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α-SYNUCLEINOPATHY PHENOTYPES

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Abstract

α-Synucleinopathies are neurodegenerative diseases characterised by the abnormal accumulation of α-synuclein aggregates in neurons, nerve fibres or glial cells. While small amounts of these α-synuclein pathologies can occur in some neurologically normal individuals who do not have associated neurodegeneration, the absence of neurodegeneration in such individuals precludes them from having a degenerative α-synucleinopathy, and it has yet to be established whether such individuals have a form of preclinical disease. There are three main types of α-synucleinopathy, Parkinson’s disease (PD), dementia with Lewy bodies (DLB), and multiple system atrophy (MSA), with other rare disorders also having α-synuclein pathologies, such as various neuroaxonal dystrophies. Multiple clinical phenotypes exist for each of the three main α-synucleinopathies, with these phenotypes differing in the dynamic distribution of their underlying neuropathologies. Identifying the factors involved in causing different α-synuclein phenotypes may ultimately lead to more targeted therapeutics as well as more accurate clinical prognosis.
**Introduction**

The α-synuclein protein is small, soluble and highly conserved, with the predominant isoform being 140 amino acids long [1]. It has the ability to adopt different conformations depending on the environment, and interacts easily with other ligands such as lipids [2]. α-Synuclein is normally localised in the central nervous system, predominantly in the presynaptic nerve terminals [1] and plays important regulatory roles, including involvement in synaptic maintenance, mitochondrial homeostasis, proteasome function, dopamine metabolism and chaperone activity [2].

α-Synucleinopathy is a term used to describe a number of neurodegenerative diseases characterised by the abnormal accumulation of α-synuclein insoluble aggregates in neuronal or glial cells. Disorders that are collectively referred to as α-synucleinopathies include Parkinson’s disease (PD), dementia with Lewy bodies (DLB) and multiple system atrophy (MSA)(Figure 1), as well as rare disorders such as various neuroaxonal dystrophies. A much lower density of abnormal α-synuclein is also present in the nervous system of approximately 10% of neurologically normal elderly individuals over 60 years of age [3], with this figure doubling for studies using immunohistochemical methods [4]. These individuals do not have marked neuronal loss or gliosis [4]. It is currently unknown if this incidental pathology represents a preclinical α-synucleinopathy. However this seems likely based upon several prospective studies, which suggest an association with early non-motor manifestations such as rapid eye movement sleep behaviour disorder, hyposmia and symptoms of autonomic dysfunction such as constipation [5].
Figure 1 – Table illustrating the differences between the three main α-synucleinopathies, Parkinson’s disease (PD), dementia with Lewy bodies (DLB) and multiple system atrophy (MSA). DLB has an older age of onset on average and is a dominant dementia syndrome, while PD has early olfactory deficits and is responsive to L-dopa treatment, with a longer average disease duration. Both PD and DLB have intraneuronal α-synuclein inclusions and neurites, while MSA has glial cytoplasmic inclusions (GCIs), neuronal intranuclear inclusions (NNIs) and early autonomic features.

Photomicrographs are of α-synuclein immunoreactive structures counterstained with cresyl violet. NCI=neuronal cytoplasmic inclusions. Light blue highlights indicate the clinical attribute that distinguishes one disease from the other two diseases.
This review will examine the different phenotypes of the neurodegenerative α-synucleinopathies and discuss their clinical similarities, differences and any pathological correlations, as well as therapeutic considerations.

The most common pure α-synucleinopathy phenotype is Parkinson’s disease

Clinical considerations

Of the three main types of α-synucleinopathy listed above, the most commonly observed and diagnosed in clinical practice is PD (Figure 1). PD is considered a relatively homogeneous clinical syndrome diagnosed by the presence of two of four cardinal motor signs (bradykinesia, rigidity, resting tremor, gait instability) that are responsive to levodopa therapy [6]. Common non-motor signs include hallucinations, olfactory disturbances, pain, constipation, sleep disturbances, depression and dementia [7]. There is evidence that many non-motor signs can occur years before the manifestation of motor symptoms [7] and may represent some of the earliest clinical symptoms of PD. Olfactory dysfunction in particular, which occurs in approximately 90% of patients, is suggested to be a useful tool for early diagnosis [7]. Cognitive impairment is common in PD, and may present in a mild form early in the disease process, particularly involving deficits in executive function, problem solving and visuospatial skills [8]. Dementia and hallucinations are typically observed later in the disease course. In longitudinally followed patients, 83% were demented and 74% experienced visual hallucinations after twenty years disease duration [9].

The average age of disease onset in PD is around 60 years, with life expectancy about 15 years from diagnosis (Figure 1). The majority of early onset patients (those diagnosed under 40 years of age) have a positive family history of PD and long
disease durations, while older patients (diagnosed over the age of 70 years) tend to have shorter disease durations and more rapid disease progression [10].

