications detailed in the resin cements International Standard document ISO 5833.

The force data was collected using the same mixing methods. Once the bone cement was fully mixed, one aliquot was transferred to two 6 ml syringes attached to 18-gauge 3.5 inch spinal needles. A Torbal FB200 force meter was used to measure the force required to push 0.2 ml through the spinal needle. Force measurements were collected in Newtons from each syringe in tandem at 1 minute intervals until the bone cement could no longer be expressed through the needle. Each experiment was run concurrently with a temperature data collection experiment providing the ambient temperature for both data sets. The results from each syringe were averaged; each experiment was performed twice.

*Statistics*

A one way ANOVA Kruzkal-Wallis test for non-parametric data with post hoc Tukey application was used to compare differences between the selected data points ($P <= 0.05$). A linear regression analysis applied to the force data controlling for time was created with a 95% confidence interval. Data points used for statistical analysis included peak temperature, setting time (measured by the time at which the temperature is midway between ambient and peak temperature), injecting time (time able to push 0.2 ml through the syringe) and force measurements.

*Results*

In accordance with ISO 5833, each experiment was run at 23° C +/- 1 ambient temperature, and between two experiments a peak temperature within a 10°C range and a setting time within 1 minute are required; if there is a discrepancy, the experiment must be run in duplicate. Henceforth, each concentration was run 4 times except from 8.32g of powder, which only needed to be run twice. The results are summarized in Table 1.

The average peak temperatures did not increase on a linear scale and were 57.4 ±2.5, 83.5 ±4, 99.2±5.1 and 98.5±4.3°C respectively for 5, 6.66, 8.32, and 10 g of powder. There was a significant difference in temperature between all concentrations except for 8.32g of powder and 10 g of powder ($P \leq 0.05$). The average setting times were 26.1 ±3.6, 20.4±3.3, 17.1±0.9, and 14.0±1.9 minutes for the same progression of concentrations. Based on the graphical information, the setting times decreased as the concentration increased. The setting time for 5g of powder was not significantly different from 6.66 g ($P>0.05$). There was no significant difference in setting times between 6.66 and 8.32g of powder and between 8.32 and 10 g of powder however, there was a significant difference between 6.66g and 10 g of powder setting times.

The injecting times decreased as concentrations increased and were 16.5±0.7, 10.5±3.5, 6.5±0.7, and 4±1.4 minutes respectively for 5, 6.66, 8.32, and 10 g of powder. There was no significant difference between 5g and 6 g of powder. No significant difference between 6.66, 8.32, or 10g of powder. See table 1 for complete results.

The linear regression analysis yielded a significant difference for all the concentrations between the intercepts calculated from the force data acquired at 1 minute intervals.

**Table 1** concentration of powder used and resulting values of the properties of the cement

| **Composition of powder (g)** | Peak Temperature (C°) | Setting Time (min) | Injection Time (min) |
|---|---|---|---|
| 5 | 60.4 [a] | 33.2 [a] | 17 [a] |
| 5 | 57.2 [a] | 30.9 [a] | 16 [a] |
| 5 | 54.5 [a] | 27.5 [a] | |
| 5 | 57.6 [a] | 28.7 [a] | |
| 6.66 | 77.7 [b,c] | 18.5 [a,b] | 8 [a,b] |
| 6.66 | 84.3 [b,c] | 26.8 [a,b] | 13 [a,b] |
| 6.66 | 86.6 [b,c] | 23 [a,b] | |
| 6.66 | 85.5 [b,c] | 24.8 [a,b] | |
| 8.32 | 102.8 [b,c,d] | 19.5 [b,c] | 7 [b,c] |
| 8.32 | 95.6 [b,c,d] | 18.4 [b,c] | 6 [b,c] |
| 10 | 92.6 [c,d] | 12.9 [c] | 3 [b,c] |
| 10 | 98.1 [c,d] | 15.7 [c] | 5 [b,c] |
| 10 | 102.6 [c,d] | 15.7 [c] | |
| 10 | 100.7 [c,d] | 18.3 [c] | |

**[a,b,c] same superscript are not significantly different from each other**

*CONCLUSIONS*: Based on these results, the concentration obtained using 8.32g of powder in 10 ml solution was chosen for the future experiments; despite the high peak temperature (which will require cooling during procedure), this concentration allows a faster setting of the bone cement, a fundamental factor to consider for a better outcome of the procedure that could be performed in sedated horses.

## EXPERIMENT IN PROGRESS (Phase 2- AIM 3)

The goal of this phase was an in-vitro comparison of respiratory mechanics between "injection laryngoplasty" and traditional laryngoplasty ("suspension laryngoplasty") performances in maintaining the arytenoid in abducted position against airflow and negative inspiratory pressures.
We used larynges stored at -20°C. The in-vitro tests were performed on 3 larynges.

Each experiment was conducted using a flow chamber system[19] that comprised a vacuum system (16-gallon vacuum cleaner) attached in parallel with a rheostat used to generate negative pressure and so produce an inspiratory flow; a PVC pipe (50mm internal diameter) containing a cycling valve (PVC

disk, 1mm radial clearance) contacting valve seals at the fully closed position to occlude air flow. The valve cycle at 2Hz intermittently obstructing airflow to mimic the inspiratory cycle at exercise. This PCV pipe is connected to the caudal aspect of the trachea by a clamp. Static flow, monitored through a rotameter (KDG Flowmaters, West Sussex UK) in line with the chamber, is then applied for 30 seconds, while pharyngeal pressure (Pp) and tracheal pressure (Tp) are recorded at 500Hz using custom software (Labview, National Instruments). For each condition, static flow is maintained for 30 s, the cycling valve is then activated (at 2Hz) and data recorded for an additional 30 s. In addition, digital recording of the laryngeal cartilage is taken to document the degree of laryngeal collapse.

*Cadaver larynges preparation*
The larynges were allowed to reach room temperature. Four different settings were obtained in sequential order from each larynx to simulate laryngeal function in

1. normal condition
2. left RLN
3. treated with injection laryngoplasty (treatment A)
4. treated with traditional laryngoplasty and left vocal cordectomy (treatment B, gold standard).

A prosthetic laryngoplasty suture was placed on both right and left arytenoids to obtain maximal abduction and simulate a normally functioning larynx during maximal exercise. The larynx was then placed into the flow chamber, and data recorded to represent a "normal condition".
The larynx then underwent the removal of the left prosthetic suture to mimic a larynx affected by RLN with left laryngeal paralysis (laryngeal function grade IV at rest, C at exercise). The flow chamber test was repeated and data recorded as "RLN larynx".

Each larynx then underwent treatment A first, followed by treatment B:

*Treatment A* ("Injection laryngoplasty" of left hemilarynx): a 6G (4.1mm diameter) plastic insertion tool was inserted through the crico-thyroid membrane and slid under the mucosa along the left ventricular fold. After reaching the ventral angle of the corniculate process, while keeping the arytenoid in 80% abduction (in reference to the right side kept in maximal abduction), PMMA was injected and the insertion tool slowly retracted. The abduction was maintained until the PMMA hardened. Digital recording for arytenoid angle determination were obtained at the end of injection and while under airflow pressure in the chamber. Data were recorded as "injection laryngoplasty".

*Treatment B* ("traditional laryngoplasty"): A prosthetic laryngoplasty suture was replaced to obtain 80% abduction of the left arytenoid using the previously described technique[19] and a left ventriculocordectomy (VC) was performed. The flow chambers tests were repeated and data recorded as "laryngoplasty with VC".

Preliminary data from the tests on the first 3 larynges are promising however the current sample size of the data does not allow appropriate comparison between treatment A (injection laryngoplasty) and treatment B (traditional laryngoplasty with VC) at this time.

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# *Harry M. Zweig Memorial Fund for Equine Research*
# *2017 Final Report for Zweig Committee*

| | |
|---|---|
| P.I.: | Dr. Lisa Fortier |
| Title: | Macrophage regulation of mesenchymal stem cell function in tissue regeneration |
| Project Period: | 1/1/16-12/31/17 |
| Reporting Period | 1/1/17-12/31/17 |

**Dr. Fortier received a no cost extension - final report below.**

***TITLE:*** *Macrophage regulation of mesenchymal stem cell function in tissue regeneration*

***PRINCIPAL INVESTIGATOR(S):*** *Lisa Fortier*

Aim 1: **Aim 1. Determine how M1/2 macrophages alter MSC phenotype and function.**
*Completed. Resulted in manuscripts 1 & 2 listed below.*

Aim 2: **Study the ability of inflammatory conditioning to produce a stable immunomodulatory MSC phenotype.**
*Completed. Resulted in manuscript 3 listed below.*

Resultant Cassano manuscript. Our results suggest MSCs can be activated by a variety of inflammatory stimuli, but the recipient injured tissue bed in chronic injuries may not contain sufficient inflammatory signals to activate MSC immunomodulatory function. Enhancement of MSCs immunomodulatory function through inflammatory priming prior to clinical application might improve the therapeutic effect of MSC treatments.

Resultant Hill manuscript. Results indicated that MSC exposure to pro-inflammatory cytokine IL-1β decreased MHC class II expression and antigenicity. Treatment of inflamed joints with allogeneic MSCs might not be contraindicated, but further investigation is warranted.
This was Dr. Hill's qualifying manuscript for the American College of Veterinary Surgeons Examination.

> Dr. Hill presented this study at the American College of Veterinary Surgeons in 2015 and received 2nd place in the basic science division.

Resultant Cassano manuscript. Results of this study indicate that in vitro inflammatory licensing agents enhanced the immunomodulatory ability of MSCs exposed to inflammatory macrophages, with little evidence that licensing creates a pro-inflammatory MSC secretome. Compared to licensing agents, exposure of MSCs to inflammatory macrophages inconsistently stimulated immunomodulatory function. This suggests that in vitro inflammatory licensing prior to clinical use could result in more consistent induction of immunomodulatory function, compared in vivo inflammatory licensing by the recipient environment.

The Cassano manuscripts formed the basis for Dr. Cassano's PhD thesis. The results were presented at several international conferences including the Orthopedic Research Society, International Cartilage Repair Society, Havemeyer Conference on Regenerative Medicine, and the Cornell Stem Cell Symposium, to name a few. Some aspect of the work supported by this Zweig grant is also presented when Dr. Fortier does CE courses.

