Human striatal recordings reveal abnormal discharge of projection neurons in Parkinson’s disease

Arun Singh a, Klaus Mewes b, Robert E. Gross c, Mahlon R. DeLong b, José A. Obeso d,e, and Stella M. Papa a,b,1

aYerkes National Primate Research Center, Emory University, Atlanta, Georgia 30329; bDepartment of Neurology, Emory University School of Medicine, Atlanta, Georgia 30329; cDepartment of Neurosurgery, Emory University School of Medicine, Atlanta, Georgia 30329; dCentro Integral de Neurociencias AC, Hospitales de Madrid Puerta del Sur, San Pablo University, Móstoles, 28938 Madrid, Spain; and eCenter for Biomedical Research on Neurodegenerative Diseases, Instituto de Salud Carlos III, 28029 Madrid, Spain

Edited by Peter L. Strick, University of Pittsburgh, Pittsburgh, PA, and approved June 27, 2016 (received for review April 28, 2016)

Circuitry models of Parkinson’s disease (PD) are based on striatal dopamine loss and aberrant striatal inputs into the basal ganglia network. However, extrastriatal mechanisms have increasingly been the focus of attention, whereas the status of striatal discharges in the parkinsonian human brain remains conjectural. We now report the activity pattern of striatal projection neurons (SPNs) in patients with PD undergoing deep brain stimulation surgery, compared with patients with essential tremor (ET) and isolated dystonia (ID). The SPN activity in ET was very low (2.1 ± 0.1 Hz) and reminiscent of that found in normal animals. In contrast, SPNs in PD fired at much higher frequency (30.2 ± 1.2 Hz) and with abundant spike bursts. The difference between PD and ET was reproduced between 1-methyl-4-phenyl-1,2,3,6-tetrahydropyridine-treated and normal nonhuman primates. The SPN activity was also increased in ID, but to a lower level compared with the hyperactivity observed in PD. These results provide direct evidence that the striatum contributes significantly altered signals to the network in patients with PD.

Motor features of Parkinson’s disease (PD) are caused by alterations in the corticobasal ganglia–thalamic network, although the role of particular circuits is unclear (1–3). The progressive degeneration of nigral neurons that massively deplete the striatum of dopamine is at center stage. Classic circuitry models of PD are based on the dopamine-depleted striatum sending disrupted commands through medium spiny neurons, the striatal projection neurons (SPNs), into the direct and indirect output pathways (4, 5). However, the role of a dysfunctional striatum has been undermined in recent years in part due to the focus on extrastriatal mechanisms, particularly the direct cortical regulation of the subthalamic nucleus (hyperdirect pathway), and the widespread effects of dopamine depletion over basal ganglia stations (6–8). In addition, the striatal changes predicted by the model lack clear functional correlates in humans. The only available data are provided by neuroimaging and show inconsistent metabolic changes (both increased or normal activity) in the putamen of patients with PD (9). On the other hand, the molecular and physiological impact of dopamine loss on striatal outputs has been difficult to determine, given the complexity of microcircuits regulating the SPN activity. Convergent cortical, nigral, thalamic, and various interneuronal signals could variably exacerbate or compensate for the lack of dopamine modulation on SPN discharges (10).

Morphological and physiological studies in animal models of PD, however, have provided significant data supporting abnormal SPN activity in the parkinsonian state. There is a major loss of dendritic spines in SPNs that is accompanied by remodeling and enlargement of postsynaptic densities of the remaining spines (11, 12). Although the mechanisms underlying spine pruning and regrowth are yet unclear, ultimately, these morphological changes involve synaptic contacts and thus have major effects on synaptic transmission (13). Indeed, changes in corticostriatal synaptic plasticity have been extensively documented in paradigms of long-term potentiation and depression (LTP and LTD) after dopaminergic lesion (14, 15). However, the activity of SPNs in vivo has been more elusive, particularly in animal models of chronic dopamine deficiency. Different from striatal interneurons, recordings of SPNs have inherent difficulties in rodents and nonhuman primates (NHP) due to unstable firing and frequent loss of unit. Nevertheless, studies in anesthetized hemiparkinsonian rats show higher SPN activity (16, 17). In the NHP model of advanced parkinsonism that more closely reproduces PD features, SPN recordings show major activity alterations (18, 19). However, it is often questionable whether large changes produced by short-term, high-toxin exposure in the animal are equivalent to the effects of slowly progressive neurodegeneration as occurs in the human disease (20). All in all, a demonstration of altered SPN discharges supporting a primary role of striatal dysfunction in PD pathophysiology is lacking. To date, the striatal activity has not been studied during the electrophysiology-guided mapping of basal ganglia in patients undergoing surgical treatments.

