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Review: Sporadic Parkinson's disease: development and distribution of α-synuclein pathology

K. Del Tredici and H. Braak

Clinical Neuroanatomy Section, Department of Neurology, Center for Biomedical Research, University of Ulm, Ulm, Germany

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Sporadic Parkinson's disease: development and distribution of α-synuclein pathology

The development of α-synuclein immunoreactive aggregates in selectively vulnerable neuronal types of the human central, peripheral, and enteric nervous systems is crucial for the pathogenesis of sporadic Parkinson's disease. The presence of these lesions persists into the end phase of the disease, a process that is not subject to remission. The initial induction of α-synuclein misfolding and subsequent aggregation probably occurs in the olfactory bulb and/or the enteric nervous system. Each of these sites is exposed to potentially hostile environmental factors. Once formed, the aggregates appear to be capable of propagating trans-synaptically from nerve cell to nerve cell in a virtually self-promoting pathological process. A regional distribution pattern of aggregated α-synuclein emerges that entails the involvement of only a few types of susceptible and axonally interconnected projection neurons within the human nervous system. One major route of disease progression may originate in the enteric nervous system and retrogradely reach the dorsal motor nucleus of the vagal nerve in the lower brainstem. From there, the disease process proceeds chiefly in a caudo-rostral direction through visceromotor and somatomotor brainstem centres to the midbrain, forebrain, and cerebral cortex. Spinal cord centres may become involved by means of descending projections from involved lower brainstem nuclei as well as by sympathetic projections connecting the enteric nervous system with postganglionic peripheral ganglia and preganglionic nuclei of the spinal cord. The development of experimental cellular and animal models is helping to explain the mechanisms of how abnormal α-synuclein can undergo aggregation and how transmission along axonal connectivities can occur, thereby encouraging the initiation of potential disease-modifying therapeutic strategies for sporadic Parkinson's disease.

Keywords: α-synuclein, autophagy, central nervous system, chaperones, dorsal motor nucleus of the vagal nerve, enteric nervous system, Lewy pathology, Parkinson's disease, peripheral nervous system, protein aggregation/misfolding

Introduction

Sporadic Parkinson's disease (PD) is a widespread human disease [1–4] that has not been reported in nonhuman vertebrates. It involves circumscribed portions of the central, peripheral, and enteric nervous systems (CNS, PNS and ENS) (Table 1; Figure 1) [5–13]. A protracted but progressive and irreversible pathological process that usually manifests itself clinically in late midlife and lasts for decades accompanies the disease (Table 2).

Despite the more obvious motor symptoms (initially asymmetric hypokinesia, cogwheel rigidity, resting

Correspondence: Kelly Del Tredici, Clinical Neuroanatomy (Department of Neurology), Center for Biomedical Research, University of Ulm, Helmholtzstrasse 8/1, 89081 Ulm, Germany. Tel: +49 731 500 63114; Fax: +49 731 500 63133; E-mail: kelly.del-tredici@uni-ulm.de

Table 1. Distribution of Lewy pathology in the enteric, peripheral, and central nervous systems together with clinical symptoms

Stage 1	Initial development of LNs somewhere in ENS, PNS or CNS
	LNs and LBs in olfactory bulb and anterior olfactory nucleus
	LNs and LBs in dorsal motor nucleus of the vagal nerve, intermediate reticular zone
	Absence of symptoms; then a phase of prodromal symptoms, for example hyposmia, autonomic dysfunction
Stage 2	LNs in ENS, in peripheral parasympathetic and sympathetic nerves, and in peripheral autonomic ganglia
	LNs and LBs in medullary nuclei of the level setting system, for example lower raphe nuclei, locus coeruleus
	Hyposmia, autonomic dysfunction, for example gastrointestinal, urinary symptoms
	Disturbed sleep, parasomnias, mood changes
Stage 3	LNs and LBs in tegmental pedunculopontine nucleus and substantia nigra, pars compacta
	LNs in spinal cord centres reached by descending projections of level setting nuclei
	LNs and LBs in upper raphe nuclei, magnocellular nuclei of the basal forebrain, hypothalamic tuberomammillary nucleus
	LNs and LBs in central subnucleus of the amygdala
	Disturbed sleep and possible early phase motor dysfunction: asymmetric tremor, rigidity, hypokinesia
Stage 4	LNs and LBs in midline and intralaminar nuclei of the thalamus
	LNs and LBs in anteromedial temporal cortex (transentorhinal and entorhinal regions, hippocampal formation, plexus of LNs in the second sector of the Ammon's horn)
	Early phase motor dysfunction: tremor, rigidity, hypokinesia
Stage 5	LNs and LBs in superordinate cortical areas for regulation of autonomic functions
	LNs and LBs in high-order sensory association areas and prefrontal fields
	Late phase motor disability: flucutation, falls, wheelchair bound or bedridden
	Cognitive impairment
Stage 6	LNs and LBs in first-order sensory association areas and premotor fields
	LNs and LBs in primary sensory and primary motor areas
	Late phase motor disability: fluctuation, wheelchair bound or bedridden
	Cognitive impairment, dementia

