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# Noradrenergic alterations in Parkinson's disease: a combined <sup>11</sup>C-yohimbine PET/neuromelanin MRI study

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## Abstract

Degeneration of the noradrenergic system is now considered a pathological hallmark of Parkinson's disease but little is known about its consequences in terms of parkinsonian manifestations. Here, we evaluate two aspects of the noradrenergic system using multimodal in vivo imaging in patients with Parkinson's disease and healthy controls: the pigmented cell bodies of the locus coeruleus with neuromelanin sensitive MRI and the density of  $\alpha$ 2-adrenergic receptors (ARs) with PET using [<sup>11</sup>C]yohimbine. Thirty patients with Parkinson's disease and thirty age- and sex-matched healthy control subjects were included. Patient's symptoms characteristics were assessed using the MDS-UPDRS scale. Patients showed reduced neuromelanin signal intensity in the locus coeruleus compared to controls, and diminished [<sup>11</sup>C]yohimbine binding in widespread cortical regions including the motor cortex as well as in the insula, the thalamus and the putamen. Clinically, locus coeruleus neuronal loss was correlated with motor (bradykinesia, motor fluctuations, tremor) and non-motor (fatigue, apathy, constipation) symptoms. A reduction of  $\alpha$ 2-ARs availability in the thalamus was associated with tremor, while a reduction in the putamen, the insula and the superior temporal gyrus was associated with anxiety. These findings highlight a multifaceted alteration of the noradrenergic system in Parkinson's disease since locus coeruleus and  $\alpha$ 2-ARs degenerations were found to be partly uncoupled. These findings raise important issues about noradrenergic dysfunctions that may encourage the search for new drugs targeting this system, including  $\alpha$ 2-ARs, for the treatment of Parkinson's disease.

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14  
15 **Running title:** Noradrenergic parkinsonian alterations

16  
17 **Keywords:** Parkinson's disease; Noradrenaline; PET/MRI; locus coeruleus;  $\alpha$ 2-adrenergic  
18 receptors

19 **Abbreviations:** ARs = adrenergic receptors; BP<sub>ND</sub> = non-displaceable binding potential; CNR =  
20 contrast-to-noise ratio; FSL = FMRIB Software Library; FWE = family-wise error; LC = locus  
21 coeruleus; LEDD = levodopa equivalent daily doses; MDS = Movement Disorder Society; MNI  
22 = Montreal Neurological Institute; MoCA = Montreal Cognitive Assessment; PDQ-39 = 39-item  
23 Parkinson's Disease Questionnaire; ROI = region of interest; TSE = turbo spin-echo; UPDRS =  
24 Unified Parkinson's disease Rating Scale; VBM = voxel-based morphometry

25

# 1 Introduction

2 Parkinson's disease is mostly characterized by the loss of dopaminergic neurons in the substantia  
3 nigra pars compacta. However, the neurodegenerative process also extends to other  
4 neurotransmission systems such as serotonin, or acetylcholine and noradrenaline, which  
5 contribute to motor and non-motor symptoms.<sup>1-4</sup> The modulation and interactions of the complex  
6 interconnected non-dopaminergic networks with dopaminergic circuits are far from being fully  
7 understood, and the cascade of perturbations in these interconnected systems and associated  
8 circuits is still a central issue. Among these neurotransmitters, the noradrenergic system is  
9 probably the most least well known.<sup>5,6-8</sup>

10 Firstly, the direct role of the noradrenergic system in various cognitive functions is well  
11 documented.<sup>9</sup> This includes specific functions like vigilance, attention and executive control, but  
12 also more transversal functions like learning, cognitive flexibility and working memory,<sup>8</sup> or an  
13 even more general role in adaptive adjustments in gain that serve to optimize performance.<sup>10</sup> It is  
14 important to point out that there is a major gap in our understanding of how these modulations  
15 apply to the motor circuits.<sup>11</sup> The alteration of these functions might thus contribute to various  
16 parkinsonian manifestations.<sup>8</sup> However, it is often under-recognized and its association with  
17 specific symptoms, in particular with the motor manifestations of the disease, is still unclear.

18 Secondly, the effect of noradrenergic dysfunction in the manifestations of the disease might also  
19 be related to its antiparkinsonian and neuroprotective properties.<sup>12</sup> The loss of noradrenergic  
20 neurons in the locus coeruleus (LC), the sole source of noradrenaline to the neocortex,  
21 hippocampus, cerebellum and thalamus (<sup>13</sup> for review) which is estimated between 20 and 90%,  
22 precedes and exceeds the characteristic loss of dopaminergic neurons in the substantia nigra.<sup>14</sup>  
23 There is evidence mainly based on animal studies that noradrenergic loss may enhance  
24 neurotoxic damage to nigrostriatal dopaminergic neurons,<sup>15,16</sup> and that, conversely, restoration of  
25 LC damaged system positively influences the recovery process of degenerated dopaminergic  
26 neurons.<sup>17,18</sup> Taken together, these observations suggest that enhancing noradrenergic  
27 neurotransmission may facilitate both specific functions affected by the disease and the recovery  
28 process of dopaminergic neurons.<sup>19</sup> Yet, therapeutic strategies targeting this system in  
29 Parkinson's disease are still currently limited.<sup>7,20</sup>

1 The use of  $\alpha 2$ -adrenergic receptors (ARs) antagonists has recently been considered as a major  
2 possible target<sup>21</sup> as it might potentiate noradrenaline availability by blocking presynaptic  $\alpha 2$ -ARs  
3 whose normal function is to regulate noradrenaline release.<sup>22-24</sup> However, direct *in vivo* evidence  
4 of alteration of  $\alpha 2$ -ARs in parkinsonian manifestations is still lacking. This critical lack of  
5 knowledge, accompanied by substantial controversies on the neurofunctional bases of the  
6 noradrenergic system and its alteration in Parkinson's disease,<sup>25,26</sup> is mainly due to the lack of  
7 specific *in vivo* molecular imaging tools in humans. To date, *in vivo* imaging studies have only  
8 used presynaptic noradrenergic tracers of the noradrenergic transporter with [<sup>11</sup>C]MeNER. These  
9 studies have shown reduced tracer uptake, suggesting a diminished density of noradrenergic  
10 terminals, mainly in the thalamus, hypothalamus, LC and raphe nuclei.<sup>2,27</sup> Regarding  $\alpha 2$ -ARs,  
11 most of our knowledge derives from animal studies or post-mortem samples of human brains.<sup>28,29</sup>  
12 Autoradiography studies suggest an alteration in  $\alpha 2$ -ARs in different brain regions (thalamus,<sup>30</sup>  
13 cerebellum,<sup>31</sup> hypothalamus,<sup>32</sup> and prefrontal cortex<sup>33</sup>). Here, we take advantage of recent  
14 methodological developments allowing *in vivo* imaging of the noradrenergic system in human  
15 with the novel PET radiotracer [<sup>11</sup>C]yohimbine,<sup>34-36</sup> now available for use in large samples of  
16 human subjects to map differences in  $\alpha 2$ -ARs distribution between parkinsonian patients and  
17 matched controls. The [<sup>11</sup>C]yohimbine binds with high selectivity to all  $\alpha 2$ -ARs subtypes.<sup>37</sup> It is  
18 displaced when there is competition at the receptor with endogenous noradrenaline, as shown by  
19 preclinical studies reporting reduced receptor binding by unlabeled yohimbine challenge,  
20 amphetamine administration or acute vagus nerve stimulation in preclinical studies<sup>37-39</sup> and  
21 human studies reporting increased receptor binding after clonidine administration.<sup>34</sup>

22 The objectives of the present study are to assess two aspects of noradrenergic neurodegeneration  
23 in Parkinson's disease by means of hybrid MRI/PET imaging: lesions of pigmented cell bodies  
24 of the LC with neuromelanin-sensitive MRI and alterations of  $\alpha 2$ -ARs density with  
25 [<sup>11</sup>C]yohimbine PET. Assessing simultaneously noradrenergic terminals (with PET) and LC cell  
26 bodies (with MRI) is of primary importance given that their respective deficits may be  
27 uncoupled.<sup>40</sup> To inform the disputed pathophysiological mechanisms of the disease, correlations  
28 between the observed noradrenergic dysfunctions and the standard clinical measures of  
29 Parkinson's disease have been systematically tested.

30

# 1 **Materials and methods**

## 2 **Participants**

3 The demographics and clinical characteristics of the participants are listed in Table 1. We  
4 enrolled 30 Parkinson's disease patients and 30 healthy controls matched for age, gender, and  
5 global cognitive functioning using the Montreal Cognitive Assessment (MoCA) score. Patients  
6 were diagnosed with Parkinson's disease for at least 1 year in accordance with the Movement  
7 Disorder Society criteria.<sup>41</sup> The Movement Disorder Society Revised Unified Parkinson's  
8 Disease Rating Scale (MDS-UPDRS) was used to assess Parkinson's disease symptoms,  
9 including separate assessments of non-motor symptoms (UPDRS-I), daily activity (UPDRS-II),  
10 motor symptoms (UPDRS-III), and motor complications (UPDRS-IV).<sup>42</sup> In addition, the 39-item  
11 Parkinson's Disease Questionnaire (PDQ-39) was used to evaluate quality of life.<sup>43</sup> All patients  
12 were assessed in the "ON" state with their usual antiparkinsonian medication. Levodopa  
13 equivalent daily dose (LEDD) was calculated according to previously published conversion  
14 rules.<sup>44</sup> Most of the patients with PD were in the early and middle stage of their disease with a  
15 mean Hoehn & Yahr score of 1.8 and MDS-UPDRS-III score in the "on" state of 17.7 ( $\pm$ 12.3).  
16 Exclusion criteria were medications that interfere with the noradrenergic system, a diagnosis of  
17 other neurological or psychiatric disorders, and the presence of major dyskinesia or tremor in the  
18 patients for technical imaging purposes. The study was conducted in accordance with the  
19 Declaration of Helsinki and approved by the local Ethical Committee in Biomedical Research  
20 (N° CPP 19\_01\_02/N°EudraCT 2018-003999-13). Written informed consent was obtained from  
21 all the subjects before the study.

## 22 **Magnetic Resonance Imaging**

23 MRI data were acquired on a Biograph mMR (hybrid MRI/PET) including a whole brain high-  
24 resolution anatomical 3D T1-weighted MPRAGE sequence in the sagittal plane (matrix size 256  
25  $\times$  256  $\times$  176, voxel size 1  $\times$  1  $\times$  1 mm<sup>3</sup>), and a 2D axial turbo spin-echo (TSE) sequence (in plane  
26 resolution 0.4  $\times$  0.4 mm<sup>2</sup>, slice thickness 3 mm, matrix size 464  $\times$  512  $\times$  15) which provided a  
27 neuromelanin-sensitive MRI.

## 1 **Voxel-based morphometry analysis**

2 To exclude the possibility that structural degeneration affects the results of [<sup>11</sup>C]yohimbine  
3 binding, voxel-based morphometry (VBM) analysis was conducted. Standard VBM  
4 preprocessing protocol of the Computational Anatomy Toolbox (CAT12, [http://www.neuro.uni-](http://www.neuro.uni-jena.de/cat/)  
5 [jena.de/cat/](http://www.neuro.uni-jena.de/cat/)) running on Statistical Parametric Mapping (SPM12; Welcome Trust Centre for  
6 Neuroimaging, University College, London, UK) was employed. Briefly, each 3D T1-weighted  
7 image was bias-corrected and segmented into grey matter, white matter and cerebrospinal fluid  
8 tissue classes using SPM's unified segmentation function. The grey matter segmentation  
9 probability maps were spatially transformed to the Montreal Neurological Institute (MNI)  
10 standard space using the ICBM-152 template according to the DARTEL approach with default  
11 settings in 1.5 mm cubic resolution, and then smoothed with a Gaussian kernel of 8 mm full-  
12 width-at-half-maximum. Lastly, to correct for individual differences in brain size, the Total  
13 Intracranial Volume (TIV) was estimated for each subject using CAT12 interface (Statistical  
14 Analyses – Estimate TIV), as the sum of the grey matter, white matter, and CSF volumes.

