

# Internal Pallidal Neuronal Activity During Mild Drug-Related Dyskinesias in Parkinson's Disease: Decreased Firing Rates and Altered Firing Patterns

J. I. Lee, L. Verhagen Metman, S. Ohara, P. M. Dougherty, J. H. Kim and F. A. Lenz

*J Neurophysiol* 97:2627-2641, 2007. First published Jan 10, 2007; doi:10.1152/jn.00443.2006

**You might find this additional information useful...**

---

This article cites 57 articles, 24 of which you can access free at:

<http://jn.physiology.org/cgi/content/full/97/4/2627#BIBL>

Updated information and services including high-resolution figures, can be found at:

<http://jn.physiology.org/cgi/content/full/97/4/2627>

Additional material and information about *Journal of Neurophysiology* can be found at:

<http://www.the-aps.org/publications/jn>

---

This information is current as of April 12, 2007 .

# Internal Pallidal Neuronal Activity During Mild Drug-Related Dyskinesias in Parkinson's Disease: Decreased Firing Rates and Altered Firing Patterns

J. I. Lee,<sup>1</sup> L. Verhagen Metman,<sup>2</sup> S. Ohara,<sup>3</sup> P. M. Dougherty,<sup>4</sup> J. H. Kim,<sup>5</sup> and F. A. Lenz<sup>5</sup>

<sup>1</sup>Department of Neurosurgery, Samsung Medical Center, Sungkyunkwan University School of Medicine, Seoul, Korea; <sup>2</sup>Rush University Medical Center, Chicago, Illinois; <sup>3</sup>Department of Neurosurgery, Kyoto Kizugawa Hospital, Kyoto, Japan; <sup>4</sup>Department of Anesthesiology, M.D. Anderson Medical Center, Houston, Texas; <sup>1,3,4,5</sup>Department of Neurosurgery, Johns Hopkins University, Baltimore; and <sup>2</sup>Experimental Therapeutics Branch, National Institute of Neurological Disorders and Stroke, National Institutes of Health, Bethesda, Maryland

Submitted 26 April 2006; accepted in final form 7 January 2007

**Lee JI, Metman LV, Ohara S, Dougherty PM, Kim JH, Lenz FA.**

Internal pallidal neuronal activity during mild drug-related dyskinesias in Parkinson's disease: decreased firing rates and altered firing patterns. *J Neurophysiol* 97: 2627–2641, 2007. First published January 10, 2007; doi:10.1152/jn.00443.2006. The neuronal basis of hyperkinetic movement disorders has long been unclear. We now test the hypothesis that changes in the firing pattern of neurons in the globus pallidus internus (GPi) are related to dyskinesias induced by low doses of apomorphine in patients with advanced Parkinson's disease (PD). During pallidotomy for advanced PD, the activity of single neurons was studied both before and after administration of apomorphine at doses just adequate to induce dyskinesias (21 neurons, 17 patients). After the apomorphine injection, these spike trains demonstrated an initial fall in firing from baseline. In nine neurons, the onset of ON was simultaneous with that of dyskinesias. In these spike trains, the initial fall in firing rate preceded and was larger than the fall at the onset of ON with dyskinesias. Among the three neurons in which the onset of ON occurred before that of dyskinesias, the firing rate did not change at the time of onset of dyskinesias. After injection of apomorphine, dyskinesias during ON with dyskinesias often fluctuated between transient periods with dyskinesias and those without. Average firing rates were not different between these two types of transient periods. Transient periods with dyskinesias were characterized by interspike interval (ISI) independence, stationary spike trains, and higher variability of ISIs. A small but significant group of neurons demonstrated recurring ISI patterns during transient periods of ON with dyskinesias. These results suggest that mild dyskinesias resulting from low doses of apomorphine are related to both low GPi neuronal firing rates and altered firing patterns.

## INTRODUCTION

Patients with advanced Parkinson's disease (PD) often respond to levodopa with rapid, random movements that are known as drug-related dyskinesias (Marsden 1994). The current model of basal ganglia function proposes that dyskinesias and other hyperkinetic disorders result from decreased neuronal firing rates in the globus pallidus interna (GPi) (Albin et al. 1989; DeLong 1990; Mitchell et al. 1989). This leads to decreased inhibitory outflow from the GPi to the thalamus that may result in increased activity of thalamocortical motor circuits and in dyskinesias (Holsapple et al. 1991; Perkel et al. 1964).

This model is based on studies in 1-methyl-4-phenyl-1,2,3,6-tetrahydropyridine (MPTP)-treated monkeys that show

parkinsonian motor behavior (Filion and Tremblay 1991; Miller and DeLong 1987), including drug-related dyskinesias (Boraud et al. 2001, 2002; Filion et al. 1991; Papa et al. 1999). High doses of dopamine agonists silence GPi neurons and lead to pronounced dyskinesias (Papa et al. 1999) as predicted by the current model of basal ganglion function (DeLong 1990). At lower doses, firing may or may show decreased rates and altered in firing patterns (Boraud et al. 1998, 2001, 2002; Filion et al. 1991).

Intra-operative administration of apomorphine in patients with advanced Parkinson's disease leads to ON with dyskinesias and to GPi firing rates that are lower than those during the OFF state (Levy et al. 2001; Merello et al. 1999). However, decreased GPi neuronal firing rates cannot completely explain drug-related dyskinesias because lesions of GPi both silence these neurons and lead to abolition in drug-related dyskinesias (Lang et al. 1997; Vitek et al. 2003). Therefore changes in measures of GPi activity other than firing rate must account for drug-related dyskinesias.

In patients with PD at the onset of apomorphine induced dyskinesias, the firing of single GPi neurons has been characterized by a change to an irregular pattern (4 neurons) (Merello et al. 1999) and by no apparent change in the firing pattern (1 neuron) (Levy et al. 2001). We now test the hypothesis that GPi neuronal activity is related to the mild transient dyskinesias that are evoked by administration of low doses of apomorphine to patients with advanced PD and drug-related dyskinesias. The results demonstrate that GPi spike trains during dyskinesias are characterized by both decreased firing rates and altered firing patterns.

## METHODS

These studies were carried out in patients with idiopathic Parkinson's disease and drug-related dyskinesias. Patients who were already scheduled to undergo unilateral pallidotomy were approached for participation in the protocol for this study. The protocol was approved and renewed annually by the Joint Committee on Clinical Investigation at Johns Hopkins University and by the Institutional Review Board at the National Institute of Neurological Disorders and Stroke. To mimic the effect of levodopa, we used intravenous apomorphine hydrochloride, a nonselective D1/D2-dopamine receptor agonist. Intravenous apomorphine has an immediate (<1 min) onset of action,

Address for reprint requests and other correspondence: F. A. Lenz, Dept. of Neurosurgery, Meyer 8-181, Johns Hopkins Hospital, Baltimore, MD 21287-7713 (E-mail: flenz1@jhmi.edu).

The costs of publication of this article were defrayed in part by the payment of page charges. The article must therefore be hereby marked "advertisement" in accordance with 18 U.S.C. Section 1734 solely to indicate this fact.

and a short (30 min) elimination half-life (Corsini et al. 1979; Gancher et al. 1989; Kempster et al. 1990; Verhagen et al. 1998). The effect size is identical to that of levodopa, but the response is short-lived.

Preoperatively, the patients' motor behavior was studied before and after the intravenous administration of apomorphine (Corsini et al. 1979; Gancher et al. 1998; Kempster et al. 1990; Verhagen et al. 1998). Intraoperatively, the patients' motor behavior and GPi neuronal firing were studied before and after administration of the "surgical" dose of apomorphine which was selected during preoperative testing.

### Preoperative studies

This phase of the study was designed to establish a safe, effective, dyskinesigenic, and well-tolerated dose for every patient, thereby limiting risks during the pallidotomy. During the month preceding surgery, patients were admitted to the National Institutes of Health Clinical Center (Bethesda, MD) for 2–4 days. Patients were started on domperidone, a peripheral dopamine antagonist, to prevent nausea (Corsini et al. 1979). Regular antiparkinsonian medications were withheld after 11 PM on the day prior to testing until completion of the preoperative evaluations on the following day. Patients then entered an apomorphine dose-finding phase, during which they received an average of six intravenous apomorphine injections. Doses were administered  $\geq 2$  h apart to a maximum of four per day.

Subjects and evaluators were blinded to the doses given, which ranged from 0.007 to 0.10 mg/kg. Doses were selected by an unblinded safety officer. A randomly inserted placebo injection was given in those cases where there was an unanticipated response at very low apomorphine doses. Vital signs were taken before and several

times after each injection. In addition, written comments from all subjective and objective observations were made continuously by the blinded evaluator monitoring the patient (Fig. 1D). These comments included the timing of onset of the ON state (see following text), the disappearance of tremor, the appearance of dyskinesias, the response duration, and the occurrence of side-effects.

Formal motor ratings, using selected items of the Unified Parkinson's Disease Rating Scale (UPDRS) (Fahn and Elton 1987), were performed to evaluate parkinsonian symptoms. The modified Abnormal Involuntary Movements Scale (AIMS) was used to rate dyskinesias. These ratings were performed before injection and every 10 min after the injection until baseline motor conditions returned (Fig. 1). Figure 1 shows results for preoperative testing in one patient. The selected UPDRS-III items included items 20, 22, 23, 26, 29, and 31, describing tremor, rigidity, fingertaps, leg agility, gait, and bradykinesia on a scale from 0 to 4 with a score of 64 indicating maximum impairment (Verhagen et al. 1998). Dyskinesias were measured in the head and neck, as well as in the four extremities, on a scale from 0 to 4, where 20 was the maximum dyskinesia score (Fig. 1A).

Apomorphine doses were increased by 0.01–0.02 mg/kg until either the maximum allowed dose was reached (6 mg) or side effects occurred. At the end of this phase, an appropriate surgical dose for intra-operative use was selected for each subject by the unblinded safety officer based on the combined criteria of minimal apomorphine-related adverse effects and maximal antiparkinsonian efficacy. The chosen dose was that just adequate to reliably induce dyskinesias and was always  $\geq 0.01$  mg/kg above the lowest effective dose. This "surgical dose" was repeated once, as the first morning dose. Whereas patients were usually injected while sitting in a straight chair to facilitate all aspects of motor ratings, the repeat injection was given

