

Interaction between A β Peptide and α Synuclein: Molecular Mechanisms in Overlapping Pathology of Alzheimer's and Parkinson's in Dementia with Lewy Body Disease

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Abstract Amyloidogenic proteins (A β peptide) in Alzheimer's disease (AD) and alpha-synuclein (α -Syn) in Parkinson's disease (PD) are typically soluble monomeric precursors, which undergo remarkable conformational changes and culminate in the form of aggregates in diseased condition. Overlap of clinical and neuropathological features of both AD and PD are observed in dementia with Lewy body (DLB) disease, the second most common form of dementia after AD. The identification of a 35-amino acid fragment of α -Syn in the amyloid plaques in DLB brain have raised the possibility that A β and α -Syn interact with each other. In this report, the molecular interaction of α -Syn with A β 40 and/or A β 42 are investigated using multidimensional NMR spectroscopy. NMR data in the membrane mimic environment indicate specific sites of interaction between membrane-bound α -Syn with A β peptide and vice versa. These A β - α -Syn interactions are demonstrated by reduced amide peak intensity or change in chemical shift of amide proton of the interacting proteins. Based on NMR results, the plausible molecular mechanism of overlapping pathocascade of AD and PD in DLB due to interactions between α -Syn and A β

is described. To the best of our knowledge, it is the first report using multidimensional NMR spectroscopy that elucidates molecular interactions between A β and α -Syn which may lead to onset of DLB.

Keywords Parkinson's disease · Alzheimer's disease · Dementia with Lewy bodies · A β peptide · α -Syn · NMR

Introduction

Alzheimer's disease (AD) and Parkinson's disease (PD) are the two most common neurodegenerative disorders characterized by insoluble protein deposits: β -amyloid lesions in AD and alpha-synuclein (α -Syn) containing Lewy bodies (LBs) in PD. α -Syn is a ubiquitous 140-amino acid protein of 18–20 kDa that is encoded by a single gene consisting of seven exons borne by chromosome 4 [1–3]. This protein is mainly intracellular and abundant in neurons, especially enriched in pre-synaptic terminals and is identified as a major component of the intracytoplasmic fibrillar LBs deposits, hallmark lesions of PD [4, 5].

Alpha-synuclein protein is composed of three distinct domains: a highly conserved amino terminal lipid-binding α -helix (residues 1–61) domain; a variable internal hydrophobic NAC (nonamyloid component) domain (residues 61–95) and a variable carboxyl terminal acidic tail (residues 95–140) composed primarily of negatively charged glutamate and aspartate residues [3]. The 35-amino acid containing NAC domain is the building block of α -Syn aggregates. The C-terminal domain contains several consensus phosphorylation sites for protein tyrosine kinases and serine/threonine

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kinases [3] and appears critical for the chaperone-like activity of α -Syn [6]. The C-terminal part of α -Syn does not associate with either vesicles or micelles and remains free and unfolded [7]. Over half of the α -Syn molecule (residues 7–87) is composed of six imperfect repeat sequence motif KTKEGV (Fig. 1) [8]. Although the function of these repeats is still unknown, the structure of α -Syn allows the protein to exhibit different conformations depending on its interacting environment, such as natively unfolded conformation in aqueous solution, α -helical conformation upon binding to lipid vesicles, or β -pleated sheets in its aggregated form, suggesting highly dynamic structural changes depending upon the local cellular milieu. According to recent NMR studies, it is reported that the lipid-binding domain of α -Syn is composed of two α -helices that are interrupted by a short break [9].

It has been suggested that there are two structurally distinct populations of α -Syn in cells, an α -helix-rich, membrane-bound form and a disordered, free cytosolic form [10]. The membrane-bound form of α -Syn represents only a fraction of total brain α -Syn and acts as a nucleating species because of its higher propensity for aggregation and its seeding abilities [10].

Alzheimer's disease is associated with the progressive accumulation of extracellular amyloid plaques and neurofibrillary tangles [11]. The major component of amyloid plaques is the 39–43 amino acids containing A peptide (Fig. 2). A β peptide has a high propensity for aggregation to form β -sheets [12]. It is proteolytically

cleaved from much larger glycoproteins known as amyloid precursor proteins, which comprise 695–770 amino acids with a single hydrophobic transmembrane region and resemble glycoprotein receptors on cell surfaces [13].

Amphipathic A β contains six negatively charged and six positively charged amino acid residues [14]. The structures of both A β 40 and A β 42 have been reported in membrane-associated and aqueous environments [15–17]. A β 40 in a membrane mimic environment exists as a random coil (residues 1–14), α -helix-I (residues 15–23), kink 25–29 and α -helix-II (residues 30–35) [15]. The function of soluble monomeric A β peptide is not known, however, oligomeric β -stranded A β conformations are neurotoxic [14]. Recent reports indicated catalytic breakdown of glycerophosphocholine (GPC) to α -glycerophosphate and choline by A β peptide (in vitro). Breakdown of GPC depends on molecular state (i.e., monomeric or oligomeric) of A β peptide [18, 19].

