Fibroblast growth factor (FGF) 8 is produced by a signaling center in the anterior telencephalon and is an important signal for patterning the mammalian cerebral cortex along the anterior/posterior axis. However, how FGF signaling is mediated during cortical development is still largely unknown. To identify the pathway of FGF signaling, we examined the activation status of Ras–ERK and PI3K/Akt pathways by immunohistochemistry. Strong staining of phosphorylated-ERK was detected in the mouse telencephalon at E10.5 in the specific area where FGF8 was expressed, while staining of phosphorylated-Akt was broader and dissimilar from that of FGF8. Misexpression of *Fgf8b* in mouse telencephalon at E10.5 caused upregulation of phosphorylated-ERK positive cells, indicating FGF8b is sufficient to activate Ras–ERK pathway. Overexpression of *Fgf8b* in the telencephalon also induced expression of candidate downstream genes of Ras–ERK pathway, such as *Sprouty2*, *Mkp3*, and *Pea3 Ets* family genes. Next we address the question of which FGF receptors mediate this signaling. To test the function of the receptors, we designed dominant negative forms of FGF receptors (dnFGFRs) which lacked a functional intracellular tyrosine kinase domain. Misexpression of one of the strongest candidates, dnFGFR3c, resulted in downregulation of Ras–ERK pathway at E10.5 and rearranged the patterning of cortex at P0, indicating that FGFR3c may be responsible for FGF signaling. These findings suggest that FGF8 secreted from signaling center regulates cortical gene expression via Ras–ERK pathway.

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Program/Abstract # 430
Characterization of glycine neurotransmitter activity during postnatal development of the rat retina
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Glycine is a major inhibitory neurotransmitter in the adult vertebrate retina. In order to better understand the glycinergic circuitry of retina, we studied the activity of the glycine transport system and the postsynaptic glycine receptor (GlyR) in the developing rat retina. The uptake of ^{3}H-glycine increased during the first 2 weeks of postnatal age, reaching maximum values at 12 days, then it decreased sharply to the adult values. Similarly, specific binding of ^{3}H-glycine to isolated retinal membranes increased from 5 to 15 postnatal days. Immunohistochemical studies were also addressed to determine the localisation of the amino acid, glycine transporter Glyt 1, and the α-subunit of the GlyR. Glycine displayed neuronal localisation. The expression of Glyt 1 and GlyR match with the biochemical studies. Immunoreactivity was mainly found at the inner layer of the retina, but migrating cells and external layer showed slight labeling. The occurrence of the glycinergic system at early postnatal ages before the presence of synapses and its correlation with the photoreceptor differentiation, support the hypothesis that glycine acts as a neurotrophic substance during retinal development.

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Program/Abstract # 431
Autonomic and sensory pancreatic nerves are differentially affected by large-scale β-cell turnover in the RIP-cmycER mouse
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The dynamics of pancreatic innervation during development and in disease are largely unexplored. Our goals were to perform a descriptive analysis of autonomic and sensory innervation in two physiological contexts: during pancreas organogenesis and maturation, and large-scale β-cell loss and repopulation in the adult. Immunohistochemistry and confocal microscopy were used to examine the autonomic and sensory innervation of the normal mouse pancreas during organogenesis (e9.5–18.5) and postnatal maturation (p0–21). Neuron populations begin to enter the embryonic pancreas between e11.5 and e14.5, and come into close apposition with α- and β-cells during late embryonic development and postnatal maturation. We also used a mouse model of β-cell regeneration in which tamoxifen administration conditionally induces widespread c-*myc*-mediated β-cell apoptosis. Repopulation of islets by β-cells occurs gradually over a two month period. We discovered that large-scale β-cell loss has cell-type-specific effects on pancreatic innervation.

