Autologous neural cells ecosystem (ANCE) transplantation as therapy for Parkinson's disease: a promising approach

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Introduction

Parkinson's disease (PD) is the 2nd most common neurodegenerative disease charaterized by a progressive loss of dopaminergic neurons. Since the 70s, many laboratories explore dopamine neurons replacement as a therapeutic strategy in animal model of PD based on stem cell transplantations. Nevertheless, controversies regarding the use of stem cells (e.g. ethical and immune limitations) restrict their application. However, these limitations can be vanished by autologous cells transplantation. Bloch J. and colleagues (2014) demonstrated the potential of the transplantation of autologous neural cells ecosystem (ANCE) in parkinsonian monkeys. The present investigation intended to assess the impact of ANCE transplantation in four monkeys (*Macaca fascicularis*) exhibiting parkinsonian symptoms, focusing on the manual dexterity and the dopaminergic system state *in vivo*.

Methods

The monkeys were extensively trained to perform fine manual dexterity tasks as well as a reach and grasp drawer task before undergoing chemical lesions of the substantia nigra pars compacta (SNpc) with the neurotoxin MPTP (1-methyl-4-phenyl-1,2,3,6-tetrahydropyridine) in order to mimick the motor symptoms of PD. During the MPTP phase, small cortical biopsies from the dorsolateral prefrontal cortex (dlPFC) were performed and the gray matter material obtained was put into culture according to the protocol developed by Brunet et al. (2005). Additionally, at all phases of the protocol, the integrity of the nigro-striatal system was followed-up by ^{18}F-DOPA PET scan. At the end of the experiment, histological readout was performed with immunohistochemistry technique in order to reveal the state of the dopaminergic system.

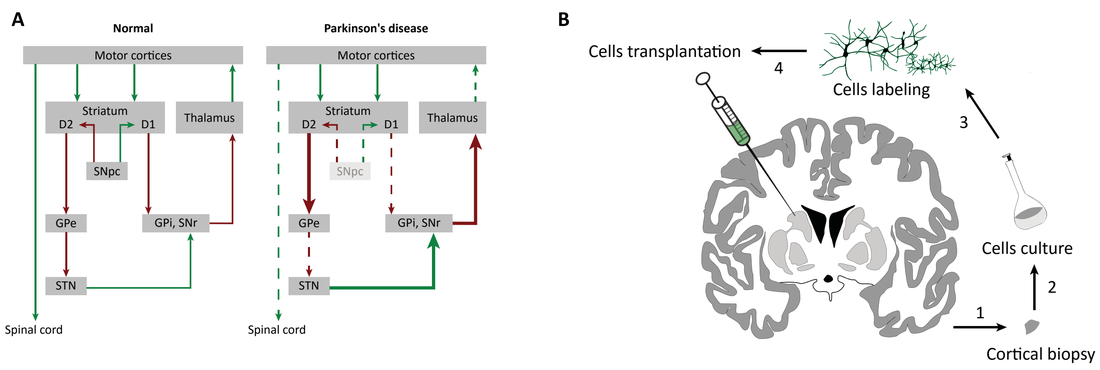


Figure 1: (**A**) Schematic illustration of the basal ganglia nuclei connectivities (red arrow represents an inhibiting synapse, green arrow represents an activating synapse). The left panel is the normal state while the right is a pathological state (hypokinetic, as in Parkinson's disease. (**B**) Therapeutic strategy of autologous neural cells ecosystem (ANCE) transplantation. The first step consists in performing a small cortical biopsy in the dorsolateral prefrontal cortex (dlPFC) in each monkey. Cells are then put in culture, labeled before being transplanted bilaterally into the striatum of the same subject (Brunet et al., 2005).

