2022 Midwest *Drosophila* Conference

October 21st-23rd, 2022
In Person and Virtual

2022 Midwest Drosophila Conference Schedule

All times are in Central Standard Time

Friday, October 21

On-line poster kick-off event/happy hour

3:00-3:30 PM Networking: Please enjoy getting to know one another!
On-line Social Networking Tables open to all to support each other
First Generation
Underrepresented in Science
LGBTQA+
Women and Non-Binary in STEM

3:30-4:15 PM poster session 1: posters 1-24

4:15-5:00 PM poster session 2: Posters 25-48

Saturday, October 22

1:00pm In-person Check-In at Allerton Park and Retreat Center

1:30-1:45 PM - Opening Remarks

1:45 – 3:00 PM Platform presentation session: *Development*

Moderator: Olivier Devergne, Northern Illinois University

1:45 PM **Prostaglandins Limit Nuclear Actin to Control Nucleolar Activity During Oogenesis**

Danielle Talbot – Laboratory of Tina Tootle
University of Iowa

2:00 PM **Control of Crag's Localization and Activity in the Polarized Deposition of Basement Membrane Proteins in Epithelial Cells.**

Hemin Shah – Laboratory of Olivier Devergne
Northern Illinois University

2:15 PM **The Histone Chaperone NASP Maintains H3-H4 Reservoirs in The Early Drosophila Embryo**

Reyhaneh Tirgar – Laboratory of Jared T. Nordman
Vanderbilt University

2:30 PM **How the Thanos Requirement Leads to an End Game on Wing Fate During Ectopic Eye Formation**

Allie Smith – Laboratory of Justin Kumar
Indiana University

2:45 PM **Trithorax is an Essential Regulator of Cardiac Hox Gene Expression and Anterior-Posterior Patterning of the Embryonic Dorsal Vessel**

Adam J. Farmer – Laboratory of Kristopher R. Schwab
Indiana State University

3:00-3:30 PM Break Opportunity for Socializing/Networking

On-line Professional Development Networking Tables:

Applying for Graduate School

Going on the job market

Working at a PUI

3:30 PM -4:45 PM Platform presentation session 2: ***Evolution and Resource Updates***

Moderator: Sumit Saurabh, Loyola University, Chicago

3:30 PM **Prevention of Ribosome Collision-induced Neuromuscular Degeneration by SARS CoV-2- encoded Nsp1**

Xingjun Wang – Laboratory of Bingwei Lu
Stanford University

3:45 PM **Rapid Evolutionary Diversification of the flamenco Locus Across simulans Clade Drosophila**

Sarah Signor
North Dakota State University

4:00 PM **Fly-CURE, a Multi-institutional CURE, Has a Positive Impact on Students' Self-efficacy, Sense of Belonging, and Interest to Pursue Additional Research Experiences**

Julie Merkle
University of Evansville

4:15 PM **BDSC Update**

Cale Whitworth
Indiana University

4:25 PM **Flybase Update**

Brian Calvi
Indiana University

4:35 PM **DGRC Update**

Arthur Luhur
Indiana University

4:45-5:00 PM short break

5:00-6:00 PM Keynote Talk: Control of the Drosophila Germline Stem Cell Lineage by Diet, Temperature, and Whole-Body Physiology

Daniela Drummond-Barbosa
University of Wisconsin

6:00 PM - 7:00 PM Dinner

7:00 PM - 8:00 PM Poster Session 1

8:00 PM -9:00 PM Poster Session 2

9:00 PM - 10:00 PM Social Time

Sunday, October 23

8:00-9:00 AM Breakfast

9:00-10:15 AM Platform presentation session 3: ***Neuroscience and Signaling***

Moderator: Seth Tomchik, University of Iowa

9:00 AM **A Role for SIFa Receptor in the Regulation of Drosophila Circadian Feeding Rhythms**

Andi Beaudouin – Laboratory of Daniel Cavanaugh
Loyola University

9:15 AM **A FMRP-Dependent Pathway for the Glial Phagocytosis of Brain Neurons**

Jagarlamudi Rincon – Laboratory of Kendal Broadie
Vanderbilt University

9:30 AM **Drosophila Models of *SNRNP200*-Associated Retinitis Pigmentosa Exhibit Photoreceptor Abnormalities**

Sara K. Mayer – Laboratory of Lori L. Wallrath
University of Iowa

9:45 AM **Velvet Ant Venom Activates Pain-Sensing Neurons Through Pickpocket and Balboa, Homologs of DEG/ENaC and ASIC Channels**

Lydia J. Borjon – Laboratory of Daniel Tracey
Indiana University

10:00 AM **Rho GTPases Play an Important Role in Germ Cell Migration**

Mia Seohee – Laboratory of Afshan Ismat
University of Saint Thomas, Minnesota

10:15-10:45 AM Break Opportunity for Socializing/Networking and group photo

On-line Science Networking Tables:

Signaling and Development

Signaling and Disease

Neuroscience and Behavior

Evolutionary Biology

10:45 AM -12:00 PM Platform presentation session 4: *Gene Expression and Models*

Moderator: Maria L. Spletter, University of Missouri Kansas City

10:45 AM **Obesity is not a Direct Cause of Infertility**

Rodrigo Dutra Nunes – Laboratory of Daniela Drummond-Barbosa
University of Wisconsin

11:00 AM **Loss of E2F Regulation on The Expression of Phosphoglycerate Kinase (Pgk) Gene Exerts Broad Effects on Metabolism**

Maria Paula Zappia – Laboratory of Maxim V. Frolov
University of Illinois Chicago

11:15 AM **Ataxin-3 Ubiquitination at Lysine 117 Impacts its Toxicity in Drosophila Models of Spinocerebellar Ataxia Type 3**

Nikhil C. Patel – Laboratory of Sokol V. Todi
Wayne State University

11:30 AM **Role Of M1BP, A Transcriptional Pausing Factor in JNK-mediated Cell Death During Eye Development**

Anuradha Venkatakrishnan Chimata – Laboratory of Amit Singh
University of Dayton

11:45 AM **CDK11 Active Site Phosphorylation is Crucial for Cell Cycle Progression**

Abdulrahman Aljabri – Laboratory of Daimark Bennett
Taibah University, KSA and University of Liverpool

12:00-12:20 PM Awards and Business Meeting

12:20 PM Departure

Posters

[1] **Metabolic Effect Interactions Between Fragile X Syndrome and Glycogen Storage Disease Type IX in Two Drosophila Disease Models**

Aashi Gurijala and Kendal Broadie
Presenting Author: Aashi Gurijala
Vanderbilt University

The *Drosophila* model for Fragile X syndrome (FXS) is well established (Coffee, 2012). A possible relationship between FXS and Glycogen Storage Disease Type IX (GSD) surfaced in a 1993 case study on GSD "patient 2" who had mutations in Phosphorylase Kinase Regulatory Subunit Alpha 2 (*PHKA2*) as well as the Fragile X Mental Retardation Protein (FMRP) KH-type RNA-binding domain. He presented symptoms far more severe than in either disease alone, including heightened intellectual disability, severe macroorchidism, physical deformations, and difficulties with communication and movement (De Boulle, 1993). We hypothesized that the interaction between FXS and GSD results in an unsustainable elevated metabolic demand due to known heightened glucose metabolism in FXS (Qin, 2002). There is no established *Drosophila* GSD model, but *PHKA2* mutant and RNAi lines are available. We first assayed neuromuscular junction (NMJ) structural differences in third instar larvae, a well established FXS phenotype, comparing mutant genotypes. We find significant differences in NMJ branch length and synaptic bouton number in *dfmr1* and *PHKA2* mutants, as well as double heterozygote animals. We next investigated GSD effects on metabolism in larvae, using *PHKA2* RNAi with MitoTracker Orange™ to assay mitochondrial changes compared to w^{1118} background controls. After starving third instars for 15 hours, we saw a significant decrease in mitochondrial fluorescence between fed and starved control larvae. In contrast, starving *PHKA2* RNAi larvae caused no effect on mitochondrial fluorescence; both were comparable to starved control larvae. Future work includes starvation experiments with knockdowns of *PHKA2* and FMRP, individually and together.

[2] **Twin Roles Of The Zinc-Finger Transcription Factor Castor: Specification Of Cardiac Cell Subtypes And Regulation Of Cardiac Progenitor Cell Division**

Abbigayle J. Gamble and Shaad M. Ahmad
Presenting Author: Abbigayle J. Gamble
Indiana State

Mutations in the zinc-finger transcription factor-encoding gene *CASZ1* lead to aberrant heart development in humans, Xenopus, and mice, indicating its conserved role in cardiogenesis. Our phenotypic analysis of a null mutation of *castor* (*cas*), the *Drosophila* ortholog of *CASZ1*, shows that *cas* has two distinct roles in heart development. First, *cas* is required for mediating all three categories of cardiac progenitor cell division: asymmetric, symmetric, and cell divisions at an earlier developmental stage. Second, *cas* prevents subsets of cells in the most anterior region of the heart, the anterior aorta, from becoming specified as *seven up*-expressing cardial cells (Svp-CCs). Svp-CCs are present in the posterior aorta and the even more posterior heart proper, regions of the heart determined by the expression of the Hox genes *Ultrabithorax* (*Ubx*) and *abdominal A* (*abd-A*). Intriguingly, both *Ubx* and *abd-A* repress *cas*, and ectopic expression of either of these two Hox genes in the anterior aorta leads to the ectopic specification of Svp-CCs there—a result which phenocopies *cas* loss-of-function mutants. Collectively, these data raise the possibility that that *Ubx* and *abd-A* specify Svp-CCs in the posterior aorta and the heart proper by repressing *cas* in those regions. In contrast, in the anterior aorta, in the absence of both *Ubx* and *abd-A*, *cas* levels may be sufficiently high to repress the Svp-CC fate. We are

presently testing this hypothesis for *cas*-mediated Svp-CC specification and attempting to elucidate the pathways through which *cas* regulates cardiac progenitor cell division.

[3] **The Impact of Repeated Alcohol Exposures on Adult Drosophila Stat92E Signaling**
Alexandria Wilson and Emily Petruccelli
Presenting Author: Alexandria Wilson
Southern Illinois University Edwardsville

Alcohol Use Disorder (AUD) is associated with repeated alcohol exposures and addiction behaviors. Annually, about 95,000 deaths in the U.S. are due to alcohol consumption (CDC, 2021). Despite the prevalence of AUD, we lack novel therapeutic approaches for treating the cellular mechanisms of the disease. Recently the conserved Janus kinase (JAK)/Signal transducer and activator of transcription (STAT) signaling pathway has been implicated in both mammalian and *Drosophila* models of AUD (Chen et al., 2021; Petruccelli et al., 2020). JAK/STAT signaling is involved in early development and innate immune signaling, but it is unclear how STAT activity influences specific ethanol-induced behaviors. To address these gaps in our knowledge, this work will capitalize on the single *Drosophila* STAT homolog, *Stat92E*, and the genetic tools available in flies. Recent findings in our lab have shown that ubiquitous expression of *Stat92E-RNAi* causes an increase in ethanol-induced locomotion, which is reminiscent of behavioral sensitization, a distinctive feature of addiction in mammals (Wilson et al., in revision 2022). To follow up on this work, we used the GAL4/UAS expression system to knockdown or overexpress *Stat92E* in glia to provide insight as to how Stat92E functions in ethanol-induced locomotion. Second, we performed DNA or RNA extractions in STAT reporter flies to investigate whether repeated ethanol exposures influence the preferential isoform usage of *Stat92E* transcripts and downstream transcriptional activity. Neither knockdown, nor overexpression, of *Stat92E* in glia caused a drastic change in ethanol-induced locomotion. This work provides better resolution of STAT signaling in the context of repeated ethanol exposure, informing the development of molecular strategies to manage AUD.