A β 40 is the major species found in plasma and cerebrospinal fluid, while A β 42 represents only about 10% of total secreted A β [20]. It is widely believed that A β 42 is responsible for the nucleation of amyloid formation, because it has a higher tendency to aggregate, and A β 42 aggregate can seed A β 40 in vitro [21]. This idea was supported by the findings that diffuse plaques, which are believed to be the precursor for senile plaques, are composed of A β 42 in Down's syndrome, whereas senile plaques themselves contain both A β 40 and A β 42 [22].

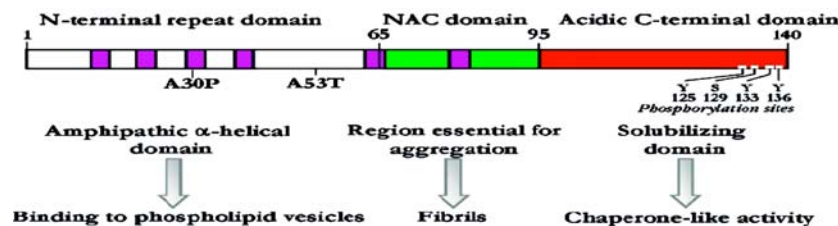


Fig. 1 Human α -Syn sequence and domains and the function of each domain are indicated. The imperfect KTKEGV repeats are shown in violet. Missense mutations A30P and A53T are also shown. Several phosphorylation sites have been detected in the

carboxyl-terminal region on Tyr-125, -133 and -136, and on Ser-129. NOTE: For interpretation of the references to color in this figure legend, the reader is referred to the online version of this article

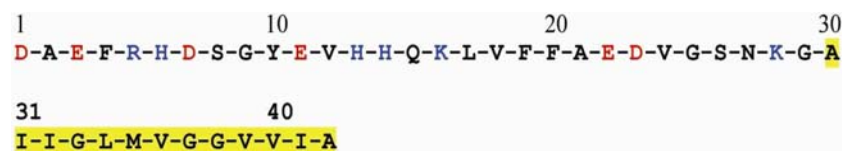


Fig. 2 Amino acid sequence of A β 42 peptide. Amphipathic A β contains six negatively charged (red) and six positively charged (blue) amino acid residues. The hydrophobic residues are

marked by yellow background color. NOTE: For interpretation of the references to color in this figure legend, the reader is referred to the online version of this article

Neurodegenerative diseases, such as AD and PD have their unique neuropathological features. Overlapping symptoms (progressive cognitive decline, fluctuating cognition, Parkinson's-like symptoms and visual hallucinations) of AD and PD are identified in dementia with Lewy bodies (DLB) disease, the second most common form of dementia after AD [23]. Neuropathological analysis of DLB cases have indicated the presence of both amyloid plaque, tangles, cortical LBs [24]. NAC, a 35 amino acid fragment of α -Syn is clinically observed in senile plaques in DLB cases [25, 26]. The identification of NAC component in the amyloid plaques positively indicates the interactions of α -Syn with A β peptide.

Studies using the α -Syn/ β -amyloid double transgenic mice demonstrated severe learning, memory and motor deficits [27]. Based on double transgenic mice animal studies, Masliah et al. had indicated that α -Syn interacts differently with A β 42 compared to A β 40 [28]. It is important to understand the molecular mechanism of A β and α -Syn interactions which might influence the pathophysiology of DLB.

In this report, the specific interactions of membrane-bound α -Syn with A β 40 and/or A β 42 are investigated using multidimensional NMR spectroscopy. The question is raised as to how NAC (a fragment of α -Syn protein) coexist in the extracellular space with the amyloid plaque in DLB patients. We hypothesize that in the diseased condition (indicated by altered membrane packing) membrane-bound α -Syn interacts with membrane-associated (extracellular) A β 40 and/or A β 42 peptides and these two peptides (A β 40 and/or A β 42) may likely cleave [19] the NAC fragment from α -Syn. Due to these interactions with α -Syn, both A β 40 and/or A β 42 eventually get precipitated [25, 26] along with the NAC component which is neuropathologically observed in DLB patients. To test this hypothesis, we have studied the interactions of A β 40 and A β 42 peptide with α -Syn in a membrane mimic sodium dodecyl sulfate (SDS) environment using multidimensional NMR spectroscopy. Based on NMR results the plausible effect of interactions between A β and α -Syn is discussed and molecular scheme leading to overlapping pathology of AD and PD in DLB is presented.

Materials and methods

Materials

Uniformly ^{15}N -labeled A β 40, A β 42, α -Syn and unlabeled A β 40, A β 42 and α -Syn were purchased from Recombinant Peptide Technologies (Atlanta, GA,

USA). Deuterated SDS ($\text{SDS}_{\text{D}25}$) was purchased from Cambridge Isotope Laboratories (Andover, MA, USA). Other chemicals and materials were purchased from Sigma (St Louis, MO, USA) and used without further purification.

