Neurophysiological correlates of bradykinesia in Parkinson's disease

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ABSTRACT

Many neurophysiological abnormalities have been described in the primary motor cortex of patients with Parkinson's disease. However, it is unclear whether there is any relationship between them and bradykinesia, one of the cardinal motor features of the condition. In the present study we aimed to investigate whether objective measures of bradykinesia in Parkinson's disease have any relationship with neurophysiological measures in M1 as assessed by means of transcranial magnetic stimulation techniques. Twenty-two patients with Parkinson's disease and 18 healthy subjects were enrolled. Objective measurements of repetitive finger tapping (amplitude, speed and decrement) were obtained using a motion analysis system. The excitability of primary motor cortex was assessed by recording the input/output curve of the motor-evoked potentials and using a conditioning-test paradigm for the assessment of short-interval intracortical inhibition and facilitation. Plasticity-like mechanisms in primary motor cortex were indexed according to the amplitude changes in motor-evoked potentials after the paired associative stimulation protocol. Patients were assessed in two sessions, i.e. 'OFF' and 'ON' medication. A canonical correlation analysis was used to test for relationships between the kinematic and neurophysiological variables. Patients with Parkinson's disease tapped more slowly and with smaller amplitude than normal, and displayed decrement as tapping progressed. They also had steeper input/output curves, reduced short-interval intracortical inhibition and a reduced response to the paired associative stimulation protocol. Within the patient group, bradykinesia features correlated with the slope of the input/output curve and the after-effects of the paired associative stimulation protocol. Although dopaminergic therapy improved movement kinematics as well as neurophysiological measures, there was no relationship between them. In conclusion, neurophysiological changes in primary motor cortex relate to bradykinesia in patients with Parkinson's disease, although other mechanisms sensitive to dopamine levels must also play a role.

Key words: Bradykinesia, motor cortex, motor control, Parkinson’s disease, motor evoked potentials

Abbreviations: abductor pollicis brevis (APB), active motor threshold (AMT), analysis of variance (ANOVA), Beck Depression Inventory (BDI), canonical correlation analysis (CCA), coefficient of variation (CV), electromyography (EMG), Frontal Assessment Battery (FAB), Fatigue Severity Scale (FSS), healthy controls (HC), Hoehn and Yahr (H&Y), input-output (I/O), inter-stimulus interval (ISI), intracortical facilitation (ICF), least absolute shrinkage and selection operator (LASSO), long-term potentiation (LTP), Montreal cognitive assessment (MoCA), motor-evoked potentials (MEP), Movement Disorder Society-sponsored revision of the Unified Parkinson's Disease Rating Scale (MDS-UPDRS), paired associative stimulation (PAS), Parkinson’s disease (PD), primary motor cortex (M1), resting motor threshold (RMT), short-interval intracortical inhibition (SICI), transcranial magnetic stimulation (TMS).

INTRODUCTION

Bradykinesia, or slowness of movement, is one of the cardinal motor features of Parkinson's disease (PD) (Berardelli *et al*., 2013; Postuma *et al*., 2015). Objective kinematic measures show that this is also accompanied by other changes, including low amplitude (hypokinesia) and a progressive reduction in amplitude and velocity during movement repetition (decrement) (Agostino *et al*., 1992 and 2003; Berardelli *et al.*, 2001; Espay *et al.*, 2009 and 2011; Kang *et al.*, 2010; Heldman *et al.*, 2014; Bologna *et al*., 2016a; Hasan *et al.*, 2017). Bradykinesia is believed to result primarily from a failure of basal ganglia output to the primary motor cortex (M1) (Berardelli *et al.*, 2001). However, there is also evidence from animal studies that there are additional intrinsic deficits in M1 that may contribute towards production of symptoms (Xu *et al.*, 2017; Pasquereau *et al.*, 2011 and 2016). The question we address here is whether these may also play some role in determining bradykinesia in human patients.

Neurophysiological studies in humans using transcranial magnetic stimulation (TMS) have revealed changes in resting measures of excitability and plasticity in M1. There is enhanced corticospinal excitability and reduced M1 inhibition (Cantello *et al.*, 2002; Currà *et al.*, 2002; Lefaucheur *et al.*, 2005; Berardelli *et al.*, 2008; Bologna *et al.*, 2016b), together with reduced long-term potentiation (LTP)-like plasticity in M1 (Morgante *et al.*, 2006; Ueki *et al.*, 2006; Schwingenschuh *et al.*, 2010; Suppa *et al.*, 2011; Kojovic *et al.*, 2012 and 2015; Kawashima *et al.*, 2013; Kishore *et al.*, 2017). Some studies have reported a weak relationship between changes in plasticity and clinical motor scores, i.e. lower plasticity associated with more severe motor symptoms (Ueki *et al.*, 2006; Kojovic *et al.*, 2012 and 2015; Kishore *et al.*, 2017).

The aim of the present study was to investigate possible relationships between movement kinematics and neurophysiological changes in the M1 of patients with PD. Movement was assessed objectively during repetitive finger tapping and the excitability and plasticity of M1 was measured in the resting state using various TMS techniques. To evaluate the effects of dopaminergic

treatment on the neurophysiological measures, we assessed patients in two separate sessions, i.e. both off and on their usual therapy. Data obtained from patients with PD were compared with those obtained from a group of healthy subjects.

MATERIALS AND METHODS

Participants

Twenty-two patients with PD, (4 females, mean age ± 1 standard deviation: 67.2 ± 10.3; Table1) and 18 healthy controls (HC) (6 females, mean age ± 1 standard deviation: 63.0 ± 11.8) were enrolled in the study. The diagnosis of PD was based on clinical criteria (Berardelli *et al.*, 2013; Postuma *et al.*, 2015). The clinical assessment included the Hoehn and Yahr (H&Y) scale, the motor section (part III) of the Movement Disorder Society-sponsored revision of the Unified Parkinson's Disease Rating Scale (MDS-UPDRS), (Goetz *et al.*, 2008; Antonini *et al.*, 2013); side of predominance of motor symptoms was considered evaluating the current most affected side. The clinical assessment also included the Beck Depression Inventory (BDI), (Beck *et al.*, 1961), the Montreal cognitive assessment (MoCA), (Nasreddine *et al.*, 2005) the Frontal Assessment Battery (FAB) (Dubois *et al.*, 2000) and the Fatigue Severity Scale (FSS) (Friedman *et al.*, 2010). The clinical assessment was performed by a clinician blinded to the experimental procedures. The experimental procedures, which adhered to the Declaration of Helsinki regulations and to international safety guidelines (Rossi *et al.*, 2009; Rossini *et al.*, 2015), were approved by the local institutional review board. All the participants gave their written informed consent to the study.

