Functional connectivity changes during epileptogenesis: a longitudinal resting-state functional MRI study

Emma Christiaen[1], Marie-Gabrielle Goossens[2], Benedicte Descamps[1], Paul Boon[2], Robrecht Raedt[2], Christian Vanhove[1]

[1]MEDISIP, Department of Electronics and Information Systems, Ghent University, Belgium

[2]Laboratory for Clinical and Experimental Neurophysiology, Neurobiology and Neuropsychology (LCEN3), Department of Neurology, Ghent University, Belgium

Introduction

Temporal lobe epilepsy (TLE) is the most common form of epilepsy in adults. Research has shown that abnormal functional brain networks could be involved in the development of epilepsy and its comorbidities[1]. Gaining more insight into these networks can be useful for the development of new therapies. Resting-state functional magnetic resonance imaging (rs-fMRI) can visualize changes in functional networks on a whole-brain level[2]. In this study, we aim to map changes in functional networks during epileptogenesis in the intraperitoneal kainic acid (IPKA) rat model for TLE using longitudinal resting-state fMRI and graph theory. Additionally we investigate whether these changes are related to the severity of epilepsy, quantified with electroencephalography (EEG).

Subjects and Methods

Twenty-four adult male Sprague-Dawley rats (276 ± 15 g body weight) were used in this study. Seventeen animals were intraperitoneally injected with kainic acid (KA) according to the protocol of Hellier et al. (1998)[3] resulting in status epilepticus (SE). The other 7 animals were injected with saline and used as a control group. Rs-fMRI images were acquired before the KA injections and at 5 time points during the development of epilepsy: 1, 3, 6, 10 and 16 weeks after SE. At each time point an anatomical TurboRARE T2 image and three resting-state blood-oxygen level dependent (BOLD) fMRI images (TR=2s, TE=20ms, 300 repetitions) were acquired on a 7T system (Bruker PharmaScan). During image acquisition animals were anesthetized with medetomidine. The fMRI images were corrected for slice timing and motion, normalized to a template, smoothed with a Gaussian kernel (FWHM=0.8 mm), and band-pass filtered (0.01-0.1 Hz) using SPM12. The mean time series of 38 predefined regions of interest (ROIs) were extracted from the preprocessed images and the Pearson correlation coefficient between each pair of ROIs was calculated and stored in a correlation matrix using a graph theoretical network analysis toolbox (GRETNA)[4]. Different thresholds were applied to the correlation matrix to remove the weakest connections, resulting in 31 correlation matrices with a density ranging from 20% to 50%. Each of these matrices was visualized as a graph in which the nodes represent the ROIs and the edges the correlation coefficients between the time series of the ROIs. Several network measures were calculated at each time point, including clustering coefficient and local efficiency (measures of segregation), characteristic path length and global efficiency (measures of integration), and small-world coefficient. The mean value was calculated over the range of densities and plotted as a function of time to visualize how the properties of the functional networks change during the development of epilepsy. After the acquisition of the fMRI images, the animals were implanted with electrodes in the left and right dorsal hippocampus. EEG was measured during 8 consecutive days and several parameters, including seizure frequency and mean seizure

duration, were calculated. The relation between these parameters and the network measures was assessed using the Pearson correlation coefficient.

Results and Discussion

In Fig. 1 the distribution of the correlation coefficients is shown at different time points during the development of epilepsy in the IPKA rat model and in control animals. The correlation coefficients shift to smaller values during epileptogenesis. This indicates that network connections progressively become weaker during the development of epilepsy. In Fig. 2A and 2B clustering coefficient and local efficiency are shown. Both decrease during epileptogenesis, indicating a decrease in segregation or local interconnectivity in the functional brain network. Fig. 2C and 2D show that characteristic path length increases and global efficiency decreases during epileptogenesis. This indicates that the integration in the brain network decreases, so there is a decrease in overall communication efficiency. The correlation between seizure frequency and 2 global network measures 16 weeks after SE is visualized in Fig. 3. Seizure frequency is positively correlated with clustering coefficient and negatively with characteristic path length at this time point. This indicates that a larger decrease in functional segregation and integration during epileptogenesis is associated with a lower seizure frequency.

Conclusion

The results of this study show that functional brain network connections progressively become weaker and that segregation and integration of the network are decreased during epileptogenesis. The decrease in segregation and integration 16 weeks after SE is negatively correlated with seizure frequency. Further research is necessary to obtain more insight into the biological mechanisms underlying these findings.

Summary for lay people

Abnormal functional brain networks could be involved in the development of temporal lobe epilepsy (TLE). In this study, changes in these networks during the development of epilepsy were investigated in a rat model for TLE. Resting-state functional magnetic resonance images were acquired at several time points during epileptogenesis to identify functional networks. Our results suggest that network connections in the functional brain network progressively become weaker during the development of epilepsy. We also find a decrease in local interconnectivity and overall communication efficiency in the network and this decrease is inversely proportional to the severity of epilepsy.