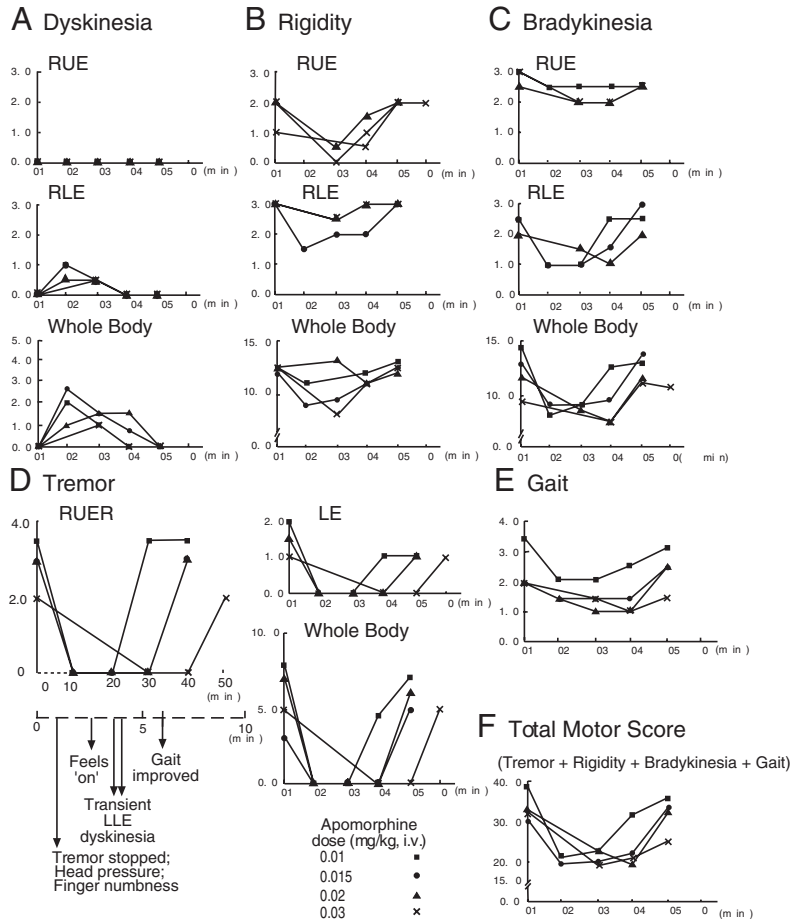


FIG. 1. Clinical assessments during preoperative apomorphine dose-finding in a single patient. Dyskinesias, rigidity, bradykinesia, tremor and gait were scored before and every 10 min after apomorphine injection. A written log was kept of all clinical changes in between rating time points as is indicated in D. This patient had transient dyskinesias when turning ON (D) but not at the formal rating time points (A). Only motor scores from the side opposite to the subsequent pallidotomy are shown here.

with the patient in the supine position to simulate the position in the operating room.

### *Operative and recording procedures*

The pallidotomy was performed as previously described (Lenz 2006). The GPI was explored with a platinum-iridium electrode etched to a tip of 3–4  $\mu\text{m}$  and coated with solder glass to give an impedance of  $\sim 2.5 \text{ M}\Omega$  (fabricated in-house by LH Rowland). The electrode was advanced toward the GPI as localized by preoperative imaging.

The microelectrode signal was amplified (DAM 80, WPI, Sarasota, FL), analog filtered (6 dB below 300 and  $> 10,000 \text{ Hz}$ ), and stored on a video tape recorder (Vetter Model 4000, Petersburg, PA). The striatum, GPe, and GPI were identified by the features of neuronal activity (DeLong 1971; Lozano et al. 1996; Vitek et al. 1998). Responses to sensory stimuli were sought for neurons in the GPe and the GPI during passive movements of upper and lower extremities, both ipsilateral and contralateral to the recording site.

There can be significant difficulties in maintaining single-neuron isolation during lengthy human single-unit recordings including intervals before and after the administration of apomorphine. Therefore small movements of the microelectrode were made throughout the recordings from a single neuron to maintain constant size and shape of the action potential, as in our previous studies (see Figs. 4 and 5) (see also Lee et al. 1999; Lenz et al. 1994; Patel et al. 2006).

The optic tract was identified by light-evoked potentials and microstimulation-evoked phosphenes ( $\leq 40 \mu\text{A}$ ). The internal capsule was identified by microstimulation-evoked muscle twitches. The physiologic map of the locations of the optic tract, internal capsule, and cellular and acellular regions were fitted to the atlas maps to determine the location of the GPI (Lozano et al. 1996; Schaltenbrand and Bailey 1959; Vitek et al. 1998). Lesions were made within the part of the GPI where neurons responded to movements of the extremities. Each thermal lesion was made to  $70^\circ\text{C}$  with the tip of the electrode 2 mm above the optic tract and to  $80^\circ\text{C}$  with the tip 3–4 mm above the optic tract.

### *Intra-operative studies*

In the operating room, the patient's motor status was continuously monitored by the neurologist who had performed the preoperative testing (LVM), and by video recordings of the patient. Prior to apomorphine injection, baseline assessments of rest tremor, rigidity, and finger tapping were performed. Tremor was recorded continuously with electromyography (EMG) from wrist flexors and extensors. The patient was instructed to report the moment of turning ON. We interpreted the intra-operative response in terms of the usual response to apomorphine in that patient.

Once initial landmarks had been identified and an appropriate GPI neuron was isolated, the predetermined surgical dose of apomorphine was injected intravenously. Close observation and frequent motor testing was performed to document the changes in motor behavior. Patients were periodically reminded to report when they turned ON. The onset of the apomorphine-evoked ON state was identified by the improvement in finger tapping, decreased rigidity, and the patient's self-report of feeling ON. Based on these combined assessments, motor behavior was categorized as OFF, ON without dyskinesias, and ON with dyskinesias. Cases in which there was a discrepancy between these assessments of the ON state are described in RESULTS.

### *Postoperative analysis*

Postoperatively, neuronal activity was analyzed for all neurons that were well isolated and that had been studied before and after the apomorphine injection. In the case of firing rates, the first analysis was carried out across 1-min segments. The action potentials of single

neurons were discriminated and digitized at 10 kHz by a standard shape-fitting package (Explorer, Brainwave, Thornton, CO), and their times of occurrence were stored at a clock rate of 10 kHz.

The coefficient of variation is a measure of ISI variance that is calculated as SD/mean of the distribution (Glaser and Ruchkin 1976; Snedecor and Cochran 1967). The coefficient of variation does not measure the effect of order of the ISIs within a spike train. Properties that dependent on ISI order within the spike train include independence of ISIs and stationary firing within a spike train.

Several aspects of random spike train activity were assessed including: the median runs test for stationary spike trains, the serial correlation coefficient for independence of sequential ISIs, and the fit to the Poisson distribution, the most common model of a random spike generator (Fine et al. 2000; Glaser and Ruchkin 1976; Snedecor and Cochran 1967). The use of several measures of random firing increased our ability to interpret models for spike train generators suggested by the data.

Stationary behavior of a spike train variable, such as firing rate, was assessed by a median runs test that measures whether fluctuations in the value of that variable over time are different from that expected at random (Bendat and Piersol 1976). The spike train was divided into 10-s sample intervals, and the firing rate was calculated for each sample interval. A run was defined as a series of contiguous sample intervals with mean firing rates that were either all above or all below the median firing rate. The number of runs in a spike train was compared with that expected for a spike train with the same number of intervals given a 0.5 probability that any interval will have a value greater than the median. If number of runs in the spike train is significantly different from the expected number, then the spike train is nonstationary.

The first serial correlation coefficient is an average measure of the likelihood that adjacent ISIs are jointly greater than or less than the mean ISI. If the coefficient is not significantly different from zero, then adjacent ISIs do not vary jointly with each other (independent ISIs) (Cox and Lewis 1966; Eckmann et al. 1987; Glaser and Ruchkin 1976; Kaluzny and Tarnecki 1993). If the coefficient is significantly different from zero, then adjacent ISIs vary jointly with each other (nonindependent ISIs). Nonindependent ISIs may have a first serial correlation coefficient that is significantly greater than zero, which indicates that adjacent intervals vary directly, i.e., both ISIs are either greater than the mean or less than the mean. Alternately, nonindependent spike trains may have a serial correlation coefficient that is significantly less than zero, which indicates that adjacent intervals vary indirectly with each other, i.e., one ISI greater than the mean and the other less than the mean.

To test for the presence of a Poisson generator, the ISI histogram was converted to a ISI cumulative density function (CDF). Each CDF bin was the sum of all ISIs in histogram bins to the left of the corresponding bin in the histogram. This sum is divided by the total number of ISIs in the spike train. The presence of this Poisson process was tested by fitting the CDF to an ideal Poisson function of the same mean (K-S statistic in Press et al. 1992).

The bursty firing of the spike trains was characterized by a technique assessing numbers of action potentials occurring over fixed intervals (Kaneoke and Vitek 1996). This technique measures the number of spikes occurring in contiguous intervals of duration equal to the inverse of the mean firing rate; a histogram was then constructed of the number of intervals with different numbers of spikes. The fit of this histogram to a Poisson density function was used to identify spike trains with a Poisson (random) distribution.

### *ISI patterns in the spike train*

To test for the presence of patterned firing in the spike trains, we used a recurrence plot approach (Eckmann et al. 1987; Kaluzny and Tarnecki 1993; cf. Dayhoff and Gerstein 1983a,b). The recurrence plot has axes consisting of ordinal numbers of ISIs in the spike train

with the first ISI at the origin (Eckmann et al. 1987; Glaser and Ruchkin 1976; Kaluzny and Tarnecki 1993). For example, both axes in Fig. 8 (*bottom*) represent ordinal numbers of ISIs in the spike train from the 1 to 250. Therefore the plot includes a point for each pair of ISI ordinals in the spike train (Kaluzny and Tarnecki 1993). A dot is placed at the point defined by any two ISIs if the pattern of ISIs is similar for the segments of the spike train starting at these two ISI ordinals (see following text). For example, the left dot in the line indicated by the arrow in the bottom recurrence plot indicates that the ISI pattern beginning at ISI ordinal number 9 (y axis) is similar to that for beginning at ISI 194 (x axis).

The similarity of the two segments is calculated from the sum of absolute distances between the corresponding, sequential ISIs in the two segments divided by the sum of the durations of ISIs in the first segment. For example, the three corresponding, sequential ISI pairs for the line at the arrow in the bottom recurrence plot of Fig. 8 are (9,194), (10,195), and (11,196). The two segments are considered to be similar if this value is less than the threshold value,  $r$ ; the number of ISIs in the segment is indicated by  $d$  (Eckmann et al. 1987; Glaser and Ruchkin 1976; Kaluzny and Tarnecki 1993). The threshold values of  $r < 0.3$  and  $d = 4$  were selected as in prior studies (Dayhoff and Gerstein 1983a,b; Eckmann et al. 1987; Kaluzny and Tarnecki 1993). The main diagonal is always completely dotted because each segment is identical to itself.

Two similar segments will result in a line of dots parallel to the main diagonal (arrows in Fig. 8, *left*) because similarity will be found for pairs of corresponding sequential ordinals within that pair of segments. The number of such lines indicates the number of similar patterns in the spike train, and the number of points in the line indicates the number of similar ISIs in a sequence. However, such sequences may occur at random in any spike train. Therefore we shuffled the data to test whether the number of recurrent patterns was significantly greater than that expected at random for the set of ISIs in the spike train.

The data are shuffled by proceeding through the spike train interval by interval and swapping pairs of neighboring intervals with a probability of 0.5 (Kaluzny and Tarnecki 1993). This process was repeated five times to generate one shuffled spike train and the associated recurrence plot. One hundred such plots of shuffled spike trains were analyzed to determine the mean and 95% confidence interval (CI) of the number of lines expected from the spike train at random. This approach leads to an estimate of variability which is not dependent on the assumption of any distribution for the number of lines. In Fig. 8 (*right*), the number of lines in the unshuffled and shuffled data are shown as a function of line length. Points in the plot of shuffled data are indicated by the presence of error bars (mean  $\pm$  95% confidence interval). The number of lines occurring in 95% of shuffles of the spike train is a direct measure of the number of lines expected at random given the ISIs in the spike train. If the error bars of the shuffled spike train do not overlap the unshuffled spike train, then there are significant recurrent patterns in the unshuffled spike train to a certainty of  $P < 0.05$ .

