Program/Abstract # 213
Drosophila apontic acts as a feedback inhibitor of JAK/STAT signaling and is required to limit an invasive cell population
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Gradients of signaling are used repeatedly in development to instruct cells to acquire different properties or behaviors dependent on the level of signaling. In the epithelium of the *Drosophila* ovary, high levels of JAK/STAT signaling specify a small number of cells to become the border cells, which must undergo a stereotyped migration. Cells with lower levels of STAT activation remain within the epithelial layer. How this gradient of signaling is translated into a binary decision to migrate or not was unclear. We found that the *apontic* (*apt*) gene is required for this process. In *apt* mutants, the migratory population was expanded and cells were unable to detach efficiently from the epithelium. This phenotype resembled gain-of-function of JAK/STAT activity. Gain of function of APT also mimicked loss of function of STAT and its key downstream target, SLBO. APT expression was induced by STAT, which was further supported by DNA binding assays. Together, this suggests that APT functions as a feedback inhibitor of the STAT pathway. We conclude that a regulatory circuit between STAT, APT, and SLBO functions to convert an initially graded signal into an all-or-nothing activation of JAK/STAT and thus into proper cell specification and migration. A mathematical model incorporating the relationships of these proteins can accurately simulate wild-type and mutant phenotypes, supporting the idea that our three component system is sufficient to translate a gradient into discrete cell identities.

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Program/Abstract # 214
Sequential actions of Pax3 and Pax7 drive xanthophore development in zebrafish neural crest
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The *Pax3/7* gene family has a fundamental and conserved role during neural crest formation. In people, *PAX3* mutation causes Waardenburg syndrome, and murine Pax3 is essential for pigment formation. However, it is unclear exactly how Pax3 functions within the neural crest. Here we show that *pax3* is expressed before other *pax3/7* members, including duplicated *pax3b*, *pax7* and *pax7b* genes, early in zebrafish neural crest development. Knockdown of Pax3 protein by antisense morpholino oligonucleotides results in defective fate specification of xanthophores, with complete ablation in the trunk. Other pigment lineages are specified and differentiate. As a consequence of xanthophore loss, expression of *pax7*, a marker of the xanthophore lineage, is reduced in the neural crest. Morpholino knockdown of Pax7 protein shows that Pax7 itself is dispensable for xanthophore fate specification, although yellow pigmentation is reduced. Loss of xanthophores after reduction of Pax3 correlates with a delay in melanoblast differentiation followed by significant increase in melanophores, suggestive of a Pax3-driven fate switch within a chromatophore precursor or stem cell. Analysis of other neural crest derivatives reveals that, in the absence of Pax3, the enteric nervous system is ablated from its inception. Therefore, Pax3 in zebrafish is required for specification of two specific lineages of neural crest, xanthophores and enteric neurons.

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Program/Abstract # 215
The zebrafish mutants lpy and myx exhibit loss of skeletogenic cranial neural crest
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The vertebrate cranial neural crest (CNC) gives rise to both non-ectomesenchymal derivatives (e.g. neurons, glia, and pigment cells) and ectomesenchymal derivatives (e.g. cartilage and bone). How the ectomesenchyme lineage of the CNC is specified is not well understood. In order to understand ectomesenchyme specification, we identified two zebrafish mutants, lpy and myx, which display specific loss of the ectomesenchyme-derived head skeleton. The expression of early neural crest markers (including sox9b, sox10, snai2, and foxd3) is reduced or completely absent in lpy embryos at early neurulation, suggesting a defect and consequent delay in neural crest specification. However, we have demonstrated that other CNC derivatives appear to be unaffected in lpy mutants, with the notable exception of melanophore pigment cells which are variably lost. Furthermore, transplantation of wild-type CNC precursors into lpy embryos shows that the *lpy* gene is required autonomously within the CNC for normal crest induction and development of the head skeleton. Thus, lpy and myx represent two potential key players in the specification and/or development of the cranial neural crest and the ectomesenchyme lineage. We will present analyses of CNC cell fate in wild-type and lpy mutants, together with more preliminary studies of the recessive myx mutant. In addition, we have mapped the semi-dominant lpy mutation to a small interval on linkage group 3 and will present progress towards identifying the *lpy* gene.

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Program/Abstract # 216
Notch resolves mixed neural identities in the zebrafish epiphysis
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Manipulation of Notch activity alters neuronal subtype identity in vertebrate neuronal lineages. Nonetheless, it remains controversial as to whether Notch activity diversifies cell fate by regulating the timing of neurogenesis or acts directly in neuronal subtype specification. Here, we address the role of Notch in the zebrafish epiphysis, a simple structure containing only two neural subtypes, projection neurons and photoreceptors. Reducing the activity of the Notch pathway results in an excess of projection neurons at the expense of photoreceptors as well as an increase in cells retaining a mixed identity. However, while forced activation of the pathway inhibits the projection neuron fate, it does not promote photoreceptor identity. As birthdating experiments show that projection neurons and photoreceptors are born simultaneously, Notch acts directly during neuronal specification rather than by controlling the timing of neurogenesis. Finally, our data suggests that two distinct signals are required for photoreceptor fate specification: one for the induction of the photoreceptor fate and the other, involving Notch, for the inhibition of projection neuron traits. We propose a novel model in