Discharge Rate of Substantia Nigra Pars Reticulata Neurons Is Reduced In Non-Parkinsonian Monkeys With Apomorphine-Induced Orofacial Dyskinesia

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Submitted 27 October 2003; accepted in final form 24 April 2004

Nevet, Alon, Genela Morris, Guy Saban, Nina Fainstein, and Hagai Bergman. Discharge rate of substantia nigra pars reticulata neurons is reduced in non-parkinsonian monkeys with apomorphine-induced orofacial dyskinesia. J Neurophysiol 92: 1973–1981, 2004. Involuntary movements (dyskinesia) are a common symptom of dopamine-replacement therapy in parkinsonian patients, neuroleptic drug treatment of mental patients, and tic disorders. Levodopa-induced dyskinesia has been shown to be associated with substantial reduction of firing rate in the internal part of the globus pallidus. This study characterizes the changes that occur in the activity of the substantia nigra pars reticulata (SNr) of non-parkinsonian (normal) monkeys with apomorphine (APO)-induced orofacial dyskinesia. We conducted extracellular recordings of SNr neurons of two monkeys before and after induction of orofacial dyskinesia by systemic administration of APO. Involuntary orofacial movements appeared a few minutes after the injections and lasted 20–40 min. Almost all recorded neurons changed their firing rate after APO injection (96%), and most declined (70%). The mean amplitude of decreases was also larger than that of increases (40 vs. 21% of the control rate). Changes in firing pattern were not significant on average. Pairs of SNr neurons were uncorrelated before APO injection, similar to the normal pallidum. However, unlike the increased correlations in the pallidum that accompany Parkinsonism, orofacial dyskinesia in non-parkinsonian monkeys was not associated with changes in correlation between SNr neurons. We conclude that normal monkeys treated with APO can model orofacial dyskinesia and tic disorders that are a consequence of dopaminergic over-activity. These symptoms appear to be more related to reduced firing rate of SNr neurons and thus to disinhibition of their targets, than to changes in pattern and synchronization.

INTRODUCTION

Involuntary movements, termed dyskinesia, are present in different forms in various clinical syndromes. The most common are Levodopa-induced dyskinesia (LID) in Parkinson’s disease (PD), tardive dyskinesia due to anti-dopaminergic treatment, and Tourette’s syndrome (TS). The pathology in PD is extensive death of dopaminergic midbrain neurons, and the dopamine precursor Levodopa is the gold standard in drug therapy of PD. However, within months or years of treatment, most patients develop adverse side effects, including dyskinesia. The dyskinetic manifestations in PD primarily include chorea (dance-like limb and trunk movements) and dystonia (abnormal posture of limbs) (Bezard et al. 2001). Tardive dyskinesia is a consequence of long-term anti-dopaminergic treatments (classic neuroleptic drugs) usually targeting mental health illnesses. Its prominent manifestation is usually orofacial (Jenner and Marsden 1987). TS is a disorder characterized by chronic tics, often accompanied by obsessive compulsive behavior and attention deficits (Jankovic 2001).

Although each of these syndromes is unique and may have a distinct pathophysiology (Mink 2003), evidence from various studies suggests that a common characteristic of the different types of dyskinesia is inappropriate dopaminergic activity. Drugs that might induce involuntary movements either increase the extracellular level of dopamine in the striatum or directly act on dopamine receptors by activating them or increasing their sensitivity (Bezard et al. 2001; Jenner and Marsden 1987). Tic disorders, mainly TS, have been shown to involve dopaminergic hyper-innervation of the striatum (Jankovic 2001; Singer et al. 2002). Consistently, the most effective anti-tic agents are dopamine-receptor antagonists (Jankovic 2001), and injection of the dopamine D1/D2 agonist apomorphine (APO) to normal human subjects and monkeys induces blinking and orofacial movements (Blin et al. 1990; Kleven and Koek 1996). In this respect, dyskinesia seems to be at the opposite extreme from PD: whereas striatal dopamine depletion in PD causes bradykinesia and akinesia, dyskinesia appears to involve an excessive effect of dopamine in this structure.