tremor, postural instability) that are associated with clinical phases of the disease, an accurate interpretation of the early prodromal dysfunctions [14, 15] and the differential diagnostics of PD as opposed to other forms of parkinsonism are complicated [16]. For this reason, autopsy-based assessment is required to confirm the clinical diagnosis and to identify individuals with early PD-related neuropathology in the broader population [17–20].

α-Synuclein and Lewy pathology

Sporadic PD still is defined neuropathologically by the continuous formation of α-synuclein immunoreactive inclusion bodies [21–28], which develop only in a few especially susceptible neuronal types within circumscribed portions of the CNS, PNS, and ENS [29–32]. The inclusions occur in the form of spherical Lewy bodies (LBs) in cell somata [33–36], as elongated spindle-shaped or thread-like Lewy neurites (LNs) in axons and dendrites [37–40], as pale bodies [25], and sometimes as particulate (granular, dot-like, punctate) aggregates [41, 42]. These lesions, subsumed under the rubric 'Lewy pathology' (LP), very likely occupy a central role in the pathogenesis of the disease [34, 35, 43–49]. Individuals who lack LP at autopsy, but who displayed symptoms resembling those seen in PD, have atypical parkinsonian disorders other than sporadic PD [50].

The natively unfolded protein α-synuclein is present in erythrocytes and platelets [51, 52] but is a predominantly neuronal protein that occurs in most, but not all, types of mature human nerve cells [53]. It is produced in the nerve cell soma and, provided it is fully functional, is redistributed from there into the axonal compartment and ultimately resides in presynaptic terminals, where it may contribute to neurotransmission and synaptic homeostasis [54–60]. The presence of a hydrophobic NAC region within the α-synuclein molecule distinguishes it from related β- and γ-synucleins, confers it with a β-sheet potential, and is required for abnormal conformational changes that can induce the process of aggregation [61–63]. It has been proposed that, in normal nerve cells, natively monomeric α-synuclein exists in dynamic equilibrium with helical tetramers that appear to be more resistant to aggregation than natively unfolded monomers [64–67], but the evidence for a predominantly tetrameric state has been challenged [68–70]. Specific types of chaperones are present in the preterminal axoplasm and appear to regulate the functional relationships between tetrameric α-synuclein and synaptic vesicle membranes [65].

Thread-like LNs generally are the first lesions that are detectable in involved regions [31, 71–74]. In many instances, it is unclear whether the filamentous

