Identifying Transcripts Involved in Neural Tube Closure Using RNA sequencing

Tianna Perosino, Lexy Kindt, Alicia Coughlin, Marshall Hampton, Jennifer O. Liang

Anencephaly is a fatal human developmental defect in which the anterior neural tube remains open. When Nodal signaling is reduced, zebrafish display a disorder similar to anencephaly. Previous work from our laboratory suggests a model where Nodal signaling acts through induction of the head mesendoderm and mesoderm. The mesendoderm/mesoderm then promotes adhesion between neural tube cells. This cell adhesion is required for the movements that bring the edges of the left and right folds together so that they fuse at the dorsal midline and close the neural tube. Consistent with a central role for mesendoderm/mesoderm, we found that Nodal signaling is required up to dome stage (4.3 hpf) for a closed neural tube, which is within the time frame when Nodal activity is required for mesendoderm/mesoderm induction. However, the actual mechanism for interaction between the mesendoderm/mesoderm and the developing neural tube is still unknown. We used RNA-sequencing to identify potential candidates within this mechanism. Embryos treated with a Nodal inhibitor at sphere (neural tube will be open) or 30% epiboly (neural tube will be closed) stages were snap frozen at three time points important for neurulation: shield (6 hpf, neurectoderm induction), tailbud (10 hpf, neural plate forms), and 7 somites (12 hpf, forebrain neural rod is closed). Resulting mRNAs were selected based on degree and significance of differential expression. Consistent with our model, we found mRNAs that are known to be regulated by Nodal signaling, including goosecoid, sox17, and bonnie and clyde. The screen also identified many mRNAs involved in Wnt, FGF, and BMP signaling, suggesting these pathways might be involved in the communication between the mesoderm/mesendoderm and the neuroectoderm.

This research was supported by NIH R15 grant 1 R15 HD068176-01A1.