[4] **Heterochromatinization of repetitive DNA is location dependent**
Alix Hathaway and Andrew M Arsham
Presenting Author: Alix Hathaway
Bemidji State University

Heterochromatin, though gene-poor, is nonetheless essential for cellular function and organismal survival and plays important roles in gene expression, genomic stability, and defense against invasive DNA. How a cell's epigenetic machinery recognizes novel threats, such as repetitive DNA, remains unclear. Here we show that the recognition and silencing of an exogenous tandem array of repetitive DNA is highly location-dependent. Using transposition mutagenesis, we inserted a reporter construct expressing the white gene adjacent to a 256-copy tandem array of the 36 nucleotide *E. coli* lac operator. Approximately 1% of the recovered flies expressed variegation, indicating that the repeat array was not itself sufficient to trigger silencing. Many of the variegating insertions are located in gene-rich euchromatic regions, often in promoters or 5' UTRs, showing that under the right conditions the lacO array can trigger the formation of ectopic heterochromatin even in actively transcribed euchromatin. A clearer understanding of what features distinguish euchromatin that is sensitive to repeat-induced silencing from that which is not will provide insight into genomes' innate pathogen-sensing mechanisms as well as the genetic mechanisms of trinucleotide repeat expansion diseases.

[5] **CRL4 E3 Ligase Mahjong/DCAF1 Knockdown Induces Minute-like Cell Competition, Unrelated To Apicobasal Cell Polarity**

Amit Kumar and Nicholas E. Baker
Presenting Author: Amit Kumar
Albert Einstein College of Medicine

Cell competition involves elimination of viable cells when present next to more fit cells. Mahjong (Mahj), a ubiquitin E3 ligase substrate receptor, binds to lethal giant larvae (*lgl*), a neoplastic tumor suppressor that regulates apicobasal polarity of the cell, and loss of either leads to cell competition. Here we show that bZip-domain transcription factor Xrp1, which eliminates *Minute* cells (heterozygous for Ribosomal protein gene mutations), is required for the competition of *mahj* mutant cells, not for *lgl* mediated competition. Xrp1 expression in *mahj* mutant cells reduces translation and growth and activates cell competition-associated signaling pathways. Knockdown of other CRL4 E ligase subunits (DDB1 and Cul4) or proteasome subunits also has *Minute*-like effects. Our findings uncouple *mahj*-mediated cell competition from apical-basal polarity and show that Xrp1 is activated by defects in protein turnover.

[6] **Warm And Cold Temperatures Have Distinct Effects During Drosophila Oogenesis And Spermatogenesis**

Ana Caroline P. Gandara and Daniela Drummond-Barbosa
Presenting Author: Ana Caroline P. Gandara
University of Wisconsin

Investigating how temperature impacts reproduction is crucial in light of the current climate crisis. *Drosophila* reproduction is highly sensitive to environmental factors; however, it remains largely unknown how adult exposure to chronic thermal stress affects gametogenesis. We incubated newly-eclosed *y w* adult flies (raised at 23°C) at 18°C (cold), 25°C (optimal), or 29°C (warm) for 20 days and found that egg production was reduced at both suboptimal temperatures through distinct cellular mechanisms. Chronic exposure of females to 18°C improved germline stem cell (GSC) maintenance, germline survival, and oocyte quality; however, follicle growth rates were reduced, slowing down egg production. By contrast, in females at 29°C, GSCs numbers and follicle growth were similar to 25°C, while death of early germline cysts and vitellogenic follicles was markedly increased, leading to fewer mature oocytes of lower quality. In adult males, exposure to 29°C dramatically decreased sperm motility and abundance within seminal vesicles after just 5 days, leading to near complete sterility after 10 days at 29°C; by contrast, male fertility remained high after 20 days at 18°C. As in females, male GSCs maintenance was improved at cold temperature. Surprisingly, cold and warm temperatures similarly disrupted spermatid elongation/individualization and decreased sperm transfer to females. These results suggest that the decrease in male fertility at 29°C is the combined result of impaired sperm entry into seminal vesicles at the end of spermatogenesis coupled to reduced sperm motility.

[7] **Elucidating The Roles of Zelda and Taranis During Late Regeneration in Drosophila Wing Imaginal Discs.**

Anish Bose and Rachel Smith-Bolton
Presenting Author: Anish Bose
University of Illinois Urbana-Champaign

Regeneration is a complex process that enables damaged tissues to be replenished and restored back to their correct morphology and function. As organs and tissues are prone to damage either by external

causes or intrinsic chronic illness, the identification of factors and mechanisms important for regeneration may have clinical significance. Our laboratory has described a regeneration-specific pathway, where the gene *taranis* was found to be important for maintaining proper cell fate identity during late regeneration in *Drosophila* third instar larval wing imaginal discs. Lowered Taranis ($tara^{1}$/+) levels causes mis-regulation of the posterior selector gene *engrailed*, as $tara^{1}$/+ mutants allow damage-induced high JNK signaling to overexpress *engrailed* expression. Overexpressed *engrailed* allows its downstream target *polyhomeotic* to initiate a negative feedback loop by which the *engrailed* locus becomes silenced in the regenerating wing pouch. Silenced *engrailed* results in impaired regeneration, characterized by anterior features being present in the posterior compartment of the poorly regenerated adult wing, collectively termed as posterior-to-anterior cell fate transformations. Taranis prevents these posterior-to-anterior cell fate changes from occurring by preventing *engrailed* from getting overexpressed. However, it is unclear which signals activate *taranis* during late regeneration, or how *engrailed* is regulated during late regeneration. Preliminary data shows that the pioneer transcription factor *zelda* is upregulated during late regeneration and under a reduced *zelda* background, *taranis* levels are also lowered, suggesting Zelda is likely an activator of *taranis*. We are also looking into the role of *zelda* during regeneration beyond activating *taranis*.

[8] **Fly Fam161 is a Centriole Protein Essential for Coordinated Behavior and Male Reproduction**
Ankit Jaiswal, Andrew Boring, and Tomer Avidor Reiss
Presenting Author: Ankit Jaiswal
University of Toledo

Fam161 is an ancient family of evolutionarily conserved proteins represented in humans with two paralogs (FAM161A and FAM161B) and one ortholog in flies (Fam161). FAM161A is a centriole lumen protein found in photoreceptors connecting cilium and is essential for maintaining human vision (Langmann et al., 2010). FAM161A is a component of the mammalian sperm typical and atypical centrioles rods however its role in reproduction is unknown (Khanal et al., 2021). We use the fruit-fly as a model to study the role of Fam161.

We hypothesized that fruit-fly Fam161 functions in centriole and connecting cilium in a cell-specific manner. Here, we study Fam161 using transgenic Fam161GFP fly and a hypomorphic mutant lacking Fam161 C-terminus *(fam161ΔC)* and found it is a centriole and connecting cilium protein essential for normal coordination behavior and reproduction. In sensory neurons, Fam161 is in the centrioles and connecting cilium in a neuron-specific type. Accordingly, Fam161*ΔC* has abnormal geotaxis, suggesting a reduced coordinated behavior. Fam161 is found in sperm giant centriole from spermatocyte to seminal spermatozoa but is absent from sperm stored in the female reproductive tract, exhibiting a novel post-ejaculation regulation. While *fam161ΔC* mutant males and females are fertile and 80% of fertilized eggs hatched larva, only 60% of the males mate with control females within two hours, suggesting that Fam161 is essential for efficient mating due to its role in fly coordination.
In the future, we want to generate a full knockout of the Fam161 gene and study its role if any in sperm competitiveness.

[9] **The Interplay Of Peroxisome And Mitochondrial Dynamics During Aging In Drosophila**
Ankur Kumar and Hua Bai
Presenting Author: Ankur Kumar
Iowa State

Damaged mitochondria are repaired and recycled through the mechanisms of mitochondrial dynamics in response to stress; this helps in restoring cellular homeostasis. Mitochondrial

dynamics have emerged as a novel regulator of aging in recent years. During aging, alterations in mitochondrial morphology and structure have been observed. Researchers have performed genetic manipulations of genes involved in the fission and fusion of mitochondria, which extended the lifespan. However, the causes of the age-dependent alteration in mitochondrial dynamics remain unanswered. Our focus is to explore the involvement of the peroxisome in maintaining mitochondrial homeostasis during animal aging. Recent studies in our lab have shown mitochondrial morphology and function alteration due to impaired peroxisomal protein import in aging oenocytes (hepatocytes) of fruit flies. We found an increase in mitochondrial size in oenocytes during fly aging. Similarly, we have found that the knockdown of Pex5 alters mitochondrial morphology. Interestingly, knocking down the genes involved in peroxisomal plasmalogen synthesis resulted in enlarged mitochondria. Our future goal is to understand how peroxisomes contribute to age-related alterations of mitochondrial dynamics and functions.

[10] **Synaptic dysfunction and neurodegeneration in a Drosophila lipid storage disease model of Niemann Pick Type C**

Anna Eberwein and Kendal Broadie
Presenting Author: Anna Eberwein
Vanderbilt University

The lipid storage disease (LSD) Niemann Pick Type C (NPC) results in progressive childhood neurodegeneration owing to loss of the NPC1 (95%) and NPC2 (5%) genes. Lipids accumulated in NPC include cholesterol, sphingosine and glycosphingolipids (GSL). We propose two linked hypotheses for the NPC disease condition; 1) GSL pathway mistrafficking of mannosyl glucosylceramide (MacCer) causes functional neurotransmission defects, and 2) this synaptic dysfunction underlies neurodegeneration. We are testing these hypotheses using an established *Drosophila* NPC disease model, with experiments focused at the well-characterized glutamatergic neuromuscular junction (NMJ) model synapse. We have found that null *npc1a* mutants exhibit increased synaptic transmission, which is phenocopied in *brainiac* (*brn)* mutants, the GSL synthesis beta-1,3-N-acetylglucosaminyltransferase that causes MacCer accumulation. Importantly, double *npc1a*; *brn* null mutants have no additional increase in synaptic transmission, indicating that the *npc1a* and *brn* gene products operate in the same functional pathway. To test synaptic dysfunction causal roles in NPC neurodegeneration, terminal deoxynucleotidyl transferase biotin-dUTP nick end labeling (TUNEL) has been used to measure cell death. We have found that *npc1a* null brains have elevated TUNEL labeling indicative of progressive neurodegeneration. We have been testing *brn* null mutants *and npc1a; brn* double nulls with TUNEL labeling, as well as rescue by application of a glucosylceramide synthase inhibitor, to determine causal roles of MacCer accumulation related to neurodegeneration progression. Taken together, these studies test MacCer functions as a key pathogenic lipid in NPC synaptic dysfunction causally linked to progressive neurodegeneration.

[11] **Yorkie Dependent Transcriptional Network Promotes Tumor Growth.**

Arushi Rai and Madhuri Kango-Singh
Presenting Author: Arushi Rai
University of Dayton

The Hippo pathway effector, Yorkie (Yki) is a key mediator of signaling interactions and transcriptional addictions in tumors cells and presents an attractive opportunity to study transcriptional dependencies in cancer cells. The RasV12 scrib-/- tumor mosaic model, a well- established model, shows activation of oncogenic Ras in the background of impaired apical-basal polarity. Previously, we have shown that in RasV12, scrib-/- cells Wingless (Wg) acts upstream of Caspases, JNK and Yki and

via its canonical and non-canonical pathways to interact with Yki to regulate the development and cancer growth. Our goal is to further understand how the two evolutionarily conserved signaling pathways i.e., Hippo and Wingless crosstalk and interact with each other to regulate tumor growth using the RasV12, scrib-/- tumor model in Drosophila imaginal discs. Our data showed that the wg transcriptional reporters and wg transcript levels both are upregulated in RasV12, scrib-/- cells. In other contexts, wg is shown as a transcriptional target of Yki. Therefore, we will test for (a) the effects of Yorkie protein, the main effector molecule of Hippo pathway, on the transcription and expression of Wg and other Wg pathway components by reporter assays, and qRT-PCR- based approaches, and (b) the effect of wg pathway components (frizzled, TCF) on the intrinsic wingless signaling and also growth of RasV12, scrib-/- tumor (c)the effect of the effect of feedback interactions that promote tumorigenesis using genetic epistasis-, and immunohistochemistry-based approaches. Here, we present our progress on the organization of the molecular network involving Wingless and Yorkie.

