

# Project Descriptions

## 1. Evolutionary, Ecological and Comparative Genomics of Malaria Vectors

**Project Mentor: Nora Besansky**

**Introduction:** By examining patterns of genetic variation across species, populations, and different regions of the genome, inferences can be made about phylogenetic relationships, population structure and history, speciation and adaptive change. These themes run through various lines of investigation pursued in my laboratory, often in collaboration with biologists in Europe and Africa and computer scientists here at Notre Dame.

**Research Projects:** Ecological genomics of *Anopheles gambiae*. This African mosquito is subject to an ongoing speciation process driven by ecological adaptation to diverse environments, resulting in increased malaria transmission spatially and temporally. We combine a “top-down” approach to look for key phenotypic (ecological, behavioral and physiological) differences and a “bottom-up” approach to scan for sequence or gene expression changes likely to be causal to ecological and reproductive divergence. Malaria vector genomics. Of the ~500 anopheline species, only two dozen are important vectors of human malaria parasites. Why some anopheline species transmit malaria while others do not is incompletely understood. Developing a better understanding of ‘vectorial capacity’ may enable its eventual manipulation in order to reduce disease burden. Dr. Besansky is coordinating a major international collaboration whose objective is the complete genome sequencing of 13 anopheline mosquito genomes, representing 26 billion base pairs, to complement and facilitate comparative genomic analysis with the three other sequenced anophelines: *Anopheles gambiae* PEST, M and S forms. Mining these data by computational approaches will allow inferences about evolutionary changes relevant to vector ability, and will enable the development of new approaches to the control of vectors whose biology is poorly understood relative to genetic and evolutionary model systems such as *Drosophila*.

**What the Student will Learn:** The student will be exposed to mosquito ecology, behavior, physiology and genomics. The student will gain an appreciation of how computer science is an essential interface not only for big comparative genomics projects, but also for smaller ones that nevertheless require sophisticated computational skills to manipulate, explore and interpret increasingly large data sets produced by current high throughput genetic technology.

## 2. Neuropeptide Modulation of Vertebrate Behaviors

**Project Advisor: Sunny Boyd**

**Introduction:** The long-term objective of this research program is to identify the interactions among chemical messengers that control behaviors. Neuropeptides and steroid hormones alter a variety of vertebrate behaviors, including parental, aggressive, and reproductive behaviors. The mechanisms of action of these compounds and the site in the brain where they act on specific behaviors are poorly understood. We currently focus

on the neurohypophysical peptides which modulate the display of vocalizations in vertebrates. Vocal behavior is a critical component in social interactions of many species, including humans.

**Research Projects:** Student projects will be designed to provide experience at both the whole-animal behavior level and also at the cellular and molecular level of investigation. Students will thus (1) analyze the effects of peptides and steroids on animal behavior, (2) localize peptides, steroids and their receptors in the brain, and/or (3) sequence genes involved in the synthesis of these factors or their receptors.

**What the Student will Learn:** Students will learn a variety of basic techniques in behavioral neuroendocrinology, including: observation of animal behavior, survival surgeries, brain sectioning, neuroanatomy, immunocytochemistry, confocal microscopy, receptor binding techniques, PCR, Northern, and *in situ* hybridization. Through development of their own research projects, students will also learn literature analysis, experimental design, data analysis and data presentation.

### **3. Membrane Trafficking, Cytoskeletal Remodeling and Signal Integration**

**Project Advisor: Crislyn D'Souza Schorey**

**Overview:** The detachment of cancer cells from the surface of a tumor, signals the beginning of a critical phase in the spread of some of the most notorious cancers – including cancers of the colon, breast and prostate. For the onset and progression of malignancy, cells from the primary tumor must acquire the ability to become motile, i.e. to breakaway from associated cells and “invade” through the surrounding tissue. The acquisition of the “invasive phenotype” in tumor cells correlates with poor prognosis in cancer patients. Our laboratory is investigating cellular changes that lead to the acquisition of the invasive phenotype and consequently to the initiation and propagation of cancers. Ultimately, the goal is to understand the most significant changes that are responsible for tumor progression.

**Project:** The student will have the choice of working on a sub-project in one of two ongoing lines of investigations in the laboratory.

