PHI ZETA RESEARCH DAY

Kansas State University College of Veterinary Medicine

EXPLORING SCIENCE. ADVANCING MEDICINE.

Phi Zeta Research Day celebrates scientific inquiry and discovery within the College of Veterinary Medicine. The event showcases innovative research from students, interns, residents, and faculty, creating a space to share knowledge, exchange ideas, and foster collaboration. Through oral and poster presentations, participants highlight work that advances veterinary medicine and strengthens scientific understanding in support of animal and human health.

Innovation, Discovery, and Veterinary Scholarship.

Phi Zeta Sigma Chapter

Phi Zeta RESEARCH DAY
KANSAS STATE UNIVERSITY

March 3, 2026

The Sigma Chapter of Phi Zeta, est. 1969

12:00 pm – 12:50 pm	*Plenary Session*
BI Auditorium	*Welcome and Introduction of the Keynote Speaker* Phi Zeta President, Nicolette Cassel, BSc BVSc MMedVet (Diagnostic Imaging) ***Keynote Speaker*** Joseph Bartges, DVM, PhD, DACVIM, SAIM (nutrition), ACVNU (founding member) "Livin' – Lovin' – Learnin' – What a long strange trip it's been."
1:00 pm – 2:30 pm	*Oral Research Presentations*
BI Auditorium	Applied Science – Large Animal
201 Trotter Hall	Applied Science – Small Animal
301 Trotter Hall	Basic Science
2:30 pm – 3:15 pm	*Royal Canin Poster Session*
BI Atrium	*Basic Science, Applied Science*
3:15 pm – 5:00 pm	*Oral Research Presentations*
BI Auditorium	Applied Science – Large Animal
201 Trotter Hall	Applied Science – Small Animal
301 Trotter Hall	Basic Science & Clinical Case Reports
5:00 pm – 5:45 pm	
BI Atrium	*Reception*
5:45 pm	*Awards Ceremony* Initiation of New Members to Phi Zeta Awards Recognizing Research and Scholarship Accomplishments Closing Comments

Phi Zeta
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KANSAS STATE UNIVERSITY

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Phi Zeta RESEARCH DAY
KANSAS STATE UNIVERSITY

March 3, 2026

The Sigma Chapter of Phi Zeta, est. 1969

APPLIED SCIENCES: LARGE ANIMAL AND LARGE EXOTICS – BI AUDITORIUM 8

Applied Sciences: Large Animal and Large Exotics – BI Auditorium

Phi Zeta RESEARCH DAY
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BI Auditorium 1:00 pm – 1:30 pm

Rebecca Bigelow

Influence of Age and Contact Status on BRD Treatment Risk Between Adjacent Hutch Calves Within a Commercial Calf Ranch

BI Auditorium 1:00 pm – 1:30 pm

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Keywords: calf health; pre-weaned calves; respiratory disease; hutch proximity; calf management

Intro/background: The health status of pre-weaned calves can impact future productivity and performance. Etiology and management practices have been studied for bovine respiratory disease (BRD), yet less is known about the impact of contact with diseased calves in close proximity. This study aimed to determine morbidity risk of calves within 12 days of an adjacent calf being treated for BRD.

Methods: This prospective observational study was conducted for 11 months at a commercial High Plains calf ranch. Electronic identification (EIDs; n = 9,676 calves) and hutch location were recorded and linked to health records. Daily calf-level records were generated, and respiratory treatments were identified to determine contact status based on adjacent calf treatments. Calves without an adjacent calf being treated in the previous 12 days were classified as “non-contact” calves. Calves with an adjacent calf treated in the prior 12 days were classified as “contact” calves. A generalized linear mixed model estimated the daily probability of treatment by contact status, age (categorized in 30-day periods), and the potential interaction among age and contact status with calf ID included as a random effect.

Results: The interaction between contact status and age significantly affected the daily probability of treatment ($P < 0.001$). The magnitude of daily risk of BRD treatment based on contact status varied by calf age with lower risk in calves < 30 days old (4 vs. 2 cases per 1,000 calf-days, contact vs. non-contact, respectively) compared to calves at 30-59 days (13 vs. 8), 60-89 days (23 vs. 7), and ≥ 90 days (10 vs. 7). Daily treatment risk increased ($P < 0.001$) if an adjacent calf was treated within 12 days, and the magnitude of difference varied with calf age.

Conclusion: Proximity between hutches may play an important role in risk of BRD and should be considered in management and prevention strategies.

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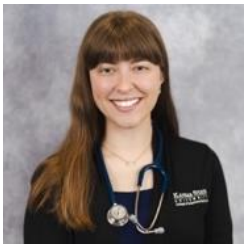
BI Auditorium 1:15 pm – 1:30 pm



Emily Evans-Stevens

Evaluation of Sanitation Procedures in Swine Nursery Facilities Utilizing ATP Bioluminescence

BI Auditorium 1:15 pm – 1:30 pm

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Keywords: Swine; nursery; sanitation; adenosine triphosphate bioluminescence

Enterotoxigenic Escherichia coli F18 (ETEC) is of high concern to the swine industry as it causes postweaning diarrhea (PWD). Environmental persistence of ETEC in poorly sanitized facilities can lead to reinfection of successive litters. Traditionally, sanitation assessment has relied on subjective visual inspection. Adenosine Triphosphate (ATP) bioluminescence offers a rapid, inexpensive, and sensitive method for evaluating cleanliness by detecting residual biological material. However, its accuracy may be affected by disinfectants. Quaternary ammonium compounds (QACs) may cause false positives, while hydrogen peroxide-based products can suppress signals ("quenching"), potentially leading to false negatives.

This study had two objectives. The first was to evaluate surface ATP concentration as a marker of sanitation efficacy using four different sanitation protocols. The second objective was to identify a superior protocol based on ability to effectively remove biological contamination following exposure to Enterotoxigenic Escherichia coli F18.

This study was conducted at a BSL-2 swine nursery facility following an in vivo ETEC challenge study. Forty-four pens, previously housing ETEC-inoculated nursery aged pigs, were assigned to one of four sanitation treatments: 1) Hot water (HW, 138.6°F) + Synergize (1:256; Neogen Corporation, Lansing, MI); 2) Cold water (CW, 73.8°F) + Synergize; 3) HW + Virkon S (1:128; Lanxess Corporation, Pittsburgh, PA); and 4) BarnStorm (1:64; Preserve International, Zepher Cove, NV) pre-treatment + HW + Synergize. Treatments were applied in a randomized complete block design (11 pens per treatment). ATP concentrations were measured before and after sanitation at five standardized pen locations using UltraSnap swabs and Hygenia SystemSURE/EnSURE Luminometer (Hygenia, Camarillo, CA). Statistical analysis was conducted using lme4 package in R, with treatment and location as fixed effects, and pen, cleaning day, and ETEC dose as random effects. Significance was declared at $P \leq 0.05$.
No significant treatment × location interactions were found, indicating treatment efficacy was consistent across locations. After sanitation, HW + Virkon S resulted in lower ($P < 0.001$) residual ATP levels and the greatest reduction in ATP compared to all other treatments. The feeder lip retained the highest residual ATP after cleaning, while the corner wall plastic divider retained the least ($P < 0.05$). ATP reductions were highest in the corner wall, followed by intermediate reductions on the feeder exterior and corner flooring, and lowest reductions on the feeder lip and center flooring.

Based on these results, practitioners should pay special attention to cleaning hard-to-reach or complex surfaces like feeder lips, where biological residues are more likely to persist. The ATP bioluminescence assay should be considered as a rapid assessment tool, while being cautious of potential interference from disinfectant residues (e.g. quenching or false positives).

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BI Auditorium 1:30 pm – 1:45 pm

Larrison Hicks

Title Effect of Direct-fed Microbial on Growth Performance and Health Outcomes in Beef-Dairy Calves in the First Three Weeks of Life

BI Auditorium 1:30 pm – 1:45 pm

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Keywords: Direct fed Microbial; Health; Growth; Beef-dairy Cross Calves

Introduction
Early-life morbidity increases costs and labor demands that can impact long-term calf well-being. Intestinal barrier function and immune function influence the risk of developing diseases, respiratory and digestive, that impact growth and health. Lactic acid-producing bacteria could help during this period by altering intestinal microflora. The purpose of this study is to evaluate the effect of lactic acid-producing bacteria on calf growth and health outcomes (respiratory and pathogenic diarrhea) in the first three weeks of age in beef-dairy cross calves.

Methods
Beef-dairy cross calves (N = 300) were enrolled in the study between May and June 2025, at a Kansas dairy. Calves were assigned randomly to receive a direct-fed microbial (DFM) or a negative control (no DFM). The DFM (BactaShield; Lactobacillus acidophilus and Lactobacillus casei; Legacy Animal Nutrition Inc.) was delivered via the milk and the starter feed; each containing (1 x 10^9CFU/animal/day). Calves were weighed at birth and at three weeks of age. Fecal scores were collected once daily using the Wisconsin fecal scoring system (0 = normal : 1 = semi-formed : 2 = runny : 3 = watery). All calves were diagnosed (either respiratory illness or diarrhea) and treated by dairy personnel according to in-house protocols. The study design was a prospective randomized control trial. Data for body weight and average daily gain were analyzed using a linear mixed model with fixed effects of treatment, age, and treatment by age interaction in R statistical software. Data for health outcomes (respiratory disease and neonatal diarrhea) were analyzed as a binary logistic regression mixed model with fixed effect of treatment. Fecal score data were analyzed as a binary logistic regression model with treatment, age, and treatment by age interaction. Models with repeated data (body weight and fecal score) include calf as a random effect. Calves that died within the first three weeks were omitted from the analysis.

Results/Conclusion
Direct-fed microbial had no effect ($P > 0.05$) on body weight or average daily gain. The number of calves treated for diarrhea was 117 for the DFM group compared to 112 calves for the non-DFM group, with no observed difference ($P > 0.05$). Respiratory treatments were not different ($P > 0.05$) between DFM and non-DFM calves, with each group having 13 cases. The probability of abnormal fecal scores increased ($P > 0.05$) with age, but there was no difference between DFM and non-DFM calves. Overall, lactic acid producing bacteria had no effect on growth performance and health outcomes in beef-dairy cross calves within the first three weeks of life.

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BI Auditorium 1:45 pm – 2:00 pm

Madeline Mancke

The Effect of Shade on Steer Performance after Terminal Sort

BI Auditorium 1:45 pm – 2:00 pm

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Keywords: feedlot; heat mitigation; heat stress; shade; steers

Background

Heat stress occurs when total environmental and metabolic thermal load exceeds an animal's ability to dissipate heat, leading to potentially negative impacts on performance and welfare. Access to shade has been associated with reductions in body temperature and respiratory rate in cattle, but previous findings on cattle performance has been inconsistent.

Methods

This pen-level randomized controlled trial evaluated the effects of shade (30 ft²/head, 100% solar block; n= 12 pens) versus no shade (n= 12 pens) on steer performance following terminal sort at a commercial feedyard in the Pacific Northwest, where animals were housed on roller-compacted concrete floors. Data collected included water consumption, feed delivery, disease events, open mouth breathing (OMB) behavior, and carcass traits.

Results

Shade reduced water consumption by 15% ($P<0.05$), and pens without shade showed a 6% decrease in feed delivery when the previous day's temperature-humidity index (THI) was ≥80 ($P<0.05$). Additionally, shade significantly reduced OMB behavior ($P<0.05$), the number of culled steers ($P<0.05$), and the incidence of dark cutters ($P<0.05$). A greater proportion of steers in shaded pens graded choice and prime compared to those in unshaded pens (93% and 91%, respectively; $P<0.05$).

Conclusion

The inclusion of shade improved animal health, welfare, carcass quality, and environmental stewardship during the summer months in a commercial feedyard setting. Shade should be considered a potentially valuable management tool for mitigating heat stress in finishing cattle.

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BI Auditorium 2:00 pm – 2:15 pm

Audrey Matheny

Generation of Cross-Neutralizing Antibodies Against Japanese Encephalitis Virus Following West Nile Virus Vaccination in Domestic Swine

BI Auditorium 2:00 pm – 2:15 pm

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Keywords: Japanese encephalitis virus; vaccine; swine; antibody

Swine are a significant agricultural species in the United States (U.S.) with projected cash receipts for 2025 to total approximately $30 billion. Stability of this industry relies on strong biosafety and biosecurity measures to protect animal health against foreign animal diseases. Japanese encephalitis virus (JEV) is a zoonotic vector-borne orthoflavivirus that can cause severe illness in humans and swine with signs ranging from mild and flu-like to fatal encephalitis. Whilst the virus is endemic throughout Central and Southeast Asia, in 2022, JEV caused a major outbreak in southern Australia that affected 80% of the country's swine farms and led to 3 - 6% production losses on affected farms due to JEV induced reproductive disease. Although there are no JEV vaccines for swine available in the U.S., there are commercial veterinary vaccines for a closely related virus: West Nile virus (WNV). To assess potential cross-protection elicited by commercially available WNV vaccines against JEV, 3-week-old domestic swine were vaccinated with one of three commercially available equine vaccines selected for evaluation following the manufacturer's recommended dosing regimen for juvenile horses: a primary dose followed by a booster dose 3 weeks later. Mock vaccinated piglets were included as negative controls. Piglets were monitored daily for any adverse reactions to vaccination, body temperatures were taken daily and weights were measured bi-weekly. Serum samples were collected from piglets prior to primary vaccination and bi-weekly starting on day 7 post primary vaccination until study termination 35 days after primary vaccination. Serum was evaluated for presence of JEV neutralizing antibodies using plaque reduction neutralizations tests (PRNTs). Serum samples from vaccinated piglets showed JEV neutralizing antibody titer levels above 1:10 indicating potential protection against JEV infection. All vaccines evaluated elicited generation of cross-neutralizing antibodies in vaccinated piglets. Mock vaccinated piglets showed JEV neutralizing antibody titers below the 1:10 threshold indicating no potential protection against JEV infection. These results demonstrate the evaluated commercially available WNV veterinary vaccines may generate cross-protective immunity against JEV in domestic swine. These vaccines, if shown to be efficacious following JEV infection, could serve as important tools for outbreak control in the event of a JEV introduction to the U.S.

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BI Auditorium 2:15 pm – 2:30 pm

Liliana Rivas

Optimal Sampling Time for Indigestible Sugar Markers of Gastrointestinal Permeability in Yearling Cattle

BI Auditorium 2:15 pm – 2:30 pm

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Keywords: Biomarker; ruminant; serum; gas chromatography

Gastrointestinal permeability refers to an animal's ability to regulate the passage and absorption of nutrients, toxins, and even pathogens from the lumen of the gut into the bloodstream. In veterinary research, a method used to assess gastrointestinal permeability involves using non-metabolizable sugars (e.g. lactulose) as serum biomarkers; however, there is uncertainty surrounding persistence of these biomarkers in the rumen. Objectives of this study were to assess the suitability of lactulose and sucralose, a novel sugar, as biomarkers of gastrointestinal permeability and to determine appropriate blood sampling time for peak concentrations in functioning ruminants. Three yearling Holstein steers (290 ± 7.86 kg) were individually fed standard grass hay, ad libitum, for 4 days before being restricted to 25% of their ad libitum intake from days 5-9 to induce gastrointestinal permeability. On day 9, an oral dose of lactulose and sucralose (0.4 mg/ kg of body weight) was given and blood sampled at time 0 and every 3 hours until 36 hours post dosing. Serum was extracted and analyzed for sugar concentration by gas chromatography- tandem mass spectrometry. Empirical means and residual standard deviations of lactulose and sucralose concentrations were calculated at each time point and a Monte Carlo simulation was conducted to estimate the probability of each sugar exceeding the limit of quantification (LOQ). The predicted probabilities revealed that at three hours both sugars had high probabilities of exceeding LOQ (Lactulose= 0.805, Sucralose= 0.905). In addition, the probability of sucralose being detected above LOQ was higher over the entire time period of 36 hours compared to lactulose (Lactulose= 0.857, Sucralose= 0.901). In conclusion, the probability of sucralose concentration exceeding LOQ was consistently higher than lactulose with time points 3 to 33 hours being equally acceptable time points for blood sampling.

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BI Auditorium 3:15 pm – 3:30 pm

Jacob Schumacher

Comparing the Use of Lidocaine-Loaded Castration Bands to Reduce Pain in Dairy and Beef-Dairy Calves

BI Auditorium 3:15 pm – 3:30 pm

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Keywords: castration; pain; welfare; cattle

Introduction
Introduction of greater number of beef-dairy calves entering the United States beef market continues to highlight the importance of pain mitigation during castration.

Materials and Methods
This randomized blinded controlled trial aimed to evaluate pain mitigation and behavior in beef-dairy (BOD) and Holstein (HOL) calves castrated with either lidocaine-loaded (LLB) or standard bands (CON). Thirty-eight bull calves weighing 83 ± 17 kg were randomly assigned to 1 of the following 2 treatment groups; castrated with lidocaine-loaded bands (BOD-LLB, n = 9; HOL-LBB, n = 9), or castrated with a standard control band (BOD-CON, n = 10; HOL-CON, n = 10). Calves' performance, lying and standing activity, kinetic gait analysis, plasma cortisol, and behavior were analyzed at multiple timepoints. Linear-mixed effects models were used to determine potential associations between outcomes and treatment, breed, time, and potential interactions.

Results
Lying and standing activity outcomes showed a significant effect of breed for the number of standing bouts per day, with the BOD group transitioning from lying to standing more frequently than the HOL group (18.3 and 17.3 bouts/day, respectively; SEM=0.49; P=0.03). Gait outcomes showed a treatment × time interaction for gait velocity and gait distance, with the CON group increasing over time. However, no gait outcome differed between LLB and CON groups at any timepoint. Neither treatment or breed had an effect on plasma cortisol, performance, or video monitoring behavior outcomes (P > 0.05).