[12] **Tracking a Fruit Fly in a Tunnel: Oxidative Stress and Potassium Channel Defects Cause Two Types of Circadian Restless Behaviors**

Atsushi Ueda and Chun-Fang Wu
Presenting Author: Atsushi Ueda
University of Iowa

The involvement of reactive oxygen species (ROS) signaling has been implicated in the circadian activity patterns of sleep-wake cycles (Kempf et al., 2019). We discovered that *Drosophila Superoxide dismutase 1 (Sod1)* mutants display fragmentated sleep (rest) state. Using the TriKinetics multibeam counter, which can monitor locomotor activity of a single fly traversing along a narrow glass tube (65 mm length, 5 mm diameter, divided into 17 adjacent zones), we show that quiescence period in *Sod1* was fragmented (< 5 min, mostly) and most activity was confined to within local movements in a single zone. In contrast, WT spend long stretch of time (up to 2 hrs) in quiescent "sleep" state during midday and midnight with no activity detected in any of the 17 zones. When active, walking back and forth along a tube length during light on and off switching time (dawn and dusk). Mutant flies of the K^+ channel gene, *Shaker* (*Sh*), which is known to exhibit reduced sleep (Cirelli et al., 2005). We found that compared to *Sod1*, *Sh* flies were often engaged in long-distance locomotion, marching along the entire tube for prolonged time periods (up to 4 hrs, especially during night time), with only brief rest periods (< 5min). With this new multibeam device, we are able to distinguish different types of locomotion and local activities and to ensure the periods of the rest state, with no activities detected along the entire tube. Future genetic and circuit analysis may reveal the molecular and cellular bases of the distinct patterns of activity and sleep defects.

[13] **The Genetic Architecture of Morphological Scaling**

Austin Wilcox and Alexander Shingleton
Presenting Author: Austin Wilcox
University of Illinois Urbana-Champaign

Morphological scaling relationships between the sizes of individual traits and the body captures the characteristic shape of a species, and the evolution of scaling is the primary mechanism of morphological diversification. However, we have almost no knowledge of the genetic architecture of scaling, critical if we are to understand how scaling evolves. Here we begin to explore the genetic architecture of *population-level* morphological scaling relationships – the scaling relationship fit to multiple genetically-distinct individuals in a population – by describing the distribution of *individual* scaling relationships – genotype-specific scaling relationships that are unseen or cryptic.

These individual scaling relationships harbor the genetic variation that determines relative trait growth within individuals, and theoretical studies suggest that their distribution dictates how the population scaling relationship will respond to selection. Using variation in nutrition to generate size variation within 197 isogenic lineages of *Drosophila melanogaster*, we reveal extensive variation in the slopes of the wing-body and leg-body scaling relationships among individual genotypes. These data allow us to predict how different selection regimes alter scaling and morphology in Drosophila and is the first step in identifying the genetic targets of such selection.

[14] **Interactive Regulation of Growth by Dorsal Proventriculus and yorkie in the Drosophila Eye**
Basavanahalli Nanjundaiah Rohith and Madhuri Kango-Singh
Presenting Author: Basavanahalli Nanjundaiah Rohith
University of Dayton
The developing eye of *Drosophila* is a well-established model for studying developmental genetic processes and growth regulation. The developmental genetic networks discovered in *Drosophila* are highly conserved in all animals including higher mammals. Our long-term goal is to understand the molecular basis of Dorsal-Ventral patterning and growth in the eye by interactions of the dorsal selector genes and growth regulatory genes. We recently identified *dorsal proventriculus* (*dve*) as a candidate for dorsal-ventral eye patterning. Preliminary data from our lab also suggests that Dve may interact with the Hippo growth regulatory pathway to control patterning and growth of the eye. We tested the interactions between Hippo pathway and dorsal-ventral patterning using the GAL4-UAS system. We tested if these pathways act independently to control eye patterning and growth or act via shared targets and regulatory interactions. In this context we investigated the effect of overexpressing Yorkie (the effector of the Hippo pathway) and Dve during larval development specifically in the dorsoventral domains of the eye imaginal discs. Over expression of Yki extends the Dve domain in the eye disc as a result of which the disc growth is enlarged, and suppresses eye differentiation, thereby suggesting that these two distinct genes may regulate a common downstream target to control the disc growth and differentiation. We have tested *wingless* (wg) a known and conserved Hippo downstream target for Dve and Yki mediated effects using reporter assays, and qRT-PCR- based approaches, and our results will be discussed.

[15] **Mutations in Lamin and How it Causes Multiple Tissue-Specific Disorders**
Bismark Acquah and Alysia Vrailas-Mortimer
Presenting Author: Bismark Acquah
Illinois State

Lamins, the major components of the nuclear lamina where they provide a platform for the binding of proteins to the chromatin and confer mechanical stability (Dittmer and Misteli.,2011). Mutations in the human LMNA gene result in at least 15 distinct disorders ranging from muscular dystrophies to neurological disorders to lipodystrophies **(**Vytopil *et al.*,2003**).** Interestingly, some mutant forms of lamin protein aggregate, which may be toxic to the cells. However, it is unknown how specific mutations in lamin give rise to tissue specific disease. We hypothesize that certain tissues are susceptible to specific lamin mutations due to the inability of tissue specific quality control mechanisms to degrade those mutant forms, leading to protein aggregation and cellular toxicity. I will be testing if tissue specific disease mutations in Lam Dm0, one of the fly homologues of LMNA, cause the protein to aggregate in muscles and/or neurons. We find out that the main forms and the farnesylated forms of the different Lam Dm0 mutant proteins have different expression patterns in the muscles of the flies. In addition, we find that the p38 MAPK (p38Kb) interacts with the CASA complex to regulate the degradation of Lam Dm0, the other fly homologue of LMNA. Future experiments will characterize how these mutant forms of Dm0 affect the functionality of the muscles and neurons in flies and if these forms can be targeted for degradation by p38Kb and the CASA complex.

[16] **Regulation of the Cell-Fate Gene engrailed During Late Regeneration in Drosophila Wing Imaginal Discs.**

Brian Park and Rachel Smith-Bolton
Presenting Author: Brian Park
University of Illinois Urbana-Champaign

Regeneration is the process of restoration of damaged tissues back to their correct structures. The regenerative process requires damage-responsive signaling pathways and factors to promote the recovery of the tissue structure and function. One such mechanism is c-Jun N- Terminal kinase (JNK) signaling, which plays a key role in stress response and regeneration. Our laboratory showed that JNK signaling can disrupt cell fate specification during late regeneration in third instar wing imaginal discs. Specifically, the posterior selector gene engrailed is susceptible to being mis-regulated by JNK signaling. High JNK signaling activity during late regeneration overexpresses engrailed. Overexpressed engrailed then activates its downstream target polyhomeotic, which in turn initiates a negative feedback loop and results in silencing. This mis-regulation of engrailed causes cell fate deformities in the regenerated adult wing, such as posterior-to-anterior cell fate transformations. However, we do not know which enhancer modules within the regulatory region of engrailed are recognized by JNK signaling targets to mis-regulate posterior cell fate during regeneration. Our objective is to identify these regulatory regions using LacZ-reporter lines that contain fragments of the 62kb engrailed regulatory region. We are also investigating the potential targets of JNK signaling that recognize the engrailed regulatory elements that ultimately cause mis-regulation of engrailed. This work will give us novel insights into how JNK signaling regulates engrailed expression during late regeneration to disrupt proper cell fate identity.

[17] **The Creation of a Null Allele of Clipper and Investigation of Its Role in Drosophila melanogaster Oogenesis**

Charlie T. Watts and Julie A. Merkle
Presenting Author: Charlie T. Watts
University of Evansville

The process by which the oocyte identity is determined in the Drosophila ovary is not well understood. A mutagenesis screen was performed on 2L to better understand the molecular mechanisms that control this process, and an allele of Clipper (Clp) was identified. Clp germline clones exhibit a phenotype consisting of 16 nurse cells, no oocyte, and a clustering of ring canals within the mutant egg chamber. Clp encodes a protein component of the Cleavage and Polyadenylation Specificity Factor (CPSF) complex that recognizes the AAUAAA signal sequence at the 3’ end of primary transcripts and recruits poly(A) polymerase and other cleavage and polyadenylation factors. Polyadenylation is important for mRNA stability, export from the nucleus, and recognition by the ribosome. To investigate how a genetic null of Clp affects oogenesis, CRISPR/Cas gene editing was used to generate a knock-out allele of Clp. A gRNA plasmid was generated using Gibson Assembly to assemble two gRNAs into the pCDF5-w backbone, while a homology domain plasmid was created by ligating homology arms that flank the Clp genomic region into pHD-EGFP-attP. These plasmids were then injected into a germline-expressing Cas9 line. Next, transformants will be screened for GFP, indicating potential CRISPR-generated alleles. These lines will then be validated and characterized. The future goal of this project is to better understand the role of Clp in oogenesis, thereby providing insight into the molecular mechanisms of oocyte identity.

[18] **Characterization Of The Role Of Notch In Drosophila Testes**

Christine Severude, Adriana Soriano, Heather Wheeler, Jennifer Jemc Miersich
Presenting Author: Christine Severude
Loyola University

The process of gonad development and gametogenesis is crucial for the propagation of our species and the conservation of biodiversity. The gene *Notch* plays an important role in these processes and is widely conserved across animal phylogeny. In humans and mice, lack of *Notch* has been associated with spermatogenesis arrest, suggesting that the differentiation and survival of male germ cells is dependent on the *Notch* signaling pathway. However, how Notch functions in this process is not well understood. Previous studies in *Drosophila* have shown that a loss of Notch signaling in the embryo results in defects in hub establishment, indicating that Notch functions in gonad development. Recent work in our lab demonstrates that increased Notch signaling in somatic cells of the testes also negatively impacts spermatogenesis, resulting in spermatogenesis arrest and infertility. Despite these important roles, no transcriptional targets of *Notch* have been identified in the testes. The goal of this experiment is to determine the transcriptional targets of Notch in the testes through RNA-sequence analysis, bioinformatics, immunohistochemistry, and in situ hybridization. These target genes will help us understand the process and regulation of spermatogenesis in *Drosophila* and across animal phyla. It will also pave the way for future research endeavors exploring the function of identified target genes. These genes could represent therapeutic targets for individuals dealing with infertility.

[19] **Dietary Restriction Fails to Extend Lifespan of Drosophila Model of Werner Syndrome**
Eileen Sember, Ranga Chennakesavula, Breanna Beard, Mubaraq Opoola, and Dae-Sung Hwangbo
Presenting Author: Eileen Sember
University of Louisville

Werner syndrome (WS) is an autosomal recessive disorder that results in premature aging and occurs in 1 in 1,000,0000 to 1 in 10,000,000 people. In humans, WS is a result of mutations that render the WRN gene, that contains a helicase and an exonuclease domain, non-functional. Currently there is no cure for WS in humans making dietary and lifestyle interventions attractive for increasing the quality and longevity of lives. Diet restriction (DR) has been shown to extend the lifespan of several model organisms including *Drosophila melanogaster*, making it a strong candidate for WS treatment. Using the null mutant flies for the gene *WRNexo*, homologous to the exonuclease domain of WRN in humans, we examined the effects of DR on lifespan and DR mediated physiological, and behavioral characteristics of starvation, oxidative stress, and sleep/activity. Surprisingly, it was found that DR had a deleterious effect on lifespan in WRN mutants. DR was also observed to have a sex-dependent reduced resistance to both starvation and oxidative stress while creating more sleep/activity disruption. Deleterious effects of DR on lifespan, physiological, and behavioral characteristics, suggest that the WRN protein is necessary for longevity benefits traditionally observed in DR as well as the WRN protein having a function in metabolic control.

