

LAUREN BONNER, Brown University
Replay of Incidentally Encoded Episodic Memories in Rats
Mentors: Cassandra Sheridan, Jonathon Crystal

Given their keen sense of smell, olfactory stimuli provide a powerful vehicle through which to examine the presence of episodic memory in rats. The central hypothesis of an animal model of episodic memory proposes that, at the moment of a memory assessment, the animal remembers back in time to the relevant event. Rats were trained on a list-encoding task consisting of a variety of olfactory stimuli. As most models of animal episodic memory rely on training, it is possible that animals are able to use their training to anticipate that the encoded information is necessary for memory assessments, therefore relying on non-episodic memory instead. In turn, administering an unexpected memory assessment of incidentally encoding information prevents the animal from knowing if encoded information is important, making it impossible for the animals to transform the information into an action plan when faced with the unexpected memory assessment. The present study used rats that had been trained to replay episodic memory through an explicitly encoded list of odors that were trial unique. In a second task, rats foraged in a radial maze in the absence of odors. In a critical test, rats foraged in the radial maze and encountered novel odors. Next, they were presented with a memory assessment that required the identification of the third-to-last odor that was encountered in the maze. It was hypothesized that novel odors would reduce the likelihood that rats automatically encoded odors for the purpose of an upcoming test of memory. Thus, continued high accuracy on unexpected memory assessments indicates replay of episodic memory. Rats performed with high accuracy on the critical unexpected memory assessment. These results suggest that rats are able to replay episodic memory of incidentally encoded information.

MADISON BOUGGESS, Vassar College
Discovering the Role of Cadherin-23 Genes in Class 1 MDDA ddaE and ddaD Neurons
Mentors: Dorian Dale and Daniel Tracey

In *Drosophila melanogaster*, or fruit flies, transmembrane channel-like (TMC) proteins are utilized for locomotion and expressed in class I multi dendritic arborization (clda) neurons of fruit fly larva. TMC exhibits the same directional sensitivity in the neurons of fruit fly larva that is seen in the mechanosensitive directionally selective hair cells in vertebrates. Tip links, the string of cadherin molecules that are in charge of opening and closing TMC channels for vertebrates, are important for TMC's function. Although, no work has been done to establish whether cadherin genes are important for how TMC proteins work for invertebrates. Therefore, to address the question, we are screening clda neurons for TMC signal intensity and neuron morphology in the presence of cadherin knockdown. We crossed *UAS-Tmc::GFP;2-21-GAL4* (TMCGFP) virgin female flies with 16 respective male lines carrying RNA interference (RNAi) in order to visualize the effect of cadherin knockdown on the clda neurons and TMC. We used a Zeiss confocal microscope with a 488 nm laser to take a z-stack in order to generate a maximum intensity projection of the ddaE and ddaD neurons in one body segment (abdominal segment 6) for each larva. Our positive controls for this experiment were larva with RNAi for the TMC protein localized in the ddaE and ddaD neurons. Our negative controls were TMCGFP virgin female fruit flies crossed with males containing an empty attP site corresponding to the experimental group. We found that larva with RNAi for the cadherin genes *Cad87A 28716*, *Cad99C 27510*, *Cad99C 35037*, *CadN2 27508*, *CadN 27503*, *Cad86C 27295*, *Cad88C 29303*, and *CadN 41982* presented a decrease of TMC signal intensity in the ddaE and ddaD neurons. Additionally, *Cad74A 27485*, *Cad86C 61280*, *27295*, *53314*, and *Cad87A 28716* are cadherin genes that produce larvae with some irregular neuron morphology. Irregularities in the neurons observed included extra dendrites and extra curls at the ends of neurons in comparison to the neurons in the negative control group. In conclusion, the absence of cadherin genes in fruit fly larva seems to affect the TMC signal intensity in ddaE and ddaD neurons used for locomotion. This decrease in TMC intensity in the clda neurons may indicate irregularly segmented movement in the larva. In the future, we plan to quantify the images taken of the neurons in order to validate our observations of low TMC signal intensity and irregular neuron morphology in specific larva with cadherin knockdown. Hopefully, from this research we can learn more about how the loss of certain genes in proprioceptors contributes to larval movement and gain insight into the mechanisms facilitating locomotion.