# *Harry M. Zweig Memorial Fund for Equine Research*
# *2017 Annual Progress Report for Zweig Committee*

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| P.I.: | Dr. Susan Fubini |
| Title: | The Relationship between obesity and Post-Operative Incisional Infections following Abdominal Surgery in the horse |
| Project Period: | 1/1/17-12/31/18 |
| Reporting Period | 1/1/17-12/31/17 |

## ***TITLE:*** *The Relationship between obesity and Post-Operative Incisional Infections following Abdominal Surgery in the horse*

## ***PRINCIPAL INVESTIGATOR(S):*** *Susan Fubini*

Aim 1. To retrospectively evaluate if there is a relationship between body mass index (BMI) and surgical site infection (SSI) in horses undergoing ventral midline celiotomy. Records of horses undergoing ventral midline celiotomy over the last six years will be reviewed. Data retrieval including age, breed, sex, body weight, withers height, lesion, hospital stay length, incisional morbidity and survival will be conducted to review via the record and telephone interview. BMI, a marker of adiposity, will be calculated for each horse and used in statistical comparisons between horses with and without SSI.

Aim 2: To prospectively evaluate whether BMI and other morphometric clinical variables and laboratory parameters related to adiposity are predictive of SSI and long-term survival in horses undergoing emergency abdominal surgery. All horses undergoing an emergency ventral midline celiotomy will have blood collected pre-operatively to measure leptin and circulating adipokine concentrations. Morphometric measurements including body weight, withers height, neck circumference and body condition score (nine point scale) will also be recorded.

Phase I

The retrospective study has gone well. Records have been reviewed from both Cornell University and the University of Georgia over a six year period. To date we have approximately 350 medical records reviewed and data collection completed for 246 cases. Calls are ongoing to owners and trainers about incisional infections. A cursory look at the data indicates an infection rate of 24% of the colic surgeries done at the Cornell Equine Hospital. This is consistent with previous reports and supports our rationale for instituting this study. Data is currently being analyzed by Drs. Hill and Hayes together. Dr. Hill will plan to present the results at the American College of Veterinary Surgeons (ACVS) meeting in Phoenix, Arizona in 2018.

Phase II

The prospective study has taken longer to get started and accumulate cases. Our surgical colic caseload has decreased in recent years, and it took a bit of time to set up accounts and a system for collecting samples. We have enrolled 10 horses from Cornell's Equine Hospital and have identified a new house officer, Laia Reig-Codina to continue work on this project. *Note: Dr. Reig-Codina is an intern. If she does not stay for a residency at Cornell we will still keep her involved in the study and may add the incoming first year resident (this would not be until July 2018). Dr. Fubini and Dr. Radcliffe will continue to oversee this part of the project. Dr. Hill is now working at a private practice in Colorado, Littleton Equine Medical Center, which typically sees a relatively high caseload of colics each year (approximately 70 surgical cases/year). To help increase the number of cases enrolled in the prospective portion of the study, cases will also be collected at this clinic and samples sent to Cornell for analysis.

a) **Reig L**, Werre SR, Brown JA. Outcome and risk factors for post-operative complications after umbilical resection in 82 foals (2017) (Under revision). Short abstract accepted for AAEP 2017 Annual Convention
b) **Reig L,** Brown JA. Pleomorphic sarcoma with giant cells in the paranasal sinuses of a 27-year-old Thoroughbred mare 2017 (In preparation).

# *Harry M. Zweig Memorial Fund for Equine Research*
# *2017 Annual Progress Report for Zweig Committee*

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| P.I.: | Dr. Heidi Reesink |
| Title: | Quantitative Computed Tomography and Bone Quality Assessment for the Prediction of Fetlock Breakdown Injuries in Racehorses |
| Project Period: | 1/1/17-12/31/18 |
| Reporting Period | 1/1/17-12/31/17 |

## ***TITLE:*** *Quantitative Computed Tomography and Bone Quality Assessment for the Prediction of Fetlock Breakdown Injuries in Racehorses*

### ***PRINCIPAL INVESTIGATOR(S):*** *Heidi Reesink*

***Aim 1.*** To compare quantitative measurements, including bone geometry, bone mineral density, bone volume fraction and trabecular thickness, between horses with PSB fracture and controls, using micro-CT and conventional CT. To correlate QCT measurements with histological parameters.

Bone morphology was analyzed in 16 TB racehorses (samples collected from 2015-2016) using micro-CT. Differences in bone geometry, bone volume fraction, and trabecular thickness between horses with PSB fracture and controls were evaluated. The main conclusions were that bone volume fraction of the PSBs was increased in fracture horses compared to controls, and the width (relative to animal body weight) of the PSBs was smaller. Sagittal sections from all 4 sesamoid bones of 16 horses have been processed for histology. Slides have been stained **(Fig 1)**, and sections from the contralateral limb of fracture horses (FXCL) and the paired limb form the sex and age matched control horses (SAMC) were blinded and are currently being scored to evaluate the degree of osteoarthritis. Staining
with Masson's Trichrome was performed to evaluate the degree of mineralization of bone ((red: highly mineralized bone, blue: new osteoid or poorly mineralized bone).

Since the start of 2017, an additional 20 horses (n=10 PSB fracture, n=10 control) have been imaged with both clinical QCT (in situ, 500μm) and micro-CT (ex vivo, 50μm), (Table 1). Dual imaging was performed to increase sample size of micro-CT measures (i.e., combine data from 2015 through 2017) as well as determine if clinically relevant QCT with lower resolution could still detect changes in bone morphology associated with increased fracture risk.

Bone volume fraction was the measure of bone morphology that most accurately classified fracture from control horses from micro-CT images. Unfortunately, bone volume fraction is not an accurate measurement in clinical QCT due to the decrease in resolution. Therefore, a new measure that also represented the density of bone (amount of bone tissue verses bone marrow and vascular space) was required for clinical QCT images. Density in QCT images was measured and directly compared to the bone volume fraction as measured in micro-CT in the same bone and were found to be highly correlated ($R^2$ = 0.87, **Fig 2A**).
Density was measured from clinical QCT images in fracture and control horses and was found to be higher in fracture horses as compared to controls **(Fig 2B)**.

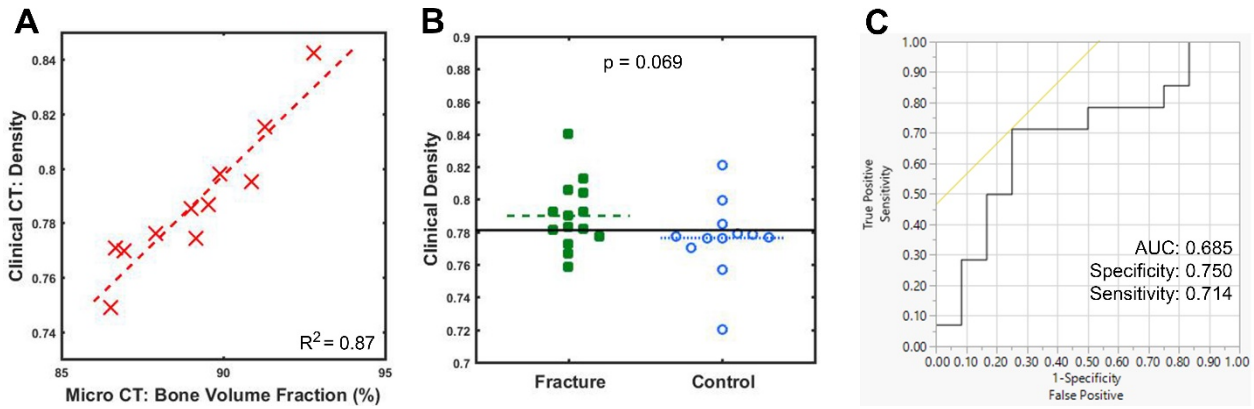


**Figure 2. (A) Bone volume fraction as measured by micro-CT was highly correlated to density as measured in the lower resolution clinical CT. Data points represent a single PSB imaged with both micro and clinical CT. (B) There is a trend towards increased density in fracture horses as compared to control horses as detected by clinical QCT. The black horizontal line represents the cut off value from the ROC curve, indicating that density accurately classified most horses with only 3 false positives and 4 false negatives. (C) The ROC curve for density from clinical CT separated fractured from control horses with a specificity of 0.750 and sensitivity of 0.714.**

***Aim 2.*** To develop a mathematical model encompassing epidemiologic variables, including exercise history and QCT imaging analysis that will detect horses at increased risk of PSB fracture.

We have developed mathematical models from micro-CT bone morphological measures to predict fracture risk in TB racehorses (see EVJ manuscript in Appendix). We are working to improve/update the models as we increase our sample size. We are also moving towards creating a mathematical model from clinical CT using bone density, measures of bone size, and animal weight. As our numbers are too small to preform accurate epidemiologic modeling, we are collaborating with co-investigators who have developed fracture risk estimations based on epidemiologic and pre-race veterinary exam findings from hundreds of horses. We are working on testing a model that incorporates our findings on bone morphology and our collaborator's epidemiologic findings and applying it to new incident horses to assess how often the model accurately classifies fracture and control horses. We envision this model could be used by owners and trainers to determine fracture risk of their horses based first on epidemiologic data, and if risk is high, pre-catastrophic imagine could be performed with clinical CT to further assess fracture risk based on bone morphology.

Catastrophic proximal sesamoid bone fracture remains the leading cause of racehorse fatalities, causing nearly 80% of all fatal musculoskeletal injuries on NY racetracks over the past 4 years. There are currently no good diagnostic tests or clinical modalities for detecting horses at increased risk of proximal sesamoid bone fracture. Catastrophic musculoskeletal injuries are of enormous impact to horseracing, both due to the potential safety concerns for horses and jockeys and also due to welfare concerns among the public.

Plans to address Specific Aims 1 and 2 have not been significantly modified. We are on track to reach our goal of imaging 30 horses before the end of 2018. In addition, samples are currently being processed, sectioned and stained for histological analysis. Plans for 2018 include continuing to image the remaining 10 horses via clinical and micro-CT, processing and staining PSBs, and scoring PSBs for OA. We expect to publish an abstract and a manuscript describing the translation of results from micro-CT to clinical CT in 2018.

***Harry M. Zweig Memorial Fund for Equine Research***
***2017 Final Report for Zweig Committee***

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| P.I.: | Dr. Tracy Stokol |
| Title: | Platelets are a Trojan Horse that deliver Equine herpes virus to endothelial cells. |
| Project Period: | 1/1/17-12/31/18 |
| Reporting Period | 1/1/17-12/31/17 |

***TITLE:*** Platelets are a Trojan Horse that deliver Equine herpes virus to endothelial cells.

***PRINCIPAL INVESTIGATOR(S):*** *Tracy Stokol*

In this proposal we had three aims.

Aim 1: Platelet infection of ECs: Here we proposed to determine the efficiency of platelet-mediated EC infection under physiological shear. In preliminary data, we had found that platelets could transfer virus infection to endothelial cells under shear. We have repeated the experimental procedures several times and cannot duplicate our promising preliminary results, despite troubleshooting experimental procedures substantially. As disappointing as this is, this is the nature of research.