The reason was generally related to limitations for recordings in the stratum due to unstable mapping techniques or surgical time restrictions. We took the unique opportunity of deep brain stimulation (DBS) surgery to record single cells in the stratum of patients, and here we report the spontaneous SPN activity in advanced PD.

Striatal single-unit recordings in patients with essential tremor (ET) and isolated dystonia (ID) were also analyzed for comparison. Patients with ET provided the counterpart for absence of (known) striatal mechanisms as negative control (21, and

Significance

This study is important because it provides the first account of the electrophysiological activity in the human striatum, and it demonstrates major and distinctive abnormalities of neuronal firing in Parkinson’s disease (PD). Up until now, circuit models of PD were based on striatal changes that were never demonstrated in patients. We compared striatal recordings across patients with PD and other neurological disorders [dystonia and essential tremor (ET)], and correlative findings in nonhuman primates. Therefore, the data provided by the present study significantly contribute to understand the role of striatal mechanisms in basal ganglia circuits and in the pathophysiology of PD. Additionally, the study originally reports altered striatal activity in dystonia and activity compatible with unchanged striatal function in ET.


The authors declare no conflict of interest.

This article is a PNAS Direct Submission.

1To whom correspondence should be addressed. Email: spapa@emory.edu.

This article contains supporting information online at www.pnas.org/lookup/suppl/doi:10.1073/pnas.1606792113/-/DCSupplemental.
patients with ID provided a different motor disorder with substantial evidence for striatal dysfunction as positive control (22). Equivalent recordings in NHPs were included for correlation of the SPN activity with normal and abnormal states. After several years in the quest for surgeries without anesthesia and with electrode trajectories passing through striatal areas according to the surgical approach, a total of 155 human SPNs were recorded, allowing comparisons across neurological disorders to characterize the SPN activity in PD.

Results

Striatal recordings were obtained in 26 patients, including those with PD (n = 11), ET (n = 10), and ID (n = 5), who were alert and resting during these recordings and had suspended their regular drug treatments on the day of surgery (see criteria for patient inclusion in Materials and Methods). Patients with ID had a segmental form: cervical and cranial dystonia. All patients had a long-standing disease, with a minimum of 10, 9, and 4 y in patients with PD, ET, and ID, respectively (Table 1).

In all patients, single-cell recording in the striatum showed variable spiking, corresponding to the characteristic firing of SPNs. However, difficulties in recording these neurons complicated data collection in all patients with PD, ET, and ID. The typically unstable SPN spiking requires longer recordings, but there is time limitations in human surgery. Recordings of spike traces were thus limited to 1–3 min in all patients. Examples of typical SPN recordings are shown in Fig. S1. Considering that the activity parameters of SPNs are phylogenetically conserved throughout vertebrates, including primates (23), to analyze the human single-cell data, we applied the same well-established criteria that are currently in use to classify striatal neurons in animal recordings. In all recordings, units were first sorted using principal component analysis (Offline Sorter, Plexon) and then classified selecting data compatible with SPN and excluding interneuron activity [first spiking interneuron (FSI) and tonically active neuron (TAN)] (SI Materials and Methods, Fig. 1, and Fig. S2). Therefore, the analyzed single-cell activity pattern represents the status of spontaneous SPN discharges in patients with different underlying disorders. According to mapping references, the total of 62 SPNs recorded in PD were located in the putamen (n = 44) or the caudate nucleus (n = 18), allowing comparisons with SPNs recorded in ET and ID. Both striatal regions could be recorded in patients with PD because the electrode trajectory toward the different surgical targets [i.e., globus pallidus internus (GPI), the subthalamic nucleus (STN) and ventralis intermedius nucleus (VIM)], imposed different routes. All 41 SPNs recorded in ET were located in the caudate nucleus because electrode tracks run commonly medial to the putamen in the thalamus-targeting DBS surgery (VIM). In contrast, most of the 52 SPNs recorded in ID were in the putamen because of the pallidal target, although some striatal cells were recorded in tracks passing through the caudate–putamen junction. To correlate data obtained in patients with data from three NHPs, the same striatal areas and procedures were used in all recordings.