The number of potential motor and non-motor clinical features that may occur in any given individual contribute to patient variability, with large clinical studies having assessed this variability in order to define subtypes of PD patients likely to share pathologic and genetic features [10]. As a result, four overlapping PD subtypes based on age of onset, severity and type of motor impairments, rate of progression, and presence or absence of significant cognitive impairment have been identified: earlier onset, tremor dominant, postural instability and gait dominant, and older onset (Table 1). The progression of pathology in these subtypes has been assessed and will be discussed next.

<table>
<thead>
<tr>
<th>Clinical</th>
<th>Early PD &amp;/or Tremor dominant</th>
<th>Older onset &amp;/or Post. instability/gait</th>
</tr>
</thead>
<tbody>
<tr>
<td>Age of Onset (years)</td>
<td>~60</td>
<td>~70</td>
</tr>
<tr>
<td>Duration (years)</td>
<td>~15</td>
<td>~5-10</td>
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<tr>
<td>Motor impairment</td>
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<td>more severe</td>
</tr>
<tr>
<td>Axial impairment</td>
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<td>yes</td>
</tr>
<tr>
<td>L-dopa responsive</td>
<td>Yes</td>
<td>Variable</td>
</tr>
<tr>
<td>Non-motor signs</td>
<td></td>
<td></td>
</tr>
<tr>
<td>Autonomic</td>
<td>Later</td>
<td>Earlier</td>
</tr>
<tr>
<td>Hallucinations</td>
<td>Later</td>
<td>Earlier</td>
</tr>
<tr>
<td>Cognitive impairment</td>
<td>Later</td>
<td>earlier</td>
</tr>
<tr>
<td>End-stage pathologies</td>
<td>Moderate LRP</td>
<td>Severe LRP + plaques ± tangles</td>
</tr>
</tbody>
</table>

**Pathological considerations**

In PD there are two essential pathological features required for diagnosis: loss of pigmented dopaminergic neurons in the substantia nigra pars compacta (SNpc) of the midbrain, and the presence of α-synuclein positive intracytoplasmic inclusions
and axons known as Lewy bodies (LBs) and Lewy neurites (LNs)\cite{6}, or collectively as Lewy-related $\alpha$-synuclein pathologies (LRP)\cite{1}\). (Figure 1).

The concept of the initiation and progression of LRP in PD was first investigated by Braak and colleagues, who proposed a staging scheme based on the topographical location of LRP in a large cohort \cite{11}. Initiation is hypothesised to occur in the peripheral mucosa and enteric nervous system, travel into the brain via the vagal and olfactory nerves, and progress to particular brain structures in a predictable pattern \cite{11}. This pattern begins in the caudal brainstem (dorsal motor nucleus of the glossopharyngeal and vagal nerves) and olfactory bulb (anterior olfactory nucleus)\( (\text{stage one}) \), ascending to the pontine tegmentum (stage two), midbrain (stage three), mesocortex and allocortex (stage 4), and finally culminating in widespread neocortical involvement (stages 5 and 6) \cite{11}. Prospective assessment of PD patients to autopsy reveals that the progression of LRP in typical cases (excluding early-onset cases) is consistent with Braak PD staging, where brainstem LRP dominates in those surviving to 5 years, by 13 years 50\% of cases have a transitional distribution to limbic regions, and by 18 years all have at least this pathological phenotype \cite{12}. It is thought that the earliest stages represent a preclinical disease phase, in the absence of any motor disability \cite{11}.

To further define the pathological variables, a number of recent clinicopathological studies have assessed the progression of pathology in the PD subtypes mentioned earlier (early onset, tremor dominant, postural instability and gait dominant and older onset) and observed subtle pathological differences (Table 1). In particular, the patterns and severity of LRP were found to be remarkably similar for both the early-
onset and tremor dominant groups, although the early-onset group had longer disease duration, suggesting that the rate of LRP formation may differ between earlier versus later onset PD. In patients with non-tremor dominant postural instability and gait dominated PD there was significantly more LRP and amyloid β plaques compared with tremor dominant or younger onset patients [13]. This suggests that the different burden of LRP deposition and the presence of amyloid β plaques correlate with a different clinical phenotype (Table 1), although the severity of amyloid deposition in such cases is less than that observed in DLB (Figure 2).

![Diagram illustrating the different pathological types of cases meeting clinical criteria for Parkinson’s disease with dementia (PDD), dementia with Lewy bodies (DLB) and Alzheimer’s disease (AD). Lewy-related pathologies (LRP) identified in sections from the temporal cortex and amygdala stained for α-synuclein immunohistochemistry and counterstained with cresyl violet. Plaques are identified in sections from temporal cortex stained for amyloid β immunohistochemistry and counterstained with cresyl violet. Neurofibrillary tangles (NFTs) identified in sections from the temporal cortex stained with a modified Bielschowsky silver stain.]