Aim 2: Platelet infection of PBMCs: Here we had proposed to assess whether platelets can transfer infection to and activate immunopurified PBMC subsets (monocytes and T cells). We are currently testing this objective, having harvested PBMCs and platelets from the same horse, exposed platelets to virus, then have assessed for virus transfer and infection of PBMCs. We have also harvested mRNA and DNA from the co-cultures to quantify virus infection and cytokine production. Experiments are ongoing and data is still being accumulated and has not been fully analyzed.

Aim 3: Inhibitory drug screen: Here, we proposed to screen a phosphodiesterase-3 inhibitor and two factor X inhibitors for their ability to inhibit EHV-1-induced platelet activation and P selectin upregulation. We have completed this aim and published our data.

Publications: Stokol T, Serpa PB, Zahid MN, Brooks MB (2016) Unfractionated and low-molecular-weight heparin and the phosphodiesterase inhibitors, IBMX and cilostazol, block *ex vivo* Equid Herpesvirus type-1-induced platelet activation**.** *Front Vet Sci.* **3**:99. doi: 10.3389/fvets.2016.00099. (Corrigendum for incorrect initials for Dr. Serpa on Feb 10, 2017, doi: 10.3389/fvets.2017.00010).

Submissions and/or External Grants Resulting from the Award: We received a Grayson proposal to test whether administration of heparin to horses can inhibit EHV-1-induced platelet activation ex vivo. The latter project is ongoing.

# *Harry M. Zweig Memorial Fund for Equine Research*
# *2017 Annual Progress Report for Zweig Committee*

| | |
|---|---|
| P.I.: | Dr. Gerlinde Van de Walle |
| Title: | Microencapsulated stem cells to promote wound healing |
| Project Period: | 1/1/17-12/31/18 |
| Reporting Period | 1/1/17-12/31/17 |

## ***TITLE:*** *Microencapsulated stem cells to promote wound healing*

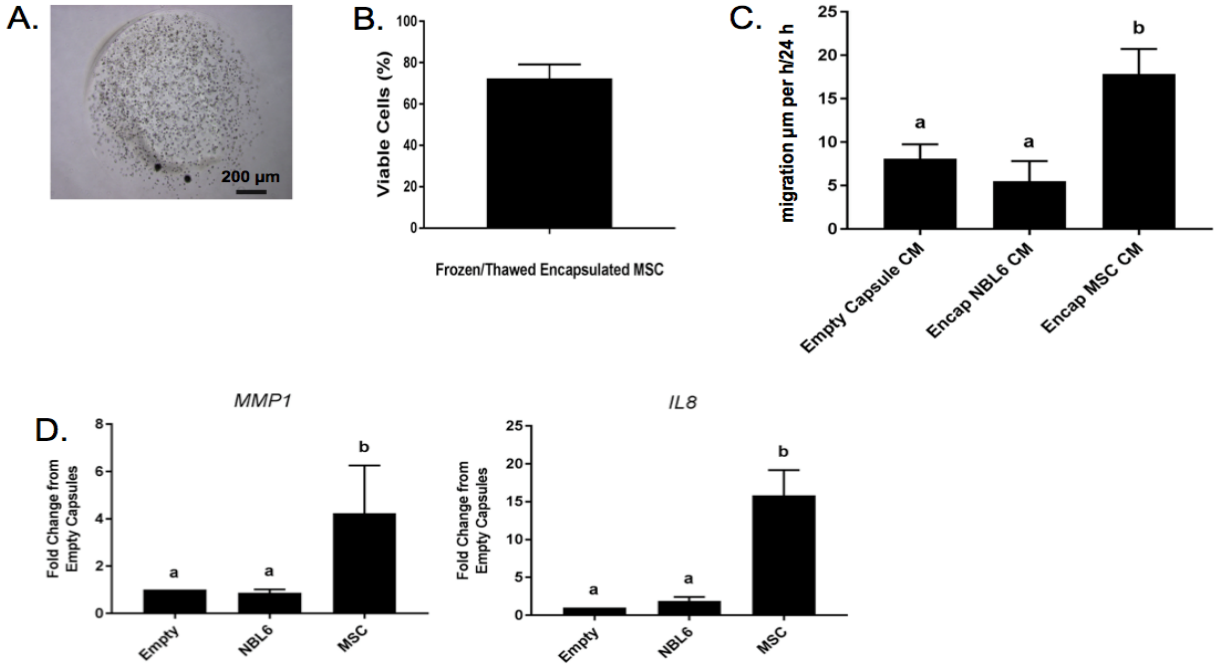
***PRINCIPAL INVESTIGATOR(S):*** *Gerlinde Van de Walle*

→ The three specific aims described in our original proposal have not been modified.

→ This year, we have made significant progress regarding the experiments described in aim 1 and have successfully completed aim 3 of our proposal.

The goal of aim 1 is to identify factors in the equine mesenchymal stem cell (MSC) secretome that contribute to wound healing, and confirm their bioactive roles. We experimentally determined that the bioactive factors are proteins, and then used both biased (antibody array) and unbiased (LC-MS/MS) approaches to define the protein composition of MSC conditioned medium (CM), which contains all factors secreted by MSC. From the lists of proteins we obtained, we focused on those previously reported in the literature to contribute to wound healing. We then optimized RNA interference assays to knock down the expression of these proteins of interest systematically, in MSC cell lines growing in culture. We have collected the CM from these MSC and are testing it in functional assays to determine how the absence of specific proteins changes the effectiveness of MSC CM to promote wound healing. We have found an active factor that is involved in wound healing in our *in vitro* assays, based on a significant reduction of fibroblast migration when this factor is silenced in the MSC CM, and are following up to determine how this factor interacts with target cells, to improve wound healing.

The goal of aim 3 is to develop and validate methods to store encapsulated MSC long term for off-the-shelf use. We have encapsulated MSC in alginate beads, and frozen them in liquid nitrogen for long-term storage. We thawed the encapsulated MSC, and determined they are viable, retain their stem cell characteristics, and still release factors that can affect target cell gene expression and migration.




Our findings that (i) we identified at least 1 specific protein in the equine MSC secretome that promotes wound healing *in vitro* and (ii) we demonstrated that encapsulated equine MSC can be frozen for long term storage, then thawed for use, are significant as they suggest that MSC have the potential to be used as an effective and practical therapy for equine cutaneous wounds *in vivo*. Defining the factor(s) responsible for the effects of MSC on wound closure will help us refine MSC treatments by screening MSC for the expression of active factor(s) prior to use, pre-treating MSC to increase the secretion of these specific active factors(s) and/or developing culture methods that maximize the secretion of these factors. Moreover, and importantly, frozen encapsulated MSC will provide clinicians with a readily available, off-the-shelve therapy that can be easily administered.

During the next year, we will work on aim 2 of the proposal, by evaluating the efficacy of microencapsulated equine MSC on wound healing using a naturally occurring wound model. We have been communicating with Dr. Bettina Wagner and will be prepared to start *in vivo* experiments using horses from her herd in the spring of 2018.

**Catastrophic Failure in Proximal Sesamoid Bones of Racehorses is Associated with Increased Bone Density**

Erin N. Cresswell1, Sean P. McDonough1, Scott E. Palmer1, Christopher J. Hernandez1, Heidi L. Reesink1
1Cornell University, Ithaca, NY
**Disclosures:** Erin N. Cresswell (N), Sean P. McDonough (N), Scott E. Palmer (N), Christopher J. Hernandez (N), Heidi L. Reesink (N)
**INTRODUCTION:** Catastrophic failure of the proximal sesamoid bone(s) in elite racehorses results in dramatic, often fatal, fractures that occur during high speed races. Proximal sesamoid bone fractures occur without prior symptoms in otherwise healthy, athletic animals. Fracture pathology is poorly understood, and it is therefore unclear what aspects of bone structure and material properties contribute to fracture risk. Understanding changes in bone morphometry that increase fracture risk could lead to screening techniques to identify horses at high risk of fracture. The goal of this study was to identify whole bone and regional changes in bone morphometry in horses that sustained a proximal sesamoid bone fracture.
**METHODS:** Proximal sesamoid bones were acquired from 3-8 year old Thoroughbred racehorses following euthanasia as part of the requirement for necropsy by the New York State Gaming Commission. Bones were harvested from four racehorses that were euthanized due to fracture of proximal sesamoid bones in one

forelimb and from four sex and age matched controls that did not undergo forelimb fracture. Where available, two proximal sesamoid bones were collected from each limb, providing 7 fractured sesamoid bones, 1 intact sesamoid in a limb that fractured, 8 intact sesamoid bones from the contralateral limb, and 14 sesamoids from controls (n=30 proximal sesamoid bones). Images of proximal sesamoid bones were collected using micro-computed tomography (voxel size of 50 microns) and images were segmented using local adaptive thresholding. Bone volume fraction (bone volume/ total volume, BV/TV) was calculated for the whole bone as well as within independent sub-regions classified by anatomical location as apical, mid-body, basilar, subchondral, medullary, and flexor (Fig 2) [1]. As no differences were found between medial and lateral sesamoids or those between left and right limbs, measures were pooled into fracture and control groups (Figure 1). The contralateral limbs from animals that had sustained a proximal sesamoid bone fracture were used to quantify regional changes in the fracture group, as the fractured proximal sesamoid bones themselves were anatomically altered and regions could not be accurately divided. Mean differences in bone volume fraction per region are reported. A t-test with unequal variance was used to determine significant changes in regional bone volume fractions between groups.

**RESULTS:** Whole bone volume fraction was increased in sesamoids of the fracture group as compared to controls (Figure 1). Additionally, BV/TV was greater in all nine sub-regions in the fracture group. Differences in regional BV/TV were greatest in the basilar region of the sesamoid bones, while changes in the apical region were not apparent (Figure 2). In the four limbs that sustained a proximal sesamoid bone fracture, 7 of the 8 proximal sesamoid bones fractured: 1 apical fracture, 2 mid-body fractures, and 4 basilar fractures. Regional BV/TV changes correspond to locations where fractures occurred in the samples of our study (largest increase in BV/TV in the basilar region, 3/4 horses in the fracture group sustained a basilar fracture).

**DISCUSSION:** Sesamoid bones in racehorses that experienced a fracture were less porous than controls. The increased bone volume at locations where fracture occurred suggests that increases in BV/TV contributed to fracture. While it is possible that the increase in BV/TV is secondary to other contributors to fracture pathology such as changes in bone tissue material properties, joint morphology, or alterations in loads across the joint, the increase in BV/TV may be a major factor in fracture etiology. BV/TV is generally thought to increase bone strength. However, the sudden nature of sesamoid bone fractures suggests that failure may be dominated by impaired fracture toughness leading to unstable crack growth. Unstable crack growth is most rapid in materials with very low porosity. More porous materials can be more resistant to crack growth because the voids within the material can arrest crack propagation [2]. Voids can therefore act as a toughening mechanism, preventing a crack from unstable propagation. We conclude that increases in BV/TV in the proximal sesamoid bones in racehorses may be an indicator of fracture risk by enabling unstable crack growth, which, in the presence of loading leads to fracture.