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Program/Abstract # 432
Homocysteine enhances cardiac neural crest cell attachment in vitro by increasing intracellular calcium levels
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Elevated homocysteine (Hcys) increases the risk of neurocristopathies. Our previous studies show Hcys inhibits cardiac neural crest (NC) cell migration (NC cells from mid-otic to the third somite axial level) in vivo. However, the mechanisms responsible for this effect are not known. Here, we evaluated the effect of elevated Hcys on the reattachment

of cardiac NC cells onto various substrates in vitro and determined if any of the observed effects might be due to altered intracellular calcium signaling. We found Hcys enhanced cardiac NC cell attachment in a dose- and substrate-dependent manner within the 30-min period allotted for the reattachment of the NC cells. Ionomycin (increases cytoplasmic levels of calcium) mimicked Hcys' effect on NC cell attachment while BAPTA-AM (a chelator of cytoplasmic calcium) or U-73122 (a phospholipase C inhibitor), blocked Hcys' effect. Neither lanthanum chloride (a general plasma membrane calcium channel blocker) nor MK801+ (an NMDA receptor blocker expressed by NC) had any effect on NC cell attachment. These results showed Hcys rapidly alters NC attachment properties by triggering an increase in intracellular calcium possibly by increasing phospholipase C activity and generating inositol triphosphate through an unknown mechanism. Hence, the teratogenic effect ascribed to Hcys may be due, in part, to perturbation of normal intracellular calcium signaling during cardiac NC cell morphogenesis.

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Program/Abstract # 433
Morphogenesis of blood vessels during mouse vasculogenesis

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The formation of embryonic blood vessels and their morphogenesis into a highly stereotyped vascular network requires precise control of endothelial cell (EC) migration, proliferation, growth and patterning. Similar to neurons, ECs exhibit growth cone-like lamellipodia that grow and extend along molecularly defined tracks, integrating both attractive and repulsive cues present in the microenvironment. Recent studies provide evidence that molecules controlling neuronal guidance and pathfinding, also play an analogous role in EC migration and organization. Here, we have identified and characterized multiple 'neuronal' molecular guidance cues that drive endothelial vessel formation during mouse vasculogenesis. Our data reveals that a striking coordination of both positive and negative cues is required to build a blood vessel at a precise location. In addition, we identify the mouse notochord as a critical source of multiple redundant negative cues, which together shape the paired dorsal aortae. We then show that anastomosis of the aortae is driven by a combination of morphogenetic movements of the embryo, as well as repression of midline repulsive cues. Finally, we investigate mouse mutants that lack the notochord, and show that absence of proper midline cues results in dramatic failure of vascular patterning. A novel *in vitro* whole embryo culture system is used to functionally assay each individual repulsive molecule. Molecular identification of the cues that attract or repel EC is critical for our understanding of cardiovascular development and will provide the foundation for the development of clinical pro- and anti-angiogenic therapies.

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Program/Abstract # 434
Notch can regulate VEGF-related signaling in embryonic vascular differentiation

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The signaling cascades that direct the morphological differentiation of the vascular system during early embryogenesis are not well defined. Both Notch and VEGF signaling are known to play a role in the formation of the vasculature in the mouse. To further understand the role of Notch signaling during endothelial differentiation, we are using a binary transgenic mouse model that expresses an activated NOTCH1 intracellular domain in the embryonic vasculature. Several defects are seen in these transgenic embryos, which do not survive after E10.5. Most notably, the vasculature of the yolk sac displays differentiation defects, with few matured vessels. Microarray analysis of RNA isolated from the yolk sac of transgenic embryos indicated aberrant expression in a variety of genes. In particular, two VEGF family members, placental growth factor (PGF) and VEGF-C, are increased significantly. This data suggests a potential vascular differentiation pathway regulated by Notch signaling. Based on these findings, a transgenic model will be generated which expresses high levels PGF in the endothelia using the binary transgenic system. Morphological and genetic analysis of the resulting embryos will allow us to determine the relevance of the overexpression of PGF in the observed NOTCH1 overexpression phenotypes. Completion of this work will provide information on cell signals and gene expression processes directing endothelial differentiation.

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Program/Abstract # 435
Control of angiogenesis and lymphangiogenesis by ephrin-B2

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The efficient transport of gases, liquids, nutrients, macromolecules and cells between distant organs is indispensable for vertebrate organisms. Blood vessels and the lymphatic