## 15 **Neuromelanin-sensitive MRI analysis**

16 To calculate signal intensity in the LC, an approach similar to that published by García-Lorenzo  
17 and colleagues<sup>6</sup> was implemented. First, two regions were manually drawn on the T1 MRI high  
18 resolution MNI template using FSLeyes (FSL v5.0.11, FMRIB, Oxford, UK). These two regions  
19 (one for each side) were defined as large 3D bounding boxes and placed around the hyperintense  
20 voxels at the lateral border of the fourth ventricle so that they included the LC, but excluded the  
21 substantia nigra which also contains neuromelanin.<sup>45</sup> In addition, we used the central reference  
22 mask freely available to normalize the intensity of the LC.<sup>46</sup> These three regions were then  
23 wrapped and resampled onto the subject's neuromelanin-sensitive T1 image using SPM12. Using  
24 FSL functions, the LC was defined as the area of the 10 voxels with the brightest intensity within  
25 each bounding box. We considered the intensity of the LC as the average of the intensities of the  
26 10 voxel region (Fig. 1). The contrast-to-noise ratio (CNR) was calculated for each LC (right and  
27 left) as  $CNR_{LC} = (S_{LC} - S_{REF}) / S_{REF}$  where  $S_{LC}$  and  $S_{REF}$  correspond to the mean signal intensity of  
28 the LC and the reference region respectively. Normalization of the LC signal to a central  
29 reference region (pons) to calculate the CR by relative difference has been the most commonly  
30 used and recommended method in previous studies.<sup>47</sup>

## 1 **[<sup>11</sup>C]yohimbine PET**

2 Radiosynthesis of [<sup>11</sup>C]yohimbine was performed as previously described.<sup>37</sup> The radiochemical  
3 purities of syntheses used for the study were greater than 95%, with molar activities of  $85 \pm 30$   
4 GBq/ $\mu$ mol at the end of synthesis. All subjects received an intravenous bolus injection of 370  
5 MBq  $\pm$  10% of [<sup>11</sup>C]yohimbine. List-mode PET data were acquired during the 90 min from the  
6 injection of the tracer. MRI data were acquired simultaneously.

7 Raw PET data were corrected for motion,<sup>48</sup> and then rebinned into 24 time frames (variable  
8 length frames,  $8 \times 15$  s,  $3 \times 60$  s,  $5 \times 120$  s,  $1 \times 300$  s,  $7 \times 600$  s) sinograms for dynamic  
9 reconstruction. Sinograms were corrected for scatter, randoms, normalization, and attenuation.<sup>49</sup>  
10 Images were reconstructed using 3D ordinary Poisson-ordered subsets expectation maximization  
11 (OP-OSEM 3D), incorporating the system point spread function using 3 iterations of 21 subsets.  
12 Reconstructions were performed with a zoom of 2 in a matrix of  $172 \times 172$  voxels, yielding a  
13 voxel size of  $2.03 \times 2.03 \times 2.08$  mm<sup>3</sup>, with a 4 mm 3D post-reconstruction Gaussian filtering.  
14 Regional time activity curves were extracted based on labelling of structural brain regions  
15 obtained with the multi-atlas propagation with enhanced registration methodology (MAPER)<sup>50</sup>  
16 and the 83-region Hammers atlas.<sup>51</sup> [<sup>11</sup>C]yohimbine non-displaceable binding potential (BP<sub>ND</sub>)  
17 parametric maps were then generated using the simplified reference tissue model (SRTM), with  
18 the corpus callosum as reference region.<sup>34</sup> PET parametric images were spatially normalized to  
19 the MNI space with parameters derived from the individual T1 MRI.

## 20 **Statistics**

21 Statistical analyses on demographic data were performed using Rstudio  
22 (<https://github.com/rstudio/rstudio>). All data were tested for normal distribution using the  
23 Shapiro-Wilk test and the Levene test for homogeneity of variance. Independent samples t-tests  
24 or Wilcoxon tests, as appropriate, were used to compare demographics group mean differences.  
25 We assessed group differences in the CNR<sub>LC</sub> with an analysis of covariance accounting for age  
26 and sex as covariates. A two-tailed t-test was generated in the SPM12 toolbox to assess between-  
27 group differences on grey matter concentration using family-wise error (FWE) correction with  $p$   
28  $< 0.05$  threshold. The extend threshold was set to 100 voxels and total intracranial volume was  
29 used as covariate of no interest.

1 SPM12 in Matlab2020a was used for between-group comparisons of [<sup>11</sup>C]yohimbine BP<sub>ND</sub> at the  
2 voxel level (2-sample t-test) controlling for age and gender. Spatially normalized parametric  
3 images were smoothed with an 8-mm Gaussian filter, and an average cortical grey-matter mask  
4 was used for explicit masking within the SPM software. The voxel-level analysis threshold was  $p$   
5  $< 0.001$  (uncorrected), and subsequently a FWE cluster-level correction at  $p < 0.05$  was applied.

6 We assessed the association of MRI and PET signals and interrogated nonparametric (partial)  
7 correlations between CNR<sub>LC</sub> and [<sup>11</sup>C]yohimbine BP<sub>ND</sub> values extracted from the ROIs,  
8 employing Spearman correlation test adjusted for age and sex.

9 We assessed the links between both noradrenergic markers and specific clinical characteristics  
10 within our group of Parkinson's disease patients by means of partial correlation coefficients  
11 adjusted for age and sex. Indeed, it is known that both variables influence the phenotypical  
12 expression of Parkinson's disease,<sup>52</sup> that healthy ageing influences neuromelanin accumulation  
13 in the LC,<sup>53</sup> and that [<sup>11</sup>C]yohimbine BP<sub>ND</sub> might be influenced by gender.<sup>36</sup> All correlations  
14 were two-sided and significance was set at  $p < 0.05$ .

15

## 16 **Results**

### 17 **Altered LC integrity in patients with Parkinson's disease**

18 Parkinson's disease patients had lower CNR<sub>LC</sub> than healthy controls ( $0.29 \pm 0.04$  vs  $0.31 \pm 0.04$ ,  
19 respectively;  $F(1, 99) = 5.72$ ,  $p = 0.019$ ). CNR was higher in the left than the right LC ( $0.32 \pm$   
20  $0.03$  versus  $0.28 \pm 0.04$ , respectively;  $F(1,99) = 22.02$ ,  $p < 0.001$ ). No significant effect of  
21 gender and no interaction were found ( $p > 0.3$ ). Because of the side effect, all other analyses  
22 have been conducted for both sides separately.

### 23 **Reduced [<sup>11</sup>C]yohimbine binding in Parkinson's disease patients**

24 Reduced binding of [<sup>11</sup>C]yohimbine was observed in the Parkinson's disease group compared to  
25 healthy controls in four significant clusters including mainly cortical regions across all lobes and  
26 the insula, the putamen and the thalamus (Table 2, Fig. 2). VBM was additionally employed to  
27 assess grey matter volume loss in Parkinson's disease patients compared to controls as a

1 potential confounding factor within the ROI. No significant effect was found between the two  
2 groups using FWE with corrected  $p$  value  $< 0.05$  in the t-test.

### 3 **Correlation between $\alpha 2$ -ARs availability and LC MRI data**

4 No significant correlation was found between regional [ $^{11}\text{C}$ ]yohimbine  $\text{BP}_{\text{ND}}$  values and  $\text{CNR}_{\text{LC}}$   
5 (all  $p > 0.1$ ).

### 6 **Correlation of neuromelanin MRI data with clinical scores in Parkinson's** 7 **disease**

#### 8 **Clinical characteristics**

9  $\text{CNR}_{\text{LC}}$  correlated negatively with LEDD (right:  $r = -0.52$ ,  $p = 0.011$  and left:  $r = -0.49$ ,  $p =$   
10  $0.017$ ).

#### 11 **Non-motor scores**

12  $\text{CNR}_{\text{LC}}$  correlated negatively with UPDRS-I total score (right:  $r = -0.43$ ,  $p = 0.035$  and left:  $r = -$   
13  $0.47$ ,  $p = 0.02$ ). We explored within this scale which items mostly driven this effect and found  
14 that apathy sub-score was negatively correlated with the right and left  $\text{CNR}_{\text{LC}}$  ( $r = -0.48$ ,  $p =$   
15  $0.018$  and  $r = -0.46$ ,  $p = 0.025$  respectively), constipation and light headedness on standing sub-  
16 scores correlated negatively with the left  $\text{CNR}_{\text{LC}}$  ( $r = -0.52$ ,  $p = 0.008$  and  $r = -0.47$ ,  $p = 0.02$   
17 respectively), while the fatigue sub-score showed significant negative correlation with the right  
18  $\text{CNR}_{\text{LC}}$  ( $r = -0.48$ ,  $p = 0.017$ ).

#### 19 **Motor scores**

20 Although we did not observe any significant correlation between  $\text{CNR}_{\text{LC}}$  and UPDRS-II and -III  
21 total scores (all  $p > 0.1$ ), we also specifically explored the relationship between  $\text{CNR}_{\text{LC}}$  and rest  
22 tremor, rigidity and bradykinesia. Those were calculated using the following items for MDS-  
23 UPDRS-III: rest tremor - 3.17, rigidity - 3.3, bradykinesia - 3.4-3.8. Interestingly, the left  $\text{CNR}_{\text{LC}}$   
24 correlated negatively with the bradykinesia sub-score ( $r = -0.42$ ,  $p = 0.043$ ) while a trend in the  
25 right side was also observed ( $r = -0.39$ ,  $p = 0.056$ ), and the right  $\text{CNR}_{\text{LC}}$  correlated positively  
26 with the tremor sub-score ( $r = 0.45$ ,  $p = 0.026$ ). Finally, we also found that the left  $\text{CNR}_{\text{LC}}$   
27 correlated negatively with the UPDRS-IV score ( $r = -0.44$ ,  $p = 0.03$ ) with a trend in the right side  
28 ( $r = -0.38$ ,  $p = 0.069$ ). Interestingly, this effect was mostly driven by the motor fluctuation sub-

1 score (sum of items 4.3 and 4.4) which showed significant negative correlation with the left and  
2 right CNR<sub>LC</sub> ( $r = -0.43$ ,  $p = 0.035$  and  $r = -0.41$ ,  $p = 0.044$  respectively) while such correlations  
3 were not observed for the dyskinesia sub-score (sum of items 4.1 and 4.2) (all  $p > 0.1$ ).

#### 4 **Correlation of $\alpha 2$ -ARs PET data with clinical scores in Parkinson's disease**

5 We found no correlation between [<sup>11</sup>C]yohimbine BP<sub>ND</sub> values in the ROI derived from the  
6 between-group SPM contrast and the total scores of the MDS-UPDRS scales. However,  
7 significant correlations were found with sub-scores within the different sections of the scale.

#### 8 **Non-motor scores**

9 The item assessing anxiety in the UPDRS-I part correlated negatively with [<sup>11</sup>C]yohimbine BP<sub>ND</sub>  
10 values in three regions within the right hemisphere : the putamen ( $r = -0.50$ ,  $p = 0.008$ ), the  
11 insula ( $r = -0.44$ ,  $p = 0.02$ ) and the superior temporal gyrus ( $r = -0.41$ ,  $p = 0.034$ ).

#### 12 **Motor scores**

13 The item assessing rest tremor in the UPDRS-III part correlated negatively with [<sup>11</sup>C]yohimbine  
14 BP<sub>ND</sub> values in the right thalamus ( $r = -0.46$ ,  $p = 0.015$ ). In other words, the lower the binding in  
15 the thalamus, the more the patients suffered from tremor.