Kinematic assessment

The participants were comfortably seated in a chair and were asked to perform repetitive finger tapping. Three 15 sec. trials were recorded from the more affected side in patients and from the

dominant side in HC. Participants were allowed to rest for 45-60 sec. between acquisition trials to avoid fatigue. Before the kinematic recordings, one practice trial was allowed for the participants to become familiar with the motor task.

Kinematic recordings were performed using an optoelectronic system (SMART motion system, BTS Engineering, Italy). Three infrared cameras followed the 3D displacement of reflective markers taped to the participant's upper limb (sampling rate of 120 Hz). We used reflective markers with a 5 mm diameter and of negligible weight. Two markers were placed on the tips of the index finger and thumb. A further three markers were placed on the hand to define a reference plane that was used to mathematically exclude possible contamination due to unwanted hand movements from repetitive finger movement recordings (Bologna *et al.*, 2016a).

Movement analysis was performed using a dedicated software (SMART Analyzer, BTS Engineering, Italy). To quantify repetitive finger movement kinematics, we used linear regression techniques to determine the intercept, which reflects the movement amplitude (degree) and velocity (degree/s), and the slope, which reflects the amplitude and velocity decrement during the movement repetition. Movement rhythm was also measured by the coefficient of variation (CV) of the inter-tap intervals (with higher values representing a lower regularity of repetitive movements), (Iansek *et al.*, 2006; Bologna *et al.*, 2016a).

TMS techniques and electromyographic recordings

Single- and paired-pulse TMS was delivered using two Magstim magnetic stimulators (Magstim Company, UK) connected to an 8-shaped coil, with the intersection of the coil held tangentially to the scalp and the coil handle positioned at a ~45° angle from the midline pointing backward. We defined the hot spot of the abductor pollicis brevis (APB) muscle, i.e. the optimal scalp position for eliciting motor-evoked potentials (MEP) of maximal amplitudes in the muscle.

We first determined the resting motor threshold (RMT) and the active motor threshold (AMT) to

the nearest 1% of the maximal stimulator output, (Rossi *et al.*, 2009; Rossini *et al.*, 2015). We then measured the MEP input-output (I/O) curve to probe M1 excitability. We used a total of 60 single pulses at six stimulation intensities, ranging in 20% increments from 80% to 180% of the RMT, delivered in groups of ten. The intensity order was randomized in order to avoid hysteresis effects (Möller *et al.*, 2009).

We also assessed short-interval intracortical inhibition (SICI) and facilitation (ICF) using paired-pulse TMS with a subthreshold conditioning stimulus (90% AMT) and a supra-threshold test stimulus (1mV MEP) with an inter-stimulus interval (ISI) between conditioning and test stimuli of 2 and 4ms for SICI and 10 and 15 ms for ICF (Peurala *et al.*, 2008; Rossini *et al.*, 2015). We choose the intensity of 90% AMT for the conditioning stimuli 2 and 4 ms ISIs because in previous studies (Peurala *et al.*, 2008) it was demonstrated that in these experimental conditions there is no overlap between SICI and short-interval intracortical facilitation. Moreover, it has been reported in PD patients OFF medication that no short-interval intracortical facilitation occur at 2 and 4 ms ISIs (Ni *et al.*, 2013). Ten trials were acquired for each ISI and intensity. SICI and ICF were expressed as the percentage ratio between the unconditioned and conditioned MEP.

To study cortical plasticity, PAS was delivered over M1 contralateral to the more affected side of the body in patients (Kojovic *et al.*, 2012 and 2015). PAS consisted of 200 electrical stimuli, delivered to the median nerve at the wrist by means of a Digitimer DS7 (Digitimer, UK), paired with TMS stimuli (adjusted to 1 mV MEP intensity), delivered over the contralateral APB hot spot (rate 0.25 Hz, electrical stimulation intensity 2-3 times the perceptual threshold) (Wolters *et al.*, 2003; Kojovic et *al.*, 2012 and 2015). Each TMS stimulus was preceded by an electrical conditioning stimulus at an ISI of 21.5 msec. We chose this specific ISI because, unlike PAS 25ms, PAS 21.5ms is not affected by cerebellar activity (Hamada *et al.*, 2012). During PAS, participants were instructed to look at their hand and to report every 20^{th} peripheral electrical stimuli they perceived in order to ensure constant attention levels and comparable conditions between sessions (Stefan *et al.*, 2004; Kojovic *et al.*, 2012 and 2015).

EMG activity was recorded from the APB and first dorsal interosseous (FDI) muscles of the more affected side in patients and of the dominant side in HC, using surface electrodes taped in a belly-tendon montage. EMG signals were amplified and filtered (20 Hz-1 kHz) using Digitimer D360 (Digitimer, UK). EMG signals were recorded and stored on a laboratory PC (sampling rate of 5 kHz) through an analog-digital converter AD1401 plus (Cambridge Electronic Design, UK) for subsequent off-line analyses performed using a dedicated software (Signal® version 4.00, Cambridge Electronic Design, UK). The MEP peak-to-peak amplitude was measured within a time window of 20-40 ms after the TMS artifact. Traces with background EMG activity exceeding 100 µV in the 200 ms time window preceding the TMS artifact were rejected online.

Experimental design

Patients underwent two sessions ('OFF' and 'ON' medication). All the patients were studied after overnight withdrawal (at least 12 hours) of their medication, in the 'practically defined OFF condition' (Defer *et al.*, 1999) or while they were on their usual therapeutic regimen ('ON' medication), expressed in terms of levodopa equivalent daily dose (LEDD) (Tomlinson *et al.*, 2010). Each session was randomly performed and counter-balanced across patients at least one week apart. Kinematic recordings and TMS measures of corticospinal and intracortical excitability were collected in each session at baseline (B). In order to assess M1 plasticity, we then performed the PAS protocol and followed up the M1 excitability changes at three time points: T1 (5min after PAS), T2 (15 min after PAS) and T3 (30 min after PAS) using single-pulse TMS. Fifteen MEPs were recorded at 1 mV intensity at each measurement time point (including baseline); for the subsequent analysis, data at T1, T2 and T3 were normalized to B. The examiners who collected the neurophysiological measures were blinded to the patients' medication status.