Sample preparation

Preparation of A β solution in SDS

For NMR studies, the uniform ^{15}N -labeled A β 40 and A β 42 were prepared with a peptide concentration of 1 mM. Each sample contained the A β peptide dissolved in 100 mM SDS in 90% MilliQ H_2O and 10% D_2O at pH 7.2. The pH measurements were uncorrected for isotope effects. The pH of these samples was adjusted using small aliquots of 0.01 M DCl or NaOD. The two samples (A β 40 and A β 42) were clean and stable. The molar ratio of peptide/ $\text{SDS}_{\text{D}25}$ was 1:200.

Preparation of α -Syn solution in SDS

For NMR studies, uniform ^{15}N -labeled α -Syn was prepared with a peptide concentration of 1 mM. α -Syn was dissolved in 100 mM $\text{SDS}_{\text{D}25}$ in 90% MilliQ H_2O and 10% D_2O at pH 7.2, the pH measurement being uncorrected for isotope effects. The pH of the sample was adjusted using small aliquots of 0.01 M DCl or NaOD. The solution was clean and stable. The molar ratio of peptide/ $\text{SDS}_{\text{D}25}$ was 1:200.

NMR experiments and data analysis

All NMR experiments were performed at 303 K. All 2D and 3D NMR experiments were carried out on a 500-MHz Bruker DRX spectrometer using a 5-mm TXI probe (Bruker, Karlsruhe, Germany). All two-dimensional spectra were collected using either 2,048 or 4,096 complex data points in F2, acquiring 256–400 increments in F1 for the total correlation spectroscopy (TOCSY) and nuclear Overhauser enhancement spectroscopy (NOESY). Spectra were acquired in the phase-sensitive mode using time proportional phase increment for quadrature detection in the t_1 dimension [29]. The solvent signal (90% H_2O) suppression was accomplished by the use of pulsed field gradients [30]. Homo-NOESY, TOCSY, heteronuclear single quantum spectroscopy (HSQC) [31] and ^{15}N filtered 3D NOESY experiments were performed for sequence-specific assignment and structural studies. NMR data was processed using nmrPipe [32] and analyzed using PIPP [33] and SPARKY [34] programs on a Silicon Graphics octane 2.

Using these NMR techniques we have studied: (a) the structural influence of α -Syn on A β 40 and/or A β 42 by monitoring the change in chemical shift and signal intensity alteration of amide protons of ^{15}N -labeled A β 40 and/or A β 42 in the presence of α -Syn using heteronuclear NMR studies; and (b) the same NMR approach was applied to monitor the structural change of ^{15}N -labeled α -Syn due to A β 40 and/or A β 42. All NMR experiments were done in membrane mimic environment.

Results

Uniformly ^{15}N -labeled A β 40, A β 42 and α -Syn were soluble at millimolar concentrations in aqueous 100 mM SDS and yielded 1D and 2D and 3D spectra of good quality. There was no evidence of gelling or precipitation of the sample over several weeks and the HSQC spectra of A β 40, A β 42 and α -Syn did not change over this period. This suggests that A β 40, A β 42 and α -Syn did not aggregate in the water/SDS micelle solution.

Figure 3 represents amide region of ^{15}N -labeled A β 40 peptide with unlabeled α -Syn (red) and without unlabeled α -Syn (blue). The assignment of well-resolved amide peaks was made using a combination of different 2D and 3D dimensional experiments. After addition of α -Syn the HSQC spectra of ^{15}N -labeled A β 40 was monitored over time. The HSQC spectra of ^{15}N -labeled A β 40 with α -Syn after 2 h show that residues E3 to G37 of ^{15}N -labeled A β 40 are perturbed to different extents as evident by change in chemical shift of amide proton. However, appreciable changes in chemical shifts are observed for residues E11 and G25 (marked with *) (Fig. 3). It is important to note that A β 40 remains in solution for 15 h after the addition of α -Syn before it oligomerizes as evident from broadening of amide peaks (figure not shown).

Figure 4 shows the amide region of ^{15}N -labeled A β 42 with unlabeled α -Syn (red) and without unlabeled α -Syn (blue). After addition of unlabeled α -Syn, the amide protons of ^{15}N -labeled A β 42 was monitored over time using HSQC spectra. α -Syn interacted strongly with A β 42 as evidenced by the major change in chemical shift of amide protons seen after 2 h. Structural alteration of A β 42 is observed by the decrease in amide peak intensity. Interactions are not specific, whole structure of ^{15}N -labeled A β 42 seems to be affected while interacting with α -Syn. It is not possible to comment on any specific region of the peptide chain as in the case of A β 40. In this case global structural change is observed.