The introduction of the 1-methyl-4-phenyl-1,2,3,6-tetrahydropyridine (MPTP)-treated monkey as a model of PD has proved instrumental in the study of this disease (Jenner 2003) as well as for LID (Bezard et al. 2001; Mitchell et al. 1992; Papa et al. 1999). In parkinsonian patients (Hutchison et al. 2003; Levy et al. 2001; Lozano et al. 2000), primates (Boraud et al. 2001; Filion et al. 1991; Heimer et al. 2002; Papa et al. 1999), and rodents (Ruskin et al. 1999), dyskinesia induced by dopamine replacement therapy (DRT) has been shown to be associated with both reduction in the firing rate of GPe neurons and with changes in their firing pattern. Animal models of other types of dyskinesia have also been introduced. These include models of tardive dyskinesia (Bedard et al. 1982; Clow et al. 1976; Klawans and Miller 1982; Lozano et al. 2000; Papa et al. 1999).

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et al. 1978) and dyskinesia induced by dopaminergic hyperactivity in otherwise drug naive (normal) animals (Jenner 2000; Mones 1972; Pearce 1999). Although single-neuron electro-physiological recordings have been conducted in rodent models of dyskinesia without striatal dopamine depletion (Ruskin et al. 2002; Waszczyk et al. 1984), such studies have not been performed on primates.

In primates, studies of LID have focused mainly on the activity of the internal segment of the globus pallidus (GPI), the larger of the two output nuclei of the BG, and have neglected the substantia nigra pars reticulata (SNr). Although these two nuclei are often clumped together as the output stage of the BG, they differ in a number of critical ways. One of these two nuclei are often clumped together as the output stage of the SNr. They differ in the type of movements associated with their activity: the SNr has been shown to be primarily related to ocular and orofacial movements (DeLong et al. 1983; Hikosaka and Wurtz 1983; Schultz 1986; Wichmann and Kliem 2004), but the GPI is mainly associated with movements of the limbs (Alexander and DeLong 1985; Anderson and Horak 1985; Georgopoulos et al. 1983).

Although the discharge activity of the SNr has never been directly recorded in dyskinetic human patients or monkeys, the SNr involvement in orofacial dyskinesia has been clearly demonstrated in lesion and metabolic studies. In rats, SNr lesions attenuate orofacial movements induced by microinjection of amphetamine to the ventrolateral striatum (Canales et al. 2000) and lesions of the SC attenuate orofacial dyskinesia induced by APO (Redgrave et al. 1980). In cats, metabolic changes were observed in the SNr and SC after orofacial dyskinesia was induced by injection of kainic acid to the striatum (Jaspers et al. 1989). Metabolic studies in dyskinetic monkeys are also suggestive of the involvement of the SNr (Mitchell et al. 1985).

In this study, we focused on the mechanisms underlying the appearance of orofacial dyskinesia in a non-parkinsonian primate. We administered APO to induce orofacial dyskinesia and recorded the simultaneous activity of several SNr neurons before, during, and after administration of the drug. Importantly, the short-lasting and rapid effects of APO enabled the recording of the same neurons before and after APO was administered. Our first goal was to investigate whether changes in the discharge rate and pattern of single SNr neurons were associated with the appearance of dyskinesia. Previous studies have indicated that the activity of normally uncorrelated neurons in the primate globus pallidus becomes highly correlated under MPTP treatment (Raz et al. 2000) and that DRT reverses this excess correlation (Heimer et al. 2002). Thus our second goal was to test the hypothesis that in addition to changes in the activity of single neurons, the appearance of orofacial dyskinesia is reflected in changes in neural correlations within the SNr.

**METHODS**

**Animals**

Two female *Macaca fascicularis* monkeys (E and G), weighing 2.5 and 3.5 kg were used in this study. Handling of the monkeys and all procedures were in accordance with the National Institutes of Health Guide for the Care and Use of Laboratory Animals (1996) and with the Hebrew University guidelines for the use and care of laboratory animals in research, supervised by the institutional Animal Care and Use Committee.

**Surgical procedures**

A square recording chamber with a 27-mm inner side was attached to the skull with an acrylic cap to allow access to the SNr. In monkey E, the recording chamber was tilted 56° laterally in the coronal plane with its center targeted at the stereotaxic coordinates of the SNr (Martin and Bowden 2000; Szabo and Cowan 1984). In monkey G, the recording chamber was placed with its center dorsal to the stereotaxic coordinates of the SNr. The chamber coordinates were verified with MRI imaging (Biostec Bruker 4.7 Tesla animal system, fast-spin echo sequence; effective TE = 80 ms and TR = 2.5 s, 13 coronal slices 1 or 2 mm wide) by alignment of the two-dimensional MRI images with the sections from the atlas (Martin and Bowden 2000; Szabo and Cowan 1984). At the end of the recording period, the chamber and cap were removed, and the skin was sutured. After a recovery period, the monkeys were sent to a primate sanctuary. All surgical and MRI procedures were performed under general and deep anesthesia.

