Sleep and Circadian Rhythm Regulation in Early Parkinson Disease

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IMPORTANT: Sleep disturbances are recognized as a common nonmotor complaint in Parkinson disease but their etiology is poorly understood.

OBJECTIVE: To define the sleep and circadian phenotype of patients with early-stage Parkinson disease.

DESIGN, SETTING, AND PARTICIPANTS: Initial assessment of sleep characteristics in a large population-representative incident Parkinson disease cohort (N=239) at the University of Cambridge, England, followed by further comprehensive case-control sleep assessments in a subgroup of these patients (n=30) and matched controls (n=15).

MAIN OUTCOMES AND MEASURES: Sleep diagnoses and sleep architecture based on polysomnography studies, actigraphy assessment, and 24-hour analyses of serum cortisol, melatonin, and peripheral clock gene expression (Bmal1, Per2, and Rev-Erbα).

RESULTS: Subjective sleep complaints were present in almost half of newly diagnosed patients and correlated significantly with poorer quality of life. Patients with Parkinson disease exhibited increased sleep latency (P = .04), reduced sleep efficiency (P = .008), and reduced rapid eye movement sleep (P = .02). In addition, there was a sustained elevation of serum cortisol levels, reduced circulating melatonin levels, and altered Bmal1 expression in patients with Parkinson disease compared with controls.

CONCLUSIONS AND RELEVANCE: Sleep dysfunction seen in early Parkinson disease may reflect a more fundamental pathology in the molecular clock underlying circadian rhythms.
In this study we set out to define the sleep and circadian phenotype of patients with early-stage Parkinson disease.

Methods

We began by studying patients newly diagnosed with PD who were recruited to a community-based incidence study in Cambridgeshire, England (Parkinsonism–Incidence and Cognitive Heterogeneity in Cambridgeshire [PICNICS]). The study was approved by the Cambridgeshire Research Ethics Committee (REC 09/H0308/7) and performed according to the Declaration of Helsinki, with all participants providing written informed consent. Patients completed the Pittsburgh Sleep Quality Index (PSQI), Epworth Sleepiness Scale (ESS), and Parkinson's Disease Questionnaire (PDQ-39) and underwent a range of other assessments (eAppendix in Supplement).

To further investigate sleep dysfunction, we carried out more comprehensive analyses on a subgroup of 30 patients with PD (16 men, 14 women) recruited consecutively from the PICNICS study, as well as 15 healthy age- and sex-matched controls (recruited via local advertising).

Sleep Questionnaires

Sleep complaints were assessed using the Parkinson's Disease Sleep Scale (PDSS), Rapid Eye Movement (REM) Sleep Behavior Disorder Questionnaire (RBDQ-HK), and ESS.

Actigraphy Assessment

Along with keeping sleep diaries, all participants wore an activity monitor (Actiwatch; Cambridge Neurotechnology) on the nondominant wrist continuously during the 14-day period prior to their polysomnography assessment.

Polysomnography

Participants were admitted for 2 consecutive nights of polysomnographic recording based on their habitual bed times (first night was an acclimatization night and not analyzed). Sleep recordings were scored visually by trained raters using the American Academy of Sleep Medicine guidelines.6 See eAppendix in Supplement for more information on the polysomnography protocol, parameters measured, and diagnostic criteria for primary sleep diagnoses.

Circadian Rhythm Analysis

A peripheral venous cannula was inserted prior to the start of sampling at 1 PM. Over the next 24 hours, patients adhered to their habitual bed times and blood was collected through a long catheter to prevent sleep disruption. Subjects remained sedentary apart from bathroom visits. Meal times were fixed and no daytime naps were allowed. Temperature was constant at approximately 21°C. Patients were not strictly shielded from external light but lighting levels were less than 5 lux once lights were turned off.

Serum melatonin and cortisol levels were measured every 90 minutes by enzyme-linked immunosorbent assays (eAppendix in Supplement). Bmal1, Per2, and Rev-Erbα gene expression was reported as a relative ratio to the constitutively expressed nonrhythmic β-actin gene every 3 hours by 1-step real-time quantitative reverse-transcription polymerase chain reactions (eAppendix in Supplement). All samples were analyzed in triplicate and averaged.

Statistical Analysis

Full details of the statistical analyses performed are given in eAppendix in Supplement.

