Final Report for NKFIH Grant #113147, entitled:
'Micro-electric imaging: modeling, source reconstruction and causality analysis for multi-electrode arrays' by Zoltán Somogyvári

As planed, during the granted work we have developed several model-based, neuro-informatical data analysis methods. Our methods are able to 1, calculate the spatio-temporal distribution of synaptic membrane currents and membrane potential on single neurons, based on multi electrode data, 2, determine the functional structure within and 3, the causal relationships between brain areas to support surgical planing in case of epileptic patients. Hereby I summarize the major achievements of the work supported by this grant.

Revealing spatio-temporal distribution of membrane current on single neurons. One of the main obstacle to decipher the information processing and the neural communication in the brain is the lack of any experimental technique which is able to measure the spatio-temporal distribution of synaptic currents on individual neurons in freely behaving animals. Thus, in cooperation with Daniel Wójcik, (Nencki Institute of Experimental Biology, Warsaw), we have developed a constrained inverse solution using kernel method to solve the Poisson inverse problem on the known, complex, branching morphology of a neuron. We called it single-neuron kernel current source desity method (skCSD). We have shown, that inclusion of the known morphology largely enhanced precision of the inverse solution and applied the skCSD method to the first available experimental data, where both the extracellular potential were measured by a 1D multi-electrode array and the morphology of the cell (a CA1 pyramidal neuron) were reconstructed. The data was provided by the lab of István Ulbert, Institute for Natural Sciences of the HAS. The current source density distribution and the spreading of the currents along the morphology were determined during the averaged action potential (Fig. 1, Cserpán et al. 2017). The paper were published in the journal eLIFE. The scripts for the analysis, written in R, were tested and made publicly available as an open source program package. A review chapter is published on the single neuron current source density analysis in a Springer volume (Somogyvári and Érdi 2016).

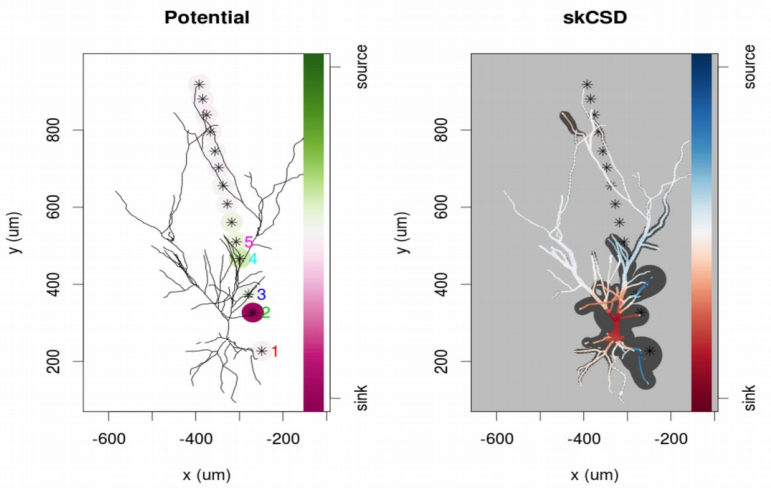

Figure 1: *Left: Electrode positions (stars) and reconstructed morphology of the pyramid cell from the CA1 region of the rat hippocampus. The color coded circles on the electrodes show the measured momentary electric potential at the moment of the peak of an action potential generated by the neuron. Right: The reconstructed current source density distribution along the dendritic tree of the neuron. Warm colors mark inward positive currents to the neuron (sink) cold colors mark the outward currents from the neuron (sources).(From Cserpán et al. 2017)*

