Assessment of Lithium Treatment using fMRI During Stroke Recovery in Rats

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ABSTRACT An important function of lithium as a neuroprotective agent has been documented previously.[1] Significant reduction in infarct size and DNA damages with the corresponding improvement of neurological scores were reported as results of post stroke lithium administration. In the current study, therapeutic efficacy of chronic lithium treatment[2] was assessed by various MRI-derived parameters (i.e., apparent diffusion coefficient (ADC), fractional anisotropy (FA), and vessel size index (VSI)) and quantified with fMRI activity using electrical stimulation of rat forelimbs. The fMRI responses were examined by measuring the local hemodynamic MRI signal intensity based on blood oxygenation level dependence (BOLD) and cerebral blood volume weighted (CBVw) responses. The ipsilesional fMRI activations in the somatosensory (SS) cortex of lithium-treated and saline-treated rats two weeks after middle cerebral artery occlusion (MCAO) were compared. ADC and FA value of local brain tissues were significantly correlated with the magnitude of fMRI responses for the lithium-treated rats while no such correlations were seen in the saline controls. However, neurological score also did not show any significant improvement with the treatment. The ipsilesional/contralesional BOLD signal intensity response ratios of lithium-treated rats were larger than those of saline-treated control rats. In contrast, the CBVw response ratios were similar between two groups. These results demonstrated that the lithium-treatment of post-stroke animal models positively enhanced BOLD fMRI response in the lesion hemisphere.

MATERIALS AND METHODS After 90 minutes of focal cerebral ischemia (MCAO), rats received a subcutaneous injection of either LiCl (n=6) (Sigma-Aldrich, St. Louis, Missouri, USA), dissolved in normal saline (injection dosage, 1 mM LiCl/kg rat, i.e. 0.3 ml of 1 M LiCl -solution for a rat of 300 grams), or the same volume of normal saline (n=6) (0.9 % NaCl) 12 hours after the operation. On day 15 following a transient MCAO, fMRI was performed in both stroke and control groups. The fMRI activation of both BOLD and CBVw responses (Gradient Echo Planar Imaging, TR/TE = 3700/15 ms for BOLD, TR/TE = 3700/11 ms for CBV, FOV = 2.5x2.5 cm^2; nine 1 mm slices, and 80x80 matrix zero filled to 128x128) was acquired using a horizontal bore 9.4T Bruker/Magnex system, equipped with a home-built surface coil. An unilateral electrical stimulation paradigm, consisting of 3 periods of 37 sec 'stimulation on' separated by 185 sec 'stimulation off,' was alternated between the left and right forepaw and was repeated at least 2 up to 5 times. Following BOLD fMRI, MION was intravenously administered (36mg (FeO_2)/kg), and the stimulation paradigm was repeated using CBVw fMRI. Functional activation maps were generated by a voxel by voxel t-test between the on and off stimulus periods. Prior to the contrast agent administration, T2 and T2* maps were created by conventional gradient echo and spin echo pulse sequences with multiple echoes where TR/TE=1000/[4, 7, 10, 13ms] and TR/TE=3000/[15, 30, 45, 60, 75, 90, 105, 120ms] were used, respectively. Thereafter, the lesion size was determined from the calculated T2 maps. For further structural analysis, ADC maps were created with a diffusion weighted EPI pulse sequence with TR/TE=3700/40ms and b=5, 300, 800, and 1200 sec/mm^2. In order to obtain FA, 6-direction diffusion tensor imaging was performed with the b values of 0 and 1200 sec/mm^2. Following a MION administration, T2 and T2* maps were created again as described above for the calculation of relative cerebral blood volume (rCBV) and VSI. Functional activation maps were computed voxel by voxel using an equivalent t-test between the on and off stimulus periods. The statistical threshold for significant activation response was $p<0.01$ with a Bonferroni correction for multiple comparisons throughout the measured brain volume. For all the structural and percent signal change analyses, ROIs were placed over the voxels pertaining to primary motor cortex (M1) and forelimb sensory areas (S1fl) based on the rat brain atlas.

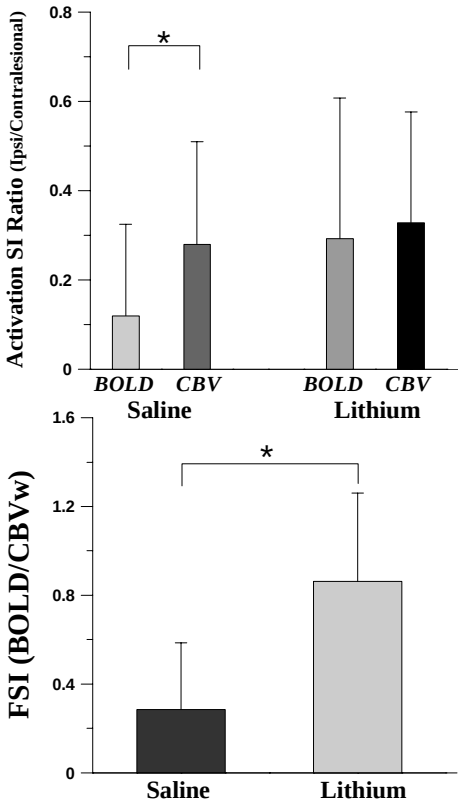

Figure 1. activation SI ratio and functional status index in somatosensory cortex.

RESULTS AND DISCUSSION Varying degree of ipsilesional fMRI responses was observed in both lithium- and saline-treated groups. Overall activation volume and magnitude were larger for saline-treated rats than the lithium-treated group in both contra and ipsilesional somatosensory cortices. However, for both BOLD and CBVw responses, the mean activated volume ratio (ipsilesional/contralesional) was higher for lithium-treated rats than controls. The CBVw activation signal magnitude ratios (Figure 1 upper panel) were significantly higher than BOLD ratios for the control animals; however, similar for the lithium-treated rats. As shown in Figure 1 (bottom panel), functional status index (FSI) which represents the mean ratio between BOLD and CBVw ipsi/contralesional activation signal intensity ratio for the individual somatosensory cortex was significantly higher for lithium-treated rats than the control group. From the acquired ADC maps, ex vacuo dilation of the ipsilesional lateral ventricles was identified for most of rats (Figure 2) while other structural damages (e.g., FA) were also evident at this late stage (2 weeks) of recovery. Among the acquired structural parameters, ADC and FA were significantly correlated with CBVw fMRI activation magnitude (i.e., area under the curve) for the lithium-treated rats while no such correlations were found for the saline controls. When compared to the contralesional values, the ipsilesional mean VSI ratio (ipsi/contra) was larger for the lithium-treated rats; however, the micro vascular volume (i.e., $\Delta R2$) ratio (ipsi/contra) was larger for control rats. These results imply that vascular transformation affects fMRI characteristis.

REFERENCES

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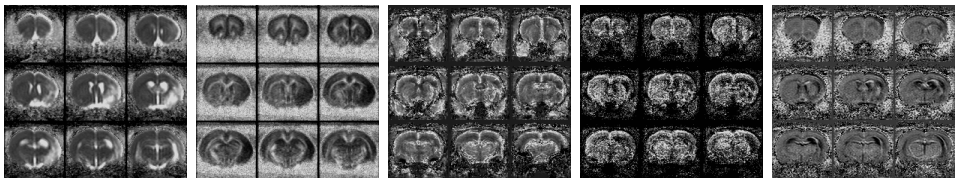

Figure 2. MRI-derived structural maps of the lithium-treated rats (from left to right: ADC, FA, rCBV, VSI, and $\Delta R2$ (or micro vascular volume))