Neuroprotective effects of heat shock proteins - an MRI study

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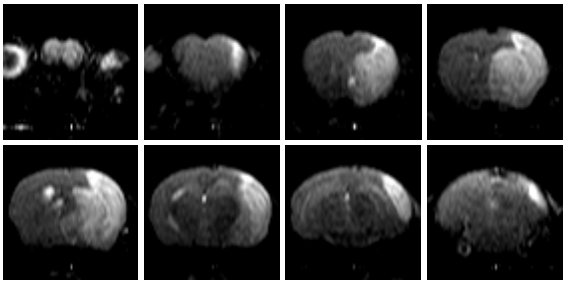
Introduction Heat shock proteins (HSPs) have been reported to increase cell survival in response to a wide range of cellular challenges. In order to investigate the mode of action of these proteins *in vivo*, transgenic mice overexpressing HSP70 have been compared to wild-type mice in a middle cerebral artery occlusion (MCAO) model of permanent cerebral ischaemia. To assess the contribution of HPSs following permanent ischaemia, difffusion maps of a 2 mm slice within the MCA territory were acquired at different time points after the onset of stroke, and the total lesion size was estimated after 24 hours of ischaemia using a multi-slice T2-weighted scan.

Methods 6 HSP70 transgenic mice and 5 WT mice were anaesthetised with 2.5% isoflurane and maintained on 1.75% isoflurane with pure oxygen. The MCA was permanently occluded by advancing a 180-µm-diameter filament into the internal carotid artery past the MCA junction. Coronal images were obtained approximately -1 mm from bregma at 1.5 hours and 24 hours after the onset of stroke. A 2.35T horizontal bore SMIS MR scanner was used with the following imaging parameters: FOV 32 mm, 2 mm slice thickness, and 128 × 64 pixels. Quantitative apparent diffusion coefficient (ADC) data were obtained using 2 trace-weighted spin-echo EPI images with b=38 and b=1187 s/mm^2. Also, at 24 hours multi-slice data were measured using a T2-weighted SE sequence with FOV 20 mm, 1 mm slice thickness, 9 slices and 128 × 64 pixels. The relative infarcted area per slice was defined as the ratio of the lesion area and the size of the contralateral hemispere. Similarly, the ADC of the affected hemisphere was expressed relative to the contralateral side.

Results

(A)



(B)

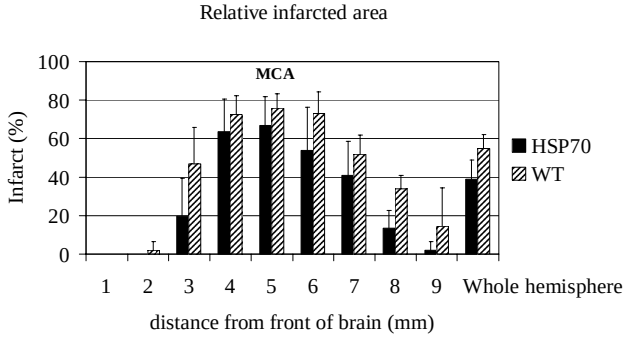


Fig. 1. A. T2-weighted images at 24 hours after onset of stroke, showing the lesion area. B. Relative infarcted area for WT and HSP70 transgenic mice.

Figure 1A shows a typical infarct caused by permanent MCA occlusion. The relative infarcted area (Fig. 1B) is larger in the WT mice than in the HSP70-overexpressing animals for every slice ($P < 0.05$ using Degrees of freedom adjusted repeated measures ANOVA). Overall, the lesion is 55% in the WT mice compared to 39% in the HSP70 mice.

At 1.5 hour after the onset of stroke, the ADC in the basal ganglia is lower for the WT mice (Fig. 2). The core and periphery show no significant differences at this stage. Between 1.5 and 24 hours, the ADC continues to decrease for the HSP70 mice, whereas for the WT mice an increase is observed ($P < 0.05$ using a paired student t-test).

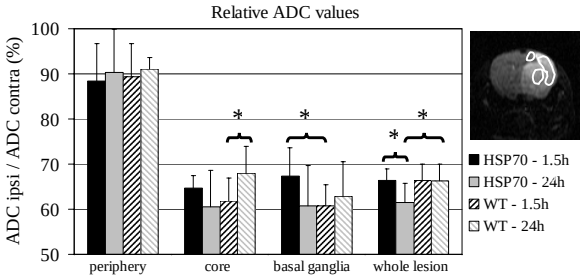


Fig. 2. Relative ADC values for selected areas within the lesion. Brackets indicate significant differences between groups ($P < 0.05$).

Discussion We have measured lesion size and ADC values in a permanent MCAO model to investigate whether heat shock proteins can protect the brain against ischaemia. With this particular model, the lesion in the HSP70 mice is 30% smaller than in the WT animals. The ADC decreases between 1.5 and 24 hours for the HSP70 mice, whereas for the WT mice the ADC levels off, or even increases between those two time points. Knight *et al.* studied the time dependence of ADC changes, demonstrating that tissues with the most severe histological damage showed the fastest ADC decrease and subsequent normalisation of ADC values. In the light of this paper, our findings suggest that the damage within the lesion may be less severe for the HSP70 mice. Thus, overexpression of HSP70 reduces the overall lesion size and may also limit the tissue damage within the lesion.

References Knight,R.A. *et al.* Magnetic resonance imaging assessment of evolving focal cerebral ischemia (1994) Stroke **25**, 1252-1262.

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