MANUEL CORTES ROMERO, Cornell University
Studying Host Preference and its Contribution to the Co-occurrence of
Entomopathogenic Nematodes
Mentors: Jonathan Enriquez Madrid and Farrah Bashey-Visser

Biodiversity plays a critical role in the maintenance of ecosystems by providing sustainable food chains and the general wellness of organisms. Biodiversity is enhanced when species can stably coexist, which can be promoted by niche partitioning. Entomopathogenic Nematodes (EPNs) are an interesting group of organisms as multiple species appear to co-occur in the same ecosystem while using the same resources. In addition, EPNs play a pivotal role in the maintenance of ecosystems. EPNs are known as obligate parasitic nematodes that will kill their insect host. Finding a worthy host to inhabit is a crucial milestone in an EPNs life cycle as they will reproduce inside the insect. They can be seen globally for agricultural purposes as they are used as environmentally friendly insecticides. A classic example that literature on these insect assassins highlights is their contribution to biological control, as EPNs have been proven to be successful biological insecticides that substitute chemical insecticides. In addition, EPNs could be using niche partitioning to decrease competition and increase the survival rate of their species. Understanding EPNs' behavior plays an important role in addressing inadequacies in existing research on biodiversity, host preference/ animal behavior, and general environment factors.

We hypothesize that EPNs are co-occurring due to their preference to infect different host species in a manner that mitigates competition with one another. This research will follow an in-depth analysis of the abundance of nematodes in different sections of both chemotaxis assays. Three EPNs will be examined: *Steinernema affine*, *S. costaricense*, and *S. krausse*. All of them have been exposed to four different host species: *Acheta domestica*, *Galleria mellonella*, *Manduca sexta*, and *Tenebrio molitor*. They will be given 24 hours to express their attraction to a host.

DAQUAN GREENWELL, Saint Francis University

Short-Term Melatonin Injections Influences on Offensive Aggression in Female Siberian Hamsters (*Phodopus sungorus*)

Mentors: Amanda Han and Gregory Demas

Organisms have physiological adaptations to increase fitness due to seasonal environmental changes such as photoperiod, temperature, and availability of food. These adaptations are primarily influenced by the surrounding endocrine systems which can cause numerous changes such as metabolism, immune responses, and social behavior. Melatonin (MEL) is a hormone secreted by the pineal gland located inside the brain and plays a critical role as the endocrine signal in seasonal changes. Our lab's previous research shows that when Siberian Hamsters (*Phodopus sungorus*) are housed in short-day photoperiods, or given timed MEL injections mimicking short days, they display increased aggression common in the non-breeding season. This raises the question: will those same behavioral changes be exhibited in a shorter injection period? The design of the study consists of a short two-week period and also a longer ten-week period of timed injections amongst three groups of hamsters. Two of the three groups were housed in a long-day photoperiod and one group was housed in a short-day photoperiod. All three groups were given timed injections that consisted of MEL or saline (SAL) for two weeks. We used a resident intruder paradigm (RIP) to measure defensive aggression and used a novel environment paradigm (NEP) to measure offensive aggression. We then collected and weighed the female reproductive tissues to assess gonadal regression. Experimental data shows that there were no significant differences in the reproductive weights between the three groups. There were no significant differences in the latency to first attack, attack duration, or chase duration among the three groups. However, a significant difference in scent-marking behavior, with NEP displaying it more often than the RIP, all other behaviors were insignificant between the two paradigms. Collectively, this data suggest that two weeks of short days or melatonin treatment is insufficient to induce changes in reproduction and behavior. Ongoing experiments will examine animals after 10 weeks of manipulation. More broadly, these findings contribute to our knowledge of how environmental cues regulate endocrine processes, which in turn, cause phenotypic changes in organisms including behavior.

ANNA KELSON, Indiana University

Comparing Metrics of Reproductive Success Between Captive and Wild Dark-eyed Juncos