Statistical analysis

Age and gender differences between PD patients and HC were evaluated using the Mann-Whitney

U test and the Fisher-exact test, respectively. The MDS-UPDRS (part III) scores in the 'OFF' and 'ON' sessions in patients were compared using the Wilcoxon test.

Group comparisons on kinematic variables and on motor thresholds between PD patients (OFF medication) and HC were performed by means of two tailed unpaired t-tests. Group comparisons on M1 excitability were evaluated using a repeated-measures analysis of variance (ANOVA) with the between-group factor 'GROUP' (PD patients 'OFF' medication and HC) and the within-group factor 'STIMULUS INTENSITY' (80%, 100%, 120%, 140% 160% and 180% RMT). When evaluating SICI and ICF, we used the within-group factor 'ISI' (2, 4 ms and 10,15 ms, respectively) in addition to 'GROUP'; SICI and ICF were analyzed in two separate ANOVAs as they represent different cortical circuits. When evaluating the effects of PAS, we used the factors 'GROUP', 'MUSCLE' (ABP and FDI) and 'TIME POINT' (T1, T2 and T3). We excluded the possible influence of handedness on movement kinematics and TMS data in patients with an additional analysis comparing the two patients subgroups (PD patients tested on the left/dominant hemisphere and PD patients tested on the right/non-dominant hemisphere) and HC (supplementary data).

To evaluate the effects of medication in patients, we added the within-group factor 'SESSION' (two levels: 'OFF' and 'ON' medication) to the various ANOVAs. Two tailed t-tests were used for post-hoc analyses in ANOVAs. Greenhouse-Geisser corrections were applied whenever we found a violation of sphericity in Mauchly's tests. Different neurophysiological variables were evaluated in separate ANOVAs.

For subsequent analysis we computed the steepness of the I/O MEP curve (i.e. the slope of the regression line across the scatter plot of the MEP amplitude - Y axis vs. the stimulation intensity - X axis) and the average percentage changes after PAS of the MEP amplitude values across the three measurement time points (T1, T2 and T3).

A multiple logistic model was also used to determine the variables best predicting disease status and medication status. For this purpose we considered the least absolute shrinkage and selection

operator - LASSO algorithm with l1-norm penalty. This procedure can be considered as a variable selection tool since it can estimate some variable coefficients to be 0 (Tibshirani, 1996).

A canonical correlation analysis (CCA) was used to assess the relationship between (i) neurophysiological measures and kinematic parameters, and (ii) neurophysiological measures and clinical-demographic data. Rather than assuming that there is a simple relationship between, say, a neurophysiological variable and a particular kinematic parameter as assumed with a linear correlation, a CCA examines whether combinations of neurophysiological measures are better predictors of kinematic variables. The procedure can reveal relationships which would otherwise be missed by simple linear correlation analysis (Hotelling, 1936). We only included variables in the CCA that had been demonstrated to be significantly different in the univariate analysis (also predictive of the disease or medication status, as demonstrated by the LASSO procedure).

Unless otherwise stated, the results are indicated as mean values ± 1 standard error of the mean. The level of significance was initially set at $P<0.05$, with the false discovery rate subsequently being applied to multiple comparisons (Curran-Everett, 2000). Data were analyzed using STATISTICA® (StatSoft, Inc) and implemented with R.

RESULTS

All the study participants completed the experimental procedure. None of the participants reported adverse effects during the experiments. No difference was found in age ($P=0.37$) or gender distribution ($P=0.23$) between PD patients and HC. As expected, the MDS-UPDRS part III score in PD patients was significantly higher in the 'OFF' medication session than in the 'ON' medication session (30.4±10.9 vs. 24.8±10.1; $P<0.001$).

PD patients 'OFF' medication vs. healthy controls

Finger tapping kinematics

The analysis yielded a significant between-group difference for movement amplitude and velocity (with lower values for both parameters being observed in PD patients than in HC (both Ps <0.01, Table 2). The analysis also revealed a between-group difference for movement amplitude slope (sequence effect), (P=0.02) with higher values being observed in PD patients than in HC. No significant difference emerged between PD patients and HC in the movement number, CV values of the inter-tap intervals and velocity slope (Table 2).

Corticospinal excitability: motor thresholds and I/O curve

The analysis did not reveal any differences in RMT or AMT between PD patients and HC (Table3). As expected, the ANOVA yielded a significant effect of the main factor 'STIMULUS INTENSITY' ($F_{5,190}$=77.26, P<0.001), with an increasing MEP amplitude being observed with increasing stimulation intensity. M1 excitability, as assessed by means of the I/O MEP curve, was greater in PD patients 'OFF' medication than in HC (Figure 1), as demonstrated by a significant interaction 'GROUP' x 'STIMULUS INTENSITY' ($F_{5,190}$=3.19, P=0.009). Lastly, the factor 'GROUP' was not found to be significant ($F_{1,38}$=2.06, P=0.15).

Intracortical excitability: SICI and ICF

When analyzing SICI, the ANOVA revealed a significant effect of the main factors 'GROUP' ($F_{1,38}$=7.89, P=0.007), with less inhibition being observed in PD than in HC. The main factor 'ISI' was also significant ($F_{1,38}$=8.87, P=0.005), indicating more profound inhibition at 2 ms than at 4 ms while there was no significant interaction 'GROUP' x 'ISI' ($F_{1,38}$=0.86, P=0.35), (Figure 1). Excitability of the intracortical facilitatory interneurons, as assessed by means of ICF, did not differ between patients with PD and HC (Figure 1), as is demonstrated by a lack of significant effect of 'GROUP' ($F_{1,38}$=0.006, P=0.93), 'ISI' ($F_{1,38}$=0.45, P=0.50) and interaction 'GROUP' x 'ISI' ($F_{1,38}$=0.11, P=0.73).

