Graft-Induced Dyskinesias in Parkinson’s Disease: High Striatal Serotonin/Dopamine Transporter Ratio

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ABSTRACT: Graft-induced dyskinesias are a serious complication after neural transplantation in Parkinson’s disease. One patient with Parkinson’s disease, treated with fetal grafts 14 years ago and deep brain stimulation 6 years ago, showed marked improvement of motor symptoms but continued to suffer from OFF-medication graft-induced dyskinesias. The patient received a series of clinical and imaging assessments. Positron emission tomography and single-photon emission computed tomography 14 years posttransplantation revealed an elevated serotonin/dopamine transporter ratio in the grafted striatum compatible with serotonergic hyperinnervation. Inhibition of serotonin neuron activity by systemic administration of a 5-HT1A agonist suppressed graft-induced dyskinesias. Our data provide further evidence that serotonergic neurons mediate graft-induced dyskinesias in Parkinson’s disease. Achieving a normal striatal serotonin/dopamine transporter ratio following transplantation of fetal tissue or stem cells should be necessary to avoid the development of graft-induced dyskinesias. © 2011 Movement Disorder Society

Key Words: Parkinson; transplantation; dyskinesias; serotonin transporter; dopamine transporter

Additional Supporting Information may be found in the online version of this article.

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Dyskinesias persisting after withdrawal of dopamine (DA) replacement medication have been reported in a number of Parkinson’s disease (PD) patients with intrastriatal grafts of fetal ventral mesencephalic (VM) tissue.1–5 A few of these patients have later required palliative deep brain stimulation (DBS) for their dyskinesias, but the results have been variable.6–8 Successful future clinical application of DA cell therapies, including transplantation of fetal or stem cells, will require an understanding of the mechanisms underlying graft-induced dyskinesias (GIDs) to be prevented or effectively treated.

We recently reported that in 2 PD patients with fetal VM transplants, GIDs were caused by serotonergic (5-HT) hyperinnervation of the striatum.5 Here, we report another patient (patient 13 in the Lund series) who received both fetal VM tissue transplantation and, later, DBS in the globus pallidus internus (GPI). Our aim was to further examine the 5-HT hypothesis and explore the interaction between the 5-HT
transmitter (SERT) and DA transporter (DAT) in the development of GIDs.

Patient, Methods, and Results

The patient, a 55-year-old man, was diagnosed with PD (right side) in 1983 and after 6 months was started on l-dopa. For 5 years he experienced a stable response to l-dopa. Between 1989 and 1990 he gradually developed peak-dose l-dopa-induced dyskinesias (LIDs). For the next 5 years his quality of life rapidly deteriorated, with daily LIDs, wearing OFF, and ON–OFF fluctuations. In 1996, he deteriorated, with daily LIDs, wearing OFF, and ON–OFF fluctuations. For the next 5 years his quality of life rapidly developed peak-dose L-dopa-induced dyskinesias (LIDs). For the next 5 years his quality of life rapidly developed peak-dose L-dopa-induced dyskinesias (LIDs).

Following transplantation, the patient showed rapid clinical improvement with decreased Unified Parkinson’s Disease Rating Scale (UPDRS) motor (part III) scores and reduced severity of LIDs. In 1997, he stopped l-dopa treatment without deterioration, completely eliminating LIDs, and since then he has had a stable performance on clinical tests (Fig. 1A). However, in 1997 he developed mild GIDs, which then progressively increased in severity over the next 6 years. In 2004, he received bilateral GPi DBS, which reduced the severity of GIDs for the next 6 months, but then he relapsed (Fig. 1B).

Currently, he is not taking any DA medication (nor anticholinergic or amantadine), but he derives motor benefit when DBS is ON (Fig. 1A). He has no OFF periods but exhibits continuous GIDs, which are insufficiently relieved by DBS (Fig. 1B). His GIDs consist of hyperkinesias, mainly in the left arm, foot, and shoulder, and dystonic movements of the right arm and are present constantly, causing disability and distress (Supplementary Video 1).

Patient 13 has been assessed with single-photon emission computed tomography (SPECT) scans before and after transplantation using 123I-FP-CIT (Fig. 2) and 14 years posttransplantation using 123I-FP-CIT to assess DAT availability (methods10,11). He has also received a series of 18F-DOPA positron emission tomography (PET) scans to measure his DA storage capacity (Fig. 1A) and 11C-raclopride (RAC) PET scans before and after a methamphetamine challenge to assess DA release from the graft (methods12).

