Pedunculopontine nucleus microelectrode recordings in movement disorder patients

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Abstract The pedunculopontine nucleus (PPN) lies within the brainstem reticular formation and is involved in the motor control of gait and posture. Interest has focused recently on the PPN as a target for implantation of chronic deep brain stimulation (DBS) electrodes for Parkinson's disease (PD) and progressive supranuclear palsy (PSP) therapy. The aim of this study was to examine the neurophysiology of the human PPN region and to identify neurophysiological landmarks that may aid the proper placement of DBS electrodes in the nucleus for the treatment of PD and PSP. Neuronal firing and local field potentials were recorded simultaneously from two independently driven microelectrodes during stereotactic neurosurgery for implantation of a unilateral DBS electrode in the PPN in five PD patients and two PSP patients. Within the PPN region, the majority (57%) of the neurons fired randomly while about 21% of the neurons exhibited 'bursty' firing. In addition, 21% of the neurons had a long action potential duration and significantly lower firing rate suggesting they were cholinergic neurons. A change in firing rate produced by passive and/or active contralateral limb movement was

observed in 38% of the neurons that were tested in the PPN region. Interestingly, oscillatory local field potential activity in the beta frequency range (~25 Hz) was also observed in the PPN region. These electrophysiological characteristics of the PPN region provide further support for the proposed role of this region in motor control. It remains to be seen to what extent the physiological characteristics of the neurons and the stimulation-evoked effects will permit reliable identification of PPN and determination of the optimal target for DBS therapy.

Keywords Pedunculopontine tegmental nucleus · Parkinson disease · Progressive supranuclear palsy · Deep brain stimulation · Basal ganglia

Introduction

Although STN deep brain stimulation (DBS) to treat advanced Parkinson's disease (PD) is a well established procedure and produces striking clinical improvement in motor symptoms such as tremor, rigidity and bradykinesia, it has only a moderate and a short term effect in improving axial symptoms like gait akinesia and postural instability (Kleiner-Fisman et al. 2003). Recently, the pedunculopontine tegmental nucleus (PPN) has been proposed (Pahapill and Lozano 2000) and examined as an alternative therapeutic target for PD, especially for patients with severe gait and postural impairment (Mazzone et al. 2005; Plaha and Gill 2005; Stefani et al. 2007). The PPN is part of the brainstem reticular formation and projects to the thalamus and spinal cord (Erro and Gimenez-Amaya 1999). It is also reciprocally connected to the basal ganglia and is believed to regulate the control of complex central nervous system functions including posture and gait (for review, see Pahapill and

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Lozano 2000). In primates, the PPN receives a major projection from the two output structures of the basal ganglia (substantia nigra pars reticulata and internal globus pallidus—GPi) and the subthalamic nucleus (STN). In turn, the PPN projects to the substantia nigra pars compacta and STN (Garcia-Rill 1991; Pahapill and Lozano 2000; Winn 2006). This organization emphasizes the potential role of the PPN in controlling basal ganglia output through subsidiary loops.

It has been shown that neurons in the PPN region degenerate in akinetic disorders such as PD and progressive supranuclear palsy (PSP) (Hirsch et al. 1987; Jellinger 1988; Zweig et al. 1989), and lesions in the PPN can lead to decreased movements (Aziz et al. 1998; Kojima et al. 1997). Therefore, it has been suggested that the PPN may be an alternative target for DBS for the treatment of PD (Jenkinson et al. 2004; Matsumura 2005; Pahapill and Lozano 2000; Plaha and Gill 2005). Indeed, recent studies have shown that low frequency stimulation (<25 Hz) of the PPN increases motor activity in a monkey model of PD (Jenkinson et al. 2004) and improves clinical motor scores in PD patients (Mazzone et al. 2005; Plaha and Gill 2005; Stefani et al. 2007).