M1 plasticity: PAS-related effects

The analysis of normalized values indicated that the MEPs increased after PAS in HC but not in patients (Figure 2). This finding is supported by a repeated-measures ANOVA, which yielded a significant effect of the main factor 'GROUP' ($F_{1,38}$=6.12, P=0.01), with lower values being observed in PD patients than in HC. The analysis also showed a significant effect for the main factor 'MUSCLE' ($F_{1,38}$=6.33, P<0.01) and for the interaction 'GROUP' x 'MUSCLE' ($F_{1, 38}$=5.82, P=0.02) with higher facilitation being observed in the APB (target muscle) than in the FDI in HC but not in PD. No significant effects were observed for the main factor 'TIME POINT' ($F_{2, 76}$=2.43, P=0.09) or for the interactions 'GROUP' x 'TIME POINT' ($F_{2,76}$=0.66, P=0.51); 'MUSCLE' x 'TIME POINT' ($F_{2,76}$=0.54, P=0.58) and 'GROUP' x 'MUSCLE' x 'TIME POINT' ($F_{2,76}$=0.76, P=0.47).

PD patients 'OFF' medication vs. 'ON' medication

Finger tapping kinematics

The analysis revealed higher movement amplitude and velocity values in the 'ON' medication condition than in the 'OFF' medication condition (both Ps<0.001). By contrast, no significant effect of medication was observed for the movement number, CV values of the inter-tap intervals and for the amplitude and velocity slopes (all Ps>0.05) (Table 2).

Corticospinal excitability: motor thresholds and I/O curve

The analysis did not reveal any differences in RMT (P=0.69) or AMT (P=0.08) between PD patients 'OFF' medication and those 'ON' medication, (Table 3).

The I/O MEP curve was less steep in patients 'ON medication' than in those 'OFF medication' (Table1). Repeated measures ANOVA showed a significant effect for the main factor 'SESSION' ($F_{1,21}$=10.90, P=0.003) and for the interaction 'SESSION' x 'STIMULUS INTENSITY' ($F_{5,}$

$_{105}$=4.66, P<0.001), indicating higher MEP amplitude values in patients 'OFF' therapy than in patients 'ON' therapy. Lastly, as expected, a significant effect was detected for the main factor 'STIMULUS INTENSITY' ($F_{5,105}$=55.68, P<0.001).

Intracortical excitability: SICI and ICF

There were no clear effects of dopaminergic medication on SICI (Figure 1). Repeated measures ANOVA revealed no statistical significance for the main factor 'SESSION' ($F_{1,21}$=1.18, P=0.28) or for the interaction 'SESSION' x 'ISI' ($F_{1,21}$=0.04, P=0.84). Lastly, the analysis revealed a significant effect for 'ISI' ($F_{1,21}$=7.47, P=0.01), thus confirming the reduced MEP amplitude values at 2ms in comparison to those observed at 4ms ISI. Similarly, there were no effects of dopaminergic medication on ICF (Figure 1) as revealed by the lack of statistical significance for the main factors 'SESSION' ($F_{1,21}$=1.08, P=0.30) and for the interaction 'SESSION' x 'ISI' ($F_{1,21}$=0.009, P=0.92). Lastly, there was no significant effect for 'ISI' ($F_{1,21}$=0.75, P=0.39).

M1 plasticity: PAS-related effects

Dopaminergic medication increased M1 plasticity in PD patients (Figure 2). This was demonstrated by a significant effect of the main factor 'SESSION' ($F_{1,21}$=4.86, P=0.03), with the post-hoc analysis yielding higher values in PD patients 'ON medication' than in those 'OFF medication' (P<0.01). There was also a significant effect of the main factor 'MUSCLE' ($F_{1,21}$=7.10, P=0.01), with higher responses being observed in the APB (target muscle) than in FDI muscle, as well as for the interaction 'SESSION' x 'MUSCLE' ($F_{1,21}$=4.50, P=0.04), as demonstrated by the increase in MEP amplitude after PAS in the 'ON' medication session observed in the ABP though not in the FDI muscle. These findings are in line with previous reports indicating that the effects of dopaminergic medication on PAS are muscle specific (Ueki *et al.*, 2006; Morgante *et al.*, 2006). Lastly, no significant effect was detected for the main factor 'TIME POINT' ($F_{2,42}$=2.48, P=0.09) or for the interactions 'SESSION' x 'TIME POINT' ($F_{2,42}$=0.18, P=0.82), 'MUSCLE' x 'TIME POINT' ($F_{2,42}$=0.03, P=0.96) and 'SESSION' x 'MUSCLE' x 'TIME POINT' ($F_{2,42}$=0.23, P=0.79).

Multiple logistic model

When considering neurophysiological data at baseline, the LASSO procedure confirmed that all the variables which differed between PD patients (OFF medication) and HC were predictors of the disease status (Amplitude intercept:-0.01; Amplitude decrement: -1.55; Velocity Intercept:-0.001; I/O MEP curve slope: 0.31; SICI: 1.16; PAS: -0.009).

When considering neurophysiological data of PD patients 'OFF' and 'ON' medication, the LASSO procedure indicated that three of the variables which differed between PD patients 'OFF' and 'ON' medication were predictors of the medication status, (Amplitude intercept: 0.05; I/O MEP curve slope: -0.34; PAS: 0.013).

Canonical correlation analysis

CCA computes linear combinations of the original kinematic and neurophysiological variables that correlate with each other. The first canonical factor consists of one combination of the original variables, the second and third factors (which are uncorrelated with the first) consists of other combinations of original variables. Note that CCA develops as many canonical factors as there are variables in the smaller of the two variable sets. Wilks' lambda test demonstrated significant effects for two canonical factors (FI=0.68; P=0.01 and FII=0.66; P=0.03 but not for FIII=0.12; P=0.60); these results show that there is an overall relationship between movement kinematics and TMS variables in PD. Table 4 shows the canonical coefficients and the canonical factor loadings for the three canonical factors (FI, FII and FIII). Canonical coefficients correspond to the values in the linear combination that generates the canonical factors from the input variables. The canonical factor loadings indicate the relationship between the canonical factor and the input variables. Amplitude decrement and PAS response had the largest contribution to FI (Table 4, Figure 3). Velocity intercept and the I/O MEP had the largest contribution to FII (Table 4, Figure 3). As further orientation to the relationships between pairs of variables, Table 5 displays the bivariate

correlation matrix and indicates: (i) the slower the movement velocity, the higher the slope of the I/O MEP curve (Figure 4) and (ii) the greater the decrement in amplitude during movement repetition the lower the PAS response (Figure 4).