18F-DOPA PET influx constants (Ki) in the left and right caudate nuclei were normal following the first posttransplantation year. 18F-DOPA uptake was higher in the right (within normal limits) than in the left (below normal levels) putamen and remained stable from the third postoperative year (increased by 77% in the left and by 49% in the right putamen from baseline; Fig. 1A). Methamphetamine challenge 5 years posttransplantation induced normal levels of DA release in the right putamen (23% reduction of RAC binding; normal range, 19.6%–27.2%), but reduced DA release in the left putamen (15%).

Fourteen years posttransplantation, 123I-FP-CIT SPECT showed that DAT availability was normal in the right caudate nucleus and putamen, slightly reduced in the left caudate nucleus, and significantly reduced in the left putamen (Fig. 3A). 18F-DOPA KiS were normal in the caudate bilaterally and in the right putamen (Figs. 1A and 3B).

Using 11C-DASB PET, a marker of SERT availability (subjects and methods5), we found increased 5-HT innervation in the grafted striatum. 11C-DASB binding was elevated by 27% in the caudate (34% left, 21% right) and 46% in the putamen (34% left, 57% right) compared with healthy normal individuals and by 101% (112% left, 91% right) and 106% (90% left, 122% right), respectively, compared with advanced PD patients (Fig. 3C–E). The ratio of 5-HT to DA innervation in the grafted putamina, estimated as the 11C-DASB/18F-DOPA binding ratio, was increased by 140% (152% left, 128% right) compared with the ratio in healthy normal individuals (259 vs 108). Moreover, SERT-to-DAT availability expressed as the 11C-DASB/123I-FP-CIT binding ratio was 0.78 for the right and 0.97 for the left putamen (normal average, 0.48).

Taken together, our imaging data indicate 5-HT hyperinnervation, high 5-HT relative to DA innervation, and increased SERT binding sites per DAT in the graft putamina 14 years after transplantation. We next tested whether the 5-HT neurons contributed to GIDs. The 5-HT1A agonist buspirone inhibits 5-HT neuron transmission via autoreceptor stimulation, and it was administered in 3 doses of 5 mg at 30-minute

<table>
<thead>
<tr>
<th>Patient number</th>
<th>13</th>
</tr>
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<tbody>
<tr>
<td>Year of transplantation</td>
<td>1996</td>
</tr>
<tr>
<td>Location of implanted sites</td>
<td>L Put + Caud/ R Put + Caud</td>
</tr>
<tr>
<td>Number of implant sites</td>
<td>5 + 2/5 + 2</td>
</tr>
<tr>
<td>Number of donors</td>
<td>4/5</td>
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<td>Size of donors</td>
<td>L Put and L Caud: mean size, 23 mm (range, 16–26 mm)</td>
</tr>
<tr>
<td></td>
<td>R Put and R Caud: mean size, 22 mm (range, 16–26 mm)</td>
</tr>
<tr>
<td>Amount of implanted tissue</td>
<td>L Put: 2.9 VM</td>
</tr>
<tr>
<td></td>
<td>R Put: 3.6 VM</td>
</tr>
<tr>
<td>Time from abortion to implantation</td>
<td>L Put: mean time, 5 h, 15 min (range, 2 h, 45 min–7 h, 30 min)</td>
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<tr>
<td></td>
<td>L Caud: mean time, 7 h (range, 5 h–8 h, 30 min)</td>
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<tr>
<td></td>
<td>R Put: mean time, 5 h, 30 min (range, 2 h, 15 min–8 h)</td>
</tr>
<tr>
<td></td>
<td>R Caud: mean time, 7 h, 15 min (range, 5 h, 15 min–9 h)</td>
</tr>
</tbody>
</table>

L, left; R, right; Put, putamen; Caud, caudate nucleus; VM, ventral mesencephalon.

*Exposed to the lazaroid tirilazad mesylate during storage and dissociation; patient given lazaroid for 3 days (Brundin et al, 2000).
intervals (methods). This treatment significantly attenuated the severity of GID for 4 hours when compared with no-drug or placebo administration (Fig. 3F and Supplementary Videos 1 and 2). The UPDRS motor scores remained unchanged at 23 (DBS-OFF) after buspirone administration, arguing against the possibility that its effect on GIDs was a result of reduced DA tone, and there were no side effects.

