THE SELECTIVE ROLE OF CORTICAL INHIBITORY INTERNEURONS IN FUNCTIONAL HYPEREMIA

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Introduction

- Acute CBF responses driven by fast changes in arteriolar diameter and related to neuronal signaling are mediated by vasoactive messengers released by excitatory and inhibitory neurons.
- CBF responses to sensory stimuli are composed of a combination of dilatory and constrictive phases.
- Inhibitory neurons (INs) release neuropeptides and nitric oxide (NO) and are thought to be able to cause dilation and constriction.
- Does selective optogenetic (OG) stimulation of INs lead to biphasic dilation and constriction of sensory-injected responses?
- Is the constriction phase specific to IN activation?
- Which IN-derived messengers contribute to vasodilation?

Methods

See Uhlirsova, Kliciù, et al. for details. Most experiments were performed in the forepaw region of the somatosensory cortex in adult mice under α-Chloralose anesthesia and neuromuscular blockade with pancuronium.

In vivo-imaging of cortical arterioles with 2-photon microscopy (2PM)

Diving arterioles were imaged at different cortical depths. Linescans were used to measure arteriolar diameter changes in response to sensory/OG stimuli.

Optogenetic Stimulation at Different Cortical Depths

Vascular responses to OG stimulation at surface (via objective) or directly in layer V (via fiber) have similar time courses and are both sensitive to TTX.

Optogenetic IN stimulation causes a biphasic dilation/constriction response. Responses to sensory and OG stimulation show a depth-dependent onset.

OG Stimulation of Inhibitory vs. Excitatory Neurons

Optogenetic stimulation of INs in VGAT-ChR2-EYFP (Zhao et al.) mice or pyramidal neurons in Thy1-ChR2-YFP (Arenkiel et al.) mice. 473 nm laser light was used for OG stimulation of INs in VGAT-ChR2-EYFP (Zhao et al.) mice or pyramidal neurons in Thy1-ChR2-YFP (Arenkiel et al.) mice under α-Chloralose anesthesia and neuromuscular blockade with pancuronium.

Validation in Awake, Head-immobilized Mice

OG stimulation in awake VGAT-ChR2-EYFP mice. Optogenetic stimulation of INs causes biphasic responses that are similar to responses to sensory stimulation.

Conclusions and Outlook

INs play a major role in shaping functional hyperemia:
- Selective OG stimulation of INs causes biphasic arteriolar responses.
- Dilation in response to sensory and OG stimulation starts in deep cortical layers.
- Release of vasoactive messengers from INs is TTX-sensitive.
- NPY is a major constrictive IN-derived messenger.
- Several messengers including VIP contribute to IN-mediated vasodilation.

Open questions:
- Sites of messenger release (dendrites or axons)?
- Propagation of dilation and constriction along arteriolar walls?
- Do IN-induced changes in extracellular [K⁺] contribute to vasodilation?
- Effect of nNOS blockade/deletion on vascular response to OG IN stimulation?

Outlook:
- Validation of the roles of NPY and VIP in awake animals.
- Analyzing the contribution of different IN subtypes to functional hyperemia.
- Establishment of a modeling framework to predict cell type-specific neuronal activity from non-invasive multi-modal data acquisition in mice and humans (see Uhlirsova et al.).

Acknowledgments

This work was supported by NH BRAIN Initiative grants U01 NS094232, UHCA1115339, NH Grants N0575198, E003790, and STO9020805, and the Research Council of Norway (223273, 223912). KJ was supported by International Headache Society and TUBITAK. MTH was supported by DFG.