**Neuronal recording**

During recording sessions the monkeys’ heads were fixed. Eight glass-coated tungsten microelectrodes (impedance: 0.3–1.2 MΩ at 1,000 Hz), confined within a cylindrical guide (1.65 mm ID), were individually advanced (EPS, Alpha-Omega Engineering, Nazareth, Israel) to the SNr. Signals from the electrodes were amplified with a gain of 10K and band-pass filtered with a 300- to 6,000-Hz 4-pole Butterworth filter (MCP+, Alpha-Omega Engineering). Extracellular action potentials were detected and classified on-line using a template-matching algorithm (MSD, Alpha-Omega Engineering). Spike-detection signals and behavioral events were logged to a data acquisition system at 12 kHz (AlphaMap, Alpha-Omega Engineering). Cells were classified as SNr neurons if they were found at the expected stereotaxic coordinates (based on the MRI and the primate atlas data) and by their electrophysiological characteristics. SNr neurons exhibit a relatively narrow spike shape and have a distinctively high firing rate (DeLong et al. 1983; Schultz 1986). Firing characteristics of neighboring neurons and fibers were also used to verify the identification of the target cells as SNr neurons. For example, adjacent to the identified SNr, and usually deeper, neurons of the oculomotor nucleus and fibers of the oculomotor nerve were often encountered, displaying characteristic prominent changes in discharge rate related to eye movements.

**Induction and recording of dyskinesia**

In each recording session, orofacial dyskinesia was induced by a single intramuscular injection of 0.1 mg/kg apomorphine (APO) HCl 1%. Neither monkey vomited or displayed symptoms of nausea, and therefore anti-emetic drugs (Gancher et al. 1989) were not required. Orofacial movements were recorded by an infrared reflection detector (Dr. Bouis, Freiburg, Germany). The infrared signal was amplified with a gain of 500, band-pass filtered with a 1- to 100-Hz 4-pole Butterworth filter (MCP+, Alpha-Omega Engineering), and sampled at 750 Hz. In addition, three video cameras recorded the monkey’s face, upper limbs, and lower limbs. The video was recorded digitally (AVer-s 2.54, AverMedia Systems, Taipei, Taiwan) for later off-line analysis. On-line detection of prominent orofacial movements was also conducted by a human observer. When such a movement was detected, a button was pressed, and this event was logged. All behavioral and neuronal data were recorded by the same system (AlphaMap, Alpha-Omega Engineering).
The monkeys were trained and engaged in a behavioral paradigm consisting of a visuo-motor delayed response task as part of a study of the normal SNr previous to this study. After these experiments were completed, the monkeys were no longer water deprived, and apomorphine injections were started. Thus the monkeys were already used to sitting in the laboratory during recording sessions but were not specifically trained to sit quietly, and occasional limb movements did occur. Because we did not observe abnormal limb movements after APO injections, and the major effect of APO was orofacial without dyskinesia of the limbs, we did not exclude limb movement periods before and after APO injections.

**Data analysis**

Spike-trains were used for further analysis only if their spike waveforms were reliably separated from those of other units during the on-line spike sorting. Each of these spike trains was then analyzed for stability. In this analysis, the rate of each unit as a function of time was displayed graphically for the entire period of recording. Only units that were judged as steady in the 15 min prior to APO injection and did not completely and irreversibly cease to discharge during the first 20 min after APO administration (6 neurons did) were considered for all further analyses. All other units were excluded to avoid possible artifacts due to electrode displacements.

The spike train of each neuron was divided into two periods: control interval—the activity in the 15 min preceding APO injection and test interval—the 15 min starting 5 min after the injection of the drug. The test interval coincided with the maximal clinical effect (see RESULTS). Changes in firing rate were determined by comparing the rates in the two intervals calculated in 200-ms bins (Mann-Whitney U test, significance at $P < 0.01$). Onset of rate changes was calculated using a rate vector starting from APO injection, in 1-s bins, filtered with a 20-s Gaussian. The rate and SD in the first 30 s were used as a reference. First, the earliest time the rate deviated by >3 SD from the reference was detected. Then the latest time there was a deviation of >1 SD from the reference, occurring earlier than the 3-SD threshold was detected and defined as the onset time of rate changes.