1) *Elucidating the mechanisms of early tumor development.* This project will make use a three dimensional cell culture models that allows cells to behave like their counterparts in epithelial glands. Using genetic manipulation as well as cell biological techniques, we have identified key components of the cellular machinery that potentially could regulate the assembly and disassembly of these structures. Thus, by inserting specific molecules in these cells we can monitor their effects on normal as well as cancer development. We specifically aim to examine these regulatory molecules for their ability to induce changes that resemble the early stages of tumor development.

2) *Elucidate mechanisms that promote tumor cell invasion.* Tumor cell invasion, a process that allows cancer cells to spread into surrounding tissues is in part dependent on structural changes that occur at the tumor cell surface. Using cell and animal model systems we are examining how tumor cells exhibit key structural changes at their migrating surface that allow them to become invasive.

**What the student will learn:** The student will learn a variety of state-of-the-art cell and molecular biological techniques. S/he will learn first hand to design experiments to address specific questions, and then how to acquire, analyze, interpret and record data. S/he will be part of a vibrant lab group consisting of post-doctoral fellows, graduate students, undergraduate students and technical staff.

#### **4. The Molecular Basis of the Mammalian Circadian Clock**

**Project Advisor: Giles Duffield**

Circadian ('about a day') rhythms are an integral component of biochemistry, physiology and behavior. Circadian clock biology is relevant to human health. Dysfunction of the circadian clock underlies several disease states, including Seasonal Affective Disorder, and sleep disorders. My lab is interested in elucidating the molecular basis of the circadian clock in mammals using a range of traditional and state of the art molecular, cellular and behavioral approaches.

The molecular circadian clock consists of an autoregulatory transcriptional-translational feedback loop composed of positive and negative regulators. Work over the last 8 years has identified 9 such components, but additional genes and modifiers are being identified. In addition, most tissues of the body harbor cell-autonomous circadian clocks. One such additional gene, identified in my laboratory using a cDNA microarray screen, is the transcriptional inhibitor Inhibitor of DNA-binding 2. It is rhythmically expressed in the master clock structure in the hypothalamic brain known as the suprachiasmatic nucleus (SCN), and throughout the body in various peripheral tissues (e.g. heart and liver). Current studies are to evaluate the role of genes such as *Id2* in the organization of the central oscillator, and to identify novel molecules relevant to cellular clock function. We are also interested in understanding how light resets the molecular clock (input), and how the clock regulates down-stream clock-controlled genes (i.e. output, hands of the clock).

My lab is using continuous activity monitoring to identify behavioral phenotypes in transgenic mouse models (e.g. *Id2* knockout mice) that are maintained under a variety of photocycle conditions, and exposed to artificial time-zone changes and acute light/pharmacologic/behavioral treatments. Results thus far have revealed that in the absence of the *Id2* gene, mice adapt to large time-zone changes (e.g. mimicking a flight from Berlin to Los Angeles) more rapidly than wildtype individuals. We are also using real-time monitoring of clock gene expression in tissues and cells derived from transgenic mice that express Firefly luciferase in a rhythmic manner. We are using DNA microarray and real-time quantitative RT-PCR analyses to identify and characterize clock regulated genes, tissue culture of immortalized fibroblasts that exhibit circadian oscillations in gene expression as a model of the *in vivo* rodent circadian clock, and traditional neuroanatomical techniques (e.g. *in situ* hybridization, immunohistochemistry, neuronal track tracing) to characterize clock gene function in the brain. These studies have been supported by the Royal Society, the Wellcome Trust and the NIMH.

## **5. Subzero Temperature Adaptations Antifreeze Proteins**

**Project Advisor: John G. Duman**

**Introduction:** My research concerns the physiological and biochemical adaptations of poikilothermic organisms to subzero temperature. Most studies are with insects and plants, but other organisms (including spiders and other terrestrial invertebrates, fungi and bacteria) are also under investigation. These organisms adapt to subzero winter temperatures by either becoming freeze tolerant (able to survive extracellular freezing) or freeze avoiding. Freeze avoiding species generally produce antifreezes, such as polyols (glycerol, etc) and/or antifreeze proteins. We have been particularly concerned with the antifreeze proteins and our studies have ranged from investigations of the function of the proteins, to hormonal and environmental cues controlling their production, to protein chemistry and molecular biology designed to determine the structure - antifreeze function relationships of the proteins. Studies with freeze tolerant organisms have concentrated on ice nucleating proteins which function to induce ice formation in the extracellular fluid at high subzero temperature, and also on antifreeze proteins. In addition to functioning as antifreezes in freeze avoiding organisms, antifreeze proteins appear to function in certain freeze tolerant organisms as cryoprotectants to inhibit the damage resulting from freezing of body water. The mechanism of this process is under investigation.

