Ipsilateral deficit revealed by operant conditioning following right middle cerebral artery occlusion in mice

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Middle cerebral artery occlusion is an animal model of brain stroke that consistently leads to a striatal infarction [1]. The striatum is involved in several aspects of cognition, including **behavioural flexibility** (i.e., the ability to adapt behaviour in a changing environment) [2].

Alongside basic sensorimotor testing, we propose an operant-based task requiring flexibility in response orientation, in order to clarify the cognitive impairments following experimental stroke.

Animals and surgery. MCAo (N=5) or sham surgery (N=5) were performed on adult C57Bl/6H mice (N=5). Under isoflurane anaesthesia, right middle cerebral artery were occluded for 30 minutes. Functional testing began 3 weeks after surgery.

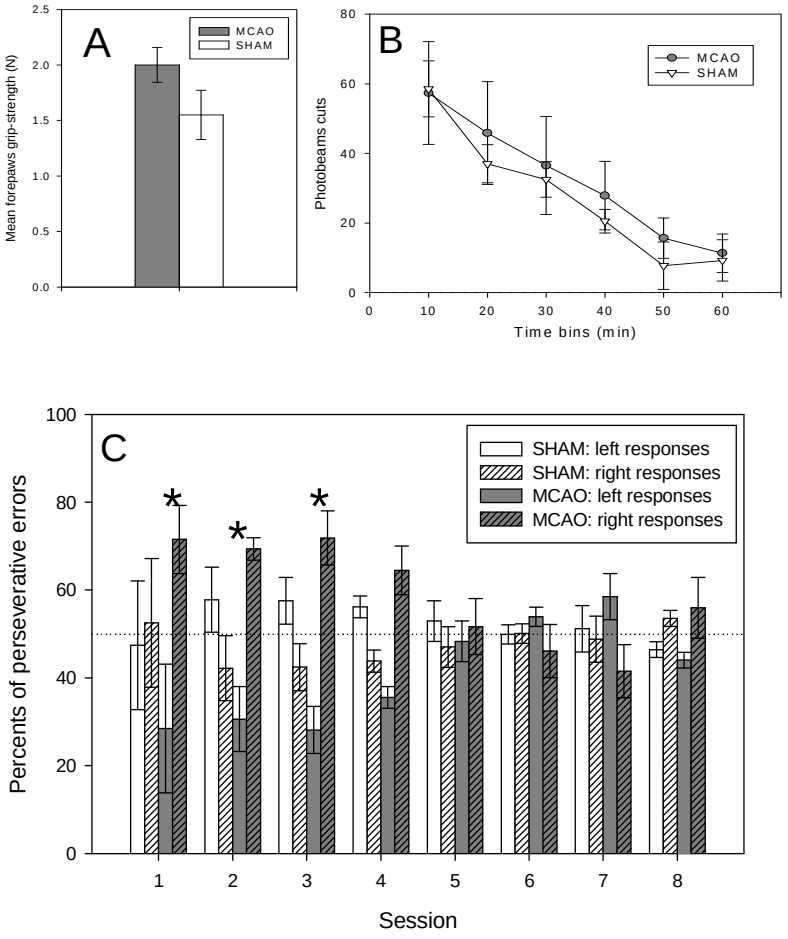
Grip-strength test. Forepaw grip-strength was measured over five trials using a dedicated apparatus (Bioseb©).

Open-field locomotion. Animals were placed in a transparent cage equipped with photoreceptors. The number of beam crossings provides a locomotion measure during an one hour session.

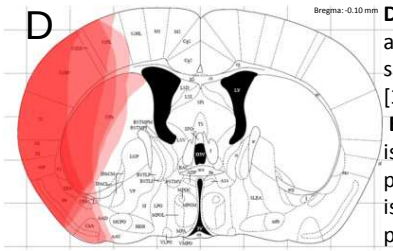
Behavioural flexibility. Conditioning took place in boxes equipped with two levers and a food-tray (Med Associates©). Following instrumental response shaping, behavioural flexibility was assessed in 8 sessions during which the two levers were available, but only one was associated with food reinforcer under a fixed-ratio 5 schedule; after each reinforcement, the active lever changed.

Histological analysis. Deeply anaesthetised mice were transcardially perfused with 4% paraformaldehyde. 50µm brain sections were incubated overnight with mouse Anti-NeuN (1:1000), then in secondary antibodies horse Anti-Mouse (1:400). After amplification with ABC reagent, cell nuclei were revealed, using DAB as chromogen. Lesion extents were estimated under microscope with Nikon NIH© software.

Statistics. Grip-strength data were treated with a non-parametric Mann-Whitney tests; a repeated measures ANOVA was applied to the measures of locomotion and operant learning.

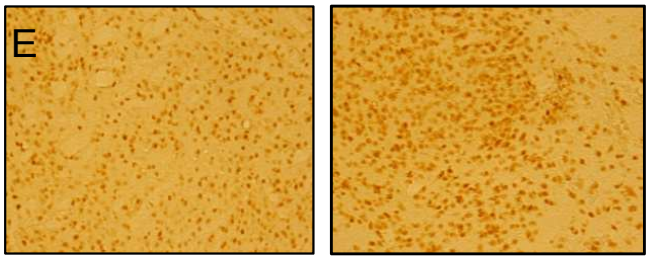


A. Grip-strength. No significant difference appears between MCAo and sham mice. **B. Open-field locomotion**. The two groups were statistically similar in their locomotor capacities. **C. Behavioural flexibility**. MCAo mice committed more perseverations in the ipsilesional hemifield compared to contralateral hemifield (indicated by * ; $p<0.05$); such a contrast was not observed in sham animals.



D. Ischemic infarct extends to a large part of the striatum, sensory and piriform cortices [3] .

E. Striatal cellular loss in the ischemic hemisphere (left panel) compared to the non-ischemic hemisphere (right panel)



If motor abilities and global learning performance were not affected *per se*, analysis of responses pattern revealed that MCAo caused an elevation of perseverative errors in the ipsilesional hemifield only, suggesting the existence of a **lateralised flexibility deficit**.

This finding could be explained either by a contralateral sensory neglect or by an inhibition deficit related to the ipsilesional side. Previous animal and human studies favour the **contralateral sensory neglect** hypothesis [4,5]. It is not excluded however that both mechanisms could be involved at different levels.

1.Belayev, L., et al., Brain Res, 1999. **833**(2): p. 181-90. 2.Ragozzino, M.E., et al., Behav Neurosci, 2002. **116**(1): p. 105-15. 3. Paxinos, G., et al., Academic Press, 2001. 4.Milton, A.L., et al., Behav Brain Res, 2004. **150**(1-2): p. 55-63. 5.Nys, G.M., et al., Brain, 2006. **129**(Pt 8): p. 2148-57.