[20] **Altered Sexual Size Dimorphism in Drosophila Via Artificial Selection**
Elizabeth Agolli and Alexander W. Shingleton
Presenting Author: Elizabeth Agolli
University of Illinois Chicago

The difference between female and male body size (sexual size dimorphism, SSD), is perhaps the most obvious way in which the sexes differ. However, while SSD can vary dramatically between closely related species, the genetic basis for variation in SSD is very poorly understood. Problematically, SSD is a characteristic of a population rather than an individual, which makes elucidating the genetics of SSD

challenging. Here we use artificial selection over 15 generations to increase and decrease SSD in an outbred population of flies, as a prelude to understand the genetic basis for changes in SSD. Using family selection, we show that selection on the difference in female and male body size among siblings leads to a rapid change in SSD. This change in SSD is primarily a consequence of changes in male body size rather than female body size. The derived lineages provide a resource for the genetic and developmental analysis of SSD and its evolution.

[21] **Mitophagy Defects In Adult Fly Neurons Lacking Vps13D Cause Age-Dependent Neurodegeneration**

Emily Rozich and Ryan Insolera
Presenting Author: Emily Rozich
Wayne State University

Mutations in the human gene VPS13D cause the adult-onset neurological disorder ataxia. Our previous work has shown that disruptions in the Vps13D gene in Drosophila neurons causes mitochondrial defects, characterized by enlarged mitochondrial morphology and impaired degradation of mitochondria through mitophagy. However, developmental lethality caused by neuronal Vps13D loss limited our understanding of the age-dependent physiological effects of Vps13D perturbation. Here, we have developed a system to temporally knockdown genes via RNAi in adult Drosophila using a modified Gal4/UAS system. Temporal control of the Gal4 repressor, Gal80, is achieved with a pharmacologically controlled destabilization-domain fused to Gal80 (Gal80-DD), expressed under the control of a pan-neuronal promoter. The method we developed allows for minimal leaky expression of UAS-driven transgenes during development, and yields adult-onset expression of Gal4 to manipulate gene expression and examine age-dependent neurological phenotypes. Optimization of the Gal80-DD tool showed that feeding animals the DD-stabilizing drug trimethoprim (TMP) during developmental stages and rearing at a reduced temperature maximally repressed Gal4 activity. Temperature shift and removal of TMP from the food activates Gal4 expression in adults. With this method, expression of UAS-driven Vps13D RNAi in adult neurons causes age-dependent accumulation of mitophagy intermediates, loss of locomotor activity, early lethality, and brain vacuolization characteristic of neurodegeneration. This system allows us to examine mechanisms of suppressing the phenotypes associated with Vps13D disruption as potential therapeutic avenues for afflicted ataxia patients. More broadly, this model will lead to understanding of the consequences of disrupted neuronal mitophagy, a common pathology associated with neurodegenerative diseases.

[22] **Emery-Dreifuss Muscular Dystrophy: The Relationship Between Two Nuclear Envelope Proteins**

Emily Symons and Lori Wallrath
Presenting Author: Emily Symons
University of Iowa

Emery-Driefuss muscular dystrophy (EDMD) is a rare muscle disease caused by several genes that encode nuclear envelope proteins, including *LMNA* and *TMEM43*. The *LMNA* gene encodes A-type lamins, intermediate filaments that form a meshwork lining the inside of the nuclear membrane. They provide structural support for the nucleus and organize the genome through contacts made with chromatin. The goal of our research was to determine if mutant lamins alter the localization of TMEM43, a known lamin binding partner. To accomplish this goal, we utilized Drosophila which has orthologues of *LMNA* and *TMEM43* called *Lamin C* and *Tmem43*, respectively. The Gal4/UAS system was used to express mutant lamins corresponding to those that cause disease in larval body wall muscle*s.* The muscles were stained with antibodies to Lamin C and TMEM43. We found that mutant

lamins cause altered nuclear shape and that mislocalization of dTMEM43 varies depending on the mutant lamin. There is a lack of correlation between the Lamin C domain affected and the mislocalization of dTMEM43. Collectively, we show that mutant lamins cause dominant nuclear shape changes, suggesting that they form an abnormal nuclear lamina, and in some cases, this alters the nuclear envelope localization of dTMEM43.

[23] **Identification Of MiRNA In Growth And Survival**
Esther Labya and Amit Singh
Presenting Author: Esther Labya
University of Dayton

Cell size plays an important role in cellular processes and function of a cell. Hence cell size and growth need to be maintained properly. Gene regulation plays an important role in regulating gene expression of different cellular processes like apoptosis, cell growth, etc. MicroRNAs (miRNAs), small single-stranded RNAs, regulate gene expression post-transcriptionally by binding to the 3'untranslated region of their target messenger RNAs (mRNAs), degrade their target and hence silence their gene expression. In a forward genetic screen, our lab has identified a micro-RNA that inhibits apoptosis in *Drosophila* eye model. Previous study shows that in the *hippo* (*hpo*) mutants, exhibit overgrowth whereas hippo gain-of-function triggers cell death in the developing eye. We studied the role of this microRNA in hippo loss-of-function and gain-of-function background. Our preliminary data suggests that gain-of-function of miRNA in the GMR>*hpo* background rescues Hpo-mediated cell death. Our working model suggests that miRNA regulates the cell growth by modulating hippo pathway.

[24] **The Role of Myc in Drosophila Wing Imaginal Disc Regeneration**
Felicity Ting-Yu Hsu and Rachel Smith-Bolton
Presenting Author: Felicity Ting-Yu Hsu
University of Illinois Urbana-Champaign

Regeneration is a complicated process through which some animals restore missing tissue upon damage. We study regeneration using a genetic ablation system in *Drosophila* wing imaginal discs. One of the genes that are important in regeneration is *Myc*, whose expression levels and transcription levels are upregulated in the regeneration blastema of wing imaginal discs. Overexpression of Myc improves wing imaginal disc regeneration, while reduction of Myc expression worsens wing disc regeneration. During normal development, one primary role of Myc is ribosomal biogenesis. However, the exact function of Myc in regeneration is unknown. Our recent findings suggest that the nucleoli sizes, indicating ribosomal biogenesis activity, in the regenerating wing pouch are similar to those in the undamaged pouch. This finding suggests that one possible role of Myc might be maintaining ribosomal biogenesis activity while the activity is reduced in other parts of the damaged disc. Thus, we aim to identify the exact role of upregulated Myc in wing disc regeneration. In addition to ribosomal biogenesis, Myc is also involved in cell competition. After tissue damage, Myc upregulation in the regeneration blastema causes increased difference in Myc expression levels between the regeneration blastema and the rest of the disc. During normal development, Myc expression level differences in the wing imaginal disc lead to cell competition, where cells with lower Myc are eliminated. Therefore, we aim to study whether cell competition takes place in regenerating wing discs and how it impacts tissue regeneration.

[25] **Oxygen regulates ecdysone levels in Drosophila but not through transcription**
George P. Kapali and Alexander W. Shingleton
Presenting Author: George P. Kapali
University of Illinois Chicago

In almost all animals, physiologically low oxygen (hypoxia) during development slows growth and reduces adult body size. There are systemic endocrine mechanisms that determine growth under hypoxic conditions; however, they are poorly understood. In *Drosophila* low oxygen elevates levels of circulating ecdysone, which slows growth and reduces final body size by suppressing the activity of the insulin/IGF-signaling pathway. How hypoxia regulates ecdysone levels is unknown. Ecdysone levels during development are, in part, regulated by the transcription of genes involved in ecdysone synthesis and degradation. A compelling hypothesis, therefore, is that oxygen affects ecdysone levels using similar mechanisms. To test this hypothesis we used q-PCR to assay the expression of genes involved in ecdysone synthesis, export, import and metabolism under normoxic and hypoxic conditions. Contrary to expectation, the transcriptional response to hypoxia does not include upregulation of ecdysone synthesis genes or downregulation of ecdysone metabolic genes. We do, however, observe an elevation in the expression of the ecdysone exporter gene *Atet* under hypoxia. This suggests that the elevation of circulating ecdysone levels under hypoxia is not achieved through an increase in ecdysone synthesis, but rather through an increase in ecdysone export.

[26] **Peanut Skin Extract Ameliorates Lead (Pb) Acetate Induced neurotoxicity and Improves Fertility in Drosophila.**

Hassan Sani and Moses Zaruwa Zira
Presenting Author: Hassan Sani
Federal University of Lafia, Nigeria

Peanut skins are nutritious by-products, containing high levels of antioxidant, resveratrol, polyphenols, flavonoids and fatty acids. This study aimed to evaluate the protective role of Peanut skin extract against Lead acetate induced neurodegeneration and its effect on fertility in *Drosophila melanogaster.* Phytochemical screening revealed the presence of phenols, flavonoids, alkaloids, saponins, steroids, terpenoids, anthraquinones and cardiac glycosides. Total phenol 156.15mg/g and total flavonoid 124.83mg/g while GC-MS analysis revealed the presence of Methyl palmitate; Ethyl palmitate; Methyl linoleate; Ethyl linoleate; Methyl oleate; Methyl heptadecanoate; 10-Octadecenoic acid, methyl ester and Octadecanoic acid, 17-methyl-, methyl ester. The extract exhibited scavenging activities against DPPH and H_2O_2 radicals. Neuronal toxicity was ameliorated through elevation in the concentration of GSH, increased SOD and GPx activities and corresponding decrease in NO and MDA production. Also the extract improved Fecundity, egg hatching rate, weight and Survival of Drosophila. Our findings revealed that peanut skin extract ameliorated lead acetate induced neurotoxicity and improve fertility in drosophila. Thus, peanut skin could be a source of therapeutic agent for fertility, and disorders related to lead toxicity.

[27] **Circadian Rhythms And The Gut Microbiome In The Fruit Fly, Drosophila Melanogaster**

Isaiah Fitzmaurice and Alder Yu
Presenting Author: Isaiah Fitzmaurice
University of Wisconsin – La Crosse

Circadian rhythms (CRs) are 24-hour oscillations of physiological rhythms that regulate behavior, physiology and metabolism in plants, animals, fungi and cyanobacteria. CRs originate within an organism and are established via regular exposure to recurring stimuli. All organisms with an intestinal tract harbor a community of bacteria called the commensal gut microbiome. The composition of the gut microbiome is largely influenced by the host CR and has been shown to cycle in mammalian models. Also, the microbiome has been shown to impact the expression of host circadian clock genes via

signaling molecules such short chain fatty acids in mammalian models. However, the cycling of the gut microbiome as well as its impact on the host CR has not been documented in non-mammalian models. The fruit fly is a good model to address this gap in knowledge, and a preliminary fly study has shown that the composition of the gut bacteria shows time-of-day variation. This study will document the cycling of the gut microbiome of flies and examine the functional consequences on the host circadian rhythm in flies without a gut microbiome. To study the cycling of gut taxa, the guts of flies with a functional and genetically ablated circadian clock will be wholly dissected out at different times of day and sequenced to examine changes in taxa, which will then be analyzed for rhythmicity. To examine functional consequences on the host CR, flies with and without a gut microbiome will be examined for activity and food-seeking behavior. Addressing this gap in knowledge will potentially update the model of circadian rhythms to include the role of the gut microbiome in all organisms with an intestinal tract.

[28] **The Role of crossveinless-c (cv-c) in Caudal Visceral Mesoderm (CVM) Migration**
Jayden Ogbodo and Afshan Ismat
Presenting Author: Jayden Ogbodo
University of St. Thomas, Minnesota

Regulation of actin polymerization occurs mainly through Rho family GTPases, of which there are three main members: Rho, Rac, and Cdc42. GTPases are regulated themselves by two types of proteins: GEFs (Guanine Exchange Factors) that convert a GTPases from an inactive to active state, or GAPs (GTPases-Activating Proteins) that inactivate GTPases. The focus of this project is on the gene *crossveinless-c* (*cv-c*), which encodes a RhoGAP protein, a GAP specific for the GTPase Rho. A previous study showed that *cv-c* mRNA was expressed in several migratory cell types in the embryo, including the caudal visceral mesoderm (CVM). The absence of *cv-c* resulted in delayed and less orderly dorsal closure by leading edge cells and an almost complete lack of migration of Malpighian tubules. In both cases, the defect in cell migration was the result of problems in actin polymerization and organization, a role that clearly fits with the function of a RhoGAP. The CVM cells migrate as a loose collective along the entire length of the embryo. Using *croc-lacZ*, a CVM cell-specific reporter, we show that, in the absence of *cv-c*, CVM cells mis-migrate. However, cellular protrusions are hard to see with *croc-lacZ*. Therefore, we are in the process of using a myristoylated-GFP into the *cv-c* mutant to visualize cell protrusions and possible changes in the actin cytoskeleton. We are also currently trying to see if there is a genetic interaction between *cv-c* and Rho in CVM migration.