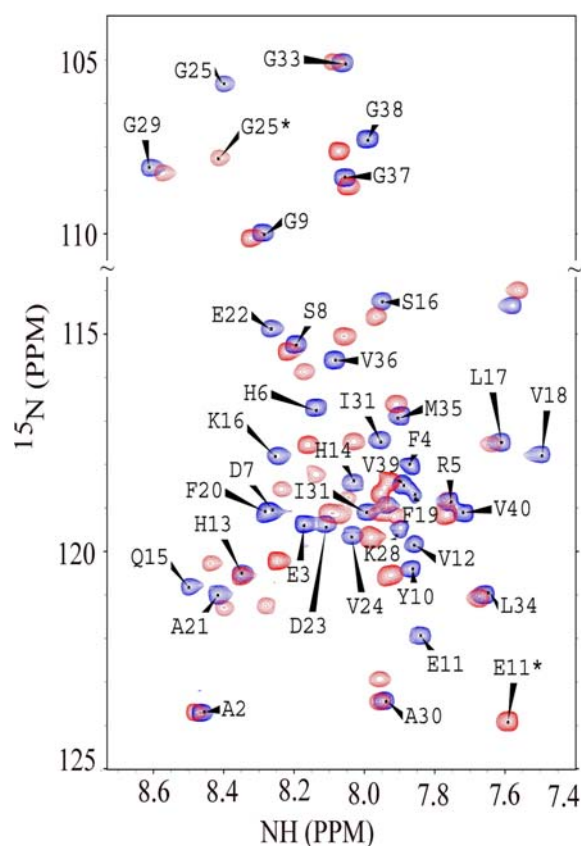


Fig. 3 HSQC spectra of A β 40, in which blue denotes the control (without α -Syn) and red denotes the A β 40 with α -Syn after 2 h. Residues marked with star (*) sign show appreciable chemical shift resulting from α -Syn. NOTE: For interpretation of the references to color in this figure legend, the reader is referred to the online version of this article

Figure 5 shows amide protons of ^{15}N -labeled α -Syn with unlabeled A β 42 (red) and without A β 42 (blue). The amide peaks of ^{15}N -labeled α -Syn were assigned by combinations of different 2D and 3D experiments that includes ^{15}N filtered HSQC–NOESY, ^{15}N filtered HSQC–TOCSY, home-NOESY. The amide peaks of ^{15}N -labeled α -Syn were monitored over time in the presence of A β 42. The HSQC spectra of α -Syn with A β 42 after 8 h show that almost all amide protons starting from K10 to A140 of A β 42 are perturbed to different extents as evident by change in chemical shift of amide proton. However, appreciable change in chemical shifts are observed for residues G63, G67, A69, G73, V74 and T81 (marked with *).

Discussion

Neurodegenerative diseases, such as AD and PD have their unique neuropathological features. “Pure” AD is characterized by presence of both diffused and neuritic

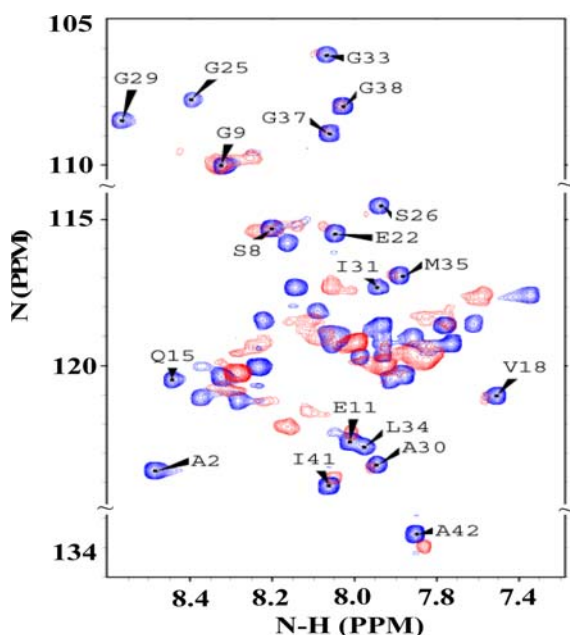


Fig. 4 HSQC spectra of Aβ42, in which *blue* denotes the control (without α-Syn) and *red* denotes the Aβ42 with α-Syn after 2 h. NOTE: For interpretation of the references to color in this figure legend, the reader is referred to the online version of this article