#### 16 **Discussion**

17 In the present study, we investigated two aspects of the noradrenergic system using multimodal  
18 *in vivo* imaging in Parkinson's disease patients and healthy controls with hybrid MRI/PET: the  
19 density of pigmented cell bodies of the LC with neuromelanin sensitive MRI and the availability  
20 of the  $\alpha 2$ -ARs with [<sup>11</sup>C]yohimbine PET. Both show marked reductions in signal intensity in  
21 patients, confirming that noradrenergic damage is profound and widespread in Parkinson's  
22 disease. Clinical, behavioral and anatomical data suggest, however, that noradrenergic  
23 dysfunction is multifaceted and may involve different circuits underlying specific motor and  
24 non-motor symptoms.

#### 25 **Noradrenergic system integrity in Parkinson's disease**

26 The reduction in signal intensity within the LC in Parkinson's disease patients confirms previous  
27 MRI studies<sup>2,40,54-57</sup> and provides a robust and replicable *in vivo* indication of the loss of

1 pigmented LC neurons previously suggested in post-mortem histological studies.<sup>58–60</sup> On the  
2 contrary, there was only very sparse evidence about the loss of  $\alpha 2$ -ARs in Parkinson's disease  
3 patients, including only one *in vitro* autoradiography study reporting a decrease of density in the  
4 prefrontal cortex<sup>33</sup> and one preliminary *in vivo* study using [<sup>11</sup>C]yohimbine in a small group of  
5 PD patients reporting a global reduction in [<sup>11</sup>C]yohimbine distribution volume in the temporal,  
6 parietal and occipital cortices, the insula and the cingulate gyrus.<sup>61</sup> Our data provide a precise  
7 mapping of [<sup>11</sup>C]yohimbine binding reduction in Parkinson's disease patients (Fig. 2). Within the  
8 parietal lobe, the angular gyrus, the superior parietal lobule and the postcentral gyrus are  
9 especially affected. The occipital cortex also shows multiple sources of binding differences both  
10 in the primary and associative visual cortices. In the frontal lobe, only motor structures (primary  
11 motor cortex, supplementary motor area and premotor cortex) and the inferior frontal gyrus  
12 reveal binding reduction in patients. The temporal lobe seems also relatively preserved since  
13 only one region in the superior temporal gyrus shows group differences. Medially, the posterior  
14 cingulate/precuneus region is particularly affected. Finally, binding reduction in Parkinson's  
15 disease also concerns the insula, the putamen and the thalamus (Fig. 2). This latter observation is  
16 in good agreement with previous post-mortem and animal studies showing severe reductions of  
17 noradrenaline levels in almost all subregions of the thalamus.<sup>30,62</sup>

18 Interpreting [<sup>11</sup>C]yohimbine binding reduction is not straightforward. Indeed, as  $\alpha 2$ -ARs are  
19 located both pre- and post-synaptically,<sup>63,64</sup> the decrease in [<sup>11</sup>C]yohimbine binding could reflect  
20 both a reduction of presynaptic  $\alpha 2$ -ARs due to the diminished innervation caused by the  
21 degeneration of noradrenergic neurons, and/or a reduction in the availability of postsynaptic  $\alpha 2$ -  
22 ARs which can be relatively independent of the degeneration of noradrenergic neurons (as  
23 observed for the dopaminergic system).<sup>65</sup> The former hypothesis predicts strong correlations  
24 between neuromelanin MRI and [<sup>11</sup>C]yohimbine PET signals. Our results are not consistent with  
25 this prediction. They are rather reminiscent of the conclusion obtained from [<sup>11</sup>C]MeNER -a  
26 marker of noradrenaline transporter availability- PET studies that deficits in noradrenergic  
27 terminals and LC cell bodies are partly uncoupled.<sup>2,40</sup> It seems that the number of noradrenergic  
28 cells in the LC would be a major determinant of  $\alpha 2$ -AR density only in the LC itself.<sup>66</sup> Our data  
29 are in line with the overall hypothesis according to which monoaminergic receptors undergo  
30 adaptive changes in Parkinson's disease that make the availability of their receptors non  
31 proportional to the extent of neuronal death of the corresponding cell bodies.<sup>65,67</sup> It is therefore

1 essential to disentangle, within the global clinical picture, the symptoms that might be more  
2 related to the loss of  $\alpha 2$ -ARs and/or to the loss of noradrenergic cells in the LC.

### 3 **Non-motor symptoms**

#### 4 **Fatigue**

5 Fatigue was found to be associated with LC neuronal loss. This is in line with the idea that  
6 fatigue results from disruption of nondopaminergic pathways, including the noradrenergic  
7 system.<sup>68-70</sup> This is not incompatible with the hypothesis that fatigue in Parkinson's disease is  
8 also associated with serotonergic dysfunction in prefrontal-basal ganglia and limbic circuits.<sup>71</sup>  
9 Indeed, it is well known that there are strong interactions between the noradrenergic and  
10 serotonergic systems (mainly through the LC-dorsal raphe nucleus connection)<sup>72-74</sup> suggesting  
11 that both are likely to contribute to the appearance of the symptom. However, the argument that  
12 fatigue characterizes a specific serotonergic phenotype of Parkinson's disease is based on  
13 associations between fatigue and other non-motor symptoms related to the degeneration of  
14 serotonergic pathways (apathy, anxiety, sleep problems and daytime sleepiness)<sup>75</sup> that are most  
15 often moderate<sup>69</sup> and that are not all reported in the present study as only apathy was also found  
16 to be associated with LC neuronal loss. Anxiety was just associated with a reduction of  $\alpha 2$ -ARs  
17 availability in the putamen, the insula and the superior temporal gyrus, but not with a reduction  
18 in signal intensity within the LC. Yet, neither LC neuronal loss nor  $\alpha 2$ -ARs availability were  
19 found to account for sleep disorders. This observation is in line with several pharmacological  
20 studies of physical exercise in healthy humans suggesting that the role of the serotonergic  
21 system in central fatigue might be overestimated<sup>76</sup> while the role of the noradrenergic system  
22 might conversely be underestimated.<sup>77</sup> In addition, given that fatigue is a non-dopaminergic  
23 symptom, the fact that Methylphenidate -a reuptake inhibitor of dopamine and noradrenaline- is  
24 proposed as a treatment against fatigue in Parkinson's disease<sup>78,79</sup> is also consistent with the idea  
25 that the pathophysiological mechanism leading to fatigue involves the degeneration of the  
26 noradrenergic system. Beyond Parkinson's disease, the association of fatigue with a loss of  
27 noradrenergic cells in the LC but not with a loss of  $\alpha 2$ -ARs in other specific brain regions is not  
28 trivial. It relates the global perception of an internal state of energy to the general function of the  
29 noradrenergic system which is to mobilize the whole brain and body for action from a unique but

1 widely distributed source. This association is not put forward in current biological theories of  
2 fatigue.<sup>69</sup>

### 3 **Apathy**

4 Apathy is a complex and multifactorial alteration of the internal and external drives of goal-  
5 directed behavior.<sup>80</sup> It has been associated with dysfunctions of different neural networks  
6 supporting emotional/affective, cognitive and auto-activation circuits,<sup>3,81,82</sup> for which meso-  
7 cortico-limbic dopaminergic and serotonergic lesions are thought to play a major role. The  
8 present study provides new insight into apathy pathophysiology and suggests that there is a role  
9 of noradrenergic alteration since we found a correlation between apathy and LC neuronal loss.  
10 This observation is reminiscent of a former recent study using pharmacological MRI and testing  
11 more specifically predictive processing (i.e., executive functions) of goal-directed behavior.<sup>83</sup>  
12 Although we cannot disentangle cognitive from motivational apathy in the present study, it is  
13 tempting to speculate that the noradrenergic dysfunction identified in our empirical observations  
14 may be more closely associated with cognitive apathy (which includes dysfunction of a set of  
15 executive mechanisms required for achieving a goal) than with motivational apathy.<sup>84</sup> Our results  
16 also strengthen the idea that fatigue is associated with apathy (both relying on the general  
17 activation function of the LC-NA system, not on specific losses of  $\alpha 2$ -ARs) but not with  
18 anxiety<sup>85</sup> (which does not involve LC neuronal loss in the present study).

### 19 **Anxiety**

20 Anxiety is commonly associated with apathy and depression to constitute a behavioral « non-  
21 motor triad » in Parkinson's disease governed by serotonergic and dopaminergic  
22 degeneration.<sup>3,86</sup> However, while anxiety symptoms are clearly linked to alterations of limbic  
23 cortico-striato-thalamo-cortical circuits, several studies have proposed that basal ganglia and  
24 brainstem nuclei could be at the root of such disorders,<sup>87,88</sup> including a loss of noradrenaline  
25 innervation in the locus coeruleus and the limbic system.<sup>87,89</sup> Our data are consistent with the  
26 involvement of noradrenergic dysfunction in anxiety as its severity is associated with a reduction  
27 of  $\alpha 2$ -ARs availability in a brain network including the putamen, the insula and the superior  
28 temporal gyrus (but not with a loss of noradrenergic cells in the LC). However, we found no  
29 evidence for a common noradrenergic dysfunction of the “non-motor triad” as they all seem to

1 have different origins: anxiety was associated with alteration of noradrenergic terminals, apathy  
2 with alteration of LC cell bodies, while depression was not linked to any of these dysfunctions.

### 3 **Constipation**

4 Constipation is one of the most common nonmotor symptoms that emerge from the very early  
5 phase.<sup>90,91</sup> The underlying mechanisms of constipation in Parkinson's disease are still unknown,  
6 but neurodegeneration of autonomic centers of both the enteric and central nervous systems are  
7 likely.<sup>92</sup> Our observation that constipation is associated with a reduction of signal intensity in the  
8 LC fits with the general idea that dysfunction of the noradrenergic system is closely linked to  
9 numerous premotor symptoms of Parkinson's disease.<sup>93-96</sup> But most importantly, it is also  
10 consistent with rodent studies providing specific evidence that: 1) the LC participates in the  
11 regulation of colorectal motility (via activation of  $\alpha 1$ -ARs in the lumbosacral defecation  
12 center),<sup>97</sup> 2) noradrenaline plays a major role in regulating colon immune homeostasis,<sup>98</sup> and 3)  
13 the reduction of noradrenaline level in the intestine causes colon inflammation,  $\alpha$ -synuclein  
14 pathology, neuronal loss and, in fine, constipation.<sup>98,99</sup>

### 15 **Motor symptoms**

16 LC neuronal loss in Parkinson's disease has been associated with the disease, but not directly  
17 with motor symptoms.<sup>6,100</sup> Here, we provide evidence for a direct link with bradykinesia, rest  
18 tremor, and motor fluctuations. By contrast, while noradrenergic denervation of the motor  
19 areas<sup>101</sup> and noradrenergic transporters density decrease in the motor cortex<sup>102</sup> have been  
20 associated with motor cortical dysfunction in Parkinson's disease,<sup>103,104</sup> the substantial reduction  
21 of  $\alpha 2$ -ARs availability in the motor cortices of Parkinson's disease patients found in the present  
22 study revealed no direct relationship with a global motor dysfunction (i.e, neither with the  
23 UPDRS-II nor with the UPDRS-III scores). However, it is possible that the potential  
24 consequences of cortical  $\alpha 2$ -ARs loss are more indirect and more specific. Indeed, as  $\alpha 2$ -ARs  
25 would be located on inhibitory interneurons, a reduced availability could contribute to the broad  
26 reduction of intracortical inhibition at rest and to the increase in cortical excitation frequently  
27 observed in PD.<sup>105-107</sup> Future studies are needed to clarify this possible masked relationship.