In cooperation with Antal Berényi's lab (Szeged University), we have shown the differences of cell-type specific input current patterns preceding and causing the action potentials during different oscillatory states of hippocampus. The layers and subfields of the hippocampus have been identified based on the recorded electrical signals, by using our electroanatomy concept and latter verified by histology. The types of the EC recorded and clustered cells were determined based on their

electrophysiological characteristics and their spatial tuning. Analyzing the temporal dynamics of the cell type specific micro-field potentials we have found, that the onset of the synaptic currents preceding the action potential was the shortest in the CA1 region: 8.9 and 11.4 ms for pyramidal and interneurons respectively). We have found longer onset times in the dentate gyrus (12.4 ms for granular cells and 16.7-31.2 ms for interneurons). Finally, the longest onset times were found in the CA3 pyramidal neurons (40 ms), while the onset times of the interneurons were shorter in this region (7.6-21 ms). As the dynamics of the total synaptic current is depends on the natural statistics of the synaptic activations, measuring the temporal aspects of net synaptic currents could lead to better understanding of the neural code, by refining our knowledge about the input-output transformation implemented by the neurons. These results were presented on the SFN Conference: Neuroscience 2015, Chicago (Somogyvári et al. 2015).
Riera et al. (J Neurophysiol 2012) initiated a debate in the literature, by claiming, that neurons show monopole currents, ie. the sum of the inward and the outward current is not 0 on a neuron at each moment. By using simulations of neural models with detailed morphology and 3D electrode measurements, we showed, that it is not necessary to assume monopole currents, a possible cause of the appearing monopole current sources is the finite sampling of the extracellular medium and the approximations used in the methods for estimating CSD. This result was published on a poster (Cserpán et al. 2016).

Membrane potential reconstruction. Two versions of the membrane potential reconstruction method, based on parallel intracellular and extracellular multi-electrode recordings were developed and applied on the first available data sets: the first version is applicable, where the spiking of the recorded neuron does not correlates to the large extracellular population activity or evoked by stimulation. In this case, we can assume, that the majority of the averaged extracellular potential was generated by the actually recorded cell, thus we are able to reconstruct the spatio-temporal distribution of the membrane potential along the actually recorded neuron.
The second version is applicable for the large, repetitive population activities. In these cases, the subthreshold somatic membrane potential of the recorded neuron can be considered as a sample from an average neuron within the population, thus the average spatio-temporal distribution of the membrane population and synaptic currents can be reconstructed.
This work have been presented as a poster on the IBRO world Congress in Korea (Somogyvári et al. 2019). The manuscript describing the new methods and the first use-cases are under preparation.

Combined micro-electric and optical imaging. Cortical functional micro-structures, such as orientation selectivity columns and pinwheeles in the primary visual cortex were generally assessed by in vivo (blood dependent) intrinsic optical signal (IOS) imaging. A new transparent cortical electrode array, which were developed by the lab of

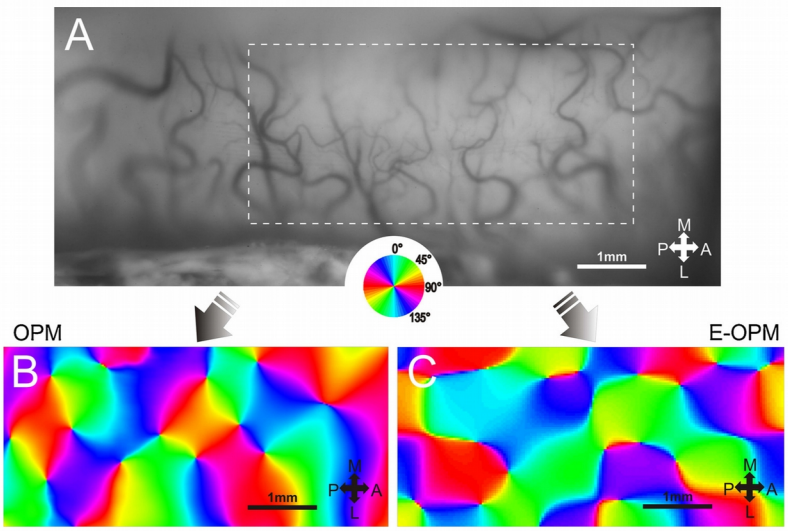

Figure 2: *Comparison between optically and electrically derived orientation preference maps. The white dashed frame on the grayscale vascular image (A) shows the position of the 32-channel microelectrode array inside the investigated A17 region. Traditionally processed orientation preference map (B) and electrical orientation preference map derived from the evoked ECoG responses to the visual stimuli (C) are shown.*

