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Extrastriatal monoaminergic dysfunction and enhanced microglial activation in idiopathic rapid eye movement sleep behaviour disorder

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Abstract

Background

The majority of patients diagnosed with idiopathic rapid eye movement sleep behaviour disorder (iRBD) progress over time to a Lewy-type α -synucleinopathy such as Parkinson's disease or dementia with Lewy bodies. This *in vivo* molecular imaging study aimed to investigate if extrastriatal monoaminergic systems are affected in iRBD patients and if this coincides with neuroinflammation.

Methods

We studied twenty-one polysomnography-confirmed iRBD patients with ^{18}F -DOPA and ^{11}C -PK11195 positron emission tomography (PET) to investigate extrastriatal monoaminergic function and microglial activation. Twenty-nine healthy controls (n=9 ^{18}F -DOPA and n=20 ^{11}C -PK11195) were also investigated. Analyses were performed within predefined regions of interest and at voxel-level with Statistical Parametric Mapping.

Results

Regions of interest analysis detected monoaminergic dysfunction in iRBD thalamus with a 15% mean reduction of ^{18}F -DOPA K_i values compared to controls (mean difference = -0.00026, 95% confidence interval [-0.00050 to -0.00002], p-value = 0.03). No associated thalamic changes in ^{11}C -PK11195

binding were observed. Other regions sampled showed no ^{18}F -DOPA or ^{11}C -PK11195 PET differences between groups. Voxel-level interrogation of ^{11}C -PK11195 binding identified areas with significantly increased binding within the occipital lobe of iRBD patients.

Conclusion

Thalamic monoaminergic dysfunction in iRBD patients may reflect terminal dysfunction of projecting neurons from the locus coeruleus and dorsal raphe nucleus, two structures that regulate REM sleep and are known to be involved in the early phase of PD. The observation of significantly raised microglial activation in the occipital lobe of these patients might suggest early local Lewy-type α -synuclein pathology and possibly an increased risk for later cognitive dysfunction.

Keywords

Idiopathic rapid-eye-movement sleep behaviour disorder,
Positron emission tomography, ^{18}F -DOPA, ^{11}C -PK11195, PD,
Dementia with Lewy bodies.

Abbreviations

PD = Parkinson's disease; REM = rapid eye movement; iRBD =
idiopathic rapid eye movement sleep behaviour disorder; ROI =
region of interest

Introduction

Pathology studies of Parkinson's disease (PD) patients have reported degeneration in noradrenergic, serotonergic, and cholinergic nuclei and pathways in both subcortical and cortical brain regions [1-4], which is suggested to be connected to a variety of non-motor symptoms commonly observed in these patients [5]. Therefore, *in vivo* molecular imaging, which has traditionally been used to assess dopamine nigrostriatal dysfunction in PD patients, has also been employed to assess changes in extrastriatal regions containing monoaminergic innervation in these patients. Main observations in early stages of PD include thalamic monoaminergic dysfunction [6-9], and increased uptake in the midbrain raphe, internal globus pallidus, anterior cingulate gyrus, and prefrontal cortex [6, 7, 10-12]. Imaging studies in PD have also observed raised levels of microglial activation in both striatal and extrastriatal regions [13-17], suggesting an important role for neuroinflammation in the progression of the disease [18].

Idiopathic rapid-eye-movement sleep behaviour disorder (iRBD), a parasomnia characterised by failure to lose muscle tone during REM sleep leading to dream enactment behaviour [19], seems to be a prodromal stage of Lewy-type α -synucleinopathies characterised by parkinsonism and dementia [20-22]. Using positron emission tomography (PET), we have recently showed

that iRBD patients have raised levels of microglial activation in the substantia nigra along with a putaminal dopaminergic deficit. These findings provide further strong evidence that a large number of iRBD patients are in a prodromal stage of parkinsonism [23]. Interestingly, IRBD patients show a variety of non-motor symptoms similar to those seen in PD patients [24, 25], which in some instances might be related to monoaminergic changes in extrastriatal regions [5]. Therefore, in the current study, we aimed to investigate whether *in vivo* PET imaging detects changes in monoaminergic innervation in extrastriatal regions in iRBD patients similar to those reported in early PD, and whether these are accompanied by increased levels of microglial activation.

