

Functional and anatomical changes of somatosensory cortex in rats recovering from severe neonatal hypoxic-ischemic encephalopathy (HIE) brain injury

Jae-Hun Kim¹, Geun Ho Im², Won Beom Jung², Moon-Sun Jang², Kyoung-Nam Kim², Julius Juhun Chung³, Sun Young Chae³, and Jung Hee Lee¹

¹*Radiology, Samsung Medical Center, Seoul, Seoul, Korea*, ²*Center for Molecular and Cellular Imaging, Samsung Biomedical Research Institute, Samsung Medical Center, Seoul, Seoul, Korea*, ³*Samsung Advanced Institute for Health Sciences & Technology, Sungkyunkwan University, Seoul, Seoul, Korea*

Introduction

Encephalopathy due to hypoxic-ischemic events occur when blood supply to the brain is interrupted, leading to global brain injury¹. Previous studies showed that hypoxic-ischemic encephalopathy (HIE) in the neonatal brain results in a prolonged evolution of injury with neurons dying days and weeks after the primary insult². However, at chronic stage, the mechanism of the spontaneous recovery from hypoxic-ischemic brain damage still remains unanswered. There were many studies of brain plasticity after focal ischemic brain damage³, but few study about brain plasticity after severe neonatal hypoxic-ischemic brain injury using MRI. In this study, we examined the functional and anatomical brain plasticity of somatosensory cortex in neonatal HIE rats model using functional MRI and DTI.

Methods

Hypoxic-ischemic encephalopathy (HIE) model in neonatal rat brain: Seven-day-neonatal rats were used to create the hypoxic-ischemic brain injury in the right hemisphere using the Rice-Vannucci model. Rats receiving the ligation of common carotid artery (CCA) in the right hemisphere were exposed to 8% oxygen at 37°C for 2.5 hours. For sham-operated group, ligation was not performed (n=6). Rats having above 80% of infarct area within the right hemisphere at 9 weeks after inducing hypoxic brain injury were selected as the HIE group (n=6).

Acquisition of MRI Data: The MRI data were obtained at 9 week after inducing hypoxic-ischemic brain injury in the HIE and sham-operated rats using a 7.0 Tesla MRI scanner (Bruker Biospin GmbH, Ettlingen, Germany). For MRI experiments, rats were initially anesthetized using an artificial ventilation system, and the body temperature was maintained at 36°C±1. T2 weighted MR (TR/TE=3000/60 ms, resolution = 120×120 μ m², slice thickness=1.5 mm), BOLD fMRI (TR/TE=1000/60 ms, resolution= 470×470 μ m², slice thickness = 1.5 mm), resting state fMRI (300 volumes) and DTI (TR/TE=4500/37 ms, resolution= 234×234 μ m², slice thickness = 0.75 mm, gradient direction = 30, gradient duration (δ) = 5ms, gradient separations (Δ) = 15ms, b-values = 1000 s/mm²) were acquired for each rat. Electrical stimulation was applied into forepaw of the rats to activate somatosensory cortex using block design (20 sec pre-stimulus, 20 sec stimulus and 40 sec post-stimulus period).

MRI data analysis: All MRI data were analyzed using AFNI and FSL software. For tractography, the Diffusion Toolkit software was used. All MRI data were spatially normalized into rat template space for analysis. To investigate the functional and anatomical changes of somatosensory cortex in rats recovering from hypoxic-ischemic brain injury, we found the brain activation responding to the electrical stimulation. With the seed ROI of somatosensory areas, which was identified by analyzing of BOLD fMRI data, we examined the functional and anatomical connectivity of somatosensory cortex using the resting state fMRI and diffusion tensor data, respectively.

Behavioral test: We performed the adhesive removal test to assess somatosensory (time to touch a tape) and sensorimotor (time to remove a tape) ability in rats before MRI experiments.

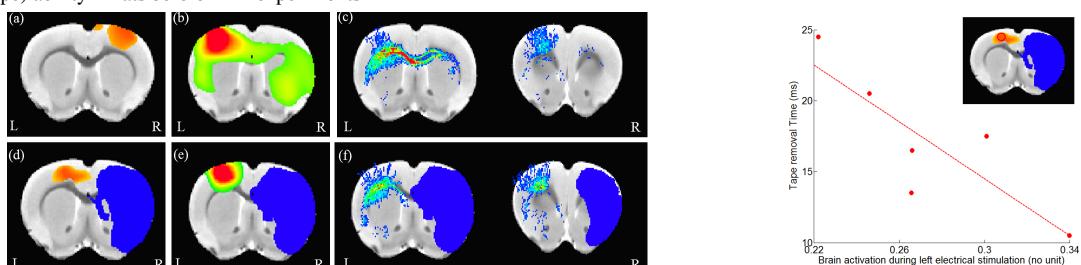


Figure 1. The functional and anatomical changes of somatosensory cortex in the sham-operated (a, b and c) and the HIE rats (d, e and f). First column represents the brain activation responding to the left forepaw stimulation (BOLD fMRI). Second column represents the functional connectivity of somatosensory cortex (Resting state fMRI). Third column represents track density maps associated with the somatosensory cortex (DTI).

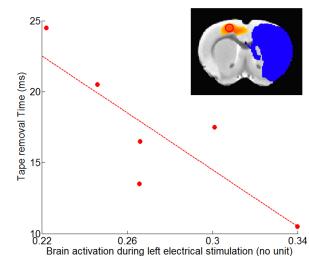


Figure 2. Correlation between the amplitude of brain activation responding to the left forepaw stimulation and the time to remove tapes in the HIE rats.

Results

BOLD fMRI (hemodynamic response): For electrical stimulation in the right forepaw, all rats showed brain activation in the somatosensory cortex in the left hemisphere. For electrical stimulation in the left forepaw, we observed the brain activation of the right somatosensory cortex in the sham-operated rats (Fig. 1a), whereas the HIE rats showed the brain activation of the left somatosensory cortex (Fig. 1d).

Resting state fMRI (functional connectivity): In resting state fMRI data, we found that the connection of the left somatosensory cortex was functionally connected to the right hemisphere including primary and secondary somatosensory cortex in the sham-operated rats (Fig. 1b). In the HIE rats, however, we found no functional connectivity between left and right somatosensory cortex (Fig. 1e).

DTI (anatomical connectivity): In diffusion tensor data, we found the same patterns of connectivity as the resting state data. Additionally, we found more fiber tracks extending into frontal brain in the HIE rats than in the sham-operated rats (Fig. 1c and f).

Behavior test: In adhesive-removal tape test, we found no significant changes of the right limb, whereas we found significant changes of the left limb between the HIE and the sham-operated rats ($P<0.01$). We found a significant negative correlation between the amplitude of BOLD activation in the left hemisphere responding to the left forepaw stimulation and tape removal time in the HIE rats ($r=-0.84$, $P<0.05$; Fig. 2).

Discussion and conclusion

In this study, we examined the functional and anatomical changes of rats recovering from severe hypoxic-ischemic injury using BOLD fMRI, resting state fMRI and DTI. In HIE rats, we found the brain activation in the left hemisphere for the left forepaw stimulation to compensate for loss function of the right somatosensory cortex showing the negative correlation of tape removal time. The inter-hemispheric connection was functionally and anatomically disconnected in the HIE rats, but we found more fiber tracks in frontal brain in the HIE rats than sham-operated rats, which may be associated with brain plasticity. Thus, we observed the inter- and intra-hemispheric brain plasticity, which refers to the ability of the brain to compensate for loss function through reorganization of neural networks.

References

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