The PPN occupies an area in the mesopontine tegmentum that contains two populations of neurons, non-cholinergic (likely glutamatergic) and cholinergic (Mesulam et al. 1989). The cholinergic neurons have larger somata, but are less numerous than the non-cholinergic neurons (Honda and Semba 1995; Rye et al. 1987; Spann and Grofova 1992). Electrophysiological studies of the PPN in non-human primates have rarely appeared in the literature (Matsumura 2005). In the PPN of the normal monkey, nearly half of the neurons recorded changed their firing rates in response to voluntary movement of the ipsi—and/or contralateral arm (Matsumura et al. 1997). In the cat it has been shown that PPN neurons change their firing activity during a conditioned movement of the forelimb (Dormont

et al. 1998) and that inactivation of the PPN by injection of muscimol (a γ -aminobutyric acid (GABA) agonist) induces prolonged arrest in the performance of the task (Conde et al. 1998). These findings suggest that the PPN is involved in the initiation and modulation of movements (see review: Matsumura 2005).

The aim of the present study was to describe the neurophysiology of the human PPN region and identify neurophysiological landmarks and characteristics that may help in localizing the target for insertion of DBS electrodes in the nucleus for the treatment of PD and PSP.

Methods

Patients

We studied seven patients, five with advanced PD and two with PSP, who were undergoing stereotactic surgery for implantation of unilateral PPN-DBS electrodes under local anesthesia. The group consisted of two women and five men who, at the time of operation, had a mean age ($\pm$ SD) of 64 ± 6.7 years and a mean disease duration of 12.4 ± 5.1 years. During surgery the patients were awake and 'off' dopaminergic medications for at least 12 h from the last oral dose of medication. Patient #7 had bilateral STN-DBS electrodes that were implanted one year before the surgery and were switched off during the recordings. Demographic details of the patients are given in Table 1. The study was performed with approval of the University Health Network Ethical Review Board, University of Toronto. Patients gave written and informed consent before surgery.

Surgical procedure and targeting

On the morning of the surgery, a stereotactic frame (Leksell G, Elekta, Inc, Atlanta, Ga) was affixed to the patients'

Table 1 Demographic and clinical features of the patients selected for surgery

Patient # and disease	Age (year) and gender	Disease duration (year)	L-Dopa therapy (year)	UPDRS III (off/on)	Gait sub-score (off/on)	Postural instability sub-score (off/on)	Freezing (off/on)	Side of brain	Span of record (mm)	Number of neurons sampled (PPN/total)
1-PD	63 M	10	5	34.5/20	2.5/0.5	3/1	Yes/yes	L	17	12/34
2-PSP	67 F	5	N/A	57.5/ (NA)	3.5 /NA	4/NA	No /NA	R	13	7/19
3-PSP	70 M	10	N/A	38.5/ (NA)	3.5/NA	3.5/NA	No/NA	L	13	3/9
4-PD	68 M	15	14	41/21	2.5/0.5	2.5/1	Yes/no	L	18 13	12/45 18/35
5-PD	67 M	21	15	42/20	2/0.5	2/1	Yes/no	L	16	12/37
6-PD	63 F	11	12	43.5/35	2/1.5	2/2	Yes/no	R	13	3/13
7-PD ^a	50 M	15	17	25.5/14	2/0.5	0/0	Yes/no	R	14	15/43

NA not applicable

^a Patient had bilateral STN-DBS

heads and preoperative magnetic resonance (MR) images were obtained (Signa, 1.5 T, General Electric, Milwaukee, Wis). To target the PPN we used 3D inversion recovery and T2-weighted axial sequences. Triplanar reconstruction of the images with the axial series parallel to the plane of the anterior (AC) and posterior commissures (PC) was carried out on a surgical neuronavigation workstation (Mach 4.1, StealthStation, Medtronic, SNT). In the dorsal ventral axis, a region that was 4–5 mm below the caudal aspect of the inferior colliculus was targeted on sagittal images. In the anteroposterior axis, our target was 2–3 mm posterior to the limit between the base and tegmentum of the pons. In the medial lateral axis, we targeted a region that was 3–4 mm medial to the lateral edge of the brainstem.