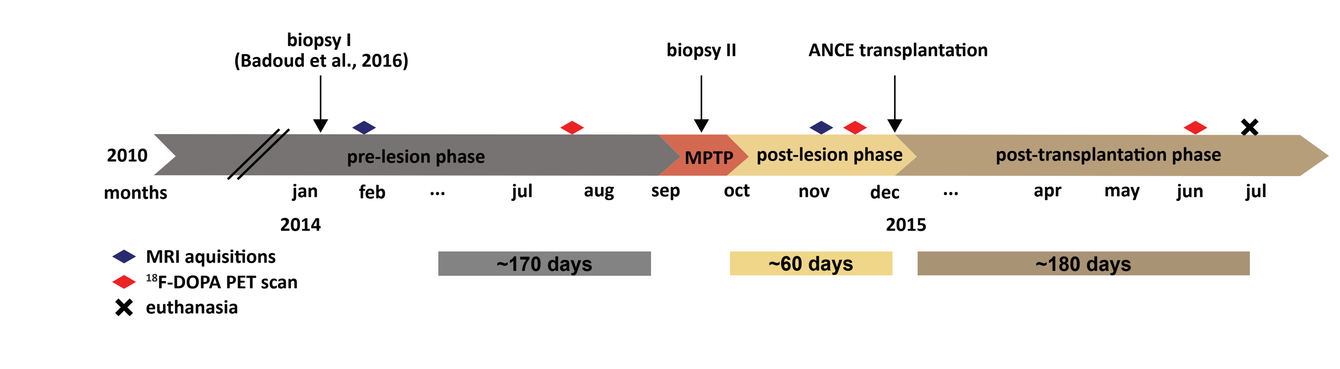


Figure 2: Experiment schedule. During the pre-lesion phase a first biopsy was performed in order to assess its potential impact on the behavioral tasks and to develop the good manufacturing practice (GMP) protocol (Badoud et al., 2016). The second biopsy, which provided the cellular material, was performed during the MPTP phase better reflecting the clinical conditions of Parkinson's disease. All four animals were subjected to the MPTP lesion and to the ANCE transplantation. Results obtained from different analyses were compared between pre- versus post-lesion and post-transplantation phases on each monkey.

Spontaneous activity

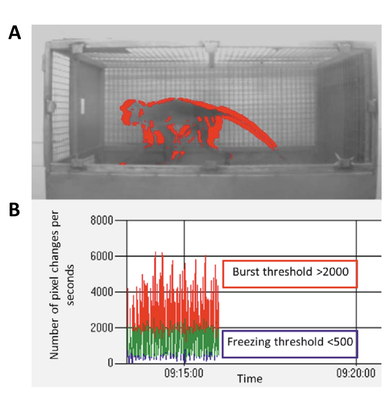
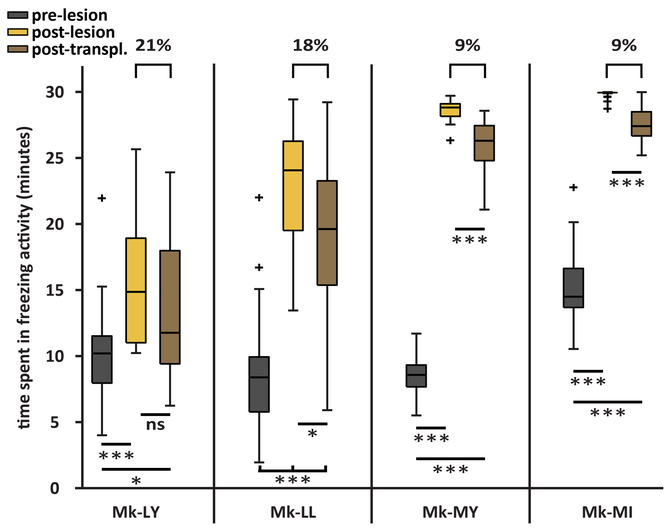


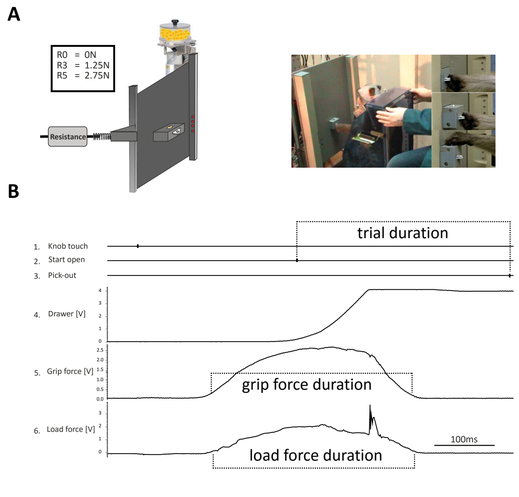
Figure 3: (**A**) Locomotor and other activities (free behaving) were assessed using the VigiePrimate image analyzer system. The system calculated in real time the quantity of movements, obtained by change of the different 256 pixels grey level. (**B**) Different thresholds defined freezing, middle and burst activities.