Methods

Study subjects

This study was conducted between March 2015 and February 2017. The study protocol was approved by the Central Denmark Region Committee on Health Research and the ethics committee of the Hospital de Clínic Barcelona. All subjects gave informed written consent according to the Declaration of Helsinki before enrolment into the study. Twenty-one iRBD patients with

a polysomnography-confirmed diagnosis were recruited from tertiary sleep clinics at Aarhus University Hospital, Denmark and the Multidisciplinary Sleep Unit of the Hospital Clínic de Barcelona, Spain. Patients from Spain flew to Denmark to undergo PET examinations. All iRBD patients underwent both ^{18}F -DOPA and ^{11}C -PK11195 PET and were compared to 29 healthy control subjects who underwent one of the two PET examinations (n=9 ^{18}F -DOPA PET and n=20 ^{11}C -PK11195 PET) [23, 26]. No patients or control subjects reported motor or cognitive problems.

Since our original investigation of nigrostriatal function in iRBD patients [23] one additional iRBD patient has been recruited to this ongoing study. Additionally, to increase the power of the ^{11}C -PK11195 PET analysis in extrastriatal areas, we have included 10 extra control subjects who had ^{11}C -PK11195 PET as part of another study performed in parallel to the current one at our Unit [26]. The PET scan was performed on the same scanner and with the same scan protocol.

PET and MRI

All subjects had PET and MRI scans performed as previously described [23]. For details, please see supplementary material.

Image quantification

Extrastriatal monoaminergic innervation was assessed with ^{18}F -DOPA PET. ^{18}F -DOPA uptake expressed as an influx constant (K_i value) reflects activity of the aromatic L-amino acid decarboxylase enzyme, which is present in most monoaminergic neurons [27, 28]. Thus, in regions with monoaminergic innervation i.e. dopaminergic, serotonergic or noradrenergic innervation, ^{18}F -DOPA PET measures the overall status of the monoaminergic system [27].

The level and extent of microglial activation was measured with ^{11}C -PK11195 PET, which frequently has been used to examine microglial activation in Lewy-type α -synucleinopathies and other neurodegenerative disorders [13-16, 29, 30].

Parametric images were generated for both tracers. The Patlak graphical approach was used to calculate the ^{18}F -DOPA influx constant (K_i value) and for ^{11}C -PK11195 binding potentials (BP_{ND}) the simplified reference tissue model using an individual reference tissue non-specific input function extracted with a supervised cluster analysis was used [31].

For details, please see supplementary material.

Region of interest analyses

Region of interest analysis was performed with PNEURO PMOD software v 3.6 (Pmod technologies Ltd., Switzerland) as

previously described [23]. For details, please see supplementary material.

Regions of interest were predefined and selected based on a combination of previous observations of ^{18}F -DOPA (monoaminergic marker) [6, 7, 10-12] and ^{11}C -DASB (serotonin transporter marker) [8, 9] changes in early stage PD, the Braak PD Lewy pathology staging scheme [32], and previous observations of raised microglial activation in PD or dementia with Lewy body patients (DLB) [13, 15-17]. Extrastriatal regions of interest sampled included: locus coeruleus, median raphe nucleus, dorsal raphe nucleus, hypothalamus, thalamus, globus pallidus internal, anterior cingulate gyrus, frontal lobe, and occipital lobe. Values from left and right side are averaged. The occipital lobe was not sampled with ^{18}F -DOPA as this was the reference region for non-specific uptake of this tracer.

Assessment of ^{18}F -DOPA K_i values and ^{11}C -PK11195 BP_{ND} in nigrostriatal regions of interest has previously been reported (iRBD $n=20$, controls $n=19$) [23]. In the current study, we assessed whole striatum ^{18}F -DOPA uptake (iRBD $n=21$), which consisted of averaged putamen and caudate ^{18}F -DOPA K_i values from both left and right side. Whole striatum ^{18}F -DOPA uptake was used to assess the relationship between dopaminergic nigrostriatal dysfunction and monoaminergic dysfunction in extrastriatal areas.

Statistical parametric mapping analysis

Following region of interest analysis, Statistical Parametric Mapping (SPM12; Wellcome Trust Centre for Neuroimaging, London, UK) was performed to localise significant changes in tracer uptake/binding at a voxel level within the previously defined extrastriatal regions of interest. Using this method, differences in ^{18}F -DOPA K_i value or ^{11}C -PK11195 BP_{ND} between iRBD patients and controls can be examined in each voxel included within the region of interest analysis. Previous ^{18}F -DOPA PET studies in PD patients have shown changes in voxels confined to smaller areas. These changes may be less extensive compared to those detected in a region of interest analysis encompassing larger volumes, however, not less important. For details, please see supplementary material.