Once the MRI based coordinates of the target were calculated, the patient was brought to the operating room and a burr hole was drilled 2 cm from the midline in front of the coronal suture. Microelectrode recordings were then conducted from 15 mm above the target and extended 2–5 mm below. In each patient except one, only one dual electrode track (consisting of two microelectrodes 600 μ m apart—see Levy et al. 2007 for details) was performed. In patient #4 a second electrode track was performed 2 mm medial to the first since the thresholds for evoking paresthesia in the first track were very low.

Microelectrode recordings and stimulation

Neuronal spontaneous firing activity and local field potentials (LFPs) were recorded simultaneously from two independently driven microelectrodes (about 25 μ m tip length, axes 600 μ m apart, about 0.2 M Ω impedance at 1,000 Hz) during the electrophysiological mapping procedure used to obtain physiological data for localizing the target for the DBS electrode placement (for further details of the procedure see: Hutchison et al. 1998). All recordings were amplified 5,000–10,000 times and filtered at 10–5,000 Hz (analog Butterworth filters: high-pass, one pole; low-pass, two poles) using two Guideline System GS3000 (Axon Instruments, Union City, CA) amplifiers. Recorded signals were digitized at 10 kHz with a CED 1401 (Cambridge Electronic Design, Cambridge, UK). In all patients, we tested the effects of a 1 s microstimulation train (200 Hz, 150 μ s pulse width at intensities up to 100 μ A) along the microelectrode trajectory at intervals of 2 mm or less.

Localization of electrode trajectory

In the present study, the approximate location of the microelectrode tracks was estimated as follows: Preoperative axial stereotactic 3D inversion recovery and T2-weighted MR images were transferred to a StealthStation workstation. Using the FrameLink 4.1 software (Mach 4.1, StealthStation,

Medtronic, SNT) these two images were merged, the fiducials of the frame were recognized, and the mean rod marking error was calculated and registered. Coronal and sagittal planes were reconstructed based on axial images. The AC and PC were then targeted in the axial plane and three additional points were plotted in the midline. Thereafter, the images were reformatted parallel to the AC–PC plane and orthogonal to the midline. Pitch, roll, and yaw were corrected in the StealthStation. The coordinate of the microelectrode track and the angles of each trajectory were entered into the neuronavigation system so that they could be reconstructed. As the PPN cannot be clearly visualized on imaging studies, we estimated the location of the region that most likely encompassed the nucleus. We defined this PPN region as the anterolateral portion of the pontine/mesencephalic tegmentum that extended from 2 mm below the inferior colliculus to the transition between the inferior and superior colliculi, according to the Paxinos and Huang atlas (Paxinos and Huang 1995). The coordinates chosen for electrophysiological recordings were similar across patients.

Data analysis

Only sites with good signal-to-noise and stable single unit recordings were analyzed. To characterize the firing activity of the recorded neurons the signals were bandpass filtered at 425–5,000 Hz and single unit activity was discriminated using the template matching tool in Spike2 (CED). Mean firing rates and measurement of firing patterns were obtained using a modification of the Kaneoke and Vitek method (Kaneoke and Vitek 1996), which uses discharge density to categorize firing patterns into random, bursty or regular. As a measure of action potential width we used the duration of the negative phase of the averaged spike waveform (see Fig. 1a). A similar method was previously used to measure spike duration of PPN neurons by Matsumura et al. (1997) and Takakusaki et al. (1997).

For examining neuronal responses to movement, movement onset was determined from accelerometer recordings and/or electromyograms and a peri-stimulus histogram constructed. Movement-related changes in neuronal firing were initially identified by visual inspection of the histograms. Then, the identified responses were evaluated statistically using nonparametric tests comparing the firing rate during a 200 or 300 ms control period and during 200/300 ms of the visually determined response ($P < 0.05$, Wilcoxon signed rank test).

