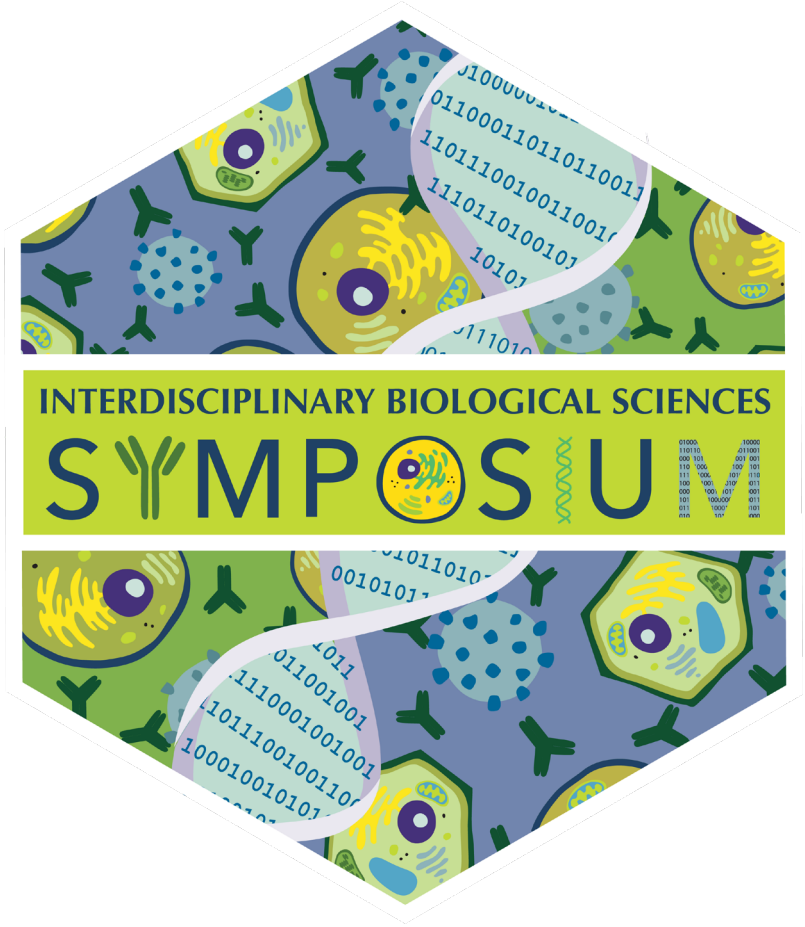
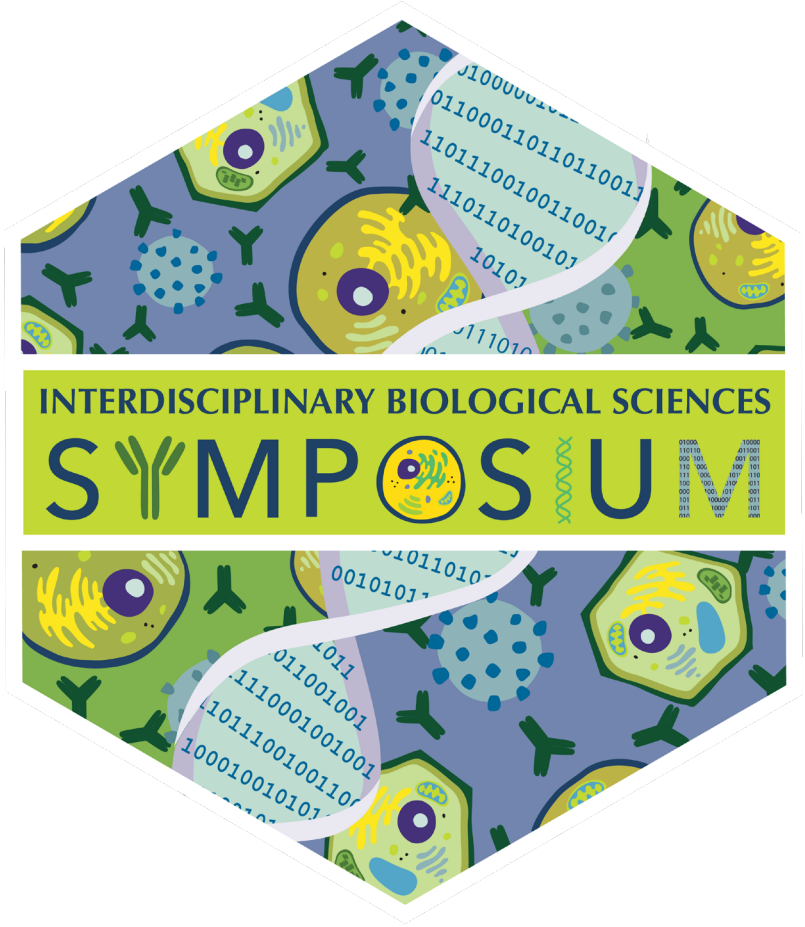


3rd Annual Interdisciplinary Biological Sciences Symposium



3rd Annual Interdisciplinary Biological Sciences Symposium

NextGen Biology: Exploring Emerging Technologies

April 16 - 17, 2025

Memorial Union - Great Hall and South Ballroom

Keynote Speakers

Wednesday April 16, 2025
Memorial Union – South Ballroom

11:00 – 12:00 – Dr. Elisa Crisci, Assistant Professor, North Carolina State University
"The PRRSV Puzzle: Next-Gen Insights into vaccine, immunity and pathogenesis"
-Introduction by Sage Becker

Elisa Crisci is a viro-immunologist who has studied swine virology and immunopathology since her DVM degree at the University of Bologna, Italy.

Crisci completed her master's degree and PhD, and also learned how to handle biolevel 3 zoonotic pathogens at the Centre de Recerca en Sanitat Animal (CRESA) in Barcelona, Spain. During her studies, she focused on animal host-virus interactions and virus-based vaccine vectors for animal diseases, and worked with different swine viruses including circovirus, influenza virus and porcine reproductive and respiratory syndrome virus (PRRSV).

After her PhD, Crisci used the pig as a large animal model for immune cell therapy, and in 2013 she joined the molecular virology division of Linköping University, Sweden, to focus on human virology using techniques such as new generation sequencing.

In 2017, she returned to veterinary virology and joined the French National Institute for Agricultural Research (INRA) in Paris, France. Later in 2018, she joined the Department of Population Health and Pathobiology at NC State's College of Veterinary Medicine as an assistant professor in virology to continue her work on virus-host interactions in pigs, with a focus on the impact of influenza and PRRSV on swine production.

2:30 – 3:30 – Dr. Benjamin Cole, Biologist Research Scientist, DOE Joint Genome Institute
"Single-cell profiling of plant / environmental interactions"
-Introduced by Sabrena Rutledge

Benjamin Cole is a Research Scientist at the DOE Joint Genome Institute. He received his Ph.D. in Biology from UC San Diego, where his research focused on understanding growth dynamics of plants during shade avoidance. He then trained as a postdoctoral researcher using time-lapse imaging and transcriptomics profiling to study diurnal and circadian rhythms in the model grass, Brachypodium Distachyon at UCSD and USC, before joining the JGI to explore functional genomics of plant/microbial interactions. His current research utilizes single-cell and spatial molecular profiling technologies to study plant responses to biotic and abiotic factors in bioenergy grasses.

Thursday, April 17, 2025
Memorial Union – South Ballroom

2:30 – 3:30 - Dr. Junhyong Kim, Co-Director, Penn Program in Single Cell Biology
"Everything, Everywhere, All at Once: From Single Cell Biology to the Theory of the Cell Type"
-Introduced by Sabrena Rutledge

Junhyong Kim, PhD is the Christopher H. Browne Distinguished Professor of Biology. He is also Secondary Professor of Computer and Information Science, the Co-Director of Penn Program in Single Cell Biology, and former Co-Director of Penn Genomics Institute and former Chair of Department of Biology. He is an expert in Genomics, Single Cell Biology, Mathematical and Computational Biology, and Evolutionary Genetics. His current research interests include using single cell technologies to generate a Cell Atlas of the human female reproductive system, single cell analysis for kidney regenerative medicine, development of novel genomic technologies and data analysis algorithms. He is one of the pioneers of the field of Single Cell Biology as well as Algebraic Statistics for biological data analysis. He is the recipient of Sloan Foundation Young Investigator Award, Yale Junior and Senior Faculty Award, J. S. Guggenheim Fellowship, and Ellison Foundation Senior Scholar Award. Dr. Kim's other honors include Visiting Fellow of the Newton Institute at Cambridge, Visiting Research Fellow at Merton College, Oxford, and Visiting Fellow at the Institut des Hautes Etudes Scientifique.

Wednesday, April 16, 2025 - Memorial Union – South Ballroom

Morning Events (South Ballroom)

8:30 – 9:00	**Breakfast, Networking, and Registration**
9:00 – 9:30	**Welcome Announcements** by Co-directors **Tessa Bierbaum, Jodi Callwood, and Ashmita Mainali**, followed by **Opening Remarks** from **Dr. Asheesh Singh**, CALS Associate Dean for Research and Discovery.

Student Oral Session 1 (South Ballroom)

9:30 – 9:45	**Morgan Vorwald** - Genetics and Genomics "A high-fat, high-sucrose diet does not exacerbate inflammatory and ER stress signaling in diaphragm of mdx mice" Advisor: Dr Joshua Selsby
9:45 – 10:00	**Claudia Carrillo** - Nutritional Science "Assessing the effects of different dietary fiber interventions on kidney health and vitamin D homeostasis in a preclinical model of type 1 diabetes" Advisor: Dr Elizabeth McNeill
10:00 – 10:15	**Bimala Acharya** – Bioinformatics and Computational Biology "Deep Learning for Pig Detection: A Step Toward Early Identification of Fall-behind Weaned Pigs" Advisor: Dr David Rosero
10:15 – 10:30	**BREAK**
10:30 – 10:45	**Pengxin Yang** - Biochemistry "Integrative profiling of transcriptome and chromatin accessibility reveals evidence associated with BCG administration induced trained immunity in porcine immune cells" Advisor: Dr Christopher Tuggle
10:45 – 11:00	**Jeong Byoung Chae** - Veterinary Medicine "Molecular Characterization of Ten Viral Pathogens Causing Calf Diarrhea in Hanwoo (Bos taurus coreanae) by Next Generation Sequencing" Advisor: Dr. Jodi McGill

Keynote Presentation 1 (South Ballroom)

11:00 - 12:00

– Dr. Elisa Crisci, Assistant Professor, North Carolina State University

"The PRRSV Puzzle: Next-Gen Insights into vaccine, immunity and pathogenesis"

-Introduction by Sage Becker

12:00 – 1:00	**Lunch**
1:00 – 1:15	**Ankur Kumar** - Genetics and Genomics "Plasmalogens Regulate Mitochondrial Dynamics during Aging in Drosophila melanogaster" Advisor: Dr Hua Bai
1:15 – 1:30	**Thị Hong Anh Pham** - MCDB "Investigating Ether Phospholipid Impact on Mitochondrial Cristae Structure" Advisor: Dr Hua Bai
1:30 – 1:45	**Madison Lane** - MCDB "Building a synthetic cuticle: Characterizing the impact of maize transcription factors on cuticle biosynthetic pathways" Advisor: Dr Marna Yandeau-Nelson
1:45– 2:00	**Joseph DeTemple** - Genetics and Genomics "Circadian rhythm differences between temperate and tropical maize inbreds in response to photoperiod" Advisor: Dr Jianming Yu
2:00 – 2:15	**Alejandro Aragon Raygoza** - GDCB "Exploring the effects of ethylene-related transcription factors during maize shoot development" Advisor: Dr. Josh Strable
2:15 – 2:30	**BREAK**

Keynote Presentation 2 (South Ballroom)

2:30 – 3:30

Dr. Benjamin Cole - Biologist Research Scientist, Joint Genome Institute
"Single-cell profiling of plant / environmental interactions"
-Introduction by Sabrena Rutledge

3:30 – 5:00 Poster Session (Great Hall Memorial Union)

Presenter Name	Poster #	Abstract title
Faith Olusegun	2	Does modularity emerge through evolution? Evidence from a Boolean network-based computational study
Alexandra Else-Keller	4	Phenotyping Differentially Abundant Proteins in Cryopreserved vs Fresh Bull Sperm
Venkata Naresh Boddepalli	6	Genetic analysis of pod traits in Iowa Mung bean Diversity Panel using image-based phenotyping
Memis Bilgici	8	No Title Provided
Wren Murzyn	10	Chemistry-specific off-target effects of a therapeutic antisense oligonucleotide for the treatment of spinal muscular atrophy
Rabsa Naseer	12	PRRSV ORF5 Ct Values: Vaccine-like Strains Show Higher rtPCR Levels
Chunoti Changwal	14	Decoy engineering: a trap for plant pathogens
Jack Moran	16	Investigating Seed Treatments and Soil Amendments to Improve the Establishment of Kentucky Bluegrass as a Perennial Groundcover
Sophie Krivograd	20	Investigating Tryglysin A resistance and gene regulation in streptococci
Meagan Putzier	22	Holder for Ultrasound Transducer to Enable Photoacoustic Imaging
Rachael Gorman	24	The interaction of the Arabidopsis xyloglucan xylosyltransferases with the COPII member SAR1 via their di-arginine motifs is critical for delivery to the Golgi.
John Ssengo	26	Dissecting Puccinia sorghi haustoria at the molecular level: A multi-omics investigation
Caden Klopfenstein	28	Solutions to Address Biases and Increase the Accuracy of Social Contact Matrices
Maddumage Dona Ginushika Premarathne	30	Alternative splicing in Gossypium hirsutum during fiber development