**Research Projects:** Current ongoing research includes: (1) structure/function relationships of insect, plant and bacterial antifreeze proteins; (2) the cloning and expression of antifreeze protein genes; (3) potential cryoprotection mechanisms of antifreeze proteins; (4) applied studies on the potential uses of antifreeze proteins in agriculture (i.e., transgenic plants which produce insect antifreeze proteins) and for the cryopreservation of biological materials; and (5) studies of antifreeze proteins in Alaskan insects. However, within the broad theme of low temperature adaptations a wide variety of student projects may be accommodated. These may range from physiological ecology to protein biochemistry.

**What the Student will Learn:** Depending on the project students will learn protein purification and characterization, including protein/protein interactions; low temperature physiology and biochemistry; cryoprotective techniques; gene cloning, sequencing, microarray, and other molecular techniques.

## **6. Genetics and Genomics of Drug Resistance and Virulence in the Malaria Parasite**

**Project Advisor: Michael Ferdig**

Malaria is flourishing in the form of drug-resistant parasites and insecticide-resistant mosquitoes. The complexity of Plasmodium parasites' life cycle and biology renders them elusive to drugs and vaccines. Currently, 40% of the global population is at risk for the disease. The nearly completed P. falciparum genome sequence, along with integrated, analytical tools, offers fresh hope for gene discovery and identification of novel control strategies. My lab is using methods to overlay critical biological processes on whole-genome data to bridge the gap between critical phenotypes, like drug resistance and virulence, and their underlying gene mutations, with the long-range goal of elucidating

new avenues of malaria intervention. We are focused on identifying genes that confer complex *P. falciparum* traits, specifically, susceptibility to antimalarial compounds and parasite proliferation in red blood cells (RBC). To do this, we study inheritance patterns of precisely measured phenotypes and high-resolution microsatellite markers to identify the genetic regions carrying genes that direct these traits' expressions. Such quantitative trait loci (QTL) profiles act as "biological filters" of massive sequencing and transcriptional databases emerging from the genome project. In this way a biological framework can be imposed on the data to narrow the search window and to pinpoint specific genes, gene interactions, pathways and transcriptional networks that drive drug responses and parasite growth.

## **7. Baculovirus recombinants capable of simultaneous expression of multiple gene products** **Project Advisor: Malcolm Fraser**

**Introduction:** The insect pathogenic Baculoviruses have been adapted for high efficiency recombinant protein production in infected insect cells and larvae. Multi-gene protein complexes such as antibodies or human pathogenic virus structures can be produced by co-infecting multiple Baculovirus recombinants, but increased efficiency of multi-gene products could be attained if all genes were expressed from a single baculovirus recombinant. We are constructing a multi-gene Baculovirus expression vectors that could be used for efficient expression of multi-gene products by adapting several non-essential regions of the virus to recombinant protein expression.

**Research Projects:** The student will begin by constructing and expanding novel Baculovirus shuttle vectors containing the gene of interest. These vectors will be used for transfection of insect cells in culture, along with Baculovirus DNAs, to generate recombinant Baculoviruses. Recombinants will be identified using fluorescent protein markers and cloned. Southern and PCR analyses will confirm the structure of the desired gene in the recombinant virus. Recombinant-infected cells will be analyzed for the expressed protein products.

**What the Student will Learn:** The student will gain experience in cloning and characterization of recombinant DNA molecules, working with animal viruses in cell culture, and the genetic and molecular characterization of DNA viruses. Subsequent analyses of protein products produced by the generated recombinants will complete the training. This will provide a useful survey of recombinant DNA and protein technologies that are standard practice in molecular genetics laboratories.

## **8. Genetic expression in butterflies under climate change** **Project Advisor: Jessica Hellmann**

**Introduction:** The geographic range limits of many species are strongly affected by climate and are expected to change under global warming. For species that are able to track changing climate over broad geographic areas, we expect to see shifts in species' distributions toward the poles and away from the equator. A number of ecological and evolutionary factors, however, could restrict this shifting process. These factors include

habitat loss and fragmentation, dispersal limitation, and our particular interest, local adaptation. We study whether populations at the northern edge of a species' range are locally adapted to the climates that occur there or if they prefer climates characteristic of the range core. These two scenarios could lead to very different outcomes under climate change with possible declines in the former and likely range shifts in the later. We pursue our research using two butterfly species that reach their northern range limit on Vancouver Island, British Columbia.