No relationship emerged in the 'ON' medication state between changes in the kinematic variables of repetitive finger tapping and changes in the excitability and plasticity TMS measures of M1 after dopaminergic medication. This result was confirmed by the CCA: Wilks' lambda test showed no significant effects of canonical factors (all Ps > 0.05).

Lastly, no significant correlations were detected between clinical scores and neurophysiological parameters (all Ps > 0.05).

DISCUSSION

The novel aspect of this study is the correlation analysis we performed between movement kinematics and neurophysiological abnormalities in the M1 of patients with PD. We found that bradykinesia features correlated with M1 excitability and plasticity abnormalities in patients. Dopaminergic therapy improved movement amplitude and speed, though not the sequence effect. Dopaminergic therapy also improved M1 excitability and plasticity, but no correlation was detected with kinematic changes.

Parkinson's disease vs. healthy subjects

Our results confirmed many previous reports in patients with PD. Finger tapping movements had a lower amplitude and velocity in patients than in healthy controls. Movement amplitude also decreased progressively during finger tapping in patients, confirming that the sequence effect is another motor feature of PD (Agostino *et al.*, 2003; Espay *et al.*, 2009 and 2011; Bologna *et al.*, 2016a). We also observed that corticospinal excitability was increased in the resting state and that

M1 plasticity was reduced in PD (Cantello *et al.*, 2002; Currà *et al.*, 2002; Lefaucheur *et al.*, 2005; Berardelli *et al.*, 2008; Bologna *et al.*, 2016b). Unlike some (MacKinnon *et al.*, 2005), but not all (Ridding *et al.*, 1995; Kojovic *et al.*, 2012 and 2015; Ni *et al.*, 2013), previous reports we also observe less effective SICI in patients with PD than in healthy subjects but no changes in ICF.

Correlation between kinematic and neurophysiological abnormalities

To our knowledge, no prior study has assessed the relation between kinematic and neurophysiological abnormalities using CCA in PD. With this approach, we were able to detect a global correlation between these two set of variables, and to identify the most influential kinematic and neurophysiological variables based on their loadings in the analysis. Our analysis demonstrates that neurophysiological abnormalities of M1 are strong predictors of altered movement kinematics in PD. This finding is in line with current models emphasizing the pathophysiological role of M1 in generating movement abnormalities in PD.

Correlates of bradykinesia (movement slowness)

M1 is a principal source of corticospinal input to control of skilled movement. Consistent with this, many previous authors have suggested that dysfunction of M1 contributes to symptoms of bradykinesia in PD (Berardelli *et al.*, 2001). Pasquereau *et al.* (2016) recently confirmed in hemiparkinsonian MPTP monkeys that the resting discharge of corticospinal neurons in M1 was lower than normal and that the correlations between changes in discharge rate and movement parameters such as direction, force and acceleration were all reduced during active movement. They concluded that a general 'hypoactivation' of M1 during movement could contribute to bradykinesia in PD.

At first sight, this data in monkey appears to be opposite to the increased I/O slope of resting corticospinal output that we observe. However, this is not necessarily the case. TMS directly stimulates axons that have synaptic inputs to corticospinal neurons. Given that axonal excitability is

unlikely to be different in PD than normal, increased corticospinal recruitment indicates that these synapses are more effective than in the control state. One possibility is that this may be an adaptation (Blesa *et al.*, 2017) that attempts to boost the power of reduced input from other areas. Indeed the present data, showing that the steeper the I/O slope the slower the movement would be compatible with a gradual recruitment of this mechanism as symptoms progress. It would also be consistent with the absence of any changes in excitability in early PD (Kojovic *et al.*, 2012 and 2015), where abnormalities of voluntary movement velocity are less prominent (Bologna *et al.*, 2016a) and with enhanced excitability in more advanced PD patients (Valls-Solé *et al.*, 1994), where voluntary movement velocity is more severely affected (Bologna *et al.*, 2016a).

An alternative hypothesis is that rather than being compensatory, changing the gain of the inputs to corticospinal neurons alters the relationship between variations in firing rate and movement parameters (Kumar *et al.*, 2010). In this case, as disease progresses, corticospinal excitability increases and bradykinesia deteriorates further.

Correlates of the sequence effect

A second feature of bradykinesia is the sequence effect, a gradual reduction in amplitude and velocity of a repetitive movement. This was not related to the I/O slope, but was negatively correlated with the PAS effect: the greater the decrement in amplitude during movement repetition the smaller the LTP-like effect of PAS. At the present time there is little information on the pathophysiological basis of decrement. On the basis of the present result we speculate that repetitive movement is assisted by short-term facilitation of movement-related synapses in M1. Reduced or absent facilitation in PD, if uncompensated by other mechanisms, might then result in a decline of corticospinal output as movement progresses, resulting in a gradual reduction of movement amplitude. The PAS effect is usually short-lived (15-30 min), and probably depends on short term synaptic effects on synaptic transmission rather than the longer-lasting processes responsible for LTP. If these short term effects employ the same mechanisms as responsible for short term

facilitation it would explain why reduced PAS is accompanied by greater decrement in volitional movements. This explanation would also be consistent with previous findings that short-term synaptic facilitation produced by 5 Hz rTMS is also reduced in PD (Gilio *et al.*, 2002). The hypothesis that reduced M1 plasticity is a possible pathophysiological mechanism of the sequence effect in PD, is supported by the observation that this abnormality is present in early PD (Kang *et al.*, 2011; Lee *et al.*, 2014; Bologna *et al.*, 2016a). Moreover, early synaptic impairment may represent the key event in patients with PD, as shown in TMS studies (Kishore *et al.*, 2012; Kojovic *et al.*, 2012 and 2015) and in both pathogenic and genetic animal models of parkinsonism (Schirinzi *al.*, 2016). In contrast to our findings, which point to a relationship between the sequence effect and M1 plasticity, Kang *et al.* (2010) observed that high-frequency rTMS of M1 did not modify the sequence effect in PD and concluded that M1 is unlikely to be involved in generating this movement abnormality in PD. The authors, however, assessed the sequence effect during the peg-board test, which does not require a fine M1 activation such as that required by repetitive finger tapping (Agostino *et al.*, 2003). Second, Kang *et al*. (2010) based their study on rTMS, which, unlike the PAS protocol we used in this study, does not involve mechanisms of sensorimotor integration (Wolters *et al.*, 2003; Classen *et al.*, 2004; Carson and Kennedy, 2013).