Thursday, April 17, 2025

Student Oral Session 3 (South Ballroom)

9:00 – 9:30	Breakfast and Networking
9:30 – 9:45	**Md Shahriar Mahmud -** Mathematics "Adaptive human behavior and delays in information availability autonomously modulate epidemic waves" Advisor: Dr Claus Kadelka
9:45 – 10:00	**Priyanka Banerjee** - Computer Science "Identifying core operons in metagenomic data" Advisor: Dr. Oliver Eulenstein and Dr. Iddo Friedberg
10:00 – 10:15	**An Phan** – Bioinformatics and Computational Biology "Prioritizing Literature Curation of Under-annotated Proteins with IgnoroMeNot" Advisor: Dr Claus Kadelka
10:15 – 10:30	**Parnal Joshi** - Bioinformatics and Computational Biology "Cross-species quantification of protein function annotations provides insights into the utility of model organisms" Advisor: Dr Iddo Friedberg
10:30 – 10:45	**Rodolfo Calderon** - MCDB "Pronephric derived nitric oxide acts in the NF-kB pro-inflammatory transcription mechanism" Advisor: Dr Raquel Espin Palazon

10:45 – 12:00 Poster Session - (Great Hall Memorial Union)

Presenter Name	Poster #	Abstract Title
Catherine Fonder	1	Evaluating the effect of shockwave impacts on the behavior of adult hippocampal progenitor cell neurospheres
Rabsa Naseer	3	PRRSV Ct Value Distributions Across Sample Types
Amaechi, Onyinyechi Judith	5	Evaluating single nucleus RNA sequencing in Gossypium (cotton) fiber
Reyna Winders	7	Investigating the Xyloglucan Xylosyltransferase Heterodimer Organization
Fred Rogers Namanda	9	FHOD3 and DIAPH3 control stress fiber organization and cell migration through shared but non-overlapping mechanisms
Oluwatobiloba Aminu	11	GWAS Approach to Identify Diagnostic Markers in Avibacterium paragallinarum
Masuma Khatun Usha	13	Rewiring endothelial cells toward definitive hemogenic endothelium
Sristi Dey	15	Impact of tryglysin on metal homeostasis in oral streptococci
Leticia Herrera	19	Analyzing Cohesion: Investigating Cdc23 S.Cerevisiae Mutations and it's Role in Chromosome Separation
Ludvin Mejia	21	Determining the role of lncRNA BCAR4 in human trophoblast cells
Matthew Wendt	23	Environmental and Genetic Factors Underlying Maize Cuticular Wax Accumulation Under Drought Stress
Jahanara Sonia	25	Translation of Newly Discovered Maize associated Tombus-Virus (MaTV) Genome: An Unsolved Mystery
Breigha Boyle	27	Exploring the Efficacy of FDA-approved inhibitors on Rift Valley Fever Virus Pathogenesis
Ushashi Bhattacharjee	29	Benchmarking BioEmu-1: Establishing a Baseline for Protein Structural Ensemble Prediction

12:00 – 1:00 Lunch

Student Oral Session 4 (South Ballroom)

1:00 – 1:15 **Nesreen Sedeek** - Neuroscience

"Differentiation and Proliferation of AHPC Neurospheres Plated on Gold Substrates Subjected to Electric Stimulation"

Advisor: Dr Donald Sakaguchi

1:15 – 1:30 **Swathi Nadendla** - BCB

"Coupling Bioinformatics approaches and Oxford Nanopore (ONT) sequencing for enhanced Diagnosis of Newcastle Disease Virus (NDV) in Poultry"

Advisor: Dr. Mohamed El-Gazzar

1:30 – 1:45	**Benjamin Jacobs** - Statistics "Heterosis gene detection for RNA-seq data without replicates: an objective Bayesian approach" Advisor: Dr. Peng Liu
1:45– 2:00	**Xinglin Jia** - BCB "CLEAR: Concise List Enrichment Analysis using R" Advisor: Dr Claus Kadelka
2:00 – 2:15	**Sabrena Rutledge** - BCB "Defining trophoblast cell lineages within the rat placenta using single-cell RNA-sequencing" Advisor: Dr Geetu Tuteja
2:15 – 2:30	**BREAK**

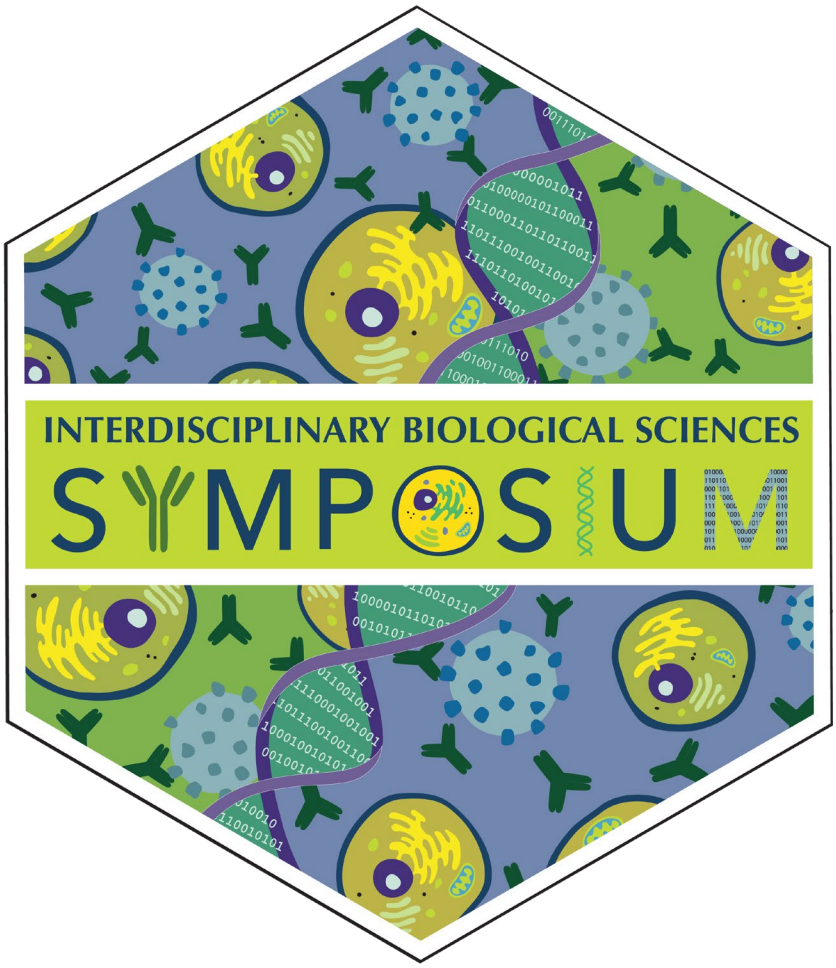
Keynote Presentation 3 (South Ballroom)

2:30 – 3:30 **Dr. Junhyong Kim - Co-Director, Penn Program in Single Cell Biology**
"Everything, Everywhere, All at Once: From Single Cell Biology to the Theory of the Cell Type"
-Introduction by Sabrena Rutledge

Closing Announcements by Co-directors **Tessa Bierbaum, Jodi Callwood, and Ashmita Mainali**, followed by **Closing Remarks** from **Dr. Jodi McGill**, Associate Dean of Research and Graduate Studies.

Awards Ceremony

- Student Oral Presentation - 1st place, 2nd place, 3rd place, people's choice
- Student Poster Presentation - 1st place, 2nd place, 3rd place, people's choice
- Lauter Award for Graduate Student Creativity and Research Excellence



Student Oral

Presentation Abstracts

A high-fat, high-sucrose diet does not exacerbate inflammatory and ER stress signaling in diaphragm of mdx mice

Morgan E Vorwald, Swathy Krishna, Ji Heun Lee, Melissa Roths, Rudy J. Valentine, Joshua T. Selsby

Genetics and Genomics Graduate Program, Department of Animal Science, Iowa State University, Ames, IA

Duchenne muscular dystrophy (DMD) is a muscle-wasting disease caused by dystrophin deficiency that leads to death from cardiac or respiratory failure. DMD is frequently complicated by insulin resistance (IR); however, the extent to which IR influences disease progression is unknown. We hypothesized that a high fat, high sucrose diet (HFHSD) and subsequent IR would cause increased inflammatory and ER stress signaling in mdx diaphragm compared to mdx mice fed a control diet (CD). To address this hypothesis, beginning at 6 w, C57 and mdx mice were fed a CD or HFHSD for 19 w, with a subset of mice from each diet also treated with prednisolone (Pred; 10mg/L). Insulin resistance was confirmed by fasting hyperglycemia and glucose tolerance tests in HFHSD mice. In dystrophin-deficient diaphragm, NFκB and phosphorylated (p)-NFκB, were decreased by dietary treatment ($p<0.01$) and further decreased with Pred ($p<0.01$). IκBα, an endogenous NFκB inhibitor, and IKKα, an IκBα inhibitor, were similar between dietary and Pred treatments. Inflammatory cytokines, including TNFα, IL-10, and IL-17, were similar between dietary treatments but significantly decreased with Pred ($p<0.01$-0.05). A HFHSD decreased ER stress markers BiP ($p<0.01$), p-Perk ($p<0.05$), and IRE1α ($p<0.05$) compared to CD-fed mice, Pred treatment increased PKR and decreased ATF4, but Perk, p-IRE1α, p-PKR, eIF2α, p-eIF2α, CHOP, and XBP1s were similar between groups. Counter to our hypothesis, a HFHSD did not increase inflammatory and ER stress signaling in mdx mice.

Assessing the effects of different dietary fiber interventions on kidney health and vitamin D homeostasis in a preclinical model of type 1 diabetes

Claudia Carrillo, Claudia Arriaga, Chloe Smith, Matthew Rowling

Interdepartmental Graduate Program in Nutritional Sciences, Department of Food Science and Human Nutrition, Iowa State University, Ames, IA

Type I diabetes (T1D) is an autoimmune disease that increases the risk of renal injury and nutritional deficiencies if left uncontrolled. Our research demonstrates that a dietary intervention with 20% resistant starch (RS) normalized growth patterns and corrected vitamin D metabolism by protecting renal health in streptozotocin (STZ)-induced T1D Sprague Dawley (SD) rats, without improving fasting blood glucose (FBG).This study aimed to evaluate the effects of a 5-day dietary intervention with 5% or 10% RS, or a whole oat (WO) diet, prior to STZ administration on kidney health, nutritional status, and growth patterns. Fifty-six SD rats (4 weeks old) were randomly assigned to one of four diets: AIN-93G with 0% RS (control), 5% RS, 10% RS, or WO diet (5% fiber). Macronutrients were adjusted to maintain isocaloric diets. After 5 days, 49 rats received an intraperitoneal (i.p.) injection of STZ, while 7 control diet rats received an i.p. injection of citrate buffered saline (vehicle) as a non-diabetic control. Three weeks later, urine, feces, and FBG were collected before euthanization after a 6-hour fast. Neither fiber diet improved FBG, but the WO diet increased weight gain, food intake, and food efficiency compared to the diabetic control (DC) and decreased urinary vitamin D binding protein excretion and serum C-reactive protein levels by 71% and 14%, respectively. Both RS diets protected renal health, decreasing urinary excretion of glucose, ketones, and volume. The 10% RS diet reduced relative kidney weight by 17% and normalized urinary creatinine. The 5% RS group maintained serum 25D levels despite elevated UPCR and vitamin D metabolite excretion. Our results indicate that low to moderate levels of RS and WO diets positively impacted T1D rats, with the timing of the fiber intervention being crucial for the observed effects.

Deep Learning for Pig Detection: A Step Toward Early Identification of Fall-behind Weaned Pigs

Bimala Acharya, Mina Shumaly, Juan Steibel, Nick Gabler, David Rosero

Bioinformatics and Computational Biology, Department of Animal Science, Iowa State University, Ames, IA, USA

Weaning is the stressful event in the life of a pig marked by drastic shift from milk to complex solid feed resulting poor feed consumption impacting the development and health of gastrointestinal track of weaned pigs. Eventually, the pigs fail to achieve growth performance compared to the rest of the herd called as fall-behind pigs. The nutritional inadequacy in fall-behind pigs leads to higher post-weaning mortality. We aim to leverage deep learning for image-based detection of pigs with high risk of falling behind. Weaned pigs (3-weeks old) in 24 pens (10 pigs/pen) were video recorded for first 4 days of arrival. Frames were extracted and annotated to train the YOLO v3 model. For evaluation, the threshold intersection over union (IOU) for the bounding boxes was set to 0.5. Other evaluation metrics were calculated as below:

Precision = TP / (TP + FP) (1)

Recall = TP / (TP + FN) (2)

F1 Score = 2 * (Precision * Recall)/ (Precision + Recall) (3)

Where, TP = True Positive, FP = False Positive, and FN = False Negative

With the developed object detection model, we reported the model's precision of 0.93016, recall of 0.90705, the F1-score of 0.91846 and the mean intersection over union of 0.73321. The model will be used further to track and identify phenotypic behaviors like: number and duration of feeding and drinking, eventually to identify pigs at high risk of falling behind.