Conclusions
Overall, this study was unable to find differences in pain outcomes between calves that were castrated with either a lidocaine-loaded or standard band.

Phi Zeta RESEARCH DAY
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BI Auditorium 3:30 pm – 3:45 pm

Brittany Smith

The Effect of Loud Versus Quiet Visitors on Chimpanzee Welfare Measured by Behavior

BI Auditorium 3:30 pm – 3:45 pm

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Keywords: Chimpanzee; welfare; noise; zoo; visitors

Chimpanzees in zoos are exposed to an unpredictable variation of noise levels. The research goal was to assess chimpanzee welfare measured by behavior in response to loud and quiet zoo visitors. It was hypothesized that when exposed to a loud group, chimpanzees would display more negative welfare indicators, such as decreased locomotion and abnormal behaviors (e.g. coprophagia). The experimental and observational unit was individual chimpanzees (n=5). Treatment groups were composed of 5 to 10 children ages 8 to 13. Chimpanzees were recorded for 5 observation days with randomized order of treatments. The Loud group's noise averaged 99.45 ± 3.73 dB while the Quiet group averaged 48.74 ± 3.41 dB. Results showed no effects of treatment, order, or their interaction on probabilities of climbing, using enrichment, or foraging at least once during the treatment period ($p<0.05$). No aberrant activities were observed. The youngest male chimpanzee had a greater predictive probability of foraging at least once during a treatment period compared to the oldest female ($p<0.05$). Changes in behavior such as percentage of time spent outdoors, moving, hiding and probability of using enrichment were likely associated with increased morning activity once they had full access to the enclosure. Overall, chimpanzee welfare was not negatively impacted by loud or quiet zoo visitors and displayed no interest in noise when given the opportunity to forage or use enrichment. Future studies may evaluate the association of chimpanzee welfare with chimpanzee sex and time
of day.

Funding: Boehringer Ingelheim Veterinary Scholars Program and Bortoluzzi Lab internal funds

Phi Zeta RESEARCH DAY
KANSAS STATE UNIVERSITY

BI Auditorium 3:45 pm – 4:00 pm

MaRyka Smith-Weishaar

Evaluating Electronic Identification for Cattle Traceability Through Analysis of Simulated Movement Networks

BI Auditorium 3:45 pm – 4:00 pm

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Keywords: Foot and Mouth Disease; Cattle; Traceability; Electronic Identification

Control of a Foot-and-Mouth Disease outbreak in the United States (US) will require animal health officials to trace the infected and at-risk animals. Infected animals can be infectious in as few as 2 to 3 days which makes rapid tracing imperative. Electronic identification (EID) and digital tracing are likely needed, however the current US animal traceability system does not require all cattle to have EID nor report all movements to a database. This project's objective was to evaluate the traceability of farms in simulated movement networks with various levels of digital tracing implementation.
InterSpread Plus was used to generate a complete 30-day national network of cattle movements. In the network, farms are nodes, and animal movements are edges. A random sample of 100 cow-calf farms, 100 markets, and 100 feedlots were chosen as farms of interest for tracing.

To create digitally traceable networks, nodes were randomly assigned to tag or not to tag using an estimated baseline level representing the current US beef cattle EID tagging rate (1-2%). Nodes were similarly assigned to report movements at the baseline (1-5%). Multiple scenarios of improved tagging and reporting at the cow-calf, market, feedlot, or stockers and dealer level were simulated. An estimated "ideal" level of improved digital tracing with 75% tagging and 75% digital reporting was also tested. For all scenarios, dairy cattle farms are 95% likely to tag and 90% likely to digitally report movements. For a movement to be included in the traceable network, the source node must tag, and the destination node must report.

The complete cattle movement network had approximately 59,000 daily movements. The baseline tagging network had approximately 1,900 daily movements represented. The first improved level had 17,000 daily movements represented. The ideal level had approximately 41,000 daily movements represented.

In the baseline level, there were 0 traceable farms of interest across all of the farm types tested. The first improved level had 14.5% of cow calf contacts found, 17.5% of market contacts, and 4.6% for feedlot contacts. As the level of tagging and reporting increased, the number of traceable farms of interest increased. With the highest level in the ideal level of tagging with 48.8% of contacts for cow-calf farms were traced, 56.4% for markets, and 43.2% of feedlot contacts. These results show the need for increased EID tag usage and movement reporting for effective tracing. The untraceable contacts may cause delays in tracing the at-risk farms which will ultimately delay and undermine the efficacy of control efforts.

Phi Zeta RESEARCH DAY
KANSAS STATE UNIVERSITY

BI Auditorium 4:00 pm – 4:15 pm

Isabell Stamm

Hepatic Ultrasonography as a Chute-side Diagnostic for Cardiac Remodeling in Feedlot Animals

BI Auditorium 4:00 pm – 4:15 pm

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Keywords: ultrasound; hepatic; feedlot; cattle; cardiac

Background – Ultrasonography is a potentially useful chute-side diagnostic tool for hepatic congestion secondary to cardiac remodeling. Cattle with hepatic congestion may be more likely to have poor outcomes.

Hypothesis/Objectives – To determine potential associations between risk for retreatment or death with caudal vena cava diameter (CVC), portal vein diameter (PV), hepatic size and echogenicity.

Animals – Enrolled feedyard cattle (n=334) identified with clinical pneumonia.

Methods – This cross-sectional observational study evaluated cattle at first or third pneumonia treatment, performing a 60 second trans-abdominal hepatic ultrasound. Measurements of PV, CVC, echogenicity, and hepatic size were conducted post-hoc. Demographic information was recorded at enrollment. After a 60-day observation period, treatments outcomes including risk for retreatment (RTx) and did not finish (DNF) were collected.

Results – In cattle at first treatment, hepatic echogenicity was positively associated with increased risk for DNF (p=0.03) when animals were late in the feeding period (>42 days); however, cattle in the early feeding period did not see this association. Hepatic size was positively associated with increased risk for retreatment when animals were late in the feeding period (p=0.02). Lastly, risk for retreatment increased concurrently with CVC diameter (p=0.014). In third treatment animals, no significant associations were identified between ultrasound measurements and treatment outcomes.

Conclusions and Clinical Importance – These results indicate hepatic ultrasound findings are associated with negative treatment outcomes within 60 days of assessment in cattle at first pneumonia treatment. Further evaluation of the usefulness of ultrasonography in feedlot cattle for early diagnosis of cardiac remodeling must be studied.

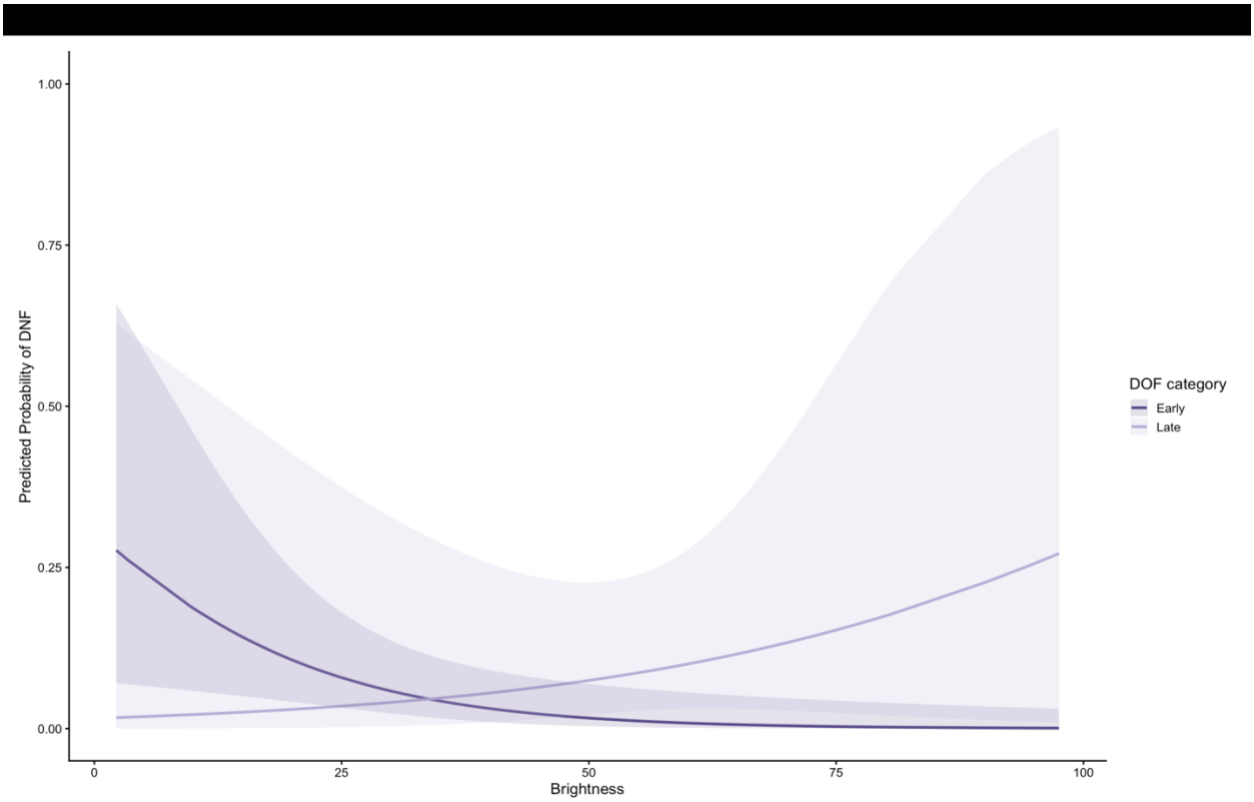

Figure 1. Predicted probability of hepatic echogenicity (mean gray value, AU) on did not finish (DNF) in first treatment cattle differentiated by early (<42) and late (>42) days on feed (n = 304). The solid lines depict the model-predicted means, shaded regions indicate the 95% CI for each category.

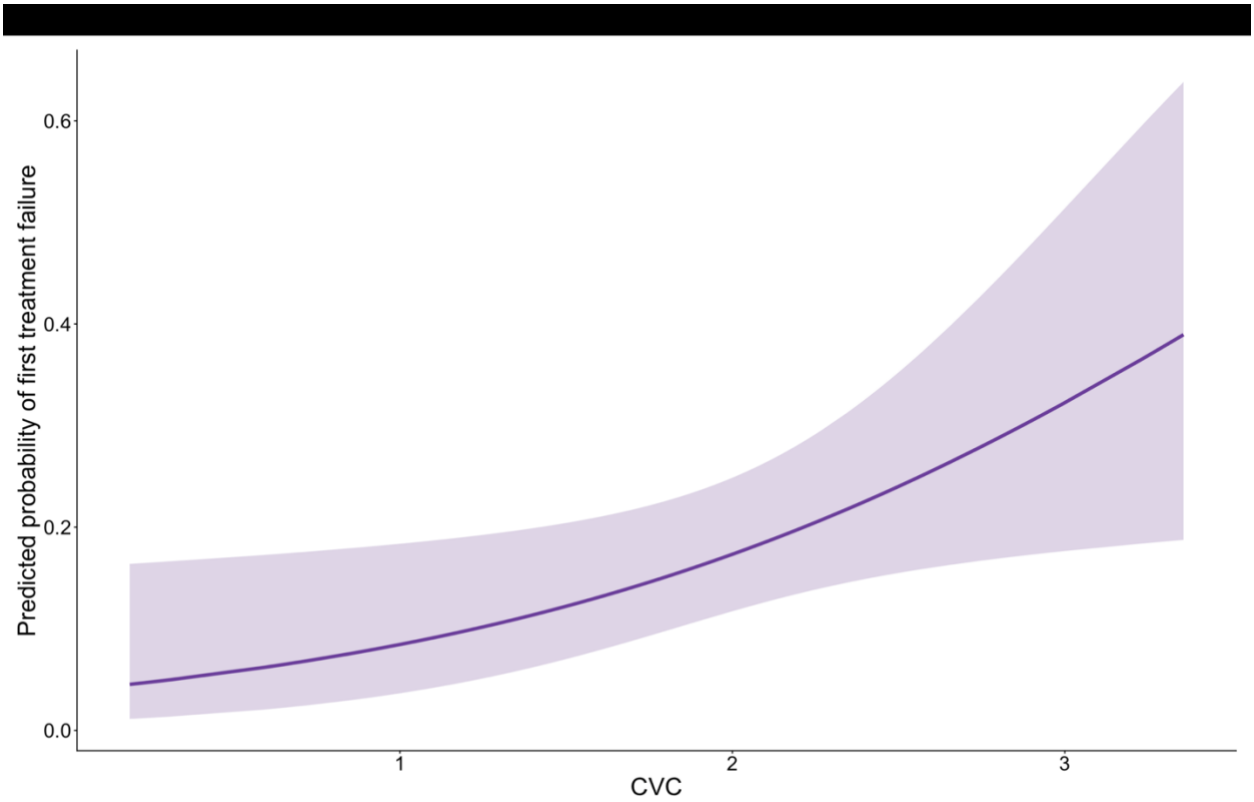

Figure 2. Relationship between risk of retreatment and caudal vena caval diameter (CVC, cm) in first treatment cattle (n = 304). The solid line depicts the model-predicted mean, and the shaded region represents the 95% confidence interval.

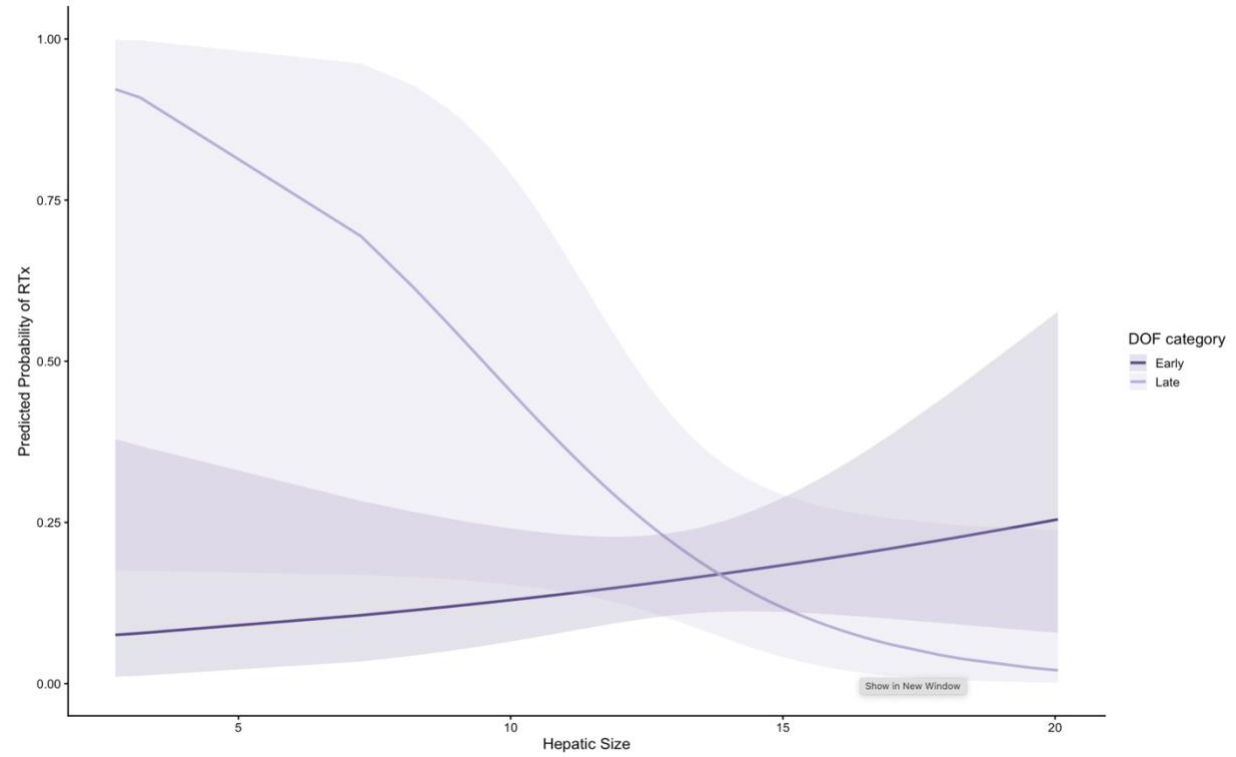

Figure 3. Predicted probability of hepatic size (cm) on risk for retreatment (RTx) in first treatment cattle differentiated by early (<42) and late (>42) days on feed (n = 304). The solid lines depict the model-predicted means, shaded regions indicate the 95% CI for each category.

Phi Zeta RESEARCH DAY
KANSAS STATE UNIVERSITY

BI Auditorium 4:15 pm – 4:30 pm

Caroline Wingert

Factors Associated with Veterinarian Career Satisfaction in Rural Practice

BI Auditorium 4:15 pm – 4:30 pm

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Keywords: Survey; maternity leave; population

The objective of this study was to determine factors associated with self-reported career satisfaction in rural veterinary practice. An anonymous, electronic survey was distributed to veterinary alumni and national/state veterinary associations. Of 841 responses, 376 met inclusion criteria. Inclusion criteria consisted of consent, rurality, and answering any question past the inclusion questions. Respondents who indicated they strongly or somewhat agreed they were satisfied were categorized as satisfied. A multivariable logistic regression model was fitted via manual forward selection to evaluate relationships between satisfaction and other survey variables (p=0.05 required for inclusion). Respondents who indicated their clinic offered maternity leave had higher odds of indicating career satisfaction than those whose clinics did not (OR=3.3,95%C.I.=8.18-372). Proximity to a large city (≥50,000) was associated with career satisfaction and those who lived 26-50 (OR=0.213,95%C.I.=0.04-0.78) and >100 (OR=0.189,95%C.I.=0.036-0.723) miles away had lower odds of career satisfaction compared to respondents who lived <25 miles from a city of >50,000. Respondents who live in cities ≥35,000 had lower odds of career satisfaction (OR=0.167,95%C.I.=0.0267-1.01) relative to respondents who live in cities of <999. Relative to respondents without family in the area, those with local family had lower odds of career satisfaction (OR=0.217,95%C.I.=0.0647-0.723). Respondents that did not charge hourly for herd health services had lower odds of career satisfaction (OR=0.343,95%C.I.=0.145-0.781) compared to respondents that charged hourly. Factors often cited as potential reasons for higher career satisfaction in rural practice were not supported by the data from this study.