For spectral analysis, spike times and unfiltered LFP data were imported into MATLAB (version 6.5, The Math Works, Natick, MA). Only data of ≥ 11 s duration (mean $\pm$ SD: 37.9 $\pm$ 19.7 s, range: 11–112 s) were analyzed. Spectral analysis of neuronal discharges and LFP recordings

Fig. 1 Width of action potentials. (a) The width of the action potential was measured as the duration of the negative phase of the averaged spike waveform (b) Distribution histogram of spike durations

was performed according to Halliday et al. (1995). After signals were down-sampled to 1 kHz, spectra of LFP power were estimated by dividing the waveform signal into a number of sections of equal duration of 1.024 s (1,024 data points, 512 point overlap), each section was windowed (Hanning window) and the magnitude of the 1,024 discrete Fourier transforms of each section was squared and averaged to form the power spectrum for frequencies up to 80 Hz with a frequency resolution of 0.97 Hz. The power was transformed to a logarithmic scale and shown in decibels (dB). Since the estimated power spectrum has a distribution which is analogous to a χ^2 distribution, the 95% confidence intervals were given on the basis of the χ^2 distribution (Jarvis and Mitra 2001) while the degrees of freedom is based on the number of windowed sections. For detecting significant neuronal oscillations we used a process of spike train shuffling (see Rivlin-Etzion et al. 2006).

Coherence and cross-correlation analysis (Halliday et al. 1995; Rosenberg et al. 1989) was used to assess the relationship between simultaneously recorded data from separate electrodes and between LFP and spike data recorded from the same microelectrode. The coherence function provides a frequency domain bounded measure of association, taking on values between 0 and 1, with 0 in case of independence and 1 in case of a perfect linear relationship of both frequency, amplitude and phase. The 95% confidence level is given by $1 - (0.05)^{1/(L-1)}$, where L is the number of disjoint sections. Cross-correlation provides a time-domain measure of dependency between random processes. Under the hypothesis that in the case of two independent processes the value of the cross-correlation function is zero, the upper and lower 95% confidence limits for the estimated cross-correlation are given by $0 \pm 1.96 (\text{variance}_{\text{cross-corr}})^{1/2}$.

Results

Action potentials and firing properties

A total of 244 neurons was recorded in seven patients. Of these, 235 were recorded while the patient was at rest and were used to characterize the baseline firing activity. Neu-

rons were classified according to the width and polarity of their action potentials. The incidence histogram of spike durations did not reveal a clear bimodal distribution. Nevertheless, we observed a marked drop in the number of neurons that had action potential durations ≥ 0.65 ms (see Fig. 1b), which suggests they may have originated from a different population of neurons that are likely to be composed of cholinergic neurons (Dormont et al. 1998; Takakusaki et al. 1997). This population consisted of 38 neurons that comprised 15.6% of the total number of neurons. These neurons were found in all the patients except one (patient #3, PSP) in whom a total of only nine neurons were recorded (see Table 1). Their mean ($\pm$ SD) spike duration was 0.84 ± 0.2 ms compared with 0.47 ± 0.1 ms for the neurons with brief spikes. In addition, these neurons had a significantly lower mean firing rate (medians: 9.3 vs. 17.3 spikes/s, $P \leq 0.001$, Mann-Whitney rank sum test) (Fig. 2b, d). This provides further support for the assumption that these cells represent a different neuronal population. The majority of these neurons (86.8%) was found within and below the PPN region. In the region of the PPN, these neurons comprised 21% of the neurons (18/85) compared to 19.5% below that region (15/77) and 6% above (5/82).

Interestingly, 16 units had positive-going action potentials and significantly higher mean firing rates (median 67.6 spikes/s, $P \leq 0.001$) (Fig. 2c, d). These units were found in all seven patients. Six of these neurons were located in the PPN region and nine were located below that region. In addition, most (88%) of these neurons displayed a regular firing pattern as determined by the Kaneoke and Vitek method (Fig. 3a). This was significantly different from the patterns of activity exhibited by the two types of neurons described above which were primarily random (65 and 68% for the neurons with short and long spike durations respectively) ($P \leq 0.001$, χ^2 -test). Comparison of the proportions of both brief-and broad-spike duration neurons exhibiting regular, random or bursty firing patterns above, within and below the PPN region, revealed a statistically significant difference ($P = 0.016$, χ^2 -test) (Fig. 3b). In the PPN region, the majority (57%) of the neurons fired randomly while about 21% of the neurons exhibited 'bursty' firing. Above and below the PPN region, the proportion of

