generates all epithelial cell subsets that comprise a fully functional thymus. Canonical (β-catenin-mediated) Wnt signaling is involved in many developmental processes, including cell survival, proliferation, migration and polarity. It has also been implicated in thymus development, where recent evidence suggests a role in epithelial cell identity. In the current work we have used a conditional knockout of APC to address the role of Wnt signaling during thymus organogenesis in mice. Loss of APC leads to beta-catenin accumulation and target gene activation, thus mimicking pathway activation via ligand binding. We have knocked out APC, and therefore activated the canonical Wnt pathway, in all thymic epithelial cells from E11.5 of embryonic development. This resulted in a dramatic alteration in thymus structure during embryogenesis. Epithelial cells within the mutant thymus ceased proliferation, no longer formed the characteristic 3D network and showed an altered epithelial marker profile. The mutant thymus was also devoid of lymphocytes, failed to form a vascular network and was encased in a dense mesenchymal capsule. These characteristics are consistent with previous evidence that activation of Wnt signaling in TECs leads to their differentiation to an alternative epithelial cell fate, possibly that of keratinocytes, and we are working to explore this further. We are also interested to determine the molecular mechanisms that underlie this altered epithelial cell fate.

doi:10.1016/j.ydbio.2011.05.598

Program/Abstract # 185
Role of enteric neurons and smooth muscle in development of zebrafish intestinal motility
Kenneth Wallace, Gillian Roach, Amy Cameron
Clarkson University, Potsdam, NY, USA

Enteric neurons and Interstitial Cells of Cajal (ICC) play a major role in coordination of adult intestinal motility. In mammalian and zebrafish embryos, intestinal motility begins before a complete array of enteric neurons differentiate. In addition, contractility is present in mice and zebrafish embryos even when enteric neurons fail to differentiate. Early motility instead appears to depend more on intrinsic smooth muscle contractility. Changes in serotonin (5HT) modulate motility in the intestine with receptors on both neurons and smooth muscle. Previously, we demonstrated the development of 5HT secreting enteroendocrine cells in the posterior intestinal epithelium along with an anterior to posterior distribution of 5HT containing enteric neurons beginning late during 4 dpf with increases at 5 dpf. Presence of enteroendocrine cells increases posterior 5HT concentration six times higher than anterior as measured by differential pulse voltammetry (DPV). As observed by 5HT immunohistochemistry and DPV, we are able to alter concentrations of 5HT pharmacologically within the intestine. In 5 dpf zebrafish embryos, we have begun to determine the effects of pharmacological alteration of 5HT concentration on intestinal motility. We find that lowering 5HT concentrations changes motility in both anterior and posterior regions however, this response appears to have a neural component in the anterior intestine but not in the posterior.

doi:10.1016/j.ydbio.2011.05.599

Program/Abstract # 186
Augmentation of Smad-dependent BMP signaling in cranial neural crests causes craniosynostosis in mice
Yoshihiro Komatsu, Paul Yu, Nobuhiro Kamiya, Yuji Mishina
School of Dentistry, University of Michigan, Ann Arbor, MI, USA
Massachusetts General Hospital, Boston, MA, USA

Craniosynostosis is a clinical condition of facial deformity caused by a premature fusion of sutures in an infant skull. Here, we show that enhanced BMP signaling through BMP type Ia receptor in neural crests causes premature suture fusion in mice. Ectopic cartilage is found in the frontal suture prior to the fusion suggesting that augmented BMP signaling altered a fate of neural crest stem cells towards chondrocytes. Notably, this phenotype is rescued in a heterozygous null background of Bmpr1a. Phosphorylated SMAD1/5/8, which is higher in the mutant mice, is restored to endogenous levels in the rescued mice. In agreement with this, treatment of selective chemical inhibitor of BMP type I receptor results in a rescue of the craniosynostosis phenotype. These findings demonstrate that augmentation of Smad-dependent BMP signaling directly leads to premature fusion of cranial sutures and appropriate levels of BMP signaling are critical to govern the skull development regulating cell fate of neural crests.

doi:10.1016/j.ydbio.2011.05.601

Program/Abstract # 187
HMG2A is required in the neural crest cells of Xenopus laevis
Simone Macciò, Marco Onorati, Riccardo Sgarra, Gloria Rosi
Università di Pisa, Pisa, Italy

