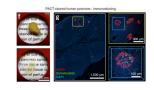


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Whole-body tissue stabilization and selective extractions via tissue-hydrogel hybrids for high-resolution intact circuit mapping and phenotyping



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Protocol status: Working

We use this protocol and it's working

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Keywords: selective extractions via tissue, tissue biomolecules during selective lipid extraction, image tissue, tissue biomolecule, processed tissue hybrid, tissue hybrid, staining protocol, selective lipid extraction, tissue fixation, body tissue stabilization, hydrogel hybrid, transparent sample, tissue, intact tissue, set of tissue fixation, macromolecule permeability, resolution imaging, endogenous fluorescence, cell visualization, scale phenotyping of whole specimen, whole specimen

#### Abstract

To facilitate fine-scale phenotyping of whole specimens, we describe here a set of tissue fixation-embedding, detergent-clearing and staining protocols that can be used to transform excised organs and whole organisms into optically transparent samples within 1-2 weeks without compromising their cellular architecture or endogenous fluorescence. PACT (passive CLARITY technique) and PARS (perfusion-assisted agent release in situ) use tissuehydrogel hybrids to stabilize tissue biomolecules during selective lipid extraction, resulting in enhanced clearing efficiency and sample integrity. Furthermore, the macromolecule permeability of PACT- and PARS-processed tissue hybrids supports the diffusion of immunolabels throughout intact tissue, whereas RIMS (refractive index matching solution) grants high-resolution imaging at depth by further reducing light scattering in cleared and uncleared samples alike. These methods are adaptable to difficult-to-image tissues, such as bone (PACT-deCAL), and to magnified single-cell visualization (ePACT). Together, these protocols and solutions enable phenotyping of subcellular components and tracing cellular connectivity in intact biological networks.

#### **Attachments**



nprot.2015.122.pdf

6.6MB



# Troubleshooting