Phi Zeta RESEARCH DAY
KANSAS STATE UNIVERSITY

BI Auditorium 4:30 pm – 4:45 pm

Jordana Zimmermann

Evaluating the Use of Bovine Rate of Consumption Index, Intake Metrics, and Bolus Outcomes as Objective Measures of Surgical Castration Pain

BI Auditorium 4:30 pm – 4:45 pm

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Keywords: Animal Welfare; Feeding behavior; Non-invasive objective monitoring; Pain assessment

Pain assessment outcomes in cattle are mostly subjective. Bovine Rate of Consumption Index (BROCI) is the velocity at which an animal eats and may provide an objective indicator of pain.

This study evaluated BROCI, intake metrics and ruminal-bolus outcomes as objective indicators of surgical castration pain.

Commercial bull-calves (n=47, 8–10 months; 281–419 kg) were housed in a pen with eight proprietary scale systems that measured BROCI, feed intake, and total time feeding. Calves were randomly assigned to treatment groups: castration without analgesia (CAST), castration with meloxicam (MEL-CAST), castration with ketoprofen (KETO-CAST), sham-castration without analgesia (SHAM), or sham-castration with meloxicam (MEL-SHAM). A subset of calves (n=25) were administered ruminal boluses that collected activity, ruminal temperature, and water intake. Measures were taken at baseline (-10d to -1d), castration (0d), and 42d post-procedure. Outcomes were analyzed using linear mixed models.

BROCI showed no significant treatment×time interaction, despite a significant timepoint effect (P≤0.001). Feed intake and total-time feeding were lower in CAST on 0d relative to other groups (P<0.01). Water intake was lower in CAST and MEL-CAST than MEL-SHAM on 0d. Bolus activity was lower on SHAM than MEL-CAST on 5d and 6d. Ruminal temperature differed among treatment groups, but all remained within normal physiological range.

This study used objective measures to assess pain and analgesic effects following surgical castration. While BROCI did not differ among treatments, feeding behavior and ruminal-bolus outcomes varied by treatment and time, supporting utility of objective outcomes as castration pain and analgesic effects indicators.

Applied Sciences: Small Animal and Small Exotics – 201 Trotter Hall

Phi Zeta RESEARCH DAY
KANSAS STATE UNIVERSITY

201 Trotter Hall 1:00 pm – 1:15 pm

Mahamudul Hasan

Identifying 'IOLA': A Targeted Approach to Detecting a Novel Pathogen in Atypical Canine Respiratory Cases

201 Trotter Hall 1:00 pm – 1:15 pm

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Keywords: CIRD (Canine Infectious Respiratory Disease); IOLA (Infectious Organism Lurking in Airways); Dual-target PCR

Background: In 2014, a novel bacterium, IOLA (Infectious Organism Lurking in Airways), was identified in respiratory samples from human patients in Japan, raising concerns about its potential role in chronic respiratory disease. This unculturable organism can currently be classified to the bacterial order Rickettsiales and comprises five distinct phylotypes. It exhibits epicellular parasitism, suggesting a previously overlooked pathogenic lifestyle. Monitoring this Rickettsiales-like agent is critical for One Health surveillance, as many members of this bacterial order are zoonotic and the discovery of a novel species in companion animals warrants investigation into potential transmission risks to humans. Recently, several U.S. states reported an unusual increase in cases of atypical canine infectious respiratory disease (aCIRD), in which many affected dogs tested negative for known respiratory pathogens. Concurrently, investigators from the University of New Hampshire reported the detection of an unidentified organism in outbreak-associated canine respiratory samples, further highlighting potential diagnostic gaps. In this context, we investigated the presence of IOLA in affected dogs in unexplained, treatment-resistant respiratory disease during the winter 2023 aCIRD outbreak.

Methods: We developed a PCR assay targeting two IOLA genes *(16S rNA and PrfA)*. The *PrfA* gene was selected as a secondary diagnostic target alongside *16S rRNA* to provide a specific indicator of the organism's pathogenic potential, as this master virulence regulator is essential for the intracellular survival and host-cell infection characteristic of virulent Rickettsiales. Assays were optimized at 58°C with 500 nM primers, and products visualized by agarose gel and capillary electrophoresis. Analytical sensitivity was 10^4 copies/µL and 10^4 copies/µL for *16S rRNA* and *PrfA* assay, respectively. Suspected amplicons were verified by Sanger sequencing and BLAST analysis. The study leveraged KSVDL's NGS surveillance (MDL-0202), which was offered free of charge to help identify unrecognized pathogens. We screened 777 veterinarian-submitted canine respiratory samples from the United States using 16S metagenomic sequencing. Samples identified by 16S sequencing contained Rickettsiales, the lowest taxonomic classification for IOLA, were tested by PCR assay targeting two IOLA genes *(16S rRNA* and *PrfA)*.

Results: Of the 777 samples screened, 55 contained sequencing reads classified as Rickettsiales. 45 of the 55 samples were negative by *16S rRNA* PCR assay, while 10 samples produced amplicons near the expected size. The *PrfA* PCR assay was negative across all samples. Sequencing representative samples from those that produced 16S amplicons confirmed nonspecific amplification. Therefore, all 55 samples were negative for IOLA identification by dual targeted PCR.

Conclusions: IOLA was not detected by dual targeted PCR in Rickettsiales contained upper respiratory samples from dogs with suspected aCIRD. The newly developed IOLA 16S PCR assay produces some non-specific amplification. This highlights the importance of using multiple targets when developing a detection method for an unculturable, uncharacterized organism.

Phi Zeta RESEARCH DAY
KANSAS STATE UNIVERSITY

201 Trotter Hall 1:15 pm – 1:30 pm

Hannah Kunzman

Complications Secondary to Castration Technique in Client-Owned Guinea Pigs (*Cavia porcellus*)

201 Trotter Hall 1:15 pm – 1:30 pm

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Keywords: abscess; edema; elective castration; incisional dehiscence; Cavia porcellus; guinea pig

Introduction: Castration is recommended in guinea pigs to facilitate group housing by preventing unwanted pregnancy, minimizing aggression, and to prevent or treat neoplasia. Two approaches for castration have been described in the literature: scrotal and abdominal. Guinea pigs are more prone to post-castration infection than other species, both anecdotally and in laboratory-controlled settings. The objective of this multi-institutional, retrospective study was to identify complications associated with two castration techniques in a clinical setting.

Methods: Medical records were acquired from three veterinary teaching hospitals for guinea pigs undergoing routine castration surgery between September 15, 2014, and September 15, 2024. Records were excluded if the surgery was performed due to an emergency or suspected reproductive disease. Sixty medical records were included in the data analysis.

Results: Sixty routine castrations were identified, with 41 scrotal and 19 abdominal approaches. Fifteen complications were identified, including anesthetic, intra-operative, and post-operative incidences. The anesthesia-related death rate was consistent with previous literature at 5% (3/60). The complication rate for animals that survived surgery was 23% (9/39) for the scrotal approach and 17% (3/18) for abdominal. There were four total intra-operative complications: three (two scrotal, one abdominal) with mild hemorrhage from the pedicle, and one (scrotal) with inadvertent extrusion of the urinary bladder through the inguinal canal. No intra-operative complications resulted in post-operative complications. There were eight post-operative complications: six scrotal approach cases, two with scrotal edema and serosanguinous discharge, and four scrotal abscesses; and two abdominal approach cases with minor incisional dehiscence. There was no significant association between the surgical approach and complication incidence using a Fischer's exact test (p=0.734).

Conclusion: Due to the increased severity of complications associated with the scrotal approach, the results of this study support abdominal castration as the preferred method in guinea pigs in a clinical setting.

Phi Zeta RESEARCH DAY
KANSAS STATE UNIVERSITY

201 Trotter Hall 1:30 pm – 1:45 pm

Jessica Mayer

Comparison of Cage Layering Configurations on Moisture Management in Clinical Patients

201 Trotter Hall 1:30 pm – 1:45 pm

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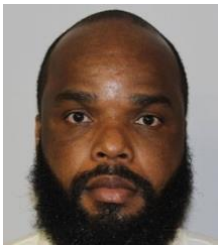
Keywords: Veterinary Nursing Care; Kennel Bedding; Patient Hygiene; Small Animal Hospitalization; Incontienence

Keeping patients dry is a vital, but often overlooked, aspect of care in veterinary medicine. Important aspects include maintaining sanitary surgical sites, IV catheters, and reduction of moisture-associated dermatitis. This study was conducted in two phases. Phase one tested eight configurations, phase two tested six. Configurations tested high- and low-absorbency pads alone, synthetic sheepskin over grate, synthetic sheepskin over pad, towel over grate, and towel over pad with phase one additionally evaluating microfleece blanket over grate and microfleece blanket over pad. Two primary measures were tested, surface area of moisture spread and % moisture (digital moisture meter). MANOVA analysis showed significant differences in cage setup configurations (Wilks' λ=.008, $F(14,174) = 123.14$, $p<.001$, partial $\eta 2$=.908). Following initial evaluation, a simulated patient model was utilized to examine fluid absorption (g) and spread of fluid (cm2). MANOVA analysis showed significant differences (Wilks' λ=.105, $F(10,82) = 17.11$, $p<.001$, partial $\eta 2$=.676). Worst performers were pads alone (11.0-17.9 g and 567.2-718.7 cm2), with the low absorbency pad being the worst overall (17.9 g & 718.7 cm2). Best performance was from synthetic sheepskin configurations, with sheepskin directly touching the pad being the best overall (3.0 g & 129.9 cm2). Veterinarians typically utilize donated materials for cage layering and patient comfort. For patients that are at a higher risk of urinating in kennels, this study demonstrates the importance of kennel materials for small animal patients in a clinical setting, with synthetic sheepskin being optimal.

Student Support: Boehringer Ingelheim Veterinary Scholars Program

Phi Zeta RESEARCH DAY
KANSAS STATE UNIVERSITY

201 Trotter Hall 1:45 pm – 2:00 pm



Aaron Mitchell

Rabies Virus Diagnostic Testing –Evidence Towards Changing the Minimum Standards for Tissue Sampling

201 Trotter Hall 1:45 pm – 2:00 pm

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Keywords: rabies; direct fluorescent antibody test; brainstem; cerebellum

Introduction/Background: The World Health Organization, the World Organization for Animal Health, and the Centers for Disease Control and Prevention recognize the direct fluorescent antibody (DFA) test as the gold standard for postmortem rabies diagnosis. U.S. national rabies diagnostic protocols recommend sampling both the brainstem and cerebellum, whereas WOAH guidelines allow composite samples that include the brainstem with the cerebellum or hippocampus. We evaluated whether brainstem tissue provides sufficient antigen for reliable DFA diagnosis by analyzing the correlation between brainstem and cerebellar DFA results from routine diagnostic cases.

Methods: We reviewed 29 DFA-positive rabies cases submitted between January 2024 and January 2026, including 12 cats, 11 skunks, three bovines, and one each of a dog, horse, and fox. We prepared brainstem and cerebellar impression smears, fixed them in cold acetone, stained them with fluorescein isothiocyanate, and examined slides under a fluorescence microscope for apple-green fluorescence indicative of rabies virus antigen, using positive controls to ensure accuracy. We graded antigen intensity on a 1+ to 4+ scale (1+ to 2+, rare to few inclusions; 3+ to 4+, numerous to abundant inclusions) and analyzed brainstem and cerebellar DFA scores using the Pearson correlation coefficient (r) and paired t- tests.

Results: Rabies antigen intensity was similar in the brainstem and cerebellum, with 18 of 29 cases (62%) recorded as 4+/4+. Pearson correlation analysis demonstrated a strong positive correlation between brainstem and cerebellar scores ($r(29) = .71$, 95% CI [.47, .86], $p < .0001$). The paired-samples t test demonstrated that antigen levels were significantly higher in the brainstem ($M = 3.86$, $SD \pm 0.35$) than in the cerebellum ($M = 3.45$, $SD \pm 0.83$), $t(28) = 3.55$, $p = .001$, 95% CI [0.18, 0.65], with a large effect (partial $\eta^2 = .31$). Brainstem DFA scores showed minimal species-related variability, with cats (11/12) and skunks (10/11) at 4+ and only occasional 3+ scores in cats, skunks, bovines, and the single dog. Cerebellar DFA scores showed modest variability, with skunks at 4+ in 10/11 cases, cats spanning 4+, 3+, and 2+ (5/12, 6/12, 1/12), and other species including bovines (2/3 at 4+, 1/3 at 1+), the dog (2+), the horse (4+), and the fox (3+).

Conclusions: The analyses show a strong positive association between rabies antigen detection in the brainstem and cerebellum, with significantly higher antigen levels in the brainstem. This correlation indicates consistent antigen detection across both tissues. These findings provide supportive evidence towards revising minimum tissue sampling standards to allow greater flexibility for independent brain stem testing when optimal collection is not feasible. This can improve diagnostic efficiency, biosafety, and field compliance without compromising diagnostic accuracy or public health protection.

Phi Zeta RESEARCH DAY
KANSAS STATE UNIVERSITY

201 Trotter Hall 2:00 pm – 2:15 pm

Julianne Nussbaum

Clinical Feasibility and of Standard Neurologic Examinations in Sonoran Desert Toads (Incilius alvarius) [GT1] [JN2]

201 Trotter Hall 2:00 pm – 2:15 pm

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Keywords: Neurologic exam, Sonoran Desert toad, amphibians, species-specific neurologic exam

Background
To evaluate the application of published standard clinical neurological exam techniques in Puerto Rican Crested toads (Peltophryne lemur) (PRCT) in Sonoran Desert toads (Incilius alvarius) and to establish expected species-specific results for the selected neurologic tests.

Methods
A research population of healthy Sonoran Desert toads was used in July 2025. Neurologic examinations based on standard tests described in PRCT were performed on each individual and evaluated for feasibility and expected results compared to other species.

Results
Nineteen toads were available, with three excluded due to mild pododermatitis. Sixteen toads were included for evaluation. Multiple tests were inconsistent or produce absent responses in this species, including menace response (0% [0/16]), facial stimulation (31% [5/16]), physiologic nystagmus (0% [0/16]), withdrawal reflex (0% [0/16]), myotatic reflexes (0% [0/16]), vent reflex and tone (0% [0/16]), tactile placing (31% [5/16]), proprioceptive placement (0% [0/16]), and hopping (25% [4/16]). The tests that were consistent and reproducible across individuals (i.e., 100% positive response) were the righting reflex, wheelbarrowing, pupil symmetry, pupillary light reflex, corneal reflex, and palpebral reflex.

Conclusions
Expected responses during neurologic examinations differed in Sonoran Desert toads compared to PRCT, with a notable difference being Sonoran Desert toads do not show a withdrawal reflex, proprioceptive placement, and hopping, as well as facial stimulation that was seen in PRCT.
It is recommended that a neurological examination be evaluated against species-specific parameters.

Phi Zeta RESEARCH DAY
KANSAS STATE UNIVERSITY

201 Trotter Hall 2:15 pm – 2:30 pm

Brooke Paisley

Trimethoprim Alone Attains High Urine Concentrations and May Be Effective for Canine Urinary Tract Infections

201 Trotter Hall 2:15 pm – 2:30 pm

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Keywords: Trimethoprim; Canine urinary tract infections; Veterinary pharmacology; Antimicrobial therapy

Antimicrobial resistant urinary tract infections in canines are becoming more common, and treatment options can be limited. Sulfonamide antimicrobials, often in combination with trimethoprim, can be used. However, the sulfonamide may produce severe adverse effects in dogs including death. Trimethoprim without a sulfonamide is routinely used in humans and may be effective in treating canine urinary tract infections.

Our objective was to evaluate trimethoprim urine concentrations after a single oral dose of trimethoprim alone. We hypothesized that trimethoprim alone can achieve and sustain concentrations in the urine to treat canine urinary tract infections. IACUC approved the study. Nine healthy client-owned dogs of varying breeds weighing over 10 kg received a single oral dose of trimethoprim 5 mg/kg (100 mg tablets) with food. Clients collected urine before administration of the dose and then 6-, 8-, 12-, and 24-hour post administration. Validated mass spectrometry, and a urine concentration versus time curve was created to analyze the concentration of the trimethoprim in the urine samples. Descriptive statistics were performed.

The mean (range) actual dose administered was 5.5 (4.5-7.2) mg/kg. The range of urine concentrations at 6 hours (69.9-413 mcg/mL), 8 hours (46.2-405 mcg/mL), 12 hours (34.5-220 mcg/mL) and 24 hours (17.2-154 mcg/mL) all exceeded 2 mcg/mL, the minimum inhibitory concentration for susceptible bacteria.

These data suggest oral trimethoprim alone may be effective when administered every 24h for canine urinary tract infections. Further research is needed to confirm the antimicrobial activity of trimethoprim in the urine.