Although we detected a correlation between the sequence effect and M1 plasticity measures, we acknowledge that alternative mechanisms may also contribute to this abnormality in PD. For example, it has been suggested that altered activity in pre-motor areas, basal ganglia or cerebellum may be responsible for the sequence effect in PD (Kang *et al.*, 2010; Little *et al.*, 2012; Tan *et al.*, 2013a, 2013b and 2015; Lee *et al.*, 2014; Steiner *et al.*, 2017)

Acute effect of dopaminergic therapy

We found that dopaminergic medication improved movement amplitude and velocity but not the sequence effect (Espay *et al.*, 2009 and 2011; Bologna *et al.*, 2016a). We also found that dopaminergic replacement normalized M1 excitability, as assessed by the slope of the I/O MEP

curve, and normalized plasticity measures (Bologna *et al.*, 2016b; Suppa *et al.*, 2017). However, there was no correlation between changes in neurophysiology and changes in movement suggesting that abnormalities in performance of movement are not due solely to deficits in M1 but probably involve distributed systems at cortical and subcortical levels. Dopamine could exert acute effects at these sites and improve movement independently of changes in M1 excitability and plasticity. For example, neuroimaging studies have shown that dopaminergic replacement induces changes not only in basal ganglia regional activity but also in pre-motor-M1 and corticostriatal connectivity in patients (Michely *et al.*, 2015). Moreover our results possibly indicate that kinematics and TMS have different sensitivity to change to dopaminergic medication (Suppa *et al.*, 2017). For example, TMS studies indicate non-linear effect of levodopa dosage on PAS-induced plasticity in humans (Monte-Silva *et al.*, 2010; Thirugnanasambandam *et al.*, 2011). It is also possible that some pathophysiological abnormalities in PD (including for example those related to altered GABA-ergic transmission) are not strictly dependent on dopaminergic loss.

Limitations of the study

Unlike electrophysiological recordings in animals or DBS recordings in patients with PD, TMS provides only indirect measures of cortical activity, which may be affected by several sources of variability. Also, it should be borne in mind that this study was performed on a relatively small sample of patients with mild/moderate PD. Since M1 excitability increases and M1 plasticity decreases as the disease worsens (Lefaucheur, 2005; Bologna *et al.*, 2016b), we cannot fully exclude that the relationships we found (either between movement slowness and the slope of the I/O MEP curve as well as between amplitude decrement and PAS response) could perhaps be the result of a common correlation with disease progression. However, this seems unlikely since we found no correlation between neurophysiological data and clinical and demographic features. Further studies on patients in different stages of PD as well as longitudinal studies are needed to investigate intra-individual correlations. This approach would also allow to better understand whether the

electrophysiological changes of M1 reflect compensatory/adaptive changes following the disease onset.

Conclusions

This study provides novel information on the role of M1 in patients with PD and a deeper understanding of the pathophysiological mechanisms that may underlie the various features of bradykinesia. The results support the hypothesis that the various movement abnormalities reflect different pathophysiological mechanisms. Namely, excitability and plasticity changes in M1 may play distinct roles. While M1 excitability changes underlie movement slowness, plasticity changes underlie the sequence effect. Further studies are needed to elucidate how neurophysiological abnormalities of M1 contribute to motor abnormalities in PD. Clarifying this issue is an important step toward the development of novel therapeutic approaches, based on non-invasive brain simulation techniques, targeting the specific neurophysiological abnormalities underlying the various bradykinesia features in PD.

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REFERENCES

Agostino R, Berardelli A, Formica A, Accornero N, Manfredi M. Sequential arm movements in patients with Parkinson's disease, Huntington's disease and dystonia. Brain. 1992;115:1481-95.

Agostino R, Currà A, Giovannelli M, Modugno N, Manfredi M, Berardelli A. Impairment of individual finger movements in Parkinson's disease. Mov Disord. 2003;18:560-565.

Antonini A, Abbruzzese G, Ferini-Strambi L, Tilley B, Huang J, Stebbins GT, et al. Validation of the Italian version of the Movement Disorder Society--Unified Parkinson's Disease Rating Scale. Neurol Sci. 2013;34:683-7.

Beck AT, Ward CH, Mendelson M, Mock J, Erbraugh J. An inventory for measuring depression. Arch Gen Psychiatry. 1961;4:561-71.

Berardelli A, Dick JP, Rothwell JC, Day BL, Marsden CD. Scaling of the size of the first agonist EMG burst during rapid wrist movements in patients with Parkinson's disease. J Neurol Neurosurg Psychiatry. 1986;49:1273-9.

Berardelli A, Rothwell JC, Thompson PD, Hallett M. Pathophysiology of bradykinesia in Parkinson's disease. Brain. 2001;124:2131-46.

Berardelli A, Abbruzzese G, Chen R, Orth M, Ridding MC, Stinear C, et al. Consensus paper on short-interval intracortical inhibition and other transcranial magnetic stimulation intracortical paradigms in movement disorders. Brain Stimul. 2008;1:183-91.

Berardelli A, Wenning GK, Antonini A, Berg D, Bloem BR, Bonifati V, et al. EFNS/MDS-ES/ENS [corrected] recommendations for the diagnosis of Parkinson's disease. Eur J Neurol. 2013;20:16-34.

Blesa J, Trigo-Damas I, Dileone M, Del Rey NL, Hernandez LF, Obeso JA. Compensatory

mechanisms in Parkinson's disease: Circuits adaptations and role in disease modification. Exp Neurol. 2017;298:148-161.

Bologna M, Leodori G, Stirpe P, Paparella G, Colella D, Belvisi D, et al. Bradykinesia in early and advanced Parkinson's disease. J Neurol Sci. 2016a;369:286-91.

Bologna M, Suppa A, Conte A, Latorre A, Rothwell JC, Berardelli A. Are studies of motor cortex plasticity relevant in human patients with Parkinson's disease? Clin Neurophysiol. 2016b;127:50-9.

Cantello R, Tarletti R, Civardi C. Transcranial magnetic stimulation and Parkinson's disease. Brain Res Brain Res Rev. 2002;38:309-27.

Carson RG, Kennedy NC. Modulation of human corticospinal excitability by paired associative stimulation. Front Hum Neurosci. 2013;7:823.