### Cross-correlation analysis

For all neurons, simultaneous EMG signals for flexors and extensors of the wrist and extensors of the elbow were digitized. Standard techniques were used to process these two signals and to compute their raw spectral estimate (Bendat and Piersol 1976; Glaser and Ruchkin 1976; Lenz et al. 1988). These raw spectral estimates were averaged across raw frequency estimates to produce the autopower and cross-power. The autopower of a signal is the square of the magnitude of the signal as a function of frequency. The cross-spectrum of the two signals is composed of the magnitude (cross-power spectrum) and phase spectrum. The square of the cross-power is often divided by product of the averages of the two autospectra to produce the coherence. The coherence is used to estimate the proba-

bility that the two signals are linearly related, which is to say that one signal could be described as a linear function of the other (Bendat and Piersol 1976). GPi activity might be related to dyskinetic movements by a model involving sensory transmission to GPi, motor output from GPi or sensorimotor integration, as in a feedback control system or network (Churchland and Sejnowski 1992). A detailed treatment of these possibilities is beyond the scope of this study.

### Statistical analysis

Student's  $t$ -test or ANOVA with post hoc tests with corrections for multiple comparisons (Tukey honestly significant difference, HSD) were used to compare normally distributed parametric data. Parametric data that were not normally distributed were tested by Mann-Whitney or Kruskal-Wallis. Nonparametric data were tested with a Fisher or  $\chi^2$  test, as appropriate. For all tests, the null hypothesis was rejected for  $P < 0.05$  (Snedecor and Cochran 1967). Statistical analysis of multiple periods within the spike train of a single neuron is described in the results section dealing with that data (*Transient periods of ON with or without dyskinesias*).

## RESULTS

Under this protocol, 28 patients received apomorphine preoperatively and 17 patients received apomorphine intra-operatively as summarized in Table 1. These patients had Parkinson's disease with long duration (6–25 yr), with midline motor symptoms indicating advanced disease (Hoehn and Yahr stage 3 or greater), and with drug-related dyskinesias (Paulson and Stern 1997). GPi neurons were not recorded in the other 11 patients during surgery because of a preoperative response characterized by either limited efficacy of apomorphine ( $n = 4$ ), violent dyskinesias ( $n = 1$ ), or because of the inability to "hold" GPi neurons during microelectrode recording ( $n = 2$ ) or other practical reasons ( $n = 4$ ).

The surgical apomorphine dose for the 17 patients ranged from 0.015 to 0.06 mg/kg [ $0.044 \pm 0.022$  (SD) mg/kg]. At this individualized dose, the mean improvement in preoperative UPDRS motor score was  $-58 \pm 16\%$  (range:  $-35$  to  $-91\%$ ) (Table 1). All 17 patients reported they were ON after the selected dose, and all but 3 developed dyskinesias. This dose was not associated with clinically relevant adverse events, such as nausea or lightheadedness. Mild paraesthesias and yawning frequently preceded the benefit, providing additional markers for the imminent onset of dopaminergic motor effects. As anticipated, apomorphine injection induced a characteristic clinical response in each individual subject.

In the operating room, all 17 patients received their surgical dose of apomorphine. Four of 17 patients received a second apomorphine injection during recording from a second neuron  $>30$  min after the first injection and after the first injection had worn off. In total, the spike trains of 21 neurons, (17 patients, 20 trajectories) were studied before and after apomorphine injections. In these neurons, GPi neuronal activity was studied before (10–30 s) and after the administration of apomorphine ( $362 \pm 192$  s).

### GPi neuronal properties: location and firing rates

The location of all 21 neurons was within the sensorimotor part of GPi as inferred from the physiologic mapping on a standard atlas map (Schaltenbrand and Bailey 1959) (*Operative procedures*). All neurons were within the borders of GPi in

TABLE 1. Patient Characteristics

Subject	Age, yr	Sex	Disease Duration, yr	H & Y (Off)	Levodopa Equivalent Dose, mg/d	Pre-Operative Apomorphine Doses (n)	Apomorphine Dose Selected for Surgery, mg/kg	Pre-Operative Improvement With Selected Dose, %
1	58	F	23	4	1250	7	0.06	43
2	65	M	14	4	650	5	0.015	35
3	52	M	17	3	650	6	0.03	62
4	66	M	6	5	1550	7	0.1	91
5	57	M	15	5	900	8	0.01	80
6	57	M	6	3	1000	7	0.04	37
7	77	M	20	4	1025	7	0.04	76
8	70	M	18	4	900	6	0.03	50
9	66	F	15	4	900	6	0.02	57
10	66	M	10	3	600	4	0.04	72
11	57	M	24	5	750	6	0.06	72
12	63	F	9	3	1150	5	0.06	61
13	43	M	10	5	3150	7	0.04	39
14	69	M	28	4	675	5	0.04	52
15	63	M	7	3	2200	6	0.06	41
16	62	M	12	4	2400	5	0.05	52
17	66	F	21	4	825	7	0.06	61
Mean $\pm$ SD	62 $\pm$ 7.8		15 $\pm$ 6.7	3.9 $\pm$ 0.75	1210 $\pm$ 719.9	6 $\pm$ 1.05	0.04 $\pm$ 0.022	58 $\pm$ 16.4

Pre-operative improvement with the selected apomorphine dose was measured by using elements of the motor subscale of the Unified Parkinson's Disease Rating Scale. The levodopa equivalent dose was calculated as described in studies of pallidotomy (Fine et al. 2000).

the 21 mm lateral plane of the atlas map (see Fig. 1 in Lemstra et al. 1999). These neurons were all within the sensorimotor portion of GPi as demonstrated by study the neuronal RFs before administration of apomorphine. These neurons had either RFs (16 neurons, see RESULTS) or had no RF themselves but were adjacent to neurons that did have RFs. RFs were not studied after the apomorphine injection in order to record as much of the ongoing neuronal activity as possible without the artifacts produced by the bilateral sensory examination.

Prior to the apomorphine injection, all patients were in their typical OFF state. Intravenous apomorphine administration led to a decrease in firing rate from baseline and a change in the subjects' clinical condition. For each neuron, mean firing rates of recorded neurons were measured in 1-min periods. Analysis of average firing rates across these 21 neurons revealed significantly higher rates immediately prior to injection ( $77.2 \pm 6.6/s$ ) than at the firing rate nadir after the injection ( $36.5 \pm 5.5/s$ ,  $P = 0.0002$ ,  $t$ -test). When normalized to the firing rate during the 1-min prior to the injection, the fall in the averaged firing rate occurred 4 min after the apomorphine injection.

Analysis within individual neurons demonstrated a significant decrease in firing rate ( $P < 0.05$ ) from the baseline to the nadir after apomorphine administration in 15 of 21 GPi neurons (maximum decrease: 19–100% of baseline, mean  $66 \pm 25\%$ ). The 15 neurons with a fall in firing rate included 9 with simultaneous ON and dyskinesias and 6 in neurons for which the onset of dyskinesias followed ON (see following text, Results: *Relationship of dyskinesias to GPi* . . .). The initial decrease in firing rate started as early as the first minute after the apomorphine injection (Fig. 2A). The nadir in firing rate preceded or was simultaneous with the onset of dyskinesias in all 15 of these neurons, suggesting that a fall in firing rate is a necessary condition for the occurrence of dyskinesias.

In the other 6 of 21 injections, apomorphine was not effective either in turning the patient ON or in leading to a decrease in firing rate. In four of these six injections, the ON state was not achieved either subjectively or objectively. During these four injections, two of the neurons recorded showed a nonsignifi-

cant decrease in the firing rate (77.8–47; 38.6–25/s), one showed a significant decrease in firing rate ( $70.1 \pm 5.7$  to  $57 \pm 6.1$ ,  $P = 0.0139$ ,  $t$ -test, *neuron 20251*), and one showed an increase in the firing rate (56.7–96). In the two remaining injections of six, the patients turned ON without dyskinesias and showed small increases in their finger tapping (15–25 and 15–20/min; see following text). These two injections did not produce decreases in firing rates (50.7–47 and 56.5–92/s). Apomorphine had produced the ON state and dyskinesias in all six patients during preoperative screening.

#### Relationship of dyskinesias to GPi neuronal spike trains

Among the 15 apomorphine injections which turned patients ON with dyskinesias finger tapping rates increased significantly

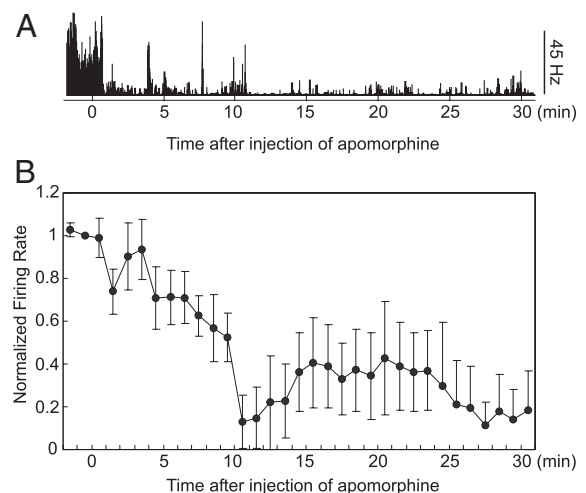


FIG. 2. Globus pallidus interna (GPi) neuronal firing rates relative to apomorphine injection. *A*: effect of apomorphine injection (at *time 0*) on firing rates in an individual GPi neuron, showing a decrease in firing rate within 30 s. The time scale is as indicated. The vertical calibration bar indicates the firing rate. *B*: effect of apomorphine injection on the average firing rate ( $\pm$ SD) for the group of 21 neurons normalized to the average baseline firing rate for each neuron.

from  $14.3 \pm 1.3$  to  $26.3 \pm 2.1$ /min (paired *t*-test,  $P < 0.0001$ ). The latency from injection to ON with dyskinesias was  $127 \pm 143$  s. The firing rate is shown as a function of time after apomorphine injection for an individual neuron in Fig. 2A. Figure 2B shows the results for all 15 neurons with firing rates normalized to 2 min before the injection. Firing rates decreased significantly from the baseline firing rate ( $84.8 \pm 8.2$ ,  $P = 0.002$ , *t*-test) to immediately after the onset of ON ( $49.9 \pm 6.4$ /s). This figure demonstrates a dramatic fall in mean normalized firing rate from the baseline to the nadir.

Figure 3A shows the neuronal firing rates of the nine neurons recorded after injections after which patients simultaneously turned ON and developed dyskinesias. This plot shows average firing rates plotted relative to the onset of ON with dyskinesias and demonstrates fall in firing rate prior to the gradual fall in firing rate that occurs around the onset of ON with dyskinesias. For these nine neurons as a group, the fall in firing rate between 15 s prior to the initial fall in firing rate versus 15 s after the initial fall was greater ( $38 \pm 25$ /s,  $P < 0.05$ , paired *t*-test) than

that around the time of ON with dyskinesias ( $12 \pm 13$ /s, mean difference per neuron:  $23.9 \pm 27.2$ /s). There was a significant fall in firing rate around the time of onset of ON with dyskinesias on within neuron analysis of all nine neurons. The possibility that this fall was due to the onset of ON, or dyskinesias, or both was studied in the 6 of 15 injections for which ON preceded dyskinesias.