**SIGNIFICANCE:** Fracture of proximal sesamoid bones accounts for 42.5% of fatalities caused by musculoskeletal injuries in racehorses [3]. Identifying bone morphological changes that increase fracture risk could lead to screening techniques to identify racehorses at a high risk of fracture, thereby enhancing animal welfare.

**REFERENCES:** [1] Anthenill, L.A., et al. (2010)Amer J Vet Res 71(7): 755-765. [2] Cook, R.B., et. al. (2009) J Biomech 42(13): 2054-2060.
[3] Johnson, B.J., et al. (1994) Equ Vet J 26(4): 327-330.

**ACKNOWLEDGEMENTS:** The authors thank the New York State Gaming Commission (NYSGC) for funding the post mortem examinations.

ORS 2017 Annual Meeting Poster No.1256

*Harry M. Zweig Memorial Fund for Equine Research*
*2017 Annual Progress Report for Zweig Committee*

| | |
|---|---|
| P.I.: | Dr. Bettina Wagner |
| Title: | Effects on equine herpesvirus (EHV) vaccination frequency on host immunity on EHV-1 |
| Project Period: | 1/1/16-12/31/17 |
| Reporting Period | 1/1/17-12/31/17 |

***TITLE:*** Effects on equine herpesvirus (EHV) vaccination frequency on host immunity on EHV-1

***PRINCIPAL INVESTIGATOR(S):*** *Bettina Wagner*

The aims have not been modified.
**Aim 1**, we will vaccinate non-pregnant mares and geldings with the same vaccination schedule (60 or 90 day intervals) as used previously for the pregnant mares to identify if pregnancy status or gender influences adverse immunity to frequent vaccination.
**Aim 2**, we will test if short vaccination intervals of 30 or 45 days will consistently induce adverse immunity in horses. For both aims, we will evaluate local and systemic EHV-1 specific antibody and cellular immune responses induced by the different vaccination protocols.

Proposed timeline: Aim 1 was planned to be performed in year 1 and Aim 2 in year 2 of the project.

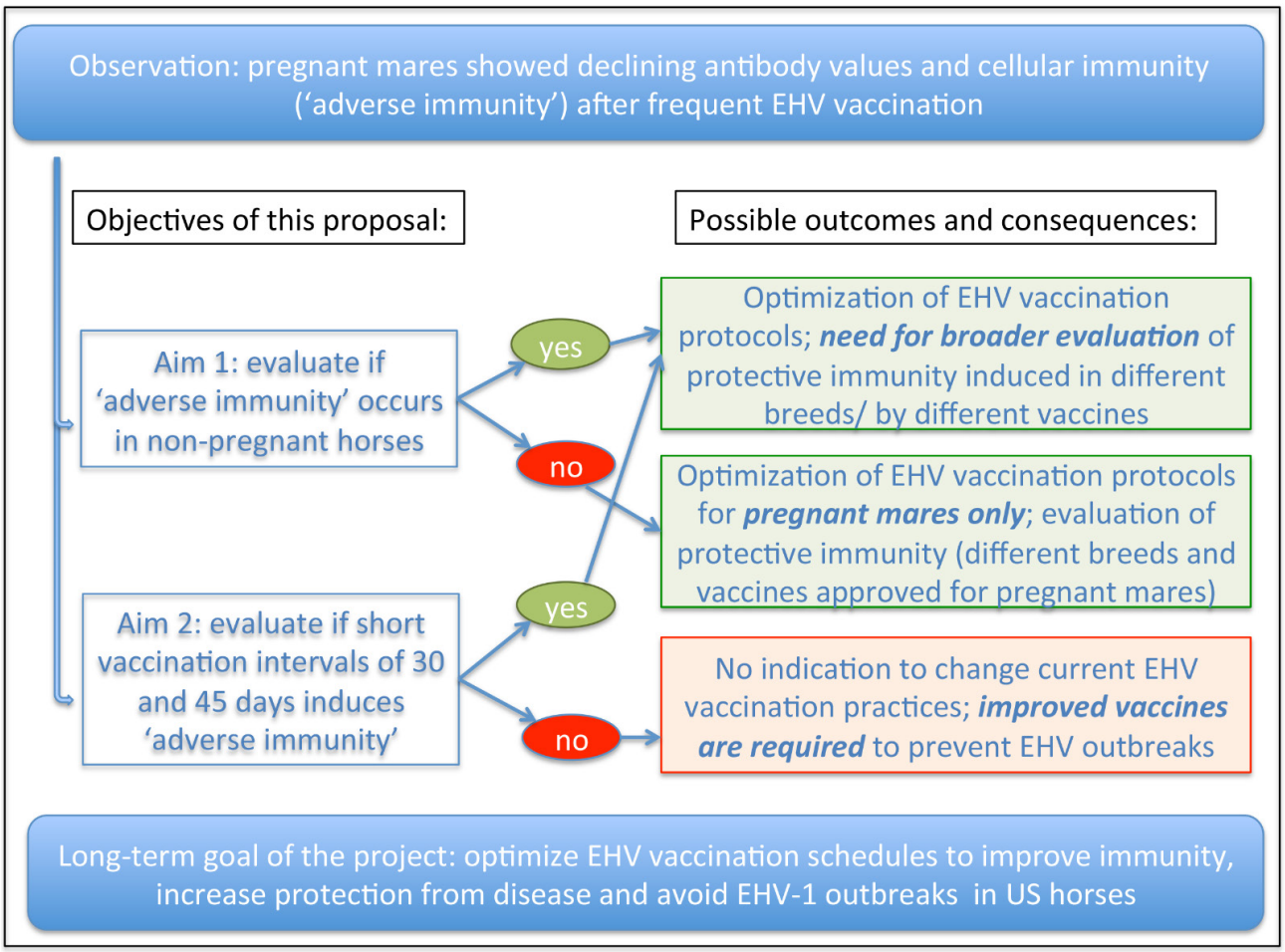
In year 1 of the project, we have performed the vaccination approach outlined in Aim 1. We have previously observed that frequent vaccination in 60-90 day intervals with a commercial, inactivated EHV vaccine decreased the humoral and cellular immune response of the vaccinated pregnant mares (Wagner et al. 2015, Vaccine 33: 5588-5597). The goal of Aim 1 is to investigate whether this unexpected immune response was due to pregnancy or is a general effect of frequent EHV vaccination on the immune response. This is of relevance because frequent EHV vaccination has been practiced in the US in recent years in response to the ongoing EHV outbreaks. However, scientific data on frequent EHV vaccination and its effect on host immunity are lacking.

In Aim 1, we performed frequent vaccination in non-pregnant horses with the same vaccine as previously used in the pregnant mare vaccination study above. Fifteen adult horses, 6 geldings and 9 non-pregnant mares were vaccinated using the vaccination schedule previously used for the pregnant mares. Horses received an initial vaccination and 5 boosters over a period of eight months. The last vaccination for the frequent vaccination experiment in Aim 1 was given on 9/6/16.

Blood and nasal secretion samples were taken from the horses at various time points throughout the vaccination study. The last sampling date for this study will be 11/21/16. Blood samples were continuously used for analysis of cellular immune parameters such as EHV-1 specific T-cells and EHV-1 induced cytokine secretion. The analysis of the cellular immunity data is ongoing. Serum and nasal secretion samples were frozen for total EHV-1 specific antibody and antibody isotype detection. The antibody measurements will be done as soon as the last study samples are collected in November followed by the analysis of the antibody data. Ongoing.

Equine industry and the equine veterinary community responded to the ongoing EHV-1 outbreaks with a tendency to increase vaccination frequency with currently approved EHV vaccines. The American Association of Equine Practitioners (AAEP) recommends EHV vaccination every 6 months for competition horses. Some vaccine suppliers recommend vaccinating every 3 months. In addition, equine events required frequent EHV-1 vaccination with documentation that the horse has been vaccinated within a certain time frame preceding the event (often less than 30 days). These

vaccination practices and requirements resulted in multiple EHV vaccinations per year for many horses. Although, we believe that every vaccination will induce increasing immunity or at least maintain the existing levels of immunity, data to support that frequent administration of EHV vaccines improves immunity and protection from disease are missing. Moreover, retrospective data analyses from neurological EHV outbreaks suggest that frequent vaccination is a potential risk factor for disease. Figure 1 describes the Aims of this project in relation to the outcomes and possible actions depending on the outcome of each of the Aims.




In parallel to this project and as a consequence of the increasing trend from equine event organizers to require EHV and Influenza vaccination in very short time frames prior to the event, the US Equestrian Federation (USEF) has set the requirement of EHV and Influenza vaccination in 2016. Competition event organizers can now require horses to be vaccinated 6 months prior to the event but not more frequently.

Figure 1: Project Aims, outcomes and consequences for EHV vaccination schedules.

During the second year of support, Aim 2 will be performed and analyzed. We will use another 10 horses for the short vaccination interval study outlined above for Aim 2. The first vaccination for Aim 2 is planned for mid January 2017. Cellular immune parameters and local and systemic antibody responses will be measured. We do currently not anticipate any modifications to the original plan outlined in the proposal.