Phi Zeta RESEARCH DAY
KANSAS STATE UNIVERSITY

201 Trotter Hall 3:15 pm – 3:30 pm

Erin Turman

Magnetic Resonance Imaging and Sequence Optimization of the Thoracic Duct in a Heterogenous Patient Population

201 Trotter Hall 3:15 pm – 3:30 pm

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Keywords: Diagnostic Imaging; Magnetic Resonance Imaging; MRI; lymphangiogram; thoracic duct

Magnetic resonance lymphangiogram (MRL) has been shown to have the ability to visualize the thoracic duct in a less-invasive manner compared to computed tomography in human patients. No published protocols for MRL are available in heterogeneous populations of canine patients. Thus, the aim of this prospective study was to develop a protocol for evaluation of the thoracic duct in a clinical patient population.

Sixteen canine patients presented to the Veterinary Health Center at Kansas State University College of Veterinary Medicine with spinal disease and were imaged using a 3.0 T MR machine. Various sequences were performed to create an optimal protocol. After scanning, the MR images were evaluated by two board-certified veterinary radiologists via the following parameters: visualization of the thoracic duct, relation of the thoracic duct to the aorta, and the location and number of anastomosing branches.

A pilot study of five patients was completed to optimize MR sequences for further research. Thirty-four (34) sequences were performed in the pilot study. Of those, 23 sequences were successful in visualizing the thoracic duct, with no individual sequence taking longer than 15 minutes to complete. The cranial-most visualization of the thoracic duct was the thoracic inlet on six different sequences (SAG 3D MYELO, Ax Cisterno FASE, T2 FASE 3D, SAG Cisterno FASE, AX FASE, SAG FASE).

Fourteen (14) of the 23 sequences with thoracic duct visualization showed anastomosing branches with a range of 1 to 6. Consistent with prior anatomic description, of the 23 sequences with thoracic duct visualization, 22 were located to the right and dorsal to the aorta. Six (6) of those 23 sequences showed some anatomic variation of the thoracic duct being present to the left and dorsal to the aorta.

MRL is feasible in clinical canine patients and should be explored as a less invasive imaging modality compared to computed tomography lymphangiogram (CTL).

Basic Science – 301 Trotter Hall

Phi Zeta RESEARCH DAY
KANSAS STATE UNIVERSITY

301 Trotter Hall 1:00 pm – 1:15 pm

Uttama Acharjee

Broad-Spectrum Antiviral Activity of 3CLpro Inhibitors against Bat Coronaviruses Utilizing ACE2 or DPP4 Receptors

301 Trotter Hall 1:00 pm – 1:15 pm

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Keywords: Bat coronavirus; ACE2 receptor; DPP4 receptor; 3CLpro; Inhibitor

Bats are recognized as natural reservoirs of diverse coronaviruses. Major human disease outbreaks caused by SARS-CoV, SARS-CoV-2, and MERS-CoV are presumed to have originated from zoonotic spillover of bat coronaviruses. Multiple bat species harbor SARS-CoV-like (Sarbecovirus) and MERS-CoV-like (Merbecovirus) viruses that use angiotensin-converting enzyme-2 (ACE2) or dipeptidyl peptidase-4 (DPP4) as host cell receptor, similar to SARS-CoV, SARS-CoV-2, and MERS-CoV. This shared receptor tropism highlights the zoonotic and pandemic potential of bat coronaviruses and underscores the importance of evaluating the antiviral activity of inhibitors against them. Coronavirus 3-chymotrypsin-like protease (3CLpro) cleaves the viral polyprotein into functional nonstructural proteins that are essential for the viral replication cycle and is conserved among coronaviruses, making it an attractive broad-spectrum antiviral target. This study aims to evaluate the antiviral activity of 3CLpro inhibitors against ACE2 or DPP4 utilizing bat coronaviruses. Ten bat coronaviruses from Betacoronavirus (Sarbecovirus and Merbecovirus) and Alphacoronavirus genera, along with SARS-CoV, SARS-CoV-2, MERS-CoV, and FIPV (Feline Infectious Peritonitis Virus), were included in this study. Their full-length 3CLpro genes were cloned and expressed for fluorescence resonance energy transfer (FRET) assay. Nine 3CLpro inhibitors, including GC376, its analogs, and Nirmatrelvir, were tested against the expressed 3CLpros in the FRET assay to determine 50% inhibitory concentrations (IC_{50}). Some compounds effective in the FRET assay are currently being evaluated for their activities in cell-based assays. X-ray crystallography of bat coronavirus 3CLpro in complex with GC376 was performed. Additionally, we analyzed approximately 220 bat coronavirus 3CLpro sequences for amino acid homology and phylogeny. Sequence analysis of these 3CLpros revealed high homology within the Sarbecovirus (90–100%) and Merbecovirus (80–95%) subgenera, and 60–90% homology within Alphacoronavirus genus. Fifty percent or less homology was observed among the different coronavirus genera. In the FRET assay, some compounds demonstrated potent broad-spectrum inhibition across all tested coronaviruses, including those utilizing ACE2 or DPP4 receptors. These preliminary results highlight the potential of 3CLpro inhibitors as antiviral candidates against bat coronaviruses with zoonotic potential.

Thomas Hoefler

LoRRecMa – A Long-Read Recombination Mapper

301 Trotter Hall 1:15 pm – 1:30 pm

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Keywords: Recombination; Bioinformatics; Herpesvirus

Evolution utilizes three major forces to mold new forms out of old material: mutation, genetic drift and selection. Adaptable diversity is selected from a pool of variants created by mutation and lost by drift. Another way to increase the genetic diversity of a population is the recombination of already existing variation. Recombination describes a molecular process in which genetic material from different sources is exchanged. Many species benefit from recombination because, in contrast to other forms of mutation, it creates new combinations of functional material and reshapes the building blocks of molecular machines.
Viruses utilize recombination to increase their already remarkable diversity. This allows them to rapidly explore new host populations, mitigate immune pressure or antiviral treatment and enhance their transmission between individuals. However, detecting recombination events in a viral population is challenging. The conglomerate of millions of individual genomes within the viral population contains only a small subset of recombinant viruses. Biophysical approaches such as the introduction of marker genes (e.g. GFP, or selection markers), are wedded to laborious purification of many individual clones. Phylogenetic methods, in contrast, depend on consensus sequences which often fail to capture recombinant minor variants.
Bioinformatic analysis revolutionized the mapping of large-scale deletions, duplications or inversions as well as recombination between two unrelated genomes (i.e. ViReMa). The detection of recombination between two closely related references, however, remains a challenge. We utilized long-read Oxford Nanopore sequencing and unique mutation profiles of co-infected strains to successfully map recombination events in herpes simplex virus type 1 (HSV-1). Using this approach, we detected distinct recombination rates depending on which strains were used for co-infection. Additionally, we find that nearly half of all recombinant genomes feature more than one recombination event. We also observe distinct junction distributions across the genome, arguing that sequence motifs play an important role in recombination. Overall, we present an unbiased method to detect recombination events that can be readily adapted to any virus species.

Phi Zeta RESEARCH DAY
KANSAS STATE UNIVERSITY

301 Trotter Hall 1:30 pm – 1:45 pm

Claire Horton

Determining the Prevalence of Influenza A in Feline Species Through Competitive Enzyme-linked Immunosorbent Assay and Polymerase Chain Reaction Methods

301 Trotter Hall 1:30 pm – 1:45 pm

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Keywords: avian influenza A; highly pathogenic avian influenza virus; feline; seroprevalence; enzyme-linked immunosorbent assay; polymerase chain reaction

Unanticipated outbreaks since March 2024 of H5N1 highly pathogenic avian influenza virus (HPAI) in feline species in the United States has caused various veterinary and public health concerns. Infections result in a high mortality rate in felines, also facilitating mutations and possible cross-species transmission. This study aimed to investigate the seroprevalence of antibodies against H5N1 and presence of viral RNA in feline species submitted for diagnostic testing through the Kansas Veterinary Diagnostic Laboratory. A total of 617 feline serums were collected in 2023 to 2024 and analyzed for the presence of Avian Influenza A antibodies. The test was performed using 2 commercial competitive enzyme-linked immunosorbent assay (ELISA) testing kits, A and B. In addition, polymerase chain reaction (PCR) tests for Avian influenza A virus targeting the M-gene were performed on 225 feline brain tissue samples submitted due to neurological signs between 2023 and 2024. Of the 617 feline samples, 3 tested positive on test A (0.49%) and 1 tested positive on test B (0.16%). In the test detection of M-gene, 10 out of 225 (4.44%) showed a positive result. The results demonstrate that HPAI can be detected in feline species by both ELISA and PCR. The findings highlight the importance of continuing surveillance, especially due to the virus's high mortality rate in felines and increased risk of zoonotic transmission. Moreover, the higher detection rate by PCR compared with ELISA supports the hypothesis that many cats may succumb to infection before seroconversion is able to occur.

Phi Zeta RESEARCH DAY
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301 Trotter Hall 1:45 pm – 2:00 pm

Prabhu Raj Joshi

Role of Beta-2 Adrenergic Receptor Signaling in Innate Immunity Against Intracellular Bacterial Pathogens

301 Trotter Hall 1:45 pm – 2:00 pm

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Keywords: B. thailandensis, β2-AR, innate immunity, pneumonia

Burkholderia thailandensis (Bt) is a Gram-negative intracellular respiratory pathogen and a model organism for studying melioidosis because of its similarities to the highly virulent *B. pseudomallei*, which causes severe pneumonia and sepsis in animals and humans. The bacterium evades phagocyte killing, proliferates within phagocytes, and leads to antibiotic-resistant infections. Lung-innervating sympathetic neurons and the adrenal gland release the neurotransmitter noradrenaline (NA) during stress, inflammation, or injury. NA acts on tissue-resident and recruited immune cells in the lungs via the β2-adrenergic receptor (β2-AR) to modulate the immune response. Our findings show that β2-AR deficiency in mice led to lethality, severe clinical signs of sepsis, hypothermia, and defective bacterial clearance in the lungs and extrapulmonary organs, compared with β2-AR-sufficient mice after intranasal infection with *Bt.* Additionally, *Adrb2*$^{-/-}$ mice showed defective immune cell recruitment and increased airway cytokine levels compared with control mice. Furthermore, systemic treatment of Bt-infected mice with a β2-agonist (salmeterol) rapidly improved survival, reduced bacterial burden in the lungs and other organs, and enhanced immune cell recruitment. Similarly, *Adrb2*$^{-/-}$ bone marrow-derived macrophages (BMDMs) infected with *Bt* showed an increased intracellular bacterial burden when compared to wild-type (WT) BMDMs. Additionally, treating *Bt*-infected WT BMDMs with a β2-AR agonist significantly decreased intracellular bacterial survival by increasing nitric oxide production and altering cytokine production. Overall, our results suggest a critical role of β2-AR signaling in lung innate immunity against intracellular bacterial infections.

Phi Zeta RESEARCH DAY
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301 Trotter Hall 2:00 pm – 2:15 pm

Sujan Kafle

Mucosal infection of CAST/EiJ Mice with Mpox Virus

301 Trotter Hall 2:00 pm – 2:15 pm

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Keywords: Mpox; Mpox virus(MPXV); CAST/EiJ mice; mucosal infection

Introduction:
Mpox, a zoonotic disease caused by Mpox virus (MPXV), primarily replicates in rodents and small mammals with occasional spillovers to humans. Historically, human-to-human transmission was limited and mainly occurred through close contact and respiratory droplets. However, during the 2022 global outbreak of MPXV clade IIb, sexual transmission, particularly among men who have sex with men (MSM), emerged as the predominant transmission route. Experimental studies investigating MPXV infection via rectal and genital mucosal routes are limited, and the need for a small animal model to better understand MPXV pathogenesis is critical. Here, we aim to investigate the susceptibility of CAST/EiJ mice to MPXV clade IIb infection via mucosal routes.

Methods:
CAST/EiJ mice (mixed sex) were infected with 2×10^5 PFU of MPXV clade IIb via intraperitoneal or mucosal (intranasal, rectal, and genital) routes. Mice were monitored daily for clinical signs and weight changes, and swabs (oropharyngeal, rectal, and urogenital) were collected to assess viral shedding. At the end of study, mice were euthanized and necropsied, and various tissues and blood were collected for virological and histopathological analyses.

Results and conclusion:
Intraperitoneal infection of mice with MPXV produced non-lethal, mild clinical disease, particularly in male mice, which showed slight body weight loss from 5 to 10 days post-infection, corresponding to increased clinical scores marked by ruffled fur and subdued behavior; viral DNA was also detected in oropharyngeal swabs during this period. In contrast, mucosal inoculation via intranasal, rectal, or genital routes did not induce obvious clinical signs, as mice remained largely asymptomatic with no skin lesions and minimal to no body weight loss. However, viral DNA was consistently detected for at least 8 days in oropharyngeal, rectal, and genital swabs in the respective inoculation groups, except in male mice infected via the intrapenile route. Viral DNA was also detected at sites other than the route of exposure. The sustained recovery of viral DNA at the mucosal inoculation sites suggests active viral replication in the associated mucosal tissues. However, it must be considered that grooming may play a role in sustained viral DNA detection in swab samples.

Phi Zeta RESEARCH DAY
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301 Trotter Hall 2:15 pm – 2:30 pm

Christine Langner

Insights into Host Responses to a Codon Pair Deoptimized SARS-CoV-2 Vaccine Candidate

301 Trotter Hall 2:15 pm – 2:30 pm

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Keywords: Codon Pair Deoptimisation, SARS-CoV-2, Immune Response, Vaccine-Host Interactions

Five years after the start of the COVID-19 pandemic, SARS-CoV-2 is still a considerable global health burden, posing an acute threat to immunocompromised individuals and causing chronic health problems. Although safe and effective vaccines have been developed, they do not protect from infection and allow for viral transmission. sCPD9, a vaccine candidate attenuated by codon pair deoptimization (CPD) of the SARS-CoV-2 nsp16 gene, confers superior protection by strong induction of mucosal immunity and promotes immune responses directed against the entire viral antigenic repertoire. Owing to excellent preclinical safety and superior efficacy, sCPD9 is currently in line for phase 1 clinical trials. However, the precise mechanism of attenuation, and how CPD influences host responses, remains elusive.

We explored early immune responses in human cell cultures and primary airway epithelial cells (HAE) using state of the art RNA sequencing methods to complement standard virological assays. Our results show that sCPD9 elicits an early host response distinct from the SARS-CoV-2 WT virus and a functional knock-out mutant of nsp16 (D130A). In this comparison, timing and intensity of the interferon response are different between viruses, as are markers for keratinisation and calcium signaling. Different cell types play distinct roles in the antiviral response – depending on whether they are infected with WT, sCPD9 or nsp16 D130A. Taken together, our results suggest that CPD may confer attenuation independently of the recoded genes' function in SARS-CoV-2. Additional bioinformatic analysis will focus on details such as basal cell turnover and differentiation, and analysis of protein expression and nsp16 complementation will further reveal effects of CPD on the host response and viral replication. To overcome batch effects inherent to human primary cell cultures, we will provide data from a Syrian hamster infection model, known to yield highly reproducible outcomes of SARS-CoV-2 infection. This model allows us to explore the interplay between a fully functional immune system and the epithelium, and may help us to uncover how CPD influences mucosal immunity. We believe that CPD has the potential to serve as broadly applicable strategy to rationally design live attenuated vaccines – for SARS-CoV-2 and future zoonotic coronaviruses.

Phi Zeta RESEARCH DAY
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301 Trotter Hall 3:15 pm – 3:30 pm

Luija Mudiyanselage

Development of a Biosaftey Level 2–Compatible Prototype Infection Model to Investigate Rift Valley Fever Virus (RVFV) Interactions with Culex tarsalis Mosquitoes

301 Trotter Hall 3:15 pm – 3:30 pm

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Keywords: Rift Valley Fever Virus; Culex tarsalis; BSL-2 infection model; RVFV MP-12

BACKGROUND: Rift Valley Fever Virus (RVFV) is an arthropod-borne virus primarily transmitted by Aedes and Culex mosquitoes. It has a tri-segmented, negative-sense RNA genome comprising small (S), medium (M), and large (L) segments. RVFV causes severe disease in livestock, including high mortality in young animals and widespread abortions. Human infections are often mild but can result in severe complications. RVFV is classified as a select agent that requires handling under enhanced biosafety level 3 (BSL-3) containment. The attenuated vaccine strain RVFV MP-12 can be handled under biosafety level 2 (BSL-2) conditions and offers a safer and less complex alternative for studying RVFV–mosquito interactions. The objective of this study was to establish an RVFV MP-12 infection model in Culex tarsalis mosquitoes and to evaluate the dynamics of viral replication and dissemination over time.
METHODS: Female C. tarsalis mosquitoes were starved for 24 h prior to infection. Mosquitoes were orally exposed for 1 h to an artificial blood meal consisting of freshly prepared RVFV MP-12 mixed 1:1 with defibrinated sheep blood, or to sheep blood alone for the mock infection, using a membrane-feeding system. Following blood feeding, fully engorged mosquitoes were sorted, and 20 individual mosquitoes per group were collected at 0, 3, 7, and 14 days post-infection (dpi). Individual mosquitoes were homogenized, viral RNA was extracted from supernatants for RT-qPCR analysis of the M and L genome segments, and infectious virus titers were determined by plaque assay. RVFV MP-12 stock, and blood meal titers were verified by back-titration via RT-qPCR and plaque assay. The entire experiment was independently repeated three times.
RESULTS: Back-titration of the RVFV MP-12 stock yielded initial viral loads with median RNA levels of 7.2 log10 RNA copies (M segment-M) and 6.3 log10 RNA copies (L segment-L). Immediately following the 1-hour feeding period (Day 0), median viral loads in engorged mosquitoes were at 4.6 log10 RNA copies (M) and 2.8 log10 RNA copies (L). By 3 dpi, a reduction in viral RNA levels was observed, with median declining 4.0 log10 (M) and 3.1 log10 (L) RNA copies, which is compatible with the elimination of the inoculum through digestion of the mosquito blood meal. An increase in viral RNA levels was observed by 7 dpi, with median RNA loads increasing to 5.5 log10 (M), 4.5 log10 (L) RNA copies, demonstrating that the virus is actively infecting and replicating in the mosquito. By 14 dpi, median viral RNA levels of 5.5 log10 (M) and 4.6 log10 (L) RNA copies remained comparable to 7 dpi levels, consistent with sustained viral replication.
CONCLUSION: Collectively, these findings demonstrate that C. tarsalis supports productive replication and long-term persistence of RVFV MP-12. This BSL-2 compatible infection model provides an invaluable model for investigating RVFV-mosquito interactions and identifying mosquito factors relevant to the development of novel transmission-blocking strategies.

