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## Neural Signals May Take a Distinct Route to the Hindbrain when Generating Spontaneous Eye Movements

The visuomotor system enables animals to move their eyes in response to visual stimulation. In vertebrates it includes the superior colliculus, homologous to the optic tectum in non-mammals, which relays signals downstream to hindbrain regions that direct eye movements. If this system or a different one accounts for eye movements that occur absent visual stimulation is unknown. We address this question by studying horizontal eye movements in larval Danio rerio (zebrafish). Zebrafish are an ideal model because the visuomotor system is well conserved in vertebrates and the entire brain can be imaged at once in small and transparent larvae. We hypothesize that the tectum does not contribute to eye movements absent visual stimulation and by extension, that zebrafish bypass it to do so. We employed two-photon calcium imaging to observe activity in the tectum positively correlated with rapid eye movements from side to side, called horizontal saccades. Larvae were immobilized in agarose dorsal side up, given space to move their eyes, and most of the brain was imaged with a custom-built two-photon laser scanning microscope. Eve movements were simultaneously recorded with a camera from below the animal. The animal was kept in darkness during the experiment to prevent visual stimulation. Analysis of these data corroborated our hypothesis and produced unexpected results. First, we did not find neurons positively correlated with horizontal saccades in the optic tectum. This finding corroborates the idea that the tectum does not contribute to saccades made in the absence of visual stimulation. Second, we observed neurons positively correlated with eye position in unexpected regions of the forebrain. This finding provides preliminary evidence of another circuit for nonvisual eye movements, one that may bypass the tectum and involve the forebrain.