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Development and Evolution

Program/Abstract #469
Functional genetic and comparative genomic analysis of vector mosquito development

Molly Duman-Scheel[a], Ellen Flannery[b], Susanta K. Behura[b], Morgan Haugen[c], Anthony Clemons[b], David W. Severson[b]
[a]*Indiana Univ. Sch of Med-South Bend At Notre Dame Medical & Molecular Genetics, South Bend, IN, USA*
[b]*University of Notre Dame, South Bend, IN, USA*
[c]*South Bend, IN, USA*

Although vector mosquitoes transmit deadly diseases, little is known about their development. Analysis of mosquito developmental genetics will advance the study of insect evolutionary development and may reveal novel vector control strategies. We recently described methods for functional analysis of embryonic genes in *Aedes aegypti*, the dengue fever vector and an emerging arthropod model. Although it was anticipated that Drosophila developmental genes and their functions would be generally well conserved in mosquitoes, our analyses are revealing striking differences. For example, knockdown of axon guidance genes in *A. aegypti* embryos indicates that nerve cord development has diverged in insects. We are now examining development of the *A. aegypti* olfactory system and salivary gland, two tissues of vector importance. Large-scale comparative analysis of developmental genes in *D. melanogaster* and three vector mosquito genomes suggests that genetic regulation of the development of these and other tissues has diverged. Lineage specific duplication, expansion, and loss of genes critical for Drosophila development were observed. Sequence analyses suggest that some processes, including salivary gland development, may be under more stringent selection forces than others. Comparative analysis of predicted miRNA binding sites suggests that the repertoire of developmental genes targeted by miRNAs is species-specific. Our comparative analyses are providing insight into the evolution of insect developmental genes and processes and will promote the design of additional functional genetic studies in mosquitoes.

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Program/Abstract #470
The evolution of a regulatory linkage mediating sexually dimorphic trait development

John C. Butts[a], Mark Rebeiz[b], Thomas Williams[c]
[a]*University of Dayton Biology, Dayton, OH, USA*
[b]*University of Pittsburgh, Pittsburgh, PA, USA*
[c]*University of Dayton, Dayton, OH, USA*

Traits culminate from the spatial and temporal expression of genes comprising a gene regulatory network (GRN). To trace GRN evolution, we studied the abdominal pigmentation of Sophophora fruit fly species where sexually dimorphic pigmentation, like that of *D. melanogaster*, evolved from a monomorphic ancestor. During the evolution of dimorphism, this pigmentation GRN was modified by the gain of dimorphic expression of the Bric-à-brac (Bab) transcription factors. In the derived dimorphic GRN, Bab represses the expression of the gene yellow whose encoded protein is required to make black pigments. The sexually dimorphic pattern of yellow expression in the *D. melanogaster* abdomen requires two cis-regulatory element (CRE) sequences, the wing element and body element. Our research asks two questions: does Bab form a direct regulatory linkage with CREs in the *D. melanogaster* yellow gene; and was this linkage (whether direct or indirect) an ancestral feature of the Sophophora pigmentation GRN? We found: that the wing element is required to repress yellow expression where Bab is present, and that Bab1 binds this CRE in vivo. Ongoing studies will determine which sequence(s) Bab1 directly binds. By comparison of orthologous yellow loci and transgenic analysis we found extensive divergence in non-coding sequences and CRE activity. Future studies will determine whether this divergence included the gain of Bab binding sites in dimorphic species or whether these binding sites were ancestral and conserved in the midst of otherwise dramatic sequence evolution.

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Program/Abstract #471
Doublesex expression is regulated by the Hox protein Abdominal-B

Wei Wang, Shun Yan, John Yoder
The University of Alabama, Tuscaloosa, AL, USA

Sexually dimorphic traits play prominent roles in animal behavior and evolution. Adult *Drosophila melanogaster* develop several sexually dimorphic traits including male-specific abdominal pigmentation and sex-specific abdominal segment number. Two principle transcription factors controlling these abdominal morphologies are the Hox protein Abdominal-B (Abd-B) and the sex-determination factor Doublesex (Dsx). These proteins function cooperatively to confer sex-specific regulation of downstream target genes such as bric a brac and wingless. However, little is known about the interaction between these key regulators. Recent evidence shows that Dsx expression is spatially dynamic throughout development. We therefore investigated Dsx expression during pupal development when Dsx and Abd-B function cooperatively to reduce segment number in male flies. We found that Dsx expression is highly dynamic in the fly abdomen during pupation with highest level in posterior segments, suggesting it may be an Abd-B target gene itself. Indeed, Dsx expression responds to alterations in Abd-B protein levels. Dsx expression is lost in Abd-B loss-of-function mutants and responds positively to elevated Abd-B protein levels. We hypothesize that Abd-B activates its necessary cofactor, Dsx, to create sexually dimorphic traits in the developing abdomen. These unex-