## 1 **Bradykinesia**

2 Bradykinesia is a multifaceted concept including potentially distinct motor abnormalities with  
3 distinct pathophysiological backgrounds that are all mixed in one single clinical term.<sup>108</sup> Here,  
4 we did not find specific correlates of bradykinesia as assessed by means of UPDRS-III in  
5 [<sup>11</sup>C]yohimbine binding data, but report a significant association with a global alteration of LC  
6 integrity. Although this observation does not address the problem of the “bradykinesia complex”  
7 (ibid), it contributes to the debate about its incomplete response to dopaminergic drugs. Indeed,  
8 levodopa only improves overall bradykinesia but does not normalize all other abnormal  
9 movement parameters.<sup>109</sup> Our data support the suggestions of several studies promoting the role  
10 of noradrenaline in the control of motor behavior<sup>11</sup> and the fact that its depletion results in motor  
11 deterioration in Parkinson’s disease.<sup>16,19,30,102</sup> It can be assumed that the role of noradrenaline is  
12 indirect. First because it would facilitate nigro-striatal dopamine transmission.<sup>110</sup> Second because  
13 the noradrenergic system would play a major role in executive functions like response inhibition  
14 whose alteration might lead to movement control disorders.<sup>111–114</sup> Future work on the  
15 noradrenergic bases of movement disorders in Parkinson’s disease should include a more  
16 detailed clinical evaluation of bradykinesia and its related features.<sup>108</sup>

## 17 **Motor fluctuations**

18 The present study pinpoints a correlation between the decrease in LC signal intensity and the  
19 severity of levodopa-induced motor complications (UPDRS-IV total score). However we did not  
20 find any relationship with  $\alpha 2$ -ARs availability change despite previous studies having reported a  
21 potent effect of  $\alpha 2$ -ARs antagonists on dyskinesia in a monkey model of Parkinson’s disease<sup>115–</sup>  
22 <sup>118</sup> as well as in patients with Parkinson’s disease when given in combination with levodopa.<sup>119</sup>  
23 In addition, although there are very few investigations of the role of the LC in motor fluctuations,  
24 LC denervation in rodents was not found to potentiate levodopa-induced motor complications.<sup>120</sup>  
25 These contradictions clearly require further investigations comparing notably Parkinson’s  
26 disease patients with and without motor fluctuations and dyskinesias. Nevertheless, the present  
27 data bring back to the forefront the issue of the links between motor and non-motor  
28 fluctuations,<sup>121</sup> and suggest that alteration of LC integrity might be a common denominator.

## 1 **Rest tremor**

2 Tremor in Parkinson's disease does not closely correlate with nigrostriatal dopaminergic  
3 deficits<sup>122</sup> and is not fully restored by dopaminergic medication.<sup>123</sup> Increasing evidence also  
4 involves the noradrenergic system in the network dynamics of parkinsonian tremor.<sup>27,122,124</sup> Our  
5 data are consistent with this view. Both the amount of neuronal loss in the LC inferred from  
6 neuromelanin MRI and  $\alpha$ 2-ARs availability in the thalamus inferred from [<sup>11</sup>C]yohimbine  
7 binding have been associated with tremor level in the present dataset. The direction of the  
8 relationship between neuromelanin MRI and clinical data indicates that tremor is higher in  
9 patients for whom LC integrity is better preserved, as previously observed in post-mortem  
10 analyses comparing tremor-dominant to akinetic-rigid patients<sup>125,126</sup> or in *in vivo* investigations  
11 using [<sup>11</sup>C]MeNER PET.<sup>124</sup> This relationship was also predicted by former observations linking  
12 the presence of tremor to a more benign course of disease<sup>127</sup> or to a relatively preserved LC-NA  
13 system.<sup>27,124</sup> Our data are in line with a group of studies implicating a cerebello-thalamo-cortical  
14 circuit in the genesis of tremor<sup>128-132</sup> as they provide evidence that  $\alpha$ 2-ARs availability in the  
15 thalamus decreases with the severity of rest tremor. This is particularly consistent with the recent  
16 observation that distribution volume ratio of noradrenaline transporters in the thalamus correlates  
17 with resting tremor amplitude in patients with Parkinson's disease, suggesting that noradrenaline  
18 transporters density is relatively preserved in this region for patients with tremor-dominant  
19 Parkinson's disease.<sup>124</sup>

## 20 **Limitations**

21 There are some limitations to this study that should be acknowledged. First, due to ethical  
22 reasons and to the need to reduce movements within the scanner for imaging small brainstem  
23 structures like the LC, patients with Parkinson's disease were scanned in the ON medication  
24 state. There is evidence from animal studies that administration of levodopa might foster the  
25 accumulation of neuromelanin.<sup>133</sup> However, these effects are expected to be relatively long-  
26 lasting rather than fluctuating between medication states. In the present study, LEDD in patients  
27 did correlate with CNRLC indicating that PD patients with higher dose of dopaminergic treatment  
28 were those with lower signal in the LC. This was observed for both sides of the LC. A second  
29 possible effect of the ON medication state is that postsynaptic  $\alpha$ 2-ARs may be

1 downregulated.<sup>134,135</sup> However, we did not find any correlation between LEDD and  
2 [<sup>11</sup>C]yohimbine BP<sub>ND</sub>, at least in the regions that have been sampled.

3 Second, it is not possible to distinguish the pre- and post-synaptic locations of the  $\alpha$ 2-ARs with  
4 [<sup>11</sup>C]yohimbine. Future studies should consider multitracer approaches with specific presynaptic  
5 tracer (such as [<sup>11</sup>C]MeNER) in order to assess whether changes in  $\alpha$ 2-ARs with Parkinson's  
6 disease mainly occur at the pre- or at the post-synaptic level (or both). Furthermore, it is also  
7 worth mentioning that [<sup>11</sup>C]yohimbine does not differentiate either between the different  
8 subtypes of  $\alpha$ 2-ARs that have been characterized pharmacology ( $\alpha$ 2A,  $\alpha$ 2B,  $\alpha$ 2C).<sup>136</sup> Therefore,  
9 there is still a strong need in the future to pursue the effort in developing specific radiotracers to  
10 further investigate this noradrenergic system *in vivo*.

## 11 **Conclusions**

12 In conclusion, the present work is the first multimodal comparative *in vivo* imaging study to  
13 assess the association between Parkinson's disease symptoms and the integrity of two aspects of  
14 the noradrenergic system: the distribution of  $\alpha$ 2-ARs in the brain and the density of cell bodies in  
15 the LC. Our results highlight the substantial and multimodal alteration of the noradrenergic  
16 system in humans and its possible role in the pathophysiology of Parkinson's disease, not only in  
17 terms of non-motor impairments (supporting the noradrenergic phenotype recently proposed by  
18 Chaudhuri and colleagues)<sup>7</sup> but also with regard to some motor aspects of the disease  
19 (bradykinesia, motor fluctuations and rest tremor). Overall, our findings may encourage the  
20 search for new drugs targeting the  $\alpha$ 2-ARs system for the treatment of Parkinson's disease.  
21 However, future studies are warranted to shed more light on the specific role of the  $\alpha$ 2-ARs and  
22 their different subtypes in the pathophysiology of the disease. In particular, further research is  
23 needed to sharpen the clinical picture and better characterize the different phenotypes of this  
24 heterogeneous and multisystem disease.

## 25 **Data availability**

26 Data are available upon reasonable request.

27

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## 7 **Competing interests**

8 The authors report no competing interests.

## 10 **References**

- 11 1. Politis M, Wu K, Loane C, et al. Depressive symptoms in PD correlate with higher 5-  
12 HTT binding in raphe and limbic structures. *Neurology*. 2010;75(21):1920-1927.  
13 doi:10.1212/WNL.0b013e3181feb2ab
- 14 2. Sommerauer M, Fedorova TD, Hansen AK, et al. Evaluation of the noradrenergic system  
15 in Parkinson's disease: an 11C-MeNER PET and neuromelanin MRI study. *Brain*.  
16 2018;141(2):496-504. doi:10.1093/brain/awx348
- 17 3. Maillet A, Krack P, Lhommée E, et al. The prominent role of serotonergic degeneration  
18 in apathy, anxiety and depression in de novo Parkinson's disease. *Brain*. 2016;139(Pt 9):2486-  
19 2502. doi:10.1093/brain/aww162
- 20 4. Bohnen NI, Kaufer DI, Hendrickson R, Constantine GM, Mathis CA, Moore RY.  
21 Cortical cholinergic denervation is associated with depressive symptoms in Parkinson's disease  
22 and parkinsonian dementia. *J Neurol Neurosurg Psychiatry*. 2007;78(6):641-643.  
23 doi:10.1136/jnnp.2006.100073
- 24 5. Malatt C, Tagliati M. The role of the locus coeruleus/norepinephrine system in the  
25 pathogenesis of neurodegenerative disorders: An update. *Curr Opin Neurol*. 2022;35(2):220-229.

- 1 doi:10.1097/WCO.0000000000001042
- 2 6. García-Lorenzo D, Longo-Dos Santos C, Ewencyk C, et al. The coeruleus/subcoeruleus  
3 complex in rapid eye movement sleep behaviour disorders in Parkinson's disease. *Brain*.  
4 2013;136(Pt 7):2120-2129. doi:10.1093/brain/awt152
- 5 7. Ray Chaudhuri K, Leta V, Bannister K, Brooks DJ, Svenningsson P. The noradrenergic  
6 subtype of Parkinson disease: from animal models to clinical practice. *Nat Rev Neurol*.  
7 2023;19(6):333-345. doi:10.1038/s41582-023-00802-5
- 8 8. Holland N, Robbins TW, Rowe JB. The role of noradrenaline in cognition and cognitive  
9 disorders. *Brain*. 2021;144(8):2243-2256. doi:10.1093/brain/awab111
- 10 9. Sara SJ. The locus coeruleus and noradrenergic modulation of cognition. *Nature Reviews*  
11 *Neuroscience*. 2009;10(3):211-223. doi:10.1038/nrn2573
- 12 10. Aston-Jones G, Cohen JD. Adaptive gain and the role of the locus coeruleus-  
13 norepinephrine system in optimal performance. *The Journal of Comparative Neurology*.  
14 2005;493(1):99-110. doi:10.1002/cne.20723
- 15 11. Waterhouse BD, Predale HK, Plummer NW, Jensen P, Chandler DJ. Probing the  
16 structure and function of locus coeruleus projections to CNS motor centers. *Front Neural*  
17 *Circuits*. 2022;16:895481. doi:10.3389/fncir.2022.895481
- 18 12. O'Neill E, Harkin A. Targeting the noradrenergic system for anti-inflammatory and  
19 neuroprotective effects: implications for Parkinson's disease. *Neural Regen Res*.  
20 2018;13(8):1332-1337. doi:10.4103/1673-5374.235219
- 21 13. Benarroch EE. The locus ceruleus norepinephrine system: functional organization and  
22 potential clinical significance. *Neurology*. 2009;73(20):1699-1704.  
23 doi:10.1212/WNL.0b013e3181c2937c
- 24 14. Braak H, Del Tredici K. Neuropathological Staging of Brain Pathology in Sporadic  
25 Parkinson's disease: Separating the Wheat from the Chaff. *J Parkinsons Dis*. 2017;7(s1):S71-  
26 S85. doi:10.3233/JPD-179001
- 27 15. Fornai F, Torracca MT, Bassi L, D'Errigo DA, Scalori V, Corsini GU. Norepinephrine  
28 loss selectively enhances chronic nigrostriatal dopamine depletion in mice and rats. *Brain Res*.