After **MPTP intoxication**: increase of the time spent in freezing activity in all four animals.

After **ANCE transplantation**: slight decrease of the time spent in freezing activity in all four animals.

Reach and grasp movements



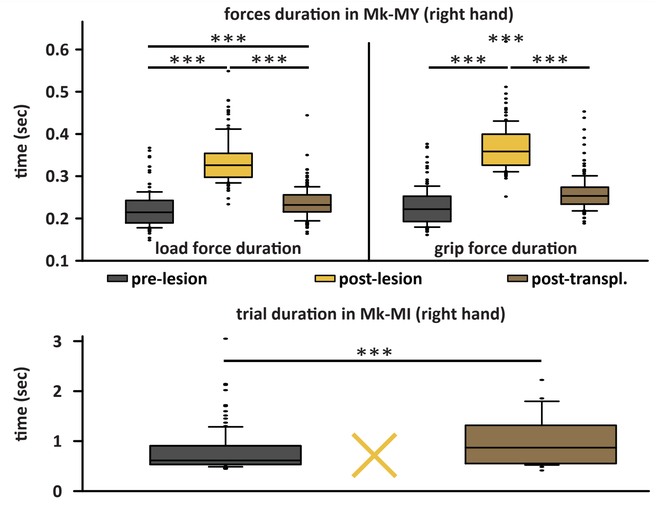


Figure 4: (**A**) Representation of the «reach and grasp drawer task» setup with the adjustable resistances in Newtons. (**B**) Raw data curves are shown for one trial (from drawer opening to the reward picking out).

After **MPTP intoxication**: in Mk-MY, increase of the load and the grip force durations at all resistances for both hands (R0, R3 and left hand not shown). Mk-MI was not able to perform the task. Mk-LY and Mk-LL were affected to a lesser extent (data not shown).

After **ANCE transplantation**: in Mk-MY, recovery of the load force durations at all resistances for both hands (R0 and R3 not shown). Monkey-MI regained its capacity to perform the task only at R0.

Fine manual dexterity

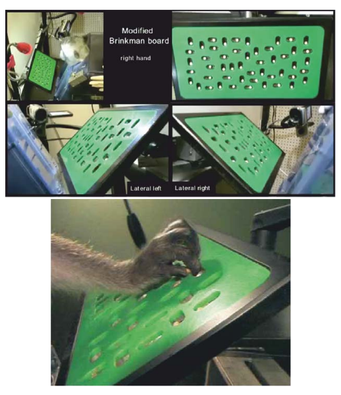
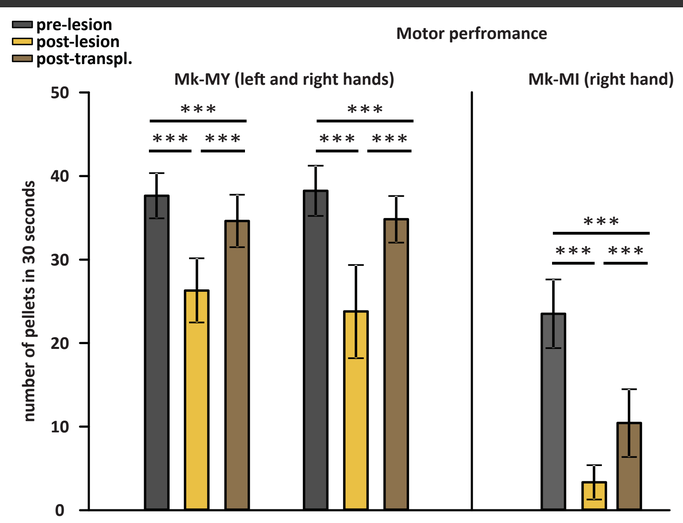