# APPENDIX B

# SUMMARY OF 2017 EXPENDITURES

| | |
|---|---|
| 2017 Research Awards | $449,675 |
| 2018 Public Relations and Administrative Budget | $29,200 |
| 2017 Incentive Awards | $20,000 |

Total Expenditures: $498,875

# APPENDIX C

# RESEARCH PRESENTATIONS

**November 29, 2017**
**Cornell College of Veterinary Medicine**
**Ithaca, New York**

9th Annual Harry M. Zweig Memorial Fund for Equine Research Poster Session & Talks

Cornell College of Veterinary Medicine Ithaca, New York

FEATURING SPEAKERS FROM CORNELL'S COLLEGE OF VETERINARY MEDICINE

**Wednesday, November 29, 2017 – S1-122**

**3:00PM Welcome!** ***Robert Weiss - Associate Dean Research/Graduate Education***

COLLEGE OF VETERINARY MEDICINE
RESEARCH PRESENTATIONS

## ***How Zweig Funding Changed Accepted Paradigms in Regenerative Medicine***

*Lisa Fortier* – James Law Professor – Large Animal Surgery
https://www2.vet.cornell.edu/research/faculty/lisa-fortier-dvm-phd

## ***The Mesenchymal Stem Cell Secretome in Equine Wound Management***

*Gerlinde Van de Walle – Zweig Assistant Professor (2017-2019)*
https://www2.vet.cornell.edu/departments-centers-and-institutes/baker-institute/about-us/faculty-staff-and-students-directory

## ***Regenerative Approaches for Recurrent laryngeal Neuropathy***

*Jonathan Cheetham – Associate Professor, Large animal Surgery*
https://www2.vet.cornell.edu/research/faculty/jonathan-cheetham-vetmb-phd-dacvs

## ***Equine Biobanking – Paving the Way to Personalized Medicine***

Marta Castelhano – Director – Cornell Veterinary Biobank
https://www2.vet.cornell.edu/research/faculty/marta-g-castelhano-dvm-mvsc

## ***Vaccination of Horses with Lyme Vaccines for Dogs Induces Short-Lasting Antibody Responses***

Cassandra Guarino – Extension Associate
https://www2.vet.cornell.edu/research/faculty/cassandra-m-b-guarino

**4:30pm-5:30pm - Poster Session and Reception – Atrium**

# *Cornell University & Harry M. Zweig Memorial Fund for Equine Research Presentations - November 29, 2017*
## *Equine researchers present to the Zweig Committee*
## *Cornell College of Veterinary Medicine, Ithaca, New York*

Faculty from Cornell University's College of Veterinary Medicine, Ithaca, New York, presented a series of equine-related research projects and lectures on November 29, 2017 at the College. The research presentations and poster session were well attended by the committee members of the Harry M. Zweig Memorial Fund for Equine Research, College faculty and staff, and others from around the university.

Speakers included *Lisa Fortier – James Law Professor – Large Animal Surgery*, *Gerlinde Van de Walle, Zweig Assistant Professor (2017-2019), Jonathan Cheetham,* Associate Professor, Large Animal Surgery, *Marta Castelhano, Director – Cornell Veterinary Biobank, and Cassandra Guarino – Extension Associate*, and their respective lectures; "How Zweig funding changed accepted paradigms in regenerative medicine", "The Mesenchymal stem cell secretome in equine wound management", "Regenerative approaches for recurrent laryngeal neuropathy", "Equine biobanking – Paving the way to personalized medicine", and "Vaccination of horses with Lyme vaccines for dogs induces short lasting antibody Reponses".

The annual meeting was held on Thursday, November 30, 2017 at the College of Veterinary Medicine, Ithaca, NY. Dean Lorin Warnick, thanked current and new members for the dedication, expertise and knowledge they bring to the Zweig Committee, which is a big part of the success of the Zweig Fund in helping to support projects related to equine research.

The Harry M. Zweig Memorial Fund for Equine Research honors the late Dr. Harry M. Zweig, a distinguished veterinarian, and his numerous contributions to the state's equine industry. In 1979, by amendment to the pari-mutuel racing and wagering law, the New York State legislature created the Harry M. Zweig Memorial Fund for Equine Research to promote equine research at the Cornell University College of Veterinary Medicine. The Harry M. Zweig Committee was established for the purpose of administering the fund and is composed of individuals in specified state agencies and equine industry positions and others who represent equine breeders, owners, trainers, and veterinarians. The Fund contributes a percentage of its revenue to support a variety of equine-related research. The Fund is proud to support the Harry M. Zweig Memorial Fund for Equine Research. This first-rate research helps to provide protection and preventative planning for the equine industry, which in turn helps to ensure a healthy and positive future for the horse racing industry.

The committee administering the fund always includes the chairman of the New York State Racing and Wagering Board or his designee, the dean of the College of Veterinary Medicine at Cornell or his designee, a member or the executive director of the Agriculture and New York State Horse Breeding Development Fund, a member or the executive director of the New York State Thoroughbred Breeding and Development Fund, and at least five New York State breeders, owners, trainers, or veterinarians in equine practice. Dean Lorin Warnick currently serves on the committee, representing the College and its many researchers who have received the Fund's support for research projects advancing equine health and athleticism.

# APPENDIX D

# AWARDS FOR 2018

# 2018 Harry M. Zweig Memorial Fund for Equine Research Awards

| **CONTINUATION** | | **ANNUAL AWARD** |
|---|---|---|
| Dr. Antczak | Functional Gene Annotation in the Horse | $60,083 |
| Dr. Cheetham | Regenerative Approach to Recurrent Laryngeal Neuropathy | $48,899 |
| Dr. Fubini | The Relationship between Obesity and Post-Operative Incisional Infections following Abdominal Surgery in the Horse. | $16,528 |
| Dr. Reesink | Quantitative computed Tomography & Bone Quality Assessment for the Prediction o Fetlock Breakdown Injuries in Racehorses | $49,643 |
| Dr. Van de Walle | Microencapsulated Stem Cells to Promote Wound Healing | $56,399 |
| | Sub-Total: | $231,552 |
| **NEW/Revised** | | **ANNUAL AWARD** |
| Dr. Cheetham | Accelerating Recovery after Laryngeal Nerve Graft | $37,997 |
| Dr. Divers | Characterizing Tropism and Transmission of Equine Parvovirus-Hepatitis (EqPV-H) | $79,006 |
| Dr. Nixon | Next Generation Arthritis Control through Lubricin and IL-1 Receptor Antagonist Overexpression in Carpal OA | $55,000 |
| Dr. Reesink | Intra-Articular Recombinant Lubricin To Restore Joint Lubrication And Prevent Osteoarthritis In Horses | $60,054 |
| Dr. Van de Walle | The Mesenchymal Stem Cell Secretome against Equine Herpesvirus Type 1 Infections | $67,136 |
| Dr. Wagner | Towards a Neonatal Vaccine against Equine Herpes Virus Type 1 (EHV-1) | $99,314 |
| | Sub-Total: | $398,507 |
| | TOTAL: | $630,059 |
| | | |

# APPENDIX E

# ZWEIG NEWS CAPSULES

Zweig

From the Harry M. Zweig Memorial Fund for Equine Research at Cornell University College of Veterinary Medicine

No. 64 October 2017

# Dr. Robert Weiss appointed as new Associate Dean for Research and Graduate Education

Dr. Robert Weiss

On October 1, 2017, Dr. Robert Weiss, professor of Biomedical Sciences, became Associate Dean for Research and Graduate Education, following the role that Dr. Bettina Wagner filled from 2014 to 2017. "I'm excited about this opportunity," Weiss says. "I'm passionate about the areas that this administrative position oversees."

Weiss's appointment as the new associate dean was a natural one. "Having served on key college and university research committees and in graduate field leadership roles, in addition to his own graduate student advising, Dr. Weiss brings a wealth of experience to the position of associate dean," says Dean Lorin Warnick. "He also is director of the Comparative Cancer Biology Program at the College and recognized for his work in fostering communication between cancer patients and researchers in the community through the Cancer Resource Center. I am very appreciative of Bob's willingness to take on the role of Associate Dean and look forward to the expertise and perspective he will bring to this critical position in the College."

Weiss is no stranger to the Zweig Memorial Fund for Equine Research; as co-chair of the College Research Council, he conducted the internal reviews for research proposals to present to the Zweig committee. "We're fortunate to have Zweig's incredible support for ground-breaking studies in equine health," he says. "Much of the work done by Zweig-funded scientists fits in with some of the key strategic research areas highlighted by Cornell's provost, including infectious disease and genomics—both of which has active representation at the College," says Weiss.

Additionally, Weiss is excited to support all scientific endeavors at CVM in his new position, and plans to continue building the College's role as a leader in key areas of research while also expanding its scientific prowess into new areas, including neurobiology and computational biology. "We'll also continue to strive to bridge together our basic and clinical researchers," he says, and build the College's work in clinical trials and partnerships with pharmaceutical companies to bolster drug development.

Weiss now also oversees graduate education at the College, which he aims to ensure remains excellent. "The College has a very supportive environment for graduate students," he says. "We're dedicated to mentoring our trainees and setting them up for career success."

Weiss's own experience with research has been rich—starting as an undergraduate at Wabash College where he developed a mathematical model for mollusk feeding behavior. As a PhD student at Baylor College of Medicine he studied oncogenic viruses, which eventually directed him to his current interest in cancer biology both as a postdoctoral fellow at Harvard Medical School, and later as a faculty member at the College. "I found it to be an incredibly complex disease with a tremendous intellectual challenge," says Weiss. "And, it has a clear health challenge—it's a disease where more work is needed." Now at the administrative helm of the College's research activities, Weiss is sure to help take on this and many other scientific challenges.

# The curious clinician: Ann Dwyer DVM ‘83

Dwyer examines a patient
(photos: Dede Hatch)

If you ask Ann Dwyer DVM ‘83, the co-recipient of the Cornell University College of Veterinary Medicine’s 2017 Salmon Award for Distinguished Alumni Service, the key to her career, she might answer: “SOAP.” No, not the stuff that lathers up your hands—but the Cornell-taught approach to problems: **S**ubjective and **O**bjective observations, turning those into an **A**ssessment, then creating a **P**lan. “Cornell trains all graduates to do this kind of analysis. In reflecting back on my career, all I have done is “SOAP” many facets of being a professional,” says Dwyer. “That method applies to patients, of course, but also to things like doing research, making a speech, leading an organization, assembling a course or designing a building. This SOAP approach to life is one that I see Cornell colleagues—teachers, administrators, classmates, students and fellow graduates carry out over and over again throughout their careers. Mine is no different.”

Dwyer is currently the co-owner of the Genesee Valley Equine Clinic in Scottsville, N.Y. and is known for her expertise in equine eye diseases, currently serving as a reviewer for Equine Veterinary Ophthalmology and has authored a number of papers and book chapters on equine ophthalmology. She received a BS in biology from Mount Holyoke College in 1975 and her DVM from the College in 1983. Dwyer is a member of the AVMA, AAEP (serving as president of that organization in 2013), NYSVMS, IEOC, the Zweig Committee, and serves as vice-chair of the College’s Advisory Council.

For Dwyer, there is “no such thing as a typical day,” in her life as an equine veterinarian and practice owner. “Some days I see herds of horses, some days I see just a few patients,” she explains. “Some days I concentrate on patient care; other days I put on my practice-owner hat and tend to taxes and budget. Some days I am away from the practice, teaching vet students or doing work for AAEP. Some days I seem to do a combination of all of the above activities and am up late at night tending to email.”

