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-dimentional 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 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.

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Program/Abstract # 189
Hox cofactor MEIS1 plays essential roles in pulmonary airway smooth muscle patterning
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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.

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Program/Abstract # 190
A novel ENU-induced neonatal death mutant mouse: Characterization and identification of responsible mutant gene
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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 spondylocostal 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 Snail 1 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.

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Program/Abstract # 191
Mesenchymal nuclear factor I B regulates cell proliferation and epithelial differentiation during lung maturation
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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, musculature, 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−/− lungs compared to wild type lungs and this increased proliferation is seen in both epithelial and