Classen J, Wolters A, Stefan K, Wycislo M, Sandbrink F, Schmidt A, Kunesch E. Paired associative stimulation. Suppl Clin Neurophysiol. 2004;57:563-9.

Currà A, Modugno N, Inghilleri M, Manfredi M, Hallett M, Berardelli A. Transcranial magnetic stimulation techniques in clinical investigation. Neurology. 2002;59:1851-9.

Curran-Everett D. Multiple comparisons: philosophies and illustrations. Am J Physiol Regul Integr Comp Physiol. 2000;279:1-8.

Defer GL, Widner H, Marié RM, Rémy P, Levivier M. Core assessment program for surgical interventional therapies in Parkinson's disease (CAPSIT-PD). MovDisord. 1999;14:572-84.

Dubois B, Slachevsky A, Litvan I, Pillon B. The FAB: a Frontal Assessment Battery at bedside. Neurology. 2000;55:1621-6.

Espay AJ, Beaton DE, Morgante F, Gunraj CA, Lang AE, Chen R. Impairments of speed and amplitude of movement in Parkinson's disease: a pilot study. Mov Disord. 2009;24:1001-8.

Espay AJ, Giuffrida JP, Chen R, Payne M, Mazzella F, Dunn E, et al. Differential response of speed, amplitude, and rhythm to dopaminergic medications in Parkinson's disease. Mov Disord. 2011;26:2504-8.

Friedman JH, Alves G, Hagell P, Marinus J, Marsh L, Martinez-Martin P, et al. Fatigue rating scales critique and recommendations by the Movement Disorders Society task force on rating scales for Parkinson's disease. Mov Disord. 2010;25:805-22.

Gilio F, Currà A, Inghilleri M, Lorenzano C, Manfredi M, Berardelli A. Repetitive magnetic stimulation of cortical motor areas in Parkinson's disease: implications for the pathophysiology of cortical function. Mov Disord. 2002;17:467-73.

Goetz CG, Tilley BC, Shaftman SR, Stebbins GT, Fahn S, Martinez-Martin P, et al.; Movement Disorder Society UPDRS Revision Task Force. Movement Disorder Society-sponsored revision of the Unified Parkinson's Disease Rating Scale (MDS-UPDRS): scale presentation and clinimetric testing results. Mov Disord. 2008;23:2129-70.

Hamada M, Strigaro G, Murase N, Sadnicka A, Galea JM, Edwards MJ, et al. Cerebellar modulation of human associative plasticity. J Physiol. 2012;590:2365-74.

Hasan H, Athauda DS, Foltynie T, Noyce AJ. Technologies Assessing Limb Bradykinesia in Parkinson's Disease. J Parkinsons Dis. 2017;7:65-77.

Heldman DA, Espay AJ, LeWitt PA, Giuffrida JP. Clinician versus machine: reliability and responsiveness of motor endpoints in Parkinson's disease. Parkinsonism Relat Disord. 2014;20:590-5.

Hotelling H. Relations between two set of variates. Biometrika, 1936; 28: 321–77.

Iansek R, Huxham F, McGinley J. The sequence effect and gait festination in Parkinson disease: contributors to freezing of gait? Mov Disord. 2006;21:1419-24.

Kang SY, Wasaka T, Shamim EA, Auh S, Ueki Y, Lopez GJ, et al. Characteristics of the sequence effect in Parkinson's disease. Mov Disord. 2010;25:2148-55.

Kang SY, Wasaka T, Shamim EA, Auh S, Ueki Y, Dang N, et al. The sequence effect in de novo Parkinson's disease. J Mov Disord. 2011 May;4:38-40.

Kawashima S, Ueki Y, Mima T, Fukuyama H, Ojika K, Matsukawa N. Differences in dopaminergic modulation to motor cortical plasticity between Parkinson's disease and multiple system atrophy. PLoS One. 2013;8(5):e62515.

Kishore A, Joseph T, Velayudhan B, Popa T, Meunier S. Early, severe and bilateral loss of LTP and LTD-like plasticity in motor cortex (M1) in de novo Parkinson's disease. Clin Neurophysiol. 2012;123:822-8.

Kishore A, James P, Krishnan S, Yahia-Cherif L, Meunier S, Popa T. Motor cortex plasticity can indicate vulnerability to motor fluctuation and high L-DOPA need in drug-naïve Parkinson's disease. Parkinsonism Relat Disord. 2017;35:55-62.

Kojovic M, Bologna M, Kassavetis P, Murase N, Palomar FJ, Berardelli A, et al. Functional reorganization of sensorimotor cortex in early Parkinson disease. Neurology. 2012;78:1441-8.

Kojovic M, Kassavetis P, Bologna M, Pareés I, Rubio-Agusti I, Berardelli A, et al. Transcranial magnetic stimulation follow-up study in early Parkinson's disease: A decline in compensation with disease progression? MovDisord. 2015;30:1098-106.

Kumar A, Rotter S, Aertsen A. Spiking activity propagation in neuronal networks: reconciling different perspectives on neural coding. Nat Rev Neurosci. 2010;11:615-27.

Lee E, Lee JE, Yoo K, Hong JY, Oh J, Sunwoo MK, Kim JS, Jeong Y, Lee PH, Sohn YH, Kang SY. Neural correlates of progressive reduction of bradykinesia in de novo Parkinson’s disease. Parkinsonism Relat. Disord. 2014;20:1376–81.

Little S, Pogosyan A, Kuhn AA, Brown P. β band stability over time correlates with Parkinsonian rigidity and bradykinesia. Exp Neurol. 2012; 236:383-8.

Lefaucheur JP. Motor cortex dysfunction revealed by cortical excitability studies in Parkinson's disease: influence of antiparkinsonian treatment and cortical stimulation. Clin Neurophysiol. 2005;116:244-53.

MacKinnon CD, Gilley EA, Weis-McNulty A, Simuni T. Pathways mediating abnormal intracortical inhibition in Parkinson's disease. Ann Neurol. 2005;58:516-24.

Michely J, Volz LJ, Barbe MT, Hoffstaedter F, Viswanathan S, Timmermann L, et al. Dopaminergic modulation of motor network dynamics in Parkinson's disease. Brain. 2015;138:664-78.

Möller C, Arai N, Lücke J, Ziemann U. Hysteresis effects on the input-output curve of motor evoked potentials. ClinNeurophysiol. 2009;120:1003-8.