[29] **Characterizing Muscle Disuse in Drosophila**
Jodi Protasiewicz and Robert Wessells
Presenting Author: Jodi Protasiewicz
Wayne State University

In individuals left bed-ridden by their medical conditions, secondary metabolic and physical complications often ensue due to extended sedentary periods, independent of the original illness or injury. *Drosophila* can be a useful tool in studying such cases due to the available genetic techniques and ability to expand studies across several different genotypes in large numbers. Therefore, we generated a model of muscle disuse via enforced restraint in order to characterize the effects of chronic sedentary behavior in flies. Fluorescent imaging of muscle structure and physical performance assays revealed a stark contrast in the overall muscle health of unrestrained versus restrained flies. Flies under restraint possess visibly damaged muscle fibers and perform significantly worse in terms of endurance, climbing speed, and lifespan as compared to unrestrained flies. These findings establish a novel restraint model in *Drosophila* with which to base future work on. Having characterized this model, we are

currently testing the potential of rescuing muscle health of restrained flies through the use of exercise mimetics.

[30] **Characterization Of Flotillin 1 In The Polarized Deposition of Basement Membrane Proteins In Epithelial Cells**

Joseph Austin and Olivier Devergne
Presenting Author: Joseph Austin
Northern Illinois University

Epithelial cells and their proper morphogenesis are critical for the proper development and physiology of metazoans. Epithelial cells exhibit polarized trafficking to transport newly synthesized proteins to their proper location such as the apical, basal, or lateral domains. One component of this apical-basal polarity is the basement membrane (BM) which is secreted basally by epithelial cells. The BM serves as a barrier with selective permeability between epithelial cells and surrounding tissues, provides mechanical support to the epithelial cells, and is important for the establishment and maintenance of epithelial cell polarity. To study the process of BM formation, we use the follicular epithelium of the *Drosophila* ovary, a monolayer epithelium, as a model system. Using this model system, we found a gene potentially involved in epithelial morphogenesis, and basement membrane protein trafficking, *Flotillin 1* (*Flo1*). Flo1 has been shown to be involved in cellular trafficking events such as exocytosis. Epithelial cells knocked down for *Flo1* lose epithelial integrity; they are irregular in shape and accumulate on top of each other. Moreover, the loss of *Flo1* leads to apical mislocalization of BM proteins. Our data suggest that *Flo1* is important for proper BM secretion and epithelium architecture. To confirm and better characterize the role of *Flo1* in epithelial morphogenesis, we generated a Flo1 gene knockout (KO). This $Flo1^{KO}$ allele was generated by complete excision of the *Flo1* coding region using CRISPR/Cas9. We will describe the different approaches that we are undertaking to understand the role of *Flo1* in epithelial cell polarity.

[31] **Gene Annotation Of Akt In Drosophila Persimilis**

Julia Kaniuk and Jennifer Mierisch/Laura K. Reed
Presenting Author: Julia Kaniuk
Loyola University and the University of Alabama

Examining gene conservation across many species is an important approach for exploring the molecular evolution of signaling pathways involved in vital cellular processes. The main goal of this project is to examine the structural conservation of genes in the insulin signaling pathway, a key metabolic pathway, across *Drosophila* species to determine how quickly this pathway is evolving. Existing evidence suggests molecules acting earlier in the insulin signaling pathway evolve more quickly than molecules acting later. *Drosophila persimilis*, the species investigated in this annotation, is a sister species of *Drosophila pseudoobscura* within the *obscura* group which diverged from the *melanogaster* group about 25 million years ago. This distant phylogenetic relationship implies an increased amount of divergence between *D. melanogaster* and *D. persimilis*. *D. melanogaster* has a well annotated genome and therefore serves as a good reference for gene annotations across *Drosophila* species. Using the UCSC Genome Browser, BLAST, and synteny to *Drosophila melanogaster*, we mapped the location of Akt in *D. persimilis* and annotated its coding exons. Our annotation showed many interesting instances of divergence, including an expanded distance between genes and lack of conservation at the beginning of the Akt coding sequence across all isoforms. Additionally, we found a well conserved non-canonical start site for one isoform of Akt which is characterized across many *Drosophila* species. This project will contribute to analysis of the

molecular evolution of insulin signaling pathway genes across *Drosophila* species by Genomics Education Partnership (thegep.org).

[32] **Active Interphase Centrosomes Correlate with Stem Cell Loss**
Julianna Hernández and Pamela K. Geyer
Presenting Author: Julianna Hernández
Unviersity of Iowa

Homeostasis of Drosophila germline stem cells (GSCs) depends upon the integrity of the nuclear lamina (NL). Loss of the Drosophila NL proteins emerin or its partner Barrier-to-autointegration factor (BAF) compromise NL structure, block germ cell differentiation and cause GSC death due to activation of the DNA damage response kinases, ATR and Chk2. Notably, checkpoint activation occurs independently of canonical triggers, such as DNA damage. To understand the mechanism of checkpoint activation, we have investigated NL dynamics in GSC mitosis, as both proteins are required for these processes. We find that in emerin mutants, non-dividing GSCs retain active centrosomes that nucleate microtubules and deform nuclear structure. Previously, we over-expressed NL structural genes to test whether NL deformation was the trigger for checkpoint activation. Having not found this to be consistent with our data, we are now testing the hypothesis that active centrosomes are responsible for checkpoint activation in emerin mutants. We are testing this model using RNA interference to deplete components of the centrosome in wild type and emerin mutant backgrounds, including components of the pericentriolar material (PCM) that is responsible for nucleating microtubules, as well as components of the centriole. Our preliminary data suggest that reduction in the PCM protein Centrosomin partially suppresses GSC loss in emerin mutants, whereas reduction of the centriolar protein Asterless does not. These results provide support for a link between defects in centrosome structure and checkpoint activation.

[33] **Elucidating The Effects Of Trc Knockdown On Glial Development And Function In Drosophila**
Karan Patel and Jennifer Jemc Mierisch
Presenting Author: Karan Patel
Loyola University

The central and peripheral nervous systems are critical for sensory perception and motor output. Glial cells perform multiple functions in the nervous system from forming the blood brain barrier and phagocytosing waste to facilitating and regulating neuronal synthesis and migration. Given the importance of glial cells in establishing a robust nervous system, it is important to understand the genes that regulate glial development, as mutations in these genes can result in defects in neural development. The fruit fly, *Drosophila melanogaster*, has proven an excellent model system for exploring the genetic mechanisms regulating glial development. Our lab has identified the fruit fly homologue of the mammalian NDR family kinase Serine Threonine Kinase 38 (STK38), known as Tricornered (Trc), as a protein needed for glial development in the peripheral nervous system. We have shown that knockdown of *trc* expression results in a reduced number of glia along peripheral nerves and in the developing eye. Larval crawling assays revealed that *trc* knockdown results in a reduced crawling speed as compared to controls. In the context of the developing eye, we are interested in characterizing how *trc* knockdown results in fewer glia. Work has demonstrated that glia are not dying, but it is unclear if they are failing to proliferate or to migrate into the developing eye disc. We are currently performing glial migration assays and assessing glial proliferation to determine if these processes are defective upon *trc* knockdown. Increased understanding of the mechanisms by which Trc promotes glial development will provide insight into how homologous proteins function in glia in other organisms.

[34] **Phenotypic Characterization and Genetic Mapping of a Drosophila Melanogaster Growth Control Mutant**

Kelsey Breneman and Joyce Stamm
Presenting Author: Kelsey Breneman
University of Evansville

This project continues the characterization of mutations from a genetic screen that targeted the left arm of chromosome 2 and aimed to identify genes associated with cell growth and proliferation. This screen was conducted as part of the Fly-CURE consortium. The parent stock had a P-element insertional mutation in the Apoptosis Inducing Factor (AIF) gene that blocks programmed cell death and contributes a *white*$^{+}$ eye color marker. Random EMS mutagenesis of this stock produced mutant lines 4.14.JS and 4.18.JS. These mutations are homozygous lethal and lead to death early in development. Mitotic recombination was therefore induced in the eye using the yeast protein FLP recombinase under the control of the eyeless promoter, generating red homozygous mutant tissue and white wildtype tissue in the eye. Experiments with 4.14.JS and 4.18.JS thus far have demonstrated complete lethality at 25ºC and rare escapers at 18ºC. These escapers generally lacked red mutant tissue in the eye compared to AIF controls. We have also conducted complementation mapping experiments using the BDSC 2L deficiency kit to narrow down the location of these mutations. Preliminary results of these experiments reveal the *Mondo* gene as a candidate for the 4.14.JS mutation, and narrow the 4.18.JS mutation to a 35 kB region in cytological region 26B1-26D7. We are now conducting additional crosses to verify the complementation mapping data, and will examine whole genome sequence data to identify the molecular nature of these mutations.

[35] **Illinois State University Confocal Microscopy Facility: Multiple Imaging Modes for Enhanced Analysis of Drosophila Tissues.**

Kevin Edwards, Isolde McKiernan, and Shaniya Barrett
Presenting Author: Kevin Edwards
Illinois State

The Confocal Microscopy Facility at Illinois State University is open to regional users and features several capabilities that allow enhanced characterization of fly tissues. 1) Resolution enhancement. We extensively tested Leica's Lightning adaptive deconvolution tool and found it greatly improved imaging of defined objects on the 0.1-1 micron scale such as protein aggregates or cytoskeletal structures. 2) Spectral analysis. Certain challenging problems such as colocalization of overlapping dyes, and precisely defining shifts in dye fluorescent properties, can be solved using "spectral fingerprinting" (Lambda-lambda plots) in which every excitation/emission combination is imaged and graphically analyzed. 3) Fluorescence lifetime imaging (FLIM). The timing of fluorescent emissions can be used to complement spectral analysis, providing a way to distinguish different chemical species with similar spectra. We find, for example, that FLIM analysis of cuticle autofluorescence can distinguish among functionally different antennal sensilla.

[36] **Enable Location Services: Confocal Microscopy-Based Screen for Determinants of Protein Pattern and Assembly.**

Ben Lundy, Brandon Bernicky, Andrew Melaga, Shaniya Barrett, and Kevin Edwards
Presenting Author: Kevin Edwards
Illinois State

The intracellular environment is highly structured, with myriad protein-protein (and -lipid) interactions controlling the subcellular location, and in turn the local concentration, activity, and interaction network, of each protein. To identify novel protein localization signals in an unbiased manner, we tested the effectiveness of screening fluorescent protein traps in live Drosophila embryos with laser scanning confocal microscopy. The traps were generated with the Hostile takeover (Hto) system, in which GAL4 drives expression of an mCherry fusion to a downstream coding region (either a full-length form or C-terminal fragment of the target protein). In a pilot screen, we recovered any embryo with a fusion protein showing a pattern distinct from that of the Starter Hto element. Of 79 selected and hatched embryos, 66 eclosed as adults, 56 were crossed, 55 transmitted the fluorescent tag to offspring, and 46 yielded stocks with new stable protein trap insertions. A majority of those fusions were concentrated in the nucleus, with several subtypes (nucleolar, non-nucleolar, and patterned), and there were several lines with distinct non-nuclear patterns (junctions, vesicles, and plasma membrane). A secondary transposition in one of the selected animals yielded an additional line with an apparent microtubule-binding fusion. Thus, live embryo Hto screening provides an efficient means of harvesting protein fragments with diverse and useful localization signals.

