Dense 4D nanoscale reconstruction of living brain tissue

P. Velicky[1,2], E. Miguel[1], J.M. Michalska[1], J. Lyudchik[1], D. Wei[3], Z. Lin[3], J.F. Watson[1], J. Troidl[3], J. Beyer[3], Y. Ben-Simon[1], C. Sommer[1], W. Jahr[1], A. Cenameri[1], J. Broichhagen[4], S.G.N. Grant[5,6], P. Jonas[1], G. Novarino[1], H. Pfister[3], B. Bickel[1], J.G. Danzl[1]

[1] Institute of Science and Technology Austria; [2] Core Facility Imaging, Medical University of Vienna, Austria; [3] School of Engineering and Applied Sciences, Harvard University, Cambridge, MA, USA; [4] Leibniz-Forschungsinstitut für Molekulare Pharmakologie, Berlin, Germany; [5] Simons Initiative for the Developing Brain (SIDB), Centre for Discovery Brain Sciences, University of Edinburgh, Edinburgh, UK; [6] Genes to Cognition Program, Centre for Clinical Brain Sciences, University of Edinburgh, Edinburgh, UK.

Abstract

Brain computation and information storage are intimately linked to the structure of a network of ~86 billion neurons in humans. Each is typically connected by thousands of information-transmitting synapses to other neurons and interacts with glial support cells. 3D reconstruction of living brain tissue down to individual synapse level would create major opportunities for decoding the dynamics and structure-function relationships of the brain's incredibly complex and dense information processing network. However, this has been hindered by insufficient 3D resolution, inadequate signal-to-noise-ratio, and prohibitive light burden in optical imaging, whereas electron microscopy is inherently static. Here we solved these challenges by developing an integrated optical/machine learning technology, LIONESS (Live Information-Optimized Nanoscopy Enabling Saturated Segmentation). It leverages optical modifications to stimulated emission depletion (STED) microscopy in comprehensively labelled tissue and prior information on sample structure via machine learning to simultaneously achieve isotropic super-resolution, high signal-to-noise-ratio, and compatibility with living tissue. This allows dense deep-learning-based instance segmentation and 3D reconstruction at synapse level incorporating molecular, activity, and morphodynamic information. LIONESS opens up major avenues for studying the dynamic functional (nano-)architecture of living brain tissue.