Firing patterns were examined by comparing the second, third, and fourth statistical central moments of the inter-spike interval (ISI) distribution (variance, skewness, and kurtosis, respectively). A statistical central moment is the average of differences between the samples and the mean taken in the power indicated by the order of the moment. The third and fourth moments were normalized to the variance taken as the onset time of rate changes.

Cross-correlation functions (Perkel et al. 1967) were calculated for the control and the test periods with a 1-ms bin size for a time window of 2 s. The SD and expected correlation were calculated for each cross-correlogram using the first 1/5 and last 1/5 of the correlogram. A correlation between a pair of neurons was considered significant when it included at least three consecutive bins that deviated from the expected correlation by at least three SD. Correlation coefficients were calculated between 15-min binary spike trains with a 1-ms bin size (the same test intervals described in the preceding text). Only neurons recorded from different electrodes were examined for correlation analysis to avoid artifacts due to spike sorting (Bar-Gad et al. 2001b).

**RESULTS**

We recorded the activity of 96 SNr neurons that were indicated on-line as reliably separated from other units and were stable for the control and the test periods (35 min). Of these neurons, 75 passed the off-line stability test and were used for subsequent analysis (monkey E, 37; monkey G, 38). The SNr neurons included in the analysis were recorded during 30 sessions (monkey E, 14; monkey G, 16), and generated 71 simultaneously recorded pairs [1 neuron may have been paired with >1 partner, thus $n$ simultaneously recorded stable neurons yielded $n(n - 1)$ pairs for the cross-correlation study (monkey E, 41; monkey G, 30 pairs).

**Orofacial dyskinesia is triggered by systemic injection of APO**

Orofacial movements were markedly increased both in amplitude and frequency soon after the systemic injection of 0.1 mg/kg APO (Fig. 1, A and B). Typically, episodes of tongue protrusions and contractions of the facial muscles were apparent within 1–3 min and could be observed for a period of 20–40 min. Off-line analysis of video recordings revealed a marked increase in the rate of blinking (Fig. 1C), with a similar time course. This is in accordance with previous studies that have shown that blinking rate is correlated with dopaminergic activity (Karson 1983; Karson et al. 1981). In monkey E, a total of 30 APO injections were given over a period of 103 days. In monkey G, a total of 26 APO injections were given over a period of 87 days. Changes in APO impact on orofacial movements and blinking did not have a consistent or significant tendency with time (calculated for 15 sessions with blinking rate analysis and for 7 sessions with infra-red analysis), and no priming effect was apparent.

**SNr neuronal firing rates change after administration of APO**

Because the main mechanism by which the SNr influences its targets is believed to be disinhibition (Hikosaka et al. 2000), we first analyzed changes in firing rates of individual SNr neurons after injection of APO. Nearly all the recorded SNr neurons displayed significant changes in their firing rate after APO injection, at the time of the appearance of orofacial dyskinesia (Fig. 2A). In both monkeys, most neurons decreased their firing rate (78% in monkey E, 61% in monkey G; Fig. 2B). The mean amplitude of the decreases was also larger than that of the increases (40 vs. 21% of the control rate; Fig. 2B). Eighty-two percent of the neurons changed their firing rate before symptoms started, and the average latency between rate change onset and tic onset was 1 min 51 s. The peak of rate changes also preceded the peak of symptoms by 4 min 36 s on average. The orofacial dyskinesia observed by the recorded infrared reflection tended to be related to the changes in the firing rate. However, we could not demonstrate a significant correlation between the rate of blinking and orofacial dyskinesia and the amplitude of changes in discharge rate of neurons recorded simultaneously (35 neurons recorded over 15 sessions with blinking, 21 neurons recorded over 7 sessions with infrared signal, $P > 0.1$). Additionally, we analyzed poststimulus time histograms (PSTHs) aligned with movements detected by the infrared reflector. However, the results were indecisive. As with the behavioral parameters,
Changes in SNr firing rate did not show a consistent trend between sessions.