## Equine interests

A love of horses has been a constant throughout Dwyer's life, from taking riding lesson from her fifth grade teacher, eventing as a teen, and teaching lessons herself while a college student at Mount Holyoke. Horses played a key role in Dwyer's self-described "tipping point" towards veterinary school—when she spent three years working at various east coast Thoroughbred racetracks as a hot walker, groom, and eventually an exercise rider. "By the time I finished my admittedly crazy, somewhat wild "adventure" on the track, I decided to throw my hat in the ring and apply to Cornell," Dwyer says. "I clearly remember typing up my application on a NON-electric typewriter in the tack room of the racing stable at Hialeah in Miami! When I sealed the envelope I think there was hay chaff inside it. To my surprise I got an interview and was accepted for the Fall of 1979."

At the College, Dwyer sunk her teeth into what would become her favorite courses--such as gross anatomy taught by Drs. Howard Evans (CALS '44, PhD '50), Alexander deLahunta (DVM '58, PhD '63), Wolfgang Sack and John Cummings (CALS '58, DVM '62, PhD '66) -- relishing the experience of sitting in lectures given by professors who "wrote the textbooks," and the camaraderie in the clinics. "All the faculty seemed to be friends and colleagues, not competitors, and they treated the students like the budding clinicians we all were," she recalls. "It was a real team feeling, quite a unique professional environment. I think it set an example for me of how things should be when a group of people come together to solve problems and help animals."

## Eying ophthalmology

When she graduated Cornell, Dwyer's parents and friends pooled their resources to buy her a direct ophthalmoscope as a graduation present. "Once I had a scope I figured I better learn how to use it, so I started looking at every horse I saw," she says. While she briefly considered pursuing specialty training in ophthalmology, "my gut told me that there was a place in the world for a general equine practitioner who had a special interest in ophthalmology. In looking at the sparse clinical literature that was available 30 years ago, my SOAP approach told me there was a need for more information on clinical problems in populations, and for relating eye problems in horses to equine health in general. So I decided to stay put in practice, but pursue every available opportunity to train in ophthalmology."

And pursue she did—even attending monthly Ophthalmology Grand Rounds at the University of Rochester Medical Center—something she continues to this day, along with serving on the Advisory Board of the Flaum Eye Institute at the University of Rochester. Dwyer also began to travel to universities in search of more ophthalmology training, spending time at University of Florida and North Carolina State, observing cases and participating in rounds.

## Staying curious

This depth of knowledge around equine ophthalmology, and the breadth of experience that veterinary medicine lends, has given Dwyer the kind of career that keeps her engaged. "Years ago I worked on an assembly line to earn money during a college summer break," she recalls. "Pretty much every day was the same. I would report for work at the appointed hour, sit down at a table and assemble traffic safety devices (blinking lanterns) … That experience taught me to appreciate the variety and the challenge that every day brings when you become a veterinarian!"

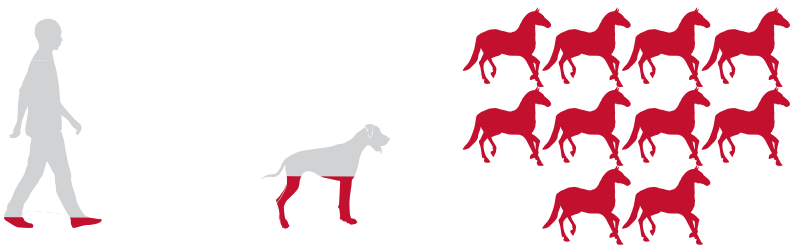
Beyond the daily excitement that veterinary medicine has given Dwyer, she points out how a Cornell-taught foundation in the field has truly set her up for a life of practical perseverance. "There is something unique about the Cornell experience that I call the 'roll up your sleeves and get it done' approach," she says. "It is eminently practical. Kind of a 'head in the clouds but boots on the ground' attitude. I have observed it throughout the university, not just in the veterinary school. To me, Cornell represents a center of excellence that keeps the focus relevant to practical problems of the world."

As Dwyer continues to manage her successful practice "one stall at a time," she's mindful of the foundation of meaning and mentorship she received at the College. "The value of having a Cornell education is that you have been given an example of mentors who are truly excellent in their respective fields but are still humble and plain spoken. They teach you to 'be in the world' and to stay curious, but to always be grateful at the same time."

# Improving the odds of equine anesthesia recovery

## BY THE NUMBERS

Horses are over 200 times more prone to anesthetic fatality during anesthesia than humans, and 20 times more so than dogs.



**0.05** out of 1,000

**0.5** out of 1,000

**10** out of 1,000

Horses face a much higher risk of dying when undergoing general anesthesia compared to humans and other domestic animals, with fatality rates reported a little shy of one percent. Many of those fatalities are attributable to orthopedic injuries that occur in the recovery room--a risk that both horse owners and veterinarians are aware of, yet have little control over.

Currently, there is no scientifically-based best practice for equine anesthesia recovery techniques. Most equine practitioners have their own specific protocols to minimize complications—primarily with the goal of minimizing a horse's startled, uncoordinated movements, which can lead to bone fractures. These methods might include rope-and-pulley systems, water-assisted recoveries, or no assistance at all. Yet, it's still unclear if these recovery methods prevent—or even cause—these injuries.

Thanks to the generous support from longtime friend and donor Richard Schechter, the College has started a study to identify the elements that are likely to ensure safe recovery from anesthesia, and to provide the evidence to justify changes in established practice, should they be necessary.

Phase one of the study is well underway. It comprises surveying equine practitioners about their methods, views, and outcomes around anesthesia recovery. The survey garnered a 43% response rate, with 380 respondents—half of which said they are willing to participate in future studies examining the topic. "This indicates there's a lot of interest with these clinicians to address this issue," says Dr. Julia Miller '12, a postdoctoral fellow working on the project.

Preliminary findings from the survey revealed that most equine practices assist patients during the recovery process, but thus far, no patterns exist in terms of whether assisted or unassisted recoveries result in better or worse outcomes.

This phase also entails filming and analyzing videos of equine anesthesia recoveries done at the Cornell University Hospital for Animals. "We'll look for a pattern after watching all these videos to determine if horses that have unacceptable recoveries fall a certain way" Miller explains. "If we do determine that, the end goal is to build a fall-prevention system."

Building and testing that system would comprise Phase two of the project, which will involve collaborations with Cornell biomechanical engineers. "The study has great potential that will require additional funding, particularly as the prototype is developed," says Miller. "Faculty are seeking additional support through research grant applications, but private funding is also encouraged from individuals who are concerned about the welfare of the horse."

Richard Schechter (r.) stands with rider Jennifer Magee and horse, Churchill's Ace, at the Wellington Winter Equestrian Festival

# Zweig funding enables advanced treatment for horses with atrial fibrillation at Cornell

Dr. Bruce Kornreich stands with On-Star the horse

*by Patricia Waldron*

When medical treatment fails, cardiologists at the Cornell University College of Veterinary Medicine can now offer a procedure that resets the quivering heart of a horse in atrial fibrillation to bring back its normal heartbeat.

Drs. Romain Pariaut, associate professor and section chief of cardiology and Bruce Kornreich, associate director of the Feline Health Center and staff cardiologist, recently performed a transvenous electrical cardioversion (TVEC) to treat a horse diagnosed with atrial fibrillation (AF), a rapid, irregular heart rhythm that causes decreased blood flow from the heart. This procedure, which involves carefully placing electrodes into the heart to reset its rhythm with an electric shock, is now available at Cornell to treat horses impacted by this condition.

"I'm very excited to be able to offer this procedure," says Kornreich. "AF is a very common condition in horses that we're often asked to diagnose and treat. This is another tool in our toolbox to convert these patients back to a normal heart rhythm."

The Equine Hospital had offered TVEC until about five years ago, when the hospital could no longer purchase the catheters needed for the procedure. The catheters recently came back onto the market, around the same time that Cornell vet students examined On-Star, a 19-year-old mare belonging to the teaching herd. "The students picked up the arrhythmia at the Cornell Equine Park and we diagnosed it as AF," says Dr. Gillian Perkins, medical director of the Equine and Nemo Farm Animal Hospital, who coordinated the procedure. "We figured this was the perfect opportunity to practice on one of our own animals so that we could offer the procedure to clients."

AF is the most common cause of an irregular heartbeat in horses, and vets often diagnose the condition in racehorses. AF occurs when the organized electrical signals that normally control heart rate and contraction become disorganized, causing a rapid and erratic heartbeat. Horses can survive for years with AF, but the condition often causes poor performance.

Traditionally, veterinarians have treated AF with quinidine, a drug that can reverse irregular heartbeat in about 85% of treated horses. The drug has several possible side effects, however, including gastrointestinal problems, low blood pressure, and even sudden death. For horses that don't respond well to quinidine, or that have had AF for several years, TVEC may be a better treatment option.

On-Star is prepared for the procedure (r.) Drs. Roberto Santilli and Romain Pariaut observe cathater placement (l.)

TVEC works just like the paddles of a defibrillator that doctors routinely use on humans, and even make the horses "jump" from the muscle spasm. In horses, however, the thick chest muscles and lungs make it impossible to shock the heart from the outside, so instead, veterinarians place three-foot long catheters tipped with electrodes into the heart via the right jugular vein.

"The most difficult part of the procedure for us is to guide the catheter," says Pariaut. "In horses it's a long path to the heart from the outside." After sedating the standing horse, they carefully maneuver one catheter into the right atrium, while a second continues its journey down into the right ventricle and up into the pulmonary artery. They use a cardiac ultrasound to monitor their progress and then perform an x-ray to verify that the metal electrodes are sandwiching the right atrium. Finally, they anesthetize the horse, stand back, and apply a carefully timed electric shock to return the heart to a normal rhythm.

On-Star's procedure took approximately four hours and required a veritable village of clinicians, including cardiologists, internists, radiologists, anesthesiologists, and licensed veterinary technicians. Combined with the expense of the catheters, the initial diagnostic evaluation, and hospitalization, the procedure costs about $3500 to $4,000.

Of course, TVEC carries its own risks, not the least of which are associated with the general anesthesia. With this in mind, Pariaut and Kornreich tried to reduce the amount of time that On-Star was under anesthesia through careful attention to electrode placement. With both TVEC and quinidine, there is also the possibility that horses will spontaneously go back into AF. One recent study found that between one third and one half of horses successfully converted from AF to normal heart rhythm will revert back after treatment. So far, On-Star is doing well and has had no complications.

Generous funding from the Harry M. Zweig Memorial Fund and the large animal medicine and cardiology sections made TVEC possible for On-Star, and for future patients diagnosed with AF. "We expect that offering this technique will draw cases to our hospital, thereby improving our ability to train large animal medicine and cardiology residents and veterinary students," says Perkins. "We are very pleased that On-Star, a member of our teaching herd, could help us move forward in this capacity."

"We're very thankful to everyone involved for their support," says Kornreich. "We are excited to have TVEC available once again, and we hope that we'll now be doing it routinely."