Phi Zeta RESEARCH DAY
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301 Trotter Hall 3:30 pm – 3:45 pm 

Nicole Ostrander

Evaluation of Porcine T Cells Following Vaccination with Commercial Flavivirus Vaccines

301 Trotter Hall 3:30 pm – 3:45 pm

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Keywords: Immunity; Flavivirus; Vaccination; Swine; Flow cytometry

Japanese encephalitis virus (JEV) is a foreign zoonotic flavivirus of significant concern to the United States (U.S.) swine industry. Currently, there are no approved veterinary vaccines or treatments for JEV in the United States. In contrast, West Nile virus (WNV), a related flavivirus and zoonotic pathogen of concern, does have approved commercial equine vaccines available. Within the Flaviviridae family, some cross-reactivity exists between these viruses, as JEV and WNV share similar structural proteins that often serve as vaccine targets. Cross-neutralizing antibodies between flaviviruses have been documented in both humans and animals following vaccination or natural infection. However, the immune response against JEV infection in swine remains poorly understood, and the mechanisms of protective immunity following vaccination have yet to be investigated. To evaluate potential cross-reactive T cell immunity, we analyzed CD4+, CD8+, and CD4+CD8+ double-positive T mon populations within peripheral blood mononuclear cells (PBMCs) isolated from swine vaccinated with commercial equine WNV vaccines. PBMCs were stimulated in vitro with JEV, and T cell activation was assessed through phenotypic characterization of naïve, central memory, and effector memory subsets. We then assessed the protective functionality of these activated T cells by measuring IFNγ expression. An observed increase in IFNγ expression in response to JEV stimulation would suggest that these T cell populations could be capable of contributing to protective immunity against JEV. Leveraging available commercial vaccines to provide cross-protection against JEV could enable a rapid response in the event of JEV introduction into U.S. swine populations.

Phi Zeta RESEARCH DAY
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301 Trotter Hall 3:45 pm – 4:00 pm

Saurav Pantha

Sexual Dimorphism in Metabolic Responses to Influenza Vaccination in a Mouse Model of Diet-Induced Obesity

301 Trotter Hall 3:45 pm – 4:00 pm

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Keywords: Diet-induced obesity; influenza; vaccination; metabolomics; B cell

Background: Our prior study showed that female mice with obesity produce significantly higher antibody responses to the influenza vaccine and are better protected compared to the males with obesity. The B-cell responses in the spleen, however, were comparable. We hypothesized that differences in B-cell metabolism and function drive sexual dimorphism in influenza vaccine efficacy during obesity. In this study, our objective was to determine sex and obesity-specific differences in influenza vaccine-induced metabolites in splenic B-cells.

Methodology: Male and female mice, with or without diet-induced obesity, were vaccinated with an inactivated pandemic 2009 H1N1 influenza vaccine twice at a 3-week interval. At 35 days post-vaccination, magnetically sorted CD19+ B-cells from the spleen were analyzed using liquid chromatography mass spectrometry. Log-transformed data were analyzed by two-way ANOVA, and $p \leq 0.05$ was considered a significant difference.

Results: Principal component analysis revealed obesity-associated metabolic reprogramming in males but not in females. There was a clear separation of the analyzed metabolites between males with or without obesity, while no such distinction was observed in females, who exhibited substantial overlap. Overall, a total of 184 biochemicals showed statistically significant sex-differences in B cells. While focusing on the sex-differences under obese conditions, pairwise comparisons identified 142 metabolites that displayed significant differences. Females with obesity had a significantly lower level for three biochemicals, and significantly higher level for 139 biochemicals compared to males with obesity.

The 3 biochemicals that were lower in females with obesity compared to the males with obesity included hypotaurine, S-(1,2-dicarboxyethyl) glutathione, and uridine 5'-monophosphate. Females with obesity had a significantly higher level of dipeptides, lipids, such as long-chain polyunsaturated fatty acids (PUFAs), acylcarnitines, acylcholines, and phospholipids, nucleotides, cofactors, and vitamins as compared to males with obesity. Dipeptides are sources of amino acids that act as metabolic fuel for the endoplasmic reticulum (ER) expansion and boost antioxidant protection. PUFAs are required for B-cell membrane remodeling, signaling, and eicosanoid-mediated regulation. Acylcarnitines and acylcholines facilitate the import of fatty acids for mitochondrial β-oxidation to meet the energy demands of highly metabolic B cells during proliferation and differentiation. Phospholipids, such as phosphatidylcholines (PC), form the lipid rafts, which concentrate and amplify the B cell signaling for proliferation, which help in the expansion of the endoplasmic reticulum and unfolded protein response, and contribute towards higher antibody production. Furthermore, phosphatidylethanolamine (PE) maintains membrane integrity and curvature required for B-cell activation and antibody production. Plasmalogens act as antioxidants and protect B cells from oxidative damage, and maintain membrane fluidity. Sphingomyelins maintain the structural organization of B cells and improve receptor signaling. Vitamins, cofactors, and nucleotides support antioxidant defense, coenzyme function, and nucleotide metabolism.

Conclusion: Our findings reveal that the higher antibody production following influenza vaccination in females, compared to males with obesity, is associated with a favorable metabolic and lipid environment that supports robust B-cell activation and differentiation. Further exploration and intervention in such metabolic pathways are needed to develop tailored therapeutics to minimize disparities in vaccine response.

Phi Zeta RESEARCH DAY
KANSAS STATE UNIVERSITY

301 Trotter Hall 4:00 pm – 4:15 pm

Ricardo Martin Vidal

Tuning nsp14 Expression Reshapes SARS-CoV-2 Evolutionary Trajectories

301 Trotter Hall 4:00 pm – 4:15 pm

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Keywords: Recoding, nsp14, evolution

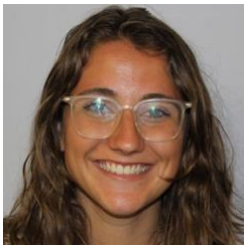
Introduction: Coronaviruses possess unusually large RNA genomes (~30 kb) that require high replication fidelity to maintain genetic stability. Unlike most RNA viruses, coronaviruses encode a proofreading exoribonuclease, nsp14, which forms a functional complex with nsp10 and increases replication fidelity by excising misincorporated nucleotides during RNA synthesis. In SARS-CoV-2, mutations in the catalytic core of nsp14 are lethal, underscoring its essential role in viral replication. However, how partial impairment of nsp14, particularly through reduced protein expression rather than complete catalytic inactivation, affects viral replication, fidelity and evolution, remains unclear. In this study, we use codon pair deoptimization (CPD), a large-scale recoding strategy, to modulate nsp14 expression without altering its amino acid sequence, enabling controlled reduction of nsp14 expression in SARS-CoV-2.

Materials & Methods: Using the ancestral SARS-CoV-2 Wuhan-01 strain, we generated codon pair-deoptimized mutants targeting either the entire nsp14 coding sequence or only the exoribonuclease domain (Exo-CPD), spanning a range of deoptimization levels defined by Codon Pair Scores (CPS). Recombinant viruses were constructed by TAR cloning and rescued in Vero E6 cells. Selected mutants were serially passaged for 30 passages under progressively increasing population bottlenecks. Viral replication kinetics were assessed by multi-step growth curves and plaque assays. Nsp14 expression was quantified by Western blotting under both low-MOI and synchronized high-MOI infection conditions. Viral genomes from different passages were analyzed by Next-Generation Sequencing (NGS) to assess mutation frequencies and evolutionary trajectories.
Results: Viruses carrying codon pair deoptimization limited to the exoribonuclease domain were efficiently rescued and exhibited replication kinetics comparable to wild type virus across all passages, with no detectable reduction in nsp14 protein levels. In contrast, only moderately deoptimized full-length nsp14-CPD mutants were viable, highlighting strict constraints on nsp14 recoding. These nsp14-CPD mutants displayed early replication defects and markedly reduced nsp14 protein levels at initial passages, followed by recovery of viral growth without reversion of the recoded sequence. Quantitative protein analysis under standardized infection conditions revealed a clear inverse correlation between the extent of codon pair deoptimization and nsp14 expression. Genome sequencing demonstrated that both, but specially nsp14-CPD mutants, accumulated more single-nucleotide polymorphisms than wild type virus at intermediate passages, consistent with impaired proofreading activity, followed by selection of putative compensatory mutations at later passages.

Conclusion: Our findings show that SARS-CoV-2 requires not only the presence of a functional nsp14 protein, but also sufficient expression levels to sustain replication fidelity and viral fitness. Partial reduction of nsp14 expression compromises genome stability and alters viral evolutionary trajectories while still permitting replication. Together, this study demonstrates that SARS-CoV-2 tolerates a only narrow range of nsp14 expression levels and highlights codon pair deoptimization as a powerful strategy to study essential viral functions and to generate attenuated viruses.

Phi Zeta RESEARCH DAY
KANSAS STATE UNIVERSITY

301 Trotter Hall 4:15 pm – 4:30 pm



Hailey Weir

Neonatal Administration of Esrogenic Ear Implants as a Non-invasive Alternative to Bovine Castration

301 Trotter Hall 4:15 pm – 4:30 pm

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Keywords: Animal Welfare; Castration; Cattle; Estrogen; Kisspeptin

Castration is a common management practice in the cattle industry performed to reduce aggression, prevent unwanted pregnancies, and improve meat quality. Targeting the development of kisspeptin neurons through neonatal exposure to estrogen may provide a practical, non-invasive alternative that improves animal welfare. The objective of this randomized controlled trial was to determine the efficacy of administering commercially available estrogenic implants (Ralgro®, Compudose®, and Encore®) in neonatal calves to suppress testicular development. Twenty-five intact Holstein bull calves (<24h of age with two descended testicles) were randomly assigned to 1 of 5 treatment groups at birth: surgically castrated control (CUT), silicone placebo implant (SIL), Ralgro® (RAL), Compudose® (COMP), or Encore® (ENC). Scrotal circumference was measured every 2 weeks from enrollment through week 56. Linear mixed-effects models were used to determine potential associations between treatment, week, and their interactions on scrotal circumference ($P \leq 0.05$). A significant treatment-by-week interaction was detected for scrotal circumference. Bulls that received RAL, COMP and ENC had smaller ($P \leq 0.03$) scrotal circumference than SIL bulls from weeks 30 to 46. Bulls that received RAL and COMP continued to exhibit smaller ($P \leq 0.02$) scrotal circumference compared to SIL bulls through week 50. No differences ($P \geq 0.22$) in scrotal circumference were observed between SIL and ENC after week 46. These results indicate suppression in testicular development however, further analyses are needed to determine the effect of estrogenic implants on reproductive potential in dairy bulls. Additional outcomes, including infrared thermography, plasma testosterone, breeding soundness examinations with semen evaluation, and testicular histology, are still being analyzed.

Alexandria Zabiegala

PD-L1 Expression in Tissues of Cats with Feline Infectious Peritonitis

301 Trotter Hall 4:30 pm – 4:45 pm

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Keywords: Coronavirus, PD-L1, Feline Infectious Peritonitis

Feline infectious peritonitis (FIP) is a fatal, systemic infection in cats caused by virulent feline infectious peritonitis virus (FIPV). Feline coronavirus (FCoV) typically circulates as feline enteric coronavirus (FECV), which is ubiquitous among cat populations, and causes asymptomatic to mild enteric disease. However, viral mutations in combination with impaired host immune response may lead to FIP in a small number of cats. Programmed death-1 (PD-1) is a major immune-checkpoint protein expressed on activated T cells, and its engagement by the ligands PD-L1 and PD-L2 on target cells leads to suppression of T-cell activity. Previously, we found that infection with FIPV, but not FECV, upregulated the expression of PD-L1 in feline cells, which can attenuate the T cell activities. We also demonstrated that feline interferons (IFNs), particularly IFN-γ, are potent inducers of PD-L1 in the cells. In this study, we assessed the expression of PD-L1 in association with FIPV in the tissues of four cats with naturally occurring FIP using histopathology, real-time RT-qPCR and confocal microscopy with specific antibodies. Using confocal microscopy, we found that FIPV and PD-L1 co-localized in the tissues, although the relative levels of PD-L1 RNA and viral RNA varied among tissues and between individual cats. The cells expressing PD-L1 were located adjacent to FIPV positive cells, which is probably due to inflammatory cytokines such as IFN-γ. These findings support our previous findings of a close association between PD-L1 and FIPV and further underscore the need to investigate the roles of PD-L1 and T cell responses in the development of FIP in cats.

Clinical Case Studies – 301 Trotter Hall

Phi Zeta RESEARCH DAY
KANSAS STATE UNIVERSITY

201 Trotter Hall 4:15 pm – 4:30 pm

Devon DiBello

Widespread Osteolysis as an Uncommon Manifestation of Equine Sarcoidosis

201 Trotter Hall 4:15 pm – 4:30 pm

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Keywords: Equine, Sarcoidosis, Granulomatous Disease, Diagnostic Imaging, Pathology

Background: Equine sarcoidosis is an uncommon, presumed immune-mediated disease characterized by granulomatous inflammation with variable systemic involvement and severity. Most commonly, inflammation affects the skin, although the disease can progress to involve lymphatics and other organ systems. The most severe form of the disease is known as generalized equine sarcoidosis. While musculoskeletal involvement is a recognized disease feature in human sarcoidosis, there are only few reports to date in horses, and none yet have correlated imaging features to histopathologic diagnosis.

Case Description: A 15-year-old Quarter Horse mare presented for evaluation of chronic left forelimb lameness. Relevant history included recurrent left forelimb edema and a chronic undiagnosed dermatitis. Lameness examination identified a grade 3/5 left forelimb lameness that markedly improved following a left palmar-digital nerve block, therefore localizing it to foot. Radiographs revealed bilateral navicular degeneration, worse on the left, and the unexpected finding of multifocal geographic osteolytic lesions throughout the phalanges. Clinicopathologic testing was consistent with chronic inflammation. Differential diagnoses for polyostotic osteolysis included round cell neoplasia, fungal osteomyelitis, and silicate-associated osteoporosis; multiple myeloma was excluded with serum protein electrophoresis, and no supporting evidence was found for infectious or toxic etiologies.

Although the lameness initially improved with corrective shoeing and anti-inflammatory therapy, the mare re-presented one month later with anorexia, lethargy, and generalized stiffness. Skin biopsies were pursued, revealing severe, diffuse granulomatous lymphoplasmacytic dermatitis. In the absence of identifiable infectious agents, equine sarcoidosis was considered the primary differential. Given the concurrence of systemic disease, a progressed form of sarcoidosis was suspected, and euthanasia was elected by the owner due to poor prognosis.

Ante-mortem radiographs revealed extensive polyostotic osteolysis affecting nearly every bone of the appendicular skeleton. Post-mortem computed tomography of the left forelimb further characterized lesion distribution, identifying predominantly cortical lytic lesions, with some extending to the corticomedullary junction and few within the medullary cavity. Cortical thinning, expansion, and articular surface involvement were observed.

Necropsy and histopathology discovered diffuse granulomatous inflammation affecting multiple organ systems, including skin, musculoskeletal, gastrointestinal, lymphatic and central nervous systems. Histologic evaluation of bone sections

corresponding to osteolysis as found on imaging confirmed granulomatous inflammation within these lesions. Collectively, findings supported a diagnosis of generalized equine sarcoidosis.

Discussion: This case documents extensive granulomatous inflammatory infiltration throughout the musculoskeletal system in a horse, previously sparsely reported as a disease feature. Additionally, this is the first report to directly correlate osteolysis detected on both radiography and computed tomography to histopathologic diagnosis.

In humans, osseous manifestations of sarcoidosis are often asymptomatic and incidentally discovered, suggesting its prevalence may be underestimated. Observations in people with bone marrow infiltration support its association with more severe systemic disease; this is also observed in the present case, with bone marrow involvement detected in a progressed disease form.

Given the reliance of horses on their musculoskeletal system, diffuse osteolysis and extensive muscular involvement raise concern for catastrophic, life-ending injury, including pathologic fracture. This consideration is particularly important prior to initiating corticosteroid therapy, the mainstay of treatment, as corticosteroids are known to exacerbate osteoporosis. Further investigation into the prevalence and clinical significance of musculoskeletal involvement in equine sarcoidosis may improve prognostication and inform clinical decision-making.

Phi Zeta RESEARCH DAY
KANSAS STATE UNIVERSITY

201 Trotter Hall 3:45 pm – 4:00 pm

Drew McNassor

First Successful Management of Confirmed Canine Malignant Hyperthermia

201 Trotter Hall 3:45 pm – 4:00 pm

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Keywords: Malignant hyperthermia, dantrolene, RYR1 mutation, hypercapnia

Background: Malignant hyperthermia (MH) is a rare, life-threatening pharmacogenetic disorder that is poorly documented in dogs. Diagnosis is typically presumptive based on clinical signs, and treatment is often limited due to medication cost and rarely pursued in
veterinary medicine.