Statistical analysis

Statistical analysis and graphical presentations were performed in Stata IC 14.2 (StataCorp LP, TX, USA) and GraphPad Prism 6 (GraphPad Software, La Jolla, CA, USA). Normal distribution of variables was examined with qnorm-plots and the D'Agostino Shapiro-Wilks test. Group differences in ^{18}F -DOPA K_i values and ^{11}C -PK11195 BP_{ND} were interrogated with a parametric two-tailed Student's t test ($\alpha = 5.0\%$) when data were normally distributed and with a non-parametric

Wilcoxon / Mann-Whitney rank test when this was not the case. Correlations between ^{18}F -DOPA K_i and ^{11}C -PK11195 BP_{ND} within regions of interest were interrogated with a two-tailed Pearson product-moment or Spearman rank correlation ($\alpha=5.0\%$). Correlations between striatal and extrastriatal ^{18}F -DOPA uptake were only performed if an extrastriatal region showed a significant change in ^{18}F -DOPA uptake in iRBD patients compared to controls.

Results

Twenty-one polysomnography-confirmed iRBD patients, 18 men and three women, with a mean age of 66.2 years ($\text{SD}=6.3$) at time of examination and 3.6 ($\text{SD}=3.4$) years from iRBD diagnosis, with a Montreal Cognitive Assessment (MoCA) score of 25.7 ($\text{SD}=2.4$) (Table 1) and 29 healthy control subjects, 21 men ($n=9$ ^{18}F -DOPA and $n=12$ ^{11}C -PK11195) and eight women ($n=8$ ^{11}C -PK11195), with a similar age ($n=9$ ^{18}F -DOPA 64.6 ± 3.6 years, $n=20$ ^{11}C -PK11195 66.8 ± 6.0 years) and MoCA score 26.5 ($\text{SD}=2.3$) were included in the current study.

Region of interest analysis detected a significant mean reduction of 15% in ^{18}F -DOPA K_i values in the thalamus of iRBD patients compared to controls (mean difference = -0.00026, 95% confidence interval [-0.00050 to -0.00002], p -value = 0.03).

In all other extrastriatal regions of interest assessed, no difference was observed between groups (Figure 1 and Table 2). Comparisons for ^{18}F -DOPA uptake at a voxel level within predefined extrastriatal regions of interest did not show any significant difference between iRBD patients and controls with the predetermined threshold ($p < 0.01$ and family-wise error correction $p < 0.05$ at the cluster level). However, when we lowered the threshold of significance to that of the region of interest analysis ($p < 0.05$) we again detected clusters of decreased ^{18}F -DOPA uptake in the thalamus of iRBD patients compared to controls, confirming the findings of the region of interest analysis.

Extrastriatal ^{11}C -PK11195 binding was similar between groups in the region of interest analysis (Figure 1). Voxel-level analysis of ^{11}C -PK11195 binding detected areas of significantly increased signal in the occipital lobe of iRBD patients compared to controls, cluster size = 11629 and 11521 voxels (Figure 2). Within these areas, voxels with the strongest difference in ^{11}C -PK11195 binding were localised to the visual-associative area bilaterally (Brodmann area 18). This finding was not correlated to the MoCA score of iRBD patients, additionally, iRBD patients who drew the cube incorrect ($n=9$) in the visuoconstructional skill test of the MoCA had similar ^{11}C -PK11195 binding in the occipital lobe cluster identified by voxel-analysis compared to iRBD patients who drew the cube

correct (n=12) (p value = 0.81, mean ^{11}C -PK11195 BP_{ND} , iRBD incorrect = 0.086, mean iRBD correct = 0.077) (Figure 2).

No correlations were observed between ^{18}F -DOPA Ki and ^{11}C -PK11195 BP_{ND} within the regions of interest investigated. However, a significant positive correlation was found between thalamic and whole striatum ^{18}F -DOPA uptake, Pearson correlation $r=0.52$ (95% CI 0.11 to 0.78), $p=0.016$ (Figure 3).

Discussion

In this study, we have used PET imaging to assess whether iRBD patients have dysfunctional monoaminergic innervation and/or the presence of microglia activation in extrastriatal brain regions similar to those reported in early stage PD. Patients with iRBD are thought to represent a prodromal phenotype of a Lewy-type α -synucleinopathy disorder and so could already have some pathological features distinctive of these disorders, namely Lewy-type α -synuclein deposition in the peripheral autonomous nervous system [33-35], reduced dopaminergic innervation in the striatum [23, 36] and microglial activation within the substantia nigra [23]. We found significantly reduced ^{18}F -DOPA uptake in the thalamus and increased

microglial activation in the occipital lobe of iRBD patients compared to controls.

