the function of other localized RNAs. We have also developed a rapid and robust assay for mapping the sequence determinants of RNA localization, and found several predicted RNA stem loops in two transcripts that are necessary and sufficient for localization. These studies indicate that localized and segregated RNA is broadly important for patterning in this embryo, and suggest how the specificity and intricacy of the observed RNA localization patterns are achieved.

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Program/Abstract # 364
Dissecting RNA localisation pathways in neuroblasts and neurons in the *Drosophila* embryo
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In the *Drosophila* embryo, neuroblasts undergo repeated rounds of asymmetric division along the apical–basal axis. This is accomplished by the unequal segregation of cell fate determinants in response to the axis of polarity. A number of mRNAs also exhibit a polarised distribution in neuroblasts, most notably *inscutable*, *prospero* and *miranda*. Although several factors have been shown to be involved in asymmetric mRNA localisation in neuroblasts, the mechanism and developmental significance of this process is not clear. We aim to identify factors involved in this localisation of mRNAs using in vivo RNAi. We are expressing dsRNA for genes potentially involved in this localisation using the maternal Nanos Gal4-VP16 driver, and looking for changes in the mRNA localisation patterns. Preliminary experiments indicate that RNA localisation is disrupted in embryos expressing dsRNA of known components involved in establishing the apico-basal axis, thereby supporting the feasibility of screening for components involved in RNA localisation using an RNAi fly bank. Using the same strategy we aim to analyse RNA localisation in axonal and dendritic processes in later embryonic stages (14–16 h). Many mRNA species are selectively localised in axonal and dendritic compartments of mammalian neurons, and this localisation plays a role in nerve outgrowth/pathfinding and local translation in the synapse. To analyse the mechanism of mRNA localisation in embryonic neurons we shall combine the MS2–GFP system of in vivo RNA labeling with blocking zygotic gene expression by RNAi.

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Program/Abstract # 365
The role of VegT in the pre-MBT development of *Xenopus laevis*
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A prevailing notion in developmental biology has been that the zygotic genome of an organism remains transcriptionally silent during development until a major embryonic transition known as the mid-blastula transition (MBT). The molecular machinery necessary for zygotic gene expression is present in early *Xenopus* embryos, but large-scale transcription does not occur until the MBT. However, our laboratory has demonstrated that Wnt/β-catenin signaling robustly induces transcription of two Nodal-related dorsal determinants, Xnr5 and Xnr6, prior to the MBT. To test whether other maternal transcription factors are active before the MBT, we examined known targets of VegT, a vegetally localized maternal T-box transcription factor, essential for the specification of mesoderm and endoderm. VegT is an especially interesting candidate because the post-MBT expression of Xnr5 and Xnr6 is regulated by VegT as well as β-catenin/Tcf. Our data show that depletion of VegT leads to ablation of pre-MBT gene expression of Xnr5 and Xnr6. Several other direct targets of VegT, including the Nodal ligand derrière, are also transcribed during pre-MBT stages and are sensitive to loss of maternal VegT function in the embryo. Furthermore, unlike Xnr5 and Xnr6, these newly identified pre-MBT genes are not sensitive to depletion of β-catenin, suggesting that the Wnt pathway is not necessary for all pre-MBT gene expression in *Xenopus*. Current and future work includes characterizing when VegT function is required during these early stages of *Xenopus* development, as well as determining the function of pre-MBT gene expression in the embryo.

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Program/Abstract # 366
Serotonin signaling regulates morphogenesis of the ciliated gastrocoel roof plate (GRP) epithelium during *Xenopus* left–right axis formation
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A cilia-driven leftward flow breaks bilateral symmetry in fish and mammalian embryos. We recently showed that flow was conserved in frog, and that inhibition of flow caused left–right (LR) defects. Early asymmetries have been reported in *Xenopus* during cleavage stages, such as asymmetric localization of serotonin, raising the questions if and how these events are connected to flow. Immunohistochemical analysis of serotonin localization revealed vesicular staining, which was enriched close to the blastocoel from cleavage to blastula stages, and at the roof of the archenteron at neurulation, i.e. during flow stages. However, no consistent asymmetries could be detected at any stage. Surprisingly, serotonin was also prominently seen along the entire length of cilia at the GRP. We cloned 11 of the 14 known serotonin receptors, of which two were expressed during early embryogenesis. One of the type 3 receptors was analyzed in depth. Morpholino knock-down as well as expression of a dominant-negative receptor resulted in LR defects. *Xnr1* and *Pitx2c* gene expression were randomized. Flow was undetectable; scanning electron microscopy revealed that the GRP did not form in injected specimens, suggesting that serotonin signaling is required for GRP morphogenesis. Our analysis of serotonin signaling during LR axis formation thus revealed that serotonin is not required at cleavage stages but is necessary for generating the ciliated epithelium which drives leftward flow.

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Program/Abstract # 367
Regulation of early *Xenopus* development by the Polycomb group protein EED
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