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# A Molecular Brace for Sprains

Dr. Lisa Fortier

*by Patricia Waldron*

Rest, ice, compress and elevate is the advice that most people get after a sprained ankle from tripping on stairs or falling on the soccer field. These measures help alleviate the pain, but no clinical data backs up this advice as the best treatment for healing or long-term joint health.

Dr. Lisa Fortier, a professor of large animal surgery just recently given the James Law professorship, is looking for new treatments for sprains. She is partnering with Dr. Hazel Szeto of Weill Cornell Medical School to test a peptide called SS-31 in horses with mild to severe ankle sprains. The peptide stabilizes mitochondria in the damaged tissue, acting like a molecular brace for the sprain, and preventing further damage, which can lead to the irreversible process of arthritis. Fortier's work will yield evidence-based therapies for treating sprains, with the ultimate goal of averting arthritis in horses, dogs and humans.

While sprains are painful and inconvenient, a more serious health problem in humans and horses is the sequela of post-traumatic osteoarthritis. Trauma from a fall, vehicle accident, or military injury can trigger this type of arthritis, which develops months, years or even decades after the initial damage.

Injury to a joint also damages the mitochondria in cartilage tissue. These organelles, which normally are packed full of waves of internal membranes, become disorganized and leaky. Without a source of energy from the mitochondria, cells die and the cartilage degrades. Developing drugs that target the cells in the cartilage, however has been a challenge for researchers. Szeto and colleagues serendipitously discovered SS-31, a small, water-soluble peptide with a unique charge density that allows it to penetrate the tissue and cross the cell membrane. It enters the mitochondria where it binds to cardiolipin, a phospholipid that makes up part of the inner membrane, thus stabilizing the membrane and restoring power to the cell.

Now, Fortier and Assistant Professor Dr. Michelle Delco are testing a single dose of the peptide in six horses that have experienced trauma to the cartilage of the hock joint, as a model for human ankle sprains. It's a proof-of-concept experiment to see if they can stabilize the tissue and prevent arthritis. "We're trying to study the early disease process, long before clinical sign associated with arthritis such as bone spurs and lameness appear," says Fortier. It's the very early stages of trauma that we and others are targeting for therapy and prevention."

The resulting treatments can be applied to human ankles, horses with stifle injuries, and dogs with cruciate ligament trauma. "That's the cool thing about our work," says Fortier. "We identify a problem in the clinic and we can take it to the laboratory and design studies to simultaneously help humans and animals."

The Harry M. Zweig Memorial Fund for Equine Research honors the late Dr. Harry M. Zweig, a distinguished veterinarian, and his numerous contributions to the state's equine industry. In 1979, by amendment to the pari-mutuel revenue laws, the New York State legislature created the fund to promote equine research at the College of Veterinary Medicine, Cornell University. The Harry M. Zweig Committee is established for the purpose of administering the fund and is composed of individuals in specified state agencies and equine industry positions and others who represent equine breeders, owners, trainers, and veterinarians.

Cornell University
College of Veterinary Medicine

Zweig News Capsule
S2054 Schurman Hall
Ithaca, NY 14853-6401



Yes Mickey, winner of the Dr. Harry M. Zweig Memorial Trot at Vernon Downs, pictured here with Mrs. Anna Zweig and Mr. Brian Zweig, along with Cornell faculty, staff, and student.

**Have you visited our Web site lately?**
*www.vet.cornell.edu/zweig*
This site provides information on the projects and publications resulting from the Zweig Memorial Fund, and demonstrates the objectives of the Fund in promoting equine health in the racing industry.

The Zweig News Capsule is published twice a year, and can be downloaded in PDF format at http://bit.ly/ZweigNews
Please encourage other equine enthusiasts to visit this site.

Zweig

From the Harry M. Zweig Memorial Fund for Equine Research at Cornell University College of Veterinary Medicine

No. 65 June 2018

# Doug Antczak gives talk on equine genetics at Arabian Horse Festival

*By Starr Todd*

The first annual Asharqia Arabian Horse Festival drew 40,000 breeders, horse owners, artists, and tourists to Dammam, Saudi Arabia for four days of horse shows, races, art exhibits, and talks, all celebrating the majestic Arabian horse.

Doug Antczak, the Dorothy Havemeyer McConville Professor of Equine Medicine with the Baker Institute for Animal Health, was an invited speaker at the festival, where he presented his research on the genetic relationships between Arabians and other horse breeds.

"Legend has it that the Arabian is the oldest recognized breed of horse," said Antczak. Bred by Bedouin tribes on the Arabian Peninsula, the horse has long been a fixture of Arabian culture. They even appear in rock art dating to 3,500 years ago. The breed has a distinctive "dished face," high tail, and arched neck, and its endurance and toughness make it popular for long-distance racing events.

Doug Antczak. Photo by University Photography

During the workshop component of the festival, Antczak spoke with horse breeders from Saudi Arabia and other countries in the Middle East, Europe, and the U.S. about the basics of horse genetics. He presented his latest research into the genetic and historical relationships between Arabians and other horse breeds. "That project has involved sample collection from horses in the Middle East, Europe, and the United States," he said. "We spent many days of travel in the Middle East meeting horsemen and gaining their trust, often over cups of tea and plates of Arabian dates."

He began the project in 2013 with Samantha Brooks, a former Cornell faculty member and colleague at the University of Florida, when they received funding from the Qatar National Research Foundation to study the genetics of the Arabian horse, dromedary camel, and Arabian oryx. Antczak first gained entry to the world of Arabian horses through his work on lavender foal syndrome, a deadly genetic defect that causes foals to be born with severe neurological problems and a lavender-tinted coat. In 2010, Antczak and Brooks discovered the mutation that causes the syndrome. They also developed a molecular genetic test that is used widely by Arabian breeders to identify carriers of the mutation. This allows breeders to select sires and dams that cannot produce affected offspring, and thus prevent the birth of foals with this lethal inherited disease.

Breeders were very interested to learn the results of Antczak's work on genetic relationships between different horse breeds. "It has already lead to new invitations to speak to other Arabian breeders," said Antczak, "and to potential new collaborations to collect samples from rare strains of Arabians that we haven't studied yet."

# Harry M. Zweig Memorial Fund for Equine Research Awards

## NEW

$37,997 to Jonathan Cheetham for "Accelerating Recovery after Laryngeal Nerve Graft"

$79,006 to Thomas Divers for "Characterizing Tropism and Transmission of Equine Parvovirus-Hepatitis (EqPV-H)"

$55,000 to Alan Nixon for "Next Generation Arthritis Control through Lubricin and IL-1 Receptor Antagonist Overexpression in Carpal Osteoarthritis"

$60,054 to Heidi Reesink for "Intra-Articular Recombinant Lubricin to restore Joint Lubrication and Prevent Osteoarthritis in Horses"

$67,136 to Gerlinde Van de Walle for "The Mesenchymal Stem Cell Secretome against Equine Herpesvirus Type 1 Infections"

$99,314 to Bettina Wagner for "Towards a Neonatal Vaccine against Equine Herpesvirus Type 1 (EHV-1)"

## CONTINUED

$60,083 to Douglas Antczak for "Functional Gene Annotation in the Horse"

$48,899 to Jonathan Cheetham for "Regenerative Approach to Recurrent Laryngeal Neuropathy"

$16,528 to Susan Fubini for "The Relationship Between Obesity and Post-Operative Incisional Infections Following Abdominal Surgery in the Horse"

$49,634 to Heidi Reesink for "Quantitative Computed Tomography and Bone Quality Assessment for the Prediction of Fetlock Breakdown Injuries in Racehorses"

$56,399 to Gerlinde Van de Walle for "Microencapsulated Stem Cells to Promote Wound Healing"

**Pictured above:** Jonathan Cheetham studies horse airways by having a horse run on a treadmill. Photo by University Photography.
**Opposite page:** A horse grazes at the Cornell Equine Park. Photo by University Photography.

# M. Kelly Young named executive director of the Agriculture & NYS Horse Breeding Development Fund

*By the Agriculture and NYS Horse Breeding Development Fund*

The Agriculture and New York State Horse Breeding Development Fund Board of Trustees, which runs the New York Sire Stakes series, unanimously approved the appointment of harness racing expert M. Kelly Young as the Fund's Executive Director.

Young, a fifth-generation participant in harness racing, grew up on a leading horse breeding farm outside Goshen. She most recently served as Deputy Director of Public Policy for the New York Farm Bureau where her legislative and regulatory portfolio has included dairy, forestry, aquaculture and food policy, and access. She has held various positions over the past decade with the Farm Bureau and has expertise in general agricultural matters, which are critical to the Fund's mission to support the state's harness racing industry.

"I am excited to help New York's storied harness racing industry grow and thrive in this new position," said Young. "I thank the Board of Trustees for their confidence in my abilities, and look forward to working with horsepersons, breeders, tracks, fans, and everyone in the industry to uphold harness racing's important place in our state's agricultural sector."

Richard A. Ball, Commissioner of the New York State Department of Agriculture and Markets and Fund Trustee, said, "Kelly's extensive knowledge and understanding of both the Standardbred breeding industry and New York agriculture make her a great fit for this position. I have no doubt that she will be a tremendous asset to the Fund. Her energy, enthusiasm, and focus will help us identify new opportunities for New York's Standardbred farms to prosper and strengthen the overall racing industry for years to come. I am excited to work with her as she takes on this new role."

Barry Sample, Chairman of the New York State Gaming Commission and Fund Trustee, said, "Kelly's knowledge of New York's harness racing is second-to-none, and her dedication to the sport is commendable as evidenced through her career including service on the New York Racing Fan Advisory Council. Her leadership skills are exactly what the industry needs at this exact time and I look forward to continuing to work with her in her new role."

Young previously served as Executive Director for the Harness Horse Breeders of New York State and was Associate Editor of The Horseman and Fair World, where she developed and wrote stories on harness racing.

Young was selected for a 2013 McCloy Fellowship in Agriculture and was a member of Class 13 of the Empire State Food and Agricultural Leadership Institute. She received multiple awards for her writing from the American Farm Bureau Federation, the U.S. Harness Writers Association, and the New York Agricultural Society.

Young is currently a member of the New York State Racing Fan Advisory Council, which provides recommendations to the New York State Gaming Commission on matters related to growing the fan base of horse racing. She also serves as president of the Saratoga Harness Hall of Fame and is a member of the U.S. Harness Writers Association. Young holds a B.A. from Boston University.

Established in 1965, the Fund's primary mission is to promote and preserve agriculture through the promotion of horse breeding and equine research in New York state. It directs the state's premier harness racing program designed to stimulate the breeding, buying, and racing of Standardbred horses in New York. Because of the state's unparalleled breeding industry, there is strong international demand for New York-bred Standardbred horses.