Fig. 2 Classification into three groups of neurons and their mean firing rates. (a–c) Superimposition of action potential waveforms obtained from three neurons displaying different spike durations and polarities (top trace) with 2 s of their spontaneous firing bandpass filtered at 425–5,000 Hz (bottom trace). **a** short duration negative spikes; **b** long duration negative spikes; **c** positive spikes. **d** Box plot of firing rates of the three types of neurons. The boxes represent the span of the first to third interquartile range in data distribution and the filled dots depict the outliers. The median is marked by a horizontal line

Fig. 3 Comparison of proportions of firing patterns for each cell type and location. **a** Stacked bar graphs showing percentage of neurons in each firing pattern category for each of the three types of neurons ($P \leq 0.001$, χ^2 -test). **b** Stacked bar graphs showing percentage of neu-

rons in each firing pattern category for both brief- and broad-spike duration neurons according to their location relative to the PPN region ($P = 0.016$, χ^2 -test)

bursting neurons was smaller (11 and 19% respectively) while more neurons exhibited random firing (65 and 75% respectively).

Neurons in the PPN region were responsive to movements

The activity of 103 neurons was recorded during contralateral limb movements. Movement tasks varied between passive and/or voluntary movements of the wrist, hand, foot, elbow and sometimes shoulder. 36% ($n = 37$) of the neurons responded to at least one type of passive or voluntary

movement of the limb. Out of the 41 neurons that were tested only for passive movements, 16 responded (39%) and out of the 50 neurons that were tested only for voluntary movements, 12 responded (24%). Twelve neurons were tested for both passive and voluntary movement, out of which, five neurons responded to passive movement only, two neurons responded to voluntary movement only and two neurons responded to both. Table 2 summarizes the results for each of the neuron types. The changes in firing rates were mostly excitatory (about 80%) and in some cases, were followed by inhibition (see Fig. 4a, c). Within

Table 2 Neuron types and movement-related activity

Note that 12 neurons that were tested for both passive and voluntary movement are included in both groups
Nr no response

Neuron type	Passive movement				Voluntary movement				All
	Increase	Decrease	Nr	Total	Increase	Decrease	Nr	Total	
A	17	4	25	46	11	2	37	50	96
B	0	1	5	6	1	1	7	9	15
C	1	0	0	1	1	0	2	3	4
Total	18	5	30	53	13	3	42	62	115

Fig. 4 Examples of movement-evoked responses of the three types of neurons. **a** An example of a type A neuron exhibiting an increase in firing rate in response to passive wrist flexion that is followed by a decrease in firing rate ($P = 0.001$, Wilcoxon signed rank test). **b** Type B neuron exhibiting increased firing rate to passive elbow movement ($P \leq 0.001$). **c** Type C neuron exhibiting increased firing followed by decreased firing in response to passive flexion and extension of the foot respectively ($P = 0.003$). All movements were contralateral to the recording side. Raster plots (*middle traces*) and stimulus histograms (*bottom traces*) were aligned to movement onset (indicated by smoothed and rectified EMG (**a, b**) or accelerometer (**c**) output) (*top traces*)

the PPN region 38% of the neurons responded to movement and similar percentages were recorded above and below the PPN region where 37 and 33% of the neurons responded respectively. Movement responsive neurons included all three neuron types. Figure 4 shows an example of responses to contralateral movement from each neuron type.

Microstimulation in the PPN region

In all patients, 1 s stimulus trains evoked paresthesia at some sites along the microelectrode trajectory. In three patients, the threshold intensities for stimulation-evoked paresthesia varied between 50 and 100 μ A (with three exceptions of 20, 25 and 25 μ A) (mean $\pm$ SD: 79.2 $\pm$ 29.6 μ A, n = 24). In the remaining four patients, the threshold intensities were lower and varied between 1 and 50 μ A (with two exceptions of 100 and 75 μ A) (mean $\pm$ SD: 18.8 $\pm$ 19.9 μ A, n = 55). The thresholds were not significantly different when stimulating within or close to the PPN region. These responses were assumed to be due to activation of ascending axons in the medial lemniscus. In addition, eye movements were evoked by microstimulation in two patients and occurred only at the maximum stimulation intensity of 100 μ A, and only within and up to 2 mm above the PPN region.

