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Functional integration of neural grafts in Parkinson's disease

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Functional imaging in a Parkinson's patient with a neural transplant shows that the graft is still functional after ten years, and that dopamine from the graft can bind to postsynaptic sites.

Parkinson's disease (PD) is a common neurological disease that affects approximately 1 in 1000 adults and is characterized by slowness of movement ('bradykinesia'), rigidity and tremor. The disease is associated with the loss of dopamine neurons in the basal ganglia, a group of forebrain nuclei that control voluntary movement. Although the early stages of PD can be treated with drugs that replace dopamine, these treatments eventually fail to control the disease symptoms, and they also produce their own debilitating side effects. Thus, there is an urgent need to develop other therapeutic strategies. Among the most promising is cell transplantation, a radical new approach that seems to offer the possibility of repairing the damage in PD, rather than merely alleviating or suppressing the symptoms. Of the various alternatives that have been explored so far, the best and most consistent results have been obtained with dopamine neurons taken from aborted human embryos at the specific developmental stage when these cells are first born and primed for active growth. The use of such cells in PD patients was pioneered over a decade ago in Sweden, and although it remains a highly experimental approach, a number of centers worldwide have now reported clinical benefits in transplantation patients¹. On page 1137 of this issue², the Swedish team presents exciting new data to show that

embryonic dopaminergic grafts can continue to release dopamine for as long as ten years after transplantation.

The new study is based on a single PD patient from a series of 17 who have received grafts of embryonic dopamine neurons over the last decade. The clinical benefit derived from these grafts has already been described^{3,4} and is thought to be due to dopamine replacement by the grafted cells. Consistent with this idea, most of the transplanted patients show a reduced dependence on L-dopa, a dopamine precursor that is widely used to treat PD patients, which works by supplementing the dopamine content of surviving dopamine terminals. Moreover, positron emission tomography (PET) scans have provided direct evidence for the presence of grafted dopamine neurons using the tracer [¹⁸F]dopa, which is taken up by presynaptic dopamine terminals.

In the normal brain, dopamine is released from nerve terminals in the basal ganglia and binds to specific dopamine receptors on the postsynaptic neurons. Free dopamine is then metabolized or removed by reuptake into the presynaptic terminals. [¹⁸F]dopa PET scans can tell us whether dopamine neurons and their associated nerve terminals are present, but they do not reveal whether dopamine from the graft is released and taken up in a controlled manner at synapses⁵. It could be that the grafted cells release dopamine in a non-specific manner such that it simply diffuses away from the transplanted cells to influence other cells in a non-regulated fashion. Post-mortem studies at another center have shown that

a positive [¹⁸F]dopa PET signal correlates with graft survival as well as with clinical effect⁶. However, post-mortem studies cannot provide direct information on graft function *in vivo*.

To address this question, Piccini and colleagues² studied an unusual patient ('number 4' in the series) who had received a graft on one side only, because both he and his clinicians were satisfied with the recovery that had been achieved. This asymmetry provided the authors with a unique opportunity for research, in that the long-term survival and growth of the graft on one side of the brain could be compared with the progression of the disease on the untransplanted side.

The authors have also used a new PET scanning protocol. In addition to doing conventional longitudinal PET scans using [¹⁸F]dopa, they have examined the presence and occupancy of postsynaptic dopamine D₂ receptors, by measuring binding with the D₂ dopamine receptor ligand [¹¹C]raclopride. The rationale for their experiment is illustrated in Fig. 1. [¹⁸F]dopa is taken up by presynaptic dopamine terminals, and the resulting PET signal reveals the presence of dopamine innervation in the healthy striatum, its loss in PD, and its replacement by grafted dopamine neurons in transplanted patients. Raclopride, by contrast, labels postsynaptic receptors on host striatal neurons. If the dopamine innervation is lost—whether experimentally or in PD—the number of postsynaptic D₂ receptors is increased, in addition to which individual receptors become supersensitive to dopamine^{7–9}. In the transplant patient², the loss of dopamine terminals on the untreated side (observed by loss of [¹⁸F]dopa signal) is accompanied by an increase in raclopride binding, indicating upregulation of postsynaptic receptor numbers. On the grafted side, in contrast, the raclopride signal has reverted to a normal level in parallel with restoration of the [¹⁸F]dopa signal. This suggests that the graft-derived terminals are releasing dopamine in sufficient quantities to suppress the upregulation of postsynaptic receptors. This is an encouraging result, although perhaps not surprising given