**Research Projects:** The student will play a key role in rearing caterpillars of our two focal butterfly species in growth chambers. These individuals are collected in the field and then exposed to a variety of conditions at Notre Dame. We will sample caterpillars at a variety of stages of development and extract their mRNA to determine which genes are expressed during larval development. These expressed proteins then will be sequenced, ultimately to generate microarrays for use in subsequent climate experiments.

**What students will learn:** The student will gain an appreciation for organismal biology and genetic techniques. The student will learn how to operate environmental chambers and care for living organisms. The student also will interact with graduate students, providing an opportunity to see she/he wishes to pursue graduate study.

## **9. The Genetic and Developmental Basis of Divergence in *Drosophila*** **Project Advisor: Hope Hollocher**

**Introduction:** My lab studies all aspects of speciation, everything from the evolution of reproductive incompatibilities to morphological differentiation. We are particularly interested in determining the genetic and developmental changes responsible for differences we observe between species to reveal general patterns underlying speciation processes. Are certain traits more prone to change during speciation than others? If so, why? Do differences between species mostly arise from changes in regulatory sequences? These are a few of the many questions we aim to answer in our research.

**Research Projects:** We have two ongoing research projects in the lab. The first focuses on the evolution of hybrid incompatibilities in African *Drosophila* (specifically, *Drosophila melanogaster* and its sister species). Using the arsenal of genetic tools available in *D. melanogaster*, we are working to identify which genes are involved in disrupting germ line development in hybrids. In addition, we have started to use microarrays to assay gene expression profiles of pure species and hybrids to investigate how the genetic cascades affecting germ line development are altered during speciation. The second project focuses on the evolution of abdominal pigmentation between different species of the *Drosophila cardini* group, which inhabit the Caribbean Islands and nearby mainland of Central and South America. Here, we are analyzing patterns of sequence variation from multiple genes involved in pigmentation to determine how these patterns correspond to changes in the developmental control of melanin synthesis and deposition in the different species.

***What the Student Will Learn:*** Students will have the opportunity to learn a variety of molecular techniques (e.g. DNA extraction, DNA amplification, cloning, and sequencing) as well as population genetic analyses used to interpret these molecular data. In addition, students will be introduced to *Drosophila* identification, dissection, tissue preparation, antibody staining and microscopy used in developmental research. Students are also instructed on how to keep a laboratory notebook, troubleshoot, and design experiments to test specific hypotheses. All students are encouraged to read and discuss the primary literature associated with their research and are given the flexibility to pursue their own lines of inquiry directed toward the overall research goals of the laboratory.

## **10. Molecular Genetic Analysis of Zebrafish Eye Development and Retinal Regeneration**

**Project Advisor: David R. Hyde**

***Introduction:*** My lab studies the mechanisms involved in eye development (both retina and lens) in zebrafish. Eye development proceeds very quickly, with a functional eye (based on behavior and electrophysiology) present within 72 hours after fertilization of the egg. Because the embryo develops external to the mother and the embryo is translucent, it is relatively straightforward to follow many of the developmental events using non-invasive microscopy. The advanced state of the zebrafish genome project, the ability to reduce the expression of embryonic genes using morpholinos and RNAi, and the panel of antibodies that my lab has generated, it is relatively straightforward to observe and perturb in precise ways the development of the functional eye. Using microarray experiments, we have identified a number of interesting genes that are candidates for being essential in lens and retinal development. We have also developed methods to kill specific neuronal populations in the retina and observe their regeneration. Microarray experiments have identified that a number of the genes differentially expressed in neuronal regeneration correspond to genes that are likely to be important in early retinal development.

***Research Project:*** Regardless of the project that the student selects, the student and I will devise experiments that incorporate molecular and genetic techniques to examine one or two genes. This analysis may include cloning a candidate gene or promoter, DNA sequencing, in situ hybridization to examine the expression pattern of the candidate gene and morpholinos to knock-down the expression of the candidate protein. The student will then use a combination of histology, immunohistochemistry, and in situ hybridization to examine the effects that result from the loss of the candidate protein. We will then examine the potential role of the candidate protein in a biological process (such as eye development or retinal regeneration) by studying the process in wild-type and morpholino-injected embryos, which will again require histology, immunohistochemistry, and in situ hybridization. Thus, the student will gain an appreciation of the relationship between gene-protein-function-role in a biological process.