- 1 1996;735(2):349-353. doi:10.1016/0006-8993(96)00891-8
- 2 16. Mavridis M, Degryse AD, Lategan AJ, Marien MR, Colpaert FC. Effects of locus  
3 coeruleus lesions on parkinsonian signs, striatal dopamine and substantia nigra cell loss after 1-  
4 methyl-4-phenyl-1,2,3,6-tetrahydropyridine in monkeys: a possible role for the locus coeruleus  
5 in the progression of Parkinson's disease. *Neuroscience*. 1991;41(2-3):507-523.  
6 doi:10.1016/0306-4522(91)90345-o
- 7 17. Paredes-Rodriguez E, Vegas-Suarez S, Morera-Herreras T, De Deurwaerdere P,  
8 Miguelez C. The Noradrenergic System in Parkinson's Disease. *Front Pharmacol*. 2020;11:435.  
9 doi:10.3389/fphar.2020.00435
- 10 18. Cui K, Yang F, Tufan T, et al. Restoration of Noradrenergic Function in Parkinson's  
11 Disease Model Mice. *ASN Neuro*. 2021;13:17590914211009730.  
12 doi:10.1177/17590914211009730
- 13 19. Delaville C, Deurwaerdère PD, Benazzouz A. Noradrenaline and Parkinson's disease.  
14 *Front Syst Neurosci*. 2011;5:31. doi:10.3389/fnsys.2011.00031
- 15 20. Laurencin C, Timestit N, Marques A, et al. Efficacy and safety of clonidine for the  
16 treatment of impulse control disorder in Parkinson's disease: a multicenter, parallel, randomised,  
17 double-blind, Phase 2b Clinical trial. *J Neurol*. Published online June 20, 2023.  
18 doi:10.1007/s00415-023-11814-y
- 19 21. Espay AJ, LeWitt PA, Kaufmann H. Norepinephrine deficiency in Parkinson's disease:  
20 the case for noradrenergic enhancement. *Mov Disord*. 2014;29(14):1710-1719.  
21 doi:10.1002/mds.26048
- 22 22. Yssel JD, O'Neill E, Nolan YM, Connor TJ, Harkin A. Treatment with the noradrenaline  
23 re-uptake inhibitor atomoxetine alone and in combination with the  $\alpha$ 2-adrenoceptor antagonist  
24 idazoxan attenuates loss of dopamine and associated motor deficits in the LPS inflammatory rat  
25 model of Parkinson's disease. *Brain Behav Immun*. 2018;69:456-469.  
26 doi:10.1016/j.bbi.2018.01.004
- 27 23. van Veldhuizen MJ, Feenstra MG, Heinsbroek RP, Boer GJ. In vivo microdialysis of  
28 noradrenaline overflow: effects of alpha-adrenoceptor agonists and antagonists measured by  
29 cumulative concentration-response curves. *Br J Pharmacol*. 1993;109(3):655-660.

- 1 doi:10.1111/j.1476-5381.1993.tb13623.x
- 2 24. Srinivasan J, Schmidt WJ. Treatment with alpha2-adrenoceptor antagonist, 2-methoxy  
3 idazoxan, protects 6-hydroxydopamine-induced Parkinsonian symptoms in rats: neurochemical  
4 and behavioral evidence. *Behav Brain Res.* 2004;154(2):353-363. doi:10.1016/j.bbr.2004.03.002
- 5 25. Borghammer P. The brain-first vs. body-first model of Parkinson's disease with  
6 comparison to alternative models. *J Neural Transm (Vienna).* 2023;130(6):737-753.  
7 doi:10.1007/s00702-023-02633-6
- 8 26. Rietdijk CD, Perez-Pardo P, Garssen J, van Wezel RJA, Kraneveld AD. Exploring  
9 Braak's Hypothesis of Parkinson's Disease. *Front Neurol.* 2017;8:37.  
10 doi:10.3389/fneur.2017.00037
- 11 27. Nahimi A, Sommerauer M, Kinnerup MB, et al. Noradrenergic Deficits in Parkinson  
12 Disease Imaged with <sup>11</sup>C-MeNER. *J Nucl Med.* 2018;59(4):659-664.  
13 doi:10.2967/jnumed.117.190975
- 14 28. Probst A, Cortés R, Palacios JM. Distribution of alpha 2-adrenergic receptors in the  
15 human brainstem: an autoradiographic study using [<sup>3</sup>H]p-aminoclonidine. *Eur J Pharmacol.*  
16 1984;106(3):477-488. doi:10.1016/0014-2999(84)90051-7
- 17 29. Pazos A, González AM, Pascual J, Meana JJ, Barturen F, García-Sevilla JA. Alpha 2-  
18 adrenoceptors in human forebrain: autoradiographic visualization and biochemical parameters  
19 using the agonist [<sup>3</sup>H]UK-14304. *Brain Res.* 1988;475(2):361-365. doi:10.1016/0006-  
20 8993(88)90626-9
- 21 30. Pifl C, Kish SJ, Hornykiewicz O. Thalamic noradrenaline in Parkinson's disease: deficits  
22 suggest role in motor and non-motor symptoms. *Mov Disord.* 2012;27(13):1618-1624.  
23 doi:10.1002/mds.25109
- 24 31. Kish SJ, Shannak KS, Rajput AH, Gilbert JJ, Hornykiewicz O. Cerebellar norepinephrine  
25 in patients with Parkinson's disease and control subjects. *Arch Neurol.* 1984;41(6):612-614.  
26 doi:10.1001/archneur.1984.04210080020007
- 27 32. Shannak K, Rajput A, Rozdilsky B, Kish S, Gilbert J, Hornykiewicz O. Noradrenaline,  
28 dopamine and serotonin levels and metabolism in the human hypothalamus: observations in  
29 Parkinson's disease and normal subjects. *Brain Res.* 1994;639(1):33-41. doi:10.1016/0006-

- 1 8993(94)91761-2
- 2 33. Cash R, Ruberg M, Raisman R, Agid Y. Adrenergic receptors in Parkinson's disease.  
3 *Brain Res.* 1984;322(2):269-275. doi:10.1016/0006-8993(84)90117-3
- 4 34. Laurencin C, Lancelot S, Gobert F, et al. Modeling [11C]yohimbine PET human brain  
5 kinetics with test-retest reliability, competition sensitivity studies and search for a suitable  
6 reference region. *Neuroimage.* 2021;240:118328. doi:10.1016/j.neuroimage.2021.118328
- 7 35. Nahimi A, Jakobsen S, Munk OL, et al. Mapping  $\alpha 2$  adrenoceptors of the human brain  
8 with 11C-yohimbine. *J Nucl Med.* 2015;56(3):392-398. doi:10.2967/jnumed.114.145565
- 9 36. Laurencin C, Lancelot S, Merida I, et al. Distribution of  $\alpha 2$ -Adrenergic Receptors in the  
10 Living Human Brain Using [11C]yohimbine PET. *Biomolecules.* 2023;13(5):843.  
11 doi:10.3390/biom13050843
- 12 37. Jakobsen S, Pedersen K, Smith DF, Jensen SB, Munk OL, Cumming P. Detection of  
13 alpha2-adrenergic receptors in brain of living pig with 11C-yohimbine. *J Nucl Med.*  
14 2006;47(12):2008-2015.
- 15 38. Landau AM, Dyve S, Jakobsen S, Alstrup AKO, Gjedde A, Doudet DJ. Acute Vagal  
16 Nerve Stimulation Lowers  $\alpha 2$  Adrenoceptor Availability: Possible Mechanism of Therapeutic  
17 Action. *Brain Stimul.* 2015;8(4):702-707. doi:10.1016/j.brs.2015.02.003
- 18 39. Landau AM, Doudet DJ, Jakobsen S. Amphetamine challenge decreases yohimbine  
19 binding to  $\alpha 2$  adrenoceptors in Landrace pig brain. *Psychopharmacology (Berl).*  
20 2012;222(1):155-163. doi:10.1007/s00213-011-2632-6
- 21 40. Doppler CEJ, Kinnerup MB, Brune C, et al. Regional locus coeruleus degeneration is  
22 uncoupled from noradrenergic terminal loss in Parkinson's disease. *Brain.* 2021;144(9):2732-  
23 2744. doi:10.1093/brain/awab236
- 24 41. Postuma RB, Berg D, Stern M, et al. MDS clinical diagnostic criteria for Parkinson's  
25 disease. *Mov Disord.* 2015;30(12):1591-1601. doi:10.1002/mds.26424
- 26 42. Goetz CG, Tilley BC, Shaftman SR, et al. Movement Disorder Society-sponsored  
27 revision of the Unified Parkinson's Disease Rating Scale (MDS-UPDRS): scale presentation and  
28 clinimetric testing results. *Mov Disord.* 2008;23(15):2129-2170. doi:10.1002/mds.22340

- 1 43. Bushnell DM, Martin ML. Quality of life and Parkinson's disease: translation and  
2 validation of the US Parkinson's Disease Questionnaire (PDQ-39). *Qual Life Res.*  
3 1999;8(4):345-350. doi:10.1023/a:1008979705027
- 4 44. Tomlinson CL, Stowe R, Patel S, Rick C, Gray R, Clarke CE. Systematic review of  
5 levodopa dose equivalency reporting in Parkinson's disease. *Mov Disord.* 2010;25(15):2649-  
6 2653. doi:10.1002/mds.23429
- 7 45. Zucca FA, Bellei C, Giannelli S, et al. Neuromelanin and iron in human locus coeruleus  
8 and substantia nigra during aging: consequences for neuronal vulnerability. *J Neural Transm*  
9 *(Vienna)*. 2006;113(6):757-767. doi:10.1007/s00702-006-0453-2
- 10 46. Dahl MJ, Mather M, Werkle-Bergner M, et al. Locus coeruleus integrity is related to tau  
11 burden and memory loss in autosomal-dominant Alzheimer's disease. *Neurobiol Aging.*  
12 2022;112:39-54. doi:10.1016/j.neurobiolaging.2021.11.006
- 13 47. Liu KY, Marijatta F, Hämmerer D, Acosta-Cabronero J, Düzel E, Howard RJ. Magnetic  
14 resonance imaging of the human locus coeruleus: A systematic review. *Neurosci Biobehav Rev.*  
15 2017;83:325-355. doi:10.1016/j.neubiorev.2017.10.023
- 16 48. Reilhac A, Merida I, Irace Z, et al. Development of a Dedicated Rebinner with Rigid  
17 Motion Correction for the mMR PET/MR Scanner, and Validation in a Large Cohort of 11C-PIB  
18 Scans. *J Nucl Med.* 2018;59(11):1761-1767. doi:10.2967/jnumed.117.206375
- 19 49. Mérida I, Reilhac A, Redouté J, Heckemann RA, Costes N, Hammers A. Multi-atlas  
20 attenuation correction supports full quantification of static and dynamic brain PET data in PET-  
21 MR. *Phys Med Biol.* 2017;62(7):2834-2858. doi:10.1088/1361-6560/aa5f6c
- 22 50. Heckemann RA, Keihaninejad S, Aljabar P, et al. Improving intersubject image  
23 registration using tissue-class information benefits robustness and accuracy of multi-atlas based  
24 anatomical segmentation. *Neuroimage.* 2010;51(1):221-227.  
25 doi:10.1016/j.neuroimage.2010.01.072
- 26 51. Hammers A, Allom R, Koepp MJ, et al. Three-dimensional maximum probability atlas of  
27 the human brain, with particular reference to the temporal lobe. *Hum Brain Mapp.*  
28 2003;19(4):224-247. doi:10.1002/hbm.10123
- 29 52. Prange S, Danaila T, Laurencin C, et al. Age and time course of long-term motor and