References

1. Chiang, S. & Haneef, Z. Graph theory findings in the pathophysiology of temporal lobe epilepsy. *Clin. Neurophysiol.* **125,** 1295–1305 (2014).

2. Hutchison, R. M., Mirsattari, S. M., Jones, C. K., Gati, J. S. & Leung, L. S. Functional Networks in the Anesthetized Rat Brain Revealed by Independent Component Analysis of Resting-State fMRI. *J. Neurophysiol.* **103,** (2010).

3. Hellier, J. L., Patrylo, P. R., Buckmaster, P. S. & Dudek, F. E. Recurrent spontaneous motor seizures after repeated low-dose systemic treatment with kainate: assessment of a rat model of temporal lobe epilepsy. *Epilepsy Res.* **31,** 73–84 (1998).

4. Wang, J. *et al.* GRETNA: a graph theoretical network analysis toolbox for imaging connectomics. *Front. Hum. Neurosci.* **9,** 386 (2015).

Acknowledgements

This research is funded by a Doctoral Grant Strategic Basic Research of Research Foundation – Flanders (FWO).

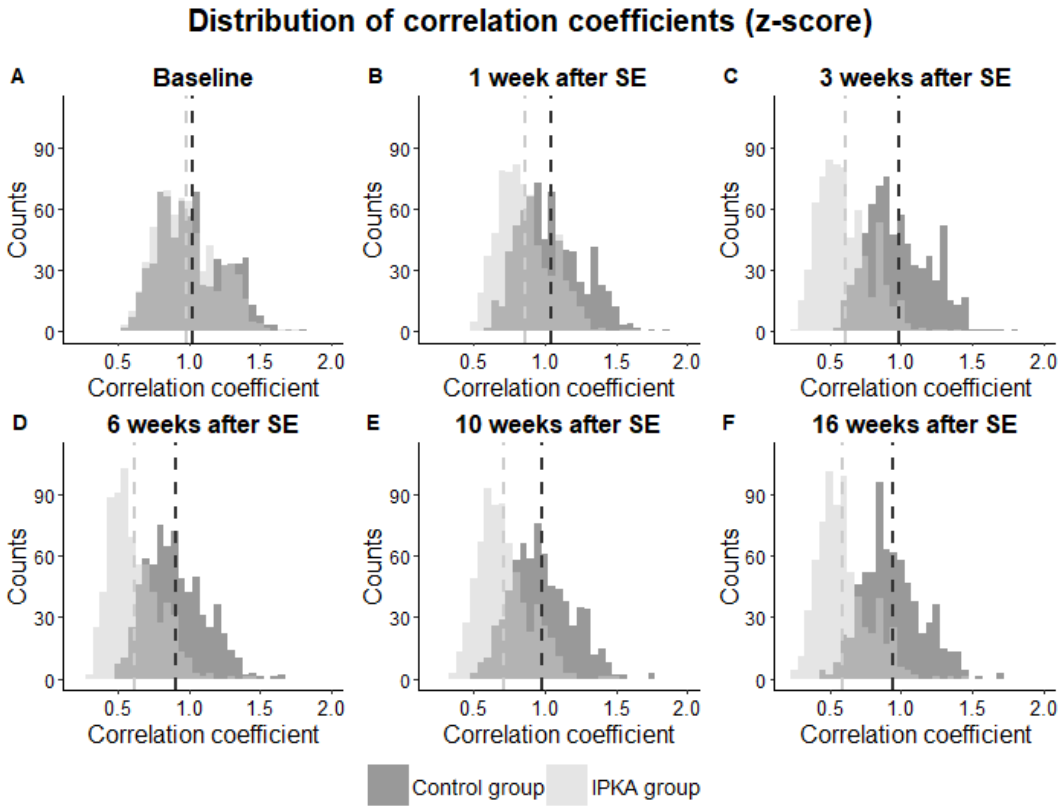

Figure 1: Distribution of correlation coefficients

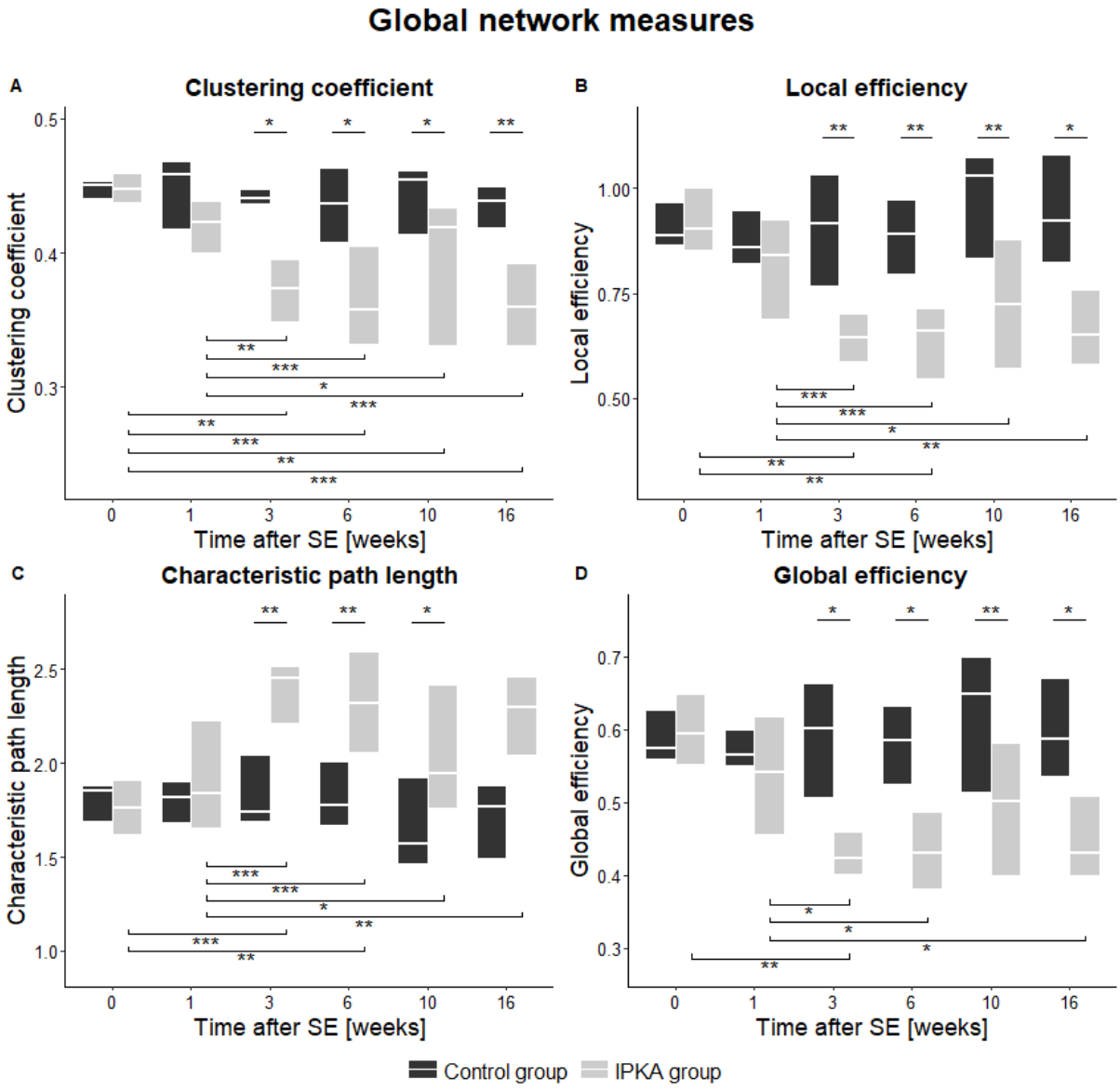

Figure 2: Global network measures

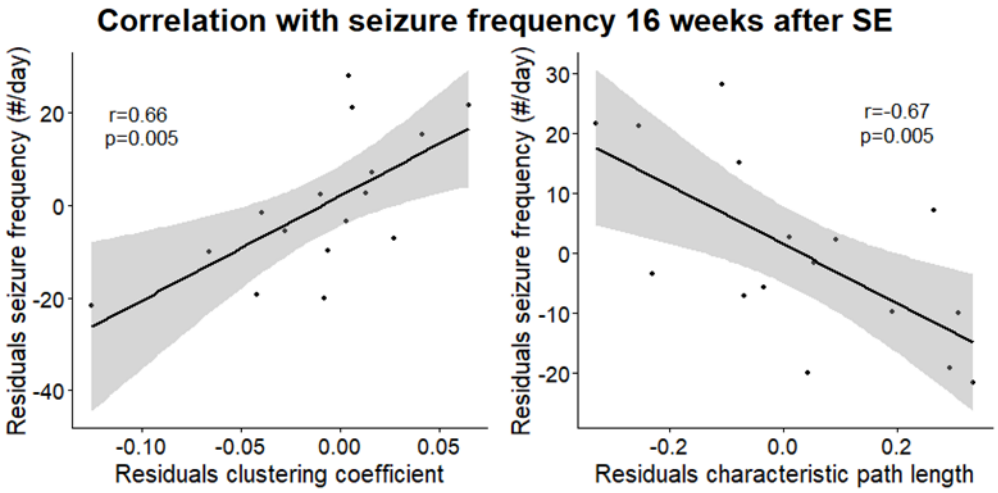

Figure 3: Correlation with seizure frequency 16 weeks post-SE