[37] **A Kinesin-like Protein Encoded by CG14535 Controls Border Cell Migration During Drosophila Oogenesis**

Leif Verace and Olivier Devergne
Presenting Author: Leif Verace
Northern Illinois University

Cell migration plays a key role in many biological processes including embryonic development, tissue morphogenesis, and wound healing. Cell migration is also involved in metastasis, a hallmark property of cancer cells. Thus, understanding how this behavior works is highly important. To study cell migration, we use a well-established model system: border cell (BC) migration in Drosophila ovaries. During oogenesis, the BCs originate within a monolayer epithelium that envelops the germline cells, forming an egg chamber (the future egg.) BCs are recruited as a cluster of epithelial cells, which delaminate from the epithelium and migrate from the anterior to posterior end through the center of the egg chamber. In a genetic screen looking for novel genes involved in BC migration, we identified a mutant KC53 which results in border cell migration defects. This mutation was mapped to a kinesin-like gene, CG14535, on chromosome 2L. I will describe the different approaches and preliminary results we have obtained to investigate the role of CG14535 in the control of BC migration. These include RNAi knockdowns, mutant clonal analysis, and the generation of a CG14535 knockout using the CRISPR/Cas9 genome editing system to produce an amorphic mutant

[38] **Scraps, An Anilin, And Nebbish, A Kinesin, Are Integral Components Of A Fox Transcription Factor-Regulated Subnetwork That Mediates Specific Cardiac Progenitor Cell Divisions**

M. Rezaul Hasan and Shaad M. Ahmad
Presenting Author: M. Rezaul Hasan
Indiana State

Fox transcription factors mediate multiple cardiogenic processes in both mammals and *Drosophila*. The *Drosophila* Fox genes *jumeau* (*jumu*) and *Checkpoint suppressor homologue* (*CHES-1-like*) mediate three categories of cardiac progenitor cell division—asymmetric, symmetric, and cell division at an earlier stage. *jumu* also regulates the expression of the kinesin Nebbish (Neb) and the activity of the kinase Polo to mediate symmetric and earlier cardiac progenitor cell divisions in a *CHES-1-like*-independent process. By comparing expression profiles, we identified *scraps* (*scra*), an anilin-encoding

gene, that like *neb*, is also transcriptionally activated by *jumu*, but not by *CHES-1-like*. Phenotypic analysis of mutations show that *scra*, like *neb*, is required for only two of the three categories of *jumu*-regulated cardiac progenitor cell division—symmetric and cell division at an earlier stage. Synergistic genetic interactions between *scra*, *neb*, *jumu*, and *polo*, and the absence of such synergistic interactions between either *scra* and *CHES-1-like* or *neb* and *CHES-1-like*, demonstrate that *scra* and *neb* are integral components of a *jumu*-regulated subnetwork mediating a specific subset of cardiac progenitor cell divisions. Preliminary data from our phenotypic analysis of other exclusively *jumu*-regulated genes suggests that the kinesin Pavarotti, the citron kinase Sticky, and the Rho GTPase Tumbleweed may be other components of this subnetwork. Using genetic interaction and rescue assays, we are attempting to position *neb* and *scra* topologically relative to each other and these other potential subnetwork components. Collectively, our results illustrate how an individual regulator can utilize different combinations of downstream effectors to control distinct developmental processes.

[39] **Mutant M.3.2 Is A Novel Allele Of Tout-velu**

MacKenzie Patterson and Jacob D. Kagey
Presenting Author: MacKenzie Patterson
University of Detroit Mercy

Abstract: Mutant M.3.2 was identified in a conditional Flp/FRT screen on chromosome 2R in the adult eye screening for mutations that disrupted cell growth, cell cycle, and organ development. This mutation was characterized and mapped as part of the Fly-CURE consortium by undergraduate researchers at the University of Detroit Mercy, Ohio Northern University, and Morehouse College. The mosaic phenotype of M.3.2 results in a deformed mosaic eye that has expansion of cuticle, a smaller adult eye, and other developmental disruptions. To identify the mutation causing these severe phenotypes we utilized deficiency mapping to locate the region on 2R. Initial mapping found a region on chromosome 2R that failed to complement. Further mapping identified M.3.2 to fail to complement two independent alleles of *tout-velu*, suggesting that M.3.2 is a novel allele of *tout-velu*$^{M.3.2}$.

[40] **Metabolic Regulation Of Blood-Progenitor Development In Drosophila**

Manisha Goyal, Bruce R Cooper, Ramaswamy Subramanian, and Tina Mukherjee
Presenting Author: Manisha Goyal
Purdue University

Much similar to mammals, *Drosophila* hematopoiesis takes place in two different waves, while the primitive wave originates during embryonic stages of development, the definitive wave takes place in a specialized larval hematopoietic organ termed the lymph gland. Various signaling cues have been shown to regulate lymph gland development, and recent studies shed light on the involvement of metabolic pathways in myeloid development. Among these metabolites, the role of reactive oxygen species (ROS) in myeloid development is well established and its aberrant generation alters hematopoiesis. Thus, maintaining homeostatic levels of ROS is very crucial for the blood-progenitor cells. Any understanding of intracellular metabolic or signaling events that enable the sustenance of this fine redox balance and blood-progenitor development remains obscure. We previously showed that, in homeostasis, myeloid-like blood-progenitor cells of the *Drosophila* larvae utilize TCA cycle to generate ROS and excessive ROS production leads to lymph gland growth retardation. Therefore, to moderate blood-progenitor ROS, *Drosophila* larvae rely on olfaction and its downstream systemic GABA. Another aspect of ROS regulation is its scavenging and we found that GABA catabolism controls antioxidant synthesis necessary to scavenge any excess ROS that is generated. GABA catabolism regulates serine levels in the lymph gland which further contributes to GSH generation. We have identified the metabolic requirement

of odor sensing and GABA in regulating redox homeostasis during *Drosophila* myeloid progenitor development, the relevance of which may be broadly conserved.

[41] **Bruno1 Is Required Throughout Drosophila Indirect Flight Muscle Development To Regulate Dynamics Of Sarcomere Assembly And Growth**
Elena Nikonova, Tobias Straub, and Maria Spletter
Presenting Author: Maria Spletter
University of Missouri Kansas City

The differential expression of structural protein isoforms influences cytoskeletal assembly and contractile properties. CELF family RNA binding proteins are important regulators of RNA processing, but we do not fully understand how misregulation of CELF proteins leads to defects in sarcomere assembly, growth and function. Bruno1 (Bru1, Arrest) encodes a CELF1/2 homolog in *Drosophila* that regulates flight muscle specific alternative splicing. Here we show that Bru1 is required throughout muscle development to regulate cytoskeletal assembly and growth dynamics. During early myofibril formation before 48h APF, using both temporally-restricted RNAi knockdown and overexpression, we show that misexpression of Bru1 leads to disorganization of the actin cytoskeleton, aberrant myofiber compaction and defects in pre-myofibril formation. Transcriptomic and proteomic analyses revealed misexpression and isoform switches in diverse structural proteins regulating sarcomere growth and actomyosin interactions. Live-imaging assays confirmed aberrant contractility of *bru1* mutant myofibers. By monitoring incorporation of fluorescent actin and myosin proteins during myofibril maturation after 56h APF in *bru1* mutant IFM, we show that lateral sarcomere growth is dramatically misregulated, leading to exacerbation of pre-existing defects, myofibril fusion and formation of hollow myofibrils. A progression in the severity of cellular and molecular phenotypes from 80h APF to adult distinguishes hypercontraction from earlier growth defects, and temporally restricted rescue can partially alleviate hypercontraction in late pupal and adult stages. Taken together, our data indicate that Bru1 regulates cytoskeletal growth and remodeling throughout myogenesis, including cytoskeletal rearrangement necessary for myofibril formation as well as the balance in length versus lateral growth of the sarcomere. Defective RNA processing due to misexpression of CELF proteins thus causes wide-reaching structural defects and progressive malfunction of affected muscles.

[42] **Exercise Mimetics As A Rescue For Mobility Phenotypes In a Drosophila Clock mutant**
Maryam Safdar and Robert Wessells
Presenting Author: Maryam Safdar
Wayne State University

Disturbances in circadian rhythms are associated with various negative health outcomes, which include an increasing incidence of chronic diseases with high societal costs. While exercise can positively impact the downstream negative effects of rhythm disruption, it is not available to all patients impacted by sleep disruptions, in part because sleep disruption itself reduces exercise capacity. Therefore, there is a need for therapeutics that bring the benefits of exercise to this population. One method to discover these therapeutic is to investigate the relationship between endurance exercise and circadian rhythm disturbances using a *Drosophila* model of rhythm loss, the Clk^{out} mutants. These mutants have been well established to have a disrupted circadian rhythm of activity and sleep. Our lab has shown them to also have the phenotypes of reduced exercise capacity, measured as post-training endurance, flight performance, and climbing speed. Rescue will be accomplished using a genetic method via Sestrin overexpression and a pharmacological rescue via Octopamine feeding. Both molecules, Sestrin and Octopamine, are well-known mediators of exercise and the methods listed above have been shown to

mimic exercise in sedentary *Drosophila*. Sestrin overexpression has also, previously, been shown to rescue neurodegenerative mobility phenotypes in *Drosophila*.

[43] **Toxicity of polyglutamine-expanded ataxin-3 is potentiated by mutating its ubiquitin-binding site 1**
Matthew V. Prifti and Sokol V. Todi
Presenting Author: Matthew V. Prifti
Wayne State University

Polyglutamine (polyQ) diseases are a family of nine neurodegenerative disorders caused by abnormal CAG triplet repeat expansions in different, widely-expressed, disease-causing genes. Long glutamine expansions are translated from these CAG repeats and cause clinically-distinct neurological disorders that include, but are not limited to, Huntington's Disease, Kennedy Disease and several spinocerebellar ataxias (SCAs). SCA3 is the most common, dominantly inherited ataxia worldwide, and is caused by a polyQ expansion in the deubiquitinating enzyme (DUB), ataxin-3 (ATX3), which is involved in in regulating protein quality control. To understand the mechanisms of disease of SCA3, a yet incurable disease, our lab has taken a systematic approach to studying the domains of ATX3 and their respective and collective roles in its toxicity. Ubiquitin-binding site 1 (UbS1), found in the catalytic domain of ataxin-3, plays a key role in its ability to function as a DUB by binding to ubiquitin and coordinating its spatial interaction with the catalytic site responsible for its overall deubiquitinating activity. We generated transgenic Drosophila lines that express pathogenic ATX3 with intact or mutated UbS1 and found that mutating UbS1 markedly exacerbates the toxicity of pathogenic ATX3. Additional experiments indicate that UbS1 mutations enhance the ability of ataxin-3 to sequester ubiquitin species in neurons, potentially segregating vital proteins away from other essential cellular pathways.

[44] **Genetic Variation in Dietary Sugar Consumption in Drosophila.**
Mubaraq Opoola, Lucas Fitzgerald, Nicholas Wright, and Dae-Sung Hwangbo
Presenting Author: Mubaraq Opoola
University of Louisville

Sugar is a key part of the daily diet; its overconsumption is often a major contributing factor to many metabolic diseases such as diabetes and cancer. Additionally, some non-metabolic diseases like high blood pressure have been impacted by high sugar consumption. In mammals, prolonged ingestion of excessive dietary sugar promotes overconsumption of the diet partially due to a desensitization of sweetness over time. Yet, its genetic and neuronal mechanisms are not fully understood. Using the fruit fly, *D. melanogaster* as a model system, we investigated the genetic variability of the trait (sugar-induced overfeeding) among eight different wild-type populations and in ~ 170 isogenic lines from the Drosophila Genomic Reference Panel (DGRP). Surprisingly, none of these lines showed an increased food consumption on a high-sugar diet (20% sucrose). This observation suggests that flies employ protective mechanisms from overconsumption of food caused by excessive sugar. A follow-up genome-wide association study (GWAS) using the data from DGRP lines identified ~ 60 genes that may have protective roles from over-consumption of a high-sugar diet.

