IN VIVO MULTIPHOTON IMAGING USING ULTRAFAST SEMICONDUCTOR DISK LASERS

Fabian F. Voigt[1,2,†], Florian Emaury[3], Philipp Bethge[1], Dominik Waldburger[3], Sandro M. Link[3], Stefano Carta[1], Alexander van der Bourg[1], Fritjof Helmchen[1,2], and Ursula Keller[3]

[1] Laboratory of Neural Circuit Dynamics, Brain Research Institute, University of Zürich, 8057 Zürich, Switzerland

[2] Neuroscience Center Zurich, University of Zürich & ETH Zürich, Switzerland

[3] Department of Physics, Institute for Quantum Electronics, ETH Zürich, 8093 Zürich, Switzerland

[†] E-Mail: voigt@hifo.uzh.ch

Keywords: Multiphoton microscopy, nonlinear microscopy, ultrafast lasers, semiconductor lasers

Over the past decades, multiphoton microscopy methods have been widely adopted in biomedical research owing to their combination of high spatial resolution, intrinsic optical sectioning, and the ability to image deep in scattering samples. Despite these advantages, the dissemination of nonlinear imaging methods is, however, limited by the high cost of ultrafast laser sources. As an attractive alternative to existing sources, we explored the imaging capabilities of a mode-locked semiconductor disk laser (SDL), which recently reached the sub-100 fs regime [1]. The laser used in this work delivers 150 mW of average power, sub-200 fs pulses at 1026 nm combined with excellent beam quality (M^2 <1.05) and a small footprint. As a proof-of-concept in vivo application, we imaged blood vessels filled with Dextran Texas Red in the brain of a mouse through an implanted chronic window (Fig 1a). Additionally, we were able to record functional signals from neocortical neurons expressing the calcium indicator R-CaMP1.07 in a similar preparation at depths of 125-300 μm (Fig 1b). Beyond two-photon excited fluorescence, we also imaged the second-harmonic generation (SHG) signal originating from the dura mater in mice (Fig 1a) and sarcomeres in the muscles of drosophila larvae (Fig 1c). As femtosecond SDLs can be tailored to a wide range of excitation wavelengths, they open up a path towards more widespread usage of multiphoton techniques in research and clinical applications by providing a cost-efficient laser source.

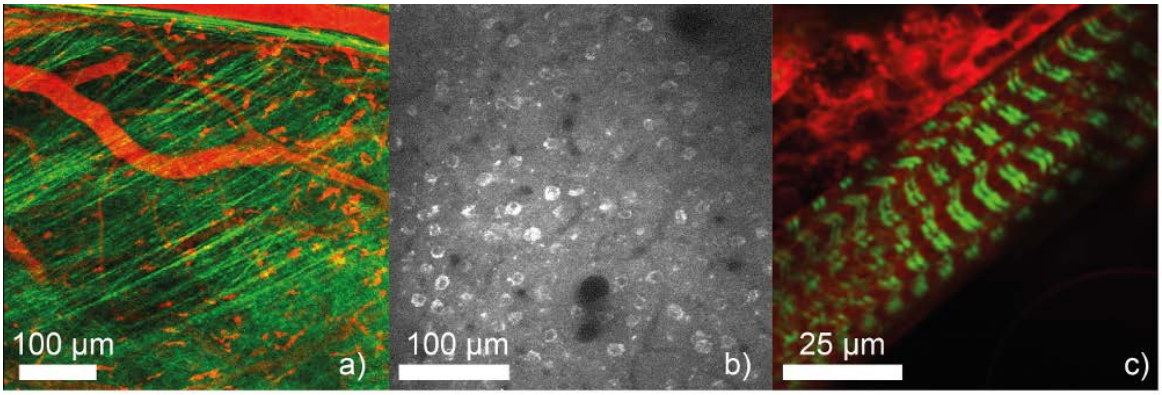

Fig. 1.: Multiphoton imaging experiments using a femtosecond semiconductor disk laser (SDL): a) In vivo imaging of blood vessels filled with Dextran Texas Red (red) in the mouse brain. b) In vivo imaging of R-CaMP1.07-positive neurons in the mouse brain. c) Two-photon excited fluorescence (red) and SHG (green) imaging in a Drosophila larva expressing mKate2 in cell membranes.

References

[1] D. Waldburger, S. M. Link, M. Mangold, C. G. E. Alfieri, E. Gini, M. Golling, B. W. Tilma, and U. Keller, "High-power 100 fs semiconductor disk lasers," Optica **3**, 844-852 (2016).