mature endocrine cell numbers, as well as gradual loss of islet cell mass. Mafa and Arx, key islet cell gene products, were also found to be Isl1 transcriptional targets (Diabetes. 2009 Sep; 58(9):2059–69 and J Biol Chem. 2011 Mar 9). The LIM domains of Isl1 can function as protein interaction interfaces for other transcription factors and co-regulators. In this study, we investigated the impact of the LIM protein co-factor, Ldb1, in endocrine pancreas formation. Ldb1 expression was detected widely during early embryogenesis in the pancreatic buds and the surrounding mesenchyme, but later became highly enriched in islets and pancreatic ducts. Loss of Ldb1 in Pax6 (Le)-Cre;Ldb1fl/fl mice was found to cause an overt reduction in insulin+ and glucagon+ cell numbers by E18.5. The remaining insulin+ cells appear dysfunctional, as they lacked Mafa and Glut2 in beta cells. Ldb1-deficient animals also suffered from overt diabetes due to greatly reduced islet cell mass. Presently, we are examining by microarray and ChIP-Seq analysis the similarities between Ldb1 and Isl1 regulation of beta cells, with the expectation that differences may be found due to the expression Lmo2 and Lmo4 in the developing pancreas.

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Program/Abstract #463
Vangl2, aPKC and VAMP1; the interactions of polarity proteins with trafficking vesicle proteins in the Xenopus oocyte
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The Xenopus oocyte is a polarized cell with a distinct animal/vegetal axis. It contains the components of both PCP and apical/basal polarity systems, including Vangl2 and flamingo (planar cell polarity proteins) and aPKC, PAR3 and 6 (apical basal polarity proteins). To begin to address the relative roles of these systems in the oocyte, we examined the localization and function of Vangl2 and aPKC. We found that: 1. Vangl2 is enriched in radially arranged islands in the animal hemisphere. 2. Vangl2 co-localizes and interacts physically with VAMP1, a component of post-Golgi membrane trafficking vesicles. 3. The localization of animalized VAMP1 islands depends upon Vangl2 protein. 4. The arrangement of VAMP1/Vangl2 complexes depends on the stable acetylated microtubule cytoskeleton 5. aPKC also physically interacts with Vangl2 6. aPKC is required for the stability of the radially arranged microtubule cytoskeleton and the location of VAMP1/Vangl2 complexes. We conclude that both maternal aPKC and Vangl2 are essential for the polarity of the Xenopus oocyte.

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Program/Abstract #464
A novel role for a Cdc42 effector protein in Xenopus neurogenesis
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Many developmental events, such as axis determination and cell movement, are dependent on the asymmetrical distribution of proteins and RNAs. In Xenopus laevis, several maternal mRNAs essential for normal development are localized to the oocyte vegetal cortex. In this work we characterize a novel cortex-enriched transcript, cdc42 effector protein 4-like (cep4l). CEPs bind cdc42 and related small GTPases, which regulate many cellular functions. cep4l is expressed in the oocyte vegetal cortex and throughout embryonic development, including expression in migratory cells during gastrulation, neural crest in neurulae and tailbud stages, and neural regions in older embryos. Misexpressed cep4l RNA causes convergent extension defects and induces ectopic neuronal marker expression, indicating a role in neurogenesis. Experiments to identify upstream and downstream pathways indicate roles for FGF as well as cdc42. Co-expression studies with another neuronal inducer, FGFl, demonstrate an enhancement of cdc42 binding and ectopic neurogenesis. The effects of both cep4l and FGFl are independent of proliferation. We also present loss of function data showing a role for cep4l in normal axial and nervous system development, as well as a requirement for FGFl-induced neurogenesis. Although the roles of small GTPases in cell division, migration, and adhesion are well-characterized, our results suggest novel roles and pathways for these proteins and their effectors in neural fate patterning and neurogenesis.

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Program/Abstract #465
Expression in dorsal–lateral regions of Drosophila early embryos is supported by Grainyhead-mediated anti-repression
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The Drosophila pre-gastrula embryo is patterned by a nuclear gradient of the transcription factor Dorsal, which supports expression of genes in defined domains along the dorsal–ventral axis. Current models postulate that limiting amounts of Dorsal establish the dorsal boundaries of gene expression. In the case of the gene intermediate neuroblast definitive (ind) which is expressed in a dorsal–lateral stripe, in addition, EGFR signaling also supports ind expression. We have evidence suggesting repressors are necessary for the sharp dorsal border of ind. A synthetic enhancer analysis of the ind enhancer located a short 12 base pair repetitive sequence (“A-box”) that mediates transcriptional repression in dorsal regions of embryos (Stathopoulos & Levine, DevBio 2005). We found that this element alone is sufficient to mediate repression in dorsal regions, furthermore, when this element is mutated in the full-length enhancer expression is expanded dorsally. We identified proteins that bind this element using affinity chromatography and mass spectrometry. One of the factors we identified is grainy head (grh), a DNA-binding protein, which surprisingly we found acts as an activator to support expression of ind and has been shown to exhibit context-dependent activation that is influenced by MAPK signaling. We believe that grh acts to inhibit the repressor that sets the dorsal border of ind by competitively binding to the A-box element. Instead of limiting activators being responsible for establishing the dorsal boundary of ind, we propose instead that expression of this gene is supported by the grh activator that functions to limit the effects of repressors.

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Program/Abstract # 466
A high throughout sequencing-based screen for sea urchin skeletal patterning genes
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A high throughout sequencing-based screen for sea urchin skeletal patterning genes was performed. This work identifies the skeletal gene regulatory network using a high-throughput sequencing based screen for sea urchin skeletal patterning genes. The screen was performed in larvae of the sea urchin Strongylocentrotus purpuratus. The screen was designed to identify genes that are required for the development of specific skeletal structures, such as spines, spicules, and test. The screen was performed using a combination of RNA sequencing and in situ hybridization. The results of the screen were analyzed using bioinformatics tools, such as gene ontology analysis and pathway analysis. The results of the screen revealed a number of novel genes that are required for the development of specific skeletal structures. This work provides a valuable resource for understanding the genetic basis of sea urchin skeletal development.