[45] **Using Drosophila To Validate Genetic Modifiers Of Muscle Laminopathies**
Nathaniel P. Mohar and Lori L. Wallrath
Presenting Author: Nathaniel P. Mohar
Unviersity of Iowa

Mutations in the human LMNA gene cause a collection of diseases known as laminopathies. These diseases affect many tissue types including striated (skeletal and cardiac) muscle. A hallmark of these striated muscle laminopathies is their phenotypic variability. Individuals with the same LMNA mutation can have clinically distinct diseases. Furthermore, individuals with the same clinical diagnosis are likely to have extensive variation in the severity of their muscle disease phenotypes. Here we describe four siblings that all possess the same LMNA mutation. Two of the four individuals have severe Emery Dreyfuss muscular dystrophy, and two have very mild muscle disease. We have modeled the LMNA mutation in the Drosophila ortholog LamC. Muscle-specific expression of mutant LamC causes many muscle defects similar to those observed in the human disease. To explain the variability among the four siblings, whole genome sequencing was performed and a variant in SMAD7 was identified that co segregates with severe disease. We have generated a Drosophila model with the identified variant through CRISPR editing of the Drosophila ortholog of SMAD7, called Dad. These flies exhibit severely reduced fertility and a partial block in oogenesis, which implies that the modified Dad fails to regulate Dpp signaling. Future work will focus on combining expression of mutant LamC with the Dad variant to look for genetic interaction through the use of muscle function assays. These studies will implicate alterations in SMAD signaling an enhancer of LMNA-associated skeletal muscle disease.

[46] **Outcomes Of Genetic Manipulations Of Nerve Ensheathment In The Fruit fly, Drosophila melanogaster**

Nelchi Prashali and Joyce Fernandes
Presenting Author: Nelchi Prashali
Miami University

In most higher organisms, the nervous system is modified throughout the life cycle to accommodate changing behaviors or in response to injury and aging. This phenomenon is called neural plasticity. Five of the eight pairs of abdominal nerves converge during metamorphosis to form the terminal nerve trunk (TNT). Each abdominal nerve is ensheathed by an external acellular layer called the neural lamella (NL) and three concentric inner layers of glial cells. Our lab is interested in how nerve ensheathment is re-organized as the nerves fuse to give rise to the TNT. During the first day of metamorphosis, NL breaks down and an associated 4-fold increase in the number of adjacent perineurial glial (PG) cells is observed. This forms the basis for our working hypothesis that NL breakdown releases proliferative signals which are received by PG cells. To test this hypothesis, NL breakdown was manipulated using the Gal4-UAS-Gal80^{ts} system to spatially and temporally prevent the action of matrix metalloproteases (MMPs) that are known to break down NL. A second hypothesis is that NL breakdown is a prerequisite for TNT formation and eclosion of the animals. A significant decrease in the number of proliferating PG cells per unit length of nerve (50 microns) is observed when NL degradation is disrupted during the earlier stages of metamorphosis. Additionally, a 30% decrease in eclosion rate of experimental animals was noted. Our project aims to test the impact of the genetic manipulations on PG cell proliferation, TNT formation, and the motor behavior of eclosion.

[47] **Nuclear Actin is a Critical Regulator of Drosophila Germline Stem Cell Maintenance**

Nicole Green, Danielle Talbot, and Tina Tootle
Presenting Author: Nicole Green
University of Iowa

While actin was observed in the nucleus decades ago, nuclear functions of actin dynamics have only recently been widely acknowledged. Our studies investigate the roles of nuclear actin in regulating stemness using a model tissue, the *Drosophila* ovary. The *Drosophila* ovary is made up of 15-20 ovarioles of sequentially developing follicles. Germline stem cells (GSCs) give rise to all germline cells

and reside in a niche at the anterior tip of each ovariole in a structure known as the germarium. Our lab previously defined several distinct pools of nuclear actin in the ovary by screening established actin labeling tools. One of these pools stained by anti-actin C4 is found in both the nucleoplasm and nucleolus of GSCs. The polymeric nucleoplasmic C4 pool is lost quickly at the 2-cell stage and the monomeric nucleolar pool persists to the 8-cell stage. This dynamic trend suggests nuclear actin, in particular polymeric actin, may play a role in regulating stemness. To test this hypothesis, we overexpressed NLS-actin constructs in germline cells of the germarium. When we increased monomeric nuclear actin (NLS-Act5C^{G13R}), but not WT actin (NLS-Act5C), there is progressive germline loss indicating defective GSC maintenance. Additionally, increasing monomeric nuclear actin disrupts nuclear architecture causing nucleolar hypertrophy, distortion of the nuclear lamina, and heterochromatin reorganization. Germline loss was rescued by simultaneous over-expression of monomeric nuclear actin and WT nuclear actin to restore polymeric nuclear actin pools. These studies indicate nuclear actin is critical for germline maintenance and provide exciting new insight into its developmental roles.

[48] **The SNARE Protein Snap29 Controls The Secretion Of Basement Membrane Proteins In The Drosophila Follicular Epithelium.**

Nusrat Jahan and Olivier Devergne
Presenting Author: Nusrat Jahan
Northern Illinois University

Basement membranes (BMs) are thin layers of extracellular matrices that localize at the basal side of epithelial cells. They are important for the establishment and maintenance of the apical-basal polarity of epithelial cells. However, little is known about the pathway that regulates the basal restriction of BM proteins. To uncover this pathway, we use the *Drosophila* follicular epithelium (FE) as a model system. To be secreted basally, BM proteins containing vesicles must be transported to the basal side of the FE. This vesicle-mediated transportation requires the fusion of vesicles with their target compartments. SNARE protein family has an active role in this fusion, suggesting its involvement in BM protein secretion. To identify specific SNAREs involved in the BM secretion, we performed a genetics screen using RNAi lines against all the *Drosophila* SNAREs. To assess the localization of BM proteins, we used two GFP-tagged BM proteins: Perlecan-GFP (Pcan-GFP) and Collagen IV-GFP (Coll IV-GFP), two main components of the BM. In this screen, we identified the SNARE *Snap29*. The loss of *Snap29* leads to the accumulation of Coll IV but not Pcan in intracellular structures suggesting its specific role in the secretion of Coll IV in the FE. However, the distribution of other apical-basal polarity markers is not affected in *Snap29* mutant cells suggesting a specific role of *Snap29* in the control of BM secretion. Altogether, our data show that *Snap29* is necessary to control the proper secretion of Coll IV, a main component of the BM proteins in the *Drosophila* FE.

[49] **MTORC2 Protects The Heart From High-fat Diet-induced Cardiomyopathy Through Mitochondrial Fission In Drosophila**

Peiduo Liu and Hua Bai
Presenting Author: Peiduo Liu
Iowa state

High-fat diet (HFD)-induced obesity has become the major risk factor for the development of cardiovascular diseases, but the underlying mechanisms remain poorly understood. Here, we use *Drosophila* as a model to study the role of mTORC2 in HFD-induced mitochondrial fission and cardiac dysfunction. We find that knockdown of mTORC2 subunit *rictor* blocks HFD-induced mitochondrial fragmentation and Drp1 recruitment. Knockdown of *rictor* further impairs cardiac

contractile function under HFD treatment. Surprisingly, knockdown of *Akt*, the major effector of mTORC2, did not affect HFD-induced mitochondrial fission. Similar to mTORC2 inhibition, knockdown of *Drp1* blocks HFD-induced mitochondrial fragmentation and induces contractile defects. Furthermore, overexpression of *Drp1* restored HFD-induced mitochondrial fragmentation in *rictor* knockdown flies. Thus, we uncover a novel function of mTORC2 in protecting the heart from HFD treatment through Drp1-dependent mitochondrial fission.

[50] **Characterizing the Role of p38Kb and GARS in CMT**

Piotr Klos and Alysia D. Vrailas-Mortimer
Presenting Author: Piotr Klos
Illinois State

Charcot-Marie-Tooth Disease (CMT) is a progressive neuropathology caused by the deterioration of neuronal function of the peripheral motor and sensory neurons. Symptoms include tripping, ankle twisting, and clumsiness as well as sensations such as pins and needles and burning pain. One gene mutated in CMT is the glycine tRNA-synthetase, GARS. We have recently found that the p38 MAPK (p38Kb), which regulates aging and age-dependent locomotor behaviors, regulates the levels of GARS during aging. Since p38Kb interacts with the Chaperone-Assisted Selective Autophagy (CASA) complex to mediate the degradation of misfolded or nonfunctional proteins, this suggests that during aging, GARS becomes damaged and is normally cleared by p38Kb and the CASA complex. Failure to clear these damaged GARS proteins potentially results in disease symptoms or worsening of symptoms. I hypothesize that p38Kb-mediated regulation of GARS degradation is crucial for maintaining proper neuromuscular function. I will utilize *D. melanogaster* to test interactions between p38Kb and GARS and how this contributes to CMT-like phenotypes in flies by determining if p38Kb can mediate the clearance of mutant GARS from the cell. We have found that overexpression of p38Kb in motor neurons leads to locomotor dysfunction, and confirmed that expression of GARS mutations in neuronal tissue alone is sufficient to cause CMT2D. Overexpression of p38Kb in muscles improves locomotor function, and newer evidence suggests that mutant GARS expression in the mesoderm is sufficient to induce neuropathology. Future research with p38Kb can help us better understand its role in progressive diseases.

[51] **Drosophila Eye Model To Study The Role Of Mnat9 In Alzheimer's Disease Related Dementia**

Prajakta Deshpande, Emily Snider, Anuradha Venkatakrishnan Chimata, Madhuri Kango-Singh, and Amit Singh
Presenting Author: Prajakta Deshpande
University of Dayton

Alzheimer's Disease (AD), a progressive neurodegenerative disorder, is manifested as extracellular accumulation of amyloid-beta-42 (Aβ42) plaques and intracellular accumulation of neurofibrillary tangles (NFTs) due to hyperphosphorylation of tau that results in destabilization of microtubules. Targeted misexpression of human Aβ42 (GMR>Aβ42) in retinal neurons of developing *Drosophila* eye results in Aβ42 plaque(s) formation, extensive neurodegeneration and mimics AD like neuropathology. However, the underlying mechanism(s) for Aβ42-mediated neurodegeneration have not been fully understood. In a forward genetic screen, we identified N-acetyltransferase 9 (Mnat9) as one of the genetic modifiers of GMR>Aβ42 neurodegenerative phenotype. Mnat9 is known to stabilize microtubules by inhibiting c-Jun-N- terminal kinase (JNK) signaling. The neurodegenerative phenotype of GMR>Aβ42 is rescued by gain-of-function of *Mnat9* whereas loss-of-function of *Mnat9* exhibits converse phenotype of enhanced neurodegeneration. Human Mnat9 also suppresses Aβ42-mediated

neurodegeneration suggesting the functional conservation. Surprisingly, Mnat9 neuroprotective function is independent of its acetylation activity. We found that Mnat9 downregulates JNK signaling pathway, which is involved in rescuing neurodegenerative phenotypes seen in GMR>Aβ42 background. Here we propose a new neuroprotective function of Mnat9 in downregulating JNK signaling pathway to ameliorate Aβ42-mediated neurodegeneration.

[52] **Fox Transcription Factors Mediate Proper Positioning Of Cardiac Cells By Restricting The Expression Of ECM Genes**

Rajnandani Katariya and Shaad M. Ahmad
Presenting Author: Rajnandani Katariya
Indiana State

The development of a complex organ requires the specification of appropriate numbers of its constituent cell types as well as their correct positioning within the organ. We previously showed that Fox transcription factors (TFs) Checkpoint suppressor homologue (CHES-1-like) and Jumeau (Jumu) determine the correct number of different cardiac cell types by regulating cardiac progenitor cell divisions. Here we show that *CHES-1-like* and *jumu* are also required for the correct positioning of these cardiac cell types: null mutations in either gene result in the misalignment and incorrect location of cardial and pericardial cells within individual hemisegments. Statistical analysis demonstrated that these positioning defects cannot be completely explained by steric constraints caused by differing number of cardial cells in contralateral hemisgments due to cell division defects. In order to discover the other cause underlying positioning defects, we compared genome-wide transcription expression profiles of purified mesodermal cells from wild-type embryos and Fox mutants to identify Fox-regulated targets. Among the 2,131 target genes we identified, genes encoding extracellular matrix (ECM) proteins were overrepresented among genes repressed by the Fox TFs. In particular, the ECM proteins Viking, Collagen type IV alpha 1, and Terribly reduced optic lobes were all overexpressed in Fox mutants. Our preliminary phenotypic analysis of these specific targets suggests that the Fox TFs bring about the correct positioning of cardiac cell types by restricting their expression: ectopic overexpression of each of these ECM genes in the mesoderm phenocopies the cardiac cell positioning defects observed in *CHES-1-like* and *jumu* loss-of-function mutants.