Results

Clinical Characteristics

Two hundred thirty-nine patients newly diagnosed with PD were recruited to the PICNICS study (Table 1). One hundred ninety-two patients (80%) returned the PSQI, 170 patients (71%) returned the ESS, and 206 patients (86%) returned the PDQ-39. Age, sex, Movement Disorders Society–Unified Parkinson’s Disease Rating Scale; PICNICS, Parkinsonism—Incidence and Cognitive Heterogeneity in Cambridgeshire; PSQI, Pittsburgh Sleep Quality Index; PDQ-39, Parkinson’s Disease Questionnaire.

*Twenty-four percent, levodopa; 16%, dopamine agonist; 5%, amantadine, and 4%, rasagiline. Twenty-nine patients (12%) were also taking antidepressants, 10 patients (4%) were taking opiates, 7 patients (3%) were taking benzodiazepines, and 3 patients (1%) were taking zopiclone.

<table>
<thead>
<tr>
<th>Variable</th>
<th>Mean (SD)</th>
</tr>
</thead>
<tbody>
<tr>
<td>Male, %</td>
<td>62</td>
</tr>
<tr>
<td>Age at diagnosis, y</td>
<td>68 (9)</td>
</tr>
<tr>
<td>Time since diagnosis, mo</td>
<td>2.2 (2.9)</td>
</tr>
<tr>
<td>Receiving dopaminergic therapy, %</td>
<td>42*</td>
</tr>
<tr>
<td>LEDD in treated patients, mg</td>
<td>316 (209)</td>
</tr>
<tr>
<td>MDS-UPDRS Part III score</td>
<td>32 (12)</td>
</tr>
<tr>
<td>Hoehn and Yahr score</td>
<td>1.8 (0.8)</td>
</tr>
<tr>
<td>ACE-R score</td>
<td>90 (7)</td>
</tr>
<tr>
<td>Global PSQI score</td>
<td>6.1 (3.9)</td>
</tr>
<tr>
<td>Total PDQ-39 score</td>
<td>26.1 (20.4)</td>
</tr>
</tbody>
</table>

Abbreviations: ACE-R, Addenbrooke’s Cognitive Examination; LEDD, levodopa equivalent daily dose; MDS-UPDRS, Movement Disorders Society–Unified Parkinson’s Disease Rating Scale; PICNICS, Parkinsonism—Incidence and Cognitive Heterogeneity in Cambridgeshire; PSQI, Pittsburgh Sleep Quality Index; PDQ-39, Parkinson’s Disease Questionnaire.
amantadine). Four patients (13%) were taking antidepressants and 2 patients (7%) were taking benzodiazepines.

Sleep Complaints and Quality of Life

In the incident PICNICS cohort, global PSQI scores ranged from 0 to 20, with 94 patients (49%) classified as poor sleepers (global PSQI score greater than 5). Poor sleepers were significantly more likely to have problems with nonmotor activities of daily living, low mood, apathy, and impaired cognition (eTable 2 in Supplement). Moreover, the PSQI score was found to be an independent risk factor for increased PDQ-39 score (β = 0.170, P = .007). In the intensive sleep subgroup, patients with PD had lower total PDSS scores compared with controls (β = −0.373, P = .01), reflecting their greater subjective sleep complaints. Specifically, they scored lower on subscores relating to sleep quality (β = −0.372, P = .01), sleep refreshment (β = −0.422, P = .005), and nocturnal motor impairment (β = −0.408, P = .007). Using the suggested RBDQ-HK cutoff score of 19, 9 patients (30%) and 1 control (7%) reported symptoms suggestive of REM parasomnia. The ESS scores were not significantly different between the groups.

Actigraphy Assessment

Patients with PD had reduced light to dark ratio (β = −0.393, P = .01), as well as higher intradaily variability (β = 0.355, P = .02) showing that they had more fragmented motor activity during the 24-hour period compared with controls. Apart from later sleep onset time (β = 0.316, P = .04), there were no actigraphy differences in nocturnal motor activity between patients with PD and controls (eTable 3 in Supplement).

Primary Sleep Diagnoses

Eight patients with PD had evidence of REM sleep behavior disorder (RBD) (Table 2). Ten patients with PD had periodic limb movements of sleep (defined as a periodic limb movement index greater than 15 per hour) compared with 5 controls. Two patients with PD had central sleep apnea (CSA), while 5 patients and 5 controls had evidence of moderate or severe obstructive sleep apnea (OSA) (moderate = 2 and severe = 3 in both groups). In all cases, individuals had not been diagnosed with these conditions prior to the sleep study. Patients with moderate or severe OSA were more likely to have increased body mass index (unpaired t test, P < .001).