PET studies of PD patients with short disease duration have previously observed reduced thalamic ^{18}F -DOPA K_i values (monoaminergic marker) [6, 7], and reduced thalamic ^{11}C -DASB binding (serotonin transporter marker) [8, 9]. Additionally, reduced thalamic ^{11}C -RTI-32 binding (noradrenergic and dopaminergic transporter marker) has also been reported in PD patients with depression [37]. Our observation of thalamic monoaminergic dysfunction in iRBD patients, detected with ^{18}F -DOPA PET, occurring prior to onset of parkinsonism, is a novel finding which could contribute to the understanding of the developing neurodegenerative process in Lewy-type α -synucleinopathies.

Whilst ^{18}F -DOPA PET allows us to assess the presence of overall thalamic monoaminergic dysfunction, it does not allow a separate evaluation of dopaminergic, noradrenergic or serotonergic function in isolation in this structure. Neither will PET evaluate glutamate, glycine or GABA levels; these transmitters are released from the subcoeruleus and gigantocellular nucleus and reach the thalamus and are essential pathways for the maintenance of normal REM sleep atonia [38, 39].

The density of dopaminergic axon terminals within the thalamus is limited with the exception of the pulvinar [40, 41]. Given this, dopaminergic changes will have a small impact on the

overall thalamic monoaminergic innervation. In contrast, both the predominantly noradrenergic locus coeruleus and the serotonergic caudal raphe nucleus send extensive projections to thalamus [40, 42], and early dysfunction in these brainstem nuclei could account for the observed reduction in thalamic ^{18}F -DOPA uptake. This may be supported by post-mortem pathological observations of Lewy-type α -synuclein pathology in these brainstem nuclei in iRBD patients [43] similar to that seen in Braak stage II PD [32]. Additionally, the locus coeruleus plays a role in REM sleep origin and maintenance [38, 44-47] and *in vivo* MRI changes in the locus coeruleus have been found in iRBD patients [48].

^{18}F -DOPA and ^{11}C -PK11195 PET detected no group-level abnormalities in either locus coeruleus or raphe nucleus in our iRBD patients, which might argue against early involvements of these structures. Still, it should be noticed that similar observations were present in PET studies of early stage PD patients. In these studies - monoaminergic and serotonergic reductions in tracer uptake in the thalamus preceded reductions in the locus coeruleus and raphe nucleus that did not occur until later disease stages [6, 7, 9]. Additionally, compensatory upregulation in the activity of the enzyme aromatic L-amino acid decarboxylase, which ^{18}F -DOPA K_i reflects, is reported in early stage PD [6, 7, 10] and this may well retain ^{18}F -DOPA K_i values within the normal range and thereby underestimate the true degree of dysfunction. Another

explanation for the apparent disparity between reduced ^{18}F -DOPA K_i values in thalamic axonal terminals but not in the cell bodies of the locus coeruleus and raphe nucleus could be impaired axonal transport leading to dying back of axons, an early abnormal phenomenon occurring in the terminals in PD [49, 50].

The clinical impact of early monoaminergic dysfunction in the thalamus in the presumed prodromal phase of Lewy-type α -synucleinopathies needs to be clarified in future studies. It might be an early sign of future development of cognitive dysfunction [51, 52] as noradrenergic innervation from the locus coeruleus is believed to facilitate prefrontal cortex function and so mediate executive behaviour [53].

Other extrastriatal regions of interest assessed in this study showed no significant decrease or increase at a group level. Previous ^{18}F -DOPA PET studies in early stage PD patients have occasionally observed increased uptake in the midbrain raphe, internal globus pallidus, anterior cingulate gyrus, and prefrontal cortex, which is interpreted as components of an early compensatory mechanism [6, 7, 10-12]. It might be that the group of iRBD patients examined in this study are more heterogeneous in terms of their prodromal disease stages (3.6 years (SD=3.4), this would decrease our power to detect subtle changes at this stage compared to a possible more homogenous study population like early PD patients.