Among these six neurons, the onset of ON could not be determined exactly in one neuron. In another two, the interval between ON and appearance of dyskinesias was too short for analysis. Among the remaining three, the firing rate of one dropped considerably around the onset of ON (Fig. 3B) but did not change from before versus after the onset of dyskinesias, (21.1 vs. 20.0/s; Fig. 3C). In the case of another neuron, the patient's self report of ON, the resolution of tremor, and faster finger tapping occurred 190 s after apomorphine injection (Fig. 4B, arrow B). The patient developed dyskinesias 80 s later, long before the significant change in firing rate for this neuron (Fig. 4B, arrow C).

A third injection demonstrated end-of-dose dyskinesias both preoperatively (Fig. 5A, 45 min) and intra-operatively (Fig. 5B, arrow C). During the preoperative testing, this patient exhibited brief (20 s) beginning-of-dose dyskinesias and longer (5 min, Fig. 5A) end-of-dose dyskinesias. Intra-operatively, there were only end-of-dose dyskinesias that began while firing rates were low (Fig. 5B, arrow C). Firing rates before and immediately after the onset of dyskinesias initially were similar (0.1 vs. 0.2/s). However,  $\sim 3$  min after the onset of dyskinesias (Fig. 5B, arrow C), the firing rates increased significantly while dyskinesias continued until the patient turned off (Fig. 5B, arrow D). This neuron demonstrates that end-of-dose dyskinesias can occur as firing rates increase from ON to OFF. In summary, the onset of ON was associated with a fall in firing rate in two of three neurons, whereas there was no change in firing rates at the onset of dyskinesias in three of three neurons.

#### Transient periods of ON with or without dyskinesias

The relationship between neuronal firing rates and motor behavior is also demonstrated by analysis of OFF, transient ON periods with dyskinesias, and transient ON periods without dyskinesias. These transient periods resulted from the low doses of apomorphine used in this study as indicated by the 6/21 intraoperative injections did not turn the patients ON. At these doses dyskinesias were mild and were often discontinuous over the interval of ON with dyskinesias. To examine properties of spike trains related to dyskinesias at these doses, we next examined changes in spike trains in transient ON periods with dyskinesias versus those ON without dyskinesias and OFF (for example, see Levy et al. 2001).

The progression of dyskinesias over these intervals was studied by examination of the intra-operative EMG, video, and audio recordings. An example of a patient with transient periods of dyskinesias during the intervals of ON without or with dyskinesias is shown by the thin line under the EMG recordings in Fig. 6A (neuron 266025). In A, transient periods of dyskinesias began  $\sim 1.5$  min from the beginning of the panel and ended  $\sim 3$  min later. The second period of dyskinesias started at 7 min prior to the end of the panel. These transient periods were analyzed if they had a duration of  $> 1$  min. Gaps

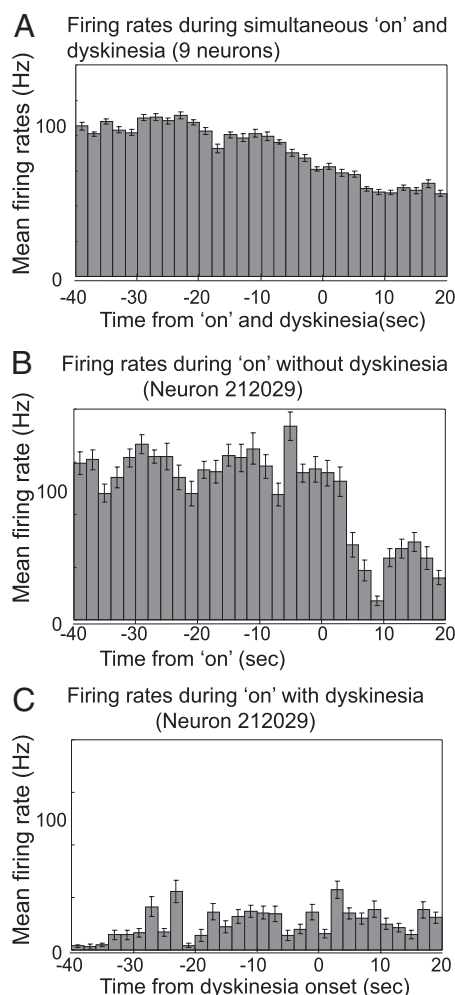


FIG. 3. GPI neuronal firing rates before and after apomorphine injection. A: average GPI neuronal firing rates relative to onset of ON for injections ( $n = 9$ ) during which ON and dyskinesias were simultaneous. B: firing rates before and after the onset of ON for an individual patient in whom the onset of ON preceded the onset of dyskinesias. The firing rate drops immediately after the onset of ON. C: firing rates around the onset of dyskinesias in the same patient as B. No decrease in firing rate is seen around the onset of dyskinesias. B and C: firing rates are averaged across 20 100-ms bins. Error bars  $\pm$  SD.

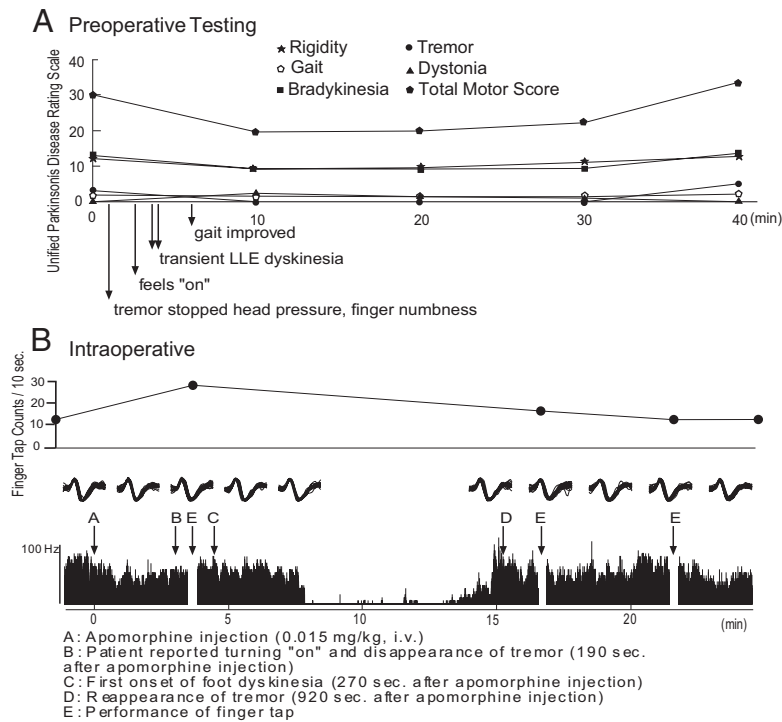


FIG. 4. Motor scores and GPI neuronal recordings following apomorphine injection in a patient in whom the onset of ON preceded that of dyskinesias. *A*: clinical events during the preoperative apomorphine injections are indicated as a function of time in the plot. The labels below this plot indicate the events during the first 10 min as the patient turned on. *B*: number of finger taps/10 s (*top*) and GPI neuronal firing in the same patient after apomorphine injection (point A). The same neuron was held throughout the recording as indicated by the constant shape of the action potential as shown above the *bottom panel*. In the operating room, it took 190 s before the tremor stopped and the patient felt ON (point B), slightly longer than during preoperative testing. As was the case preoperatively, this was shortly followed by the occurrence of dyskinesias (point C). No reduction in firing rate was seen until ~4 min later. After ~13 min the firing rate began to increase and the tremor reappeared (point D).

in the record correspond to measurement intervals for finger tapping as in Figs. 4 and 5.

The spike trains for the 15 neurons were studied during injections that clearly turned patients ON with dyskinesias. Additionally, we studied two neurons which clearly changed from OFF to ON without dyskinesias (see preceding text, *GPI neuronal* . . .). Among these neurons, we identified 17 transient periods of ON without dyskinesias and 25 transient periods of ON with dyskinesias (Fig. 6).

The variability within spike trains for a single neuron in one motor state is indicated (see following text) by the large numbers of nonstationary spike trains measured across different periods of one type and by differences in binary variables between different epochs of one type in one neuron. To address this variability, multiple periods within the spike train of a single neuron were classified into periods of OFF, ON with dyskinesias, and ON without dyskinesias (Levy et al. 2001). For each period, continuous variables (including average firing

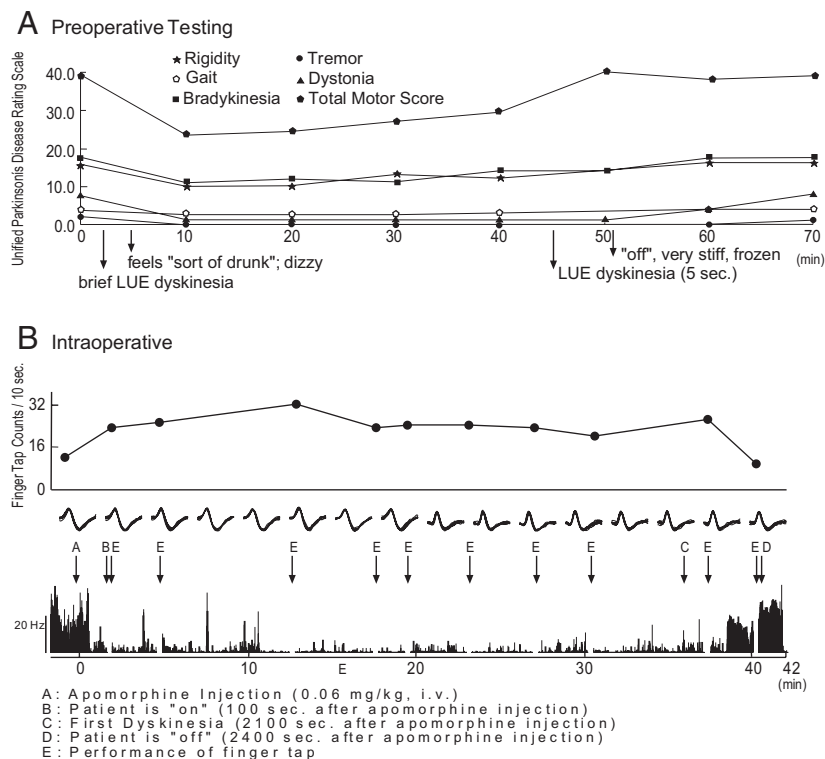


FIG. 5. Motor scores and GPI neuronal recordings following apomorphine injection in a patient in whom onset of ON preceded that of the dyskinesias which occurred at the end of the dosage interval. Conventions as in legend for Fig. 4.



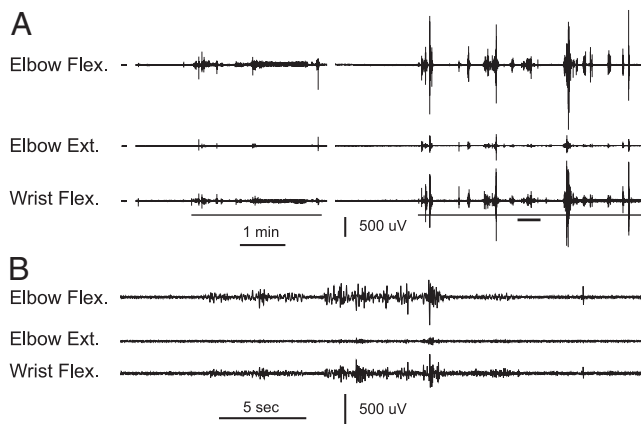


FIG. 6. EMG recordings over a 12-min period of ON time including ON without dyskinesias and ON with dyskinesias. *Bottom trace*: expended time scale image of the interval indicated by the line below wrist flexor in A. See text.

rates, etc.) and binary variables were calculated. To include variability between and within neurons, we used linear and logistic regression models with robust variance estimates (LRM) (Dingle et al. 2002; Statacorp 2005). This technique was used to compare the means of the parametric variables and the odds of the binary variables across the three periods. The robust variance estimates appropriately account for correlation across periods within a single neuron. For a binary variable, such as stationary firing during ON versus OFF the odds ratio (OR) is a measure of the relative likelihood of stationary firing during ON versus OFF. In this example, the OR was calculated as the proportion with stationary / nonstationary for ON divided by that for OFF.