***What the Student Will Learn:*** The student will be exposed to modern techniques in molecular genetics, including the generation and analysis of transgenic zebrafish, generation and analysis of transient mutants using RNAi and morpholinos, cloning of

genes and analysis of their promoters, immunohistochemistry to examine cell-specific expression of proteins. They will be exposed to experimental design and the use of stringent controls, interpretation of data, and assembling the data into a presentation format (manuscripts or meeting posters). The students will also have the opportunity to see how their molecular genetic analysis relates to the physiological and cell biological studies that are being performed by graduate students and postdocs in the lab.

## **11. Modulatory Effect of Intrinsic and Environmental Cues on Circadian Rhythms of Visual Sensitivity**

**Project Advisor: Lei Li**

**Introduction:** One of our research projects is to study intrinsic and environmental cues that play a role in the regulation of circadian rhythms of visual system functions. Like humans, the behavioral visual sensitivity of zebrafish (our study model) fluctuates between day and night. They are most sensitive in the late afternoon and least sensitive in the early morning. A number of factors, such as daily light-dark cycles, vitreal dopamine and the input from olfactory bulbs may affect the circadian rhythms of visual sensitivity. We are interested in understanding how that happens.

**Research Project:** To investigate mechanisms underlying multiphasic opsin gene expression in rod photoreceptor cells in transgenic zebrafish.

**What the Student Will Learn:** You will be exposed to modern bio-techniques, such as time lapse imaging, immunochemistry and molecular biology. You will learn how to design and perform experiments, how to interpret the data, and how to assemble the data into a presentation format, i.e., research articles or meeting posters.

## **12. Utilization and Metabolism of Vitamin A in Insects**

**Project Advisor: Joseph E. O'Tousa**

**Introduction:** Most animals cannot synthesize Vitamin A and so must obtain it directly or indirectly from plant sources. In insects, vitamin A is used only in vision where it serves the role of the chromophore for all the visual pigments. However, different chemical forms of retinal are used by different insects and there are dramatic differences in the ability of different species to store these compounds. This project will define the chemical composition of the retinal stored in four representative insects from different orders by quantitative HPLC, and use genetic and genome information to determine how specific genes are involved in vitamin A utilization.

**Research Projects:** The student will execute one or more of the following projects: 1. Extraction and HPLC analysis of retinoid and carotenoid compounds from different tissues of the following insects: house fly, honey bee, beetle, moth. 2. Test the ability of specific Vitamin A derivatives to act in formation of visual pigments in the *Drosophila* model. 3. Identify the *Drosophila* transporters and retinoid binding proteins responsible for movement of various forms of vitamin A to the retina. 4. Characterize the

relationships the vitamin A transporters from other insects using genomic sequence information.

***What the Student will Learn:*** The student will produce novel information on the process by which vitamin compounds are extracted from diet and used productively by different animals. They will gain an appreciation of the benefits associated with the use of *Drosophila melanogaster* genetic and molecular analyses in addressing current questions in basic insect biology. Finally they will see the rationale for, and insights to be gained from, comparative analysis of genomic information.

### **13. Identifying signaling pathways involved in ErbB2-mediated protection from detachment-induced cell death**

**Project Mentor: Zachary T. Schafer**

***Introduction:*** It has recently been determined that mammary epithelial cells must inhibit classical cell death (apoptosis) *and* rectify metabolic defects in order to survive detachment from the extracellular matrix. However, oncogenes like ErbB2 will permit the survival of detached epithelial cells. ErbB2 affects numerous signaling pathways that regulate both apoptosis and metabolism, but it is not yet known which pathways are critical for the ability of ErbB2 to permit the survival of detached cells. Unmasking the survival signaling pathways that operate downstream of ErbB2 could potentially reveal novel targets or strategies used to selectively eliminate detached cancer cells.

***Research Projects:*** In order to address this question, a collaboration between my laboratory and the laboratory of Amanda B. Hummon in the Department of Chemistry and Biochemistry has been initiated. The Hummon laboratory has substantial expertise in RNAi based screening approaches and my laboratory is proficient in conducting cell biological approaches to understand cell death and metabolism. Thus, an siRNA based screen (using Qiagen siRNA libraries) of detached ErbB2 expressing mammary epithelial cells will be initiated in the Hummon lab to identify genes that are critical for the ErbB2 based rescue of apoptosis (caspase glo assay) and metabolism (ATP assay) Candidate genes will then be validated in the MCF-10A 3D cell culture assay and in soft agar transformation assays in my lab. We will be looking for candidate genes that may be involved solely in either the apoptotic regulation or the metabolic regulation and for “master regulators” that may affect both apoptosis and metabolism.

