Program/Abstract #487
Molecular anatomy of the developing limb bud in the coqúi frog, *Eleutherodactylus coqui*
Joshua Gross[a], Ryan Kerney[b], James Hanken[c], Clifford Tabin[d]
[a]*University of Cincinnati, Cincinnati, OH, USA*
[b]*Halifax, NS, Canada*
[c]*Cambridge, MA, USA*
[d]*Boston, MA, USA*

The vertebrate limb demonstrates remarkable similarity in basic organization across phylogenetically disparate groups. To gain further insight into how this morphological similarity is maintained in different developmental contexts, we explored the molecular anatomy of size-reduced embryos of the direct-developing Puerto Rican coquí frog, *Eleutherodactylus coqui*. Coquí exhibits a basal anuran limb structure, with four toes on the forelimb and five toes on the hind limb. We investigated the extent to which coquí limb bud development conforms to the model of limb development derived from amniote studies. We characterized dynamic patterns of gene expression for 13 critical patterning genes across three principal stages of limb development. The expression patterns of most genes are essentially unchanged compared to amniote species. For example, an EcFgf8-expression domain is evident within the apical ectodermal ridge (AER). This likely defines a putatively functional AER signaling domain, despite the absence of a morphological ridge in coquí embryos. However, two genes, EcMeis1 and EcAlx4, demonstrate altered domains of expression, which imply a potential shift in gene function between coquí frogs and amniote model systems. Unexpectedly, several genes thought to be critical for limb patterning in other systems, including EcFgf4, EcWnt3a, EcWnt7a and EcGremlin, produce no evident expression pattern in the limb at the stages we analyzed. Thus, while this analysis substantiates the existence of a core set of ancient limb-patterning molecules, which likely mediate identical functions across diverse vertebrate forms, it also reveals plasticity in the genetic control of a conserved morphological pattern across evolutionary time.

doi:10.1016/j.ydbio.2011.05.549

Program/Abstract #488
Comparison of circadian gene expression in the eye and pronephros of *Xenopus laevis*: More like a mammal than a fish?
Matthew Redmann[a], Kristen Curran[b]
[a]*Appleton, WI, USA*
[b]*University of Wisconsin-Whitewater, Whitewater, WI, USA*

Failure of adult organs and tissues to synchronize to the local time leads to jet lag, increased risk of heart disease, and liver failure in mammals. We are interested in when during development embryonic organs can synchronize with the external environment. Eyes are directly entrained by light from the environment. In mammals, internal organs are synchronized to the external environment via the brain. Zebrafish organs are synchronized directly by light. Embryonic eyes of *X. laevis* exhibit a mature circadian rhythm at stage 41. We are currently comparing the time of day dependent expression of 3 circadian genes, xNocturnin, xBmal1, and xPRX2 in embryonic eyes and pronephri. xBmal1 is part of the central oscillator while xNocturnin and xPRX2 are acted upon by this oscillator. In order to elucidate the rhythmic expression of these genes we dissected eyes and pronephri at four times of day. We next evaluated the level of gene expression at each time point via quantitative Real Time PCR. We have found that genes like xNocturnin and xBmal1 show rhythmic expression in the eye. Rhythmic expression of xPRX2 in the eye is scarcely detectable. xBmal1 and xPRX2 show low levels of rhythmic expression in the pronephros when compared to the eye. Interestingly, the peak of xBmal1 and xPRX2 expression was 4–6 h later in the pronephros when compared to the eye. The delay in onset of peak expression observed in the pronephros suggests peripheral organs synchronize to the external light cues indirectly via the brain like adult mammals, instead of through direct detection of light cues as reported in zebrafish.

doi:10.1016/j.ydbio.2011.05.550

Program/Abstract #489
Role of Plakophilin-3, a desmosomal catenin, in *Xenopus laevis* development
William Munoz[a], Kyucheol Cho[b], Moonsup Lee[c], Hong Ji[c], Kris Vleminckx[d], Malgorzata Kloc[e], Pierre McCrea[c]
[a]*MD Anderson Cancer Center Biochemistry and Molecular Biology, Houston, TX, USA*
[b]*Salk Institute for Biological Studies, La Jolla, CA, USA*
[c]*MD Anderson Cancer Center, Houston, TX, USA*
[d]*Ghent University, Ghent, Belgium*
[e]*Methodist Hospital Research Institute, Houston, TX, USA*

Desmosomes contribute to the exchange of cell–cell signals, and the physical integrity of junctions and tissues. They are comprised of proteins such as transmembrane cadherin super-family members, and intracellular components including homologous catenins (plakophilins and plakoglobin), desmoplakin and intermediate filaments. Catenins possess a central Armadillo domain, bracketed by less conserved amino- and carboxy-terminal tails. The most prominent family member is beta-catenin, which acts in varying intracellular compartments, and is a key player in both normal development and human disease. This work instead focuses upon Plakophilin-3 (Pkp-3), a catenin that we hypothesize provides key functions in differing cellular contexts. For example, Pkp-3 is resident at both desmosomal junctions and in the cytosol, with our recent data further hinting at its function(s) in the nucleus. We will present our characterization of Pkp-3 in early vertebrate embryos of *Xenopus laevis*. This will include Pkp-3 temporal and spatial profiling, and knockdown phenotypes indicating its requirement in amphibian development. Effects include touch hyposensitivity, and reductions in certain neural tracts, cilia and ectodermal integrity. While Pkp-3 knock-out mice exhibit epithelial fragility, these further phenotypes were not reported. Our goal is to deepen the cellular and developmental understanding of Pkp-3, with future work focusing upon its poorly understood role(s) in the nucleus. This will provide the basis to ultimately address if Pkp3's role in gene regulation is in some manner linked to that at cell–cell junctions.

doi:10.1016/j.ydbio.2011.05.551

Program/Abstract #490
***Xenopus* germline nanos1 is translationally repressed by a novel structure-based mechanism**
Xueting Luo
University of Miami Cell Biology, Miami, FL, USA

The translational repressor Nanos is expressed in the germline and stem cell populations of jellyfish as well as humans. Surprisingly, we observed that unlike other mRNAs, synthetic nanos1 RNA translates very poorly if at all after injection into *Xenopus oocytes*. The current model of simple sequestration of nanos1 within germinal granules is insufficient to explain this observation and suggests that a second level of repression must be operating. We find that an RNA