Firing rates were higher in OFF (76/s, LRM, Table 2) versus both transient periods without dyskinesias (53/s) and transient periods with dyskinesias (58/s). Differences between transient periods without dyskinesias versus transient periods with dyskinesias were not significant as reported in previous studies measuring transient periods (Levy et al. 2001).

#### Measures of random spike trains: stationary firing rates, ISI independence, and Poisson distributions

As measured by a median runs test, firing rates were stationary during OFF periods (9/17) less often versus transient periods with dyskinesias (19/25, LRM see Table 2) but not versus transient periods without dyskinesias (14/17). Stationary spike trains were not different between ON periods without

or with dyskinesias. Spike trains can be nonstationary as a result of either more runs or fewer runs than expected at random. Nonstationary spike trains had fewer runs than expected at random for OFF intervals (8/8), transient periods without dyskinesias (2/3), and transient periods with dyskinesias (4/6). Differences between these proportions were not significant (OR < 1.5,  $P > 0.16$ , LRM). Therefore spike trains during dyskinesias were commonly stationary. When they were nonstationary these spike trains had longer runs above or below the median than expected at random.

The periods described by independent ISIs (serial correlation coefficient) were less common among neurons/periods during OFF (5/17) versus during transient periods with dyskinesias (16/25, Table 2) but not versus during transient periods without dyskinesias (11/17). When nonindependent, ISIs were less likely to be related indirectly during OFF (0/17) than during ON with (6/17) or without dyskinesias (6/25); this indicates that during ON long ISIs were likely to be followed by short and vice versa.

Poisson distributions described the firing rate less commonly than expected at chance ( $P < 0.01$ , binomial) during the OFF periods (0/17), the periods of ON without dyskinesia (1/17), and the transient periods with dyskinesia (1/25). The early latency cumulative distribution functions (Fig. 7C, lowest row) were shifted to the right so that the shortest ISIs were longer than that expected by a Poisson distribution. This pattern was seen to a greater extent than expected by chance ( $P = 0.01$ , binomial) during OFF intervals (17/17), ON without dyskinesia intervals (16/17), and ON with dyskinesia intervals (24/25).

Therefore spike trains were often random as measured by stationary firing rates and ISI independence, during OFF, and ON with or without dyskinesias. However, the data were infrequently consistent with the Poisson model, which is the most common model of random process. Therefore we examined other variables bearing on the nature of the spike train generator.

The coefficient of variation (CV, standard deviation/mean) was computed as a basic measure of ISI variance. The CV of OFF intervals (0.83, Table 2) was significantly less versus that of ON with dyskinesias (1.13) but not versus transient periods without dyskinesias (0.98). Differences between ON periods with and without dyskinesias were not significant.

#### Models of generators of the spike train

The autocorrelogram can be used to characterize processes generating spike trains (Glaser and Ruchkin 1976; Perkel et al.

TABLE 2. Results of statistical testing of periods of OFF and ON with and without dyskinesias by linear and logistic regression models with robust variance estimates (LRM)

Outcome	ON vs. OFF			Dyskinesia vs. OFF			Dyskinesia vs. ON			Overall $P$
	Diff/OR	Variability	$P$	Diff/OR	Variability	$P$	Diff/OR	Variability	$P$	
Firing rate	-23.7	8.6	0.007	-19.1	7.9	0.050	-4.6	7.9	0.377	0.021
CV	0.15	0.07	0.041	0.29	0.09	0.006	-0.14		0.232	0.006
Stationary firing	4.15	3.63	0.104	2.81	1.17	0.013	1.47	1.09	0.602	0.042
ISI independence	4.40	3.44	0.058	4.27	2.69	0.022	1.03	0.78	0.968	0.044

ON, ON without dyskinesias. Diff/OR, Diff used for firing rate and CV, OR (odds ratio) used for stationary firing and interspike interval (ISI) independence. For ON versus OFF, Diff refers to the mean for ON periods minus the mean for OFF and OR for stationary firing refers to the likelihood of stationary firing during ON relative to that for OFF periods.  $P$  is the  $P$  value as in descriptions of the logistic regression model (LRM) test (Dingle et al. 2002; Stata Corp 2005).

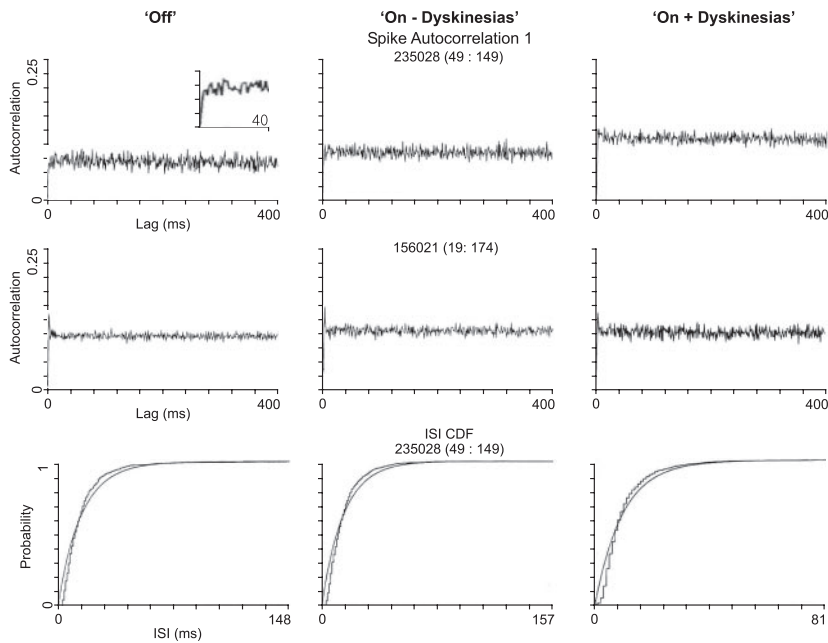


FIG. 7. Examples of autocorrelograms for the firing of neurons in GPI. *Top*: example of a neuron with autocorrelograms having constant postspike firing probability during OFF, ON – dyskinesias, or ON + dyskinesias. *Middle*: neuron having a brief postspike increase in firing probability. *Bottom*: interspike interval (ISI) cumulative distribution for this data (stepwise plot) and for a Poisson process of the same mean as the present data (smooth plot). The data distribution is right shifted with respect to the Poisson of the same mean. The postspike silent period is seen at the left end of the ISI globus pallidus interna (CDF) plots (*bottom row*) and in the *inset* in the *top left panel*, which is a large-scale version of the left end of the same ISI histogram.

1967). Across neurons and periods, all autocorrelograms demonstrated an initial postspike refractory period which is seen at the far left of each autocorrelogram [Fig. 7, *top* and *middle rows*, consistent with Nambu and Llinas (1994)]. This effect is seen in the *inset* (Fig. 7, *top row, left*), which shows the postspike inhibitory peak at an expanded scale. The postspike inhibition is also indicated by the frequent occurrence of a right shift of the short latency ISI CDF distribution (stepped trace, Fig. 7, *bottom*) relative to the Poisson distribution (smooth).

At longer lags, the autocorrelograms were remarkably flat (Fig. 7, *top*) for the OFF intervals (13/17), the transient periods without dyskinesias (13/17), and the transient periods with dyskinesias (22/25). The flat autocorrelation functions indicate ISI durations are evenly distributed which is consistent with the high ISI variance of these spike trains. These functions are also consistent with a model in which the spike generator is independent of feedback or feedforward spike-related processes (Perkel et al. 1967).

Short-latency peaks after the initial inhibition occurred in the autocorrelograms for all three periods for two neurons (156021, 212029), i.e., OFF and transient periods with and without dyskinesias. This peak is seen at the far left of the autocorrelograms in Fig. 7, *middle row* (neuron 156021). These peaks had maxima of less than twice the mean firing probability at latencies of 5–0 ms and durations of <10 ms at half-maximum amplitude. A third neuron (22028) had such a short-latency peak during the ON with dyskinesias interval only. The autocorrelograms with a short latency peaks may be the result of the shadowing effect by which a neuronal refractory period is followed by a short peak (Bar-Gad et al. 2001).

We next examined the possibility that a bursting generator explained firing of the GPI neurons. By the Kaneoke and Vitek method of burst detection, three neurons were found to have bursty firing (Kaneoke and Vitek 1996): neuron 147062 during OFF, 217041 during ON without dyskinesias, and 235028 during ON with dyskinesias.

We next examined recurrence plots to determine whether the data were consistent with a generator producing patterned

firing (Eckmann et al. 1987; Kaluzny and Tarnecki 1993). The recurrence plots in Fig. 8 (*left*) show short lines parallel to the diagonal (see arrows and *Recurrence plots*); this indicates recurrent spike train ISI patterns. Recurrent patterns (Fig. 8, *right*) were significantly greater than random when the raw (unshuffled) data were above the 95% confidence interval of the shuffled data (error bars). In the case of four neurons, there were significant recurrent patterns as illustrated in Fig. 8 (*right*) in the longest transient period of ON with dyskinesias recorded for that neuron (neurons 156021, 212029, 147062, 178030) but not in periods of OFF or ON without dyskinesias (Fig. 8, *right, top 2 panels*). The spike trains of these four neurons during ON with dyskinesias demonstrated ISI dependence in 3/4 neurons and with a non-Poisson distribution (4/4 neurons). The presence of significant recurrent patterns with nonindependence of ISIs and a non-Poisson distribution strongly suggest that these spike trains were the result of a pattern generator.

Significant recurrent patterns occurred only during transient periods of ON with dyskinesias. This consistency of spike trains with recurrent patterns is significantly different from our expectation that these spike trains will occur equally in all three types of intervals, i.e., OFF, transient periods of ON with and without dyskinesias ( $P = 0.01$ , combinatorial analysis) (Snedecor and Cochran 1967). This analysis suggests that spike trains of a subpopulation of neurons during dyskinesias result from a generator that produces patterns of ISI intervals (Dayhoff and Gerstein 1983b; Kaluzny and Tarnecki 1993).

#### RF-related neuronal activity

Among the 16 neurons with RFs, the location of the RF was on the upper extremity (8 neurons) or lower extremity RFs (5 neurons) or both (Baron et al. 2000). Five neurons had no RFs. None of the binary and parametric variables described in the preceding text were significantly different between neurons with RFs versus neurons without RFs. RF examinations were not carried out after apomorphine injection to minimize interference with recordings of ongoing neuronal activity.

