

# Rapid and Efficient Clearing of Brain Tissues for 3D Imaging



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Identification and exploration of detailed organization of organs or whole body at cellular level are fundamental challenges in biology. Conventionally this task has been approached by serial sections, labeling of specific targets, and the reconstitution of them into 3-dimensional structures. These are labor- and time-consuming process, and the accuracy is highly limited. Advent of tissue clearing techniques, such as CLARITY, PACT, CUBIC and iDISCO, has revolutionarily improved the efficacy of volume imaging mainly by elimination of tissue sectioning and reconstitution steps. However, currently available protocols for organ clearing require considerably long process time, making it difficult to implement tissue clearing techniques to the lab. Here, we present a rapid and highly reproducible ACT-PRESTO (active clarity technique-pressure related efficient and stable transfer of macromolecules into organs) method that renders tissue or whole-body clearing within a day while preserving tissue architecture and protein-based signals derived from endogenous fluorescent proteins. Moreover, ACT-PRESTO is compatible with conventional immunolabeling methods and expedites antibody penetration into thick specimens by applying pressure. Speed and consistency of this method will allow high-content mapping and analysis of normal and pathological features in intact organs and bodies.

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