**Discussion**

Here we have shown that GIDs, which respond to a 5-HT<sub>1A</sub> agonist that acts to inhibit transmitter release from 5-HT neurons, are associated with 5-HT hyperinnervation and a high SERT-to-DAT ratio in the grafted striatum. The patient had more pronounced dyskinesias in the left side of the body, in line with
our finding that 5-HT hyperinnervation was most pronounced in the right putamen. This difference was probably because the right putamen was grafted with more VM tissue than the left putamen (Table 1).

We found a 46% increase in putaminal $^{11}$C-DASB binding in our grafted patient (patient 13) compared with normal controls; this increase was significantly lower than that observed in our 2 previous cases (patient 7, 172%; patient 15, 77% increase). However, the severity and frequency of GIDs were similar in patients 13 and 15. Interestingly, patients 13 and 15 showed a comparable increase in the $^{11}$C-DASB/$^{18}$F-DOPA binding ratio (patient 13, 140%; patient 15, 146% increase, compared with the normal ratio), suggesting that it is not just the 5-HT hyperinnervation per se but the proportion of 5-HT-to-DA innervations within the grafted striatum that is the most important factor in the development of GIDs.

The results from this case together with our previous observations$^5$ indicate that the occurrence of GIDs is mediated by an interaction between the graft-derived excess of 5-HT terminals and the DA terminals in the striatum. SERT and DAT seem to be the key mediators in this mechanism. Patient 13 expressed elevated numbers of SERT per DAT sites in the putamen. SERT can take up DA from the extracellular space, and DA is then stored in the presynaptic 5-HT terminals and released as a “false” transmitter.$^{13,14}$ In addition, we propose that the increased 5-HT release from striatal hyperinnervation acts directly on DA terminals to induce further DA release via action on and reversal of DAT.$^{13,16}$ This would result in an abnormal, impulse-independent release of DA at the synapse, leading to development of GIDs. The dysregulation of synaptic levels of DA caused by 5-HT hyperinnervation can be reversed by administration of low doses of 5-HT$_{1A}$ agonists like buspirone. By activating inhibitory 5-HT autoreceptors, buspirone attenuates the transmitter release from 5-HT neurons, thereby leaving most of the synaptic DA to be normally regulated from the DA terminals (Fig. 4). The clinical correspondence of such interference is the significant attenuation of GIDs.

In this patient, GIDs showed a slow and gradual increase, in contrast to the rapid and marked improvement of PD symptoms following transplantation. This gradual buildup of GIDs is most likely explained by the emergence of unfavorable 5-HT-to-DA and SERT-to-DAT ratios because of gradual expansion of the graft-induced 5-HT innervation over time to generate hyperinnervation patterns in the reinnervated areas.$^{19,20}$ In line with this scenario, although DBS in GPi improved motor function, GIDs were not alleviated.

The development of GIDs is a serious adverse event following intrastriatal transplantation of fetal VM tissue in PD patients. We have demonstrated virtually complete suppression of GIDs by systemic administration of the 5-HT$_{1A}$ agonist, buspirone. Our findings...
strongly support the notion that 5-HT neurons are crucially involved in the pathogenesis of GIDs and should lead to new, optimized protocols for safer and more effective application of DA cell replacement strategies in PD patients. Our data indicate that functional imaging provides the means of monitoring such protocols by assessing the key modulators (SERT and DAT) underlying the development of GIDs. We propose that near to normal $^{11}$C-DASB/$^{18}$F-DOPA (about 108) and $^{11}$C-DASB/$^{123}$I-FP-CIT (about 0.48) ratios associated with robust DA ($^{18}$F-DOPA) restoration will provide long-lasting relief from PD symptoms and avoid the occurrence of GIDs.

Acknowledgments: We thank patient 13, whose participation made this study possible.
Legends to the Video

Supplementary Video 1. Patient 13 after administration of placebo. Video sequences were captured between 150 and 180 minutes after the beginning of the trial.

Supplementary Video 2. Patient 13 after administration of 5-HT1A agonist (buspirone). Video sequences were captured between 150 and 180 minutes after the beginning of the trial.

References

15. Jacocks HM 3rd, Cox BM. Serotonin-stimulated release of [3H]dopamine via reversal of the dopamine transporter in rat