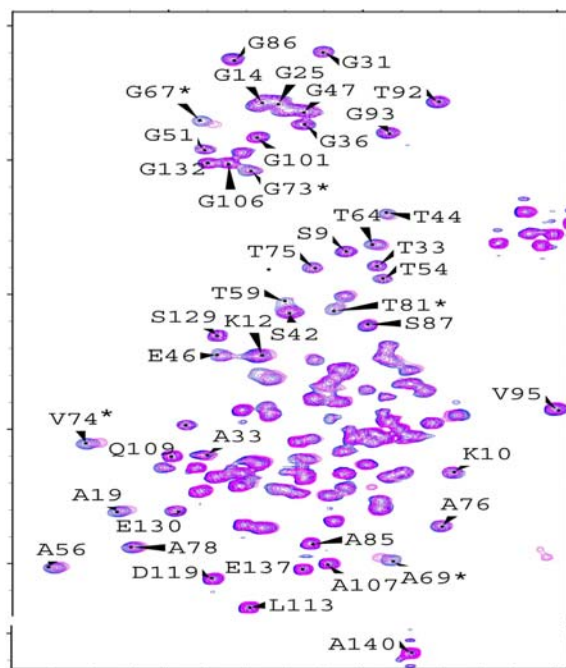


Fig. 5 HSQC spectra of α-Syn, in which *blue* denotes the control (without Aβ42) and *red* denotes the α-Syn with Aβ (1–42) after 8 h. Residues marked with *star* (*) show appreciable chemical shift resulting from α-Syn. NOTE: For interpretation of the references to color in this figure legend, the reader is referred to the online version of this article

plaques in the neocortex (Fig. 6A) and the substantia nigra (SN) typically shows only mild or no neuronal loss (Fig. 6B, C) and lacks abnormal α-Syn inclusions

or neuritis (Fig. 6B, D). On the other hand, “pure” PD without dementia is characterized by rarely any β-Amyloid plaques in the neocortex (Fig. 7A) and some scattered LBs and α-Syn (Fig. 7B). The SN in PD without dementia is specifically characterized by severe neuronal loss and deposition of LBs and α-Syn-positive neurites (Fig. 7C, D).

Dementia with LB patients (second major cases of dementia after AD) have been found with neuropathological features of both AD and PD [35–37]. DLB cases have frequent deposits of Aβ peptide (Fig. 8A) as well as abnormal α-Syn-positive neocortical neuronal inclusions (LBs) and neuritis (Fig. 8B). The SN in DLB shows a modest neuronal loss with scattered LBs (Fig. 8C) as well as a moderate number of α-Syn-positive neurons and neuritis (Fig. 8D). In DLB cases, there is the overlapping of neuropathological, neuropsychiatric and neurochemical features of AD and PD (Table 1) [24]. Doctors and clinical experts previously used other terms to describe DLB, i.e., diffuse Lewy body disease [38], Lewy body dementia [39], cortical Lewy body dementia [40] and senile dementia with Lewy body [41]. Most clinicians now follow DLB as the generic term for all these cases as recommended by an international consortium in 1996 [23].

As mentioned earlier, Aβ and α-Syn are involved separately in pathogenesis of AD and PD, respectively. Table 1 clearly indicates extensive overlapping of neuropathological, neuropsychiatric and neurochemical characteristics of AD and PD in DLB, therefore it is reasonable to say that both α-Syn and Aβ are involved in DLB. The identification of the NAC fragment in DLB patients is supportive of this hypothesis. Our NMR data show that both ¹⁵N-labeled Aβ40 and Aβ42 interacts with membrane-associated α-Syn. However, α-Syn induced structural alteration is more profound in Aβ42 compared to Aβ40. This observation is supported by Masliah et al. in their transgenic mice studies [28]. In Aβ40–α-Syn interaction studies (Fig. 3), few residues of Aβ40 show appreciable change in chemical shift and Aβ40 remain in solution for 15 h, while ¹⁵N-labeled Aβ42 interacts strongly with α-Syn and undergoes major structural change to oligomerize and eventually precipitates quickly within 4 h. This experimental result supports the previous notion that Aβ42 is more pathogenic than Aβ40 [42].

Previous studies using solid phase binding assay have speculated that residues (81–95) of α-Syn interacts with Aβ peptide [43]. Although, solid phase binding method cannot provide the specificity of the binding site, in that respect NMR is extremely helpful to indicate the specific binding site of interaction