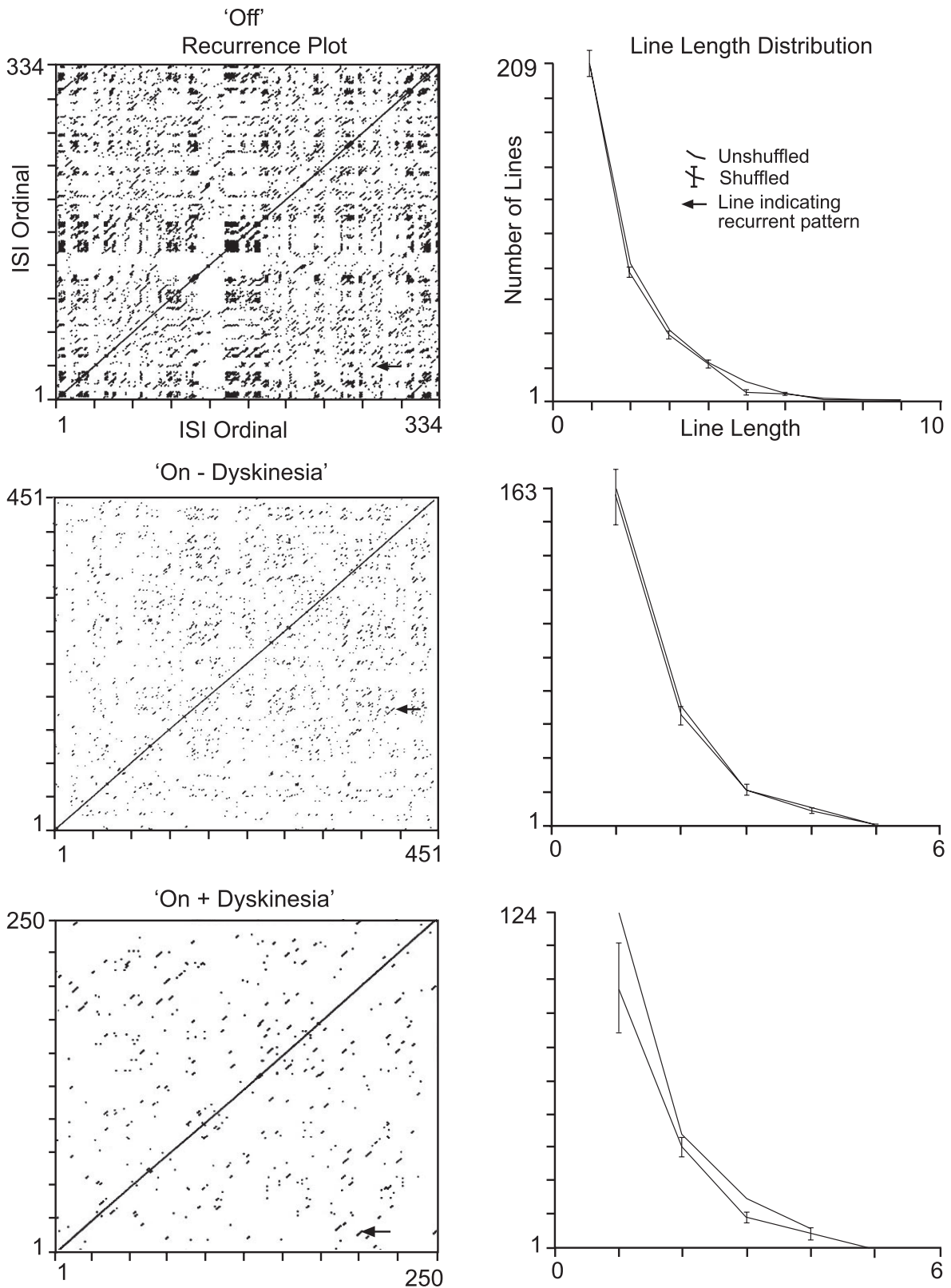


FIG. 8. Recurrence plots and line length distributions for the spike train of a neuron (178030) during OFF, transient periods without and with dyskinesias. See text.

#### DISCUSSION

The results of this study support our hypothesis that GPi neuronal activity is related to the mild transient dyskinesias

that result from low doses of apomorphine in patients with advanced PD. In most patients, ON and dyskinesias began simultaneously and were associated with a significant fall in

firing rates. In some patients, the onset of dyskinesias followed the onset of ON and the firing rate did not fall at the onset of dyskinesias. The firing rates during the OFF state were higher than firing rates during transient periods of ON, either without or with dyskinesias. These results suggest that a fall in firing rate is associated with the onset of ON but not the onset of dyskinesias. Transient periods of ON with dyskinesias were characterized by stationary firing rates, independent ISIs, and greater ISI variability. Therefore decreases in firing rate associated with the ON state are combined with changes in the firing pattern that may produce transient ON periods with dyskinesias (Suarez et al. 1997; Vitek et al. 1999).

#### Methodological considerations

The effects of injection of the surgical apomorphine dose were measured preoperatively for each patient (Corsini et al. 1979). This approach not only provided an optimal individual apomorphine dose, but it allowed us to predict the intraoperative response. The increased degree of certainty about the clinical effects was useful in the interpretation of less common responses, such as end-of-dose dyskinesias (Fig. 5B) (Marsden et al. 1982).

The total number of neurons recorded in this study was limited because we studied the activity of single neurons during both OFF and ON periods. To our knowledge, five human GPi neurons have previously been studied in both these periods. This approach substantially reduces the variability associated with study designs in which different groups of neurons form the populations for each period OFF, and ON without or with dyskinesias.

The variability of the data was also decreased by use of a model that accounted for differences within neurons during periods of OFF, and ON with or without dyskinesias. "Intermittent" periods with apomorphine-induced dyskinesias have previously been studied during ON periods without dyskinesias (Levy et al. 2001). We have now approached this data with a statistical model that appropriately accounts for correlation across periods within a single neuron, i.e., linear and logistic regression models with robust variance estimates (see *Transient periods of ON with or without dyskinesias*).

Previous studies of spike train firing patterns during drug-related dyskinesias in MPTP monkeys have reported decreased random firing patterns by a global measure of random firing (Boraud et al. 2001, 2002). Many different properties of a spike train can be characterized as random firing (Cox and Lewis 1966). To facilitate comparison with the recurrence plot analysis, we have measured three of these features including stationary firing in the spike train, ISI independence, and the presence of a Poisson process—the most common model of a random process.

In the present results, the independence of ISIs and the stationary spike trains during dyskinesias suggest that a global measure of random firing might be increased after apomorphine, contrary to the results with the global measure used in MPTP monkeys (Boraud et al. 2001, 2002). However, the number of neurons with Poisson firing distributions was unchanged, suggesting that random firing was unchanged. During periods of ON with dyskinesias the neurons with recurrent firing patterns were characterized by nonindependent ISIs and by a non-Poisson distribution. These characteristics are consistent with nonrandom patterning of the spike train but are unlike the majority of our results during dyskinesias. Therefore the present results suggest a complex interaction between random properties of the spike train and dyskinesias.

#### Apomorphine, GPi activity, and dyskinesias

Our findings at the transition from OFF to ON are compatible to those in previous studies. Prior to apomorphine administration the neuronal firing rate in GPi during the OFF state was 77/s, comparable to neuronal activity in patients with PD reported by others (Favre et al. 1999; Hutchison et al. 1994; Sterio et al. 1994) (Table 3), and in MPTP monkeys (Boraud et al. 1998; Filion and Tremblay 1991) (Table 4). In the MPTP monkeys, firing rates of GPi neurons were higher than those in normal monkey by an average of 20–30% (Levy et al. 2001).

In contrast to most human studies, the present investigation utilized intravenous apomorphine at doses just adequate to induce dyskinesias (Table 3). This strategy led to 15 injections that produced ON with dyskinesias and 6 injections (6/21) that did not produce ON or dyskinesias. This strategy also produced

TABLE 3. Overview of GPi firing rates in patients with PD

Reference (Year)	Subjects (n)	GPi Firing Rate			
		Intact state	Parkinsonian State	ON after Apo	ON with Dyskinesias
Hutchinson (1997)	PD patients (14)	N/A	85 ± 19 (Population study) (75) 85 (Time course study) (10)	34 ± 22 (18) 49 (10)	3 subjects developed dyskinesias; rates N/A
Levy et al. (2001)	PD patients (15)	N/A	72 ± 2.9 (population study) (93)	39 ± 2.5 (51)	40.4 ± 5 (n = 30), ~67 μg/kg sc. Rate during on-dyskinesias = on + intermittent dyskinesias
Merello et al. (1999b)	PD patients (1)	N/A	75.8 ± 15.1 (1)	41.7 ± 9.3	41.7 (probably dyskinesic)
Merello et al. (1999a)	PD patients (5)	N/A	143.8 ± 55.6 (5)	54.2 ± 19.2	54.2 ± 19.2 ~43 μg/kg sc
Stefani et al. (1999)	PD patients (3)	N/A	65.3 ± 12.2 (3)	15.1 ± 5.6 (3)	N/A (nondyskinetic dose)
Stefani et al. (1997)	PD patients (2)	N/A	45 ± 9.3 47 (1)	20 N/A	N/A (nondyskinetic dose) 0 (n = 1). (dyskinetic dose) (no temporal relation of drop in firing rate to onset of dyskinesias)
Present study	PD patients (17)	N/A	77.2 ± 6.6 (21)	N/A	36.5 ± 5.5 (40 ± 22 μg/kg dyskinesic dose)

Values are means ± SD in s<sup>-1</sup>. Parentheses enclose number of patients for the second, fourth, and fifth columns. ~, dose/kg estimated from dose assuming a 70-kg man; PD, Parkinson's disease; GPi, globus pallidus internus; apo, apomorphine; sc, subcutaneous administration.

TABLE 4. Overview of GPi firing rates in non-human primates

Name (Year)	Subjects	GPi Firing Rate, s <sup>-1</sup>			
		Intact State	Parkinsonian State	ON after Levodopa/Apo	ON with Dyskinesias
Nini et al. (1995)	African green monkey (3)	53 (175)	76 (154) MPTP	N/A	N/A (No dopaminergic agent given)
Miller and DeLong (1987)	<i>Macaca mulatta</i> (1)	79.5 (51)	91.5 (79) MPTP	N/A	N/A (No dopaminergic agent given)
Filion and Tremblay (1991)	<i>M. fascicularis</i> (5)	78 ± 26 (105)	95 ± 32 (166) MPTP	N/A	N/A (No dopaminergic agent given)
Boraud et al. (1998)	<i>M. mulatta</i> (2)	80.5 ± 20.7 (50)	106 ± 29.4, 42, MPTP on non-dyskinesogenic oral levodopa 10 mg/kg	47.4 ± 23.0 (78)	N/A
Filion et al. (1991)	<i>M. fascicularis</i> (1)	apo ~20 µg/kg no behavioral change, 200µg/kg chewing & agitation, no "change in pattern or rate" 3	N/A	N/A	58% reduction (18), apo ~20–40 µg/kg (induction of associated with only a partial decrease of activity in GPi, not with complete silence)
Pappa et al. (1999)	<i>M. mulatta</i> (4)	N/A	46.3 ± 8.1, (14) MPTP & dyskinesogenic oral levodopa 1.25–2 gm/day	26 ± 3.1	7.6 ± 1.5 additional decline when dyskinesias started in response to sc levodopa 125–200 mg on a day when oral levodopa withdrawn.
			46 ± 8.2, (10), as above.	27 ± 3.4	N/A (nondyskinetic dose). Low dose sc levodopa 50–100 mg.
Boraud et al. (2001)	<i>M. fascicularis</i> (3)	63–67 bursty (16) to 40–53 regular post apo	80–83 (27)	19–30 (7) apo dyskinesias (0.1 mg/kg)	"Significant modification . . . of the firing pattern (increase in the number of random neurons)". (7)
Present study	PD patients (17)	N/A	77.2 ± 6.6 (time course study) (21)	N/A	36.5 ± 5.5 (40 ± 22 µg/kg dyskinesic dose)

Parentheses enclose *n* values. MPTP, 1-methyl-4-phenyl-1,2,3,6-tetrahydropyridine. ~, estimated dose/weight for monkeys.

a decrease in GPi firing rate in our dyskinetic PD patients that was smaller than those in a study of PD patients receiving nondyskinetic subcutaneous doses of apomorphine [see Table 3 (Levy et al. 2001)]. This strategy produced a decrease in GPi firing rate that was quantitatively similar to that found in MPTP monkeys after infusion of nondyskinetic doses of subcutaneous apomorphine or oral levodopa (Boraud et al. 1998; Filion and Tremblay 1991) (Tables 3 and 4). Therefore the present doses of apomorphine were just adequate to produce dyskinesias consistent with previous studies.