Increased Firing Frequency of SPNs in PD. SPNs of patients with PD were spontaneously overactive, firing at much higher frequencies in comparison with other patient groups (examples in Fig. 1 A–D).

In the baseline parkinsonian state in absence of antiparkinsonian drugs and in the resting condition, the mean firing frequency of SPNs was 30.2 ± 1.2 Hz (between 13.5 and 47.9 Hz). Similar increases of activity were found in both putamen and caudate areas, but putamen neurons fired at slightly higher frequency than those in the caudate nucleus ($P < 0.01$; Fig. 1 E).

The SPN activity in patients with PD was increased by more than 10-fold compared with the very low firing frequencies (2.1 ± 0.1 Hz) found in patients with ET ($P < 0.001$). In addition, the SPN activity in patients with PD was 3-fold higher than in patients with ID (9.3 ± 0.6 Hz), thus establishing a distinct level of hyperactivity in PD (Fig. 2 A).

The very low SPN firing found in patients with ET is congruent with the classic description of normal activity in animals (24), which was also shown here in the normal NHP using the same procedures and data analyses as in human recordings (SI Materials and Methods). These correlative findings suggest that the striatal function is spared in ET and support the use of this comparison as a substitute for normal subjects. The comparison between patients with ET and ID showed SPN firing frequencies moderately increased in ID (Fig. 2 A).

The escalation of frequency changes from ET to ID and again to PD reveals two pathological states of the striatum with SPN activity changing in the same direction but reaching an exaggerated level in PD. The strict comparison of only putaminal neurons increases the difference between PD and ID because SPNs of patients with PD had higher firing rates in the putamen than the caudate nucleus.

The SPN firing frequency in the parkinsonian macaques was markedly increased by comparison with the normal animal (23.3 ± 1.6 Hz and 1.6 ± 0.1 Hz, respectively; $P < 0.001$; Figs. 1 F and 2 B).

In NHPs [one normal macaque and two advanced parkinsonian macaques modeled with chronic 1-methyl-4-phenyl-1,2,3,6-tetrahydropyridine (MPTP) administration], a total of 52 SPNs were recorded (control NHP, $n = 18$; parkinsonian NHPs, $n = 34$). The SPN firing frequencies in these animals were in the range previously reported for the parkinsonian and normal NHP conditions (18). The increase in SPN activity from normal to advanced parkinsonian NHPs was similar to that from patients with ET to PD. Furthermore, the MPTP-induced change in SPN