Changes in Sleep Architecture

On detailed polysomnography studies, patients with PD exhibited increased sleep latency (β = 0.333, P = .04), reduced sleep efficiency (β = −0.411, P = .008), increased stage 1 sleep (β = 0.307, P = .03), and reduced REM sleep (β = −0.363, P = .02) (Table 3). Two patients with PD exhibited no REM sleep whatsoever. These aspects of the sleep architecture were similarly affected in patients with and without OSA (eTable 4 in Supplement).

In patients with PD, there was no significant correlation between sleep efficiency and total PDSS score (Pearson correlation, r = 0.156, P = .42). The RBDQ-HK was poor at correctly identifying patients with polysomnography-confirmed RBD (positive predictive value, 33%), but significantly better at correctly excluding the condition (negative predictive value, 76%).

To investigate the effect of dopaminergic medications on sleep architecture, patients with PD were divided into subgroups according to whether they were taking levodopa, dopamine agonists, or neither. Accepting the small subgroup sizes, differences between groups were not statistically significant (eTable 5 in Supplement).

Excessive Daytime Sleepiness

Patients with PD had a tendency towards hypersomnolence compared with controls, considering both mean sleep latency (MSL) (β = −0.259, P = .09) and individual nap opportunities (repeated-measures 2-way analysis of variance, F(1,38) = 3.985, P = .053). All 4 of the patients with severe excessive daytime sleepiness (MSL less than 5 minutes) were in the PD group. No sleep-onset REM episodes were observed. There was a strong correlation between ESS score and reduced MSL (Spearman correlation, r = −0.548, P = .002). Reduced MSL was associated with dopamine agonist use (unpaired t test, P = 0.04).

Table 2. Primary Sleep Diagnoses in Patients With PD vs Controls

<table>
<thead>
<tr>
<th>Primary Sleep Disorder</th>
<th>Patients With PD (n=30)</th>
<th>Controls (n=15)</th>
<th>Univariatea</th>
<th>Multivariateb</th>
</tr>
</thead>
<tbody>
<tr>
<td>RBD</td>
<td>27</td>
<td>0</td>
<td>.04</td>
<td>&gt;.99</td>
</tr>
<tr>
<td>PLMSf</td>
<td>33</td>
<td>33</td>
<td>&gt;.99</td>
<td>.56</td>
</tr>
<tr>
<td>RLSe</td>
<td>3</td>
<td>0</td>
<td>&gt;.99</td>
<td>&gt;.99</td>
</tr>
<tr>
<td>OSAe</td>
<td>17</td>
<td>33</td>
<td>.47</td>
<td>.33</td>
</tr>
<tr>
<td>CSA</td>
<td>7</td>
<td>0</td>
<td>.54</td>
<td>&gt;.99</td>
</tr>
<tr>
<td>EDSd</td>
<td>30</td>
<td>13</td>
<td>.28</td>
<td>.40</td>
</tr>
</tbody>
</table>

Abbreviations: CSA, central sleep apnea; EDS, excessive daytime sleepiness; OSA, obstructive sleep apnea; PD, Parkinson disease; PLMS, periodic limb movements of sleep; RBD, rapid eye movement sleep behavior disorder; RL, restless legs syndrome.

a Fisher exact test used.

b Logistic regression used, adjusting for age and sex (and, in the case of OSA, the effect of body mass index).

c Significant difference at .05 level.

d Periodic limb movement index greater than 15 per hour.

e Moderate or severe OSA (apnea-hypopnea index greater than 15 events per hour).

f Defined as mean sleep latency less than 8 minutes.
Reduced Melatonin Production
The time-dependent variation in melatonin concentration was statistically significant in controls ($F_{16,376} = 1.873, P = .03$) but not in patients with PD ($F_{16,328} = 0.738, P = .47$) (Figure 1A). Patients with PD had a reduced area under the curve ($\beta = -0.341, P = .048$) and a reduced melatonin nadir ($\beta = -0.461, P = .004$) (eTable 6 in Supplement). There was a significant main group effect on melatonin concentration ($F_{1,33} = 4.532, P = .04$). There was no change in the timing of melatonin onset or offset. Cosinor analysis confirmed a robust circadian melatonin profile in the majority of participants—only 5 patients (18%) and 1 control (7%) were arrhythmic—and there was no evidence of a phase shift in patients with rhythmic vs nonrhythmic clock gene expression profiles.