Case Description: A 6-month-old, male intact Whippet (7.4 kg) presented for surgical ligation of a patent ductus arteriosus (PDA). General anesthesia was maintained with isoflurane after an uneventful induction. Difficulty ventilating was observed early despite
mechanical ventilation. Severe hypercapnia was identified with end-tidal carbon dioxide (EtCO2) values of approximately 80-90mmHg. Increasing positive inspiratory pressure (~20–25 cmH2O) and atracurium administration failed to improve ventilation. Airway
suctioning, endotracheal tube replacement, and conversion to a different ventilator were attempted; however, EtCO2 increased to 110–115 mmHg and remained refractory to aggressive hyperventilation. Concurrently, body temperature demonstrated a progressive increase, reaching 102–103ºF (38.9-39.4ºC), which was considered inappropriate given the patient's size, open thoracic cavity, and discontinuation of active warming. Based on the severe, refractory hypercapnia, incongruent temperature trends, and lack of response to ventilatory interventions, MH was suspected. Isoflurane was discontinued, and anesthesia was maintained with a
propofol continuous rate infusion. The breathing circuit and ventilator were also replaced. Based on prior laboratory work, dantrolene was emergently compounded from generic dantrolene sodium capsules using 20% mannitol as an excipient to achieve an approximate concentration of 1 mg/mL. Two intravenous doses of dantrolene (approximately 2.7 mg/kg each) were administered 15 minutes apart. EtCO2 decreased to 70-80 mmHg within 5–10 minutes following the first administration and then continued to decline to 40–50 mmHg within 5–10 minutes after the second dose. Concurrently, the patient's body temperature began to decrease following dantrolene administration. Anesthetic recovery was uneventful. Post-operative serum biochemistry revealed creatine kinase (CK) elevation (2,190 U/L on the day of surgery, increasing to 7,871 U/L the following day), consistent with significant perioperative muscle injury and metabolic stress. Subsequent genetic testing identified a rare RYR1 variant (p.Asp4259Asn). The clinical presentation, response to dantrolene, and genetic findings confirm a diagnosis of MH, representing the first reported sequence-confirmed MH-positive canine to survive an MH reaction.

Discussion: Despite its rarity, MH can occur in dogs without prior anesthetic exposure and may be identified through prompt anesthetic vigilance. Severe, refractory hypercapnia and inappropriate hyperthermia given the surgical and anesthetic conditions are features consistent with MH. Furthermore, rapid clinical response associated with dantrolene therapy, postoperative evidence of rhabdomyolysis, and identification of a rare RYR1 mutation further strengthen the

diagnosis of MH. Hypoventilation, equipment failure, and hemorrhage were all considered as potential causes of the observed clinical signs; however, these were deemed unlikely
due to lack of clinical improvement despite appropriate corrective interventions. In addition to providing clinically and genetically supported evidence of MH susceptibility in a canine patient, this case also demonstrates that dantrolene can be emergently compounded to therapeutically effective concentrations and may be lifesaving when commercial intravenous formulations are cost prohibitive or unavailable.

Phi Zeta RESEARCH DAY
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201 Trotter Hall 4:00 pm – 4:15 pm

Julianne Nussbaum

Apitherapy: Clinical Use and Application for a Case of Liposarcoma in a Guinea Pig (Cavia porcellus)

201 Trotter Hall 4:00 pm – 4:15 pm

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Keywords: liposarcoma; apitherapy; guinea pig; bee sting; honey infusion

Introduction: Apitherapy is the use of bee products for therapeutic purposes. Within modern medicine, apitherapy has been applied to both human and veterinary medicine, with the applications ranging from wound healing to assistive oncologic treatment. The case described utilizes apitherapy, both bee stinging therapy and intravenous honey infusion, as adjunctive therapy to primary surgical mass removal for a cervical liposarcoma.

Case Description: Pablo, a 4.5-year-old male guinea pig, was presented to the KSU VHC as a referral for a suspected cervical abscess. Upon evaluation, surgical removal of the abscess was elected. During the surgical procedure, the mass was appreciated to be more condensed with more well-demarcated margins than expected. The mass was submitted for histopathological review and returned as an adipocyte neoplasm, most consistent with liposarcoma. The owner was offered multiple therapeutic options, including further surgical resection with radiation therapy, further surgical resection alone, active surveillance for mass regrowth, or apitherapy. The owner elected apitherapy.
The therapeutic plan was based on Batson 2025. Pablo presented for two treatments two weeks apart. Upon presentation, he was premedicated with 5 mg/kg diphenhydramine intramuscularly. He was sedated with 1 mg/kg midazolam, 6 mg/kg ketamine, and hydromorphone 0.3 mg/kg intramuscularly. An intravenous catheter was placed in the front cephalic vein to assist with the intravenous honey infusion. Pablo was maintained on isoflurane gas during the bee stinging therapy. A pilot hole was created in the previous surgical site to assist with stinger placement. Two bee stings were performed for 20 seconds per sting. Upon completion of therapy, Pablo was recovered and started on an intravenous Manuka honey therapy at 3 ml/kg. The honey infusion solution was composed of 1 part Manuka honey to 2 parts saline and placed on a fluid warmer to ensure that no crystallization was appreciated before administration. Prior to infusion therapy and every 20 minutes during the therapy, blood glucose measurements were performed to ensure that hyperglycemia or rebound hypoglycemia was not appreciated. During both treatments, Pablo recovered smoothly with no complications. During the initial treatment, Pablo's blood glucose reached 355 mg/dL, at which time infusion therapy was elected to be discontinued. Pablo is reported to be doing well at home with no evidence of regrowth and will be presenting in February for a survey CT scan to evaluate for tumor regrowth or metastasis.

Discussion: This case demonstrates the use of apitherapy as adjunctive therapy for a cervical liposarcoma in a guinea pig. Within guinea pigs, liposarcomas are characteristically locally invasive, rarely metastasize (have been reported to metastasize to the liver, lung, and bone), and surgical resection with radiation therapy is the treatment of choice. The use of apitherapy has been a documented therapeutic option for liposarcoma in a guinea pig. The use of intravenous honey possesses therapeutic properties of anti-proliferative effects, anti-inflammatory, and anti-tumor effects, while the use of bee stinging therapy additionally has anti-inflammatory effects. Overall, the use of apitherapy to help prevent tumor regrowth in a cervical liposarcoma demonstrates the potential for apitherapy to be used in conjunction with routine oncologic practices for exotic companion species.

Applied Science Poster Presentation – BI Atrium

Phi Zeta RESEARCH DAY
KANSAS STATE UNIVERSITY

Haitham Alenaemy

Evaluation of the Antimicrobial Activity of Clove Bud Oil and Cinnamaldehyde Against Major Liver Abscess-Causing Bacterial Pathogens in Cattle

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Keywords: Liver abscess; Feedlot cattle, Cinnamaldehyde, Clove bud oil, Antimicrobial acitivity

Liver abscesses (LA), a bacterial infection, are a major health and economic concern for the beef and dairy industries. The condition is typically initiated by *Fusobacterium necrophorum* invasion of the liver and is often complicated by co-infections with *Trueperella pyogenes* and or *Salmonella enterica*. High-grain diets predispose cattle to ruminal acidosis, increasing the risk of LA. Currently, tylosin phosphate is the most common feed additive used for prevention; however, the use has raised concerns regarding antimicrobial resistance (AMR) in cattle production systems. To explore natural alternatives, this study evaluated the *in vitro* antimicrobial activities of clove bud oil (CBO) and cinnamaldehyde (CIN) against key LA-causing pathogens, including *F. necrophorum* subsp. *necrophorum*, *F. necrophorum* subsp. *funduliforme*, *T. pyogenes*, and *S. enterica* serovar Lubbock, using four complementary assays: agar well diffusion, broth macrodilution, broth microdilution, and *in vitro* rumen fermentation. Across all assays, CIN demonstrated greater antimicrobial efficacy than CBO. In the agar well diffusion assay, CIN produced significantly larger, dose-dependent inhibition zones (all $P < 2.2 \times 10^{-16}$), with the highest susceptibility observed in *Fusobacterium* spp. and *T. pyogenes*. Broth microdilution confirmed CIN's potency, showing significantly lower minimum inhibitory concentrations (MICs) ($P = 3.45 \times 10^{-14}$) than CBO, requiring up to four-fold lower concentrations for complete growth inhibition. In the *in vitro* rumen fermentation assay, CIN (200–400 µL) rapidly and sustainably suppressed *Fusobacterium* populations within 12 hours ($P < 10^{-24}$), whereas CBO produced only partial inhibition. The efficacy of both compounds was reduced in the presence of lysine, indicating substrate-dependent effects. Collectively, these results demonstrate that cinnamaldehyde is a highly effective *in vitro* inhibitor of major LA-associated pathogens, exhibiting dose-dependent activity and superior performance compared to clove bud oil, highlighting its strong potential as a natural alternative feed additive for controlling liver abscesses in cattle.

Phi Zeta RESEARCH DAY
KANSAS STATE UNIVERSITY

Vani A.

Investigating the Evolutionary History and Recombination Events of Porcine Reproductive and Respiratory Syndrome Virus (PRRSV)

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Keywords: PRRS; recombination and field infection

Investigating the evolutionary history and recombination events of porcine reproductive and respiratory syndrome virus (PRRSV)

Introduction: Porcine reproductive and respiratory syndrome (PRRS) is the most devastating disease in the United States swine industry with PRRS virus (PRRSV) type 2 being the most predominant strain. Recombination is an important evolutionary mechanism in RNA viruses, and this study is an attempt to understand those evolutionary patterns in PRRSV and identify eventual recombination events between vaccine strains and field isolates of PRRSV type 2.

Methods: Forty-seven isolates of PRRSV collected during the period 2007-2021 were sequenced. The library preparation was performed using the Nextera XT DNA Library Prep kit, and 150 bp paired-end sequencing was performed on an Illumina MiSeq instrument. Sequences from six vaccine strains (Ingelvac MLV, Ingelvac ATP, Fostera PRRS, Prime Pac PRRS RR, Prevacent PRRS, and PRRSGard) were retrieved from NCBI and other laboratories. Bioinformatics analysis was primarily conducted using CLC Genomics Workbench 25.0.1. Following the removal of adapters, short reads, and low-quality sequences, reads were mapped to PRRSV 1 (NCBI Reference Sequence: NC_001961) and PRRSV type 2 (NCBI Reference Sequence: NC_038291) reference genomes. The optimal reference genomes, identified via BLAST, were then used for read mapping, and consensus sequences were extracted at 30X coverage. After the multiple sequence alignment and phylogenetic analysis of all the samples, vaccines, and some representative PRRSV sequences from NCBI, the samples were assessed for recombination using Simplot++ v1.3 and GARD from Datamonkey v2.0 based on the insights from the phylogenetic tree.

Results: Most field isolates were found within PRRSV type 2 correlating with geographic region and sample collection times. One clade corresponded to samples from Colorado, with sub-clades corresponding to different collection times. Two other distinct clades were observed, both corresponding to Kansas and adjacent Nebraska regions, again with sub-clades corresponding to different collection times. Two novel recombination events were identified during the present investigation, one between a vaccine and a previously sequenced field isolate found in NCBI, and another between two of the field isolates sequenced in this study. The two novel recombinants included GenBank ID-KT257984 - a recombinant between Sample-33 and Sample-45 with a single recombination breakpoint in the ORF3 region (occurred in the USA) and GenBank ID-PQ316100 – a recombinant between GenBank ID-OP866751 and Fostera Vaccine with two recombination breakpoints in the region of ORF1a and one each in the regions of ORF1b and ORF2 (occurred in China).

Conclusion: Monitoring PRRSV recombination by sequencing in the field is very crucial for developing effective disease management strategies and enhanced genomic surveillance will significantly aid the swine industry in combating this devastating disease.

Kylie Bitcon

Exploring the Impact of an Early Research Experience on Veterinary Career Paths

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Keywords: mentored veterinary research experience; graduate school; residency; peer-reviewed publications

Introduction: Summer veterinary research scholars programs (VRSP) provide valuable hands-on mentored research experience to preclinical veterinary students with exposure to a variety of career options, but little data exist regarding their impact. It was hypothesized that a higher proportion of veterinarians who participated in VRSP would pursue a graduate degree, internship, residency, or peer-reviewed publication than non-participants.

Methods: As a cohort study, an online survey was made available to veterinarians who graduated in 2000-2024 from Kansas State University, Mississippi State University, University of Missouri, or University of Wisconsin. Data were analyzed with logistic regression, with VRSP participation, school, and school-by-VRSP interaction as fixed effects.

Results: The survey was completed by 1,546 veterinarians, including 370 VRSP and 1,176 non-VRSP alumni. Participation in VRSP was associated with increased odds of pursuing a master's degree (OR 3.3, $P < 0.01$), internship (OR 2.3, $p < 0.01$), residency (OR 3.8, $p < 0.01$), peer-reviewed publication (OR 7.4, $p < 0.01$), PhD (OR 8.6, $p < 0.01$), and pursuing a career in academia (OR 2.7, $p < 0.01$). Additionally, more VRSP participants reported having a research component to their current career than non-participants (OR 3.5, $p < 0.01$).

Conclusion: Early exposure to research through VRSP is successfully impacting alumni careers, contributing to the education and advancement of veterinary professionals in research, clinical practice, and academia. Based on the favorable outcome measures from this study, continued support of these programs is essential for the matriculation of high-quality veterinary scientists.

Student Support: NIH T35 Program T35OD029981

Phi Zeta RESEARCH DAY
KANSAS STATE UNIVERSITY

Manickam Dhandapani

Artificial Intelligence and Machine Learning Tools for Antimicrobial Resistance Detection in Bacteria from Food Animal Production: A Systematic Review

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Keywords: Antimicrobial Resistance; Food animal production; Artificial Intelligence; Machine Learning

Antimicrobial resistance (AMR) is a major global health concern, particularly in food animal production systems where antimicrobial use is widespread. Foodborne pathogens and commensal bacteria associated with livestock can acquire and disseminate resistance genes through horizontal gene transfer and via contaminated farm environments, processing equipment, and interconnected agro-ecosystems. The spread of AMR within food animal systems poses significant risks to animal health, food safety, and public health. Current AMR surveillance relies largely on phenotypic antimicrobial susceptibility testing (AST), which is labor intensive and time consuming. We hypothesize that artificial intelligence (AI) and machine learning (ML) based tools can enhance AMR detection by enabling faster, scalable, and integrative analysis of complex biological and epidemiological data. The objective of this systematic review was to investigate and summarize the available tools for detecting AMR in food animals, with an emphasis on emerging computational approaches. A comprehensive literature search was conducted across PubMed, Scopus, and IEEE databases, covering studies published between 2010 and 2025. The searches yielded 2290, 3950 and 985 articles from PubMed, Scopus and IEEE, respectively, for a total of 7225 publications. Studies will be included if they reported the use of phenotypic, genomic, bioinformatics, and or computational approaches for AMR detection in food producing animals. Our systematic review identified a wide range of AMR detection tools, including phenotypic assays, whole genome sequencing based methods, rule based genomic tools such as AMRFinderPlus and ResFinder, and emerging AI and ML approaches. Phenotypic assays, including disk diffusion, broth microdilution, E-test, and agar dilution, provide direct information on bacterial resistance profiles but require substantial time, labor and laboratory resources. Genomic and bioinformatics tools allow rapid identification of known antimicrobial resistance genes from sequencing data; however, their dependency on curated reference databases limits their ability to detect novel or complex resistance mechanisms. Recent advances in AI and ML have created new opportunities for AMR prediction and surveillance in food animal production systems. AI and ML approaches, including random forests, support vector machines, XGBoost, neural networks, and deep learning models such as DeepARG, can analyze large scale genomic and metagenomic datasets, detecting subtle genomic signatures, predict emerging AMR traits, and providing actionable information for farm level surveillance and One Health initiatives. In conclusion, AMR detection in food animals increasingly depends on integrating phenotypic testing with genomic, bioinformatics, and AI and ML based tools. Integrating these tools can enhance surveillance accuracy and inform decision making to mitigate AMR. Future research should focus on scalable, robust platforms that combine phenotypic validation with genomic and AI and ML driven predictive models to address emerging antimicrobial resistance in livestock.

Chris Jung

Partner Insights on the Collaborative Impact of Outreach Care

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Keywords: Community Outreach; Outreach Services; Street Medicine

Partner Insights on the Collaborative Impact of Outreach Care

Chris Jung, Dr. Ron Orchard
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The purpose of this study was to build on an ongoing photovoice research project being done to explore barriers to veterinary care and community outreach services in unhoused populations of Topeka, Kansas. This part of the research explored the collaborative aspect of community outreach from the perspective of outreach partners. A focus group of three Mobile Access Partner (MAP) participants was created. This group consisted of one nurse, one nurse practitioner, both from Stormont Vail Mobile Services and a Deputy Director at the Topeka Rescue Mission. This focus group was created by reaching out to MAP partner participants for availability and interest in a focus group on a normally held “MAP” day at the Salvation Army of Topeka. Though thoughtful questioning, the focus group was asked four main questions regarding their role and impact in the communities they work with. These interactions were recorded, and later a inductive thematic analysis was performed on the recordings and consistent themes emerged: the foundational nature of trust in all levels of outreach services; using outreach in response to systemic neglect and against strategies of keeping those experiencing homelessness invisible; the strength of the human-animal bond and it’s use as a pathway for future services; the challenges of transitioning to long-term housing in the absence of community; using outreach to combat burnout and compassion fatigue; scalability and future of outreach services; and changing opinions and stigmas around homelessness. Results of the focus group demonstrated multiple and very personal motivations of partner participation in caring for the unhoused and highlighted methods to increase impact and care strategies of community outreach care.