Integrative profiling of transcriptome and chromatin accessibility reveals evidence associated with BCG administration induced trained immunity in porcine immune cells

Kristen A. Byrne, Pengxin Yang, Juan Steibel, Christopher Tuggle, Crystal L. Loving

Department of Animal Science, Iowa State University, Ames, IA, 50011, USA; Department of Biochemistry, Biophysics and Molecular Biology, Iowa State University, Ames, IA, 50011, USA.

The traditional view that only adaptive immunity has memory has been challenged in the past decade. Trained immunity, or innate immune memory, means that innate immune cells (e.g., monocytes), develop long-term functional reprogramming after initial stimulation that results in elevated response to secondary stimuli. Bacillus Calmette Guérin (BCG) induced trained immunity has been reported in humans, porcine, and other species, and BCG vaccination has been shown to provide long-term non-specific protection to humans. In vivo BCG exposure enhances cytokine production to TLR stimulation by porcine monocytes, yet changes in genome-wide regulatory networks remain understudied. To address this, we profiled transcriptomic and chromatin accessibility changes in monocytes recovered from BCG-treated or non-treated pigs. Three-day-old piglets were inoculated with BCG or PBS. Six weeks later, monocytes were isolated for ATAC-seq or stimulated ex vivo for four hours with either media alone or LPS for transcriptomics. We identified 3150 differentially expressed genes whose response to LPS stimulation was altered by prior BCG exposure and 2410 differentially accessible ATAC-seq peaks (DAP) due to prior BCG inoculation. GO analysis identified terms linked to metabolic shifts in BCG-trained monocyte RNA. RNA-seq and ATAC-seq signals at the closest gene were moderately significantly correlated, highlighting the role of chromatin accessibility in elucidating the mechanisms driving gene expression. Transcription factor binding motif analysis of the DAP predicted TFs regulating genes whose expression was associated with BCG training. Notably, no TF binding motifs were enriched in regions of increased chromatin accessibility following BCG inoculation. Our results reveal novel insights into the gene expression dynamics and chromatin accessibility landscapes associated with trained immunity in pigs. These findings deepen insights into innate immune memory in pigs, advancing veterinary and comparative immunology applications.

Molecular Characterization of Ten Viral Pathogens Causing Calf Diarrhea in Hanwoo (Bos taurus coreanae) by Next Generation Sequencing.

JeongByoung Chae, Joon-Seok Chae

Department of Veterinary Microbiology and Preventative Medicine, Iowa State University, Ames, IA

Calf diarrhea is a major issue in the cattle industry, leading to economic losses worldwide. Infectious pathogens are key contributors to this disease. This study examined the prevalence of seven pathogens—bovine rotavirus (BRV), bovine coronavirus (BCV), bovine viral diarrhea virus types 1 and 2 (BVDV1/2), Cryptosporidium parvum, Giardia spp., and Eimeria spp.—in Hanwoo calves using PCR. A total of 810 fecal samples were analyzed, including 526 normal and 284 diarrheic samples. BRV (14.0%), C. parvum (9.8%), and Eimeria spp. (1.9%) showed significant associations with diarrhea severity. Additionally, 21 pathogen-negative samples were subjected to high-throughput sequencing, revealing nearly complete genomes of 10 viruses: bovine astrovirus, bovine enterovirus, bovine kobuvirus, bovine nebovirus, bovine norovirus, bovine boosepivirus B, bovine parechovirus, bovine torovirus, C. parvum virus 1, and hunnivirus. This study provides the first report of hunnivirus in the Republic of Korea and expands knowledge of calf diarrhea-associated viral diversity. The findings enhance our understanding of pathogen prevalence and molecular characteristics, highlighting the value of metagenomic sequencing in identifying unknown causes of calf diarrhea and improving disease diagnostics.

Plasmalogens Regulate Mitochondrial Dynamics during Aging in Drosophila melanogaster

Ankur Kumar, Hua Bai

Genetics and Genomics, Iowa State University

Healthy mitochondria play an essential role in maintaining cellular homeostasis. Mitochondrial structure and function are impaired during aging, likely due to dysregulated mitochondrial fission and fusion processes. Through TEM and confocal imaging analysis, we observe that mitochondrial size increases in aged oenocytes, the hepatocyte-like cells in Drosophila. Interestingly, mitochondria from aged oenocytes do not exhibit paraquat-induced mitochondrial fission, unlike those in young flies. Additionally, aged flies show mitochondria with decreased fission compared to young flies.

To investigate the mechanisms underlying this decline in mitochondrial fission during aging, we identified that disruptions in Kua/PEDS1 (Plasmanylethanolamine desaturase), a key enzyme in plasmalogen biosynthesis, inhibit mitochondrial fission. We find that the alterations of plasmalogen biosynthesis enzymes such as Kua/PEDS1 blocks paraquat-induced mitochondrial fission and the recruitment of fission factor Drp1 to mitochondria. Mass spectrometry analysis revealed a significant decrease in mitochondrial plasmalogen C18:0/C18:1-PE levels in aged flies.

To understand the involvement of plasmalogens in mitochondrial fission, we performed lipid pull-down assays using synthetic biotinylated C18:0 plasmalogen PE (pPE) and identified interacting mitochondrial fission proteins such as Drp1, mff/Tango11, and other mitochondrial proteins. Further validation using crude mitochondrial extracts from Kua/PEDS1 mutants confirmed the decrease of Drp1 in mitochondrial fractions, indicating a critical role for plasmalogens in recruiting fission proteins.

Mitochondrial dynamics, encompassing fission and fusion processes, are essential for maintaining cellular homeostasis. Age-dependent alterations in these dynamics, including increased mitochondrial size, are closely associated with mitochondrial dysfunction. Our findings underscore the pivotal role of plasmalogens in maintaining mitochondrial dynamics during aging, with implications for understanding their involvement in age-related diseases, including cardiomyopathy, respiratory dysfunction, and neurodegeneration.

Investigating Ether Phospholipid Impact on Mitochondrial Cristae Structure

Thi Hong Anh Pham, Min Jung Kwon, Hua Bai

Molecular, Cellular, Developmental Biology Graduate Program, Genetics, Development, and Cell Biology, Iowa State University, Ames, IA, USA

The intricately folded inner membranes of mitochondria, known as cristae, are essential for optimizing oxidative phosphorylation (OXPHOS) by concentrating respiratory complexes to enhance ATP production. Their architecture is regulated by phospholipids with cone-shaped structures, which influence curvature and stability. Plasmalogens, a class of ether phospholipids enriched in mitochondrial membranes, are proposed to maintain cristae integrity by supporting ATP synthase dimerization. We hypothesize that plasmalogen deficiency disrupts this process, leading to altered cristae morphology and impaired OXPHOS efficiency. To test this, we used biotinylated plasmalogen pulldown assays in mouse cardiac tissue, followed by TMT-multiplexed mass spectrometry, identifying 70 proteins that interacted with plasmalogens and were pulled down. Of these, 75% localized to mitochondrial membranes, predominantly within OXPHOS complexes. Western blotting confirmed significant interactions with ATP synthase (Complex V), a key driver of cristae curvature through dimerization. Using a plasmalogen-deficient model (PEDS1-KO), transmission electron microscopy (TEM) revealed that cristae lost their tubular sharp-tip structure, adopting a more rounded morphology. Blue Native-PAGE (BN-PAGE) further showed reduced ATP synthase dimerization, correlating with impaired OXPHOS efficiency. These findings define a plasmalogen-associated protein interactome within cristae, suggesting that plasmalogens shape mitochondrial function through both structural lipid effects and protein interactions, with their loss impairing ATP synthase dimerization and cristae integrity. Future studies will target specific ATP synthase membrane subunits using in silico docking, predict plasmalogen binding sites, engineer mutant cell lines to test their impact, and use lipid-protein interaction assays to confirm direct binding. These insights may guide strategies to preserve mitochondrial function in aging and disease.

Building a synthetic cuticle: Characterizing the impact of maize transcription factors on cuticle biosynthetic pathways

Madison Lane, Jingjing Tong, Noah Wallberg, Sofia Torres, Jasmine Reyes, Erin Sparks, Marna D. Yandeau-Nelson

Molecular, Cellular & Developmental Biology Graduate Program, Department of Genetics, Development and Cell Biology , Iowa State University, Ames, IA

The hydrophobic cuticle, which covers aerial portions of plants, is the first line of defense against environmental stresses, including drought, UV radiation, and insects and pathogens. This cuticle is comprised of a cutin polyester matrix that is infused with and laid atop by cuticular waxes, comprised of differing combinations of very long chain fatty acids (VLCFAs), hydrocarbons, aldehydes, alcohols, wax esters, and ketones depending on the species, organ and stage of development. The biosynthesis of the cuticle can be broken down into five regulatory modules, including fatty acid elongation, cutin monomer synthesis, the decarbonylative and reductive modules within cuticular wax biosynthesis, and transport. We have identified candidate transcription factors (TFs) through multi-omics approaches predicted to be associated with cuticle composition. The potential functions of these TFs in the regulation of cuticle biosynthesis and deposition are being evaluated via a synthetic biology approach using a transient gene expression system within maize and Arabidopsis root protoplasts. Roots are an ideal system to evaluate the regulation of cuticle biosynthesis because they do not synthesize a cuticle thus any cuticle production is attributable to the gene expression induced by heterologously expressed candidate TFs. Using this approach, we've shown that both previously reported cuticle TFs and TFs not previously reported to be involved in cuticle formation can activate genes involved in cuticular wax biosynthesis. The TFs validated via this approach are now being expressed in stable Arabidopsis transgenic lines for functional analyses.

Circadian rhythm differences between temperate and tropical maize inbreds in response to photoperiod

Joseph DeTemple, Jacob Hinrichsen, Dongdong Li, Greg Schoenbaum, Jamie O'Rourke, Michelle Graham, Xianran Li, Michael Muszynski, Jianming Yu

Genetics and Genomics Graduate Program, Department of Agronomy, Iowa State University, Ames, IA

The floral transition of maize, where the shoot apical meristem switches from vegetative to reproductive growth, is controlled by networks of genes that capture, process, and interpret environmental signals such as photoperiod. A more complete picture of network dynamics is critical for fully understanding the responses of temperate and tropical lines to inductive and non-inductive photoperiods. We set out to answer three questions: 1) which genes are most important within the photoperiod pathway of the maize flowering time gene network, 2) which developmental stages are most important for photoperiod gene expression, and 3) which time of day is most important for capturing expression differences. To answer these questions, we grew the inbred lines B73, Oh43, Mo17, TX303, Ki3, CML277, CML52, and Tzi8 under 15h/9h light/dark conditions in growth chambers, collected leaf tissue at V2-V10 stages at 3 different timepoints (6am, 2pm, and 10pm), and ran qPCR reactions for 8 genes of interest (prrtf1, cca1, toc1, gi1, col3, elf3.1, mads69, and pebp8) that lie within the circadian rhythm and photoperiod pathways of the maize flowering time gene regulatory network. Phenotypes collected throughout the experiment include V-stage date, flowering time, plant height, leaf number, leaf area, and biomass measurements of different plant organs. General circadian rhythm patterns are presented, along with genotypic differences. Overall, this experiment deepens our understanding of which genes are most important in regulating flowering time differences between temperate and tropical maize lines, which developmental stages are most critical for the floral transition, and which time of day expression differences are strongest.

Exploring the effects of ethylene-related transcription factors during maize shoot development

Alejandro Aragon Raygoza, Mary Galli, Norman Best, Erica Unger-Wallace, James Satterlee, Charlie Beeler, Jeffrey Yen, Erik Vollbrecht, Andrea Gallavotti, Josh Strable

Postdoc, Department of Genetics, Development and Cell Biology, Iowa State University, Ames, IA

The plant hormone ethylene is a key developmental regulator that acts largely to restrict growth. But our understanding of ethylene function in cereal crops is markedly limited. Here, we characterized mutations in maize ETHYLENE INSENSITIVE3(EIN3)-LIKE1 (ZmEIL1) and ZmEIL9 genes that encode co-orthologs of the Arabidopsis EIN3/EIL1 hub transcription factors. By stacking independently derived mutations in ZmEIL1 and ZmEIL9, we uncovered vegetative shoot phenotypes associated with mis-regulated growth that are distinct from Arabidopsis ein3;eil1 mutants. We show that Zmeil1;Zmeil9 double mutants are insensitive to the ethylene precursor ACC in separate germination and growth assays. By utilizing single-cell transcriptomics, we identified tissue- and cell-specific genetic signatures in Zmeil1;Zmeil9 and normal shoots with or without exogenous ACC. We found that differentially expressed genes in hormone pathways are highly correlated with altered accumulation of hormone pathway intermediates in Zmeil1;Zmeil9 shoot apices. We leveraged DAP-seq to identify genome-wide binding positions of ZmEILs. Approximately 85% of ZmEIL1 and ZmEIL9 bound filtered-genic regions overlap, suggesting co-regulation of a large portion of putative target genes, including a proportion of genes that were identified as differentially expressed in our single-cell data. Genes directly bound and modulated by ZmEILs include ZAR and RTL family members that are negative regulators of ethylene signaling, and that also have important roles in organ growth. Our study shows that ZmEIL genes are central regulators of shoot growth and, by resolving cellular and tissue heterogeneity of ethylene response, significantly advances our understanding of ethylene signaling in plants.