LFP oscillatory activity in the PPN region

Spectral analyses of the local field potentials revealed oscillatory activity in the beta frequency range (15–30 Hz) in the PPN region of three PD patients (#1, 4 and 7). In all three patients, a significant coherence in the beta range between the two microelectrodes was also observed. It was not possible to adequately assess the existence of LFP oscillatory activity in the other four patients due to suboptimal slow wave recordings in those cases. Figure 5a demonstrates LFP beta oscillations recorded from the two microelectrodes (in patient #1), and their significant coherence. In these patients, the LFP power in the beta frequency range varied with depth along the microelectrode track. Beta oscillations were most prominent in the region of the PPN (Fig. 5b) and up to 4 mm below. Frequencies between 35 and 50 Hz were sometimes encountered in the LFP but the coherence in these frequencies rarely reached significance (this is notable in Fig. 5a). Similarly, oscillations at frequencies lower than 15 Hz, although occasionally observed, were not a consistent feature in the PPN region. Spectral analyses of the neuronal firing in the PPN region failed to show significant oscillatory activity in the beta frequencies. Moreover, coherence and cross-correlation analyses did not reveal a significant relationship to the simultaneously recorded LFP.

Comparison of PD versus PSP

No obvious differences were noted in any of the findings between the two groups of patients other than a lower number of neurons encountered in the PSP patients compared to all but one of the PD patients.

Discussion

The present study provides new data documenting various electrophysiological characteristics of the PPN region in PD and PSP patients. The lower number of cells recorded from the PPN region in the two PSP patients (see Table 1) suggests a lower density of neurons which might reflect the large degree of neuronal degeneration and brainstem atrophy that occurs in PSP (Dickson et al. 2007; Hirsch et al. 1987). We did not observe any obvious neurophysiological differences between the two patient groups. However, in view of the small number of neurons in the 2 PSP patients this comparison may not be very meaningful.

Analysis of spike duration and shape revealed three types of neurons. The existence of short and long duration negative going action potentials in this region agrees with the findings of previous in vitro and in vivo animal studies. In vivo recordings from the PPN in rats (Takakusaki et al. 1997), cats (Dormont et al. 1998) and monkeys (Matsumura et al. 1997) revealed the existence of two types of neurons: ones which fire at low rate and exhibit a long duration spike, and ones which fire at higher rates and exhibit a short duration spike. Yet, the incidence histogram of spike durations in our study did not reveal a clear bimodal distribution as previously demonstrated in the rat PPN (Takakusaki et al. 1997). That study, however, utilized intracellular recordings and therefore cannot be directly compared to our extracellular recordings. Moreover, the incidence histogram in our study is consistent with data from extracellular recordings in the monkey PPN that failed to exhibit a bimodal distribution (Matsumura et al. 1997).

It has been proposed on the basis of in vitro studies that the neurons with broad spikes are cholinergic (Dormont et al. 1998; Takakusaki et al. 1997). Although the exact borders of the PPN remain indistinct, the cholinergic neurons are a very obvious component of the PPN (Winn 2006). Indeed, the neurons with broad spikes in our study were found primarily within the region of the PPN and below it. The region below the PPN is likely to be the adjacent laterodorsal tegmental nucleus which is known to contain cholinergic neurons that are interconnected with the cholinergic population in the PPN (Winn 2006). These findings, together with the low rate of firing, provide further support for the assumption that these neurons are cholinergic. In our study, the majority of the neurons in the PPN

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Fig. 5 Beta oscillatory activity in the PPN region. **a** An example of power spectra of the LFPs recorded from each of the two adjacent microelectrodes (*upper* and *middle* traces) and the corresponding coherence function (*lower* trace). Dotted lines represent 95% confidence limits. **b** An example of LFP power changes along the microelectrode track, above within and below the PPN region. The LFP power was normalized and expressed as the percentage of the total power between 5 and 50 Hz

region exhibited random firing while about 21% of the neurons had a 'bursty' firing pattern. This finding is supported by earlier studies in rats (Scarnati et al. 1987), cats (Garcia-Rill et al. 2004) and monkeys (Matsumura et al. 1997) which describe a small population of bursty PPN neurons, whereas the majority of the neurons have been reported to fire in an irregular pattern.