Changes in firing pattern of SNr neurons are variable

In addition to changes in firing rate, akinesia (Ruskin et al. 2002; Vitek and Giroux 2000) and dyskinesia (Boraud et al. 2001; Hutchison et al. 2003; Ruskin et al. 2002) are thought to be associated with changes in the firing pattern of neurons in basal ganglia output nuclei. Auto-correlation functions are affected by the firing rate of the cells (Bar-Gad et al. 2001a). Therefore to track consistent changes in discharge patterns across the entire neuronal population, we first examined the properties of the distribution of interspike intervals (ISIs) of each neuron before and after APO administration and averaged across the population (Table 1). The first central moment of the ISI histogram is equivalent to the mean firing rate (see preceding text for changes in firing rate). The second moment is the variance, and this parameter increased significantly. To further examine this change in pattern, we calculated the variance separately for neurons that increased and neurons that decreased their firing rates. This analysis revealed that only the population of decreasing-rate (increased ISI) neurons had a higher variance of ISI after APO. However, the variance also remained higher after normalizing it to the rate (coefficient of variation increased by 0.33 on average, Mann Whitney U test, \( P < 0.05 \)). The third (skewness) and fourth (kurtosis) moments characterize the asymmetry and the proportions of the peak versus the marginal area of the distribution, respectively. Over the population, average skewness and kurtosis of ISI histograms were higher after APO injection (Table 1, Fig. 3A), but these changes were not significant (Mann Whitney U test, \( P > 0.05 \)).

We also examined the average skewness of density histograms, indicating burstiness level (Kaneoke and Vitek 1996). The average skewness of density histograms did not significantly change (Table 1), indicating that the proportion of bursty neurons did not change after APO injection and development of orofacial dyskinesia.

Pair-wise neuronal correlation

Simultaneously recorded neurons have been shown to be significantly correlated in the globus pallidus of tremulous parkinsonian patients (Hurtado et al. 1999; Levy et al. 2002), and MPTP-treated monkeys (Raz et al. 2000), in contrast to the uncorrelated activity found normally (Nini et al. 1995; Raz et al. 2000). To examine whether changes in firing synchrony play a role in the pathophysiology of orofacial dyskinesia in non-parkinsonian monkeys, we examined the cross-correlations between simultaneously re-
corded SNr neurons, as well as the correlation coefficients, before and after APO administration. The activity of the neurons in the normal phase was found to be mostly uncorrelated (Table 1, Fig. 3B, top). This is in accordance with early studies of the SNr in rats (Wilson et al. 1977) and with previous results obtained in the primate GP (Nini et al. 1995; Raz et al. 2000). However, unlike the case in Parkinsonism and its MPTP model, the fraction of correlated pairs of neurons and the average correlation coefficient did not change after APO injection (Fig. 3B, bottom, Table 1).

**TABLE 1. Summary of patterns and correlation changes related to apomorphine injection**

<table>
<thead>
<tr>
<th>Pattern</th>
<th>ISI STD</th>
<th>ISI Skewness</th>
<th>ISI Kurtosis</th>
<th>Density Skewness</th>
<th>Correlated Pairs, %</th>
<th>Correlation Coefficients</th>
</tr>
</thead>
<tbody>
<tr>
<td>Monkey E Before APO</td>
<td>28.2 ± 4.8</td>
<td>5.4 ± 0.6</td>
<td>90.1 ± 21.3</td>
<td>1.6 ± 0.06</td>
<td>2.4</td>
<td>-0.0001 ± 0.0002</td>
</tr>
<tr>
<td>After APO</td>
<td>65.3 ± 22.1*</td>
<td>6.5 ± 1.4</td>
<td>249 ± 175.1</td>
<td>1.5 ± 0.07</td>
<td>2.4</td>
<td>0.0007 ± 0.0003</td>
</tr>
<tr>
<td>Monkey G Before APO</td>
<td>66.8 ± 24.3</td>
<td>8.4 ± 3.4</td>
<td>560.3 ± 454</td>
<td>1.6 ± 0.07</td>
<td>6.7</td>
<td>0.0001 ± 0.002</td>
</tr>
<tr>
<td>After APO</td>
<td>207 ± 82.7*</td>
<td>12.7 ± 4.4</td>
<td>1104 ± 625.2</td>
<td>1.6 ± 0.06</td>
<td>6.7</td>
<td>0.0006 ± 0.0009</td>
</tr>
</tbody>
</table>