- 1 nonmotor complications in Parkinson disease. *Neurology*. 2019;92(2):e148-e160.  
2 doi:10.1212/WNL.0000000000006737
- 3 53. Shibata E, Sasaki M, Tohyama K, et al. Age-related changes in locus ceruleus on  
4 neuromelanin magnetic resonance imaging at 3 Tesla. *Magn Reson Med Sci*. 2006;5(4):197-200.  
5 doi:10.2463/mrms.5.197
- 6 54. Li Y, Wang C, Wang J, et al. Mild cognitive impairment in de novo Parkinson's disease:  
7 A neuromelanin MRI study in locus coeruleus. *Mov Disord*. 2019;34(6):884-892.  
8 doi:10.1002/mds.27682
- 9 55. Prasuhn J, Prasuhn M, Fellbrich A, et al. Association of Locus Coeruleus and Substantia  
10 Nigra Pathology With Cognitive and Motor Functions in Patients With Parkinson Disease.  
11 *Neurology*. 2021;97(10):e1007-e1016. doi:10.1212/WNL.00000000000012444
- 12 56. Sasaki M, Shibata E, Tohyama K, et al. Neuromelanin magnetic resonance imaging of  
13 locus ceruleus and substantia nigra in Parkinson's disease. *Neuroreport*. 2006;17(11):1215-1218.  
14 doi:10.1097/01.wnr.0000227984.84927.a7
- 15 57. Ohtsuka C, Sasaki M, Konno K, et al. Differentiation of early-stage parkinsonisms using  
16 neuromelanin-sensitive magnetic resonance imaging. *Parkinsonism Relat Disord*.  
17 2014;20(7):755-760. doi:10.1016/j.parkreldis.2014.04.005
- 18 58. Chan-Palay V, Asan E. Alterations in catecholamine neurons of the locus coeruleus in  
19 senile dementia of the Alzheimer type and in Parkinson's disease with and without dementia and  
20 depression. *J Comp Neurol*. 1989;287(3):373-392. doi:10.1002/cne.902870308
- 21 59. German DC, Manaye KF, White CL, et al. Disease-specific patterns of locus coeruleus  
22 cell loss. *Ann Neurol*. 1992;32(5):667-676. doi:10.1002/ana.410320510
- 23 60. Zarow C, Lyness SA, Mortimer JA, Chui HC. Neuronal loss is greater in the locus  
24 coeruleus than nucleus basalis and substantia nigra in Alzheimer and Parkinson diseases. *Arch*  
25 *Neurol*. 2003;60(3):337-341. doi:10.1001/archneur.60.3.337
- 26 61. Nahimi A, Kinnerup MB, Ostergaard K, et al. In-vivo imaging of  $\alpha_2$  adrenoceptors in  
27 Parkinson's disease with  $^{11}\text{C}$ -yohimbine. *European Journal of Neurology*. 2017;24(S1):594-  
28 595. doi:10.1111/ene.13368

- 1 62. Pifl C, Hornykiewicz O, Blesa J, Adánez R, Cavada C, Obeso JA. Reduced  
2 noradrenaline, but not dopamine and serotonin in motor thalamus of the MPTP primate: relation  
3 to severity of parkinsonism. *J Neurochem.* 2013;125(5):657-662. doi:10.1111/jnc.12162
- 4 63. Civantos Calzada B, Aleixandre de Artiñano A. Alpha-adrenoceptor subtypes.  
5 *Pharmacol Res.* 2001;44(3):195-208. doi:10.1006/phrs.2001.0857
- 6 64. Timmermans PB, van Zwieten PA. Mini-review. The postsynaptic alpha 2-  
7 adrenoreceptor. *J Auton Pharmacol.* 1981;1(2):171-183. doi:10.1111/j.1474-  
8 8673.1981.tb00509.x
- 9 65. Kaasinen V, Vahlberg T, Stoessl AJ, Strafella AP, Antonini A. Dopamine Receptors in  
10 Parkinson's Disease: A Meta-Analysis of Imaging Studies. *Mov Disord.* 2021;36(8):1781-1791.  
11 doi:10.1002/mds.28632
- 12 66. Klimek V, Ordway GA. Distribution of alpha 2-adrenoceptors in the human locus  
13 coeruleus. *Brain Res.* 1996;741(1-2):263-274. doi:10.1016/s0006-8993(96)00937-7
- 14 67. Quentin E, Belmer A, Maroteaux L. Somato-Dendritic Regulation of Raphe Serotonin  
15 Neurons; A Key to Antidepressant Action. *Front Neurosci.* 2018;12:982.  
16 doi:10.3389/fnins.2018.00982
- 17 68. Herlofson K, Kluger BM. Fatigue in Parkinson's disease. *J Neurol Sci.* 2017;374:38-41.  
18 doi:10.1016/j.jns.2016.12.061
- 19 69. Siciliano M, Trojano L, Santangelo G, De Micco R, Tedeschi G, Tessitore A. Fatigue in  
20 Parkinson's disease: A systematic review and meta-analysis. *Mov Disord.* 2018;33(11):1712-  
21 1723. doi:10.1002/mds.27461
- 22 70. Schapira AHV, Chaudhuri KR, Jenner P. Non-motor features of Parkinson disease. *Nat*  
23 *Rev Neurosci.* 2017;18(7):435-450. doi:10.1038/nrn.2017.62
- 24 71. Pavese N, Metta V, Bose SK, Chaudhuri KR, Brooks DJ. Fatigue in Parkinson's disease  
25 is linked to striatal and limbic serotonergic dysfunction. *Brain.* 2010;133(11):3434-3443.  
26 doi:10.1093/brain/awq268
- 27 72. Mongeau R, Blier P, de Montigny C. In vivo electrophysiological evidence for tonic  
28 activation by endogenous noradrenaline of alpha 2-adrenoceptors on 5-hydroxytryptamine

- 1 terminals in the rat hippocampus. *Naunyn Schmiedebergs Arch Pharmacol.* 1993;347(3):266-  
2 272. doi:10.1007/BF00167444
- 3 73. Aston-Jones G, Akaoka H, Charléty P, Chouvet G. Serotonin selectively attenuates  
4 glutamate-evoked activation of noradrenergic locus coeruleus neurons. *J Neurosci.*  
5 1991;11(3):760-769. doi:10.1523/JNEUROSCI.11-03-00760.1991
- 6 74. Baraban JM, Aghajanian GK. Suppression of firing activity of 5-HT neurons in the dorsal  
7 raphe by alpha-adrenoceptor antagonists. *Neuropharmacology.* 1980;19(4):355-363.  
8 doi:10.1016/0028-3908(80)90187-2
- 9 75. Chaudhuri A, Behan PO. Fatigue and basal ganglia. *J Neurol Sci.* 2000;179(S 1-2):34-42.  
10 doi:10.1016/s0022-510x(00)00411-1
- 11 76. Meeusen R, Watson P, Hasegawa H, Roelands B, Piacentini MF. Central fatigue: the  
12 serotonin hypothesis and beyond. *Sports Med.* 2006;36(10):881-909. doi:10.2165/00007256-  
13 200636100-00006
- 14 77. Klass M, Roelands B, Lévénéz M, et al. Effects of noradrenaline and dopamine on  
15 supraspinal fatigue in well-trained men. *Med Sci Sports Exerc.* 2012;44(12):2299-2308.  
16 doi:10.1249/MSS.0b013e318265f356
- 17 78. Mendonça DA, Menezes K, Jog MS. Methylphenidate improves fatigue scores in  
18 Parkinson disease: a randomized controlled trial. *Mov Disord.* 2007;22(14):2070-2076.  
19 doi:10.1002/mds.21656
- 20 79. Lou JS. Fatigue in Parkinson's disease and potential interventions. *NeuroRehabilitation.*  
21 2015;37(1):25-34. doi:10.3233/NRE-151238
- 22 80. Pagonabarraga J, Kulisevsky J, Strafella AP, Krack P. Apathy in Parkinson's disease:  
23 clinical features, neural substrates, diagnosis, and treatment. *Lancet Neurol.* 2015;14(5):518-531.  
24 doi:10.1016/S1474-4422(15)00019-8
- 25 81. D'Iorio A, Maggi G, Vitale C, Trojano L, Santangelo G. "Pure apathy" and cognitive  
26 dysfunctions in Parkinson's disease: A meta-analytic study. *Neurosci Biobehav Rev.* 2018;94:1-  
27 10. doi:10.1016/j.neubiorev.2018.08.004
- 28 82. De Waele S, Cras P, Crosiers D. Apathy in Parkinson's Disease: Defining the Park

- 1 Apathy Subtype. *Brain Sci.* 2022;12(7):923. doi:10.3390/brainsci12070923
- 2 83. Hezemans FH, Wolpe N, O'Callaghan C, et al. Noradrenergic deficits contribute to  
3 apathy in Parkinson's disease through the precision of expected outcomes. *PLoS Comput Biol.*  
4 2022;18(5):e1010079. doi:10.1371/journal.pcbi.1010079
- 5 84. Béreau M, Van Waes V, Servant M, Magnin E, Tatu L, Anheim M. Apathy in  
6 Parkinson's Disease: Clinical Patterns and Neurobiological Basis. *Cells.* 2023;12(12):1599.  
7 doi:10.3390/cells12121599
- 8 85. Béreau M, Castrioto A, Lhommée E, et al. Fatigue in de novo Parkinson's Disease:  
9 Expanding the Neuropsychiatric Triad? *J Parkinsons Dis.* 2022;12(4):1329-1337.  
10 doi:10.3233/JPD-213116
- 11 86. Thobois S, Ardouin C, Lhommée E, et al. Non-motor dopamine withdrawal syndrome  
12 after surgery for Parkinson's disease: predictors and underlying mesolimbic denervation. *Brain.*  
13 2010;133(Pt 4):1111-1127. doi:10.1093/brain/awq032
- 14 87. Thobois S, Prange S, Sgambato-Faure V, Tremblay L, Broussolle E. Imaging the  
15 Etiology of Apathy, Anxiety, and Depression in Parkinson's Disease: Implication for Treatment.  
16 *Curr Neurol Neurosci Rep.* 2017;17(10):76. doi:10.1007/s11910-017-0788-0
- 17 88. Carey G, Görmezoglu M, de Jong JJA, et al. Neuroimaging of Anxiety in Parkinson's  
18 Disease: A Systematic Review. *Mov Disord.* 2021;36(2):327-339. doi:10.1002/mds.28404
- 19 89. Remy P, Doder M, Lees A, Turjanski N, Brooks D. Depression in Parkinson's disease:  
20 loss of dopamine and noradrenaline innervation in the limbic system. *Brain.* 2005;128(Pt  
21 6):1314-1322. doi:10.1093/brain/awh445
- 22 90. Postuma RB, Berg D. Advances in markers of prodromal Parkinson disease. *Nat Rev*  
23 *Neurol.* 2016;12(11):622-634. doi:10.1038/nrneurol.2016.152
- 24 91. Chaudhuri KR, Healy DG, Schapira AHV, National Institute for Clinical Excellence.  
25 Non-motor symptoms of Parkinson's disease: diagnosis and management. *Lancet Neurol.*  
26 2006;5(3):235-245. doi:10.1016/S1474-4422(06)70373-8
- 27 92. Cersosimo MG, Benarroch EE. Neural control of the gastrointestinal tract: implications  
28 for Parkinson disease. *Mov Disord.* 2008;23(8):1065-1075. doi:10.1002/mds.22051