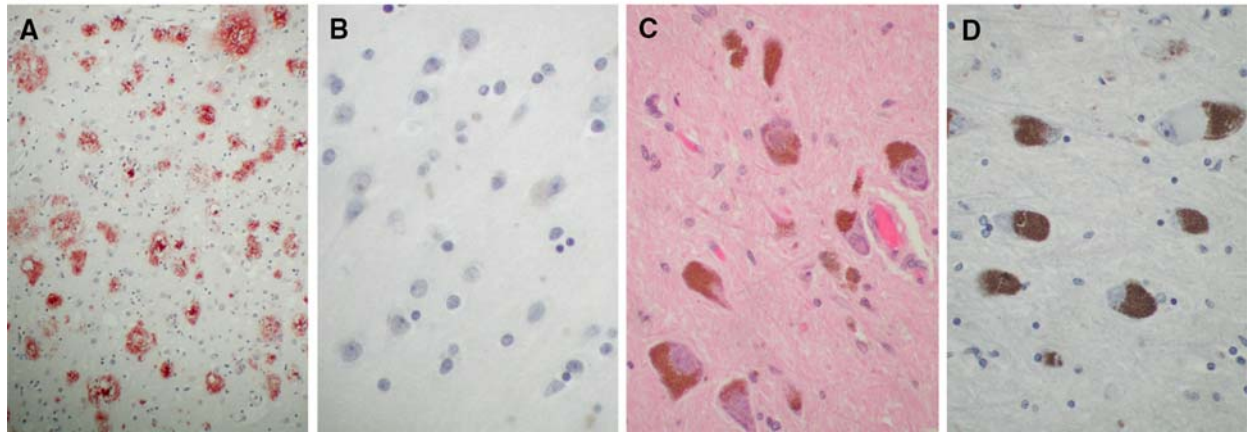
AD

Fig. 6 Micrographs of “pure” AD (A–D) brain demonstrating characteristic neuropathological changes. There are frequent deposits of β -amyloid protein in the neuropil (A) resulting in both diffused and neuritic plaques. There are no α -Syn-positive

neuronal inclusions or neurites in the neocortex (B). The SN typically shows only mild or no neuronal loss by H&E stains (C) and also lacks any abnormal α -Syn inclusions or neurites (D)

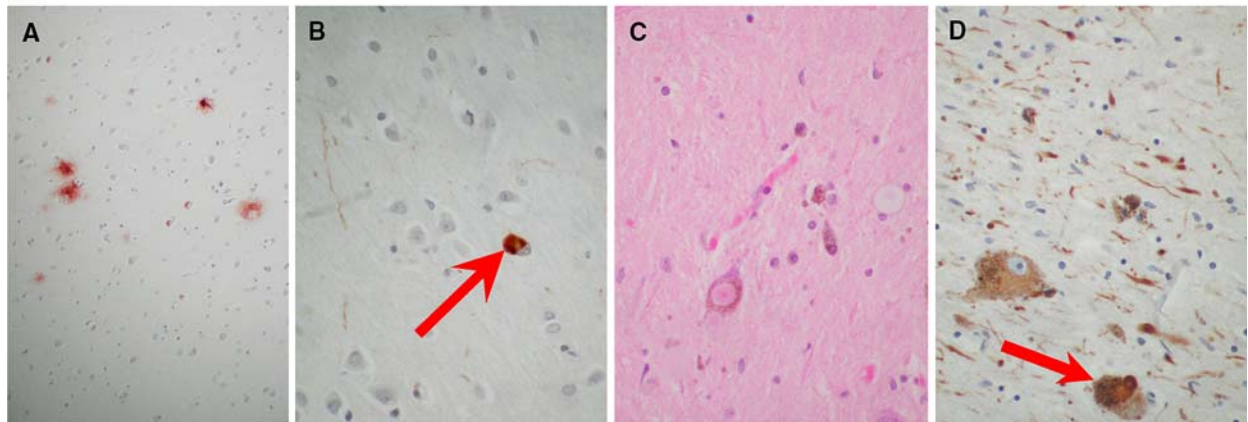
PD

Fig. 7 Micrographs of “pure” PD (A–D) brains demonstrating characteristic neuropathological changes. In “pure” PD cases without a history of dementia, there may be rare, widely scattered β -amyloid plaques in the neocortex (A). Using α -Syn immunohistochemistry, one can usually find neocortical LB and

α -Syn neurites (B) (marked by an *arrow*). In the SN, there is always severe neuronal loss and some remaining neurons may contain LB (C), while α -Syn immunostaining shows α -Syn positive neurons and numerous α -Syn-positive neurites (D) (marked by an *arrow*)

between the proteins. We have found that when labeled α -Syn interacts with A β 42 (Fig. 8), the following residues G67, G73 and V74 (marked with * in Fig. 8) show change in chemical shift and decrease in amide peak intensity. It may be noted that these three residues belong to NAC component of α -Syn. The oligomerization of α -Syn in the presence of A β 42 prevented further NMR studies to monitor any change in chemical shift of α -Syn.

The interaction between A β and α -Syn might be important in the pathogenesis of DLB because NAC constitutes up to 10% of the SDS-insoluble protein in the amyloid plaque. Also α -Syn is expressed particu-

larly in regions of the brain where AD lesions are abundant. The complex sequence of events leading to the formation of amyloid plaques with NAC component is uncertain. The proposed model (Fig. 9) indicated the sequence of events in DLB.

Proposed model for overlapping pathocascade of AD and PD in DLB

The complex sequence of events leading to the formation of amyloid plaques with NAC and intracellular LBs formation in DLB is under investigation. Concerning the role of specific molecular interactions

