Rapid and Uniform Staining of Thick Biological Tissues with Antibody using Electro-magnetic Focused Immuno-histoChemistry

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Understanding the structure of a complex biological system at the cellular level requires 3D anatomical and phenotypical maps. This task has dramatically advanced by recent surges of various tissue clearing methods. Labeling of thick tissue with an antibody, however, still depends on the slow diffusional process, thus suffer from inefficient and uneven penetration of macromolecules into thick tissues. Although a few methods have developed to overcome this problem, they are still laborious and require specialized equipment. Here, we present a novel technique for rapid and uniform penetration of antibody into thick biological tissues using the focused electro-magnetic forces. This technique that we called the **EFIC** (Electro-magnetic Focused Immuno-histoChemistry) achieves the focused electric and magnetic field into a certain area, thus concentrates antibody into the direction of thick biological samples, enabling rapid, uniform, and complete staining without sample distortion. Using the **EFIC** method, we could stain thick brain tissues uniformly and rapidly (up to 3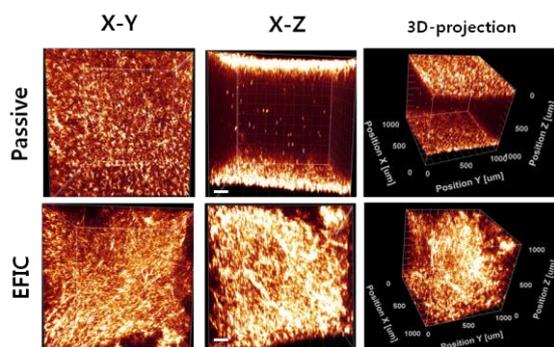 mm deep sample within 4 h) with only a limited amount of antibody (typically 50 μg/reaction). We have successfully applied **EFIC** to formalin-fixed postmortem human brain tissues and simultaneous multi-color staining with 4 different antibodies. We also developed a novel optical clearing solution named **perfect-MATCH** to minimize deformation artifacts that are due to the harsh treatment and transient sample swelling. Together, our **EFIC and perfectMATCH** are optimized to preserve all necessary 3D molecular and structural information for high-resolution fluorescence imaging.

Figure 1. EFIC *vs.* passive staining of anti-GFAP antibody in 4 hours

[1]K.Chung *et al*, Nature 497(2013) 332-7

[2]S.Y kim *et al,* Proc NatlAcad Sci U.S.A 112(2015) E6274-83

[3]E.Lee *et al*. Sci Rep. 6(2016)18631