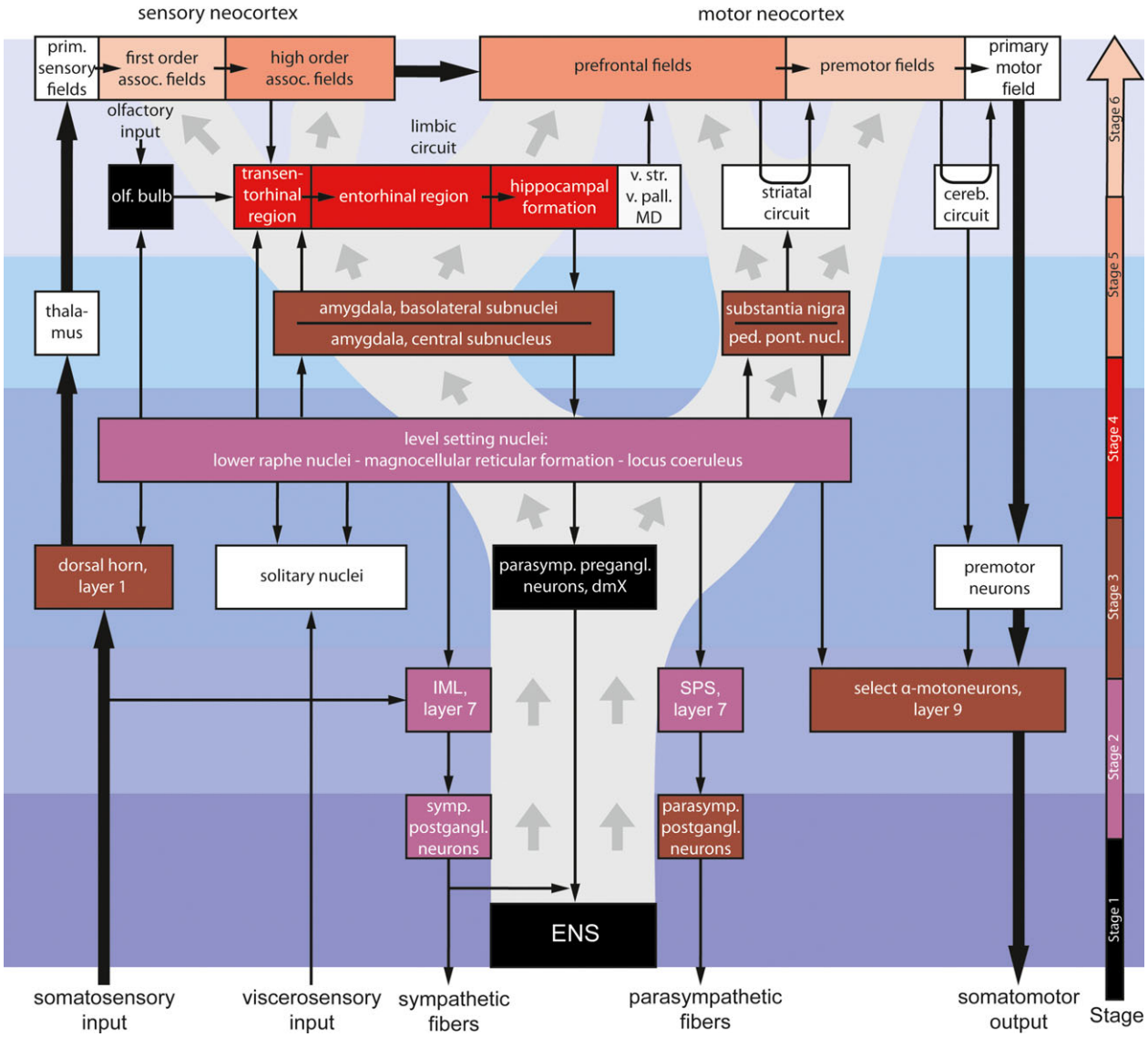

Figure 1. Diagram showing possible routes for the transmission of α-synuclein pathology within the human nervous system between stages 1 and 6 of the PD-associated pathological process. Four degrees of blue background shading indicate the enteric and peripheral nervous systems (dark blue), portions of the lower brainstem and spinal cord (medium blue), structures in the upper brainstem, thalamus and amygdala (light blue) and the cerebral cortex (sky blue). Initial predilection sites during stage 1 are the olfactory bulb (black), anterior olfactory nucleus and dorsal motor nucleus of the vagal nerve (black). In the brain, the disease process progresses in a chiefly caudo-rostral trajectory. At stage 2, lesions are seen for the first time in the level setting nuclei: the locus coeruleus, magnocellular nuclei of the reticular formation, and lower raphe nuclei (violet). A bifurcation of the pathological process may occur during stage 3 (brown). The pathological process reaches centres of the central autonomic network (central subnucleus of the amygdala, components of the limbic circuit) and progresses into centres of the somatomotor system (pedunculopontine tegmental nucleus and pars compacta of the substantia nigra). The cerebral cortex first becomes involved at stage 4, beginning with the temporal transentorhinal region (dark red) and reaches, thereafter, the entorhinal region and the Ammon's horn (dark red). The higher order association areas of the neocortex become involved in stage 5 (red), followed by the first-order association areas and primary fields in stage 6 (light red). The spinal cord becomes involved during stage 2 possibly via descending projections from involved lower brainstem nuclei (violet) as well as by sympathetic projections connecting the enteric nervous system (black) with postganglionic peripheral ganglia (violet) and spinal cord preganglionic nuclei (violet). Prim. sensory fields, primary sensory fields; first-order assoc. fields, first-order association fields; olf. bulb, olfactory bulb; v. striatum, ventral striatum; v. pallidum, ventral pallidum; MD, mediodorsal nuclei of the thalamus; cereb. circuit, cerebellar circuit; ped. pont. nucl., pedunculopontine tegmental nucleus; parsymp. pregangl. neurons, dmX, parasympathetic preganglionic neurons of the dorsal motor nucleus of the vagal nerve; IML, intermediolateral nucleus; SPS, sacral parasympathetic nucleus; symp. postgangl. neurons, sympathetic postganglionic neurons; ENS, enteric nervous system.

Table 2. Nonselected autopsy cases (n = 2366) showing sporadic PD stages in different age groups by decade

	LP	*Stage*						
Age	*0*	*1*	*2*	*3*	*4*	*5*	*6*	*Totals*
0–9	7	0	0	0	0	0	0	0
10–19	22	0	0	0	0	0	0	0
20–29	66	0	0	0	0	0	0	0
30–39	95	0	0	0	0	0	0	0
40–49	170	0	0	0	0	0	0	0
50–59	316	3	4	2	1	0	0	10
60–69	465	5	2	8	2	5	0	22
70–79	498	11	10	15	17	12	1	66