DLB

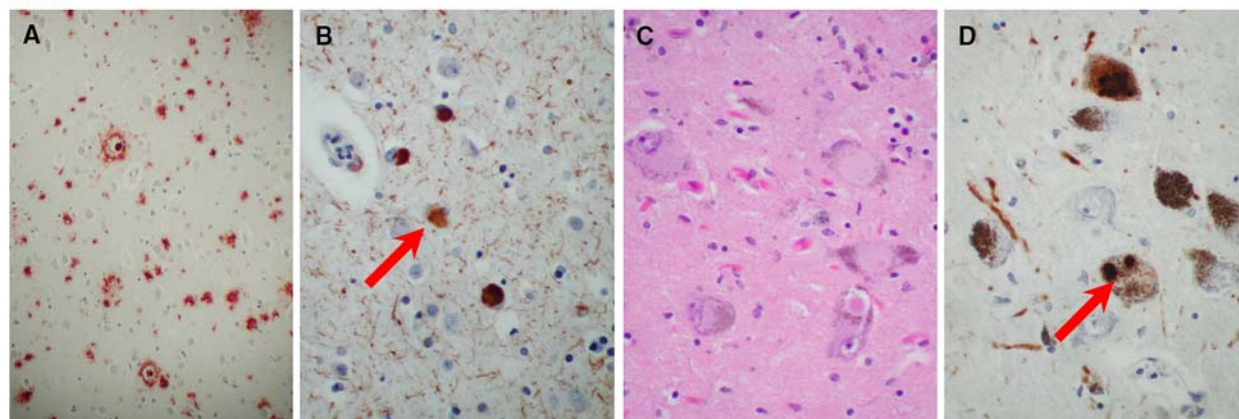


Fig. 8 Micrographs of DLB (A–D) brains demonstrating characteristic neuropathological changes. Most DLB cases have frequent deposits of β -amyloid plaque (A) and may have numerous abnormal α -Syn-positive neocortical neuronal inclu-

sions (Lewy bodies) and neurites (Lewy neurites) (B) (marked by an *arrow*). The SN may show modest neuronal loss with scattered LB (C) as well as moderate numbers of α -Syn positive neurons and neurites (D) (marked by an *arrow*)

Table 1 A comprehensive note on AD, DLB and PD

	AD	DLB	PD
Neuropsychiatric symptoms			
Visual hallucinations	+	+++	++
Delusions	++	+++	+
Depression	++	++	++
Apathy	++	++	+
	Hallucinations in late stages of disease	Persistent hallucinations early in course of disease	In association with anticholinergic dopaminergic drugs
Tremor	–	++	+++
Rigor	+	+++	+++
Bradykinesia	+	+++	+++
	Rare, usually mild in late stages	Similar severity as in PD, pronounced rigidity and bradykinesia	First manifestation of disease, initially often asymmetric
Fluctuation of cognition	+	+++	–
Neuropsychology	Early impairment of declarative memory and retention	Early disturbances in attention, visuo-perceptive functions	Impaired executive functions
Neuroimaging			
Global brain atrophy	++	++	–
Medial temporal lobe atrophy	+++	+	–
Occipital hypoperfusion		+++	+
Impaired dopaminergic activity	–	+++	+++
Neuropathology and chemistry			
Senile plaque density	+++	++	–
Tangle density	+++	++	–
Subcortical LB	–	++	+++
Cortical LB	–	+++	+
Cholinergic deficit	++	+++	+
Dopaminergic deficit	–	++	+++
Genetics			
Overrepresentation Apo ϵ 4	++	++	–

+++ indicates typical manifestation of disease, ++ indicates usually present, + indicates present, – indicates absent

between the A β peptide and α -Syn, the following sequence of events are proposed to describe the cross influences of A β and α -Syn in DLB. This putative

cascade is illustrated in Fig. 9. Pre-synaptic terminals are highly enriched in α -Syn [44]. α -Syn is found to be implicated in synaptic vesicle formation, axonal