Zoltán Fekete and the measurements which were performed by Zsolt Borhegyi and Zoltán Kisvárday, made possible the determination of those functional patterns based on the electrical activity and its comparison to the parallel optical imaging. Thus, we calculated the orientation preference map through electric imaging, based on the local field potential recordings and compared to the orientation preference maps calculated from the parallel intrinsic optical imaging, recorded through the transparent electrode array. Based on the comparison of the orientation maps obtained from optical an electrical recordings, the observed structural similarity raises the possibility to construct a unified imaging system which unifies the high spatial resolution of the optical imaging and the high temporal resolution of the electric imaging method (Fig. 2, Zátonyi et al. 2017, Zátonyi et al. 2018).

Furthermore, a user friendly graphical interface was developed for easy exploring the data recorded by subdural grid electrodes. The interface makes easy to calculate the kCSD and create power maps on different frequency regimes based on both LFP and CSD as well as segmenting the electrodes by the coherence based clustering. The method were applied on recordings in rat cortex during ketamine induced oscillations, to determine the cortical structures and areas from the measurements with the transparent cortical surface electrode grid, parallel to the intrinsic optical signal measurement. The methodology and the first results were published on a conference and it is already submitted to a methodological journal (Fedor et al. Submitted.).

Causality analysis between electrical signals of neural activity and the intrinsic optical signal of the neural tissue. Causal relationship between local field potential (LFP) and intrinsic optical signal (IOS) in evoked epileptiform activity in vitro brain slices was investigated. The parallel IOS and LFP recordings were performed by Sándor Borbély and Ildikó Világi. As far as we know, this work was the first conclusive application of the Sugihara's new causality method, the cross-convergent mapping (CCM) in neuroscience. As CCM is the first causality analysis method which can reliably detect the circular connection, we were in the position of investigating the question, whether only the evoked epileptic activity causes the intrinsic optical signal IOS, or there is a feedback mechanism as well, and the ion concentration changes measured by the IOS influence the termination or the renewal of the epileptic activity.
During preprocessing, two components of the IOS signal have been distinguished: a faster, activity dependent component (IOSh) which changes its sign between transmitted and reflected light measurements thus it is related to the reflectance or the dispersion of the tissue and a slower component (IOSl), which is negative in both cases, thus can be attributed to the increase of the absorption of the tissue. We found only unidirectional causal drive from the electric towards the optical signal, but this work demonstrated several phenomena which are instructive for further investigation: We found, that the correlation was small between the LFP and the IOSh at the time of the actual causal effect and the peaks of the cross correlation function did not reflect the actual causal dependency in this case. In stead, the temporal derivative of the IOSh was correlated with the LFP power at the time delay of the causal peak. Based on these observations, a simple model have been set up to describe the dependency of the IOSh on the LFP power and IOSh was reconstructed, based on the LFP signal. Besides the actual results, we believe that this study demonstrates, that it is possible to calculate the causality between two data series with drastically different time scales and provides useful know-how for application of causality analysis for any field of science (Benkő et al. 2019).

