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MASTER PROJECT: Immunolabelling of cleared thick three-dimensional brain sections

Fluorescent-protein expression in genetically identified cell populations, along with advances in tissue clearing [Con15, Ri15] and three-dimensional (3-D) imaging [Ra19] allow reconstructions of large tissue volumes, permitting studies of organisation and cellular circuitry at an unprecedented detail.

In our team, we developed an ultrafast, universal clearing agent that renders various tissues transparent within minutes, is non-toxic and preserves the tissue volume and fluorescence of most of the fluorophores and fluorescent proteins studied. In many applications, however, it would be desirable to counterstain autofluorescent or genetically encoded fluorophores with specific immunofluorescence to obtain a contextual information. Yet, the use of such probes in thick tissue has proven difficult and often requires optimization, following the limited depth penetation of primary and secondary antibodies and deletion of immunofluorescence by clearing agents. [Sou16, Pav18]

The current project aims at the optimization of antibody penetration and *post-hoc* labelling strategies in thick cleared sections of the rodent cortex and cerebellum and their observation with confocal, 2-photon and light-sheet microscopies.

Students should have a strong interest in experimental neuroscience, biophysics, or cell biology. The ideal candidate has already practical lab skills for, a background in quantitative biology, and a deep desire to understand the principles underlying the functioning of the nervous system. Cell culture, microscopic imaging and programming skills are beneficial. For applications we request: (i) a statement of motivation, (ii) a curriculum vitae, (iii) the names and emails of 2 academic references.

Further reading

[Con15] Constantino et al. (2015) Sci Rep. https://doi.org/10.1038/srep09808
[Pav18] Pavlova et al. (2018) Hippocampus https://doig.org/10.1002/hipo.22951
[Ra19] Rakotoson et al. (2019) Front Neuroanat. https://doi.org/10.3389/fnana.2019.00077
[Ri15] Richardson & Lichtman (2015) Cell. https://doi.org/10.1016/j.cell.2015.06.067
[Sou16] http://blogs.nature.com/methagora/2016/02/methods-and-probes-for-cleared-tissue-an-imperfect-table.html

Contact

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