Monte-Silva K, Liebetanz D, Grundey J, Paulus W, Nitsche MA. Dosage-dependent non-linear effect of L-dopa on human motor cortex plasticity. J Physiol. 2010;588:3415-24.

Morgante F, Espay AJ, Gunraj C, Lang AE, Chen R. Motor cortex plasticity in Parkinson's disease and levodopa-induced dyskinesias. Brain. 2006;129:1059-69.

Nasreddine ZS, Phillips NA, Bédirian V, Charbonneau S, Whitehead V, Collin I, et al. The Montreal Cognitive Assessment, MoCA: a brief screening tool for mild cognitive impairment. J Am Geriatr Soc. 2005;53:695-9.

Ni Z, Bahl N, Gunraj C, Mazzella F, Chen R. Increased motor cortical facilitation and decreased inhibition in Parkinson's Disease. Neurology 2013; 80:1746-53.

Pasquereau B, Turner RS. Primary motor cortex of the parkinsonian monkey: differential effects on

the spontaneous activity of pyramidal tract-type neurons. Cereb Cortex 2011;21:1362-78.

Pasquereau B, DeLong MR, Turner RS. Primary motor cortex of the parkinsonian monkey: altered encoding of active movement. Brain. 2016;139:127-43.

Peurala SH, Müller-Dahlhaus JF, Arai N, Ziemann U. Interference of short-interval intracortical inhibition (SICI) and short-interval intracortical facilitation (SICF). Clin Neurophysiol. 2008;119:2291-7.

Postuma RB, Berg D, Stern M, Poewe W, Olanow CW, Oertel W, et al. MDS clinical diagnostic criteria for Parkinson's disease. Mov Disord. 2015;30:1591-601.

Ridding MC, Inzelberg R, Rothwell JC. Changes in excitability of motor cortical circuitry in patients with Parkinson's disease. Ann Neurol. 1995;37:181-8.

Rossi S, Hallett M, Rossini PM, Pascual-Leone A; Safety of TMS Consensus Group. Safety, ethical considerations, and application guidelines for the use of transcranial magnetic stimulation in clinical practice and research. Clin Neurophysiol. 2009;120:2008-39.

Rossini PM, Burke D, Chen R, Cohen LG, Daskalakis Z, Di Iorio R, et al. Non-invasive electrical and magnetic stimulation of the brain, spinal cord, roots and peripheral nerves: Basic principles and procedures for routine clinical and research application. An updated report from an I.F.C.N. Committee. Clin Neurophysiol. 2015;126:1071-107.

Schirinzi T, Madeo G, Martella G, Maltese M, Picconi B, Calabresi P, et al. Early synaptic dysfunction in Parkinson's disease: Insights from animal models. Mov Disord. 2016;31:802-13.

Schwingenschuh P, Ruge D, Edwards MJ, Terranova C, Katschnig P, Carrillo F, et al. Distinguishing SWEDDs patients with asymmetric resting tremor from Parkinson's disease: a clinical and electrophysiological study. Mov Disord. 2010;25:560-9.

Stefan K, Wycislo M, Classen J. Modulation of associative human motor cortical plasticity by attention. J Neurophysiol. 2004;92:66-72.

Steiner LA, Neumann WJ, Staub-Bartelt F, Herz DM, Tan H, Pogosyan A, et al. Subthalamic beta dynamics mirror Parkinsonian bradykinesia months after neurostimulator implantation. Mov Disord. 2017;32:1183-1190.

Suppa A, Marsili L, Belvisi D, Conte A, Iezzi E, Modugno N, et al. Lack of LTP-like plasticity in primary motor cortex in Parkinson's disease. Exp Neurol. 2011;227:296-301.

Suppa A, Bologna M, Conte A, Berardelli A, Fabbrini G. The effect of L-dopa in Parkinson's disease as revealed by neurophysiological studies of motor and sensory functions. Expert Rev Neurother. 2017;17:181-192.

Tan H, Pogosyan A, Anzak A, Foltynie T, Limousin P, Zrinzo L, et al. Frequency specific activity in subthalamic nucleus correlates with hand bradykinesia in Parkinson's disease. Exp Neurol. 2013a. 240:122-9.

Tan H, Pogosyan A, Anzak A, Ashkan K, Bogdanovic M, Green AL, et al. Complementary roles of different oscillatory activities in the subthalamic nucleus in coding motor effort in Parkinsonism. Exp Neurol. 2013b. 248:187-95.

Tan H, Pogosyan A, Ashkan K, Cheeran B, FitzGerald JJ, Green AL, et al. Subthalamic nucleus local field potential activity helps encode motor effort rather than force in parkinsonism. J Neurosci. 2015; 35:5941-9.

Thirugnanasambandam N, Grundey J, Paulus W, Nitsche MA. Dose-dependent nonlinear effect of L-DOPA on paired associative stimulation-induced neuroplasticity in humans. J Neurosci. 2011;31:5294-9.

Tibshirani R. Regression Shrinkage and Selection via the Lasso. Journal of the Royal Statistical

Society. 1996; 58: 267–88.

Tomlinson CL, Stowe R, Patel S, Rick C, Gray R, Clarke CE. Systematic review of levodopa dose equivalency reporting in Parkinson's disease. Mov Disord. 2010;25:2649-53.

Ueki Y, Mima T, Kotb MA, Sawada H, Saiki H, Ikeda A, et al. Altered plasticity of the human motor cortex in Parkinson's disease. Ann Neurol. 2006;59:60-71.

Valls-Solé J, Pascual-Leone A, Brasil-Neto JP, Cammarota A, McShane L, Hallett M. Abnormal facilitation of the response to transcranial magnetic stimulation in patients with Parkinson's disease. Neurology. 1994;44:735-41.

Wolters A, Sandbrink F, Schlottmann A, Kunesch E, Stefan K, Cohen LG, et al. A temporally asymmetric Hebbian rule governing plasticity in the human motor cortex. J Neurophysiol. 2003;89:2339-45.

Wu T, Hallett M, Chan P. Motor automaticity in Parkinson's disease. Neurobiol Dis. 2015;82:226-34.

Xu T, Wang S, Lalchandani RR, Ding JB. Motor learning in animal models of Parkinson's disease: Aberrant synaptic plasticity in the motor cortex. Mov Disord. 2017;32:487-97.