Results are given as means ± SE. SD (standard deviation) was calculated as the square root of the variance normalized by \((n-1)\). Number of neurons for Monkeys E and G were 37 and 38, respectively and number of pairs were 41 and 30, respectively. *P < 0.05.
We report results regarding changes in SNr electrophysiological activity using a primate model of orofacial dyskinesia induced by systemic injection of APO. The orofacial movements we observed were stereotypic nonrhythmic contractions of orofacial muscles and tongue protrusions. Because the animals were not dopamine-depleted, we believe that this model represents orofacial tic disorders better than other forms of dyskinesia. Our results indicate that SNr neurons change their firing rates after APO injection, and their average rate is reduced (Fig. 2, Table 1). Changes in the activity of the basal ganglia (BG) output nuclei have been hypothesized to mediate the appearance of dyskinesia. However, only pallidal neurons of dyskinetic primates have been studied so far, and these studies were performed on dopamine-depleted animals. Dyskinesia was previously observed in non-parkinsonian monkeys treated with dopamine agonists (Pearce 1999). However, the only study that has recorded neuronal activity in normal monkeys treated with dopamine agonists (Boraud et al. 2001) focused on pallidal neurons (and not SNr), and dyskinesia was not observed in that study.

It has been claimed that the decrease in the mean firing rate of BG output nuclei results in reduced inhibition of their targets and, as a consequence, in reduced inhibition of involuntary movements (DeLong 1990). The release of involuntary eye-movements by muscimol inhibition of SNr activity is in line with this model (Hikosaka and Wurtz 1985). Our results are in accordance with this model and extend the observation that dyskinesia due to dopamine-replacement therapy (DRT) is associated with decreased firing rates in the GPi of parkinsonian patients (Levy et al. 2001; Lozano et al. 2000; Merello et al. 1999; Vitek et al. 1999), MPTP-treated primates (Boraud et al. 1998; Filion et al. 1991; Papa et al. 1999) and the SNr and entopeduncular nucleus (analogue of GPi) of rodents with nigrostriatal pathway lesions (Murer et al. 1997; Ruskin et al. 2002). These results also validate previous studies of the SNr of intact rodents treated with dopamine agonists (Ruskin et al. 2002; Waszczak et al. 1984) and emphasize the relevance of rodent studies for primates and humans. Moreover, although most of the neurons in this study exhibited a decreased discharge rate, a substantial minority showed significant increases. This is also in accordance with previous studies of the rodent SNr (Murer et al. 1997) and the primate pallidum (Matsumura et al. 1995).

It has been shown that GPi lesions may abolish dyskinesia in PD (Hutchison et al. 2003; Lang 2000; Vitek et al. 2003). Thus reduction of discharge rate cannot be its sole cause, and changes in firing pattern probably also play a role. Changes in ISI distributions of neurons in BG output nuclei were observed in nigrostriatal lesioned rats with dopamine agonists-induced rotations (Ruskin et al. 2002). Changes in discharge regularity were found in the globus pallidus of MPTP-treated monkeys with APO-induced dyskinesia (Boraud et al. 2001), and long pauses were reported to appear in the GPi of dystonic parkinsonian patients (Hutchison et al. 2003; Vitek et al. 1999). However, we did not observe similar consistent changes in SNr firing pattern in our model. Whereas the variance of ISI histograms significantly changed, these changes were only significant in neurons that decreased their firing rate following APO administration. Changes in other parameters we examined were statistically nonsignificant (Fig. 3A, Table 1).

The spiking activity of pairs of pallidal neurons in normal monkeys was reported to be uncorrelated (Nini et al. 1995; Raz et al. 2001). Our results show that SNr neurons follow this rule as well. However, although studies of pallidal neurons of primates with MPTP-induced akinesia (Raz et al. 2000) and tremulous parkinsonian patients (Hurtado et al. 1999; Levy et al. 1978 A. NEVET, G. MORRIS, G. SABAN, N. FAINSTEIN, AND H. BERGMAN J Neurophysiol • VOL 92 • OCTOBER 2004 • www.jn.org

FIG. 3. A: an example of interspike interval (ISI) distribution of a neuron (monkey G) before and after apomorphine injection. B: cross-correlations among 4 simultaneously recorded SNr neurons (monkey E) before (top) and after (bottom) apomorphine injection. Top right of each square: the ordinate represents conditional rate (in spikes/s); bottom left: ordinate is given in SD units. The SD was calculated using the 1st and last 5th of the vector. Both parts present the correlations of the same neurons (mirror imaged).