80–89	450	10	13	14	14	16	8	75
90–100	90	4	0	4	3	3	0	14
Totals	2179	33	29	43	37	36	9	187

LP, Lewy pathology.

lesions are dendritic or axonal inclusions; however, inasmuch as in mature neurons α-synuclein is localized chiefly in the axon, it is possible that the initial thread-like lesions preferentially develop in preterminal portions of the axonal compartment [75], thereby leaving uninvolved only the presynaptic terminals and the initial axon segment [31]. This means that the protein would have to undergo a slight relocalization prior to its aggregation within the axon. The very first small aggregates may possibly be transported in an anterograde as well as retrograde direction but rapidly consolidate to form larger LNs that remain stationary in the axon.

Shortly after the appearance of axonal LNs, additional aggregates develop and rapidly accumulate in the *somatodendritic compartments* of involved neurons. The sources of this abnormal material are unclear. The initial aggregation process most likely results in the reduction of monomeric α-synuclein in the axon, a state that could rapidly induce renewed production of monomers in the soma, which then – at the wrong location – form oligomers, insoluble protofibrils and, subsequently, insoluble fibrillar somatic LBs and dendritic LNs [58, 76–78]. In addition to α-synuclein, the aggregates can contain a variety of other components, including ubiquitin, tyrosine hydroxylase, synaptophysin, heat shock and autophagosomal proteins [79–86].

The insoluble intra-axonal and cytoplasmic aggregates possess amyloid characteristics [87]. More importantly, they are resistant to proteasomal recycling, autophagy or other endogenous cellular removal mechanisms [86, 88–94]. The larger spindle-shaped axonal aggregates, which can also be connected to each other, are distributed across extensive portions of the axon, and do not shift in an anterograde or retrograde direction [40, 73, 74, 95–97]. These spindles likely reduce normal axonal transport functions [73, 96, 98, 99].

Postmitotic neurons suspend their cell cycle for purposes of survival [100] and possess numerous surveillance mechanisms for repairing transient damage and eliminating defective proteins [101, 102]. For these reasons, it is unlikely that such cells would 'permit' spontaneous aggregation of α-synuclein molecules that subsequently cannot be degraded or exocytosed. Indeed, spontaneous aggregation of α-synuclein barely occurs under optimal experimental conditions, where amyloid-like fibrils develop only after prolonged incubation [56, 69, 103]. Moreover, were the natively unfolded protein to be present in tetrameric form, its tendency to resist aggregation would be, presumably, even greater. Because there are major differences between the axonal and somatodendritic compartments, the mechanisms by which axonal compartments deal with misfolded and aggregated proteins merit closer study.