***What the Student will Learn:*** The student will learn a variety of cutting edge techniques in both my lab and in the Hummon lab. Working in the Hummon lab will expose students to techniques in global molecular profiling and will allow students to gain experience in acquiring and analyzing large quantities of data. Working in my lab will afford students the opportunity to learn the mammary 3D cell culture system and will allow students to become familiar with a wide variety of techniques to study the cell biology of cancer.

## 14. Mycobacteria

**Project Advisor: Jeffrey Schorey**

Mycobacteria have a long history as pathogenic organisms and are the etiological agents of such well known diseases as tuberculosis and leprosy. Tuberculosis is a particularly deadly disease accounting for over 2 million deaths annually and is the 2nd leading cause of death due to an infectious organism. A further concern in recent years has been the dramatic increase in the number of individuals infected with multi-drug resistant strains of *Mycobacterium tuberculosis*. Other pathogenic mycobacteria include *M. avium*, one of the most common opportunistic pathogens in AIDS patients within the United States and *M. Leprae*, the causative agent of leprosy.

My lab focuses on the interaction between mycobacteria and its' host cell the macrophage. As intracellular pathogens, mycobacteria require invasion of macrophages for their survival. However, macrophages, which function as part of the innate immune system, also serve an essential role in controlling a mycobacterial infection. Interestingly, macrophages infected with pathogenic, relative to non-pathogenic mycobacteria, show limited production of inflammatory mediators (i.e. cytokines, chemokines, nitric oxide, etc.) which are required to control bacterial growth. However, the molecular mechanisms responsible for this difference in macrophage response is not well defined. Our studies have identified a number of macrophage-signaling pathways activated upon mycobacterial invasion including the mitogen activated protein kinases and have shown that production of inflammatory mediators are dependent on the activation of these pathways. Moreover, we have determined that macrophages infected with pathogenic *M. avium* and *M. tuberculosis* strains show only limited activation of these signaling systems. Studies are ongoing to further characterize the macrophage signaling molecules activated upon mycobacterial infection, how these responses differ upon infection with pathogenic and non-pathogenic mycobacteria and to characterize the mycobacterial components which initiate or inhibit these macrophage signals. We are particularly interested in studying the importance of glycopeptidolipids (a major surface component of *M. avium*) in modulating macrophage-signaling responses and in mycobacterial pathogenesis.

## 15. Genetic Models in *Drosophila* for Human Disease

**Project Advisor: Robert Schulz**

My research involves using the fruit fly *Drosophila melanogaster* to generate and study genetic models for human disease, especially as they relate to abnormalities in heart, blood cell, and muscle development. *Drosophila* is a superb model organism for these investigations because of its short generation time, the numerous experimental approaches available, the known DNA sequence of its genome, and the wealth of genetic and information resources on hand and emerging. Studying problems in *Drosophila* development has clear ramifications for our understanding of human development and disease due to the substantial conservation of genes between the two species. That is, greater than 60% of all *Drosophila* genes have homologues in humans, including many genes known to be causal of or associated with specific diseases. Research projects

ongoing in my lab include identification and analysis of (1) genes required for heart formation, relevant to the study of congenital heart disease in humans, (2) genes controlling blood cell development, with relevance to our understanding of human leukemias, and (3) signaling pathways controlling indirect flight muscle formation and function, relevant to the understanding of certain human muscular dystrophies. Genetic, developmental, and cell biological approaches are used in these analyses.

## **16. Molecular genetics of mosquito disease vectors**

**Project Mentor: David Severson**

**Introduction:** My lab employs genetic and genomics approaches to investigate the molecular basis for a variety of phenotypes observed in mosquito species. Our primary organism is the yellow fever and dengue virus vector mosquito *Aedes aegypti*. We have multiple projects involving this mosquito that include: 1) enhancing the status of the *A. aegypti* genome project; 2) investigating the molecular and cellular factors determining susceptibility to infection by dengue virus; 3) evaluating population genetics in disease endemic field sites that include Trinidad, Haiti, and India; and 4) examining the molecular biology of other important phenotypes such as autogeny, development, and meiotic drive. We are also investigating the molecular genetics of reproductive diapause in the West Nile virus vector mosquito *Culex pipiens*.