The older onset group of PD patients has a more complex disease course, with a
shorter disease duration and more rapid cognitive decline. Like the non-tremor
dominant postural instability and gait dominated PD group, they have been found to
have higher amounts of cortical LRP and additional neuropathologies, particularly
those associated with AD [13-15](Table 1). PD patients with dementia have up to 10-
fold higher amounts of cortical LRP compared to those without dementia, and there is
a correlation between the severity of LRP and AD pathology in these patients [13-15].
The more rapid disease course, along with higher amounts of pathological deposits
and additional neuropathologies, suggests an even faster rate of LRP deposition that
appears to be linked to multiple pathologies in older onset PD patients. These cases
have similarities to those described below with a dementia dominant phenotype
(Figure 2).

**How does dementia with Lewy bodies fit in?**

**Clinical considerations**

The second main α-synucleinopathy syndrome diagnosed by the presence of LRP is
DLB. It is differentiated from PD clinically by the predominance of dementia that
occurs as the presenting symptom or within the first year of PD [16]. Patients are
classified as PD with dementia (PDD) if dementia occurs a year or more after a
clinical diagnosis of PD [16], although in patients where the dementia and movement
disorder occur closely together, the distinction between PD and DLB can be difficult.
The separation between PDD and DLB is considered by some to be artificial since it
implies different disease mechanisms, which may ultimately have different
therapeutic targets.

Despite more refined diagnostic criteria for DLB [16], large autopsy studies indicate
that it is often misdiagnosed in the clinical setting [17], remaining under-diagnosed in milder cases of dementia and over-diagnosed in more severe cases of dementia [17]. Many older individuals also have more than one neurodegenerative syndrome. For this reason, the clinical diagnosis of DLB remains problematic and any clinical subtyping even more difficult without neuropathological confirmation.

**Pathological considerations**

There is little difference in the distribution or severity of LRP between PDD and DLB but many patients with DLB have substantial cortical amyloid deposition [18](Figure 2), much like the older-onset group of PD patients mentioned above. Three distinct pathological stages of DLB have been identified (brainstem, transitional and neocortical), which, when combined with grading Alzheimer-type pathology, give a clinical likelihood of DLB [16]. It is known that LRP occurs in the amygdala in many patients with clinically and pathologically confirmed AD [19], and as such it is proposed that these cases should be considered another distinct clinicopathological entity (Figure 2).

Clinicopathological studies of cases with dominant dementia and LRP have noted substantial pathological variability and assessed their clinical correlates. Four distinct pathological phenotypes can be identified (Figure 2): pure DLB, DLB plus AD, AD plus amygdala LRP and pure AD [20-22]. These studies have confirmed the presence of LRP in the amygdala in approximately 60% of AD cases, and that the vast majority of LRP cases with dominant dementia have amyloid deposition. This clinicopathological approach has established that pure pathological DLB, despite the presence of substantial plaque pathology, has a clinical picture closer to that of PDD.
than to AD, with initially more executive dysfunction rather than memory deficits [20]. Patients with DLB plus AD experience a more rapid decline and shorter disease duration, and patients with AD plus amygdala LRP have high rates of depression [23]. These studies demonstrate that several pathological factors influence the clinical phenotype of these dementia cases, including the infiltration and severity of LRP and the coexistence of AD pathologies.

Multiple system atrophy is very different

Clinical considerations

MSA is the third main α-synucleinopathy, with some fundamental differences in clinical and pathological presentation to the other α-synucleinopathies, PD and DLB. MSA patients are diagnosed at a similar age as PD (around 60 years old), but have significantly shortened disease duration (6-9 years)(Figure 1).

MSA may be divided into two clinical phenotypes. Patients who present predominantly with parkinsonian symptoms (including bradykinesia, rigidity, tremor and postural instability) are classified as MSA with parkinsonism (MSA-P)[24]. Patients who present predominantly with a cerebellar syndrome are classified as MSA with cerebellar signs (MSA-C). In MSA-C the most common symptom is gait ataxia, which often occurs together with speech and limb ataxia, and cerebellar oculomotor dysfunction [24]. There is no difference in disease duration between MSA-P and MSA-C. Both phenotypes manifest considerable changes in dominant clinical symptoms over time, which may eventually result in a different classification [24].
Autonomic symptoms are common to both variants and involve the gastrointestinal, cardiovascular and urogenital systems. Bladder and erectile dysfunction have been identified as some of the earliest clinical signs, although additional symptoms include dysphagia, bowel dysfunction and orthostatic hypotension [24]. Cognitive impairment has recently been recognised as a feature of MSA, having previously been regarded as an exclusion criteria for diagnosis [24]. Clinicopathological studies have identified some degree of cognitive disability in 14-18% of cases [25, 26] and found it to be more severe and widespread in patients with MSA-P than in patients with MSA-C. Due to the overlap of symptoms, PD and MSA are commonly misdiagnosed, however MSA symptoms respond poorly, if at all, to treatment with levodopa [27](Figure 1).

