

## Histopathological evaluation on infarct core, peri-infarct border and intrahemispheric remote area in T2 weighted and diffusion weighted MRI images in the rat brain.

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### Introduction

Recently, magnetic resonance imaging (MRI) has become the most important tool for acute stroke. Also, diffusion weighted imaging has been used to decide the indication of thrombolytic therapy in acute cerebral infarction. Temporal changes on infarct region in T2 weighted and diffusion weighted MRI images have been discussed well in humans. However, regional histopathological changes especially in peri-infarct border showing apparently normal on the MRI images have not been discussed well. The purpose of this study is to evaluate the regional histopathological changes in infarct core, peri-infarct border and intrahemispheric remote area based on the T2 weighted and diffusion weighted MRI images using unilateral middle cerebral artery occlusion rat model.

### Methods

Under pentobarbital sodium anesthesia, the right middle cerebral artery of male Sprague-Dawley rats (250-300g in weight) was occluded permanently by inserting a 4-0 nylon monofilament suture via the right common carotid artery. After 24 hr, coronal section imaging of T2 weighted image (FSE, TR=4000ms, TE=106ms, FOV=10x5cm, matrix=192x192, slice thickness=3mm) and diffusion weighted image (Single Shot Fast Spin Echo (SSFSE); less susceptibility artifact, TE=108ms, FOV=10x10cm, matrix=96x96, slice thickness=3mm, b=900s/mm<sup>2</sup>) which we made our original device, 3 inch surface coil up and down arranged, as dual array coil were taken using 1.5T MRI (SIGNA HORIZON LX Ver.8.2.5, GE) under pentobarbital sodium anesthesia. Immediately after the MRI imagings, the rats were decapitated and brains were removed. Brains were fixed with 10% formalin solution overnight, thereafter, each specimen was cut into coronal slabs. Histology sections were stained with hematoxylin-eosin. We analyzed the 12 rats which showed high intensity areas on unilateral caudate putamen, insular and parietal cortex (large middle cerebral artery area) in T2 weighted and diffusion weighted MRI images except for rats with caudate putamen infarction only or normal findings. Based on the MRI images, we performed histopathological evaluation in the following three regions; infarct core (T2 and diffusion weighted high intensity areas), peri-infarct border (cortex adjacent to infarct core that appeared normal on both T2 and diffusion weighted images), and intrahemispheric remote area (frontal cortex within the anterior cerebral artery territory) on the same coronal slices (1). The definition of necrotic neuron was derived from criteria formulated by Farber (2), Eke (3) and Garcia (4) and was based on the identification of either pyknosis/eosinophilia neurons (red neurons) or complete loss of hematoxylinophilia (ghost neurons). Other cellular alterations such as dark, scalloped and swollen neurons were included in the neuronal counts. In each rat coronal histology section representing the level of the anterior commissure was examined at a magnification of x400. Using digital still camera interfaced with an Olympus microscope, medial and lateral region from the neocortex on those three region in each rat were collected. We quantified the number of necrotic and intact neurons, and calculated the percentage of necrotic neurons.

### Results

In all examined rats, the extent of high intensity areas in diffusion weighted images were almost as same as in T2 weighted images after 24 hrs of occlusion. However, the extent of infarctions were different in each rat. For histopathological evaluation, there were no intact neurons in the infarct core showing high intensity areas in diffusion and T2 weighted images. In MRI normal regions, there were no necrotic neurons in the intrahemispheric remote areas, although isolated or widespread necrotic neurons with intact neurons were found in peri-infarct borders. The percentage of necrotic neurons in peri-infarct borders were different in each rat (maximum: 51.3 %, minimum: 7.1 %, mean: 23.9%(SD:14.1%)). The more percentage of

necrotic neurons peri-infarct borders had, the more extent areas infarct core had.

### Discussion

In acute cerebral infarction, the neurological findings and extent of infarction on MRI images are sometimes inconsistent. Also, in acute embolic stroke, final outcome of the area in ischemic sublethal injury is sometimes indeterminate just after the thrombolytic therapy or natural reperfusion. In this study, we evaluated regional histopathological changes especially in peri-infarct border showing apparently normal on the MRI images using unilateral middle cerebral artery occlusion rat model. In peri-infarct border that appeared normal on both T2 and diffusion weighted images, intact neurons and necrotic neurons were coexisted. The peri-infarct border were presumed to be incomplete infarction (5), in other word, viable tissue; ischemic penumbra (6). The percentage of necrotic neurons in the peri-infarct border were tended to be higher in rats with more extended infarct core. Further studies that measure both hemodynamic response and neuronal activity with perfusion and functional MRI would allow a more accurate characterization.

### References

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