### Table 1. Characteristics of patients

<table>
<thead>
<tr>
<th>Case</th>
<th>Sex</th>
<th>Age, y</th>
<th>Disease duration, y</th>
<th>Score</th>
<th>DBS target</th>
</tr>
</thead>
<tbody>
<tr>
<td>Parkinson’s disease</td>
<td></td>
<td></td>
<td></td>
<td></td>
<td></td>
</tr>
<tr>
<td>1</td>
<td>F</td>
<td>78</td>
<td>10</td>
<td>36/21</td>
<td>GPi</td>
</tr>
<tr>
<td>2</td>
<td>M</td>
<td>68</td>
<td>13</td>
<td>30/17</td>
<td>GPi</td>
</tr>
<tr>
<td>3</td>
<td>M</td>
<td>73</td>
<td>13</td>
<td>38/23</td>
<td>GPi</td>
</tr>
<tr>
<td>4</td>
<td>M</td>
<td>67</td>
<td>22</td>
<td>37/27</td>
<td>GPi</td>
</tr>
<tr>
<td>5</td>
<td>M</td>
<td>63</td>
<td>16</td>
<td>18/5</td>
<td>GPi</td>
</tr>
<tr>
<td>6</td>
<td>F</td>
<td>68</td>
<td>25</td>
<td>64/29</td>
<td>VIM</td>
</tr>
<tr>
<td>7</td>
<td>F</td>
<td>58</td>
<td>24</td>
<td>29/19</td>
<td>VIM</td>
</tr>
<tr>
<td>8</td>
<td>M</td>
<td>78</td>
<td>16</td>
<td>27/25</td>
<td>VIM</td>
</tr>
<tr>
<td>9</td>
<td>M</td>
<td>52</td>
<td>13</td>
<td>62/21</td>
<td>STN</td>
</tr>
<tr>
<td>10</td>
<td>M</td>
<td>56</td>
<td>11</td>
<td>38/19</td>
<td>STN</td>
</tr>
<tr>
<td>11</td>
<td>M</td>
<td>63</td>
<td>10</td>
<td>38/17</td>
<td>STN</td>
</tr>
<tr>
<td>Isolated dystonia</td>
<td></td>
<td></td>
<td></td>
<td></td>
<td></td>
</tr>
<tr>
<td>12</td>
<td>M</td>
<td>55</td>
<td>4</td>
<td>16</td>
<td>GPi</td>
</tr>
<tr>
<td>13</td>
<td>F</td>
<td>69</td>
<td>9</td>
<td>10</td>
<td>GPi</td>
</tr>
<tr>
<td>14</td>
<td>F</td>
<td>63</td>
<td>4</td>
<td>18.5</td>
<td>GPi</td>
</tr>
<tr>
<td>15</td>
<td>M</td>
<td>58</td>
<td>8</td>
<td>5.5</td>
<td>GPi</td>
</tr>
<tr>
<td>16</td>
<td>F</td>
<td>70</td>
<td>6</td>
<td>5.5</td>
<td>GPi</td>
</tr>
<tr>
<td>Essential tremor</td>
<td></td>
<td></td>
<td></td>
<td></td>
<td></td>
</tr>
<tr>
<td>17</td>
<td>M</td>
<td>71</td>
<td>14</td>
<td>59</td>
<td>VIM</td>
</tr>
<tr>
<td>18</td>
<td>M</td>
<td>79</td>
<td>9</td>
<td>50</td>
<td>VIM</td>
</tr>
<tr>
<td>19</td>
<td>F</td>
<td>74</td>
<td>NA</td>
<td>41</td>
<td>VIM</td>
</tr>
<tr>
<td>20</td>
<td>M</td>
<td>71</td>
<td>NA</td>
<td>48</td>
<td>VIM</td>
</tr>
<tr>
<td>21</td>
<td>M</td>
<td>72</td>
<td>NA</td>
<td>46</td>
<td>VIM</td>
</tr>
<tr>
<td>22</td>
<td>F</td>
<td>78</td>
<td>20</td>
<td>84</td>
<td>VIM</td>
</tr>
<tr>
<td>23</td>
<td>M</td>
<td>80</td>
<td>40</td>
<td>77</td>
<td>VIM</td>
</tr>
<tr>
<td>24</td>
<td>M</td>
<td>62</td>
<td>NA</td>
<td>30.5</td>
<td>VIM</td>
</tr>
<tr>
<td>25</td>
<td>F</td>
<td>68</td>
<td>53</td>
<td>34.5</td>
<td>VIM</td>
</tr>
<tr>
<td>26</td>
<td>F</td>
<td>78</td>
<td>13</td>
<td>57</td>
<td>VIM</td>
</tr>
</tbody>
</table>

Patients with PD were assessed by Unified Parkinson’s Disease Rating Scale III (motor score off/on medication). Patients with ID and ET were evaluated off medication with the Fahn-Marsden Scale and Tremor Rating Scale, respectively. In PD, ID, and ET groups, age averaged 66 ± 2.5, 63 ± 3, and 73 ± 1.7 (±SEM) y old, respectively (nonsignificant difference). DBS target: GPI, VIM, and STN. M, male; F, female; NA, not available.
firing frequency was computed to determine the mean predicted probability of PD. This analysis that included all 155 SPNs from 26 patients with ET, ID, or PD showed that the increase of SPN firing frequency to a minimum of 13 Hz (the minimum observed in the parkinsonian macaque) significantly predicted PD in patients (logit model, \( \chi^2: 151.6, P < 0.001, 93\% \) accuracy; Fig. 2C).