**Pathological considerations**

The pathological hallmark of all clinical subtypes of MSA is the presence of α-synuclein-positive glial cytoplasmic inclusions (GCIs) in oligodendroglia (Figure 1), observed in a widespread distribution throughout the brain (Figure 3). In addition to GCIs, α-synuclein-positive neuronal cytoplasmic, neuronal nuclear and glial nuclear inclusions may be observed in MSA [28](Figure 1). The clinical subtypes of MSA, MSA-P and MSA-C are generally reflective of the brain regions with significant pathological change (Figure 3). In MSA-P, the striatonigral regions are predominantly affected (Figure 3), while in MSA-C it is the olivopontocerebellar regions (Figure 3), however there may be considerable overlap [28]. The greatest severity of GCIs are found in the putamen, pons, motor cortices and underlying white matter, as well as the cerebellar white matter (Figure 3). Degeneration in striatonigral or olivopontocerebellar regions is seen in association
with the GClIs (Figure 3). Neuronal loss is observed in a number of brain regions including the putamen, SNpc, pons and cerebellar Purkinje cells (Figure 3). Grading systems have been proposed with cases assigned a separate severity grade between 0-3 for both MSA-P and MSA-C pathology, reflecting the considerable overlap between these subtypes (Figure 3). For MSA-P, grade I pathology is mainly confined to the SNpc, with slight changes in the putamen. There is an increase in the severity of pathology in both regions by grade II, with mild changes in the caudate and external globus pallidus. By grade III pathology is severe in the striatum with substantial neuronal loss in the globus pallidus and caudate [29]. For MSA-C, grade I has minimal loss of Purkinje cells accompanied by myelin pallor in the cerebellum with variable cell loss in the SNpc. Grade II involves widespread pathology in the cerebellum, minimal changes in the pons and inferior olives, and again variable cell loss in the SNpc. By grade III considerable changes are observed throughout the cerebellum, pons and inferior olives, and again there is variable degeneration of the SNpc [29]. Supporting the idea that the clinical phenotype reflects the distribution of pathology, one study found more severe bradykinesia correlated with striatonigral degeneration, and cerebellar signs correlated with more frequent olivopontocerebellar degeneration [30]. However, grading of neuronal loss revealed that striatonigral and olivopontocerebellar regions were equally affected in almost half of the cases [30]. This may represent a pathological dynamic reflective of the overlap of symptoms between clinical phenotypes, which over time may be significant enough to warrant a change in classification.
Figure 3 – Photomicrographs of the substantia nigra (SN in A-D), putamen (E-H) and cerebellum (I-L) of representative controls (A, E, I) and cases with early and mild (B, F, J) or late and severe (C, G, K) MSA. Cases with MSA-P have substantial degeneration in the putamen (F, G) in addition to the substantia nigra (B, C), while cases with MSA-C have substantial degeneration in the cerebellum (J, K) in addition to the substantia nigra (B, C), as shown in haematoxylin and eosin stained sections. α-Synuclein immunoreactive glial cytoplasmic inclusions are observed in all regions of neurodegeneration in MSA (D, H, L). cp=cerebral peduncle.
Conclusions

α-Synuclein deposition is considered the basis for neurodegeneration in all three α-synucleinopathies. Despite similarities in the progression of spread there are significant pathological differences between the Lewy body diseases and MSA, including the cell type involved (neurons in PD and DLB and oligodendroglia in MSA) and the degree of neuronal loss (only in selected regions in PD but widespread throughout many regions in MSA). These fundamental differences suggest the involvement of distinct pathological mechanisms in the progression of both diseases, possibly involving another member of the synuclein family that has not been fully investigated.

Further studies are required on all α-synucleinopathy clinical phenotypes in order to understand the clinical and pathological associations more fully. In particular, it is important that the subtleties of the pathological differences observed in the clinical PD phenotypes be further clarified in order to develop more targeted treatments appropriate for the predominant symptom, in addition to giving disease sufferers a more accurate disease prognosis. Working to improve the clinical recognition of DLB will enable further establishment of the relationships between the DLB pathological subtypes to the progression of clinical disease. Efforts to reconcile the clinical and pathological subtypes of MSA due to the dynamics of symptom shift is another research challenge in this area.

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References