**Research Project:** Students will work with me to select a project that best suits their interests relative to research efforts we have ongoing at the time. For example, this may involve advanced characterization of individual genes, development and testing of constructs for gene silencing and subsequent phenotype evaluation, participation in whole transcriptome microarray analyses or NextGen sequencing efforts, or field population assays.

**What the Student Will Learn:** The student will employ the scientific method to further our understanding of some aspect of mosquito molecular genetics. This will include experimental design, hypothesis testing, data analysis, and data interpretation. In addition, the student will be exposed to a variety of contemporary molecular techniques as well as the opportunity to be involved in laboratory procedures for rearing large numbers of mosquitoes. The student will gain an appreciation and understanding of the relationship between molecular and whole organismal biology.

## **17. Regulation of Microtubule-Based Membrane Transport**

**Project Advisor: Kevin T. Vaughan**

**Introduction:** The movement of membranes, chromosomes and organelles in the cell is deliberate and highly orchestrated. Most of this movement is mediated by a novel class of "molecular motor" proteins which bind to these cargo types and carry them to their destination along cytoskeletal filaments. At the end of transport, the motors release the cargo and reset for a new round of transport. The precise delivery of membranes and chromosomes to the correct destination at the correct time is tightly regulated. The interests of the laboratory are centered on how motors identify the correct cargo, and how motor function is regulated by phosphorylation. The motor protein we are focused on

currently is the microtubule-based motor cytoplasmic dynein. This motor is responsible for many aspects of membrane transport and chromosome segregation during mitosis. We are working currently on several cytoplasmic dynein and dynactin subunits whose functions are regulated by phosphorylation. We are also working on how the dynactin complex functions as a dynein receptor on chromosomes and organelles. Recently, we delineated the chain of events which lead to motor protein loading and the onset of motility. Our lab integrates the use of conventional cell biology approaches (advanced quantitative imaging, biochemistry, DNA cloning and mutant analysis and live cell analysis) with advanced proteomic methods (mass spectrometry), functional genomics (intelligent design chimeras) and kinomics (signal transduction complexity). These cutting edge approaches provide students with training in multi-dimensional dissection of complex and highly integrated systems.

**Research Project:** Students can pursue one of several projects which include: 1) analysis of protein kinase function by drug treatment and analysis of proteins with mutated phosphorylation sites, 2) live-cell imaging analysis of cells lines expressing GFP-tagged cell surface adhesion molecules.

**What the student will learn:** Each project will incorporate a mixture of advanced molecular biology (RT-PCR, cloning, etc.), immunocytochemistry and immunofluorescence microscopy, live-cell imaging, biochemistry of culture cell lines, and sophisticated software analysis. The particular project will be tailored to the interests and goals of the student.

## **18. Comparative Analysis of Photoreceptor Cell Types in the Compound Eyes of Insects**

**Project Advisor: Michelle Whaley**

**Introduction:** Analysis of the genome content of many different organisms now allows comparative studies into the expression of genes in particular tissues and cell types. Members of the rhodopsin family of visual pigments are expressed in subsets of photoreceptor cells to tune the photoreceptor to colors of light. Analysis of this gene family in many insects, including ants, butterflies, fruit flies and mosquitoes has shown a wide scope of the type and number of these genes in different genomes. In this project we seek to understand the cellular architecture of the mosquito compound eye relative to the expression of these individual genes.

**Research Projects:** The student will play a key role in one or more of the following projects: 1. Gene cloning work to prepare gene expression constructs to allow visual pigments found in mosquitoes to be expressed and characterized in *Drosophila melanogaster*, 2. *Drosophila* genetic crosses and mating schemes to characterize transgenic strains and place transgenes in required genetic backgrounds, 3. histological examination of the retinal organization in mosquitoes, providing description of the cellular anatomy and the cells expressing particular visual pigments.

**What the Student will Learn:** The student will gain an appreciation of the similarities and differences in genome structures of different insect species, and how these are

manifested in differences in the development and organization of the insect retina. The student will be exposed to hypotheses on how these adaptations benefit the behavior and life cycle strategies of various species. The student will also learn genetic and molecular analysis in the widely used model organism *Drosophila melanogaster*.