Research Grant: ASPCA
Student Support: Boehringer Ingelheim Veterinary Scholars Program

Phi Zeta RESEARCH DAY
KANSAS STATE UNIVERSITY

Hayden Kunnary

Title Evaluation of Machine Learning Models for TT-POCUS Based Prognostic Prediction of Did-Not-Finish in High-Risk Feedyard Cattle Diagnosed with Bovine Respiratory Disease

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Keywords: Bovine pneumonia; thoracic ultrasound; point-of-care; prognosis; chute-side

Background: Bovine respiratory disease (BRD) remains the leading cause of morbidity, mortality, and economic loss in U.S. feedyard cattle. Clinical examinations and visual appraisal are routinely used to identify cattle requiring treatment, yet these approaches have limited ability to predict long-term outcomes, particularly in animals that have already received multiple treatments. Early identification of cattle at increased risk of becoming did-not-finish (DNF; those that are unable to finish a 60-day post evaluation window) is critical for guiding decisions about recoverable versus non-recoverable animals, optimizing treatment strategies, allocating resources efficiently, and improving animal welfare. Targeted thoracic point-of-care ultrasound (TT-POCUS) provides objective, chute-side assessment of pulmonary pathology and has demonstrated associations with health outcomes, highlighting its potential value as a prognostic tool.

Objective: Determine accuracy machine learning models incorporating TT-POCUS derived variables to predict DNF outcomes compared to actual outcomes in feedyard cattle with history of multiple BRD treatments and currently showing clinical signs.

Methods: TT-POCUS data were collected during chute-side examinations of feedyard cattle with multiple treatment (n=151) and used as input features for supervised classification models developed in Python. Algorithms evaluated included logistic regression, decision tree, random forest, gradient boosting, perceptron, and neural network models. Model performance was assessed using area under the receiver operating characteristic curve (AUC), overall accuracy, sensitivity, specificity, positive predictive value (PPV), negative predictive value (NPV), and F1 score. Performance metrics were compared to evaluate model discrimination, balance between sensitivity and specificity, and practical prognostic utility.

Results: Model performance varied across algorithms, with AUC values ranging from 0.54 to 0.84. Overall accuracy ranged from 0.58 to 0.76, with the perceptron and random forest models achieving the highest accuracy (0.76), indicating that these models correctly classified most cattle. Positive predictive value (PPV) ranged from 0.73 to 0.84, reflecting the models' ability to correctly identify cattle that ultimately became DNF when classified as high risk. Negative predictive value (NPV) ranged from 0.43 to 0.70, with the random forest and perceptron models demonstrating the strongest performance for correctly identifying cattle likely to finish. F1 scores, reflecting the balance between precision (PPV) and recall (sensitivity), were highest for the random forest (0.83) and perceptron (0.82) models, indicating robust performance in identifying DNF cattle while limiting misclassification.

Conclusion: Machine learning models leveraging TT-POCUS derived variables demonstrate clinically relevant prognostic performance for predicting DNF outcomes in high-risk feedyard cattle. High accuracy, along with PPV, NPV, and F1, underscores the models' ability to correctly identify both cattle likely to fail and those likely to finish. These metrics highlight the practical utility of TT-POCUS based models for informed chute-side decision making, enabling early identification of high-risk cattle and supporting recoverable versus non-recoverable treatment decisions.

Phi Zeta RESEARCH DAY
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Jonana Magoga

Effects of Pharmacological Zinc Withdrawal Programs on Growth Performance and Plasma Zinc in Nursery Pigs

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Keywords: Growth performance; nursery pigs; plasma Zn; Zn oxide.

Background: Zinc oxide (ZnO) is commonly fed at pharmacological levels to nursery pigs to enhance growth and reduce post-weaning diarrhea, but abrupt removal can impair feed intake and gut function. Implementing a gradual ZnO withdrawal strategy could mitigate these effects; however, limited data are available. This study evaluated the impact of different pharmacological Zn withdrawal programs on growth performance and plasma Zn concentration of nursery pigs.

Methods: At weaning, 360 pigs (initially 12.5 ± 0.06 lb) were allotted to one of six dietary treatments (12 replicate pens with 5 pigs per pen). Treatment diets were fed in three phases (phase 1, d 0 to 11; phase 2, d 11 to 24; phase 3, d 24 to 45), followed by a common Phase 4 diet (d 45 to 55). Treatments included: (A) no pharmacological Zn (110 mg/kg Zn provided in the premix); (B) 3,000 mg/kg Zn in Phase 1, then 110 mg/kg for phases 2 and 3; (C) 3,000 mg/kg in Phase 1, 2,000 mg/kg in Phase 2, then 110 mg/kg in phase 3; (D) 3,000 mg/kg in phase 1, 2,000 mg/kg in phase 2, and 1,000 mg/kg for 7 d in phase 3 followed by 110 mg/kg for the remainder of phase 3; (E) 3,000 mg/kg in phase 1, 2,000 mg/kg in phase 2, and 1,000 mg/kg for 14 d in phase 3 followed by 110 mg/kg for the remainder of phase 3; and (F) 3,000 mg/kg in phase 1, 2,000 mg/kg in phase 2, and 1,000 mg/kg throughout phase 3. Growth performance was measured and blood was collected from one pig per pen on days 0, 11, 24, and 45 to measure plasma Zn concentration.

Results: During phase 1, pigs fed pharmacological Zn had increased ADG ($P < 0.001$), ADFI ($P = 0.005$), and G:F ($P = 0.008$). In phase 2, ADG ($P = 0.005$) was lowest, and ADFI tended to be lowest ($P = 0.065$) for the treatment previously fed 3,000 mg/kg and then reduced to 110 mg/kg of Zn. No treatment differences were observed during the total 3-week phase 3 feeding period, nor the common period ($P > 0.10$). Body weight (BW) was greater ($P < 0.05$) on d 11 and 24, and tended to be greater ($P < 0.10$) on d 31 and 38 for pigs that remained on the higher Zn treatments. For the overall study (d 0 to 55), ADG, ADFI, G:F, and BW on day 55 did not differ between treatments. Plasma Zn concentrations did not differ among treatments at d 0 as expected, but were greater in pigs fed pharmacological Zn at d 11 ($P < 0.001$). On days 24 ($P < 0.01$) and 45 ($P = 0.002$), plasma Zn concentrations were higher in pigs receiving pharmacological Zn for longer durations.

Conclusion: While pharmacological Zn improves early nursery performance, abrupt withdrawal subsequently impairs growth. Gradual withdrawal strategies maintain higher plasma Zn concentrations, but the initial growth advantages were not sustained through the end of the nursery period.

Shelby Noel

Variations in Sperm Morphology Across the Slide

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Keywords: Cattle; Sperm; Morphology; Bull

Bull breeding soundness exams are an important tool for identifying subfertile bulls, with sperm morphology serving as a key determinant of reproductive potential. It is recommended that morphological assessments use eosin-nigrosin stained semen smeared on to a slide and evaluated at 1000x magnification. The percentage of morphologically normal sperm is typically determined by recording the number of morphologically normal cells out of 100-200 total cells within a single region. However, this method does not account for variation across different regions of the slide. The objective of this study is to determine if sperm morphology percentage varies significantly across regions of the semen smear. We hypothesize that sperm morphology percentages will vary depending on the region of the slide being examined. To investigate this, cover-slipped smears of stained semen were divided into three equal sections, from the inoculation site to the end of the viable sample (defined by either the cover slip boundary or sample endpoint). Each section was evaluated independently by trained examiners with non-target sections masked and each zone assigned a unique identifier to ensure blinding. Preliminary results showed many slides had similar percentages of normal/abnormal morphology across zones, though numerical discrepancies were observed. Notably, several slides yielded “failing” results in one zone and “passing” in another based on a cut-off of 70% morphologically normal sperm. These findings highlight a potential source of variability in sperm morphology assessments. Statistical analysis of the results will determine the model adjusted effect of slide region on sperm morphology.

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Amanda Roth

Evaluating Fecal Diagnostics for *Echinococcus Multilocularis* in Coyotes

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Keywords: Taenia; Echinococcus multilocularis; zoonotic; tapeworm; PCR

Echinococcus multilocularis is a zoonotic tapeworm expanding southward in North America. It causes severe disease in humans and dogs, and its eggs are microscopically indistinguishable from those of non-zoonotic Taenia species. The gold standard for diagnosis is post-mortem examination of canine intestines for adult tapeworms; this is clinically impractical. Therefore, a non-invasive test with equivalent sensitivity and specificity is needed. Fecal and intestinal samples from ethically harvested coyotes in four Midwestern states were collected for egg detection and DNA extraction. Intestinal tracts were examined using the sifting, filtration, and counting technique (SFCT) to recover taeniid adults. Feces were processed by centrifugal fecal flotation, followed by sedimentation, to detect taenia-type eggs. Copro-PCR, targeting taeniid-specific genes, was also conducted. Adult taeniids were recovered from 151 of 204 coyotes. Of the157 fecal samples tested by flotation and sedimentation, 113 were from coyotes confirmed to harbor adult taeniids. Flotation detected taenia-type eggs in 43 of 113 samples (38% sensitivity), and sedimentation detected eggs in 46 of 113 (40%). Only 89 fecal samples had sufficient material for PCR, of which 62 were adult-positive. Within this subset, PCR detected taeniid DNA in 20 samples (32% sensitivity). Combining all three methods increased detection to 51%, although this was still lower than the recovery rate for adults. PCR was likely limited by poor DNA extraction from resilient taeniid eggs. Overall, post-mortem adult cestode recovery remains the most sensitive and specific method, highlighting the need for improved fecal diagnostics in surveillance of *Echinococcus spp.*

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Brooke Shenkenberg

Application of Bioreactor Technology for Swine Inactivated Streptococcus suis Vaccine Production

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Keywords: Bioreactor; Streptococcus suis; Inactivated Vaccine

Streptococcus suis (S. suis) is a Gram-positive bacterium that causes considerable economic loss in the swine industry worldwide. It is endemic in nearly all countries with mortality rates up to 65% during outbreaks. No broadly effective vaccines are commercially available for S. suis. To address outbreaks, custom-made S. suis vaccines for affected farms or areas are frequently developed to obtain targeted treatment against this disease. This kind of vaccine production, proven more effective against S. suis, involves the isolation, cultivation, purification and bacterial inactivation of herd-specific strains. The inactivated S. suis is formulated with vaccine additives or adjuvants that magnify protective immune responses for more effective vaccines.

Fermenters or bioreactors are used to upscale bacterial cultivation for vaccine production. These biomanufacturing devices provide an environment that support optimal bacterial growth by providing precise control and monitoring, including pH, temperature, oxygen and nutrient supply. Using a 3-liter bioreactor, we cultivated a S. suis serotype 2 isolate with the goal of optimizing S. suis bacterial growth with minimal additives for simpler downstream processing in herd-specific inactivated vaccine production. Initial fed-batch bioreactor run parameters include the use of Todd-Hewitt broth (THB) at 37 degree Celsius temperature, pH maintained at 7.4 with sodium hydroxide, 25 rpm impeller agitation and 100% compressed air at 0.3 standard liter per minute (SLPM) for aeration, no anti-foam addition and various glucose amount supplementation for 24-hour bioreactor cultivation. Optical density at 600 nm (OD600) and colony-forming unit (CFU) were performed to determine bacterial growth. Further optimization of S. suis bioreactor culture parameters resulted to shortening cultivation period to 16 hours with OD600 up to 2.7 and 1.5 x 10^13 total CFU in a single 3-liter bioreactor run. This amount is equivalent to 15,000 doses at 1 x 10^9 CFU per dose. These results show potential in the use of bioreactors in the rapid and efficient production of inactivated vaccines in event of S. suis disease outbreaks.

Katherine Shirley

Assessment of Bovine Respiratory Disease Treatment Outcomes in Stocker Cattle Following 3- or 6-Day Post-treatment Intervals Using Pradofloxacin for Initial Treatment

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Keywords: Antimicrobial; Bovine Respiratory Disease; Post-treatment Interval

The number of days between initial and subsequent respiratory disease treatments could impact antimicrobial use and cattle health outcomes; however, an optimal post-treatment interval (PTI) has not been identified for pradofloxacin. The objective was to determine the effect of 3- or 6-day PTI on first treatment failure (FTF), case fatality risk (CFR), chronic risk (CR), and days until death (DTD) following pradofloxacin treatment for bovine respiratory disease (BRD). A randomized controlled trial was conducted at a commercial stocker operation in Kansas. Cattle identified with BRD (n=400) were randomized to a 3- or 6-day PTI and followed for 60 days to monitor health outcomes. Generalized linear mixed effects models were used to determine potential effects of PTI, while accounting for weight, days on feed, and metaphylaxis status on treatment outcomes. No differences were detected in FTF, CFR or DTD between PTI groups. Cattle with a 3-day PTI had a higher probability of CR, compared to 6-day PTI (28% ± 6% vs 18% ± 4%, respectively; P= 0.01). This study concluded that cattle assigned to a 3-day PTI have a greater probability of reduced success following initial treatment. Further research is needed to determine the optimal post-treatment window for cattle with BRD treated with antimicrobials.

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Kendra Siefker

Intraruminal Inoculations of Bacterial Pathogens to Induce Liver Abscesses in Steers

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Keywords: Beef; feedlot; liver abscess; microbiology

Liver abscesses, a notable economic concern to the beef industry, reduce feed efficiency and carcass value. High-grain diets induce ruminal acidosis, damaging ruminal epithelium, allowing bacterial transfer from the rumen to the liver initiating abscess formation. Fusobacterium necrophorum subsp. necrophorum (FNN) is the primary causative agent. Secondary pathogens, F. necrophorum subsp. funduliforme (FNF), Trueperella pyogenes (TP), and Salmonella enterica (SE), are frequently isolated from abscesses. However, their independent role in abscess formation is unknown. In this study, we examined their ability to induce liver abscesses and evaluated the prevalence of these species throughout the digestive tract. Forty steers, split into four groups, were adapted to a high-grain diet. Each group was inoculated into the rumen with FNN (positive control), FNF, TP or SE on days 0 and 7. On day 27, steers were necropsied to collect livers, gut contents and tissues. Samples were analyzed for bacterial prevalence and concentrations using culture- and qPCR-based methods. Liver abscesses occurred in only one steer in the FNN group (8.5 Log10 CFU/g). In FNF and SE groups, inoculated species were detected in healthy livers of 1 and 3 steers, respectively. The prevalence of FNN and FNF were greater in ruminal epithelium and contents than the SE and TP. Prevalence of FNN and FNF were similar between healthy rumen and rumenitis tissue. The SE prevalence was low in all tissues. TP was not detected in any samples. Overall, we found inoculated FNN and FNF invaded the gut tissues, but low abscess prevalence in the positive control, rendered the study inconclusive on secondary pathogens' ability to independently cause liver abscesses.

Regan van den Elsen

Assessment of Canine Cancer Risk and Sites of Superfund and PFAS Contamination in Kansas

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Keywords: Canine; Cancer; Sites; Contamination; Kansas

Companion animals make excellent sentinel species for their human counterparts due to their shared environments. Per- and polyfluoroalkyl substances (PFAS) have recently been linked to several types of human cancer. Superfund sites are identified by the Environmental Protection Agency (EPA) as contaminated sites risking human health. By understanding the distribution of canine cancer in Kansas we looked to highlight areas of high cancer prevalence and determine if there is a visual association with the known PFAS contamination sites (n=30) and Superfund sites (n=19).
We hypothesized that canine cancer cases are clustered and there is an association between increased cases and the location of contaminated sites. Data on histological diagnoses of carcinomas, sarcomas, and mast cell tumors in dogs from 2021-2024 were collected from the K-State Veterinary Diagnostic Lab (n =4486, 22.02% carcinomas, 38.43% sarcomas, 39.55% mast cell tumors). The 3 most reported breeds were the Labrador Retriever, Boxer, and Pit Bull Terrier. The median age was 9 years old (range 0-24 years).
In this preliminary study, ArcGIS was used to create choropleth maps to visualize the location of increased cancer cases by zip code. Spatial autocorrection showed the distribution of cases was clustered. The GIS maps of raw and population normalized canine cancer cases were visually evaluated for overlap with the locations of the Superfund and PFAS contaminated sites. Areas of increased canine cancer prevalence were best associated with contaminated sites in the raw data. Our next step is to detect the presence of contaminants such as PFAS in dogs from potential hotspots identified in this study.

Basic Science Poster Presentation – BI Atrium

Ashton Foster

Sex and Circulating Adiponectin Levels Regulate Expression of Adiponectin R1 Receptors in Skeletal Muscle Resistance Arterioles

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Keywords: Adiponectin; Microcirculatory; Arterioles

Adiponectin signals through two transmembrane receptors, adiponectin R1 (AdipoR1) and adiponectin R2 (AdipoR2). In the endothelium and vascular smooth muscle, both AdipoR1 and AdipoR2 are expressed, and contribute to adiponectin signaling. The purpose of the current work was to investigate the relationship between circulating adiponectin and expression of adiponectin receptors in skeletal muscle arterioles. To acutely reduce adiponectin, Adiponectin Tam-Cre/Lox mice were fed a tamoxifen-containing diet for either 1 or 3 weeks. One week of tamoxifen feeding reduced circulating adiponectin by 54% and 52%, in male and female mice, respectively. 3 weeks of tamoxifen feeding reduced circulating adiponectin by 86% and 94%, in male and female mice, respectively. In mice with full circulating adiponectin (no tamoxifen) and partial reduction of circulating adiponectin, AdipoR1 levels were lower in arterioles from female mice as compared to those of male mice ($p<0.05$). Partial reduction of circulating adiponectin (1 week of tamoxifen) reduced AdipoR1 expression in arterioles from both male and female mice, but this reduction did not reach statistical significance ($p=0.06$), but did not change AdipoR2 expression. Surprisingly, nearly complete reduction of circulating adiponectin (3 weeks of tamoxifen) did not alter AdipoR1 expression in arterioles from either female or male mice. These results suggest that AdipoR1 expression in skeletal muscle arterioles is related to circulating adiponectin levels, but the relationship is not linear, suggesting that other physiological parameters, such as sex hormones, act in concert with adiponectin to regulate AdipoR1 expression in skeletal muscle arterioles.