Mentors: Sarah Wanamaker, Dr. Ellen Ketterson

Environmental cues are essential triggers of reproduction in many organisms. In female songbirds, their social setting, available nesting sites, and food quality and quantity are known to dictate reproductive timing and decision making. High population density suppresses reproduction; however, there is debate regarding why this is. Some studies suggest that the suppressive effect of high population density is thought to be caused by per-pair food supplies decreasing, while others suggest that territorial aggression leads to low reproductive success. Songbirds notoriously struggle to reproduce in captivity, perhaps because it is impossible to perfectly recreate complex natural reproductive cues in captive settings. However, few direct comparisons have quantified the differences in captive versus wild metrics of reproductive success in passerines, and it is unclear precisely what hinders reproductive success in captive individuals. In this study we compared clutch size, hatch rate, and the proportion of nestlings that fledged the nest between captive and wild populations of non-migratory Dark-eyed Juncos across three breeding seasons and examined potential causes of differences in reproductive success. Captive birds ($n = 87$, mean = 3.13 ± 0.90) had smaller clutches than wild birds ($n = 115$, mean = 3.67 ± 0.81 , $p < .001$). Additionally, of nests that were not abandoned or predated prior to hatching, captive nests had lower hatch rates ($n = 51$, 0.72 ± 0.28) than wild nests ($n = 49$, mean = 0.89 ± 0.17 , $p < .001$). Captive nests that fledged at least one young ($n = 22$, mean = 0.69 ± 0.26) also fledged a smaller proportion of nestlings than wild nests ($n = 21$, mean = 0.93 ± 0.16 , $p = 0.001$). Despite abundant food supplies, which often enhance reproductive success in high-density populations, captive juncos fared worse than wild juncos at each stage of the nesting process.

PAIGE PHIPPS, Cal Poly Humboldt

Male mice behavior: Exploring the connection between vocal signaling and olfactory cues

Mentors: Sierra McAlister, Laura Hurley

Multiple sensory channels, (e.g. olfaction and audition), can influence behavior during opposite sex interactions. Male mice express vocal (ultrasonic vocalizations: USVs) and non-vocal behaviors toward females for communication. We aimed to understand the influence of female olfactory cues on male vocal and non-vocal behavior. Our behavior set up involved a cage with a divider that separated males from females and was used to control the presence of olfactory cues. The divider had a hole in the middle, which could be covered, that allowed direct contact between mice. A speaker was used to play back female rejection calls (squeaks). There were 4 experimental conditions in which the mice were exposed to 1) female soiled bedding with open hole, 2) no soiled bedding and covered hole, 3) no soiled bedding with open hole and 4) female soiled bedding with covered hole. We scored behaviors such as: digging, rearing, grooming, and investigating were measured in number and duration. The behaviors that showed changes across experimental groups were digging and rearing. Both the number of digging bouts and the total duration of digging were lower when the hole was exposed. In contrast, the total duration but not the number of rearing bouts was higher when the hole was exposed. The behaviors that didn't change across treatments were grooming and investigating. These results suggest that having direct contact with females is an important driver of male behavior, as opposed to olfactory cues alone.

Santonio Resonno Jr, Southern Illinois University-Carbondale
Frequency of Affiliative Behavior Among Non-Orphaned and Orphaned African
Savanna Elephants (*Loxodonta africana*)
Mentor: Daniella E. Chusyd

African savanna elephants (*Loxodonta africana*) live in families composed of related adult females and their offspring. Older elephants provide guidance for the younger elephants allowing them to receive proper socialization. Due to poaching, culls, and human-elephant conflict, many young elephants are orphaned and thus do not receive the social guidance from the species' traditional herd composition (i.e., grandmother, mother, older cousins, and siblings). Therefore, this study aimed to investigate whether orphaned elephants, compared to non-orphaned elephants, display a similar frequency of affiliative behaviors. Videos of orphaned and non-orphaned elephants were watched, and the behavior of the focal elephant was documented every five seconds. Affiliative behavior was defined as when the focal elephant rubbed their body against, rested their head on, intertwined their trunk with, or put their trunk to another elephant's mouth or body. The age class and sex of the focal elephant were documented and efforts were made to match the sex and age class of non-orphaned to orphaned elephants. Age classes were defined as infants (0-4 years), juveniles (5-8 years), and subadults (9-15 years). The amount of times the focal elephant displayed affiliative behavior would be divided by the time they were observed out of an hour leaving you with the rate of affiliative behavior. A total of 54 elephants were used, 35 of which were non-orphaned elephants and 19 as orphaned elephants. Non-orphaned elephants showed greater frequency of affiliative behavior per hour compared to orphaned elephants in this population ($P=0.006$). These results support the importance of living in a traditional herd to foster social behaviors.