Development and application of a new causality analysis method to human neurophysiological signals. Studying Sugihara's causality analysis led us the development of a new causality analysis method, based on the manifold dimensions. We have developed the Dimensional Causality (DC) analysis method devised to detect and quantify the probability of all possible types of causal relationships between two time series: independence, direct or circular causal connection, and the existence of a hidden common cause (Benkő 2018). To our best knowledge, no single method

existed before, which can detect and distinguish all these possible causal relationships, based on time series observations from deterministic dynamical systems. To detect these relations between two time series, Takens' embedding theorem (Takens 1978) is used to reconstruct the attractors of the underlying systems. The new method is based on the subadditivity of the system's attractor dimensions, where the key is the dimension of the joint attractor of the two systems. We showed that the relations between the joint and individual dimensions unequivocally determine the causal relations between the dynamical systems. The probability of the different causal relations is obtained via Bayesian inference. We validated our method on simulated examples of 'classic' chaotic dynamical systems, such as nonlinearly coupled logistic functions, coupled Lorentz-systems and Hindmarsh-Rose models. Besides the tests on simulated dynamical systems, the EEG recordings during photostimulation as a part of the standard epileptic investigation protocol, provided us a good opportunity to test the applicability of our method on realworld data, in cooperation with the research group of Dániel Fabó (National Institute of Clinical Neuroscience). DC method confirmed our hypothesis about the increase of common cause probability between the two hemispheres during flashing light stimulation suggesting the applicability of the method in clinical scenarios.

The significance of our method was demonstrated by applying it to electrocorticographic (ECoG) data recorded by subdural grid electrodes during presurgical investigation. Examining four distinct areas of epileptic activity in the patient's brain, the possible focus of epileptic seizure was identified; an area which drives the others, meanwhile the existence of a common drive is identified between the driven areas. These results suggest that common causes revealed by DC method contains relevant information for the diagnosis of epileptic patients in various clinical settings. The first complete version of the paper was uploaded to the arxiv preprint server and was submitted to the best interdisciplinary journals. It was rejected so far, however we still hope we will be able to publish it in a high impact journal (Fig. 3, Benkő et al. 2018).

A concept paper on the topological approaches to the causality analysis have been published in the high impact journal Physics of Life