**Increased Burst Firing of SPNs in PD.** SPNs of patients with PD at rest fired not only faster, but frequently with brief spike bursts composed of 10.5 ± 1 spikes, 7 ± 0.3 ms intraburst interspike intervals (ISIs), and 72 ± 9 ms duration (Fig. 3A). Firing of bursts was found in 50% of the recorded SPNs in patients with PD, which were designated as “bursty” SPNs at a minimum of 1 detected
burst. The rate of burst activity per bursty SPN in patients with PD was 9.5 ± 1.2 bursts per 10 s. Burst activity was present to a similar extent in putamen and caudate SPNs of patients with PD (P = 0.3). This activity pattern was not present in SPNs of patients with ET in whom the characteristic occasional isolated SPN spikes were followed by prolonged silences compatible with the low firing rate of these neurons in the normal animal. Firing of bursts was also detected in SPNs recorded in patients with ID (Fig. 3B). However, compared with patients with PD, the SPN burst activity was lower in patients with ID (firing of bursts was found in 29% of the recorded SPNs in patients with ID at a rate of 4.8 ± 0.8 bursts per 10 s, P < 0.001; Fig. 3 C and D). The parameters for burst detection using the “surprise” method (25) were selected with strict limits for unequivocal classification of short duration spike bursts in all groups of patients. This method avoided the detection of longer spiking epochs as bursts in the typically irregular but overactive spike train of SPNs in PD that would have resulted in overestimation of burst activity (SI Materials and Methods). Nevertheless, we applied additional methods of burst analysis, including some that are less influenced by varying firing frequencies (26–28). The percentage of neurons that fired with bursts was similar, but the burst rate differed across detection methods, revealing the influence of frequency. However, significant differences in SPN burst activity between PD, ID, and ET were consistently found with all detection methods (Fig. S3). Therefore, there is a clear increase in the SPN burst activity in PD. Notably, the progression of burst firing from ET to ID and further to PD was similar to changes in the frequency domain, suggesting that common mechanisms probably generate the higher firing rates and bursting.

SPNs of the studied parkinsonian macaques also fired with bursts, as detected with the same burst parameters and algorithms as used for analysis in SPNs of patients. Burst firing in SPNs of these animals was as frequently detected as previously reported (19) (41% of the recorded SPNs in the two MPTP-treated NHPs fired with bursts at the rate of 4.4 ± 0.6 bursts per 10 s). Also in agreement with previous data, spontaneous firing of bursts in SPNs was not observed in the normal animal during recordings at rest (SPN burst activity has been described in normal primates in association with active movement) (29). This difference in the SPN activity pattern between the normal and parkinsonian state in NHP reproduces the changes observed between patients with ET and PD (Fig. 3 C and D).

Discussion

This is a systematic report of human SPN recordings. We found that SPN discharges in advanced PD are characterized by increased activity (~30 Hz) and frequent spike bursting firing. This characterization is relative to our findings in neurological patients with ET and ID and to comparison with the normal and parkinsonian states in NHPs. As a surrogate for normal control subjects, patients with ET are affected by a disorder that has not been associated with striatal pathology or dysfunction. Briefly, the pathophysiology of ET involves the cerebellum and connected brainstem areas where imaging and postmortem studies have shown Purkinje cell loss and other histological abnormalities (30, 31), probably leading to oscillatory activity in the cerebellolothalamocortical circuit (32). On the other hand, patients with the isolated (cervical and cranial) dystonia included in this study offer a disorder with largely recognized striatal mechanisms. Thus, ID provided a test for specificity of SPN alterations in PD, particularly because these two disorders may express some common clinical features. Importantly, primary dystonia is not associated with cell degeneration but microstructural, synaptic, and circuit abnormalities, some of which are influenced by dopamine signaling (22). Furthermore, imaging studies have shown altered dopamine D2 receptor binding in the putamen and associated network changes in patients with dystonia, including its focal forms (33).