Heat-induced stress reactions in Spadefoot Toads

Carlos Vaca Angus, Ellie Shell, Cris Ledon-Rettig

Climate change may have detrimental effects on mating behaviors and populations in amphibians. Climate instability has increased as a result of anthropogenic activity. Amphibians are sensitive to unstable climates as they are exothermic and rely on bodies of water to breed. Amphibia are most at risk of extinction and can be used as a keystone class. We evaluated trends in hormonal stress reactions using a waterborne assay across Spadefoot toads (genus *Spea*). Individuals were submerged in water separating them into two groups. Our experimental group was mostly kept at room temperature for analysis and experienced a heat-exposure condition for a short period. Our control condition was sustained at room temperature for the duration of the entire experiment. These conditions were evaluated at 3 timepoints. These timepoints reflected stress reactions upon first exposure to submersion: baseline. Heat or control exposure: temperature-dependent. And a submersion after a 2-hour period: recovery. We replicated the experiment after 8 days and had two trials. Our first trial had corticosterone elevated at baseline. Our second trial showed that the heat condition had elevated levels of corticosterone overall, but no significant differences between time points. The initial elevation seen in the first trial was likely a result of the toads experiencing stress related to being in an unfamiliar environment.

ANA SOFIA VELARDE CHONG, Cal Poly Humboldt
The Effect of Anthropogenic Activity & Variation in Body Condition on Maternal
Behavior in *Macaca fascicularis*
Mentors: Elizabeth Coggeshall, Michael Wasserman

Human driven environmental change has increased encounters with free roaming animals, which has resulted in anthropogenic presence and activity inadvertently affecting surrounding life. Long-tailed macaques (*Macaca fascicularis*), the focus of this study, are one of the most widely distributed primate species across the globe and consequentially experience high exposure to anthropogenic presence. This study aims to investigate the effect of anthropogenic activity and variation in body condition on maternal behavior in the long-tailed macaques. Data was collected in 2019 via video footage in the northeastern Amnat Charoen Province of Thailand within a Buddhist monastery forest which is home to ~1000 free-ranging macaques. Behavioral data was collected from 10-minute focal follows of mother-infant dyads. Body condition scores were determined using a novel body condition scale that was created based on this population and is scored from 1 to 5. We found that individuals who were scored as group 3 with optimum body condition, as well as individuals that were exposed to a medium range of anthropogenic activity had significantly higher time engaging in cling carriage, infant on nipple, proximity to infant, restraining infant, touching infant, and infant on ventrum behaviors. Our results lead us to believe that primates living in anthropogenic landscapes are susceptible to body condition changes. This can impact their behavior and physiology, subsequently impacting their offspring's behavior and physiology. This population and investigation offers insight into how individuals adapt in human centered environments and highlights the positive and negative behavioral and physiological tradeoffs that are made to survive. It is crucial to study the ways in which human populations are interacting with and affecting the biosphere in order to maintain stable populations and environments.

SYMPHONY WESTON, Truman State University

The Effects of Microbial Symbiotes on Entomopathogenic Nematodes.

Mentors: Morgan Familo, Farrah Bashey-Visser

Multiple species coexist and compete for resources with bacteria and their hosts in nature. The health and behavior of animals can be influenced by their symbiotic bacteria, which can be harmful or mutualistic. Depending on the genetics of the host and microbes, these relationships can change very quickly. One such organism is entomopathogenic nematodes, which have a co-evolved mutualistic relationship with the bacteria that live inside of them. These bacteria aid the nematodes in all aspects of life - competitively, nutritionally, behaviorally, and reproductively. The competition between other nematodes and bacteria strains occurs inside of their insect host. Our experiment looked at how much the bacteria, *Xenorhabdus*, affected the nutrition and behaviors of the nematodes, *Steinernema*, by placing different strains of nematodes and bacteria in the same environment (plate) together. As past literature suggests, the nutritional specificity between these nematode-bacteria pairs could allow them to exploit their host environment without aiding competing nematode-bacteria pairs. This was analyzed on a series of Lipid Agar plates acting as a nutrient-deficient environment for the nematodes. We observed how entomopathogenic nematodes developed and behaved when encountering the bacteria of competing nematodes over the course of 10 days. Each day the plates were scored for developmental stage and any notable behavioral or physical changes. In total, 10 nematode stocks consisting of three species (*S. costaricense*, *S. kraussei*, and *S. affine*) were experimentally paired with 10 different strains of bacteria. We hypothesize that nematodes will do better on bacteria associated with their own species than of a different nematode species.