Just as there are mechanisms to repair damage in postmitotic nerve cells or to stabilize partially folded protein intermediates [104, 105], mechanisms might also exist that support, and possibly accelerate, the processes leading to the production of abnormal α-synuclein aggregates. Instead of guardian chaperones, for instance, endogenous *or* exogenous proteins could 'coax' or 'mislead' the protein α-synuclein into undergoing conformational changes and then building pathological aggregates [106]. Such putative molecules possessing what we would call 'Siren-like' functions may waylay the host nerve cells during an assault on axons and thereby annul the otherwise unlikely scenario of spontaneous aggregation [107].

Specific types of nerve cells develop PD-associated Lewy pathology during the prodromal and clinical disease phases

As pointed out previously, very few types of nerve cells within the entire human nervous system are capable of developing LP, whereas other neuronal types, including directly adjacent nerve cells, remain intact for the

duration of the illness. Only projection cells with a disproportionately long, thin, and weakly myelinated or nonmyelinated axon become involved; by contrast, interneurons and projection cells with short axons or those with a heavily myelinated axon are not susceptible [30, 31, 108]. The specific regional distribution of the lesions within portions of the nervous system is a mirror of this selective vulnerability.

Because LP formation is not limited to dopaminergic projection neurons of the substantia nigra but also occurs in glutamatergic, noradrenergic, serotonergic, histaminergic and cholinergic projection cells, neurotransmitters *per se* are not adequate predictors of which neurons are predisposed or resistant to the pathological process. During the past decade, PD has come to be recognized as a multisystem disorder rather than a chiefly monosystemic disorder with selective dysfunction and loss of nigral neurons [11, 12, 20, 109–113].

The combination of especially susceptible neuronal types and the peculiarities of their distribution pattern make it possible not only to differentiate sporadic PD from other synucleinopathies but also to recognize mild forms of the pathological process in nonsymptomatic individuals. Those lacking clinically manifest motor symptoms but displaying LP in the same neuronal types and at the same sites as PD patients most likely represent individuals in a prodromal phase of PD [19, 112, 114–130].

Cases in an early phase are exceptionally valuable because they are temporally close to the prevailing circumstances at the outset of the disease process. Thus, evaluation of this phase is crucial to all other efforts directed at replacing currently symptomatic treatments with potentially causally effective therapies [131–135].

The pathological process underlying PD consists of a very early nonsymptomatic period, followed by a prodromal phase, often characterized by olfactory dysfunction [118, 136–144], autonomic dysregulation [145–156], as well as sleep, and/or mood disturbances [134, 157–159]. This phase transitions into a phase that is accompanied by the classical somatomotor symptoms and impaired cognitive functioning (Table 1) [2, 139, 159–166]. The duration of each phase varies considerably from one individual to another [134, 139, 167–169]; nevertheless, by the time of the initial PD diagnosis, when patients have crossed the threshold to clinically manifest somatomotor disease, the pathological process within the nervous system is already remarkably advanced (Table 1).

Interindividual differences with respect to disposition, pre-existing comorbidities, and environmental influences most probably have an impact on the threshold at which the conversion to the final phase occurs as well as on the reported phenotypical heterogeneity of initially presenting symptoms, disease course, and rates of disease progression [169–172]. Grading according to six neuropathological stages can incorporate all three phases, provided the assumption is correct that nonsymptomatic, prodromal, and symptomatic motor phases combined represent a biological continuum [19, 20, 31, 71, 112, 134, 171, 173–177].