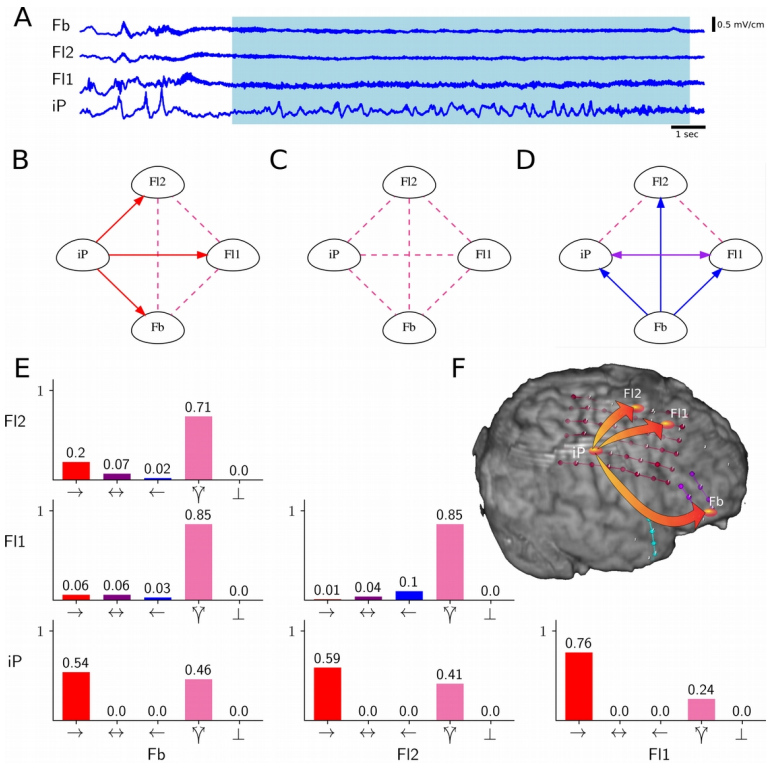

Figure 3. Cortical connectivity during epileptic seizure identified by the Dimensional Causality method (A) CSD signal from 4 areas where epileptic activity was observed. The blue selection shows the analyzed time period of the seizure. Basically two types of connectivity were detected for seizures and a third type of connectivity for interictal conditions (B, C). (B) Maximal a posteriory probability (MAP) causal connection structure for the example seizure in A. Red arrows mark unidirectional relations and the pink dashed lines mark the detected common cause relations. (Seizure type 1, n = 6) (C) MAP causal connection structure for seizure type 2 (n = 10) (D) MAP mean causal connection structure interictal sections (n = 16). Blue arrows mark unidirectional relations and purple arrows denotes circular causal relations. (E) Causal relation probabilities of the seizure showed on A and B. (F) The inferred driver and driven areas represented on the brain surface for the same seizure as on A, B, E (From Benkő et al. 2018).

Reviews as a comment. In this paper we showed, that many of the new causality analysis methods, including Sugihara's cross convergent mapping and our DC method are based on topological concepts rooting back to Takens' time delay embedding theorem. We emphasized, that this is a very powerful theorem in principle, but its application remained restricted due to the difficulties on evaluation and quantification of topologies. The new causality analysis methods, where the attractor reconstruction is applied to two time series instead on one and the topological relation between them is evaluated and quantified, set a new horizon to the dynamical systems' theory and brought the "topological renaissance" to the field (Somogyvári and Érdi, 2017). A review chapter on the data analysis techniques and challenges in epileptic EEG signals have been published in the Springer volume entitled Computational Neurology and Computational Psychiatry (Benkő et al. 2017).

Information theoretical analysis of morphological structures. We have applied our information-theoretical analysis methods to structural patterns revealed by anatomical tissue staining in collaboration with Orsolya Kántor and Béla Völgyi. Bayesian information criterion was applied to determine the optimal model for the spatial distribution of the tissue nonsepcific alkaline phosphatase (TNAP) in the internal plexiform layer of the retina. By applying our model fitting methods, we were able to identify more synaptic layers (11) based in this single staining, than it was known before. Moreover, our results indicate the specific role of TNAP in diabetes by resulting specific changes in the brain TNAP patterning (Kántor et al. 2015a).

Our information-theoretical analysis of the retinal structure have been extended, to compare layering structure of the retina of different species, based on TNAP staining. Based on the similarities and differences in the layering and TNAP staining structure, a hierarchical relation tree have been set up, expressing the group structure of the retinal TNAP structures between species. (Kántor et al. 2016a, Kántor et al. 2015b).

Similar Bayesian information criterion based analysis of connexin36 gap junctions identified 3 subpopulations of synapses within the synaptic pedicles of cone receptor cells in the retina, and helped to determine the spatial clustering of connexin36 gap junctions across human retinal ganglion cell dendritic arbors. We showed, that Cx36 plaques had a clear tendency to form clusters and particularly favored terminal dendritic segments. (Kántor et al. 2016b)

Causality analysis of ecosystems. Due to the generality of the question to be answered and the methodology of causality analysis, it can be applied in many different fields of science. The directed interactions have been determined among the variables of a lake ecosystem, via the Sugihara causality analysis method. Causality analysis indicated that the observed eutrophication signals were induced by climate change, which altered the phosphate, the fito- and the zooplacton interactions. The causality analysis showed, that the observed significant changes in the phytoplankton and zooplankton community and the eutrophication signals (increasing dominance of Cyanobacteria) were related to climate change directly. Our study also demonstrated the strong coupling between planktonic and sedimentary processes in a deep lake. Our results partially explain why short-term experiments for understanding the effect of climate change often fail, or provide contradictory results: there might be substantial delay in cause-consequence relationships in lake ecosystems (Selmeczy 2018).

Dissemination. An educational chapter have been written in Hungarian about the biophysics of the nervous system, in which we reviewed the different levels of single neuron and network models, learning algorithms and methods (Somogyvári and Zalányi 2016).