[53] **Methanol Leaf Extract of Carissa edulis (Vahl) Affects Reproductive Fitness of Drosophila**

Rashidatu Abdulazeez, Nadia E. Usman, Haruna O. Suleiman, Hajara Ibrahim, Dalhatu M. Shehu, Iliyasu S. Ndams, and Nuhu M. Danjuma
Presenting Author: Rashidatu Abdulazeez
Ahmadu Bello University Zaria, Nigeria

Carissa edulis is a spiny evergreen shrub, which is consumed by man, birds, and insects because of its nutritional and medicinal values. The study was aimed at investigating the effect of methanolic extract of *C. edulis* leaves on the reproductive fitness of Harwich strain *Drosophila melanogaster*. Various doses (149, 298 and 595 mg/Kg) of *C. edulis* extract were incorporated into the diet of the fruit flies. Fecundity and filial generation outputs were assessed. The health of each filial generation was evaluated using climbing assay. Although not significantly different, fruit flies fed with 149 mg/Kg of *C. edulis* extract laid higher number of eggs compared to other doses and the control. The 298 mg/Kg supported the highest number of emerged and healthy flies while 595 mg/Kg had the least number in most generations. The control group, however had the highest eclosure rate across the three generations. *C. edulis* MeOH leaf extract at lower concentrations improved reproduction in Harwich strain *D. melanogaster.*

[54] **The G-Signaling Protein Rcp controls the polarized basement membrane deposition in epithelial cells**

Rebecca Brnot and Olivier Devergne
Presenting Author: Rebecca Brnot
Northern Illinois University

Epithelial tissues, the most common tissue type in the human body, form the outer layer of the skin and organs. These tissues are composed of epithelial cells, and their function relies heavily on their cellular architecture. The basement membrane (BM), a specialized sheet within the extracellular matrix, is a vital structure for the establishment and maintenance of epithelial architecture. BM proteins produced in epithelial cells are secreted only to the basal side of these cells through a dedicated biological pathway. Despite its important role in epithelial cell organization and polarity, the biological pathway dedicated to the BM polarized secretion is poorly understood. To study BM deposition, we use the follicular epithelium (FE) of the *Drosophila* ovary as a model system. In a genetic screen looking for new genes involved in the proper placement of BM proteins, we identified a new gene, *Rcp* (Receptor component protein), involved in G-protein signaling. The loss of *Rcp* leads to apical deposition of BM proteins at the FE without primarily affecting the localization of apical and lateral polarity proteins, indicating that *Rcp* specifically controls the proper placement of BM proteins. Interestingly, *Rcp* is the first component of a signaling pathway that has been implicated in BM polarity. Moreover, using immunostaining and super-resolution microscopy, we determined that *Rcp* localizes in the cytoplasm and nucleus of epithelial cells. Altogether, our data indicate a specific role for *Rcp* in the organization of epithelial architecture by regulating the polarized deposition of BM proteins.

[55] Poster Withdrawn

[56] **Wolbachia infection confers olfactory changes and modification of glutamic acid decarboxylase (GAD) in Drosophila melanogaster**

Sarah Boothman and Jonathan Minden
Presenting Author: Sarah Boothman
Carnegie Mellon University

Infection with parasites and intracellular microbes can lead to behavioral changes within the host. The "Behavioral Manipulation Hypothesis" claims these pathogens evolved the ability to modulate these behaviors to facilitate successful transmission from host to host (Thomas et al., 2005). However, there is much debate over the validity of this hypothesis, prompting the need for in-depth studies of mechanisms underlying microbe manipulation of host behavior. We use *Drosophila melanogaster* and its obligate intracellular symbiont *Wolbachia pipientis* as a model for this phenomenon, as multiple host effects have been observed upon infection, including changes in behavior, mating, and fitness (Goodacre & Martin, 2012; Bi & Wang, 2019). In our work, we have specifically observed an increase in olfactory perception towards food odors upon *Wolbachia* infection. Preliminary experiments using pharmacological agents have suggested the involvement of GABA signaling in this behavioral change. Accordingly, we have discovered post-translational modification (PTM) of glutamic acid decarboxylase (GAD) in *D. melanogaster* heads upon *Wolbachia* infection using two-dimensional difference gel electrophoresis (2D-DIGE). Modification and regulation of this protein is largely understudied in *Drosophila*, so our current work aims to identify the type of PTM conferred upon infection and discover how it may contribute to the observed change in olfactory behavior. The results of this work will provide insight into the molecular mechanisms by which *Wolbachia* modulates *Drosophila* biology to better understand how pathogens manipulate host behavior more broadly.

[57] **Polycomb Patterns The Anterior Embryonic Dorsal Vessel By Repressing Abdominal-A Expression**

Sumaiya Islam and Kristopher R. Schwab
Presenting Author: Sumaiya Islam
Indiana State

Embryonic development requires the precise regulation of transcription which is partially controlled by the actions of the Trithorax group (TrxG) and Polycomb group (PcG) proteins. We have previously identified *trithorax* (*trx*) as a positive regulator of *Hox* expression within the *Drosophila* embryonic dorsal vessel (the linear heart tube). The loss of *abdominal-A (abd-A)* expression within the *trx* mutant induces a striking homeotic transformation of the heart-proper into an aortic fate. Since *trx* activates cardiac *abd*-A expression, the repressive PcG genes may silence *abd-A* within the anterior regions of the dorsal vessel. Here, we show that *Polycomb (Pc)*, a gene that encodes an essential component of the Polycomb Repressive Complex 1 (PRC1), represses the anterior *abd-A* expression within the dorsal vessel. The de-repression of *abd-A* throughout the *Pc* mutant dorsal vessel induces several cellular and morphological abnormalities that deform the cardiac patterning. Since *abd-A* is the primary homeotic selector of the posterior heart-proper region, it is likely that the loss of *abd-A* repression has induced the homeotic transformation of the entire dorsal vessel into a heart-proper-like fate. These data suggest that *trx* and *Pc* antagonize the regulatory activities of one another ensuring that *abd-A* expression and heart-proper development is restricted to the posterior dorsal vessel.

[58] **Central Circadian Clock Control of Drosophila Feeding and Activity Rhythms**

Sumit Saurabh, Ruth Meier, Liliya Pireva, and Daniel Cavanaugh
Presenting Author: Sumit Saurabh
Loyola University

Animals including humans are deeply attuned to their environments and exhibit rhythmic behaviors that follow a 24 hour light-dark cycle. *Drosophila* behavioral rhythms are governed by a set of genes e.g., *period* (*per*) and *timeless* (*tim*) that are present in ~150 of 250,000 neurons in the central nervous system. These neurons constitute a core central clock network that can be functionally and anatomically subdivided into seven different neuronal subsets - dorsal neurons (DN1, 2, and 3), dorsolateral neurons (LNds), small and large ventrolateral neurons (sLNVs and lLNvs) and lateral posterior neurons (LPNs). We asked if different circadian behaviors e.g., feeding and activity, are differentially regulated by these neurons. Using GAL4-inducible gene knockdown via the CRISPR-Cas9 gene editing system, we eliminated *per* and *tim* in selective subsets of clock neurons. We find that free-running feeding rhythms require molecular clock function within multiple individual clock cell populations, and furthermore that the severity of the effect varies according to the cell population targeted. Genetic silencing utilizing overexpression of the Kir2.1 potassium channel recapitulated the effects of gene knockdown. These results parallel those observed when using locomotor activity as a behavioral endpoint, suggesting that circadian control of these two distinct behavioral outputs diverges in downstream circadian output cells rather than in cells of the core clock network.

[59] **The Serine-like Protease masquerade (mas) Plays an Important Role in Tracheal Tube Formation**

Victoria Kurdyumov and Afshan Ismat
Presenting Author: Victoria Kurdyumov
University of St. Thomas, Minnesota

The embryonic trachea is a convoluted series of epithelial tubes that provide oxygen to all cells and tissues in the embryo. Proper formation of these tubes involves several distinct cellular processes, including invagination, collective migration, and cell intercalation. *Masquerade* (*mas*) is a serine-like protease that is expressed in the embryonic trachea and has been shown to be involved in proper axonal guidance and somatic muscle attachment, perhaps acting as a competitive antagonist to other serine proteases in the extracellular matrix. Our preliminary data suggests that *mas* also plays an important role in tracheal tube formation. In the absence of mas, we found that a significant percentage of mas mutant embryos had major defects in the trachea. Specifically, individual tracheal metameres had missing dorsal trunk or dorsal branches while the rest of the trachea formed normally. An apoptosis marker, Cleaved Caspase-3 (CC3) revealed that apoptosis is likely not the cause of these missing branches. Moreover, upon closer examination of the dorsal trunk, we found that, even when the lumen was missing, a marker for tracheal nuclei was still present. We are currently in the process of marking the entire membranes of all tracheal cells to see if there is any change to cellular protrusions or membrane degradation. We are also in the process of generating untagged and GFP-tagged over-expression constructs of *mas* to be able to examine what happens in the trachea with too much Mas present. Overall, this work will be instrumental to our understanding of tracheal tube formation.

[60] **Effects of Proline on Heavy Metal Survivability in *Drosophila melanogaster***
Matthew Lowe, Eric Kilbourn, and Jason M. Tennessen
Presenting Author: Matthew Lowe
Indiana University

Heavy metals such as arsenic, lead, and cadmium are contaminants prevalent in many environments throughout the world. These chemicals have been identified by the World Health Organization as posing severe health risks to humans. Previous results in our lab have shown decreased proline levels in *D. melanogaster* when exposed to arsenic. The mutant strain, $slgA^{1}$, is a genetic line of flies unable to catabolize proline due to a mutation in the gene encoding for proline dehydrogenase, resulting in increased proline levels. We use this to examine the effects of increased proline levels on heavy metal exposure. Comparing the survival of $slgA^{1}$ and w^{1118} flies exposed to heavy metals, we observed $slgA^{1}$ flies were significantly resistant to heavy metal-induced lethality. Supplementing w^{1118} flies with proline failed to produce any resistance to arsenic exposure. GC-MS analysis of w^{1118} flies after exposure to arsenic with proline supplementation showed increased proline levels compared to flies exposed to arsenic only; this was not observed in unexposed flies supplemented with proline.

[61] **PrecisionTox – Using *Drosophila* to Redefine Chemical Safety Testing**
Shannon R. Smoot, Emma Rose Gallant, Jessica M. Holsopple, Ellen M. Popodi, Brian Oliver, Thom C. Kaufman, Jason M. Tennessen
Presenting Author: Shannon R. Smoot
Indiana University

Human industries generate over 350,000 chemicals, however, many of these compounds have not been adequately studied for environmental safety or effects on human health. To address this pressing, global issue, the European Union recently funded a pilot study by the PrecisionTox consortium, with the goal of establishing a high-throughput chemical safety screening pipeline. This international consortium, which consists of labs from 14 universities, is tasked with using six model organisms/systems (*C. elegans, Daphnia, Drosophila,* zebrafish*, Xenopus,* and human cell lines) to determine how 250 candidate compounds disrupt gene expression and metabolic flux. The Tennessen lab, in collaboration with Brian Oliver's Lab at NIH and the Bloomington *Drosophila* Stock Center, are responsible for conducting the *Drosophila* chemical exposures. Towards this goal, we have established a high-

throughput exposure protocol for generating dose-response curves and collecting samples for RNAseq and untargeted metabolomics. Our initial studies are focused on an initial library of 50 compounds. Many of these compounds have never been studied in *Drosophila* and our preliminary studies reveal several unexpected phenotypes. Notably, we find that many chemicals produce dramatic sex-specific differences in terms of lethality and feeding behavior. In collaboration with Brian Oliver, we are now investigating the molecular mechanisms that underlie these sex-specific differences.