Adaptive human behavior and delays in information availability autonomously modulate epidemic waves

Md Shahriar Mahmud, Solomon Eshun, Baltazar Espinoza, Claus Kadelka

Applied Mathematics, Department of Mathematics, Iowa State University, Ames, IA

The recurrence of epidemic waves has been a hallmark of infectious disease outbreaks. Repeated surges in infections pose significant challenges to public health systems, yet the mechanisms that drive these waves remain insufficiently understood. Most prior models attribute epidemic waves to exogenous factors, such as transmission seasonality, viral mutations, or implementation of public health interventions. We show that epidemic waves can emerge autonomously from the feedback loop between infection dynamics and human behavior. Our results are based on a behavioral framework in which individuals continuously adjust their level of risk mitigation subject to their perceived risk of infection, which depends on information availability and disease severity. We show that delayed behavioral responses alone can lead to the emergence of multiple epidemic waves. The magnitude and frequency of these waves depend on the interplay between behavioral factors (delay, severity, and sensitivity of responses) and disease factors (transmission and recovery rates). Notably, if the response is either too prompt or excessively delayed, multiple waves cannot emerge. Our results further align with previous observations that adaptive human behavior can produce non-monotonic final epidemic sizes, shaped by the trade-offs between various biological and behavioral factors--namely, risk sensitivity, response stringency, and disease generation time. Interestingly, we found that the minimal final epidemic size occurs on regimes that exhibit a few damped oscillations. Altogether, our results emphasize the importance of integrating social and operational factors into infectious disease models, in order to capture the joint evolution of adaptive behavioral responses and epidemic dynamics.

Identifying core operons in metagenomic data

Priyanka Banerjee, Xiao Hu, Oliver Eulenstein, Iddo Friedberg

Computer Science Graduate Program, Department of Computer Science, Iowa State University, Ames, IA

Motivation: An operon is a functional unit of DNA whose genes are co-transcribed on polycistronic mRNA, in a co-regulated fashion. Experimental approaches are the most reliable way to identify operons; however, performing experiments to characterize all operons is not feasible. The challenge of discovering operons is compounded when working with metagenomic data due to genomic information loss and misassembly producing chimeric contigs. Metagenomic data contain rich information that cannot be gleaned from clonal cultures; therefore, it is necessary to investigate how well we can predict operons in metagenomic data.

Method: Here we present a Pipeline for Operon Exploration in Metagenomes, or POEM to identify operons in metagenomic data. POEM initially determines whether adjacent gene pairs are operonic or non-operonic. Subsequently, POEM introduces the concept of a core operon to facilitate the reconstruction of the operonic structure. A core operon comprises a set of intra-operonic gene pairs that have orthologs in several species in the metagenome and are concatenated using guilt-by-association. Our method considers the evolutionary conservation of co-transcribed genes in the microbial community species, which is valuable for understanding the genetic potential of a microbial community.

Results: POEM shows a high overlap in the count of functions between the core operons and the true operons indicating that while not all genes in an operon can be recovered in a metagenome, the basic functionality enabled by core operons can be recovered. We conclude that POEM effectively analyzes metagenomes, identifying multi-gene functions and potential neofunctionalization.

Prioritizing Literature Curation of Under-annotated Proteins with IgnoroMeNot

An Phan, Claus Kadelka, Iddo Friedberg

Bioinformatics and Computational Biology Graduate Program, Department of Mathematics, Iowa State University, Ames, IA

Protein function annotation databases are crucial resources for understanding protein function, but they exhibit bias towards well-studied proteins of established biomedical interest. The biocuration process of reviewing scientific publications and adding knowledgebase entries maintains rigorous quality standards but remains time-consuming and challenging. We hypothesize that extensive manual effort creates curation delays and priority criteria that perpetuate annotation bias by disproportionately affecting less annotated proteins. To investigate this hypothesis, we analyzed the growth over time of Gene Ontology (GO) annotations of all proteins in the human proteome. By quantifying the information captured in GO annotations and collecting the publication timestamps, we discovered that hundreds of human proteins have not been revisited by biocurators for more than a decade since the last time the proteins were reviewed. Furthermore, we showed that there are protein-rich small-scale studies that have been published after the most recent annotations were assigned to the less annotated proteins, but were not reviewed by biocurators, possibly due to not being a priority. Therefore, there is a certain subset of long-time less annotated proteins, which we call "under-annotated" proteins, that have been studied in more recent small-scale studies, and could be reviewed by biocurators. To address the problem of under-annotated proteins, we introduce a web tool that helps biocurators prioritize the literature curation of proteins. Our work underscores the complex relationship between publication patterns, curation practices, and protein knowledge representation in biological databases, revealing potential opportunities for improving the balanced coverage of functional annotation across the human proteome.

Cross-species quantification of protein function annotations provides insights into the utility of model organisms

Parnal Joshi, Qi Li, Iddo Friedberg

Bioinformatics and Computational Biology Graduate Program, Department of Veterinary Microbiology and Preventive Medicine, Iowa State University, Ames, IA

The accurate annotation of the biological function of proteins is an open problem in life science. While improvements in sequencing techniques have facilitated novel data generation, translating sequence data into biologically meaningful functions remains a challenge. Comprehensive knowledge of protein function is crucial to understanding life's mechanisms and for practical applications such as characterizing disease genotypes and phenotypes, identifying antimicrobial resistance, and selecting targets for drug design. Biomedical researchers have been utilizing model organisms as proxies for humans for centuries, including, starting in the 20th Century, for the study of protein function. However, the contribution of model organisms to our understanding of protein function and its applicability in humans has not yet been systematically quantified.

Here, we developed an information-theoretic framework for comparing Gene Ontology annotations between species. Our analysis shows the sum of contributions of experimental annotations from five model species surpasses the total information gained from human-specific annotations. Next, we measured the functional similarity between annotations of orthologous proteins across species. Consistent with previous studies, we observed no correlation between function similarity of experimental annotations and sequence divergence across species. This observation highlights the limitations of assays performed on different biological systems. Individual model species contribute partially to protein function, and when experimental annotations are combined from multiple model organisms, they can provide us with a multifaceted understanding of the workings of a protein. We conclude that the lion's share of experimental annotations across model species are not redundant but complementary. Our assessment of the annotation landscape allows us to better identify avenues for experimental research in model organisms, and provide suggestions for more judicious biocuration.

Pronephric derived nitric oxide acts in the NF-kB pro-inflammatory transcription mechanism

Rodolfo Calderon, Raquel Espin-Palazon, Xiaoyi Cheng, Clyde Campbell

Molecular, Cellular and Developmental Biology Graduate Program, Department of Genetics, Development and Cell Biology, Iowa State University, Ames, IA

In humans, the mature hematopoietic system is supplied from a set of hematopoietic stem cells (HSCs) that reside in kidney marrow. These HSCs are responsible for maintaining a balanced environment of blood lineages. Identifying the necessary components of hematopoietic development is understood to be essential for effective treatment of blood illnesses. Zebrafish are a great tool for studying embryonic hematopoietic development due to highly shared blood development. The significance of classical inflammatory components in developmental hematopoiesis was recently appreciated, demonstrating the importance of the canonical role of the nuclear factor-kappaB (NF-kB) transcription factor family member heterodimer p65-p50. Recently, published work from the Espin-Campbell lab showed a temporally detailed role of the heterodimer during the early stages of hematopoietic development when cells are differentiating from an endothelial to hematopoietic identity. Still, what causes the dynamic activity of NF-kB activity is not completely understood. In this project, we are working to understand this more clearly. Literature shows the ability of nitric oxide to modulate the production levels of hematopoietic precursor cells during the activity window of NF-kB. Preliminary data collected in our lab demonstrates a strong relation of nitric oxide levels with NF-kB activity through analysis by various in vivo and ex vivo methods at multiple stages of hematopoietic development which we can characterize by NF-kB activity. Most excitingly through this work, we have found the developing kidney to be a potentially unappreciated component of hematopoietic development by localization of the nitric oxide synthase activity and proximity to hematopoietic emergence presenting a possible importance of signaling from non-vascular tissue to HSC establishment.

Differentiation and Proliferation of AHPC Neurospheres Plated on Gold Substrates Subjected to Electric Stimulation

Nesreen Sedeek[1], Sajid M. Uchayash[2], Long Que[2], and Donald S. Sakaguchi[1]

[1] Genetics, Development, and Cell Biology, Neuroscience program, Iowa State University, Ames, IA
[2] Electrical and Computer Engineering, Iowa State University, Ames, IA

Neurodegenerative diseases are a significant global health challenge due to the lack of effective treatments. These diseases are characterized by the progressive loss of neurons, impaired neurogenesis, and chronic neuroinflammation, leading to cognitive decline and functional impairment. Neural stem cells (NSCs) hold promise for regenerative therapies due to their ability to self-renew, differentiate into neural lineages, and secrete neurotrophic factors that mitigate oxidative stress and neuroinflammation. However, harnessing the full potential of NSCs for therapeutic applications requires precise control over their behavior, including migration, differentiation, and integration into damaged neural circuits. Electrical stimulation (E-stim) has emerged as a powerful tool to modulate NSC behavior, with studies demonstrating its ability to enhance cell migration, differentiation, and viability in a voltage-dependent manner.

This study investigates the effects of biphasic E-stim—a form of E-stim characterized by alternating cycles of electrical current—on a population of multipotent adult NSCs originally isolated from the rat dentate gyrus of the hippocampus. These adult hippocampal progenitor/stem cells (AHPCs) are grown as self-assembling neurospheres (NS) and were cultured on smooth and nanostructured gold substrates. AHPC NSs were subjected to biphasic E-stim at 20 or 40 mV, at 1 Hz, for 10 minutes daily over 5 days in vitro. A Propidium Iodide assessment confirmed the biocompatibility of the devices, with cell viability exceeding 98% and cell death below 2%. Immunostaining for TuJ1, a marker of immature neurons, revealed that 20 mV E-stim significantly enhanced neuronal differentiation compared to 40 mV, suggesting that higher voltages may suppress

neurogenesis. No significant differences were observed between smooth and nanostructured substrates, indicating that substrate topography did not markedly influence the outcomes under these conditions.

These findings highlight the potential of biphasic E-stim to enhance neuronal differentiation in a voltage-dependent manner. In future experiments, we aim to further investigate the effect of 20 mV E-stim on AHPC differentiation using additional cell type-specific markers, including RIP for oligodendrocytes, GFAP for astrocytes, and MAB2ab for maturing neurons. This research provides a foundation for developing more effective strategies for neural regeneration and disease modeling using electrical stimulation.

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Coupling Bioinformatics approaches and Oxford Nanopore (ONT) sequencing for enhanced Diagnosis of Newcastle Disease Virus (NDV) in Poultry

Swathi Nadendla[1], Maria Chaves[2], Eman Gadu[2] & Amro Hashish[2]

[1]Bioinformatics and Computational Biology Graduate Program, Department of Veterinary Diagnostic and Production Animal Medicine, Iowa State University, Ames,IA
[2]Department of Veterinary Diagnostic and Production Animal Medicine, Iowa State University, Ames,IA

Objective
Newcastle Disease virus causes respiratory, systemic illness in poultry, controlled through modified live vaccines. Rapid and accurate genetic characterization is critical for distinguishing wild type from vaccinal strains. Current typing uses genotype-specific primers for the Fusion (F) gene, requiring multiple steps that delay results, underscoring the need for a streamlined workflow. ONT enables real-time, long-read sequencing, but high background noise from host DNA in clinical samples necessitates enrichment. Effective enrichment relies on optimized primer design, yet existing pipelines need improvement. This study aims to harness ONT to develop a genotype independent workflow providing a scalable diagnostic approach.
Methods
To achieve a universal workflow for NDV we propose two enrichment approaches, a k-mer-based primer design pipeline and a nested PCR by using a comprehensive database. Conserved regions across the F gene were identified using a custom script, divided into overlapping segments enabling the primer design. To capture diversity, degeneracy was added. This design generates a broadly effective primer sets that needs to be further optimized insilco.
Results
The k-mer pipeline produced 38 primers, while nested PCR utilized two sets of inner and outer primers. Targeted amplicons were sequenced using MinION and data was processed through an inhouse analysis pipeline enabling continuous Fastq file pickup, base calling, read classification generating alignment statistics allowing rapid identification of NDV within hours. This live analysis facilitates in improved detection compared to traditional post run bulk analysis. While this pipeline currently supports real-time detection, further optimization is needed to reliably characterize genotypes.
Conclusion
This combined approach involves targeted enrichment, nanopore sequencing, and real-time analysis has the potential to become a field-adaptable solution. Our work demonstrates that next-generation technologies tailored to pathogen-specific challenges can significantly enhance diagnostic accuracy.

Heterosis gene detection for RNA-seq data without replicates: an objective Bayesian approach

Benjamin Jacobs & Peng Liu

Department of Statistics, Iowa State University, Ames, IA

'Mid-parent heterosis' refers to the condition that a hybrid has a mean that differs from the average of its two parent means for a phenotypic trait. With RNA-seq data, the detection of genes exhibiting mid-parent heterosis helps investigate the molecular causes of the well-known phenomenon of heterosis, also known as hybrid vigor. A key parameter that characterizes mid-parent heterosis genes is the 'mid-parent fold-change', defined as the ratio of the average of the two parental mean expression levels over the hybrid's mean expression level. The estimation of mid-parent fold changes is challenging in unreplicated RNA-sequencing experiments. We present an objective Bayesian method to construct credible intervals for mid-parent fold changes in RNA-seq data, based on the Jeffreys prior for the negative binomial distribution. When the dispersion can be assumed to be some known, small value, these credible intervals' coverage rates are nearly the same as nominal, justifying their use as confidence intervals. These confidence intervals can provide estimates of the fold changes and can be used to derive p-values to test for mid-parent heterosis while controlling the false discovery rate (FDR). We present results from simulation studies to assess the efficacy of our proposed method.

