Brain Tissue Ischemic Transitions during Permanent Middle Cerebral Artery Occlusion (pMCAO) in Rats

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INTRODUCTION

Therapeutic intervention for acute stroke presupposes the presence of salvageable tissue that can be rescued from further or evolving injury. Recent clinical trials suggest that evidence of salvageable tissue, classically known as the "ischemic penumbra" by qualitative diffusion/perfusion mismatch is indicative of greater neurologic improvement when recanalization occurs [1,2]. However, comparison of qualitative diffusion/perfusion mismatch with PET oxygen extraction fraction (OEF) showed that the MRI defined mismatch is inaccurate [3] which we believe may be resolved by the correlation between quantitative measurements of perfusion by arterial spin labeling (ASL) and apparent diffusion coefficient (ADC) in voxel plots as previously reported by Shen et al [4]. Using these methods, our aim was to identify tissue ischemic transitions for up to three weeks after permanent middle cerebral artery occlusion (pMCAO) in the rat.

MATERIALS AND METHODS

Sprague-Dawley rats (n = 6) 10 to 15 weeks in age were used for this study. Isoflurane (1:1 O_2/N_2O) anesthetized rats were intubated, mechanically ventilated and femoral catheters were inserted. Baseline images were obtained following which the rats underwent pMCAO by the insertion of 4-0 suture into the internal carotid artery. Body temperature was maintained at 37 ± 0.5 ˚C using warm air, regulated with a rectal temperature probe. During each MRI study, $PaCO_2$, PaO_2, MABP, HR and rectal temperature were monitored at the time of the initial scans after pMCAO but not in later scans on subsequent days. The rats were subjected to pMCAO followed by MRI scans hourly for the first four hours and then their brain perfused-fixed for neuropathological analysis or they were scanned at one hour after pMCAO and recovered for scans at 7, 14 and 21 days later followed by brain perfusion-fixation after 21 days.

MR studies were performed on a 4.7-Tesla, 40cm bore Bruker Biospec system, equipped with a 12 cm diameter shielded gradient insert. A two coil system was used, a 72 mm volume coil and an actively-decoupled surface coil (2-3cm ID). ADC maps were acquired in the x, y or z direction with a single shot, SE-EPI sequence with a TR = 2 s, 64 x 64 matrix, FOV = 2.3 cm, 90˚ flip angle, b = 10, 500, 1500 s/mm^2, Δ = 15 ms, δ = 5 ms, and 16 averages. ADC = $-\ln(S_i/S_0)/(b_i - b_0)$, where $b_i = \gamma^2 G_i^2 \delta^2 (\Delta - \delta/3)$. Continuous ASL was used to quantify CBF [5]. A single shot, SE-EPI sequence with a TR = 2 s, 64 x 64 matrix, FOV = 2.3 cm, 2 s labeling pulse, with labeling applied ± 2 cm from the imaging plane. Maps of T_1 were generated from spin-echo images with variable *TR* (*TR* = 9100, 8500, 7900, 7300, 6700, 6100, 5500, 4900, 4300, 3700, 3100, 2500, 1900, 1300, 700, 100 msec, FOV = 2.3 cm, 4 averages, 164 x 64 matrix). CBF maps were generated from: $CBF = \lambda \cdot (T_{1obs} \cdot 2\alpha)^{-1} \cdot (M_C - M_L) \cdot (M_C)^{-1}$, where M_C and M_L are the magnetization intensities from the control and labeled images, respectively. A spatially constant value of 0.9 mL · g^{-1} was assumed for the blood brain partition coefficient for water (λ) and a spin-labeling efficiency (α) of 0.7 was assumed.

RESULTS AND DISCUSSION

A representative data set from an animal subjected to pMCAO shows that pre-stroke CBF was highly heterogeneous ranging from 220-300 mL/100g/min with higher CBF in the thalami, consistent with isoflurane anesthesia [6]. Following pMCAO, CBF fell in the ipsilateral hemisphere in the parietal, cingulate, and pyriform/amygdala cortices and remained reduced in these areas for up to 12 days post pMCAO. Pre-stroke ADC values had a mean of 0.78 ± 0.06 x 10^{-3} mm^2/s, similar to previously published data [4]. After pMCAO, ADC decreased to 0.54 ± 0.08 x 10^{-3} mm^2/s up to 7 days. At 14 and 21 days following occlusion ADC increased to 1.6 ± 0.04 x 10^{-3} mm^2/s. Voxel plots relating cerebral blood flow (CBF) and apparent diffusion coefficient (ADC) with pMCAO without reperfusion revealed a reduction in CBF without changes in ADC but by 21 days a progressively greater number of voxels characterized by low CBF and high ADC indicating tissue infarction and dissolution.

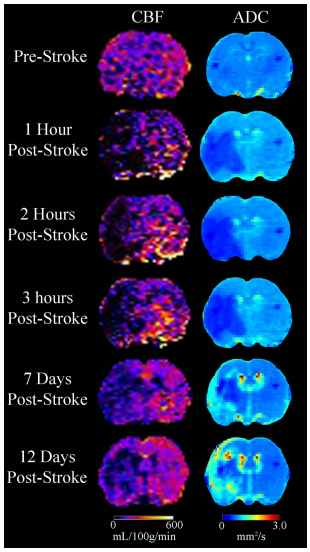

Fig. 1. Representative CBF and ADC maps from one animal before and after pMCAO.

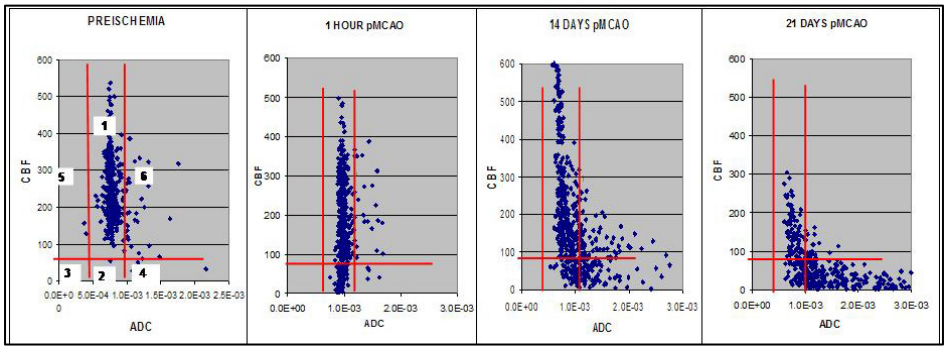

Fig. 2. Quantative CBF versus ADC voxel plots in a rat with scans before and after pMCAO illustrating the ischemic transitions of the tissue through the various phases numbered in the left panel. 1 = Normal ADC, CBF; 2 = ischemic penumbra, ischemic CBF, normal ADC; and 3 = ischemic core, low CBF , low ADC. We hypothesize that 4-6 represents various phases of infarction: 4 = low CBF, high ADC; 5 = normal to high CBF, low ADC; and 6 = hyperemic infarction, normal to high CBF, high ADC. Vertical red lines upper and lower bounds (± 2SD) of ADC and the horizontal line the lower threshold of CBF (- 2SD).

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