THE PROGRESS OF ELEMENTAL ANOMALIES OF HIPPOCAMPAL FORMATION IN PILOCARPINE MODEL OF TEMPORAL LOBE EPILEPSY — X-RAY FLUORESCENCE MICROSCOPY STUDY

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Although epilepsy has been a serious problem of clinical neurology for many years, the mechanisms of its pathogenesis are still not fully understood. The analysis of nervous tissue from the period of epileptogenesis is possible based on the animal models of the disease. Animal models of epilepsy help better understand the mechanisms leading to spontaneous seizure activity, allow observations of the progress and character of seizures as well as evaluation of the action of new antiepileptic drugs [1, 2]. The most frequently occurring type of epilepsy in adults is the temporal lobe epilepsy (TLE) and the most frequently used and highly isomorphic with human cases of TLE animal model is one with seizures induced with pilocarpine.

Administration of pilocarpine in rats evokes sequential behavioral and electrographic changes that can be divided into three distinct periods: an acute period that builds up progressively into a limbic *status epilepticus* (24 h), a silent (latent) period with progressive normalization of EEG and behavior (from a few to a few dozen days) and a chronic period with spontaneous recurrent seizures [3].

The main purpose of the present investigations was the analysis of the dynamics of elemental changes observed in rat hippocampus as a result of pilocarpine induced seizures. For the topographic and quantitative elemental analysis of tissues, taken from animals 3 hours (SE3H group) and 1 (SE24H), 4 (SE4D) and 7 (SE7D) days from pilocarpine administration, X-ray fluorescence microscopy was applied. The measurements were carried out at HASYLAB beamline L and at ANKA beamline FLUO. The 17 keV beams focused using the polycapillary optics to 15 and 12 μm were used for the study. The analysis of the differences in the hippocampal accumulation of S, K, Ca, Fe, Cu and Zn between the analyzed animal groups showed that seizure induced excitotoxicity, mossy fiber sprouting and iron induced oxidative stress are the mechanisms involved in the neurodegenerative processes which may finally lead to spontaneous seizures in the chronic period of pilocarpine model.

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References

[1] W. Loscher, "Animals models of epilepsy for the development of antiepileptogenic and disease-modifying drugs. A comparison of the pharmacology of kindling and post-status epilepticus models of temporal lobe epilepsy," *Epilepsy Res.* **50** (2002) 105 – 123.

[2] A.K. Sharma, R.Y. Reams, W.H. Jordan, M.A. Miller, H.L. Thacker, P.W. Snyder, "Mesial temporal lobe epilepsy: pathogenesis, induced rodent models and lesions," *Toxicol. Path.* **35** (2007) 984 – 999.

[3] F.A. Scorza, R.M. Arida, M. da Graca Naffah-Mazzacoratti, D.A. Scerni, L. Calderazzo, E.A. Cavalheiro, "The pilocarpine model of epilepsy: What have we learned?," *An. Acad. Bras. Cienc.* **81** (2009) 345 – 365.