CLEAR: Concise List Enrichment Analysis using R

Xinglin Jia[1], An Phan[1], Karin Dorman[2] & Claus Kadelka[3]

[1]Bioinformatics and Computational Biology Graduate Program; Department of Mathematics, Iowa State University, Ames, IA
[2]Department of Mathematics, Iowa State University, Ames, IA
[3]Department of Statistics, Iowa State University, Ames, IA

High-throughput methods provide genome-wide data for genes or other molecular features. Since biological functions rely on interacting proteins rather than individual genes, gene set analysis (GSA) is crucial for interpreting large-scale datasets. Model-based GSA methods such as GenGO and MGSA use probabilistic approaches to infer active biological categories. GenGO identifies Gene Ontology (GO) categories using a generative model that accounts for noise and overlapping GO terms to reduce redundancy. MGSA extends this framework by introducing a Bayesian network, simultaneously inferring all categories and improving robustness against noise. These methods return concise, non-redundant gene sets, unlike traditional approaches (e.g., Over-representation Analysis (ORA) and Gene Set Enrichment Analysis (GSEA)), which test sets individually.

However, GenGO and MGSA rely on binary gene activation states determined by arbitrary thresholds rather than continuous test statistics such as effect sizes or p-values. Some MGSA extensions incorporate Gene Ontology topology or additional constraints to improve performance but disregard statistical information associated with genes. We propose Concise List Enrichment Analysis using R (CLEAR), a Bayesian model-based method that directly models gene-level statistics instead of binary activation states.

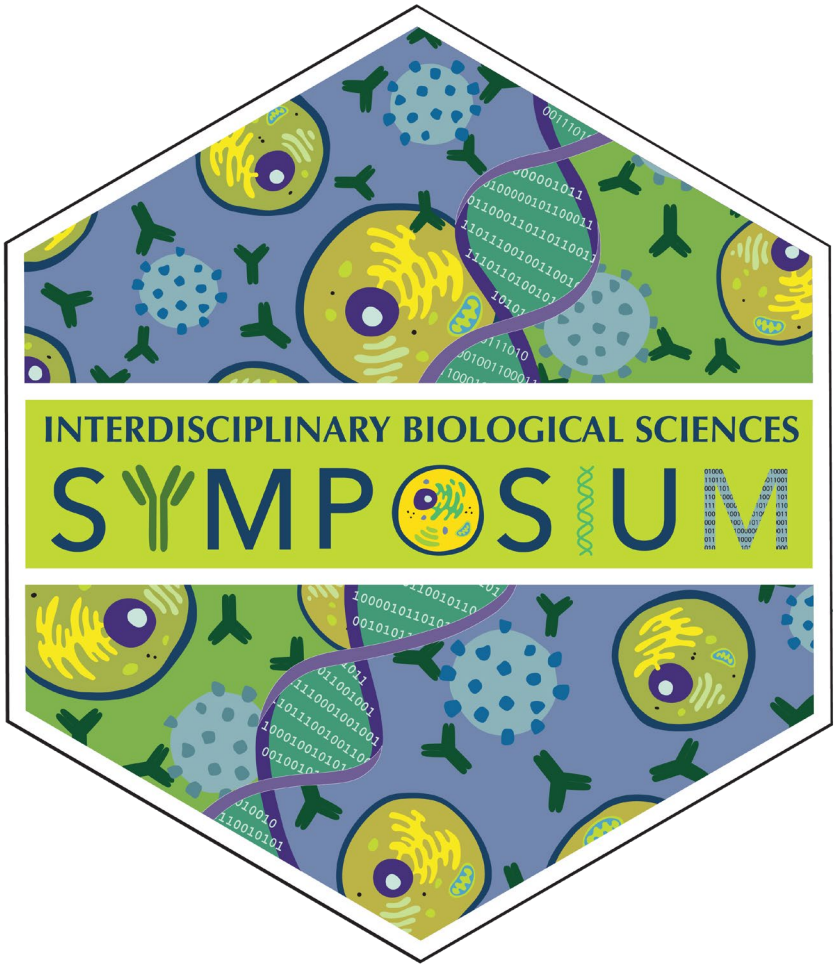
CLEAR assumes gene statistics follow distinct distributions under alternative and null hypotheses, enabling a more sensitive and nuanced interpretation of gene variation. This probabilistic, continuous framework improves robustness and interpretability. Using in silico and real datasets, CLEAR demonstrates higher sensitivity and improved interpretability by reducing redundancy and preserving signals. In conclusion, CLEAR is a powerful gene set enrichment method that leverages gene-level statistics to identify relevant gene sets with greater precision.

Defining trophoblast cell lineages within the rat placenta using single-cell RNA-sequencing.

Sabrena Rutledge[1], Marija Kuna[2], Michael J. Soares[2] & Geetu Tuteja[1]

[1]Bioinformatics and Computational Biology Program; Department of Genetics, Development, and Cell Biology, Iowa State University, Ames, IA
[2]Department of Pathology and Laboratory Medicine, Institute for Reproductive and Developmental Sciences, University of Kansas Medical Center, Kansas City, KS

The placenta is crucial during pregnancy, and its improper formation is linked to disorders affecting the mother and fetus. Trophoblast cell invasion into maternal tissues is essential for remodeling blood vessels to ensure sufficient blood flow to the placenta. The depth of invasion in rats is similar to what is observed in humans, making the rat a useful model for investigating invasive trophoblast cells. However, the developmental trajectory of invasive trophoblast cells in rats is poorly understood. This study uses single-cell RNA sequencing (scRNA-seq) to annotate the invasive trophoblast cell types, which arise from a compartment in the rat placenta termed the junctional zone. Placentation sites were collected at gestation day (gd) 14.5, and junctional zone compartments were isolated for scRNA-seq. Intact placentation sites were collected and frozen for histological analysis. scRNA-seq data was processed and analyzed with CellRanger and Seurat. The resultant 22 clusters were annotated as ten cell types based on the expression of known marker genes and an annotation tool we have developed that leverages human gene expression data. Focusing on trophoblast cells, re-clustering identified 14 new subpopulations. Pair-wise differential expression analysis organized half of these subpopulations into a differentiation trajectory, indicating both progenitor-like proliferative and mature trophoblast cells in the gd 14.5 junctional zone. Subpopulation-specific markers were validated using in-situ hybridization, confirming their spatial localization and supporting computational predictions. Assays of the junctional zone at other gestational stages will further refine trophoblast cell trajectories and will improve our understanding of invasive trophoblast cell lineage development.



Student Poster

Presentation Abstracts

Evaluating the effect of shockwave impacts on the behavior of adult hippocampal progenitor cell neurospheres

Catherine Fonder, Md Fazlay Rubby, Long Que, Donald S. Sakaguchi

Traumatic brain injuries (TBIs) affect millions of individuals annually, which can result in short-term impairments affecting movement, cognition, and communication, with severe cases resulting in permanent disabilities or even death. Unfortunately, there is a lack of understanding cellular mechanisms underlying the effects of TBIs, leading to inadequate treatment options. In our study, we evaluated the effects of shockwave impacts on adult rat hippocampal progenitor cells (AHPC) cultured as three-dimensional neurospheres, which we used as an in vitro brain model, for enhancing our understanding of the mechanisms of TBI. AHPCs are multipotent neural cells found in the adult brain and can differentiate into the three primary neural cell types of the central nervous system in vitro, making them ideal candidates for these shockwave experiments. AHPCs were exposed to a single session of 100 shockwave pulses at a frequency of 5 Hz at either mild (2 bar) or moderate (4.5 bar) strength. Cell viability was assessed three days post-exposure and immunocytochemical analysis was performed seven days post-exposure. These analyses showed there was a negative impact of the shockwave exposure on cell viability, proliferation, and differentiation of the cells within the neurospheres with increased shockwave blast strength. RNA was also isolated at seven days-post exposure and the qPCR gene expression analysis appeared to confirm some of the immunocytochemistry results. These results suggest AHPC neurospheres could be used to develop a TBI model and that some of the pathology observed following TBI may result from decreased cell viability and differentiation.

Does modularity emerge through evolution? Evidence from a Boolean network-based computational study

Faith Olusegun, Claus Kadelka

Boolean networks offer a simple way to represent complex biological systems, including gene regulatory networks. Nodes represent genes, a directed network, known as wiring diagram, describes their interactions, and Boolean update functions encode the regulatory logic. At any time step, a node takes on a binary value, which indicates whether the gene is "on" or "off". Time is discretized as well. Modularity is thought to simplify complex biological processes by separating them into smaller sub-units. Traditionally, highly connected components of the wiring diagram are known as modules. A recent, more rigorous definition of modularity identifies the strongly connected components of the wiring diagram as modules. To investigate whether this type of modularity emerges through evolution in molecular systems, we conducted a series of random Boolean network experiments. Since we do not know how evolution optimizes gene regulatory networks, we examined various objectives like maximizing phenotypical robustness or dynamical complexity. By considering different weights for these objectives, we obtained a variety of fitness functions. Inspired by natural selection, we evolved a population of random Boolean networks over many generations, preferentially selecting those with higher fitness for recombination. Recombination and mutation were parametrized in various ways. We tracked changes in the number of modules over many generations and found that modularity increased only for certain fitness functions. Additionally, assumptions about recombination and mutation influenced the emergence of modularity. Overall, these preliminary results indicate that modularity, as measured by the number of strongly connected components, can emerge through evolutionary processes, motivating further investigation.

PRRSV Ct Value Distributions Across Sample Types

Rabsa Naseer, Michael Zeller, Phillip Gauger, Giovani Trevisan

Accurate and timely diagnosis of PRRSV is crucial for effective disease management. Reverse transcription quantitative polymerase chain reaction (RT-qPCR) is a key diagnostic tool, measuring the cycle threshold (Ct) value to detect the virus. This study analyzes the distribution of Ct values across sample types, PRRSV ORF5 lineages, and years, providing insights into diagnostic utility. Data was sourced from the Iowa State University Veterinary Diagnostic Laboratory (ISU VDL), encompassing years of PRRSV testing records. This repository offers access to historical and current PRRSV diagnostic results, including specimen type, Ct value, collection date, ORF5 sequence lineage, and associated metadata. The dataset includes over 31,000 sequences, with information on specimen type, Ct value, collection date, ORF5 sequence lineage, and associated metadata. Analyses focused on cases with positive Ct values (<38) and ORF5 sequences. Data was divided by sample type, emphasizing lung, serum, oral fluid, and processing fluid. Analyses were conducted in R to compare Ct value distributions by specimen type, year, and lineage. Lung samples had the lowest Ct values, with a median of 18.7, compared to serum (24.9), processing fluid (27.5), and oral fluid (29.3). Median Ct values remained stable over the years for most lineages and specimen types. However, Lineage 1C exhibited variability, with median Ct values for lung samples ranging from 16.7 to 19.6 over time. Lineages 1C.2 and 1C.3 showed similar variability, with median Ct values for lung samples ranging from 16.7 to 21.4 and 16.5 to 19.5, respectively. This analysis reveals key trends in viral detection and load, based on sample type. The findings highlight the importance of specimen type and viral lineage in Ct values, offering insights into sampling strategies, diagnostic needs, and further molecular characterization. These insights help optimize testing strategies and biosecurity measures. While preliminary, the study provides valuable guidance for improving diagnostic protocols and ensuring timely PRRSV detection.

Phenotyping Differentially Abundant Proteins in Cryopreserved vs Fresh Bull Sperm

Alexandra Keller, George Perry, Karl Kerns

Semen cryopreservation is increasingly utilized in the livestock industry to preserve and distribute genetics from high indexing genetic sires. This technology has provided the ability to share high value genetics around the world and decrease biosecurity concerns involved with transporting live animals. It is not well understood, though, how cryopreservation affects the quality of preserved cells, but it is known that quality markers such as motility and membrane viability are decreased upon thawing. Mass spectrometry (MS) analysis was employed to identify distinct proteomic differences between fresh and cryopreserved sperm cells. Extracts were submitted for MS analysis at the Iowa State University Proteomics Core. A total of 2411 proteins were identified between both treatments. Of those, 402 were unique to the fresh replicates and 57 were unique to cryopreserved replicates. Principle component and cluster analysis revealed 154 proteins with differential abundances ($p<0.05$). In regard to those, 106 proteins were less abundant in cryopreserved samples and only 48 more abundant. Based on these identified proteins, three were selected for further phenotyping. Cells were mounted on ploy-L-Lysine slides and images were acquired with a Leica DM6 THUNDER upright microscope. Localization was seen in the equatorial segment by cytochrome C, RPS3A, and DDOST. Additional localization was seen in the apical ridge by labeling with PRS3A and DDOST and unique labeling in the principal piece of some cells was seen by cytochrome-c. Future steps involve elucidating localization differences between these three proteins in fresh and cryopreserved bull sperm by additional immunocytochemistry and validation of differential abundances by western blot analysis. This research aims to identify protein targets for optimization of cryoprotectants to maintain optimum cell quality and thereby ensure the reproductive potential of cryopreserved semen used in artificial insemination.