- 1 93. Braak H, Del Tredici K, Rüb U, de Vos RAI, Jansen Steur ENH, Braak E. Staging of  
2 brain pathology related to sporadic Parkinson's disease. *Neurobiol Aging*. 2003;24(2):197-211.
- 3 94. Taylor TN, Caudle WM, Shepherd KR, et al. Nonmotor symptoms of Parkinson's disease  
4 revealed in an animal model with reduced monoamine storage capacity. *J Neurosci*.  
5 2009;29(25):8103-8113. doi:10.1523/JNEUROSCI.1495-09.2009
- 6 95. Butkovich LM, Houser MC, Chalermphanupap T, et al. Transgenic Mice Expressing  
7 Human  $\alpha$ -Synuclein in Noradrenergic Neurons Develop Locus Coeruleus Pathology and  
8 Nonmotor Features of Parkinson's Disease. *J Neurosci*. 2020;40(39):7559-7576.  
9 doi:10.1523/JNEUROSCI.1468-19.2020
- 10 96. Braak H, Ghebremedhin E, Rüb U, Bratzke H, Del Tredici K. Stages in the development  
11 of Parkinson's disease-related pathology. *Cell Tissue Res*. 2004;318(1):121-134.  
12 doi:10.1007/s00441-004-0956-9
- 13 97. Nakamori H, Naitou K, Horii Y, et al. Roles of the noradrenergic nucleus locus coeruleus  
14 and dopaminergic nucleus A11 region as supraspinal defecation centers in rats. *Am J Physiol*  
15 *Gastrointest Liver Physiol*. 2019;317(4):G545-G555. doi:10.1152/ajpgi.00062.2019
- 16 98. Song S, Tu D, Meng C, et al. Dysfunction of the noradrenergic system drives  
17 inflammation,  $\alpha$ -synucleinopathy, and neuronal loss in mouse colon. *Front Immunol*.  
18 2023;14:1083513. doi:10.3389/fimmu.2023.1083513
- 19 99. Song S, Jiang L, Oyarzabal EA, et al. Loss of Brain Norepinephrine Elicits  
20 Neuroinflammation-Mediated Oxidative Injury and Selective Caudo-Rostral Neurodegeneration.  
21 *Mol Neurobiol*. 2019;56(4):2653-2669. doi:10.1007/s12035-018-1235-1
- 22 100. Ye R, O'Callaghan C, Rua C, et al. Locus Coeruleus Integrity from 7 T MRI Relates to  
23 Apathy and Cognition in Parkinsonian Disorders. *Mov Disord*. 2022;37(8):1663-1672.  
24 doi:10.1002/mds.29072
- 25 101. Gaspar P, Duyckaerts C, Alvarez C, Javoy-Agid F, Berger B. Alterations of  
26 dopaminergic and noradrenergic innervations in motor cortex in Parkinson's disease. *Ann*  
27 *Neurol*. 1991;30(3):365-374. doi:10.1002/ana.410300308
- 28 102. Sommerauer M, Hansen AK, Parbo P, et al. Decreased noradrenaline transporter density  
29 in the motor cortex of Parkinson's disease patients. *Mov Disord*. 2018;33(6):1006-1010.

- 1 doi:10.1002/mds.27411
- 2 103. Pasquereau B, DeLong MR, Turner RS. Primary motor cortex of the parkinsonian  
3 monkey: altered encoding of active movement. *Brain*. 2016;139(Pt 1):127-143.  
4 doi:10.1093/brain/awv312
- 5 104. Lindenbach D, Bishop C. Critical involvement of the motor cortex in the  
6 pathophysiology and treatment of Parkinson's disease. *Neurosci Biobehav Rev*. 2013;37(10 Pt  
7 2):2737-2750. doi:10.1016/j.neubiorev.2013.09.008
- 8 105. Gd A, A L. Methylphenidate increases cortical excitability via activation of alpha-2  
9 noradrenergic receptors. *Neuropsychopharmacology: official publication of the American*  
10 *College of Neuropsychopharmacology*. 2006;31(3). doi:10.1038/sj.npp.1300818
- 11 106. Strafella AP, Valzania F, Nasseti SA, et al. Effects of chronic levodopa and pergolide  
12 treatment on cortical excitability in patients with Parkinson's disease: a transcranial magnetic  
13 stimulation study. *Clin Neurophysiol*. 2000;111(7):1198-1202. doi:10.1016/s1388-  
14 2457(00)00316-3
- 15 107. Ridding MC, Inzelberg R, Rothwell JC. Changes in excitability of motor cortical  
16 circuitry in patients with Parkinson's disease. *Ann Neurol*. 1995;37(2):181-188.  
17 doi:10.1002/ana.410370208
- 18 108. Bologna M, Espay AJ, Fasano A, Paparella G, Hallett M, Berardelli A. Redefining  
19 Bradykinesia. *Mov Disord*. 2023;38(4):551-557. doi:10.1002/mds.29362
- 20 109. Bologna M, Paparella G, Fasano A, Hallett M, Berardelli A. Evolving concepts on  
21 bradykinesia. *Brain*. 2020;143(3):727-750. doi:10.1093/brain/awz344
- 22 110. Lategan AJ, Marien MR, Colpaert FC. Suppression of nigrostriatal and mesolimbic  
23 dopamine release in vivo following noradrenaline depletion by DSP-4: a microdialysis study.  
24 *Life Sci*. 1992;50(14):995-999. doi:10.1016/0024-3205(92)90093-5
- 25 111. Criaud M, Laurencin C, Poisson A, et al. Noradrenaline and Movement Initiation  
26 Disorders in Parkinson's Disease: A Pharmacological Functional MRI Study with Clonidine.  
27 *Cells*. 2022;11(17):2640. doi:10.3390/cells11172640
- 28 112. Spay C, Albares M, Lio G, et al. Clonidine modulates the activity of the subthalamic-

- 1 supplementary motor loop: evidence from a pharmacological study combining deep brain  
2 stimulation and electroencephalography recordings in Parkinsonian patients. *J Neurochem.*  
3 2018;146(3):333-347. doi:10.1111/jnc.14447
- 4 113. Ye Z, Altena E, Nombela C, et al. Improving response inhibition in Parkinson's disease  
5 with atomoxetine. *Biol Psychiatry.* 2015;77(8):740-748. doi:10.1016/j.biopsych.2014.01.024
- 6 114. Xu B, He T, Lu Y, et al. Locus coeruleus integrity correlates with inhibitory functions of  
7 the fronto-subthalamic "hyperdirect" pathway in Parkinson's disease. *Neuroimage Clin.*  
8 2022;36:103276. doi:10.1016/j.nicl.2022.103276
- 9 115. Henry B, Fox SH, Peggs D, Crossman AR, Brotchie JM. The alpha2-adrenergic receptor  
10 antagonist idazoxan reduces dyskinesia and enhances anti-parkinsonian actions of L-dopa in the  
11 MPTP-lesioned primate model of Parkinson's disease. *Mov Disord.* 1999;14(5):744-753.  
12 doi:10.1002/1531-8257(199909)14:5<744::aid-mds1006>3.0.co;2-7
- 13 116. Grondin R, Hadj Tahar A, Doan VD, Ladure P, Bédard PJ. Noradrenoceptor antagonism  
14 with idazoxan improves L-dopa-induced dyskinesias in MPTP monkeys. *Naunyn Schmiedebergs*  
15 *Arch Pharmacol.* 2000;361(2):181-186. doi:10.1007/s002109900167
- 16 117. Fox SH, Henry B, Hill MP, Peggs D, Crossman AR, Brotchie JM. Neural mechanisms  
17 underlying peak-dose dyskinesia induced by levodopa and apomorphine are distinct: evidence  
18 from the effects of the alpha(2) adrenoceptor antagonist idazoxan. *Mov Disord.* 2001;16(4):642-  
19 650. doi:10.1002/mds.1148
- 20 118. Savola JM, Hill M, Engstrom M, et al. Fipamezole (JP-1730) is a potent alpha2  
21 adrenergic receptor antagonist that reduces levodopa-induced dyskinesia in the MPTP-lesioned  
22 primate model of Parkinson's disease. *Mov Disord.* 2003;18(8):872-883. doi:10.1002/mds.10464
- 23 119. Rascol O, Arnulf I, Peyro-Saint Paul H, et al. Idazoxan, an alpha-2 antagonist, and L-  
24 DOPA-induced dyskinesias in patients with Parkinson's disease. *Mov Disord.* 2001;16(4):708-  
25 713. doi:10.1002/mds.1143
- 26 120. Marin C, Aguilar E, Bonastre M. Effect of locus coeruleus denervation on levodopa-  
27 induced motor fluctuations in hemiparkinsonian rats. *J Neural Transm (Vienna).*  
28 2008;115(8):1133-1139. doi:10.1007/s00702-008-0060-5
- 29 121. Martínez-Fernández R, Schmitt E, Martínez-Martin P, Krack P. The hidden sister of

- 1 motor fluctuations in Parkinson's disease: A review on nonmotor fluctuations. *Mov Disord.*  
2 2016;31(8):1080-1094. doi:10.1002/mds.26731
- 3 122. Isaias IU, Benti R, Cilia R, et al. [123I]FP-CIT striatal binding in early Parkinson's  
4 disease patients with tremor vs. akinetic-rigid onset. *Neuroreport.* 2007;18(14):1499-1502.  
5 doi:10.1097/WNR.0b013e3282ef69f9
- 6 123. Eggers C, Kahraman D, Fink GR, Schmidt M, Timmermann L. Akinetic-rigid and  
7 tremor-dominant Parkinson's disease patients show different patterns of FP-CIT single photon  
8 emission computed tomography. *Mov Disord.* 2011;26(3):416-423. doi:10.1002/mds.23468
- 9 124. Kinnerup MB, Sommerauer M, Damholdt MF, et al. Preserved noradrenergic function in  
10 Parkinson's disease patients with rest tremor. *Neurobiology of Disease.* 2021;152:105295.  
11 doi:10.1016/j.nbd.2021.105295
- 12 125. Paulus W, Jellinger K. The neuropathologic basis of different clinical subgroups of  
13 Parkinson's disease. *J Neuropathol Exp Neurol.* 1991;50(6):743-755.
- 14 126. Jellinger KA. Post mortem studies in Parkinson's disease--is it possible to detect brain  
15 areas for specific symptoms? *J Neural Transm Suppl.* 1999;56:1-29. doi:10.1007/978-3-7091-  
16 6360-3\_1
- 17 127. Thenganatt MA, Jankovic J. Parkinson disease subtypes. *JAMA Neurol.* 2014;71(4):499-  
18 504. doi:10.1001/jamaneurol.2013.6233
- 19 128. Antonini A, Moeller JR, Nakamura T, Spetsieris P, Dhawan V, Eidelberg D. The  
20 metabolic anatomy of tremor in Parkinson's disease. *Neurology.* 1998;51(3):803-810.  
21 doi:10.1212/wnl.51.3.803
- 22 129. Deiber MP, Pollak P, Passingham R, et al. Thalamic stimulation and suppression of  
23 parkinsonian tremor. Evidence of a cerebellar deactivation using positron emission tomography.  
24 *Brain.* 1993;116 ( Pt 1):267-279. doi:10.1093/brain/116.1.267
- 25 130. Mure H, Hirano S, Tang CC, et al. Parkinson's disease tremor-related metabolic network:  
26 characterization, progression, and treatment effects. *Neuroimage.* 2011;54(2):1244-1253.  
27 doi:10.1016/j.neuroimage.2010.09.028
- 28 131. Helmich RC, Hallett M, Deuschl G, Toni I, Bloem BR. Cerebral causes and

- 1 consequences of parkinsonian resting tremor: a tale of two circuits? *Brain*. 2012;135(Pt  
2 11):3206-3226. doi:10.1093/brain/aws023
- 3 132. Ni Z, Pinto AD, Lang AE, Chen R. Involvement of the cerebellothalamocortical pathway  
4 in Parkinson disease. *Ann Neurol*. 2010;68(6):816-824. doi:10.1002/ana.22221
- 5 133. Sulzer D, Bogulavsky J, Larsen KE, et al. Neuromelanin biosynthesis is driven by excess  
6 cytosolic catecholamines not accumulated by synaptic vesicles. *Proc Natl Acad Sci U S A*.  
7 2000;97(22):11869-11874. doi:10.1073/pnas.97.22.11869
- 8 134. Zhang XL, Wang GB, Zhao LY, et al. Clonidine improved laboratory-measured decision-  
9 making performance in abstinent heroin addicts. *PLoS ONE*. 2012;7(1):e29084.  
10 doi:10.1371/journal.pone.0029084
- 11 135. Wang Y, Wang HS, Wang T, Huang C, Liu J. L-DOPA-induced dyskinesia in a rat  
12 model of Parkinson's disease is associated with the fluctuational release of norepinephrine in the  
13 sensorimotor striatum. *J Neurosci Res*. 2014;92(12):1733-1745. doi:10.1002/jnr.23439
- 14 136. Bylund DB. Pharmacological characteristics of alpha-2 adrenergic receptor subtypes. *Ann*  
15 *N Y Acad Sci*. 1995;763:1-7. doi:10.1111/j.1749-6632.1995.tb32386.x

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## 18 **Figure legends**

19 **Figure 1 Data pipeline.** [<sup>11</sup>C]yohimbine and LC Neuromelanin data were acquired  
20 simultaneously from hybrid PET/MRI. *Left side:* [<sup>11</sup>C]yohimbine BP<sub>ND</sub> parametric maps were  
21 used for contrasting the healthy control and the Parkinson's disease groups. The outcome of the  
22 voxel-based analysis allowed identification of four significant clusters that provided the ROI for  
23 the analysis of the links between [<sup>11</sup>C]yohimbine BP<sub>ND</sub> and clinical scores in patients. *Right side:*  
24 LC identification was performed for each individual from neuromelanin sensitive T1 images  
25 with a three-steps procedure. First, the regions of interest were manually drawn as large 3D  
26 bounding boxes in the MNI space, then wrapped and resampled onto the subject's native space.  
27 Finally, the clusters of connected voxels with the highest signal intensity were selected to define  
28 LC areas. The CNR between the LC and the reference region was calculated to estimate LC

1 signal intensity and these values were referred to the clinical scores of the Parkinson's disease  
2 patients.