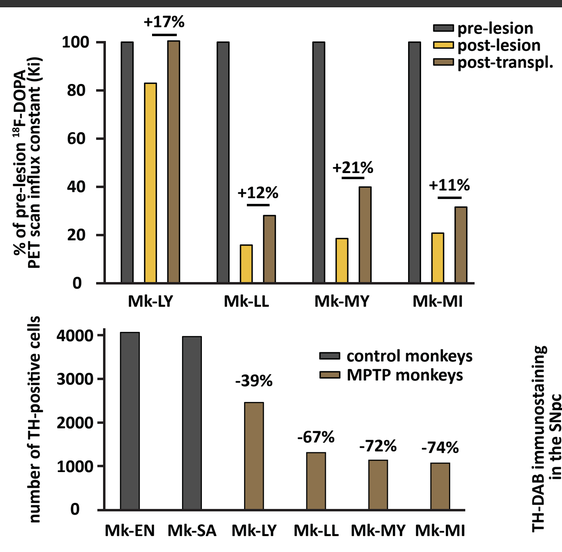
Figure 5: The «Modified-Brinkman board task», in which the monkey has to retrieve banana food pellets using the precision grip (opposition of thumb and index finger), from 25 horizontal and 25 vertical slots. Dimension of slots: 15 mm long, 8 mm wide and 6 mm deep.

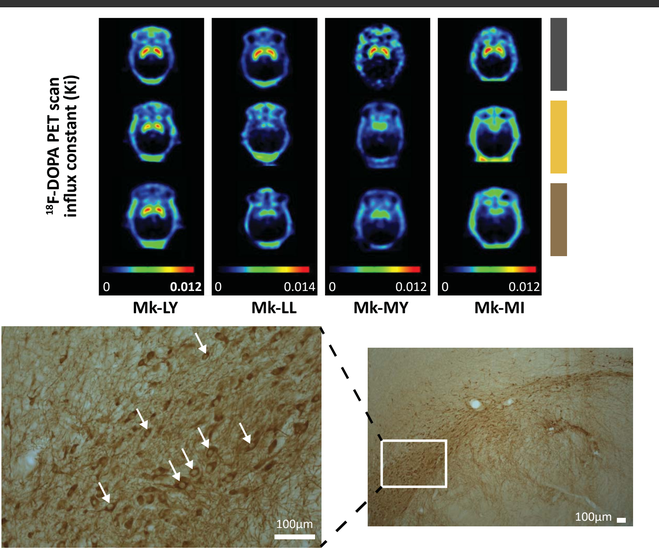


After **MPTP intoxication**: impairments of fine manual dexterity in Mk-MY and Mk-MI, whereas Mk-LL and Mk-LY were affected to a lesser extent (data not shown).

After **ANCE transplantation**: improvement of the fine manual dexterity in Mk-MY and Mk-MI (6 months post-transplantation).

Dopaminergic state





After **MPTP intoxication**: dramatic decrease of striatal uptake (loss>80%) in all animals except in Mk-LY (17%).
After **ANCE transplantation**: all subjects showed a significant but limited increase of their striatal uptakes, ranging from 11% to 21%.

In the **4 parkinsonian monkeys**: in the SNpc, the number of the dopaminergic neurons (tyrosine hydroxylase -positive neurons) was 70% lower than in two healthy monkeys (Mk-SA and Mk-EN) in all the four animals except in Mk-LY.

Discussion

Out of the four animals, two were severely affected by the MPTP lesions whereas the other two exhibited mild symptoms. Furthermore, the ^{18}F-DOPA striatal uptake was reduced by about 80% in three of them. Six months following ANCE transplantations, all monkeys presented significant improvement of their motor impairments. This functional recovery was accompanied by an increase of ^{18}F-DOPA striatal uptake. These results were consistent with the number of dopaminergic neurons within the SNpc. Taken together these new data open new therapeutic perspectives for the ANCE approach regarding neurodegenerative disorders like Parkinson's diseases.

Bloch, J., Brunet, J.-F., McEntire, C. R. S., & Redmond, D. E. (2014). Primate adult brain cell autotransplantation produces behavioral and biological recovery in 1-methyl-4-phenyl-1,2,3,6-tetrahydropyridine-induced parkinsonian St. Kitts monkeys. Journal of Comparative Neurology, 522(12).
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