Evaluating single nucleus RNA sequencing in Gossypium (cotton) fiber

Corrinne Grover**,** Marc Libault, Sandra Thibivilliers, Jonathan F. Wendel, **Onyinyechi J. Amaechi**

Cotton fiber is a seed trichome found among members of Gossypium (cotton). The cotton genus arose about 5-10 million years ago, and among its >55 species, four were independently domesticated for the fiber trait. As is common among crop species, two of these domesticated species, G. hirsutum and G. barbadense, are derived from a monophyletic polyploid lineage whose traits such as fiber quality and productivity, outperform their domesticated diploid counterparts. Notably, cotton fibers are single cells which undergo several stages of development that are distinct from the surrounding shorter "lint" cells, which also populate the ovular surface. Single-nucleus RNA sequencing (snRNA-seq) is a technique developed to survey expression from individual cells while also addressing the problem of extracting transcriptome data from hard-to-dissociate cells, such as individual fiber cells from their adjacent lint cells. Here, we survey the suitability of snRNA-seq to survey cotton fiber expression in polyploid G. hirsutum at two timepoints early in fiber development. Initial analyses suggest that the single-end nature of snRNA-seq combined with shorter read length (90 nucleotides) may complicate analyses in a polyploid species, which may require optimization of existing pipelines. Complementing each snRNA-seq sample is simultaneously generated snATAC-seq, which may also require optimization of bioinformatic pipelines before application as a survey of chromatin accessibility. Once the bioinformatic challenges of working in a polyploid species are overcome, these data will be used to compare gene expression and chromatin availability in individual cell populations for early DPA between TM1 (older cultivar) and CSX8308 (modern elite line).

Genetic analysis of pod traits in Iowa Mung bean Diversity Panel using image-based phenotyping

Venkata Naresh Boddepalli, Kim Daeun, Talukder Z Jubery, Baskar Ganapathysubramanian, Arti Singh

Mung bean (Vigna radiata (L.) R. Wilczek) is a promising crop for the plant-based protein industry due to its digestible proteins, short growth cycle, and nitrogen-fixing ability. This study utilized image-based phenotyping to evaluate pod-related traits in 372 genotypes from the Iowa Mungbean Diversity Panel (IMDP) grown across three locations in the 2022-2023 seasons. Using an Epson scanner (300 dpi), 20 pods per genotype were imaged, and traits like pod length, width, curvature, and seeds per pod were analyzed via image processing (CRAFT method) and machine learning. High correlations were found between image analysis and manual measurements for pod length (r = 0.92) and seeds per pod (r = 0.74). Genome-wide association Study (GWAS) using GAPIT and BLINK models identified 23 quantitative trait loci (QTL), with one significant SNP (5_35265704) associated with all four traits, explaining 12-45% phenotypic variance. A gene within this QTL, Vradi01g00001116, showed strong homology with genes in Arabidopsis, cowpea, common bean, and soybean, particularly the Arabidopsis ortholog AT4G27260 (GH3 family), which influences organ elongation and pod development. This research provides insights to develop genetic markers for breeding mung bean cultivars adapted to the US Midwest.

Investigating the Xyloglucan Xylosyltransferase Heterodimer Organization

Reyna Winders, Jordan Julian, Olga Zabotina

In plants, the cell wall is composed of three different groups of polysaccharides – cellulose, hemicelluloses, and pectins. The hemicellulose xyloglucan is synthesized by specific enzymes localized in the Golgi that connect monosaccharides. These membrane-bound enzymes are known as xyloglucan xylosyltransferaseses (XXTs). There are five Arabidopsis XXT isomers that attach xylose to specific glucose residues in the glucan backbone. XXTs carry out a biosynthetic reaction between UDP-xylose and a glucan backbone, where the xylosyl residue is transferred to a glucose residue, forming the branch. XXT2 binds xylose to the first and second glucose, whereas XXT5 binds xylose to the third glucose in the glucan backbone. These two enzymes produce a glucan backbone branched with three xyloses and one unbranched glucose (XXXG). Our lab has recently demonstrated that XXT2 and XXT5 form a heterodimer. Using X-ray crystallography, we obtained the structure of the heterodimer of XXT2 XXT5. Our findings in the structure suggest that the stem region, the flexibly random coiled area of the enzyme, and the catalytic domain of XXT2 tightly interacts with the catalytic domain of XXT5. Different mutations have been generated in the stem region of XXT2 with the goal of investigating this interaction between XXT2 and XXT5. These mutants will be used to confirm the interaction in the heterodimer using in vitro pull-down assays. We hypothesize that the specific mutation on the stem region of XXT2 will disrupt XXT2 XXT5 heterodimer formation.

—No Topic---

Memis Bilgici, Thomas Lubberstedt, Hirsch, C, Yandeau-Nelson

--No abstract---

FHOD3 and DIAPH3 control stress fiber organization and cell migration through shared but non-overlapping mechanisms

Fred Rogers Namanda, Azarnoosh Foroozandehfar, Gopal Niraula, Ian C. Schneider

Cell morphology is primarily determined by the organization of the filamentous actin (F-actin) cytoskeleton. Cell morphology influences essential processes like cell migration, allowing wounds to heal and cancer to metastasize. Cells adopt particular morphologies and migrate in response to interactions with the extracellular matrix (ECM), which presents not only chemical cues but also mechanical cues such as ECM stiffness that regulates cytoskeletal structure and dynamics. It is well known that cell cytoskeleton has different F-actin structures, such as branched and linear F-actin bundles. Formins, a family of F-actin regulatory proteins, are key mediators of linear F-actin assembly into stress fibers. While prior studies have highlighted the roles of formins in F-actin regulation, the specific contribution of FHOD3 and DIAPH3 to the assembly and maintenance of stress fiber subpopulations and how they control cell morphology and migration remains poorly understood. In this work, we explore how FHOD3 and DIAPH3 contribute to the assembly and maintenance of stress fiber subpopulations and their role in controlling cell morphology and migration. We characterize formin expression patterns across different cell types and correlate these expression patterns of formins to different morphological features to identify formins heavily involved in driving cell morphology. We identified FHOD3 and DIAPH3 as highly correlated with cell elongation, suggesting they regulate parallel stress fiber assembly. We demonstrated that FHOD3 knockdown dramatically decreased the parallel F-actin stress fiber length in a stiffness-and cell-type dependent manner, while the length of the perpendicular F-actin stress fibers remained roughly the same. DIAPH3, on the other hand, regulates both parallel and perpendicular stress fibers. These results strongly suggest that FHOD3 and DIAPH3 regulate overlapping, but distinct F -actin stress fibers networks. Further, we examine the effect of FHOD3 and DIAPH3 knockdown on cell migration, which shows that these formins diminish cell migration speed significantly. Future work will focus on understanding how FHOD3, DIAPH3, and other formins regulate directed migration in response to aligned collagen fibers and how they may control traction force generation by regulation of distinct F-actin stress fiber populations.

Chemistry-specific off-target effects of a therapeutic antisense oligonucleotide for the treatment of spinal muscular atrophy

Robert Kaas, **Wren Murzyn**, Eric W. Ottesen, Ravindra N. Singh

Designing therapeutic molecules while retaining specificity within a broad concentration range remains a challenging task. Nusinersen (F18MOE) is an FDA-approved compound for the treatment of spinal muscular atrophy (SMA), the leading genetic cause of infant mortality. The therapeutic merit of F18MOE is based on its ability to restore inclusion of Survival Motor Neuron 2 (SMN2) exon 7 during pre-mRNA splicing. F18MOE is an 18-mer antisense oligonucleotide (ASO) encompassing phosphorothioate (PS) backbone and 2′-methoxyethyl (MOE) modification. F18MOE targets intronic splicing silencer N1 (ISS-N1) discovered in Singh lab. Many ISS-N1-targeting ASOs with different chemistries, including 2′-O-methyl (OMe) and morpholino (PMO) have been shown to restore SMN2 exon 7 inclusion. We performed RNA-Seq to capture transcriptome-wide off-target effects of three ISS-N1-targeting ASOs, namely F18OMe, F18MOE and F20PMO, encompassing OMe, MOE, and PMO chemistries, respectively. Here we present chemistry-specific off-effect of F18MOE on splicing of exon 2 of POLR2H gene, which codes for a protein subunit of RNA and DNA polymerases. We generated minigene to uncover the role of specific sequences responsible for the off-effect of F18MOE. Our results uncovered positions within target sequence more prone to tolerating mismatch mutations. Our findings are instructive in designing future ASO-based therapies for diseases requiring splicing correction.

GWAS Approach to Identify Diagnostic Markers in Avibacterium paragallinarum

Oluwatobiloba Aminu, Mostapha Shelkamy, Amro Hashish, Eman Gadu, Yuko Sato, Mohamed El-gazzar

Objectives: Infectious coryza, caused by Avibacterium paragallinarum (AP), leads to significant economic losses in the poultry industry by reducing egg production. Current diagnostics cannot differentiate pathogenic from non-pathogenic strains leading to inaccurate diagnosis. We hypothesize that pathogenic strains harbor unique genetic markers—potential diagnostic targets—absent in non-pathogenic strains and vice versa. Here, we propose a genome-wide association study (GWAS) to identify these markers.
Methods: We analyzed 82 pathogenic and 14 non-pathogenic whole genome sequences from the NCBI database, representing known serovars of A. paragallinarum. Genomes were annotated with Prokka, and pangenome analysis was conducted using Panaroo on the resulting GFF files. The gene presence/absence matrix served as input for a linear mixed model (LMM) GWAS, which controlled for population structure using a similarity matrix derived from core gene alignments.
Results: Pangenome analysis identified 4102 genes, including 1460 core genes (present in ≥99% of sequences). Rarefaction analysis, fitted with Heaps' law, yielded an alpha of 2.0 indicating a closed pangenome. Functional annotation via eggnog mapper revealed, most of the protein as functionally unknown (labelled S) and other proteins primarily involved in transcription, translation, and cell wall biogenesis. The LMM analysis of 2631 variants identified three significant genes: the dmlR-arg-PdmlR_1 cluster, mdaB, and group_444. The dmlR-arg-PdmlR_1 cluster includes argP, a LysR-type transcriptional regulator involved in arginine and lysine metabolism, and mdaB, a NADPH quinone reductase that protects cells from oxidative stress. group_444 remains functionally unannotated.
Conclusion: Our GWAS has identified candidate gene targets for developing PCR diagnostics that differentiate pathogenic from non-pathogenic strains of A. paragallinarum. Future work will explore core gene variability and refine phenotype definitions to enhance GWAS analysis.

PRRSV ORF5 Ct Values: Vaccine-like Strains Show Higher rtPCR Levels

Rabsa Naseer, Michael Zeller, Phillip Gauger, Giovani Trevisan

Various strains of porcine reproductive and respiratory syndrome virus (PRRSV) may cause differences in the severity of clinical signs, adverse animal health impacts, and economic losses in swine populations. Understanding the relationship of cycle threshold (Ct) values between wild-type and vaccine-like strains is essential for assessing viral dynamics and identifying samples to investigate potential recombination events that may pose future clinical risk. Data was sourced from the Iowa State University Veterinary Diagnostic Laboratory (ISU VDL). This repository offers access to historical and current PRRSV diagnostic results, including specimen type, Ct value, collection date, sequence, lineage, and associated metadata. The dataset included only Lineage 5A (L5A) and Lineage 1D (L1D) sequences from 2018 to 2024, as these lineages possess robust diagnostic data and associated vaccine strains; Ingelvac and Prevacent PRRS modified live virus (MLV) vaccine strains, respectively. The data was divided by sample type, with an emphasis on lung and serum samples. Analyses focused on positive Ct values (<38). Sequences were classified as vaccine-like or wild-type based on a 98% threshold comparison to the relevant vaccine strains. Statistical analyses in R compared Ct value distributions by classification and lineage. Statistically significant differences in median Ct values between vaccine-like and wild-type samples were identified at a 0.05 significance level. Annotated phylogenetic trees showed distinct clusters for wild-type and vaccine-like sequences, indicating phylogenetic differentiation. Wild-type sequences had lower Ct values, suggesting higher viral loads. However, some vaccine-like strains also had low Ct values, warranting further investigation and surveillance. Understanding the implications of Ct value distribution could help inform the early detection of strains with more severe clinical implications and viral behavior, enabling the rapid development of effective biosecurity and management strategies against PRRSV in swine.

Rewiring endothelial cells toward definitive hemogenic endothelium

Masuma Khatun Usha, Radwa Barakat, Elizabeth Snella, Xiaoyi Cheng, Clyde A. Campbell, Raquel Espin-Palazon

The hemogenic endothelium (HE) is the developmental intermediate that gives rise to hematopoietic stem and progenitor cells (HSPCs), originating from endothelial cells (ECs) lining the floor of the dorsal aorta (DA). While HE is identifiable by co-expression of endothelial and hematopoietic markers, its precursors (pre-hemogenic endothelium, pre-HE) remain elusive due to the lack of specific markers that can effectively identify ECs with hemogenic potential from non-hemogenic counterparts. Our lab's custom NF-kB reporter zebrafish embryos are the first tool to distinguish pre-HE from non-hemogenic ECs in vivo. However, this tool is unavailable in human systems, highlighting the need for a universal surface marker for pre-HE. To address this, we identified the top 10 upregulated DEGs in NF-kB^{+}; kdrl^{+} pre-HE vs. NF-kB^{-}; kdrl^{+} non-hemogenic ECs and performed WISH in 16–24 hpf zebrafish embryos. We found that lyve1b (lymphatic vessel endothelial hyaluronan receptor-1) presented the most robust and specific expression within the developing vascular cord. We then analyzed their expression in our scRNAseq atlas of 16–24 hpf embryonic hematopoietic cells, confirming their restricted co-expression in a specific EC sub-cluster (lyve1b+ cluster). Furthermore, the lyve1b positive ECs co-express NF-kB during pre-HE stage and are non-mitotic at that stage. However, LYVE1 in mammals has been extensively used as a marker of lymphatic endothelium. Literature shows that lymphatic vessels arise from specialized angioblasts with hemogenic potential repressed by Prox1. In agreement, we found Prox1 ablation in zebrafish increased HE and impaired lymphatic vessel development. In conclusion, identifying a pre-HE marker could advance pluripotent stem cell (PSC)-derived hematopoiesis and improve disease modeling, cell therapy, and transplants.