Analysis of ^{11}C -PK11195 showed similar binding potentials in iRBD patients and controls in all regions of interest assessed (Figure 1). However, voxel-wise analysis, which enables the detection of more localised changes in tracer binding, particularly in large cortical areas, identified bilateral clusters of significantly raised levels of microglial activation in the occipital lobe (visual associative cortex) of iRBD patients compared to controls (Figure 2). This is a novel finding in iRBD, which is in line with observations in PD [15-17], and fits well with previous observations of diffuse occipital glucose hypometabolism [54-56] and reduced blood flow in iRBD patients [57]. Though, it should be noted that some studies did not observe this reduced occipital blood flow [58, 59]. Occipital neuroinflammation in iRBD may suggest the presence of local Lewy-type α -synucleinopathy and possibly reflect early signs of future cognitive changes. This view is supported by previous ^{11}C -PK11195 PET studies observing raised occipital microglial activation in patients with PD dementia [60, 61] and DLB [16]. However, the effective clinical relevance of our finding remains to be investigated. It should be noted that iRBD patients with no cognitive complaints often show abnormalities linked to visuospatial dysfunction and the occipital lobe on neuropsychological testing [62, 63]. In our cohort, the occipital neuroinflammation observed in iRBD patients did not correlate

with the MoCA score, moreover, similar levels of microglial activation was found between patients who drew the cube correct or incorrect in the MoCA visuoconstructional skill sub-item (Figure 2). Our findings could suggest that a raised level of microglial activation is not associated with cognitive decline at this prodromal stage of PD/DLB. However, it is also possible that we were underpowered to detect a significant correlation and that we need a larger study population with a wider range of MOCA scores. More specific and sensitive measures of visuospatial skills should be performed in future studies. Besides this, it should be mentioned that a previous observation in PD patients did not find an association between levels of microglial activation and clinical test scores [17], though, DLB patients in comparison to PD patients do have significantly higher levels of microglial activation [16], which could suggest that microglial activation is more strongly activated when cognition is severely impaired. Alternatively, occipital neuroinflammation at this prodromal stage could contribute to the vivid imagery of the dreams occurring in REM sleep, which is thought to be related to occipital lobe activation in REM sleep.[64].

This study has limitations which should be acknowledged: First, ^{18}F -DOPA PET enables us to obtain important knowledge

about the overall function of the monoaminergic innervation in the brain structures but does not allow examination of each separate component of monoaminergic neurotransmitter systems in brain structures with mixed dopaminergic, noradrenergic and serotonergic innervation. Second, ^{18}F -DOPA region of interest results might be prone to a statistical type I error since correction for multiple testing was not performed, yet our finding of reduced ^{18}F -DOPA in the thalamus is in line with previous observations in early stage PD patients and therefore most likely a genuine finding. Third, ^{11}C -PK11195 yields a lower specific to background signal ratio than newer translocator protein ligands; this might have reduced sensitivity to detect subtle increases in microglial activation. Nevertheless, we chose ^{11}C -PK11195 since it allows us to compare our observations with those of previous studies in Lewy-type α -synucleinopathies. Moreover, we avoid the issue of newer microglial tracers where translocator protein polymorphisms carried by individual subjects influence the tracer binding affinity, which potentially can lead to a selection bias. Finally, a relatively small number ($n=9$) of ^{18}F -DOPA controls were included.

In summary, this study observed reduced monoaminergic axonal terminal function in the thalamus of iRBD patients, a finding which suggests dysfunction of monoaminergic terminals projecting from the locus coeruleus and raphe nucleus. These

nuclei regulate REM sleep and are observed to contain Lewy-type α -synuclein pathology in iRBD patients and in the early developing disease phase of PD. Furthermore, clusters of significantly raised microglial activation were present bilaterally in the occipital lobe of our patients, a finding which might reflect early Lewy-type α -synuclein pathology and possibly an increased risk for later cognitive dysfunction and altered perception. These findings shed light on early disease processes in developing Lewy-type α -synucleinopathies, but the full clinical impact of these findings require clarification by adequate follow-up studies of these patients.