Elevated Serum Cortisol Levels
The time-dependent variation in cortisol concentration was not statistically significant in either group owing to large interindividual variation (Figure 1B). Patients with PD had an increased acrophase ($\beta = 0.502, P = .001$), increased amplitude ($\beta = 0.485, P = .002$), and increased area under the curve ($\beta = 0.615, P < .001$) (eTable 7 in Supplement). There was a significant main group effect on cortisol concentration ($F_{1,30} = 15.720, P < .001$). There was no change in the timing of cortisol onset or offset. Cosinor analysis revealed that 11 patients (41%) and 1 control (7%) had arrhythmic cortisol profiles, and there was no evidence of a cortisol phase shift in patients with PD compared with controls (Mann-Whitney test, $P = .36$). There was no correlation between cortisol and objective sleep measures.

Peripheral Clock Gene Expression Differences
The main finding was a lack of time-dependent variation in Bmal1 expression in patients ($F_{7,175} = 0.794, P = .59$) compared with controls ($F_{7,84} = 2.229, P = .04$) (Figure 2A). Neither Per2 or Rev-Erbα showed statistically significant time-dependent variation in patients or controls, despite visually similar 24-hour expression profiles in both groups (Figure 2B and C). No interaction was found between group and time for any of the 3 genes studied; however, patients with PD did have increased expression of Per2 and Rev-Erbα at 4 AM (unpaired $t$ test, $P = .04$ and $P = .03$, respectively). There was no evidence of a phase shift in Bmal1, Per2, or Rev-Erbα on cosinor analysis (Mann-Whitney test, $P = .86, P = .44$, and $P = .84$, respectively). Overall, few patients showed peripheral clock gene expression profiles that were rhythmic: 2 patients (7%) and no controls (0%) for Bmal1, 3 patients (10%) and 3 controls (20%) for Per2, and 5 patients (22%) and 3 controls (20%) for Rev-Erbα. There were no significant differences in sleep architecture in patients with rhythmic vs nonrhythmic clock gene profiles.
Discussion

In this study, we have confirmed that sleep complaints are common in patients newly diagnosed with PD and correlate significantly with poorer quality of life. We have found that patients with PD have an abnormal sleep macro-architecture including increased sleep latency, reduced sleep efficiency, and reduced

Figure 1. Twenty-Four-Hour Melatonin and Cortisol Rhythms in Patients With Parkinson Disease (PD) vs Controls

These graphs show the mean (SEM) serum melatonin and cortisol concentrations at each time point. In healthy individuals, melatonin levels typically rise in the late evening while cortisol levels peak in the early morning. A, Significant group effect on melatonin concentration on repeated-measures 2-way analysis of variance and lack of a statistically significant time-dependent variation in melatonin concentration over the 24-hour sampling period. Patients with PD also had a reduced area under the curve and a reduced melatonin nadir. There were individual missing melatonin data points in 6 patients with PD (1.1% of total dataset) and 1 control (0.4% of total dataset). B, Significant group effect on cortisol concentration on repeated-measures 2-way analysis of variance. Patients with PD also had an increased acrophase, increased amplitude, and increased area under the curve. There were individual missing cortisol data points in 7 patients with PD (1.3% of total data set) and 4 controls (1.6% of total data set).

Figure 2. Twenty-Four-Hour Clock Gene Expression in Patients With Parkinson Disease (PD) vs Controls

These graphs show the mean (SEM) normalized gene expression levels for the 3 clock genes studied at each time point in peripheral blood mononuclear cells. We sought to investigate whether patients with PD exhibited the same peripheral clock gene expressions oscillations as one would expect in healthy individuals. Loss of the time-dependent variation in Bmal1 was seen in patients with PD over the 24-hour period (A), together with higher expression of Per2 and RevErbα at 4 AM (B and C, respectively). There were individual missing data points in 2 patients with PD (0.8%) and no controls.
REM sleep and that this relates to alterations in relevant circu-
lating hormone profiles. These abnormalities are also linked to
differences in peripheral clock gene expression.

Almost half of our newly diagnosed PD cohort were poor
sleepers according to the PSQI, a validated sleep question-
naire that has been recommended for PD research.9 In the
population-based Norwegian ParkWest study, only 17.8% of pa-
tients with early untreated PD were found to have sleep prob-
lems based on the Neuropsychiatric Inventory sleep subscore.10
Our findings more closely resemble the frequency of sleep
problems reported in prevalent cohorts.11 There was discord-
bance between subjective and objective sleep measures in our
study, suggesting that other factors may influence patients’ per-
ceived sleep quality. For instance, we found that poor sleep-
ers were more likely to exhibit low mood, apathy, and im-
paired cognition.