Decoy engineering: a trap for plant pathogens

Chunoti Changwal, Alexandra Margets, Jessica Foster, Roger Innes, Thomas J Baum

Plants have an innate ability to recognize and respond to pathogens. By exploring such recognition mechanisms, one can devise novel specificities for pathogen recognition. For example, plants possess specific disease resistance (R) genes that recognize strains of the bacterial pathogen Pseudomonas syringae. During infection, bacterial proteins are injected into the plant cell, where their action is recognized by resistance proteins, which triggers an immune response. One of these secreted proteins is AvrPphB, a cysteine protease effector. In Arabidopsis, this protease effector cleaves the plant protein kinase PBS1, which in turn activates the R protein RPS5 leading to strong defense responses. A similar mechanism also exists in soybean. Building on this knowledge, we are developing a system to detect plant pathogens by modifying the protease cleavage site within PBS1 to be recognized by protease effectors from other pathogens.We are applying this concept to create new disease resistance in soybeans against the soybean cyst nematode.

Impact of tryglysin on metal homeostasis in oral streptococci

Sristi Dey, Britta Rued

Streptococci are one of the key contributors to the functional oral microbiome and play essential roles in maintaining oral health. These bacteria produce a variety of natural products termed as Ras-RiPPs (Radical S-adenosylmethonine ribosomally translated and post-translationally modified peptides) that are controlled by Rgg-type quorum sensing systems. Our research primarily focuses on tryglysins, a class of Ras-RiPPs that inhibits the growth of oral streptococci. To further explore the effects and mechanism of tryglysins in the oral environment, we performed RNA-sequencing on sensitive and producer species treated with subinhibitory concentrations of tryglysin A. These results were then confirmed by qRT-PCR. RNA-seq and qRT-PCR results revealed that Streptococcus mitis treated with tryglysin A induced differential responses in genes related to zinc uptake, survival under stress conditions, and metal homeostasis disruption. S. mutans, another important dental pathogen, exhibited increased expression of stress response related genes such as those involved in purine biosynthesis and ribosome function. In contrast, the tryglysin A producing strain S. ferus had altered gene activity involved in zinc and drug efflux transport. The downregulation of certain genes confirmed by qRT-PCR suggests the necessary suppression of their expression to manage stress and promote cell survival during tryglysin treatment, while overexpression of genes implicates adaptation effects to counteract the treatment of tryglysin. These findings demonstrate the ability of tryglysins to alter metal homeostasis, trigger stress responses, and modulate variations in gene activity across different strains of Streptococci.

Investigating Seed Treatments and Soil Amendments to Improve the Establishment of Kentucky Bluegrass as a Perennial Groundcover

Jack Moran, A. Susana Goggi, Ken J. Moore, Shui-zhang Fei

Kentucky bluegrass (KBG) has poor seed establishment in the fall when used as a perennial groundcover in corn production. This study was conducted to investigate the effect of various seed treatments and soil amendments on the establishment of KBG under drought and non-drought conditions, simulated in a growth chamber. The effect of seed treatments, soil amendments, and irrigation frequency on KBG germination and shoot dry weight were measured over 21 days in a controlled environment at 21 °C, 50% RH, and exposure to a constant red light. The treatments were the Hydroloc seed treatment, a lime soil amendment, the Pivot Bio seed treatment, an ammonium nitrate soil amendment, a gibberellic acid seed treatment, osmotic seed priming, and an untreated control. The layout was a randomized complete block design, with two irrigation frequencies (restricted and full irrigation) and four replications (blocks). The irrigation treatments were applied to whole plots and the seed treatments were applied to subplots. The entire experiment was repeated four times. Irrigation affected the germination of all the seed treatments. The control and Hydroloc treatments did not have significantly different dry shoot weights, while all the other treatments had significantly different dry shoot weights when comparing the irrigation regimes. The Hydroloc treatment significantly outperformed all the other treatments in regard to the restricted and full irrigation regime. These results indicate that the Hydroloc seed treatment improves KBG germination and shoot dry weight in drought and non-drought conditions, promoting KBG establishment in a wide range of soil moisture conditions.

Coupling Bioinformatics approaches and Oxford Nanopore (ONT) sequencing for enhanced Diagnosis of Newcastle Disease Virus (NDV) in Poultry

Swathi Nadendla, Maria Chaves**,** Eman Gadu, Amro Hashish

Objective: Newcastle Disease virus causes respiratory, systemic illness in poultry, controlled through modified live vaccines. Rapid and accurate genetic characterization is critical for distinguishing wild type from vaccinal strains. Current typing uses genotype-specific primers for the Fusion (F) gene, requiring multiple steps that delay results, underscoring the need for a streamlined workflow. ONT enables real-time, long-read sequencing, but high background noise from host DNA in clinical samples necessitates enrichment. Effective enrichment relies on optimized primer design, yet existing pipelines need improvement. This study aims to harness ONT to develop a genotype independent workflow providing a scalable diagnostic approach.
Methods: To achieve a universal workflow for NDV we propose two enrichment approaches, a k-mer-based primer design pipeline and a nested PCR by using a comprehensive database. Conserved regions across the F gene were identified using a custom script, divided into overlapping segments enabling the primer design. To capture diversity, degeneracy was added. This design generates a broadly effective primer sets that needs to be further optimized insilco.
Results: The k-mer pipeline produced 38 primers, while nested PCR utilized two sets of inner and outer primers. Targeted amplicons were sequenced using MinION and data was processed through an inhouse analysis pipeline enabling continuous Fastq file pickup, base calling, read classification generating alignment statistics allowing rapid identification of NDV within hours. This live analysis facilitates in improved detection compared to traditional post run bulk analysis. While this pipeline currently supports real-time detection, further optimization is needed to reliably characterize genotypes.
Conclusion: This combined approach involves targeted enrichment, nanopore sequencing, and real-time analysis has the potential to become a field-adaptable solution. Our work demonstrates that next-generation technologies tailored to pathogen-specific challenges can significantly enhance diagnostic accuracy."

Analyzing Cohesion: Investigating Cdc23 S.Cerevisiae Mutations and it's Role in Chromosome Separation

Leticia Herrera, Mohan Gupta

Through our project, we are researching how genome-editing mutation using CRISPR/Cas9 of the yeast Saccharomyces cerevisiae (also known as baker's yeast) cohesion complex may reflect and illuminate upon which cell gene mutations are responsible for causing human congenital disabilities and cancers. Employing the CRISPR/Cas-9 genome editing technique, phospho-mutants were created for various proteins within the cohesin complex and screened for altered sensitivity to Carbendazim (a fungicide that prevents microtubules from correctly separating). A mutant of the Cdc23 cohesion gene yielded sensitivity to this drug, indicating potential defects. Therefore, the primary goal of our project is to isolate the mutation of interest and confirm that it is affected during cellular separation due to the given mutation. To accomplish this, a Cdc23 mutant yeast strain is mated with a wild-type MAT-α strain to form zygotes. These zygotes are then picked up using a microneedle under a microscope and grow into colonies, producing diploid cells. The diploids are then transferred to a nutrient-poor sporulation medium to trigger meiosis, resulting in tetrads containing four spores, including one spore with the Cdc23 mutation of interest. The four spores are then separated under a microscope and grown into individual colonies. A selection plating series will be executed based on the Cdc23 mutant strain to confirm the presence of the mutation of interest. Finally, PCR amplification and subsequent Sanger sequencing will verify the mutation's presence and effect on the cell.

Investigating Tryglysin A resistance and gene regulation in streptococci

Sophie A. Krivograd, Britta E. Rued

Streptococci are a Gram-positive bacterial species that are highly prevalent within the oral microbiome and can contribute to disease. Multiple species of streptococci including Streptococcus mutans and Streptococcus ferus produce modified peptides known as RaS-RiPPs (Radical S-adenosylmethionine ribosomally translated and postranslationally modified peptides). RaS-RiPP expression is controlled via Rgg-like quorum systems which rely on the binding of short hydrophobic peptides (SHPs) to a Rgg transcriptional regulator. Once bound, SHPs upregulate the expression of streptococcal Ras-RiPPS. One class of Ras-RiPPs, tryglysins, have been found to inhibit growth of other streptococci species like S. mitis; however, the mechanism behind this inhibition is unknown. To examine the mechanism of action in sensitive oral species, we performed a plate-based screen to isolate streptococcal strains resistant to tryglysins. This was performed in S. mitis, a species that is normally highly sensitive to inhibition by tryglysins. We identified several S. mitis strains that were fully resistant to Tryglysin A (TryA), which we refer to as sup1-10 and sup2-20. These strains possessed numerous point mutations, and several of these point mutations were being reconstructed for examination of their role in TryA resistance. Additionally, to further understand the role of TryA we examined the response of the producer species S. ferus to the peptide, and identified that own regulation genes next to a distinct Rgg system, RggA, were downregulated during TryA exposure. To better understand this connection, reporter plasmids for these genes were constructed in wild-type S. ferus and S. ferus ΔrggA. These will then be tested with addition of tryglysin. This experiment will determine if tryglysin impacts RggA signaling or if RggA is a standalone regulator. Understanding how resistance to tryglysin can arise and how tryglysin plays a role in gene regulation is crucial in determining if tryglysin can be used in future studies as an anti-microbial agent.

Determining the role of lncRNA BCAR4 in human trophoblast cells

Ludvin Mejia, Haninder Kaur, Amanda Kirk, Geetu Tuteja

Trophoblast cells in the placenta play a key role in establishing the uterine-placental interface, which is essential for fetal development and a successful pregnancy. Disrupted trophoblast cell function can lead to pregnancy complications such as preeclampsia, intrauterine growth restriction, and recurrent pregnancy loss. Long non-coding RNAs (lncRNA) have been associated with different pregnancy complications, yet the mechanism by which they regulate gene expression is poorly understood. Here, we focus on Breast Cancer Anti-Estrogen Resistance 4 (BCAR4), a lncRNA exclusively expressed in the placenta in healthy tissues. Interestingly, BCAR4 has been shown to promote breast cancer cell proliferation and migration, but its role in trophoblast cells remains unknown. The objective of this study was to investigate the role of BCAR4 in a primary trophoblast cell line. Initial experiments using quantitative polymerase chain reaction (qPCR) showed a significant increase (Fold ≥ 2, p-value < 0.05) in BCAR4 expression during trophoblast differentiation. Next, BCAR4 expression was knocked down using short-hairpin RNA (shRNA), resulting in impaired trophoblast differentiation. To gain insight into BCAR4's mechanisms of action, we utilized RNA fractionation to determine its subcellular localization in trophoblast cells. We observed a higher abundance of the BCAR4 transcript in the cytoplasmic fraction compared to the nuclear fraction. Localization to the cytoplasmic fraction suggests that BCAR4 interacts with cytoplasmic molecules. Future experiments will further investigate the mechanisms by which BCAR4 regulates trophoblast differentiation. These experiments include generating and analyzing RNA-sequencing (RNA-seq) data to identify molecular pathways disrupted by BCAR4 knockdown and using Comprehensive Identification of RNA Binding Proteins (ChIRP) to identify BCAR4's binding partners. Understanding how BCAR4 regulates trophoblast differentiation may provide insight into the mechanisms underlying pregnancy-related disease.

Environmental and Genetic Factors Underlying Maize Cuticular Wax Accumulation Under Drought Stress

Wendt Matthew M, Hattery. T, Hirsch. C, Yandeau-Nelson

One mechanism by which plants can protect against drought and other stresses is the hydrophobic cuticle, which is composed of cutin and very long-chain waxes and coats the epidermis of aerial plant tissues. The quantity and types of cuticular waxes are impacted by genetic and environmental factors, modifying the cuticle's efficiency as a hydrophobic barrier to non-stomatal water-loss. Here, we analyze the genetic and environmental factors underlying cuticular wax composition. Using cuticular wax composition data (45 metabolites) on silks across 448 inbred lines of the Wisconsin Diversity Panel (WiDiv), we identified candidate genes explaining variation in waxes between environments and across abiotic environmental gradients by estimating the contribution of specific alleles to either genotype or genotype-by-weather effects via multi-locus GWAS. Characterization of genes that contribute to the genetic architecture of cuticular waxes between environments will enable the design of weather resilient or "climate-smart" plants that remain healthy and productive under environmental stress.

The interaction of the Arabidopsis xyloglucan xylosyltransferases with the COPII member SAR1 via their di-arginine motifs is critical for delivery to the Golgi.