Contributors

NP, AI, DJB, ET, KØ, and MGS designed the study. MGS, AI, MS, MO, KBS, AG, DV, PP, JS, CG, and NP collected the data. All authors contributed to data analysis and the writing of the manuscript

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Tables

Table 1

Demographic and clinical characteristics

	iRBD patients (n=21)
Female / male	3 / 18
Age (years)	66.2 (6.3)
Time since iRBD diagnosis (years)	3.6 (3.4)
Unified Parkinson's Disease Rating Scale (part III)	3.4 (2.3)
Montreal Cognitive Assessment Score	25.7 (2.4)
MoCA score < 26 and <21	n=10 and n=1
SCOPA-AUT score	16.6 (8.1)
University of Pennsylvania Smell Identification Test	19.0 (7.5)

Values are mean and brackets indicate standard deviation.

iRBD = idiopathic REM sleep behaviour disorder

Table 2

^{18}F -DOPA Ki values and ^{11}C -PK11195 binding potentials in regions of interest in iRBD patients and control subjects

ROI	^{18}F -DOPA Ki value					^{11}C -PK11195 BP _{ND}		
	Controls	iRBD	Difference	Percentage change	p value	Controls	iRBD	p value
	Mean (95% CI)	Mean (95% CI)				Median	Median	
Locus coeruleus	0.00428 (0.00373 to 0.00483)	0.00423 (0.00378 to 0.00469)	-0.00005 (-0.00081 to 0.00071)	-1.2%	0.90	0.0830	0.0524	0.99
Median raphe	0.00543 (0.00408 to 0.00679)	0.00521 (0.00462 to 0.00580)	-0.00022 (-0.00140 to 0.00096)	-4.1%	0.70	0.160	0.2166	0.80
Dorsal raphe	0.00613 (0.00549 to 0.00677)	0.00581 (0.00524 to 0.00637)	-0.00032 (-0.00113 to 0.00061)	-5.3%	0.48	0.2304	0.2180	0.80
Hypothalamus	0.00514 (0.00454 to 0.00574)	0.00490 (0.00457 to 0.00524)	-0.00024 (-0.00085 to 0.00037)	-4.6%	0.43	0.0705	0.0180	0.46
Thalamus	0.00176 (0.00156 to 0.00195)	0.00149 (0.00135 to 0.00163)	-0.00026 (-0.00050 to -0.00002)	-15.0%	0.03	0.2048	0.2180	0.89
Globus pallidus internal	0.0034 (0.0029 to 0.0038)	0.0038 (0.0035 to 0.0042)	0.00046 (-0.0002 to 0.0011)	13.7%	0.14	0.1350	0.1390	0.89
Anterior cingulate gyrus	0.00196 (0.00170 to 0.00222)	0.00207 (0.00184 to 0.00229)	0.00011 (-0.00027 to 0.00048)	5.6%	0.55	0.1060	0.0883	0.37
Frontal lobe	0.00104 (0.00092 to 0.00115)	0.00107 (0.00095 to 0.00119)	0.00004 (-0.00016 to 0.00023)	3.6%	0.70	0.0484	0.0387	0.45
Occipital lobe						0.0314	0.0470	0.10

iRBD = idiopathic REM sleep behaviour disorder (n=21).

Controls ^{18}F -DOPA (n=9). Controls ^{11}C -PK11195 (n=20).

Figures

Figure 1

^{18}F -DOPA K_i values and ^{11}C -PK11195 binding potentials in regions of interest

Scatter-plot with individual subjects ^{18}F -DOPA K_i values in regions of interest, blue symbols are controls ($n=9$) and red symbols are iRBD patients ($n=21$), bar indicates the mean value of the group and whiskers are one standard deviation (A). ^{11}C -PK11195 BP_{ND} in regions of interest, blue symbols are controls ($n=20$) and red symbols are iRBD patients ($n=21$), bar indicates the median value of the group (B). All values are provided in Table 2.

Figure 2

Increased occipital ^{11}C -PK11195 BP_{ND} in iRBD patients compared to controls

Localisation of voxels with significantly increased ^{11}C -PK11195 BP_{ND} in the iRBD group compared to controls identified by Statistical Parametric Mapping analysis (A) and scatter-plot of individual ^{11}C -PK11195 BP_{ND} across the two significant clusters localised by Statistical Parametric Mapping analysis (B). Bar indicates the mean value. Red dots indicate iRBD subjects who drew the cube incorrect in the visuoconstructional skill test of the Montreal Cognitive Assessment.

Region	Peak-level				Cluster-level		
iRBD (n=21) compared to controls (n=20)							
	T-score	<u>Coordinates</u>			p-value	p-value	Cluster size
		X	Y	Z	(FWE-corrected at cluster level)	(uncorrected)	(voxels)
Left visual associative cortex	5.03	-24	-100	-10	0.000	0.000	11629
Right visual associative cortex	4.89	27	-95	-7	0.000	0.000	11521

Figure 3

Correlation between ^{18}F -DOPA Ki values in thalamus and whole striatum in iRBD patients

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