Regional distribution pattern of the α-synuclein lesions within the nervous system in PD

Within the brain, the nonrandom development of the pathological process generally progresses caudorostrally through susceptible regions of the lower brainstem (the dorsal motor nucleus of the vagal nerve, lower raphe nuclei, magnocellular nuclei of the reticular formation, locus coeruleus) into midbrain tegmental nuclei (pedunculopontine nucleus, dopaminergic projection neurons of the substantia nigra, nuclei of the upper raphe system) and noncortical centres of the forebrain (amygdala, hypothalamic tuberomammillary nucleus, magnocellular nuclei of the basal forebrain, midline and intralaminar nuclei of the thalamus). Then, it reaches the cerebral cortex (transentorhinal and entorhinal regions, Ammon's horn, cortical visceromotor areas, and finally the entire neocortex) (Figure 1) [47, 71, 134, 173, 176–178].

Cases with the earliest signs of α-synuclein immunoreactivity frequently display LP both in anterior olfactory structures (that is, in mitral cells and tufted cells of the olfactory bulb and in projection neurons of the anterior olfactory nucleus) and in preganglionic parasympathetic neurons of the dorsal motor nucleus of the vagal nerve, including the adjoining intermediate reticular zone, during stage 1 [19, 31, 42, 71, 137, 138, 179, 180]. Thread-like LNs appear in unmyelinated long and thin axons that course in small bundles obliquely through the medulla oblongata and as spindle-shaped dendritic LNs and globular somatic LBs in visceromotor projection neurons of the vagal dorsal

motor nucleus. The motor cells in the ambiguus nucleus that send well-myelinated thick axons to striated muscles of the proximal oesophagus are spared [71, 179].

The nerve cells within the olfactory epithelium have not been reported to develop α-synuclein aggregates [181]; but even if the contrary were to prove true, the presence of subtle LP might well escape neuropathological detection because the nerve cells there are constantly being replaced and defective or damaged neurons are subject to removal from the functional neuroepithelium. This region, which is directly connected to the CNS and, at the same time, provides direct access to the upper respiratory tract, has effective mechanisms for repulsing invading pathogens [182].

It should also be noted that stage 1 involvement can be confined to the olfactory bulb alone [31, 175, 183]. For this reason, the possibility cannot be excluded that a toxin or pathogen may penetrate beyond the olfactory epithelium into the olfactory bulb, where it initiates the formation of α-synuclein aggregates or, alternatively, whence α-synuclein aggregation is induced in more distant interconnected regions [167, 184–188]. The site or region where the earliest aggregates occur is important for potential therapeutic strategies. In this context, the intimate spatial relationships between the nervous system and external as well as internal surfaces of the body (CNS – olfactory epithelium, ENS – gastrointestinal lumen) as potential entry points for environmental toxins and pathogens bears repeating [106, 189]. Swallowed nasal secretions, for example, come directly into contact with the upper gastrointestinal tract [78, 139, 167, 168, 190, 191].

The pathological process in the brain progresses into the serotoninergic nuclei of the lower raphe system, the magnocellular portions of the reticular formation, and the noradrenergic locus coeruleus in stage 2 [31, 71, 192]. Descending projections from all of these 'level setting' nuclei regulate medullary and spinal cord centres for somato- and viscerosensory input and viscero- and somatomotor output (Figure 1) [193–195]. Short and especially strong projections reach the dorsal motor nucleus of the vagal nerve. Spinal cord involvement begins during stage 2 with spindle-shaped axonal LNs in long descending projections generated from the involved supraspinal level setting nuclei that terminate in layer 7 preganglionic sympathetic (IML) and parasympathetic (SPS) nuclei (Figure 1) [7, 13, 19]. This sequence of spinal cord involvement indicates that aggregated α-synuclein could progress in an anterograde direction to spinal cord axonal networks in lateral portions of layer 7 (visceromotor autonomic system; IML, SPS) [13, 196, 197].

During stage 3, the somatodendritic compartments of neurons in the spinal cord that receive supraspinal projections from the level setting nuclei develop LP: dorsal horn layer 1 projection neurons (nociception) [198] and layer 9 ventral horn α-motoneurons, including the motoneurons of Onuf's nucleus (somatomotor system) (Figure 1). As in stage 2, the pathology could progress anterogradely and trans-synaptically into the somatodendritic compartments of vulnerable nerve cells [13]. A route that would involve retrograde axonal transport of the lesions to level setting nuclei and to layer 1 of the dorsal horn could originate in the plexus of the ENS. From there, aggregated α-synuclein could spread to preganglionic sympathetic neurons of the IML in layer 7 via postganglionic sympathetic projection neurons located in the coeliac ganglion (Figure 1) [7, 19, 198, 199].