The Fund provides assistance to county agricultural societies to maintain and repair racing facilities and also contributes to both the 4-H program and the Dr. Harry M. Zweig Memorial Fund for Equine Research at Cornell University.

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# Equine stem cells rein in bacteria

*By Melanie Greaver Cordova*

Researchers are exploring the use of stem cells to treat skin wounds in horses with techniques that may eventually translate to human patients.

Rebecca Harman '92, M.S. '11 and a team of researchers in the Van de Walle Lab at the College of Veterinary Medicine are finding that factors secreted by adult stem cells, also known as mesenchymal stomal cells (MSC), are able to fight bacteria commonly found in skin wounds.

Bacteria often complicate the treatment of chronic skin wounds in humans, driving a need for new therapies that reduce bacteria in wounds. Although previous research has explored the therapeutic value of MSC in wound healing, few studies examined the potential for MSC to inhibit bacterial growth. Harman and the Van de Walle Lab are examining the antibacterial properties of equine MSC secreted factors, like antimicrobial peptides, to develop therapies for horses and to serve as a model for human studies.

"This equine skin wound healing model offers a readily translatable example for MSC therapies in humans," said Harman, a research support specialist at the Baker Institute for Animal Health. "Although mice are smaller and less expensive model organisms, the horse is more physiologically relevant when it comes to human skin wound healing."

MSC are already commonly used as a biologic therapy for equine joint and tendon injuries. Practitioners isolate MSC from bone marrow and inject them at injury sites. However, bone marrow extraction is an invasive technique, and injection-site complications – such as immune responses against the MSC – may reduce the efficacy of the therapy.

Harman's work sidesteps these issues by isolating MSC from blood rather than bone marrow, making the collection of MSC less invasive. In addition, she is applying the factors secreted by MSC, rather than the cells themselves, to the wounds, which reduces the chance of a host immune response against the therapy.

MSC provide a range of benefits that extend beyond those of conventional antibiotics. Their secreted antimicrobial peptides can directly inhibit the growth of bacteria in skin wounds, and other secreted factors fight bacteria indirectly by attracting resident immune cells that are primed to clear pathogens.

By further experimenting with different delivery methods, the lab's research may make things easier on the practitioner as well. The antimicrobial peptide molecules secreted by MSC have proven to be fairly stable and can maintain their activity through a variety of conditions, such as extended freezing or being dried into a powder. This enables long-term storage options that are more efficient for practitioners than having to isolate MSC and collect secreted factors every time a wound needs to be treated.

The research team will soon partner with Bettina Wagner, chair of Population Medicine and Diagnostic Sciences at the College, to begin in vivo testing on her equine herd. Every summer, Wagner's Icelandic horses naturally develop skin wounds as part of an allergic reaction. The wounds will be directly treated with the secreted factors of MSC and the lab will monitor bacteria levels over time to see if there are differences between treated and untreated wounds.

"What we learn from the Icelandic herd about the wound-healing properties of MSC secreted factors could reasonably be tested in human medicine," said Harman.

Rebecca Harman. Photo by Rachel Philipson

# Longtime committee member steps down

Paul Mountan DVM is stepping down this year from the Harry M. Zweig Committee. After 22 years of service, Mountan has made his mark on the committee with his leadership and dedication.

Members of the committee had nothing but praise for Mountan and expressed gratitude for his work over the years.

"No way to replace an old-school, wise, and practical veterinarian with a golden touch on this committee. Your presence will be missed!" said Gabriel Cook.

"In your long and storied career, you have really helped support the health and welfare of horses through your work as a practitioner, your influence in the sport horse arena, and your committee work for the Zweig Fund. Salut!" said Ann Dwyer. Patricia Wehle seconded these comments.

"You are a great mentor and will be missed," said Robert Tugel.

"Paul has been a standout member of the Zweig Committee for a long, long time. We will miss him!" said William Wilmont, with which Brian Zweig wholeheartedly agreed.

Pictured on this page: Paul Mountan DVM after a trail ride with Theresa, a retired Argentine polo mare he adopted from her former owner after her eye was injured in a polo game. Photo provided.

ZWEIG 40 years CORNELL

*To mark the 40th anniversary of the Zweig Fund and its partnership with Cornell University, we have many events planned for late 2019, including research presentations by faculty supported by the fund, lectures on other equine-related research topics, a tour of the new facilities, and a reception to commemorate the event.*

*Wednesday, November 13, 2019*
*Cornell University College of Veterinary Medicine | Ithaca, New York 14853*


# Zweig Memorial Trot 2018

Vernon Downs, Vernon, N.Y. | Friday, August 17, 2018 | Post time: 6:45 p.m.

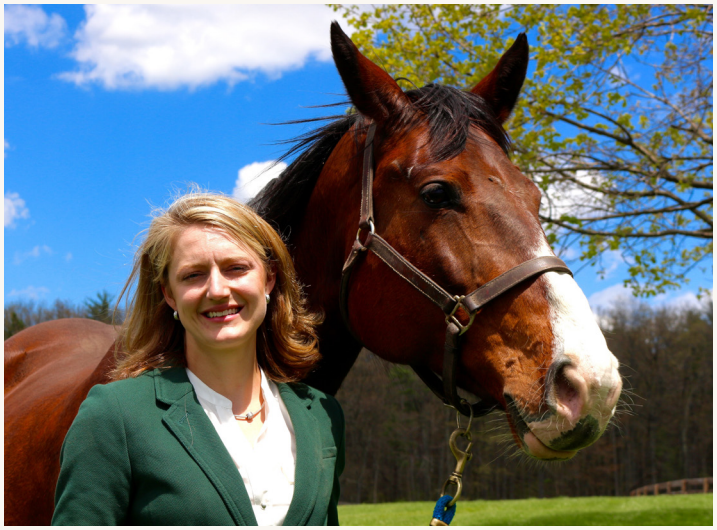
Information: 1-877-888-3766 | www.vernondowns.com/racing

# Project pioneers equine osteoarthritis therapy

*By Elvina Yau and Melanie Greaver Cordova*

Heidi Reesink is doing big-picture research at a microscopic level to improve equine joint health. "Horses are often confronted with bone and joint injuries that have limited treatment options," said Reesink, assistant professor at the Cornell University College of Veterinary Medicine. "I want to develop therapies that will restore joint function and prevent the development of career-ending arthritis."

Zweig awarded funds to Reesink for her study "Intra-Articular Recombinant Lubricin to Restore Joint Lubrication and Prevent Osteoarthritis in Horses," which will determine if recombinant lubricin is an effective therapy for horses. Naturally produced by the body, lubricin, a sugar-coated glycoprotein, helps lower friction in joints and protects cartilage. Reesink's project looks to use recombinant lubricin – lubricin produced in the lab – in treating joint inflammation and lameness.



"We're working on ways to restore effective joint lubrication through sugar-rich lubricating molecules," she said in reference to the glycoprotein. Lubrication is paramount to joint health; joint fluid nourishes and preserves cartilage structure for decades. Cartilage and bone injury can lead to lameness, which is the leading cause of retirement in horses. Around 60 percent of those cases are due to arthritis, said Reesink.

The two-year study will analyze how hyaluronic acid and recombinant lubricin behave in horses. "My long-term objective is to translate lubricin therapy to equine clinical patients," said Reesink.

Although results have been promising in rodent and mini pig models, this therapy has yet to be tested in horses, in part due to technical challenges associated with producing sufficient quantities of recombinant lubricin in the lab for such a large animal, and because of competing demands for testing recombinant lubricin in human clinical trials.

"As a large animal orthopedic surgeon and clinician scientist, my objective is to translate promising basic research from the bench to stallside," said Reesink. Her work has been largely interdisciplinary, and she's collaborated with researchers across the university, specifically chemical and biomedical engineers, to apply engineering principles to the field of osteoarthritis and joint lubrication. She currently runs an additional Zweig-funded study to leverage these collaborations in a project called "Quantitative Computed Tomography and Bone Quality Assessment for the Prediction of Fetlock Breakdown Injuries in Racehorses."

*Photos by Elvina Yau*

## IN CASE YOU MISSED IT

Lecture Schedule at the 9th Annual Harry M. Zweig Memorial Fund for Equine Research Poster Session & Talks, held on Wednesday, November 29, 2017:

- How Zweig Funding Changed Accepted Paradigms in Regenerative Medicine (Lisa Fortier, James Law Professor of Large Animal Surgery)
- The Mesenchymal Stem Cell Secretome in Equine Wound Management (Gerlinde Van de Walle, Harry M. Zweig Assistant Professor in Equine Health)
- Regenerative Approaches for Recurrent Laryngeal Neuropathy (Jonathan Cheetham, Associate Professor, Section of Large Animal Surgery)
- Equine Biobanking - Paving the Way to Personalized Medicine (Marta Castelhano, Director of the Cornell Veterinary Biobank)
- Vaccination of Horses with Lyme Vaccines for Dogs Induces Short-Lasting Antibody Responses (Cassandra Guarino, Extension Associate)

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# Welcome to our new Zweig Committee members

**Chad Brown**

While in high school, Brown got his first experience around horses working with standardbred trainer Paul Kelley. While at Cornell University, Brown worked summers for Hall of Fame trainer Shug McGaughey. While intending to undertake veterinary studies - even interning with Steve Allday - he instead took fulltime employment with Hall of Fame trainer Bobby Frankel following graduation.

Brown opened his own racing stable in November 2007. In ten years, Brown has achieved over 1,000 wins including eight Breeders Cup races and the 2017 Preakness Stakes. This year's Preakness success follows a career year for Brown. In 2016 he was the leading trainer in America by earnings and number of graded stakes wins, and earned the Eclipse Award for Outstanding Trainer.

**M. Kelly Young**

In the two decades since she graduated from Boston University with a biology degree, Young has worked on horse or farming issues. She first started as an Associate Editor with The Horsemen and Fair World, before moving on to the position of Executive Director of the Harness Horse Breeders of New York State. For the last decade, Young has been affiliated with the New York Farm Bureau, working up to her most recent position as Deputy Director of Public Policy from Associate Director of Communications and Senior Associate Director for National Affairs.

Young is a three-time winner of the John Hervey Award for Journalistic Excellence, awarded by the United States Harness Writers Association and is presently the President of the Saratoga Harness Hall of Fame. Young is a fifth-generation horseperson and grew up on Castleton Farm of New York, a Standardbred breeding farm located just outside Goshen, NY.

**Visit us online**
vet.cornell.edu/zweig

Our site provides information on the projects and publications resulting from the Zweig Memorial Fund, and demonstrates the objectives of the Fund in promoting equine health in the racing industry. The Zweig News Capsule is published twice a year, and can be downloaded at bit.ly/ZweigNews. Please encourage other equine enthusiasts to visit the site.