DISCUSSION

We report results regarding changes in SNr electrophysiological activity using a primate model of orofacial dyskinesia induced by systemic injection of APO. The orofacial movements we observed were stereotypic nonrhythmic contractions of orofacial muscles and tongue protrusions. Because the
al. 2002) show enhanced correlations that disappear after optimal DRT (Heimer et al. 2002), we show here that SNr neurons of non-parkinsonian primates with APO-induced dyskinesia do not display altered correlations. Integrating the current results with those of previous studies suggests that low dopaminergic activity is associated with correlated tonic discharge of the neurons in the output nuclei of the BG and akinnesia, whereas increasing the level of dopamine decorrelates the neuronal activity and enables initiation of voluntary movements. However, when the dopaminergic activity is elevated beyond a critical level, the rate of discharge of neurons in these nuclei decreases, and involuntary movements appear.

The SNr of primates has not been studied in the context of movement disorders as extensively as its counterpart, the GPi. However, the involvement of the SNr in the pathophysiology of orofacial dyskinesia was directly shown in several animal studies (Canales et al. 2000; Jaspers et al. 1989; Redgrave et al. 1980). It may be hypothesized that the SNr and the GPi contribute differentially to different types of dyskinesia. The SNr, which is known to be associated with orientation movements (Hikosaka et al. 2000), orofacial movements (DeLong et al. 1983; Schultz 1986), and sensory gating (Fendt et al. 2001; Koch et al. 2000), is very probably involved in orofacial dyskinesia. The GPi, on the other hand, being primarily related to limb movements (Anderson and Horak 1985; Georgopoulos et al. 1983; Mitchell et al. 1987), appears to be more involved in dyskinesia of the limbs and trunk in the form of chorea and dystonia (Bezard et al. 2001). The different roles of the dual facial representation in the GPi and the SNr should be explored in future studies.

Both the GPi and the SNr are affected directly by dopaminergic afferents (Lavoie et al. 1989; Prensa et al. 2000) and indirectly through striatal influence. The SNr of primates has been shown to be directly influenced by dopamine (Ruffieux and Schultz 1980). This may explain the appearance of orofacial dyskinesia in normal primates treated with dopamine agonists (Jenner 2000; Mones 1972; Pearce 1999). Moreover, the SNr is anatomically adjacent to the dopaminergic neurons of the substantia nigra pars compacta (SNc), and in vitro studies have confirmed dopamine receptor expression by SNr neurons (Levey et al. 1993; Mrzljak et al. 1996). The SNr may therefore be directly influenced by somato-dendritic release of dopamine (Cheramy et al. 1979; Cobb and Abercrombie 2002). This relationship may be the cause of orofacial dyskinesia in patients suffering from dopaminergic over-activity (Jankovic 2001). However, in the parkinsonian state, after the death of dopaminergic neurons of the ventral tier of the SNc, the SNr no longer receives this direct dopaminergic input. DRT may compensate for this loss in addition to its effect on the striatum. When the influence of DRT on the striatum is adverse, the SNr may still benefit from the direct compensation it receives, making it less adversely affected than the GPi. This hypothesis may explain the fact that LID in PD tends to be in the form of dystonia and chorea of the limbs and trunk (Vidalihat et al. 1999) more often than tic disorders or tardive dyskinesia, which tend to be more orofacial (Fahn et al. 1998; Leckman et al. 2001). Finally, because dyskinesia appears to be the result of reduced inhibition of BG targets, we suggest that increasing their inhibition may ease the symptoms of this disorder. This is supported by results of deep brain stimulation of thalamic nuclei in TS (Temel and Visser-Vandewalle 2004; Vandewalle et al. 1999).

ACKNOWLEDGMENTS

We thank Dr. Y. Ben-Shafl for helpful discussions, assistance with analyses, and fruitful comments on earlier versions of this manuscript, V. Sharkan-ski for technical support, G. Goelman for MRI, and O. Karasik for contributing to blinking off-line detection and monkey training.

GRANTS

This study was partly supported by a Center of Excellence grant administered by the Israeli Science Foundation, the German-Israel Binational Foundation, and the Federal Ministry of Education and Research–Israel Ministry of Science Israel-Germany collaboration in medical research.

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