In the brain, the caudo-rostral trajectory extends in stage 3 towards the mesencephalic tegmentum and basal portions of the prosencephalon – at this point, the pathological process bifurcates, entering, first, the central subnucleus and, subsequently, the basolateral subnuclei of the amygdala (Figure 1). The central subnucleus of the amygdala is reciprocally connected with the level setting nuclei and with the vagal dorsal motor nucleus (Figure 1) [37], thereby providing dense supervening limbic input to these supraspinal nuclei [200, 201]. Essential somatomotor centres also become involved at this stage, namely, the tegmental pedunculopontine nucleus [202] and the pars compacta of the substantia nigra (Figure 1). The pathological process in the substantia nigra may be induced via its interconnectivities with the central subnucleus of the amygdala or via direct coeruleo-nigral and pedunculopontine-nigral projections [203, 204]. During stage 3, LP also develops in the serotoninergic nuclei of the upper raphe system, the histaminergic tuberomammillary nucleus and the cholinergic magnocellular nuclei of the basal forebrain [31, 71, 173].

The routes described up to this point are also used by neurotropic viruses (e.g., pseudorabies) which, in experimental animals, have been inoculated into the gastric epithelium and progress sequentially through the ENS, the vagal nerve (including the dorsal motor

nucleus) and the nuclei of the level setting system to arrive at the central subnucleus of the amygdala and beyond [167, 205–209]. Nonetheless, the viruses utilized reach all of these sites in a matter or days [210], whereas the PD-related pathological process takes years to do so [139, 211].

Additional regions of the forebrain and, for the first time, a specific portion of the cerebral cortex, the transentorhinal region, become involved in stage 4. This specialized region of the cortex mediates between the temporal allocortex (hippocampal formation and entorhinal region) and the high-order association areas of the temporal neocortex. The region is highly developed only in higher primates, and above all in humans [212, 213]. The fact that the region is lacking in most experimental animals is probably the reason why the routes taken by the viral agents do not progress beyond the level of the amygdala [188]. By contrast, in humans, the transentorhinal region develops the most severe LP within the entire cerebral cortex [31].

The locus coeruleus, the basolateral nuclei of the amygdala, and the magnocellular nuclei of the basal forebrain are bidirectionally connected with the transentorhinal region, and these connectivities could lead the pathological process to the cortex. As if through a bottle-neck, the data stream from neocortical high-order sensory association areas must transit through the transentorhinal/entorhinal regions and, from there, is transferred via the perforant path to reach the hippocampal formation (Figure 1) [214, 215]. Via the perforant path and, additionally, along axons generated from the tuberomammillary nucleus to the second sector of the Ammon's horn, the pathological process could enter the hippocampal formation [31, 39].

The fullest topographic extent of the PD-related process is reached during stages 5 and 6. Beginning with the transentorhinal region, LP gradually penetrates the entire neocortex [31, 116, 216]. During stage 5, the lesions develop not only in all high-order sensory association areas but also in prefrontal fields of the neocortex (at first, in superordinate fields that process interoceptive data and regulate visceromotor functions). In stage 6, the pathological process finally makes inroads into the neocortical first-order sensory association areas, premotor fields, and even the primary sensory and motor fields (Figure 1) [31, 71]. The small- to medium-sized pyramidal cells that contain α-synuclein aggregates occur chiefly in neocortical layers V and VI [160, 217].

It still is unknown whether the involved pyramidal cells generate cortico-striatal or cortico-cortical projections, or both. The progression of the disease process throughout the neocortex, however, appears to take place via cortico-cortical projections directed from high-order association areas (that is, the prefrontal fields) to first-order association areas (that is, premotor fields) and, from there, to primary areas. These projections possess diffuse synaptic contacts in neocortical layers I–III and V–VI but very few in layer IV [218–220]. Involvement of these interconnectivities is consistent with the fact that layer IV remains virtually intact in PD, whereas LP develops mostly in layers V and VI. Thus, we have been inclined to view the disease process within the cerebral cortex as most likely progressing chiefly via these cortico-cortical pathways, supported additionally by coeruleo-cortical projections and projections from upper raphe nuclei and magnocellular nuclei of the basal forebrain (Figure 1).