HMGAs are small DNA binding proteins that use conserved “AT-hook” motifs to interact with DNA to modify chromatin architecture and assist in gene expression. Two HMGAs, HMG1 and HMG2A, have been described in mammals, encoded by separate genes. These genes are highly expressed in proliferating and undifferentiated tissues during embryogenesis, but not in adult tissues, where they are re-activated in tumor progression. In Xenopus, only the hmg2a (Xhmga2) gene is described and localized transcripts are first detected at neurula stages, in the presumptive central nervous system (CNS) and eye field, as well as in the neural crest cell (NCC) presumptive territory. At later stages, Xhmga2 mRNA is detected in the CNS, in the otic vesicles, in migrating neural crest cells and their derivatives, in the notochord and in the medio-lateral mesoderm. We are currently addressing the possible role of Xhmga2 in NCCs. We have injected two different morpholinos targeting Xhmga2 transcripts in the anterior neural region and have observed that injected embryos have a severe disruption of the branchial arches, that are missing or greatly altered. Analysis of NCC molecular markers shows severe downregulation or absence of Xtwist, and Xdll4 expression at the tailbud stage. Extensive cell death occurs in the regions normally occupied by NCC injected embryos. Injection of control mismatched or standard morpholinos did not lead to similar alterations. These data suggest that Xhmga2 is required for NCC survival, possibly during the epithelial–mesenchymal transition and or migratory phase of NCC towards the branchial pouches.

doi:10.1016/j.ydbio.2011.05.600
The mechanism by which extracellular signals, like secreted Fibroblast Growth Factors (FGFs), are transduced intracellularly to control cell polarity and shape is not well understood. We study this process in the mechanosensory lateral line organ of zebrafish, a system composed of mechanosensory organs called neuromasts, positioned along the trunk of the fish. During development, the lateral line is formed by the lateral line primordium (plLp), a patterned group of ~100 cells that migrates along the trunk of the zebrafish. The plLp is organized into rosettes, each representing a proto-neuromast. Within each rosette, cells are polarized, with apical ends constricted and nuclei basally localized. New rosettes are generated by incorporation and organization of new cells into nascent rosettes in the leading edge, while mature proto-neuromasts are deposited from the trailing edge. Previous studies demonstrated that rosette renewal is dependent on FGF signaling, but the intracellular changes controlled by FGF are not known. We found that the Ras-MAPK pathway mediates intracellular transduction of the FGF signal. Using live imaging and 3-dimensional reconstruction, we have shown that Ras-MAPK signaling is required for cells in the leading edge of the plLp to apically constrict and form new rosettes. Furthermore, we show that Ras-MAPK signaling is upstream of Rho-kinase, which specifically activates apically localized myosin regulatory light chain (MRLC), a critical step for proper apical constriction. Based on this data, we propose Ras-MAPK signaling is downstream of FGF in the leading portion of the plLp and it is required for phosphorylation of MRLC through Rho-kinase.

doi:10.1016/j.ydbio.2011.05.602

Program/Abstract # 189
Hox cofactor MEIS1 plays essential roles in pulmonary airway smooth muscle patterning
Elizabeth Hines\textsuperscript{a}, Lan Yi\textsuperscript{b}, Xin Sun\textsuperscript{b}

\textsuperscript{a}University of Wisconsin-Madison Genetics, Madison, WI, USA
\textsuperscript{b}Laboratory of Genetics University of Wisconsin-Madison, Madison, WI, USA

Breathing is a complex physiological process that requires the coordinated function of numerous lung tissues. The main bronchi are surrounded by a precise juxtaposition of smooth muscle and cartilage that produce a balance of airway rigidity and elasticity, facilitating air passage to the lungs. The molecular mechanisms that control the patterning of upper airway mesenchyme are not well understood. Here we show that Meis1, a Hox cofactor, is required for the proper development of airway smooth muscle and cartilage. In mice, inactivation of Meis1 produces an increase in airway smooth muscle and a corresponding decrease in cartilage. Results from microarray analysis indicate that among other genes, components of the Wnt signaling pathway are down regulated in Meis1 mutant lungs. Current studies are investigating if MEIS1 primarily acts through regulation of WNT signaling during early lung development. Findings from this investigation will advance the current understanding of molecular mechanisms that drive the coordinated development of airway smooth muscle and cartilage.

