HEMI-PARKINSON'S DISEASE RAT MODEL: Correlation Between Behaviour, Histology and MRI

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INTRODUCTION AND AIMS

Parkinson's Disease (PD) is a progressive neurodegenerative disorder characterized by the degeneration of dopaminergic neurons in the substantia nigra pars compacta, that produces a cascade of functional changes affecting the whole basal ganglia network [1]. The disrupted activity of pallidal and subthalamic neurons is considered to be responsible of PD symptoms including rigidity, tremor and akinesia. In PD patients the ability to perform movement sequences is greatly improved by auditory or visual cues. Bilateral lesions of the striatum results in impaired ability of self-initiated movements and preserved ability in externally cued movements [2]. The present experiment investigated the capability to switch from an internally to an externally cued task and vice versa, both in intact and unilateral 6- hydroxyl dopamine (6-OHDA) lesioned rats. The resulting nigro-striatal dopaminergic fibres degeneration correlates with neuronal loss in the ipsilateral substantia nigra. When 6-OHDA induced degeneration exceeds 70% an asymmetric and quantifiable motor asymmetry occurs [3-4] In this work, we have developed a specific behavioural paradigm in a hemi-PD rat model coupled with immuno-histological staining and MRI methods, to compare anatomical and physiological data. This animal model should be useful to investigate difficulties raising in PD patients in shifting between mental sets.

METHODS AND RESULTS

Male albino Wistar rats (n=8) were divided in two groups that followed two different training tasks using an operant chamber equipped with 3 horizontally placed levers, one adjacent to the others. A green LED was located above each lever. There were 2 speakers associated with two different tones. A food pellet dispenser was located to the opposite side of the chamber. The animal had to hold pressed the central lever at the presentation of the first tone until the occurrence of the second one. Pressing down of the second lever enabled the rats to receive the reward. The internally cued condition, task A (n=4) let the animals free to choose the lateral lever to press. The externally cued condition, task D (n=4) requested the rats to press the left or the right lateral lever in response to the occurrence of the corresponding LED. An incorrect response was not rewarded. After the training stabilization, two rats from each group underwent surgery. The animals were anesthetized and placed in a stereotaxic apparatus to receive unilateral right-side injection of 6-OHDA (5µg/µl, 3µl) through a Hamilton syringe (26G). After one-week recover period rats were tested again. The unlesioned (B, n=2) and lesioned (C, n=2) rats previously trained in the free condition were switched in the cued behavioural set. Inversely, the unlesioned (E, n=2) and lesioned (F, n=2) rats previously trained in the cued behavioural set were switched in the free condition. Performances were evaluated according to: percentage of correct responses (CR), reaction time (RT), calculated from the occurrence of the second cue and the release of the central lever, movement time (MT), ranging from the release of the central lever and pressing down of the lateral one. Statistical analysis was assessed by Anova post hoc Newmann Keuls test. Final confirmation of size and location of the lesion was achieved with *post-mortem* MRI and histological examination subsequent to cerebral tissue fixation. The whole brain was scanned with a 2.35 T MRI Biospec equipped with a TX-only volume coil and a RX-only surface coil of 2.5 cm in diameter. GEFI images (TR=3000 ms; TE=40 ms; FOV=1.5 cm^2; 256*256; slice thickness=1.1mm; NEX=18; TAQ=3h15min) were acquired covering the whole brain. Finally coronal sections (40µm) were cut using a freezing microtome and the sections were processed for histological (cresyl violet) or immuno-histological staining (TH^+).

The data reported in Fig. 1a, show the improved capability to recover from the lesion-induced neglect (*P<0.001, F vs D and E) to ipsilateral execution in the internally cued condition. The presence of the external stimuli prevented the performance deficit recorded in the free condition and removed (P=ns, C vs B and A) any ipsi- or contralateral differences in CR and RTs (Fig. 1b, §P<0.001, F vs D and E). The specificity of the free condition behavioural set was apparent in MTs trend to slow down in lesioned rats as well (°P<0.005, left F vs right F and D). As shown in Fig. 2, we found a correspondence between the MRI and the histological section, showing a lesion localized in the striatum with an evident shrinkage of the whole striatum and an enlargement of the ipsilateral ventricle. As shown in Fig. 2C, the lesion location and 6-OHDA dose induced nearly a complete loss of TH^+ immunoreactivity in the right striatum. Fig. 3 shows a significant retrograde dopaminergic denervation in the ipsilateral substantia nigra.

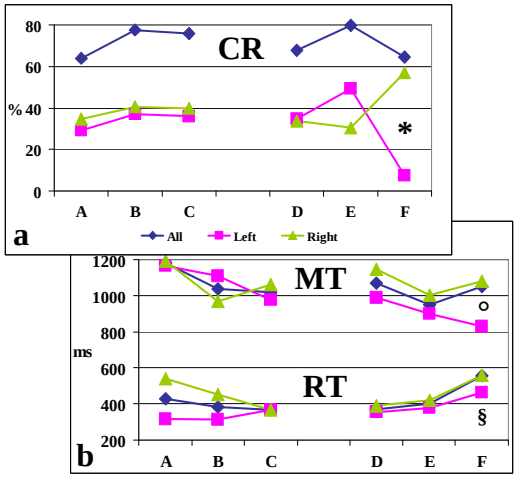

Fig.1. Effects of 6-OHDA striatal lesion on behavioural performance.

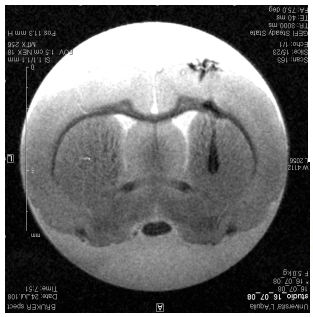

Fig. 2 Co-registered sections in the striatum obtained from (A) MRI, (B) CV and (C) TH+ histology.

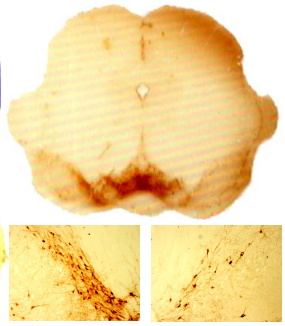

Fig. 3 TH+ section of the substantia nigra.

CONCLUSIONS

We made a comparison of behavioral, histological and MRI data. The behavioral set allowed investigating both cognitive and motor lesion-induced impairment in the hemi PD rat model. Our final aim is to use this methodology to evaluate *in vivo* anatomical and physiological manipulations in relation with behavior in the rat. These different methodological approaches should be useful both to investigate the impaired ability to initiate rather than maintain a new strategy as occurs in PD patients, and to test the hypothesis that basal ganglia play a role in the temporal encoding of behavioural control.

REFERENCES

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