Developing new models of α-synuclein spreading along axonal connectivities

We formerly speculated that an environmental or neurotropic pathogen might adhere to the mucous membrane of the upper gastrointestinal tract and/or nose and penetrate into neuronal processes of neurons there, where it might trigger a conformational change in α-synuclein. Then, via transneuronal and retrograde axonal transport along unmyelinated axons of the vagal nerve, α-synuclein aggregates from the ENS could reach the preganglionic neurons of the dorsal motor nucleus of the vagal nerve [116, 167, 168, 210, 211, 221–224]. Inasmuch as the vagus nerve exerts its influence chiefly on the distal portions of the oesophagus and following proximal portions of the stomach [225–227], these regions are among the more likely sites for a transfer and propagation of small α-synuclein aggregates between interconnected neurons. Indeed, we recently reported a case in which LP was present only in the olfactory bulb and in the submucosal plexus of the lower oesophagus [13]. Of course, it is impossible to extrapolate to a general rule from isolated occurrences [12, 228, 229]. At the same time, however, experimental models have made it possible to detect seeding mechanisms [230, 231] and spreading

over considerable distances by α-synuclein aggregates from the periphery via the vagus nerve to the CNS following intragastric, intraduodenal, and peripheral vagal nerve innoculations [199, 232–234]. There is some experimental evidence for retrograde axonal transport from the PNS to the CNS [235], but the connectivities between the ENS and the CNS via the vagus nerve probably play a pivotal role in the progression of PD [236, 237].

Experimental models involving the ENS and CNS have received additional support from a very recent large-scale epidemiologic evaluation of vagotomies that were performed in the past to treat peptic ulcers [238]. Full vagotomy is defined by resection of both vagal trunks, whereas selective vagotomy entails resection of only terminal branches supplying the fundus and corpus of the stomach [238–240]. The results of the study illustrate that the risk of having developed sporadic PD at follow-up 10 years later was significantly reduced in individuals who had undergone full truncal vagotomy but remained nearly similar to the risk of the general population in individuals having undergone selective vagotomy [238].

Taken together, the latest evidence indicates that the brain regions and the types of nerve cells that become sequentially in PD involved are anatomically interconnected, often over long distances, and that physical contacts between nerve cells as well as axonal transport are involved in PD pathogenesis [31, 221, 241]. Misfolded and slightly aggregated α-synuclein seeds can spread trans-synaptically along multisynaptic pathways and function, probably in a strain-dependent manner, as self-propagating pathogens for disease progression [70, 78, 99, 222, 223, 242–261]. Whether neuronal loss or impairment of cellular functions, the disease-related damage revolves around superordinate centres of the limbic, visceromotor, and somatomotor systems (Table 1, Figure 1). With the exception of olfaction and portions of the pain system (that is, the large neurons in layer 1 of the spinal cord), the sensorium is affected to a lesser extent [10, 262–267].

Studies of pathological mechanisms that underlie the conformational change and aggregation of α-synuclein, as well as those involving the formation of pathogens that can be transported axonally and transmitted trans-synaptically to interconnected nerve cells, where they induce α-synuclein misfolding and aggregation in a previously uninvolved cell, cannot be performed on formalin-fixed autopsy tissue. Nonetheless, experimental cellular models can only approximate the cellular milieu found in the human nervous system, where LP (as opposed to 'Lewy-like' lesions in nonhuman models) develops only in a few susceptible types of long-lived projection neurons. Resected tissue culled from surgical interventions on the human gastrointestinal tract, for example, contains functional groups of postmitotic myenteric plexus nerve cells [268–270]. Although admittedly challenging to design, a human-based ENS model might yield even more accurate insights into the mechanisms of α-synuclein aggregation and, possibly, also transmission in PD [78, 116, 225, 226].

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Author contributions

Both authors contributed to the conception of the text. The first author drafted and revised the manuscript. The senior author designed and drafted the figure.

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