For neurons recorded during simultaneous onset of ON and dyskinesias after administration of apomorphine, mean firing rates were consistent with previous reports (Table 3). The firing patterns of GPi neurons have previously been examined (4 neurons, 4 patients) during the OFF state and after administration of apomorphine induced dyskinesias (see Table 3) (see also Merello et al. 1999). In that study, firing rates during ON with dyskinesias were ~50% below those during the OFF state, and the spike train was characterized by an irregular pattern. Firing rates during periods of dyskinesias were not different from those during ON periods without dyskinesias. Similarly, after the transition from OFF to ON with "mild dyskinesias," a single neuron was reported to have a firing rate of 40% below baseline (60 Hz), apparently without any change in the firing pattern (Levy et al. 2001).

High doses of apomorphine produced "clear dyskinesias" and a reduction in GPi firing rates, during which neurons were "nearly completely silenced" (Table 4) (see also Filion et al. 1991). This is consistent with another study reporting that 'firing rates declined profoundly in almost all (GPi) neurons' during administration of high doses of levodopa (Table 4) (Papa et al. 1999; Poewe and Granata 1997). In MPTP monkeys, the administration of apomorphine at doses as little as 20 µg/kg produced dyskinesias and relatively small reduction in GPi firing rates (65%) (Filion et al. 1991). Boraud reported reliable dyskinesias and a 50% reduction in firing rates at apomorphine doses of 0.1 µg/kg in MPTP monkeys (Table 4). The present results during mild, transient dyskinesias may represent one end of a continuum, the other end of which is characterized by the intense, constant dyskinesias induced at high doses of levodopa or apomorphine (Filion et al. 1991; Papa et al. 1999; S. M. Papa, personal communication).

A relationship between decreasing GPi neuronal firing rates and more severe hyperkinetic movement disorders is consistent with the literature of these disorders (Hutchinson et al. 2003). A similar relationship between dystonia and GPi neuronal firing rates was found in an individual patient in whom firing rates decreased successively as dystonia became more severe in response to repeated voluntary movements (Lenz et al. 1998). Irregular firing patterns were characterized by "low frequency

modulation and pauses" and were more pronounced than in PD and less pronounced than in hemiballismus but did not increase with increasingly severe dystonia (see also Hutchison et al. 2003). Similarly, GPi neuronal firing rates were correlated inversely with severity of dystonia in a population of patients undergoing pallidal surgery for dystonia (Starr et al. 2005). In that study, irregularity of the spike train was higher than in PD or normal macaques but did not apparently change with the severity of dystonia. Finally, increased low-frequency, nonperiodic neuronal activity was observed in a GPi recipient nucleus of thalamus and was correlated with and led EMG during dystonia (Lenz et al. 1999). These results demonstrate that GPi neuronal firing patterns combined with a low firing rate are common features of different hyperkinetic movement disorders.

In the present results, a group of neurons had spike trains with nonindependent ISIs, non-Poisson distributions, and recurrent patterns of ISIs (Fig. 8). These GPi neurons may be influenced by a generator which produces recurrent patterns of ISIs during transient periods of ON with dyskinesias (Dayhoff and Gerstein 1983b; Eckmann et al. 1987; Kaluzny and Tarnecki 1993). Recurrent patterns have been found in the spike trains of both motor and sensory systems in both vertebrates and invertebrates (Dayhoff and Gerstein 1983b).

In sensory systems, different patterns of thalamic spike train activity result from different stimuli (Lee et al. 2005) and influence the synaptic efficacy of the excitatory thalamo-cortical synapse (Swadlow and Gusev 2001). In addition, different patterns of thalamic or peripheral nerve microstimulation can influence the quality and intensity of the evoked sensation (Lundberg et al. 1992; Patel et al. 2006). In motor systems, patterned spike trains have been found have been shown to influence the efficacy of excitatory synapses (Tsukada and Sugano 1978; Wiersma 1953).

Patterned firing in invertebrates has also been shown to influence the efficacy of inhibitory synapses and neuromuscular junctions (Perkel et al. 1964; Wiersma 1953). Therefore the patterned firing of the neurons in the inhibitory GABAergic projection from GPi to the pallidal recipient zone of the thalamus might exert a pattern dependent effect on postsynaptic elements in the thalamus. The patterned firing of neurons in GPi may likewise be dependent on inhibitory mechanisms. Inhibitory inputs to GPi from the striatum produce an inhibitory postsynaptic potential usually followed by rebound firing; this could account for the patterned firing in the present study (Kita et al. 2005; Nambu and Llinas 1994). These inhibitory inputs may be decreased in parkinsonism and increased during in drug-related dyskinesias (Albin et al. 1989; DeLong 1990), which suggests that these inputs should be increased after administration of apomorphine. If that is the case, then these inhibitory inputs may produce recurrent patterns characterized by a firing pattern/sequence of a pause followed by a sequence of short ISIs.

Of course, the presence of changes in firing patterns during apomorphine-induced dyskinesias does not prove that these changes are responsible for dyskinesias. For example, administration of apomorphine to normal monkeys produces decreases in firing rate and changes in neuronal firing patterns without any behavioral manifestation (Boraud et al. 2001, 2002). A relationship between patterns of

GPi neuronal activity and EMG during hyperkinetic disorders has been suggested based on visual and cross-correlation analysis of EMG with neuronal activity in the GPi and in a GPi recipient nucleus of the thalamus (see RESULTS (see also Lenz et al. 1999; Vitek et al. 1999)). Evidence for a causal relationship is the striking and long-lasting decrease in dyskinesias and hemiballismus that follows lesions of these neurons during GPi pallidotomy (Baron et al. 2000; Fine et al. 2000; Lang et al. 1997). Patterned firing of neurons in GPi may explain other hyperkinetic disorders that are effectively treated by pallidotomy, such as hemiballismus (Suarez et al. 1997; Vitek et al. 1999).

#### ACKNOWLEDGMENTS

We thank L. Rowland for excellent technical assistance.

#### GRANTS

This work was supported by National Institute of Neurological Disorders and Stroke Grants RO1 NS-38394, NS-40059 to F. A. Lenz, and NS-39933 to P. M. Dougherty.

#### REFERENCES

- Albin RL, Young AB, Penney JB.** The functional anatomy of basal ganglia disorders. *Trends Neurosci* 12: 366–375, 1989.
- Bar-Gad I, Ritov Y, Bergman H.** The neuronal refractory period causes a short-term peak in the autocorrelation function. *J Neurosci Methods* 104: 155–163, 2001.
- Baron MS, Vitek JL, Bakay RA, Green J, McDonald WM, Cole SA, DeLong MR.** Treatment of advanced Parkinson's disease by unilateral posterior GPi pallidotomy: 4-year results of a pilot study. *Mov Disord* 15: 230–237, 2000.
- Bendat JS, Piersol AG.** *Random Data*. New York: Wiley, 1976.
- Boraud T, Bezard E, Bioulac B, Gross CE.** Dopamine agonist-induced dyskinesias are correlated to both firing pattern and frequency alterations of pallidal neurons in the MPTP-treated monkey. *Brain* 124: 546–557, 2001.
- Boraud T, Bezard E, Bioulac B, Gross CE.** From single extracellular unit recording in experimental and human Parkinsonism to the development of a functional concept of the role played by the basal ganglia in motor control. *Prog Neurobiol* 66: 265–283, 2002.
- Boraud T, Bezard E, Guehl D, Bioulac B, Gross C.** Effects of L-DOPA on neuronal activity of the globus pallidus externalis (GPe) and globus pallidus internalis (GPi) in the MPTP-treated monkey. *Brain Res* 787: 157–160, 1998.
- Churchland PS, Sejnowski TJ.** *The Computational Brain*. Cambridge, MA: MIT Press, 1992.
- Corsini GU, Del Zompo M, Gessa GL, Mangoni A.** Therapeutic efficacy of apomorphine combined with an extracerebral inhibitor of dopamine receptors in Parkinson's disease. *Lancet* 1: 954–956, 1979.
- Cox DR, Lewis PAW.** *The Statistical Analysis of Series of Events*. London: Chapman and Hall, 1966.
- Dayhoff JE, Gerstein GL.** Favored patterns in spike trains. I. Detection. *J Neurophysiol* 49: 1334–1348, 1983a.
- Dayhoff JE, Gerstein GL.** Favored patterns in spike trains. II. Application. *J Neurophysiol* 49: 1349–1363, 1983b.
- DeLong MR.** Activity of pallidal neurons during movement. *J Neurophysiol* 34: 414–427, 1971.
- DeLong MR.** Primate models of movement disorders of basal ganglia origin. *Trends Neurosci* 13: 281–285, 1990.
- Dingle PJ, Heagerty A, Liang K-Y, Zeger S.** *Analysis of Longitudinal Data*. New York: Oxford, 2002.
- Eckmann JP, Kamphorst SO, Ruelle D.** Recurrence plots of dynamical systems. *Europhys Lett* 4: 973–977, 1987.
- Fahn S, Elton RL.** UPDRS development committee: unified parkinson's disease rating scale. In: *Recent Developments in Parkinson's Disease*, edited by Fahn S, Marsden CD, Calne DB, Goldstein M. Florham Park, NJ: MacMillan Health Care Information, 1987, p. 153–164.
- Favre J, Taha JM, Baumann T, Burchiel KJ.** Computer analysis of the tonic, phasic, and kinesthetic activity of pallidal discharges in Parkinson patients. *Surg Neurol* 51: 665–672, 1999.