The positive-going action potentials may possibly have originated from large diameter axons of passage as extracellularly recorded positive going potentials are generally assumed to be axonal. They had a significantly higher mean firing rate and a regular firing pattern. These electrophysiological properties are different than those of the PPN neurons previously described and are more similar to those seen in the GPi in PD patients (Hutchison et al. 2003; Starr et al. 2005; Tang et al. 2005). Therefore, the positive-going

action potentials might be from GPi afferents to the PPN region (Shink et al. 1997) or perhaps from ascending sensory axons of the medial lemniscus which lies laterally to the PPN (Olszewsky and Baxter 1982).

Previous electrophysiological studies in cats and monkeys have identified PPN neurons that exhibit changes in firing rate in response to limb movement and include neurons with both brief and broad spikes (Dormont et al. 1998; Matsumura et al. 1997). In monkey, PPN neurons have also been found to respond with either an increase or a decrease in firing rate to voluntary saccades (Kobayashi et al. 2002). Mazzone et al. (2005) reported finding 6 neurons (of a total of 27 studied) in the PPN of two PD patients that responded with a "slight inhibition or inhibition followed by excitation" to a single-joint movement, but no further details were provided (Mazzone et al. 2005). It is important to note

that the PPN region targeted in the current study (PPTg according to Paxinos and Huang 1995) may be different from the region studied by Mazzone et al. (see also Yelnik 2007; Zrinzo et al. 2007a, b). Our study clearly demonstrates the existence of movement-responsive neurons in the human PPN, consistent with the findings of animal studies. Moreover, we found that both short and long spike duration neurons responded to movements. It has been reported that PPN inputs to the basal ganglia are mediated by both cholinergic and glutamatergic synapses (Lavoie and Parent 1994). Taken together, these findings suggest that both groups of neurons may contribute to basal ganglia-related motor functions.

In the three PD patients analyzed, the LFPs in the PPN region exhibited oscillatory activity in the beta range (15–30 Hz). Similar oscillations have been previously observed in the STN and GPi of PD patients (Brown et al. 2001; Kuhn et al. 2005; Levy et al. 2002; Weinberger et al. 2006) and are suggested to play an ‘antikinetic’ role in PD (Brown 2003). It is important to note, however, that the neuronal firing did not show oscillations in these frequencies. The absence of oscillatory firing may be due to limited sampling or may reflect a lack of coupling between dendritic membrane potential oscillations and the soma. Since the beta oscillations were maximal in the PPN region they are unlikely to reflect far field potentials from distant structures (e.g. STN, cortex). The elevation of the beta frequency band LFP activity around the region of the PPN suggests that this activity may be useful in determining the final positioning of the DBS electrode intra-operatively, but further studies are required to examine the consistency of this feature. In a recent study by Androulidakis et al. (2008), LFP recordings from DBS electrodes in the PPN region in six PD patients revealed 7–11 Hz oscillations that were prominent only after treatment with levodopa, suggesting that these oscillations may represent a physiological pattern of activity in this region (Androulidakis et al. 2008). However, inconsistent with our finding, that study did not reveal beta oscillations in the ‘off’ dopaminergic state. This discrepancy might be due to the difference in the region targeted in that study, which is similar to that reported by Mazzone et al. (see above).

In summary, the electrophysiological characteristics of the PPN region described in this paper provide further support for the proposed role of this region in motor control. However, in contrast to microelectrode recordings in thalamus, pallidum and STN, we did not find distinctive and definitive neurophysiological characteristics and clear nuclear boundaries for PPN. This is consistent with the absence of precise anatomical borders to the PPN (Winn 2006). Thus microelectrode recordings and microstimulation-induced effects in PPN, although helpful, do not provide the same degree of confirmation of the target as they

do in these other regions although future studies may reveal additional features distinctive for the PPN which would be useful in localizing the region and identifying an optimal target for DBS therapy.

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