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Xinyu Fu

Generation of Recombinant Adeno-Associated Virus as Vaccine Against Canine Distemper

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Keywords: Canine Distemper; Vaccine; Recombinant Adeno-Associated Virus; Serotype; Protective Antigen

Introduction
To address the persistent global challenge of canine distemper virus (CDV) outbreaks—including cases in vaccinated dogs—and to overcome the antigenic divergence between existing America-1 lineage vaccines and circulating wild-type CDV strains, we propose developing a novel, single-dose vaccine utilizing an adeno-associated virus (AAV) vector to deliver protective antigens from a contemporary CDV isolate. We hypothesize that such an AAV-vectored vaccine will elicit strong and long-lasting immunity in dogs, offering effective protection against emerging epidemic CDV strains.

Methods
In this study, we selected the Hemagglutinin (H) and Fusion (F) protein genes from a recent field CDV strain and cloned them into AAVpro helper free series, which allowed for the preparation of recombinant AAV (rAAV) particles of serotypes 2, 5, 6, 8, or 9. Following transfection of AAVpro 293T cells with these constructs, rAAVs were harvested 4–7 days post-transfection, and their viral titers were quantified by qPCR. To identify the most efficient AAV serotype for gene delivery in canine cells, various canine cell lines were transduced with equal amounts of rAAVs, and H and F protein expression were evaluated by western blotting of cell lysates and supernatants.

Results
We successfully constructed AAV-CDV-H and AAV-CDV-F recombinant constructs and rescued the corresponding recombinant AAVs. The transduction efficiency of AAV serotypes 2, 6, 8, and 9 was evaluated in canine muscle, trachea, and thymus cell lines. Among the tested serotypes, AAV2 exhibited the highest expression level of CDV-H protein across all three canine cell types, indicating superior transduction efficiency.

Conclusion
These findings identify AAV2 as the most efficient AAV serotype for delivering CDV antigens in canine cells. This study provides a strong foundation for future in vivo evaluation and supports the development of a single-dose AAV-vectored vaccine against canine distemper.

Emmalyn Greeves

Glioblastoma U251 Cells Express Opioid Receptors

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Keywords: U251 cells; Astrocytes; RT-PCR; Western blotting; immunocytochemistry.

Abstract: Glioblastoma (GB) is a type of central nervous system cancer originating from glial cells. It is a very aggressive rapidly growing brain tumor. More than 13,000 Americans are diagnosed with GB every year. When in treatment, GB patients receive pain medications such as oxycodone or morphine for severe headaches. GB is highly invasive. Oxycodone is known to induce proliferation of breast and colon cancer cells. In the present study, our overarching goal is to understand if oxycodone has similar effects on GB cell proliferation. In order to respond to oxycodone and morphine, expression of opioid receptors by GB is a pre-requisite. In the present study, we used U251 cells derived from a malignant astrocytoma of a 75-year-old male patient to examine expression of opioid receptors (mu, kappa, and delta receptors abbreviated as MOR, KOR, and DOR), and salient astrocyte specific markers. Our data showed that U251 cells express astrocyte specific markers (GFAP, vimentin, and glutamate synthetase); and MOR and KOR. Results of this study will be presented at the Phi Zeta 2026 meeting.

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Claire Horton

Determining the Prevalence of Influenza A in Bovine Species Through Competitive Enzyme-Linked Immunosorbent Assay and Polymerase Chain Reaction Methods

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Keywords: avian influenza A; highly pathogenic avian influenza A virus; bovine; seroprevalence; enzyme-linked immunosorbent assay; polymerase chain reaction

Recent outbreaks of H5N1 highly pathogenic avian influenza A virus (HPAI) in ruminant species, especially dairy cows in the United States have caused various public and veterinary health concerns. Infection in dairy cows can result in severe diseases, and the virus may infect many other ruminant species. This study investigated the seroprevalence of antibodies against H5N1 in ruminants submitted for diagnostic testing through the Kansas Veterinary Diagnostic Laboratory. A total of 1,237 ruminant serum samples were collected between 2023 and 2024 including 641 samples from dairy cows and 596 from goats, sheep, and deer. The test was performed using 2 commercial competitive enzyme-linked immunosorbent assay (ELISA) testing kits, A and B. Additionally, polymerase chain reaction (PCR) tests for HPAI targeting the M-gene were performed on milk and nasal swabs from dairy cows. Among all serum samples, 0.40% (5/1,237) tested positive on test A, while 0% were positive on test B. Of the dairy cows, 0.31% (2/641) were positive on test A; none tested positive on test B. Among goats, sheep, and deer, 0.50% (3/596) were positive on test A, and 0% on test B. In PCR testing, 0.08% (3/3,941) of samples from dairy cows returned positive results. The results demonstrate that HPAI virus can be detected in ruminants by PCR and antibodies to influenza A can be determined by ELISA in different samples. With the 2 ELISA testing kits, a difference in sensitivity was found, highlighting the importance of using a test with an adequate sensitivity rate for surveillance testing. The difference in the PCR results and the ELISA results indicate a higher prevalence of active influenza infection verses animas that have seroconverted. This highlights the importance for continuing surveillance for HPAI in ruminant species, particularly dairy cows, as viral adaptations could increase zoonotic potential and increase spread of the virus.

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Zane R. Kohl

Susceptibility of U.S. Domestic Sheep to Nairobi Sheep Disease Virus

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Keywords: Viruses; Animal Diseases; Molecular Diagnostics; Sheep

Introduction: Nairobi sheep disease virus (NSDV), also known as Ganjam virus, is a tick-borne pathogen of the Orthonairovirus genus and Bunyaviridae order. The tri-segmented NSDV genome is approximately 18-19 kb and consists of small (S), medium (M), and large (L) negative-sense RNA segments. NSDV causes Nairobi Sheep Disease (NSD) in sheep and goats, characterized by acute hemorrhagic gastroenteritis with high fever, abortion, and mortality approaching 90% in naïve populations. Indigenous flocks in India reportedly have reduced mortality. NSD outbreaks caused devastating economic losses. NSDV circulates in the Haemaphysalis longicornis tick, a vector newly established in the United States, thus elevating concerns of the introduction NSDV into naïve small ruminants in North America. Despite its global significance, experimental studies evaluating the susceptibility and pathogenesis of NSDV in ruminants are limited, as are research tools such as validated molecular diagnostic methods, thus making virus detection and the understanding of pathogenesis challenging. Understanding the susceptibility of U.S. domestic sheep is critical to inform further surveillance, outbreak preparedness, and biosecurity measures, given the expanding vector range and the risk of damage to naïve livestock. The American Polypay sheep utilized are a 4-breed composite representing common breeds in the U.S. The purpose of this study was to evaluate the susceptibility and disease progression of NSDV in US domestic sheep and develop diagnostic tools for pathogenesis studies and surveillance programs.

Methods: NSDV is a foreign animal disease, and this pilot study was performed in the Biosecurity Research Institute, a BSL3 containment facility at Kansas State University. Three groups of Polypay sheep (n=12) were intradermally inoculated with Ganjam virus (IG619 strain) at 10^2, 10^3, or 10^4 PFU. Daily monitoring using standardized scoring assessing temperature, diarrhea, appetite, hydration, and overall condition was used to determine humane endpoints. Daily clinical sampling included, blood and nasal, oral, and rectal swabs. Hematology, serum chemistry, and coags evaluate host physiological responses throughout the study, and extensive postmortem pathological analysis at study endpoints assess disease severity. Virology endpoints include RT-PCR testing, virus isolation, and serology. An evaluation of magnetic particle extraction methodology allowed for successful recovery of viral RNA from inactivated, non-infectious samples released from the BSL3 facility. Research goals warranted the design and development of new PCR assays validated in this study.

Results: All animals developed high persistent fevers, 3-4 days post challenge (DPC), and succumbed to their infection reaching humane euthanasia endpoints between 6 and 8 DPC. Hemorrhagic diarrhea was infrequent. All animals developed severe pancytopenia (3-4 DPC), coagulopathy (5 DPC), and terminally elevated hepatic and renal biochemical markers. Early pathological assessment identified mild to moderate, multifocal mucosal hemorrhages throughout the

gastrointestinal tract and myocardial hemorrhage and icterus. The development, validation, and research tools for the determination of virological endpoints are ongoing.

Conclusions: This study demonstrates that Polypay sheep are susceptible to NSDV, and regardless of dose administered, developed severe acute systemic febrile illness. Introduction of this pathogen into small ruminants and permissive vectors in North America could result in significant clinical and economic consequences. Ongoing work is necessary for continued understanding of viral pathogenesis, successful vaccine development, and the development of diagnostic and surveillance tools to mitigate risk and damage caused by this pathogen globally.

Ji Yoon Kim

Reprogramming Replication Fidelity for One-to-Stop Vaccine Development

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Keywords: Herpes Simplex Virus; Vaccine; one-to-stop codons

Introduction: Herpes simplex virus type 1 (HSV-1) is a highly prevalent human pathogen, estimated to infect approximately two-thirds of the global population. Although infections are often mild, HSV-1 infection can cause severe diseases, including encephalitis and keratitis, underscoring the need for an effective vaccine. However, despite more than 70 years of intensive research, no HSV-1 vaccine has been clinically approved. Repeated failures of subunit and inactivated vaccines in the past have shifted the field towards live-attenuated vaccine (LAV) development strategies, which are well suited to elicit a strong, diverse and long-lasting immune responses. Recently, novel LAV candidates using the one-to-stop (OTS) strategy have been developed against RNA viruses. By introducing synonymous codon changes that are one random mutation away from a stop codon, this strategy increases the likelihood of premature termination, thereby reducing viral fitness while retaining immunogenicity. Here, we present a novel HSV-1 vaccine candidate that applies the OTS strategy by exploiting the error-prone replication of exonuclease-deficient HSV-1 mutants.

Methods: Using en passant mutagenesis, a two-step markerless Red recombination system, we generated exonuclease-deficient HSV-1 mutants, EAFL and DAEAQH. UL52, which encodes the primase subunit of the HSV-1 helicase-primase complex, was replaced with recoded UL52 containing one-to-stop codons targeting serine and leucine codons (SL OTS) or all possible codons (ALL OTS). Wild-type and recombinant viruses were rescued in Vero cells and serially passaged ten times. Viral phenotypes were assessed by plaque size assays and multistep growth kinetics (MOI = 0.001). Viral mutation rates and genetic stability of the recoded region were assessed by next-generation sequencing.

Results: End-point viral titers of UL52-recoded HSV-1 were significantly reduced compared to HSV-1 harbouring native UL52, demonstrating the attenuating effect of the OTS strategy. These effects were especially pronounced in the error-prone, exonuclease-deficient viruses EAFL and DAEAQH. Increased viral titers of OTS mutant viruses were observed after ten passages but did not fully return to wild-type HSV-1 levels. Reduced plaque size and impaired growth kinetics further confirmed attenuation of OTS mutant viruses. Sequencing analyses revealed increased mutation rates in exonuclease-deficient viruses EAFL and DAEAQH, while demonstrating genetic stability of the recoded UL52 region.

Conclusion: The OTS method is an effective approach for generating live-attenuated vaccine candidates for RNA viruses. However, the high-fidelity replication machinery of DNA viruses has limited the applicability of OTS strategies that rely on spontaneous mutations for attenuation. Here, we demonstrate that exonuclease-deficient viruses can circumvent this barrier. Exonuclease-deficient HSV-1 exhibits elevated mutation rates and, when combined with OTS-recoded UL52, shows highly attenuated replication in vitro. Together, these findings indicate that the OTS strategy can be extended to DNA viruses and represents a promising approach for the long-overdue development of an HSV-1 vaccine.

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Shristy Budha Magar

Sex-Specific Effects of Obesity on Fecal Microbiota During Acute Low- and High-Dose Influenza A Virus Infection in Mice

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Keywords: Diet-induced obesity; influenza A virus; gut microbiota; sex difference

INTRODUCTION: Influenza A viruses (IAVs) cause seasonal epidemics with potential threat to cause pandemics. Obesity is associated with alterations in gut microbiomes, which impact influenza pathogenesis. The objective of this study was to investigate sex-specific differences in (i) gut microbiome of obese and non-obese mice, and (ii) gut microbial alterations caused by the lethal or sublethal doses of H1N1 IAV during the acute phase of virus pathogenesis.

METHODS: Male and female C57BL/6 mice were treated with either a control diet (CD, 10kcal% fat) or high-fat diet (HFD, 60kcal% fat) for 13 to 14 weeks. Diet-induced non-obese and obese, male and female mice were infected either with a high-dose (10^3 $TCID_{50}$) or a low-dose ($10^{1.5}$ $TCID_{50}$) of mouse-adapted A/California/04/2009 H1N1 IAV. Fecal pellets were collected before infection (i.e., baseline) and at 3 days post-infection (dpi), and the V3-V4 region of the 16S rRNA gene was amplified and sequenced (Illumina MiSeq). Raw reads were processed using quantitative insights into microbial ecology 2 (QIIME2) and R software. Taxonomy was assigned with SILVA database. The alpha diversity was measured using Kruskal-Wallis test and beta diversity was measured using principal coordinates analysis (PCoA) and significance was measured at $p<0.05$.

RESULTS: First, we compared the sex-specific effects of obesity. Before virus infection, males with obesity had significantly higher Firmicutes (90.41% vs 73.01%) and F/B ratio; lower Actinobacteria (3.32% vs 11.38%); and different beta diversity compared to non-obese males. In females, obesity was associated with significantly lower Actinobacteria (2.27% vs 7.45%) and different beta diversity. After a high-dose infection, no significant phylum-level changes from baseline were observed at 3 dpi in males with obesity. However, in females with obesity, Proteobacteria were significantly decreased (0.05% vs 0.26%). We did not see any significant difference in Firmicutes/Bacteroidetes ratio (F/B), alpha diversity and beta diversity in both male and female with obesity after high-dose infection. After a low-dose infection, males with obesity had significantly decreased Actinobacteria (2.79% vs 3.44%) and F/B ratio, and increased Verrucomicrobiota (1.40% vs 0.41%). The Shannon index was significantly increased in males with obesity. Although we did not see any significant changes in phylum, Chao1 for richness and observed indices were significantly decreased in females with obesity. We also did not find any significant difference in beta diversity in both and male and female mice with obesity after low dose infection.

CONCLUSION: Our data suggest that both sex and obesity influence alterations in gut microbiota during the acute phase of low- and high-dose IAV infection. Although microbial changes during high-dose remained largely stable, low-dose infection led to pronounced microbial alterations. These insights may guide sex-specific strategies for improving immune and metabolic health in obesity.

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Molly Millett

Amplifying the Voices of the Unhoused: Exploring Barriers and Trust in Veterinary Care with Photovoice

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Keywords: Photovoice; Spectrum of care; Human-animal bond

This study explores the lived experiences of unhoused individuals who receive veterinary and outreach services while caring for companion animals. Through a photovoice methodology, participants used disposable cameras to document daily life with their pets, offering visual testimony to the barriers and adaptations that define their relationship to care. Ten participants were selected during outreach events hosted by Street Dog Coalition, a transdisciplinary program providing veterinary and social services to unhoused pet owners, in Topeka, Kansas. Over 200 photographs were collected and analyzed using thematic qualitative methods. The images revealed patterns of mutual caregiving, with pets serving as emotional anchors, protectors, and motivators for survival. Themes of shelter instability, institutional exclusion, and resource scarcity were common, yet participants also documented moments of joy, pride, and resilience. Tents became symbols of domesticity, forest edges transformed into safe havens, and dogs emerged as central to participants' identity and routine. This visual narrative highlights both the systemic neglect faced by unhoused populations and their creative, compassionate strategies for care. The findings underscore the importance of integrating animal companionship into service models and policy, and demonstrate the value of image-based inquiry in amplifying marginalized voices.

Maxwell Parr

Acute Deletion of Adiponectin Increases Expression of Sphingosine-1-Phosphate Receptors in Skeletal Muscle Arterioles

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Keywords: Microcirculation; Homeostasis; Adiponectin; Sphingosine-1

In the endothelium, adiponectin has been shown to modulate sphingolipid metabolism through ceramidase activity, increasing sphingosine-1-phosphate (S1P) levels, supporting vascular homeostasis. We have shown that pharmacological inhibition of S1P synthesis in skeletal muscle arterioles from wild-type (WT) mice recapitulates impaired endothelium-dependent flow-induced dilation observed in skeletal muscle arterioles from acute AdipoKO mice. Because impairment of flow-induced vasodilation in arterioles from acute AdipoKO may be due to loss of signaling through the ceramide-S1P-S1P receptor axis, we evaluated expression of S1P receptor 1 (S1PR1) in skeletal muscle arterioles from male and female WT and AdipoKO mice using fluorescent immunohistochemistry. 3 weeks of tamoxifen feeding reduced circulating adiponectin by 86% and 94%, in male and female mice, respectively. Expression of S1PR1 tended to be lower in arterioles from female WT mice as compared to those from male WT mice ($p=0.10$). Surprisingly, acute knockout of adiponectin increased expression of S1PR1 in arterioles from both male and female mice, with a greater increase occurring in arterioles from female mice. These results suggest that removal of circulating adiponectin, and downregulation of S1P signaling may result in a compensatory upregulation of S1PR1 expression. Further work is needed to determine how this increase in S1PR1 influences vascular reactivity and vascular homeostasis in the skeletal muscle microcirculation.