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Dear Nandita Singh,

Please accept our manuscript entitled "Healthy brain-pituitary slices for electrophysiological investigations of pituitary cells in teleost fish" by Romain Fontaine, Kjetil Hodne and Finn-Arne Weltzien, for consideration of publication in JoVE.

The patch-clamp technique is a powerful tool with unprecedented temporal resolution and sensitivity, allowing investigations of electrical properties from intact whole cells down to single ion channels. This technic is particularly relevant to study pituitary endocrine cells, which possess important exocytosis mechanisms. While the limited number of studies conducted on pituitary cells from fish have been performed on dissociated primary cells, we present here a detailed technique to prepare healthy brain-pituitary slices, using the medaka as model. Following slice preparation, we demonstrate how to conduct electrophysiological recordings using the perforated patch-clamp technique that is more reliable to study small cells such as the pituitary cells than whole cell configuration. We also emphasize the importance of correctly adjusting parameters such as pH and osmolality in the different slicing and recording media to what is



found in the model organism.

Believing that these findings may interest a large scientific audience in the field of basic and applied endocrinology, we would like to publish them in your Journal.

Yours sincerely,

Finn-Arne Weltzien