We found that 8 patients in our intensive sleep subgroup
had RBD, which is not surprising since RBD can predate the
typical motor features of PD by many years.12 In line with other
studies,13 OSA was no more common in nonobese patients with
PD than in the general population. Similarly, CSA was uncom-
mon in idiopathic PD and its presence should raise the possi-
bility of an atypical parkinsonian syndrome such as multiple
system atrophy.14 Finally, we replicated our previous find-
ings, showing that significant excessive daytime sleepiness can
be present from the earliest stages of disease and is associ-
ated with dopamine agonist use.15

Pooled analysis has not been undertaken on previous
case-control studies investigating sleep architecture in PD
owing to differences in methods and parameters measured.16 Bušková and colleagues17 compared 15 patients
with early untreated PD vs 15 matched controls and found
that patients with PD had a tendency for reduced sleep effi-
ciency, increased wakefulness, and reduced REM sleep.
Yong and colleagues18 reported similar findings in their
prevalent cohort and Diederich and colleagues19 concluded
that there was a progressive destructuring of the sleep
architecture in patients with more advanced PD.

We hypothesized that the sleep-wake disturbances in early
PD might reflect a disruption to the neural circuitry control-
ing circadian rhythms. Decline in SCN activity is already be-
lieved to be responsible for reduced melatonin output and
sleep-wake disruption in older healthy adults.20,21 There is evi-
dence from neuropathological22 and imaging23 studies that
the hypothalamus is affected in PD. Moreover, it has been shown
that mice overexpressing α-synuclein exhibit a reduced SCN
firing rate, potentially weakening their ability to communi-
cate neural and hormonal signals from the central clock.24

We sought to investigate this further using hormone and
clock gene assays. We found a sustained elevation of serum
cortisol levels in patients with PD, in line with previous re-
search reporting basal hypercortisolemia in PD.25 We also found
that patients with PD had reduced circulating melatonin lev-
els compared with elderly controls, similar to a previous
study.26 Although Fertl and colleagues27 found no differ-
ces in melatonin output in 9 patients with de novo PD com-
pared with controls, they did report that patients with PD with
a nontremor-dominant phenotype (who may have more ex-
tensive pathology) had lower melatonin levels. Differences in
melatonin output significantly correlated with reduced slow-
wave and REM sleep in our study, which fits well with trials
showing that exogenous melatonin administration increases
both sleep propensity and REM sleep continuity in patients
with PD.28,29

To our knowledge, only 1 previous study has analyzed pe-
ripheral clock gene expression in patients with PD. Cai and
colleagues30 quantified Per1 and Bmal1 expression in leuco-
cytes of 17 patients with PD and 16 controls at 4 overnight time
points. Similar to our findings, they discovered reduced Bmal1
expression compared with controls (but only in drug-naïve pa-
tients). One possible explanation for altered Bmal1 expres-
sion in PD is that dopamine is capable of regulating BMAL1/
CLOCK heterodimer activity,31 meaning that dopamine
deficiency might directly affect this central component of the
molecular clock. Alternatively, damage to the SCN itself could
be responsible for clock gene dysregulation. However, the pre-
cise nature and extent of circadian rhythm disruption in PD
needs to be investigated further because sleep may be crucial
in the clearance of degradation products of neural activity that
accumulate during wakefulness,32 and circadian disruption
may accelerate PD-related pathology.33

One of the main strengths of this study is the recruitment
of a community-based incident cohort of newly diagnosed PD
cases. This is the first study, to our knowledge, to analyze pa-
tients and controls alongside 24-hour hormone and clock gene
rhythms to try and understand the circadian correlates of sleep
dysfunction in PD. We assessed sleep phenotype in a variety
of different ways to probe different aspects of the PD sleep
phenotype.

The major limitation of our study is the fact that the num-roid of participants undergoing comprehensive sleep assess-
ment was relatively small. Ideally, we would have liked to study
all patients over time but this was not practical since many pa-
tients were unwilling to undertake intensive studies of this
nature. It also was not possible to include only unmedicated pa-
tients in an epidemiological study of this type where many
patients start therapy at the time of diagnosis. There were no
significant differences in sleep complaints between those in
the intensive sleep subgroup compared with the PICNICS co-
hort as a whole, nor were there significant differences in sleep
architecture between patients taking different combinations
of dopaminergic drugs. Furthermore, our hormone and clock
gene measurements were based around patients’ habitual bed-
times rather than a free-running environment and were not
carried out at exactly the same time as polysomnography as-
sessments.

Conclusions

In summary, our study has defined the sleep phenotype of an
early-stage PD cohort and related this to the patient’s clinical
characteristics, quality of life, and underlying circadian
rhythms. In doing so, we have provided preliminary evi-
dence that PD-related sleep dysfunction may reflect a more fun-
damental pathology in the circadian system.
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Acquisition, analysis, or interpretation of data: All authors.

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