- Filion M, Tremblay L.** Abnormal spontaneous activity of globus pallidus neurons in monkeys with MPTP-induced parkinsonism. *Brain Res* 547: 142–151, 1991.
- Filion M, Tremblay L, Bedard PJ.** Effects of dopamine agonists on the spontaneous activity of globus pallidus neurons in monkeys with MPTP-induced parkinsonism. *Brain Res* 547: 152–161, 1991.
- Fine J, Duff J, Chen R, Chir B, Hutchison W, Lozano AM, Lang AE.** Long-term follow-up of unilateral pallidotomy in advanced Parkinson's disease. *N Engl J Med* 342: 1708–1714, 2000.
- Gancher ST, Woodward WR, Boucher B, Nutt JG.** Peripheral pharmacokinetics of apomorphine in humans. *Ann Neurol* 26: 232–238, 1989.
- Glaser EM, Ruchkin DS.** *Principles of Neurobiological Signal Analysis*. New York: Academic, 1976.
- Holsapple JW, Preston JB, Strick PL.** The origin of thalamic input to the "hand" representation in the primary motor cortex. *J Neurosci* 11: 2644–2654, 1991.
- Hutchison WD, Lang AE, Dostrovsky JO, Lozano AM.** Pallidal neuronal activity: implications for models of dystonia. *Ann Neurol* 53: 480–488, 2003.
- Hutchison WD, Lozano AM, Davis KD, Saint-Cyr JA, Lang AE, Dostrovsky JO.** Differential neuronal activity in segments of the globus pallidus in patients with parkinson's disease. *Neuroreport* 5: 1533–1537, 1994.
- Kaluzny P, Tarnecki R.** Recurrence plots of neuronal spike trains. *Biol Cybern* 68: 527–534, 1993.
- Kaneoke Y, Vitek JL.** Burst and oscillation as disparate neuronal properties. *J Neurosci Methods* 68: 211–223, 1996.
- Kempster PA, Frankel JP, Stern GM, Lees AJ.** Comparison of motor response to apomorphine and levodopa in Parkinson's disease. *J Neurol Neurosurg Psychiatry* 53: 1004–1007, 1990.
- Kita H, Tachibana Y, Nambu A, Chiken S.** Balance of monosynaptic excitatory and disynaptic inhibitory responses of the globus pallidus induced after stimulation of the subthalamic nucleus in the monkey. *J Neurosci* 25: 8611–8619, 2005.
- Lang AE, Lozano AM, Montgomery E, Duff J, Tasker R, Hutchinson W.** Posteroventral medial pallidotomy in advanced Parkinson's disease. *N Engl J Med* 337: 1036–1042, 1997.
- Lee J, Dougherty PM, Antezana D, Lenz FA.** Responses of neurons in the region of human thalamic principal somatic sensory nucleus to mechanical and thermal stimuli graded into the painful range. *J Comp Neurol* 410: 541–555, 1999.
- Lee JI, Ohara S, Dougherty PM, Lenz FA.** Pain and temperature encoding in the human thalamic somatic sensory nucleus (ventral caudal): inhibition-related bursting evoked by somatic stimuli. *J Neurophysiol* 94: 1676–1687, 2005.
- Lemstra AW, Verhagen ML, Lee JI, Dougherty PM, Lenz FA.** Tremor-frequency (3–6 Hz) activity in the sensorimotor arm representation of the internal segment of the globus pallidus in patients with Parkinson's disease. *Neurosci Lett* 267: 129–132, 1999.
- Lenz FA.** Ablative procedures. In: *Handbook of Neurology. Parkinson's Disease and Related Disorders*, edited by Aminoff MJ, Boller F, Swaab TC. Amsterdam: Elsevier, in press.
- Lenz FA, Jaeger CJ, Seike MS, Lin YC, Reich SG, DeLong MR, Vitek JL.** Thalamic single neuron activity in patients with dystonia: dystonia-related activity and somatic sensory reorganization. *J Neurophysiol* 82: 2372–2392, 1999.
- Lenz FA, Kwan HC, Martin RL, Tasker RR, Dostrovsky JO, Lenz YE.** Single neuron analysis of the human ventral thalamic nuclear group: tremor-related activity in functionally identified cells. *Brain* 117: 531–543, 1994.
- Lenz FA, Suarez JI, Metman LV, Reich SG, Karp BI, Hallett M, Rowland LH, Dougherty PM.** Pallidal activity during dystonia: somatosensory reorganization and changes with severity. *J Neurol Neurosurg Psychiatry* 65: 767–770, 1998.
- Lenz FA, Tasker RR, Kwan HC, Schnider S, Kwong R, Murayama Y, Dostrovsky JO, Murphy JT.** Single unit analysis of the human ventral thalamic nuclear group: correlation of thalamic "tremor cells" with the 3–6 Hz component of parkinsonian tremor. *J Neurosci* 8: 754–764, 1988.
- Levy R, Dostrovsky JO, Lang AE, Sime E, Hutchison WD, Lozano AM.** Effects of apomorphine on subthalamic nucleus and globus pallidus internus neurons in patients with Parkinson's disease. *J Neurophysiol* 86: 249–260, 2001.
- Lozano A, Hutchison W, Kiss Z, Tasker R, Davis K, Dostrovsky J.** Methods for microelectrode-guided posteroventral pallidotomy. *J Neurosurg* 84: 194–202, 1996.
- Lundberg LER, Jorum E, Holm E, Torebjork HE.** Intra-neural electrical stimulation of cutaneous nociceptive fibres in humans: effects of different pulse patterns on magnitude of pain. *Acta Physiol Scand* 146: 41–48, 1992.
- Marsden CD.** Problems with long-term levodopa therapy for Parkinson's disease. *Clin Neuropharmacol* 17, Suppl 2: S32–S44, 1994.
- Marsden CD, Parks JD, Quinn N.** Fluctuations of disability in Parkinson's disease- clinical aspects. In: *Movement Disorders*, edited by Marsden CD, Fahn S. London: Butterworth Scientific, 1982, p. 96–122.
- Merello M, Balej J, Delfino M, Cammarota A, Betti O, Leiguarda R.** Apomorphine induces changes in GPi spontaneous outflow in patients with parkinson's disease. *Mov Disord* 14: 45–49, 1999.
- Merello M, Lees AJ, Balej J, Cammarota A, Leiguarda R.** GPi firing rate modification during beginning-of-dose motor deterioration following acute administration of apomorphine. *Mov Disord* 14: 481–483, 1999.
- Miller WC, DeLong MR.** Altered tonic activity of neurons in the globus pallidus and subthalamic nucleus in the primate MPTP model of parkinsonism. In: *The Basal Ganglia: Structure and Function II*, edited by Carpenter MB, Jayaraman A. New York: Plenum, 1987, p. 415–427.
- Mitchell IJ, Jackson A, Sambrook MA, Crossman AR.** The role of the subthalamic nucleus in experimental chorea. *Brain* 112: 1533–1548, 1989.
- Nambu A, Llinas R.** Electrophysiology of globus pallidus neurons in vivo. *J Neurophysiol* 72: 1127–1139, 1994.
- Nini A, Fiengold A, Slovin H, Bergman H.** Neurons in the globus pallidus do not show correlated activity in the normal monkey but phase-locked oscillations appear in the MPTP model of parkinsonism. *J Neurophysiol* 74: 1800–1805, 1995.
- Papa SM, Desimone R, Fiorani M, Oldfield EH.** Internal globus pallidus discharge is nearly suppressed during levodopa-induced dyskinesias. *Ann Neurol* 46: 732–738, 1999.
- Patel S, Ohara S, Dougherty PM, Gracely RH, Lenz FA.** Psychophysical elements of place and modality specificity in the thalamic somatic sensory nucleus (ventral caudal, vc) of awake humans. *J Neurophysiol* 95: 646–659, 2006.
- Paulson HL and Stern MB.** Clinical manifestations of parkinson's disease. In: *Movement Disorders*, edited by Watts RL, Koller WC. New York: McGraw Hill, 1997, p. 183–199.
- Perkel DH, Gerstein GL, Moore GP.** Neuronal spike trains and stochastic point processes. I. The single spike train. *Biophys J* 7: 391–418, 1967.
- Perkel DH, Schulman JH, Bullock TH, Moore GP, Segundo JP.** Pacemaker neurons: effects of regularly spaced synaptic input. *Science* 145: 61–63, 1964.
- Poewe W, Granata R.** Pharmacologic treatment of parkinson's disease. In: *Movement Disorders*, edited by Watts RL, Koller WC. 1997, p. 201–220.
- Press WH, Teukolsky SA, Vetterling WT, Flannery BP.** *Numerical Recipes in C*. Cambridge, UK: Cambridge, 1992.
- Schaltenbrand G, Bailey P.** *Introduction to Stereotaxis With an Atlas of the Human Brain*. Stuttgart, Germany: Thieme, 1959.
- Snedecor GW, Cochran WG.** *Statistical Methods*. Ames, IA: Iowa State University Press, 1967.
- Starr PA, Rau GM, Davis V, Marks WJ, Jr, Ostrem JL, Simmons D, Lindsey N, Turner RS.** Spontaneous pallidal neuronal activity in human dystonia: comparison with Parkinson's disease and normal macaque. *J Neurophysiol* 93: 3165–3176, 2005.
- Stata Corp.** *Stata Statistical Software*. College Station, TX: StataCorp LP, 2005, release 9.
- Stefani A, Mazzone P, Bassi A, Bernardi G, Altibrandi MG, Peppe A, Pierantozzi M, Stanzione P.** Electrophysiological and clinical desensitization to apomorphine administration in parkinsonian patients undergoing stereotaxic neurosurgery. *Exp Neurol* 156: 209–213, 1999.
- Stefani A, Stanzione P, Bassi A, Mazzone P, Vangelista T, Bernardi G.** Effects of increasing doses of apomorphine during stereotaxic neurosurgery in Parkinson's disease: clinical score and internal globus pallidus activity. Short communication. *J Neural Transm* 104: 895–904, 1997.
- Sterio D, Beric A, Dogali M, Fazzini E, Alfaro G, Devinsky O.** Neurophysiological properties of pallidal neurons in Parkinson's disease. *Ann Neurol* 35: 586–591, 1994.

- Suarez JI, Metman LV, Reich SG, Dougherty PM, Hallett M, Lenz FA.** Pallidotomy for hemiballismus: efficacy and characteristics of neuronal activity. *Ann Neurol* 42: 807–811, 1997.
- Swadlow HA, Gusev AG.** The impact of “bursting” thalamic impulses at a neocortical synapse. *Nat Neurosci* 4: 402–408, 2001.
- Tsukada M, Sugano N.** Effect of correlated adjacent interspike interval sequences of the input on the output response in the computer-simulated neuromuscular system. *Biol Cybern* 29: 69–73, 1978.
- Verhagen ML, Del Dotto P, van den MP, Fang J, Mouradian MM, Chase TN.** Amantadine as treatment for dyskinesias and motor fluctuations in Parkinson’s disease. *Neurology* 50: 1323–1326, 1998.
- Vitek JL, Bakay RA, Freeman A, Evatt M, Green J, McDonald W, Haber M, Barnhart H, Wahlay N, Triche S, Mewes K, Chockkan V, Zhang JY, DeLong MR.** Randomized trial of pallidotomy versus medical therapy for Parkinson’s disease. *Ann Neurol* 53: 558–569, 2003.
- Vitek JL, Bakay RA, Hashimoto T, Kaneoke Y, Mewes K, Zhang JY, Rye D, Starr P, Baron M, Turner R, DeLong MR.** Microelectrode-guided pallidotomy: technical approach and its application in medically intractable Parkinson’s disease. *J Neurosurg* 88: 1027–1043, 1998.
- Vitek JL, Chockkan V, Zhang J-Y, Kaneoke Y, Evatt M, DeLong MR, Triche S, Mewes K, Hashimoto T, Bakay RAE.** Neuronal activity in the basal ganglia in patients with generalized dystonia and hemiballismus. *Ann Neurol* 46:35, 1999.
- Wiersma CA.** Neural transmission in invertebrates. *Physiol Rev* 33: 326–355, 1953.