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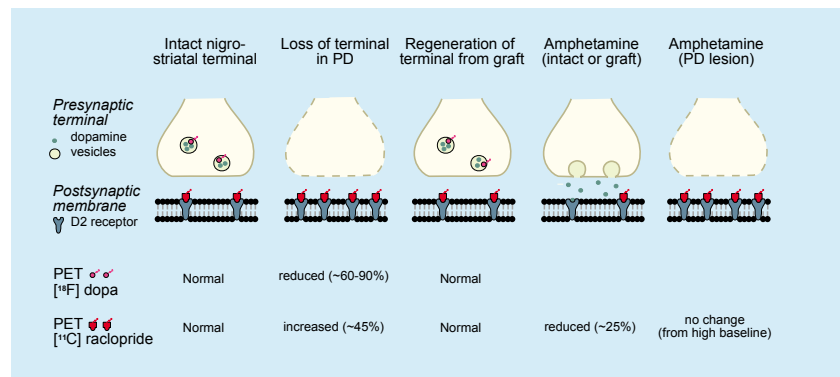


Fig. 1. Schematic illustration of the normal dopamine terminal in the basal ganglia, the pre- and postsynaptic changes associated with PD and cell transplantation, and the sites of accumulation of the two PET ligands, [^{18}F]dopa and [^{11}C]raclopride (see text for details).

this patient's much improved clinical response with progressively lower doses of oral L-dopa therapy.

The really novel feature of the present study, however, is the additional scan following administration of the stimulant drug, amphetamine. Amphetamine potentiates the release of dopamine, which then competes with raclopride for binding to postsynaptic receptors (see Fig. 1). The raclopride signal is therefore reduced by amphetamine in proportion to the amount of dopamine that is released. On the untreated side in this patient, the loss of endogenous dopamine terminals makes amphetamine ineffective, and raclopride binding does not change. By contrast, the treated side shows a 27% reduction of raclopride binding after amphetamine administration. This is very similar in magnitude to the 23% reduction found in the brains of healthy control subjects, and it indicates that dopamine can be released from graft-derived synaptic terminals to bind to host-derived postsynaptic sites. Moreover, it confirms that the grafted neurons continue to function for ten years after transplantation, suggesting that they are not vulnerable to the pathological process that destroyed the original dopamine innervation.

Although the present report is based on only a single case, it nevertheless provides a strong demonstration that embryonic grafts can restore regulated release of dopamine at synaptic sites. Like the earlier single post-mortem cases that provided the first clear evidence of graft survival in PD patients^{6,10}, the new study is likely to be seen as an important step in the development of neuronal transplantation therapy. Nevertheless, there are some important issues that must now be considered. Most obviously, results from one patient (who was selected based on his good clinical outcome) tell us nothing about the reproducibility of the technique. This is a pressing concern in view of the double-blind placebo controlled trial of human fetal dopamine cell transplants that has recently been completed in the US. Although this study has so far only been reported in the media and in abstract form (C.R. Freed *et al.*, *Soc. Neurosci. Abstr.* 25, 212, 1999), it appears that significant clinical benefit was only found in younger patients; moreover, clinical outcome correlated only poorly with the small changes in PET [^{18}F]dopa signal. However, the authors of this latter study implanted fewer cells and used a protocol markedly different from that used by the

Swedish group; together these differences may account for the more modest results of both the clinical and PET assessments.

Clearly, further clinical trials are needed to optimize the transplantation procedure, and the present study establishes a rigorous standard against which any new cellular therapy for PD will need to be assessed. There are currently a number of alternative cell sources being considered for use in the transplantation of PD, including embryonic pig tissues, neural precursor cells and genetically modified cell lines¹¹. Although these alternative sources of cells pose many problems that need to be resolved, some have already progressed to early clinical trials^{12,13}. In any situation designed to treat PD, an effective therapy is likely to be one that not only releases dopamine diffusely but also does so in a controlled and regulated manner. The novel scanning protocol introduced by Piccini and colleagues offers a way to visualize directly the extent to which this is achieved in the living brain, and will thereby complement measures of clinical benefit in refining effective novel treatments on a rational basis.

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