Concurrent with the premise, the present recordings showed that the SPN activity in ET matched that of the normal state in NHP (low, irregular single spiking below 2–3 Hz). ET could, therefore, represent the “normality” status of the human SPN firing, and its comparison revealed the development of profound changes in PD. In addition, NHP with severe MPTP-induced parkinsonism exhibited striatal activity changes similar to those observed in patients with PD (18, 19), thus establishing a clear parallelism between normal-to-dopamine lesion animal states and ET-to-PD human disorders. Accordingly, SPN hyperactivity and burst firing likely are abnormal neuronal activity characteristics of the parkinsonian state. These results represent a finding for PD that is not totally unexpected. Similar changes in neuronal activity have been described in the STN and Gpi of patients with PD (34) and throughout the motor circuit in the NHP MPTP model (35).

SPNs lack autonomous activity and, thus, the increased firing that we observed in the parkinsonian state may be interpreted as mediated by dopamine loss inducing changes in other neurotransmitter systems. Particularly, the corticostriatal glutamatergic input is up-regulated in animal models of PD (36, 37). Patch-clamp recordings show increased frequency and amplitude of glutamatergic spontaneous excitatory postsynaptic currents (sEPSCs) in SPNs of rodents with 6-hydroxydopamine lesions (38, 39). Furthermore, the ratio of NMDA-to-AMPA receptor currents and their subunit composition in SPNs are altered in this model, thereby indicating postsynaptic glutamatergic changes (40). In line with the experimental data, motor cortex excitability and metabolic activity are enhanced in patients with PD (41, 42). The origin of SPN bursting activity is more conjectural. Rapid spiking followed by slow afterhyperpolarization (AHP) generating interburst silences, as shown in cholinergic neurons (43), may cause SPN bursts. The fact that burst firing was present in half of the recorded neurons suggests a mechanism potentially related to a dopamine receptor subtype. Notably, most bursty SPNs in the parkinsonian NHP exhibited a D1 dopamine receptor (D1R) response to levodopa (19) that could be related to up-regulation of slow AHP (44)

Fig. 2. Increase of SPN firing frequency in PD. The mean firing frequency of SPNs in PD compared with ET and ID (A), and a similar change in the parkinsonian NHP compared with the normal NHP (B). Analysis of the probability of predicting PD by the SPN firing frequency (C). The probability to predict PD based on SPN frequencies ≥13 Hz (minimal value in NHP) had 93% accuracy. SPNs recorded in patients with PD, ID, and ET are plotted in blue, gray, and black, respectively. The area between the dashed lines represents overlapping values between PD and ID. *P < 0.001 versus ET or normal animal (ANOVA followed by Bonferroni post hoc test or unpaired t test). **P < 0.001 versus ET and ID probability together (logit model). Error bars represent SEM.
mine loss (18). Also it is noteworthy that optogenetic stimulation drugs undergo similar up-regulation of activity following dopaminergic D2 dopamine receptor (D2R) and giving rise to the hypokinesia is associated with increased activity of substantia nigra pars compacta (SNc) with the present data, experimental evidence indicates that dopamine loss in striatal circuits has remained an important role in the origin of bradykinesia/akinesia. In line with loss of D1R signaling. Other convergent signals on SNpc that may also participate in burst generation include muscarinic M1 and GABA receptors. M1 receptors control slow inactivating K channels of the KCNQ type involved in the “up state” silencing of SNpc (45), and GABA receptors control timely changes in membrane hyperpolarization, as described in the globus pallidus (16). Thus, SNpc bursts in PD may ultimately result from the complex interplay of signals in striatal microcircuits. Network mechanisms may also intervene, given that burst firing is increased across basal ganglia stations after dopamine depletion (46).

