Two-Photon Imaging Unit

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> Head of Unit: Balázs Rózsa, MD, PhD

Mission statement

o further understand the computational mechanism of the brain we need complex techniques to be able to measure from large population of neurons, but also with great details and subcellular resolution. The main goal of our group is to adopt and develop novel imaging techniques for scientific researches which make this quest attainable. With our novel developments it became possible to follow the activity of the entire dendritic tree of a neuron in three dimensions or to record hundreds of cell simultaneously in an intact neuronal network (Katona et al. 2012 Nature Methods, Katona et al. 2011 PNAS). Having these techniques in hand, we turn to various biological questions that could be only addressed with these novel tools. We are interested in better understanding the role of the integration on a single neuron interacting with a large network. For example we are studying the dendritic integration on a single neuron during sharp wave ripple oscillation (Chiovini et al. 2014 Neuron). A larger scale project is to find out the role of a single neuron within different network ensembles during learning tasks performed by the animal. Specifically weare investigating of the neuronal level coding of the visual cortex during different visual stimulation and behavioral paradigms. The Two-Photon Imaging Center is closely collaborating with research at the PázmányPéter Catholic University and also with the multidisciplinary research team of Femtonics Ltd.

3D two-photon imaging

To understand the fast computational mechanisms of the brain, one needs to be able to perform rapid measurements at several sites along a single neuron as well as to image large populations of neurons. Traditional two-dimensional measurements are severely limited for such kinds of endeavors since neurons are located in three dimensions. To overcome this problem, we have developed new solutions to perform three-dimensional functional imaging with large scanning ranges along the z direction (Katona et al. 2012 Nature Methods, Katona et al. 2011 PNAS). With our three-dimensional microscopes we are able to maintain point or trajectory scanning which in combination with the ~800 µm penetration depth of two-photon technology makes our methodology very convenient for *in vivo* measurements of neuronal populations, too.

In vivo measurement of network activity

Population activity has long been studied in the visual cortex. We conduct *in vivo* two-photon imaging of cell assemblies in the V1 area by using bolus loading and genetic approaches. Neuronal network responses are recorded during different visual stimuli. In addition, active cells are selected based on the previously recorded somatic activity and their dendritic responses are followed along with the network activity in three dimensions by using whole-cell patch clamp techniques.

Two-photon uncaging

Two-photon uncaging takes advantage of the high spatial and temporal resolution of two-photon excitation to study dendritic integration, a post-synaptic mechanism. Used in combination with two-photon imaging, two-photon uncaging provides an opportunity to study the long-term structural and functional consequences of stimulation of structures such as dendritic spikes and dendritic spines. Besides performing experiments we develop new caging compounds and use these for our new measurements.

In vitro measurement of spontaneous neuronal network activity

It is thought that sharp wave-ripples (SPW-R) activity is involved in the process of memory consolidation. The properties of SPW-R events are similar to what was found in vivo. We investigate spontaneous single cell (pyramidal cell and interneuron) neuronal activities during SPW-R in the hippocampus CA1 and CA3 region under in vitro conditions. Fast spiking, PV+ basket cells as the clockworks for neuronal oscillations are important elements of hippocampal neuronal networks. Thus, beside the pyramidal cells we focus on PV+ interneurons to reveal the dendritic calcium dynamics during SPW-R (Chiovini et al. 2014, Neuron).

Selected publications:

- B. Chiovini, G. F. Turi, G. Katona, A. Kaszas, D. Palfi, P. Maak, G. Szalay, M. F. Szabo, Z. Szadai, Sz. Kali and B. Rozsa. Dendritic spikes induce ripples in parvalbumin interneurons during hippocampal sharp waves. Neuron (2014)
- Gergely Katona, Gergely Szalay, Pál Maák, Attila Kaszás, Máté Veress, Dániel Hillier, Balázs Chiovini, E. Sylvester Vizi, Botond Roska & Balázs Rózsa. Fast two-photon in vivo imaging with three-dimensional random-access scanning in large tissue volumes, Nature Methods (2012)
- G. Katona, A. Kaszás, G. F. Turi, N. Hájos, G. Tamás, E. S. Vizi, B. Rózsa.
 Roller Coaster Scanning reveals spontaneous triggering of dendritic spikes in CA1 interneurons, Proc. Natl. Acad. Sci. USA, Volume 108, No. 5 (2011), Page 2148-2153
- B. Chiovini, G. F. Turi, G. Katona, A. Kaszás, F. Erdélyi, G. Szabó, H. Monyer, A. Csákányi, E. S. Vizi, B. Rózsa (2010). Enhanced Dendritic Action Potential Backpropagation in Parvalbumin-positive Basket Cells During Sharp Wave Activity, Neurochemical Research Volume 35, Number 12, 2086-2095