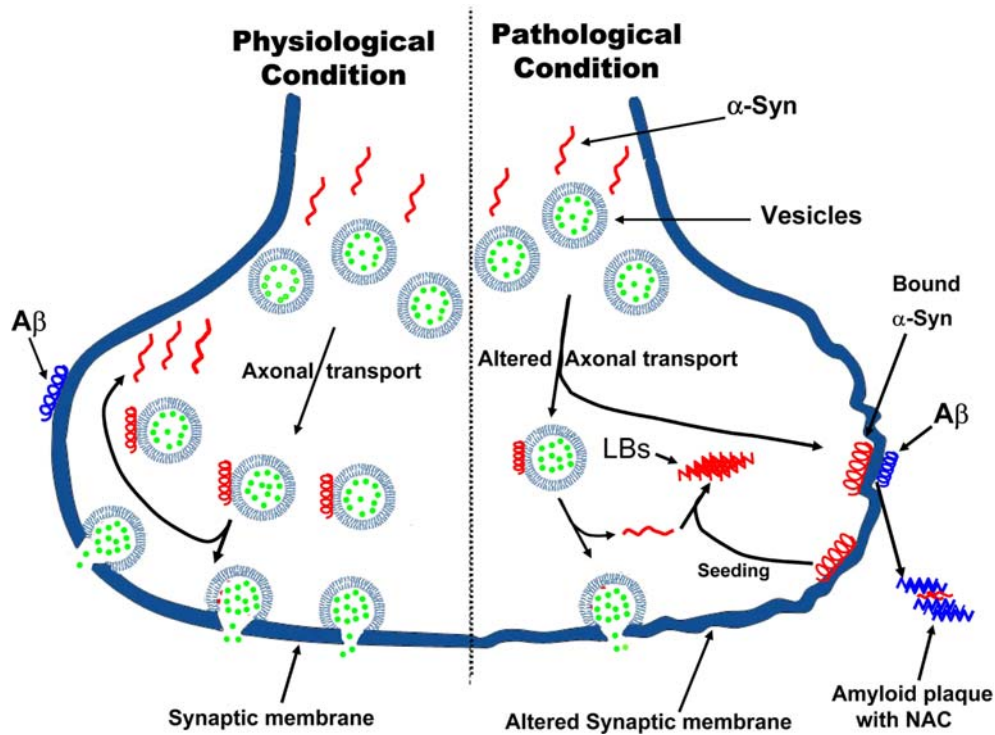


Fig. 9 Schematic diagram of potential interaction between α -Syn and A β peptide in DLB (**A**). In normal neuron random coiled, α -Syn is transported to the synaptic terminal, assembled on the surface of the synaptic vesicle as stable α -helix. Cytoplasmic α -Syn does not normally interact with A β that is within the pre-synaptic membrane. (**B**) Abnormal accumulation of free cytoplasmic α -Syn on the pre-synaptic membrane occurs due to altered axonal transport, defective proteolysis decreased

number of synaptic vesicles or other factors. Cytosolic random coiled α -Syn gets transformed into stable membrane-bound α -helical form. This membrane-bound α -Syn comes in contact with membrane-associated A β peptide. A β peptide interacts catalytically with α -Syn and causes conformational changes generating NAC fragments. Membrane-bound α -Syn also induces seeding of cytosolic α -Syn to form LB

transport and dopamine synthesis and metabolism [45]. It exhibits different conformations depending on its interacting environment [45]. In normal condition, unstructured α -Syn is associated with dopamine-containing vesicles and involved in vesicle transport process. It adopts α -helical conformation while associated with vesicles until the vesicles are fused with the synaptic membrane. It is released to the cytosol as unstructured form and participates in ubiquitin–proteasome degradation pathway [46]. In normal conditions, the synaptic membrane integrity is maintained hence it is reasonable to assume that after the vesicles are fused to the membrane, α -Syn is completely released to the cytosol. Hence, in normal condition, bound α -Syn is absent in the synaptic membrane. It has been pointed out that membrane-bound α -Syn has higher aggregation propensity [10] hence the possibility of aggregation of cytosolic α -Syn is diminished. Thus in normal condition, there are no intracellular LBs.

Due to neuronal damage, there is an increased synthesis of proteins to maintain and support the lost synaptic function. In diseased condition in an undis-

closed mechanism the α -Syn associated with vesicle transport are not completely released back to cytosol and a fraction of the α -Syn is associated with the synaptic membrane depending on the extent of synaptic membrane defect. Synaptic bound α -Syn induces cytosolic α -Syn to aggregate as intracellular LBs. These synaptic membrane-bound α -Syn can also interact with membrane-associated A β 40 and A β 42 peptides [47]. When membrane-bound α -Syn comes in contact with membrane-associated A β peptide they interact with each other at multiple locations. A recent report suggests that α -Syn aggregation may occur on membrane surfaces and the interaction of membrane-bound α -Syn with A β peptide are also indicated [48]. We have shown that α -Syn interacts with A β 40 and A β 42 differently. Interaction of synaptic membrane-bound α -Syn with A β 42 is more profound and it oligomerizes A β 42. These toxic oligomeric A β 42 affect the synaptic membrane integrity. While interacting with α -Syn, oligomeric A β 42 [14] cleaved the NAC fragment from α -Syn, which is seen in the extracellular space along with amyloid plaque.

The catalytic type activity of A β 40 is much lower than A β 42, hence A β 40 is less efficient [19] to cleave NAC fragment from bound α -Syn. It may be noted that NAC content in the amyloid plaque is low as availability of membrane-bound α -Syn is lower to interact with A β peptide. Hence it needs careful neuropathological analysis to identify the NAC fragment in the amyloid plaque in DLB patients. Figure 9 shows the molecular pathway for the interactions of A β peptide with α -Syn, however, in a clinical situation, it may take years or decades for intracellular α -Syn to be available in extracellular space due to altered membrane and interact with A β peptide.

To conclude, there is ample evidence to propose that both A β peptide and α -syn interact with each other in a specific manner. The presence of multiple sites of interaction between α -Syn and A β might be important in determining the overlapping pathocascades of AD and PD seen in DLB. To prevent the interaction of membrane-bound α -Syn with A β peptide would be an important therapeutic research goal. Different neuroprotective gangliosides may play an important role in preventing these interactions. We are working to test the influence of different ganglioside [17] in preventing the overlapping pathology of AD and PD on double transgenic mice.

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