3  
4 **Figure 2 Statistical parametric maps comparing Parkinson's disease patients to control**  
5 **subjects.** Decrease in [<sup>11</sup>C]yohimbine binding is observed in patients with Parkinson's disease  
6 with regard to healthy control subjects.

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**Table I Demographics and clinical characteristics of participants**

	Healthy Controls	Parkinsonian patients	Statistics
N	30	30	
Sex (female/male)	12/18	12/18	
Age (years)	60.3 ± 8	60.1 ± 7.5	ns <sup>a</sup>
MoCA	27.9 ± 1.5	27.9 ± 2	ns <sup>b</sup>
<b>Parkinson's disease characteristics</b>			
Disease duration (years)		6.5 ± 4	
LEDD (mg/day)		1006 ± 703	
Hoehn & Yahr		1.8 ± 0.4	
PDQ39		39.6 ± 22.1	
MDS-UPDRS I		8.5 ± 5.1	
MDS-UPDRS II		7.1 ± 4.3	
MDS-UPDRS III		17.7 ± 12.3	
MDS-UPDRS IV		2.3 ± 3	

10 Data are presented as mean ± standard deviation.

11 <sup>a</sup>Parametric test (Student's t-test).

12 <sup>b</sup>Non-parametric test (Wilcoxon test).

13

1 **Table 2 SPM results of the two samples t test on [<sup>11</sup>C]yohimbine binding**

Areas	BA	Side	MNI coordinates			T value	P corr cluster	Cluster size
			x	y	z			
<b>Reduced [<sup>11</sup>C]yohimbine binding in Parkinson's disease versus healthy controls</b>								
Angular Gyrus	40	R	46	-38	52	5.37	0.000	3775
Superior Temporal Gyrus	22	R	52	-12	0	5.08		
Postcentral Gyrus (Opercular)	48	R	50	-8	16	5.06		
Inferior Frontal Gyrus (pars opercularis)	44	R	46	14	10	4.22		
Insula	13	R	36	-12	6	4.18		
Precentral gyrus	4	R	40	-26	58	3.97		
Putamen	-	R	30	-10	6	3.90		
Postcentral gyrus	3	R	52	-14	30	3.73		
Thalamus (PuM)	-	R	6	-24	4	5.18	0.000	1354
Insula	13	L	-32	-6	16	4.59		
Precentral Gyrus (Premotor cortex)	6	L	-48	-4	12	3.84		
Posterior Cingulate Cortex (Dorsal)	31	R	2	-36	40	4.95	0.000	3836
Superior Parietal Cortex	7	R	18	-76	40	4.55		
Lateral Occipital Cortex	19	L	-10	-82	36	4.43		
Cuneus	18	R	4	-80	34	4.41		
Posterior Cingulate Cortex (Ventral)	23	R	8	-52	10	4.37		
Posterior Cingulate Cortex (Mid Cingulate)	23	R	8	-20	46	4.28		
Lateral Occipital Cortex	19	R	32	-80	26	4.27		
Lingual Gyrus	18	L	-6	-72	0	4.07		
Precentral Gyrus	6	R	4	-22	60	3.89		
Lingual Gyrus	17	R	4	-66	12	3.87		
Precuneus	31	L	-6	-52	38	3.80		
Precuneus	31	R	2	-56	42	3.80		
Superior Parietal Cortex	7	L	-32	-50	56	4.41	0.013	614
Angular Gyrus	40	L	-42	-52	46	4.23		
Postcentral Gyrus	3	L	-50	-14	38	3.76		
Postcentral Gyrus	4	L	-36	-28	56	3.66		

2  
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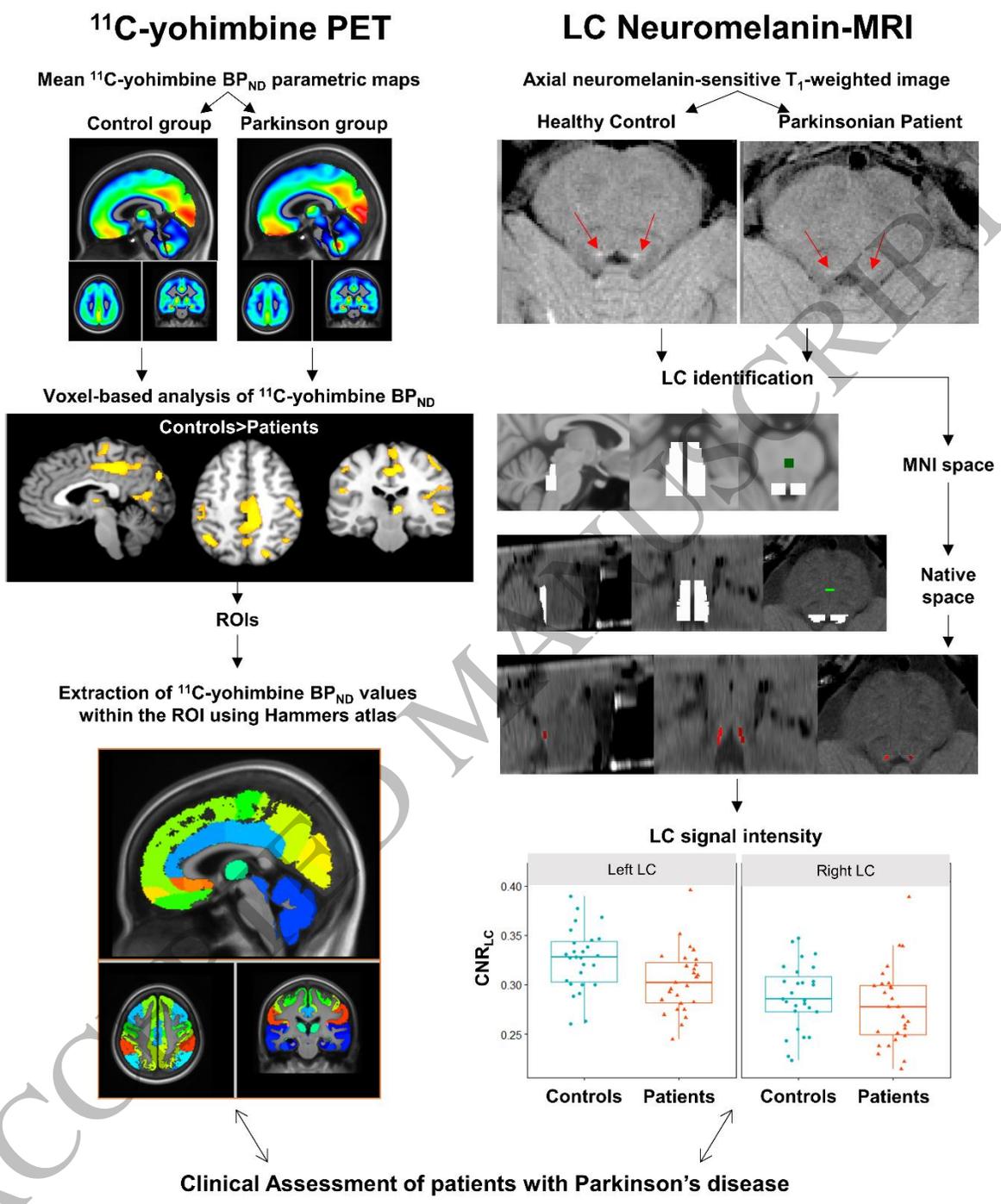


Figure 1  
159x188 mm ( x DPI)

1  
2  
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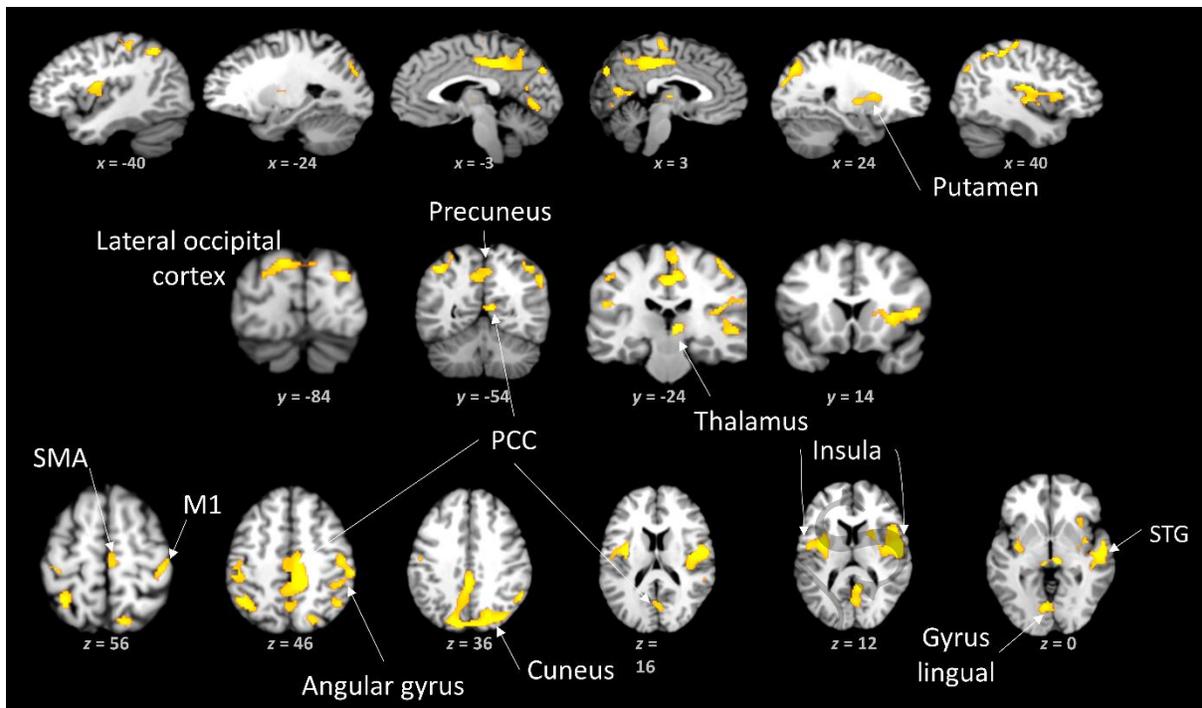


Figure 2  
159x93 mm (x DPI)

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ACCEPTED MANUSCRIPT