Regardless of the specific mechanisms causing the SNpc activity changes found in PD, these firing abnormalities likely play an important role in the origin of bradykinesia/akinesia. In line with the present data, experimental evidence indicates that dopamine depletion is associated with increased activity of SNpc expressing D2 dopamine receptor (D2R) and giving rise to the indirect striatopallidal projection, and such enhancement is associated with reduced movement capacity (15, 47, 48). Therefore, increased activity in the indirect circuit might be proposed as one possible mechanism for our findings; however, the current technology does not allow distinguishing between SNpc expressing D1R or D2R in humans. Interestingly, the available NHP data indicate that SNpc that are differentially modulated by dopaminergic drugs undergo similar up-regulation of activity following dopamine loss (18). Also it is noteworthy that optogenetic stimulation of direct or indirect pathway in rodents was shown to produce different behavioral outputs (47), but also elicited both excitations and inhibitions in the basal ganglia output nuclei (49). Moreover, both pathways are simultaneously activated during initiation of a motor task, indicating a cooperative participation (50). As new light is shed on the dichotomous (direct/indirect) canonical model (2, 3), clearly, one important step forward would be to identify the SNpc subpopulation changes that occur in NHP models with the same SNpc hyperactivity as in human PD. Until these data become available, we tentatively posit that PD akinesia may be related to hyperactive SNpc interfering with the fine coordination of discharges within and between output pathways that is necessary for normal movement.

Finally, the striatal abnormalities of ID that include dopaminergic and cholinergic signaling (51), and its clinical overlap with PD would predict that the SNpc activity might also be increased in ID. Consistently, the present SNpc recordings in ID patients showed increased firing frequencies and spike bursts compared with PD, supporting a pattern of striatal dysfunction in ID. The escalation of SNpc firing changes from ID to PD is compatible with a continuum in pathophysiologic mechanisms from the normal to the dystonic, parkinsonian, and dyskinetic states (52). In sum, the findings reported here expand the abnormalities in the human striatum associated with the parkinsonian and dystonic states further validating experimental results. More importantly, the SNpc firing alterations found in patients with PD reveal the development of major striatal dysfunction in PD. The impact of dopamine loss in striatal circuits has remained an unresolved issue in PD pathophysiology, and this study, showing marked SNpc changes in patients, provides support for a significant striatal role in network abnormalities.

Materials and Methods

Patients. The Institutional Review Board of Emory University reviewed and approved the study, and all patients gave their informed consent. Patient inclusion was based on the following conditions: (i) an established, unequivocal diagnosis made by a movement disorder specialist, (ii) recordings in DBS surgery without general anesthesia or sedative drugs, and (iii) electrode trajectories passing through the striatum as permitted by the individual surgical approach and the intended mapping of the DBS target area. Because selection of patients for surgery depended solely on medical indication for DBS treatment, their clinical characteristics could not be matched across groups (Table 1). Patients were asked to relax and not to move during the recordings to obtain data of spontaneous neuronal firing at resting conditions and avoid movement-related activity. Tremor at rest was not observed during the striatal recordings in the majority of patients, including many patients with PD. The recording method was the same not only across the groups of PD, ID, and ET patients, but also in nonhuman primates. As SNpc recording is difficult with rapid movement of the electrode, the electrode was lowered very slowly with a hydraulic microdrive in patients to simulate the small steps (10–20 μm; NANN Instruments) used in animal recordings. Typically, spiking could be found variably in the striatal tracks as the electrode advanced at a slow speed. Neurons with very low activity or pausing as the electrode was advanced could have been undetected similarly in patients with PD, ID, and ET. After listening to or seeing a spike, the movement stopped to record the activity independent of how fast or slow the spiking was. Then, the position of the electrode was slightly adjusted for best unit isolation, and after firing stability was verified, data were saved for 1–3 min. We established this minimum duration of the spike train to be accepted for analysis to avoid very short recordings that are inadequate to determine the firing frequency of neurons with variable activity. Single spike traces where the unit could be classified directly on-line as TAN interneurons by the typically large and long waveform (>1.8 ms) were not saved (53). All other spike trains were stored for off-line analysis.

NHP. Studies in NHPs (three adult macaques) were conducted in accordance with the NIH Guide for the Care and Use of Laboratory Animals (54).

Details of methods and data analyses are provided in SI Materials and Methods.

ACKNOWLEDGMENTS. This study was supported by NIH Grants NS045962 and NS073994, National Center for Research Resources RR000165, Office of...