doi:10.1016/j.ydbio.2011.05.603

Program/Abstract # 191
Mesenchymal nuclear factor I B regulates cell proliferation and epithelial differentiation during lung maturation
Yu-Chih Hsu\textsuperscript{a}, Christine Campbell\textsuperscript{b}, Cindy Bachurski\textsuperscript{c}, E. David Litwack\textsuperscript{d}, Jason Osinski\textsuperscript{e}, Dan Wang\textsuperscript{f}, Song Liu\textsuperscript{g}, Richard M. Gronostajski\textsuperscript{h}

\textsuperscript{a}University at Buffalo, Buffalo, NY, USA
\textsuperscript{b}Cincinnati Children's Hospital Research Foundation, Cincinnati, OH, USA
\textsuperscript{c}Office of Biorepositories and Biospecimen Research, Rockville, MD, USA
\textsuperscript{d}Buffalo, NY, USA
\textsuperscript{e}Roswell Park Cancer Institute, Buffalo, NY, USA
\textsuperscript{f}University At Buffalo (SUNY) Biochemistry, Buffalo, NY, USA

The nuclear factor I (NFI) transcription factor family consists of four genes (Nfia, Nfib, Nfic and Nfix) in vertebrates that regulate the development of the brain, lungs, muscle, and other organ systems. Nfib is expressed in both lung mesenchyme and epithelium and we showed previously that mice lacking Nfib have severe lung maturation defects and die at birth. Here we show that Nfib specifically in lung mesenchyme controls late epithelial and mesenchymal cell proliferation and differentiation. There is excessive cell proliferation in E18.5 Nfib\textsuperscript{-/-} lungs compared to wild type lungs and this increased proliferation is seen in both epithelial and

\textsuperscript{a}University of Wisconsin-Madison Genetics, Madison, WI, USA
\textsuperscript{b}Laboratory of Genetics University of Wisconsin-Madison, Madison, WI, USA

Program/Abstract # 190
A novel ENU-induced neonatal death mutant mouse: Characterization and identification of responsible mutant gene
Chun-Ta Ho\textsuperscript{a}, John T. Kung\textsuperscript{b}, Pei-Hsin Huang\textsuperscript{c}

\textsuperscript{a}National Taiwan Univ., College of Med Pathology, Grad Institute of Pathology, Taipei, Taiwan
\textsuperscript{b}Graduate Institute of Pathology, College of Medicine, National Taiwan University, Taipei, Taiwan

ENU mutant mice pedigree-131 (P131) presents neonatal death linked with short tail via a Mendelian autosomal-recessively inherited pattern, which serves as a mouse model for human hereditary neonatal death. We have established that the short tail phenotype in P131 mutants is attributed to spondylodental dystosis and coccygeal agenesis, and neonatal death in P131 mice was due to general hypoxia resulting from primary defective lung development. The candidate mutant gene responsible for P131 phenotype has been mapped to mouse chromosome 6 and is pinpointed to a locus presumably encoding F-box and leucine-rich repeat protein 14 (Fbxl14). P131 mutant harbors a single base mutation in fbxl14, resulting in a mutated protein in which Thr370 is replaced with Ala. We provided ultra-structural analysis of P131 lung, corroborating that relative lack of type I pneumocytes transdifferentiation is the primary pulmonary defect underlying collapsed lung and hypoxia. To definitively prove that Fbxl14 is the authentic mutant gene responsible for P131 phenotype, we have successfully generated fbxl14 -transgene in P131 background and have rescued the P131 mice from neonatal death and short tail. Both wild-type and mutant Fbxl14 could form SCF complex with Skp1 and Cul1; but the global short-lived protein degradation is not affected by over-expressing mutant Fbxl14, suggesting that only certain specific substrates recognized by SCF Fbxl14 could be affected in the ubiquitin-proteosome degradation pathway. Indeed, Snail1, as one of the candidate substrates of SCF Fbxl14, was degraded with a much slower kinetic in the presence of Fbxl14(Thr370Ala) via a proteosome-dependent process. In consistency with defective Snail1 degradation mediated by point-mutated Fbxl14, ENU-P131 neonates have significantly increased expression of Snail1 in the nucleus of pulmonary pneumocytes but not mesenchyme. Snail1, a well-established EMT mediator participating in neural crest migration during embryonic development, has never been reported to participate in the development of lung. How early Snail1 participates in mammalian lung developmental process remains to be investigated.

doi:10.1016/j.ydbio.2011.05.604