Ning Zhang, Jordan D. Julian, **Rachael K. Gorman**, Olga A. Zabotina

Golgi-localized Xyloglucan Xylosyltransferases (XXT2 and XXT5) participate in xyloglucan biosynthesis, and to do this, they require the proper localization. COPII complex is responsible for cargo protein delivery from ER to Golgi, which is recruited by the complex member proteins Sec23, Sec24 and Sar1. Also, the N-termini of glycosyltransferases (GTs) are important for their transportation and localization. In this study, we demonstrated that XXTs interact with SAR1 protein in the COPII complex but not with Sec24, which was previously reported to be the main recruiter of cargo proteins into COPII-coated vesicles. The mutation of the arginine to glutamine residues of di-arginine motifs in the N-termini of XXT2 and XXT5 caused protein mislocalization and significantly reduced the strength of the interaction with SAR1. These mutations caused 90% of XXTs either to remain in the ER or to localize to non-Golgi small compartments. In turn, such mislocalization significantly suppressed the xyloglucan biosynthesis in Arabidopsis mutant plants (xxt1xxt2 and xxt3xxt4xxt5), failing to restore their root phonotypes to normal. Our results are the demonstration of cargo proteins' interaction with Sar1 proteins and the critical role of di-arginine motifs in this interaction. These results provide new insights into the mechanism of ER to Golgi delivery of plant GTs, which significantly advances our understanding of polysaccharide biosynthesis in Golgi and the enzymes responsible for it.

Translation of Newly Discovered Maize associated Tombus-Virus (MaTV) Genome: An Unsolved Mystery

Jahanara Sonia, Seema Rani Raychaudhuri, Zachary Lozier

The Maize-associated tombusvirus (MaTV) genome, along with eleven related tombusvirids that we propose belong in a new genus (Lozier et al., 2024), features an unusually long 5' untranslated region (UTR) (622 nt) with multiple AUG triplets unlikely to serve as start codons. Unlike other tombusvirids that rely on a 3' cap-independent translation element (CITE), MaTV lacks a clear 3' CITE, suggesting the 5' UTR may function as an internal ribosome entry site (IRES). To investigate this, we constructed MaT-Luc-MaT, a luciferase reporter flanked by MaTV's 5' and 3' UTRs. In wheat germ extract (WGE), this construct failed to translate, unlike a control construct with Barley yellow dwarf virus (BYDV) UTRs, which translated efficiently due to BYDV's 3' CITE. To explore whether essential translational elements reside in MaTV's coding region, we tested a full-length MaTV transcript in WGE. Translation of ORF1, typically efficient in other tombusvirids, did not occur. However, replacing MaTV's 5' UTR with a short, AUG-free UTR restored ORF1 translation — but only from capped RNA. Interestingly, the wild-type MaTV genome produced low levels of a protein matching the expected size of the coat protein (CP), from both capped and uncapped transcripts, suggesting weak IRES activity upstream of the CP ORF. This resembles the IRES reported for turnip crinkle virus. The translation of essential ORFs 1 and 2, crucial for replication, remains unexplained, indicating that MaTV likely employs an unconventional, yet-to-be-identified translation mechanism.

Dissecting Puccinia sorghi haustoria at the molecular level: A multi-omics investigation

John Ssengo, Christian Montes-Serey, Justin Walley, Shawn Rigby, Christine Deal, Steven Whitham

Rust fungi are biotrophic pathogens that depend on living host cells for survival. They form haustoria, which are specialized structures that extract nutrients from host cells and deliver effectors that facilitate immune evasion. Currently, the molecular mechanisms governing haustoria development and function remain poorly understood. Transcriptomic studies have been used to predict haustorial-expressed gene activity, overlooking protein-level regulation. To investigate the haustorial proteome and phosphoproteome, we are employing fluorescence activated cells sorting (FACS) to purify intact haustoria from Puccinia sorghi, which causes common rust (CR) in maize (Zea mays). Haustorial-expressed proteins are then identified and quantified using mass spectrometry. In preliminary experiments, we have identified 786 proteins, including 93 unique candidate effectors validating this approach for providing insights into haustoria proteome composition. These results lay the groundwork for phosphoproteome analysis, which will highlight post-translational modifications that may influence protein activity, signaling, and maize-fungus interactions. By integrating proteomic and phosphoproteomic data with transcriptomics, we plan to create a haustorial gene regulatory network. We expect that the detailed molecular characterization of the protein composition of P. sorghi haustoria will advance knowledge of fungal virulence mechanisms and inform strategies to mitigate common rust in maize.

Exploring the Efficacy of FDA-approved inhibitors on Rift Valley Fever Virus Pathogenesis

Breigha Boyle, Lauren Bloodgood, Brianna Jones, Todd Bell

Rift Valley Fever virus, in the Bunyavirales order and Phenuiviridae family, is an emerging zoonotic pathogen and public health threat. We have previously published that combination kinase inhibition suppresses the pathogenesis of Rift Valley Fever virus (RVFV) through diminished ability for viral replication. Globally, there is a lack of available antiviral treatment options for this family of viruses. We are now working to repurpose FDA-approved inhibitors to create a multimodal treatment option for RVFV. We initially performed an analysis via reverse phase protein microarray (RPPA) to identify phospho-signaling changes associated with early, mid, and late infection phases. We found several pathways upregulated in early infection with the most consistent upregulation happening in translational control pathways and cell cycle control pathways. We then sourced FDA-approved inhibitors for the signaling pathways most active during early infection. Using luminescence-based viability bioassays, we defined the cytotoxicity and efficacy concentration of these inhibitors in vitro first alone and then in subsequent combinations that mimic the findings from our earlier paper. To demonstrate proof of concept in human cells, we recreated these experiments in a human liver cell line, Huh 7s, to show similar efficacy in vitro. We have confirmed that we can significantly reduce the viral replication of RVFV in vitro using combination kinase inhibition from readily available FDA-approved kinase inhibitors with relatively low toxicity concerns. Our short-term future goals include exploring these drugs and drug combinations in vivo as well as collaborating with a nanoparticles team to optimize our drug delivery system. Our longer-term goals include exploring whether these drug combinations would be effective in infections by other members of the Bunyavirales order both in vitro and in vivo. Further, we are starting a collaboration to better understand how we can incorporate artificial intelligence to optimize identification of antiviral drug targets.

Solutions to Address Biases and Increase the Accuracy of Social Contact Matrices

Caden Klopfenstein, Claus Kadelka

The PREM framework, widely used for projecting social contact matrices globally, relies on twenty-year-old data from eight European countries. It may no longer accurately reflect contemporary social mixing patterns. This study aims to critically assess the biases inherent in these contact matrices when applied to current population age structures, focusing on the effect of outdated data and misreporting trends. Preliminary analysis suggests that these biases can lead to inaccurate epidemic projections, affecting the estimated basic reproduction number (R₀) and the efficacy of targeted interventions. For instance, imbalanced contact matrices can significantly impact R₀ estimates and delay peak incidence in diverse demographic settings. We investigate how the synthetic PREM contact matrices deviate from empirical contact patterns, particularly when compared to alternative approaches like Arregui et al.'s demographic adaptation methods. This comparison highlights the need for updated contact matrices that reflect modern demographic shifts and behavioral changes. To address these limitations, we propose integrating dynamic demographic data and socioeconomic factors into PREM's matrices, ensuring reciprocity in contact patterns. This approach aims to improve the reliability of epidemic models and public health interventions by correcting initial biases caused by outdated data and misreporting trends, ultimately enhancing the effectiveness of disease modeling and policy design. By modernizing contact matrices through empirical validation and adaptive demographic scaling, we can better simulate outbreak dynamics and public health strategies, ultimately providing more accurate projections for pandemic preparedness in diverse demographic settings.

Benchmarking BioEmu-1: Establishing a Baseline for Protein Structural Ensemble Prediction

Ushashi Bhattacharjee, Tirtho Roy, Koushik Howlader, Sayantan Chakraborty, Tanusree Bhattacharjee, Adrito Roy

Our study establishes a comprehensive baseline for evaluating BioEmu-1—a deep-learning approach for generating diverse protein structural ensembles—using high-resolution experimental data from the Protein Data Bank (PDB). Our primary objective is to set up standardized benchmarks for BioEmu-1 that will serve as a reference point for future studies and model refinements. We curate a dataset of proteins with documented dynamic conformations and preprocess the PDB structures to ensure consistency through alignment and the removal of non-essential elements. BioEmu-1 is applied to the corresponding protein sequences to generate ensembles of predicted structures. These predictions are quantitatively compared to experimental data using metrics such as Root Mean Square Deviation (RMSD), Template Modeling Score (TM-score), and Global Distance Test - Total Score (GDT-TS). An automated analysis pipeline, developed with Python and integrated with molecular visualization tools, facilitates the efficient computation and visualization of these metrics. The baseline framework established herein not only provides critical insights into the current performance of BioEmu-1 but also lays the groundwork for future studies aimed at advancing computational protein structure prediction and dynamics analysis.

Alternative splicing in Gossypium hirsutum during fiber development

M.D.G.P Premarathne, J.F. Wendel, C.E. Grover

Gossypium hirsutum is a tetraploid fiber crop that provides material for textiles. Fiber development occurs as individual epidermal cells of the ovule progress through four overlapping stages: initiation, elongation, secondary wall synthesis, and maturation. In each stage, coordinated gene expression contributes to the developmental programme, ultimately influencing fiber quality and productivity. The goal of this study is to characterize and quantify sub genome-specific alternative splicing (AS) events and how they change during fiber development. We utilized RNA sampled daily from G. hirsutum TM-1 from 6 to 24 days post-anthesis (DPA) and sequence the transcriptome using Illumina and PacBio Iso-Seq platforms. The high depth of the short reads provided greater transcriptome coverage to use in isoform quantification, whereas the long reads enabled full-length transcript identification. StringTie2 was used in conjunction with the existing annotation to assemble and quantify transcripts. A total of 86,228 different transcripts were identified across the fiber development stages. Integrating short and long read sequences using StringTie2 significantly improved transcript discovery compared to single platform approaches or existing genome annotations. Identified transcripts were of high precision and confidence, providing a comprehensive resource for studying AS in cotton fiber development. This study is expected to provide a foundation for understanding AS diversity during cotton fiber development. Future research will focus on stage specific AS events across G. hirsutum subgenomes at each DPA and how isoforms from each sub genome are differentially used during fiber development.

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2nd Annual Biological Sciences Oral and Poster Winners

April 2024

Oral Presentation

1st place - Megan DeTemple - Plant Biology

2nd place - Yunhui Qi - Statistics

3rd place - Xiaoyi "Chelsea" Cheng - MCDB

Poster Presentation

1st place - Matthew Wendt - Genetics and Genomics

2nd place - Ashley Paulsen - Genetics and Genomics

3rd place - Molly Kroeger - ImBio and David Hall - Genetics and Genomics

Honorable Mention

An Phan - BCB

Alyssa Hohman - Genetics and Genomics

Abbigail McCune - Genetics and Genomics

Pengxin Yang - Biochemistry

People's Choice

Mubashrah Mahomood - Animal Physiology

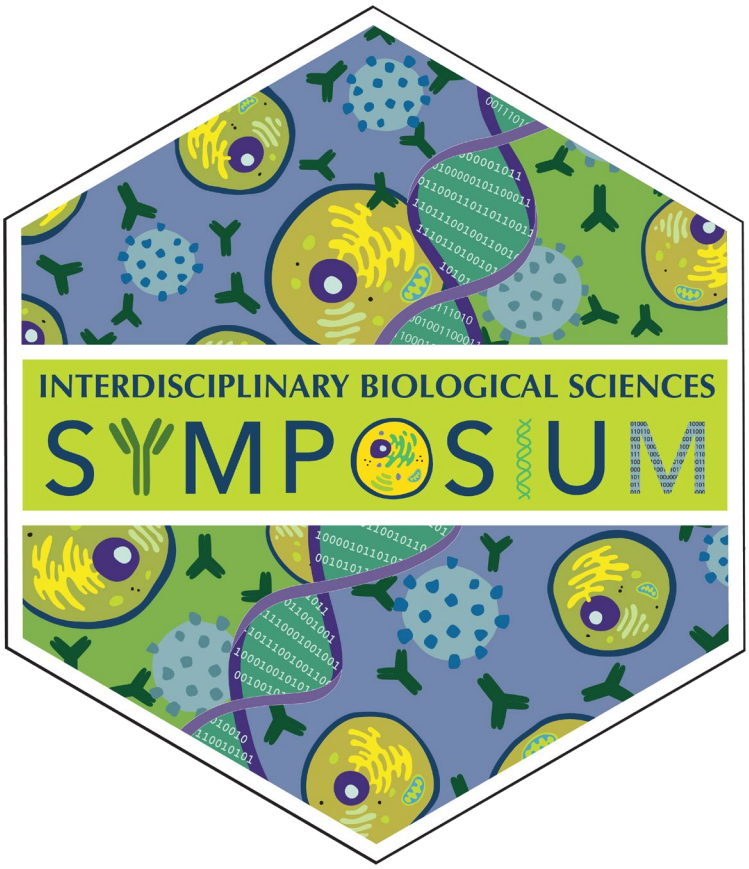
Sabrena Rutledge – BCB

Thank you for joining us for the 3rd annual Interdisciplinary Biological Sciences Symposium.

Looking forward to next year and hope to see you there.

Sincerely,

The 3rd Annual Interdisciplinary Biology Sciences Symposium committees



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