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The Aggregation of Tau Protein in Alzheimer's Disease

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ABSTRACT: Currently, neurodegenerative diseases affect approximately 50 million people and are a significant cause of death and disability worldwide. One of the most well-known neurodegenerative diseases is Alzheimer's Disease. Alzheimer's Disease (AD) is characterized by the presence of neurofibrillary tangles made up of filamentous Tau aggregates and ß- amyloid depositions and is a relatively common cause of dementia in older adults. AD is known as a tauopathy, a family of neurodegenerative diseases characterized by tau neurofibrillary tangles. Neurofibrillary tangles are thought to be toxic aggregates of Tau protein that occur after tau disengages from microtubules. In neurons, microtubules are responsible for transporting substances to different parts of the cell. They exhibit dynamic instability, meaning they constantly grow and shrink. Under normal physiological conditions, Tau protein, a neuronal microtubule-associated protein, promotes microtubule self-assembly and stabilizes microtubules. Tau's intracellular interactions and functioning is regulated by phosphorylation, a post-translational modification. However, Tau undergoes hyperphosphorylation and aggregation under pathological conditions to form neurofibrillary tangles, leading to neurotoxicity. This synaptic dysfunction and loss of microtubule stability eventually lead to the neurodegeneration characteristic of tauopathies. To find new therapeutic targets, the loss of normal tau function and gain of toxic tau function must be investigated. This review will discuss the current models of Tau aggregation, the Tau pathology that causes Alzheimer's Disease, and current therapeutic strategies to treat tauopathies.

KEYWORDS: Biomedical and health sciences, genetics and molecular biology of disease, Tau, tauopathies, aggregation.

Introduction

In addition to toxic Tau pathology, AD can also be caused by beta-amyloid aggregates that form deposits in the brain leading to neurodegeneration. Because beta-amyloid is still considered causative, it has been the target of several therapeutics. However, at least four anti-amyloid antibodies have failed in phase III trials in different Alzheimer's disease settings. Three BACE inhibitors and two γ-secretase inhibitors, which act on amyloid processing, have also failed. In some cases, these treatments were even associated with worsening cognition.[1] After failed therapy for amyloid, Tau became an alternative target for therapeutics to treat AD as it is a proximal mediator of neurodegeneration and causative of cognitive decline. Nonetheless, the key to understanding Tau's pivotal role in AD begins with its regular function in neurons to stabilize microtubules.

Microtubules are part of the cytoskeleton, a structural network within the cell's cytoplasm. In neurons, microtubules transport materials from the cell body to the axon terminals at the synapse, and they also define axons and dendrites.[2] They are composed of alpha- and beta-tubulin subunits assembled into approximately thirteen linear strands called protofilaments; these protofilaments bind together to form the hollow, tube-like structure of the microtubule.[2] Microtubules are constantly growing and shrinking, which happens when the alpha and beta tubulin subunits associate and dissociate from the plus end (the end that grows more rapidly) of the protofilament. This phenomenon is known as "dynamic instability."[3] Dynamic instability is a useful biological mechanism because it allows the microtubules to reorganize the cytoskeleton when necessary quickly.[4] In neurons, the dynamic instability of microtubules upon which axonal transport occurs is suppressed by the microtubule-associated protein Tau. Tau protein promotes microtubule self-assembly and stabilizes microtubules that have already formed.[5] Tau's other functions include neuronal cell signaling, nuclear function, and maintenance of the neuronal cytoskeleton.[5] However, an essential facet of Tau that places it at the center of research surrounding tauopathies and neurodegenerative disorders is the insoluble lesions it forms in disease.

Tau can undergo several post-translational modifications to regulate its functioning through the brain. A critical post-translational modification to analyze is phosphorylation, the addition of a phosphate group to serine, threonine, and tyrosine residues. Phosphorylation regulates Tau functioning under normal conditions; however, under pathological conditions, hyperphosphorylation can occur. As this happens, Tau loses its normal physiological function and aggregates, resulting in toxicity, for example, causing synaptic dysfunction. According to Goedert *et al.*,[6] hyperphosphorylation results in the reduced ability of tau to interact with microtubules, which is most likely necessary for its ordered assembly into oligomers, proto-fibrils, fibrils, tangles, paired helical filaments, and neurofibrillary tangles. Most importantly, neurofibrillary tangles, made up of fibrils of hyperphosphorylated tau, are a biomarker of Alzheimer's disease and other related tauopathies. This review paper will summarize the literature that aims to address the mechanisms of Tau aggregation and seeding, and current therapeutic strategies that target tauopathies will be described.

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Discussion

Structure of Tau:

Tau assembles and stabilizes microtubules by interacting with the tubulin subunits in the microtubules at various microtubule binding regions. The repeat region is a region of Tau which is tightly and specifically bound in the core of the paired helical filament and is believed to be the microtubule binding domain. The microtubules binding regions differ across the six Tau isoforms in the human brain by having varying three or four repeats in the C-terminal region.[7] Furthermore, the N-terminal inserts may help regulate Tau's dynamic behavior and function during axonal transport.[8]

When investigating Tau's primary structure, it is crucial to note that Tau's primary structure can, in fact, cause disease. For example, Tau isoforms containing either three (3R) or four (4R) microtubule binding regions usually are in a one-to-one ratio. But, there are splicing defects in familial tauopathies such as frontotemporal dementia or corticobasal degeneration, which skew the ratio of 3R to 4R, either increasing the amount of 3R or 4R.[9] Furthermore, missense and silent tau primary structure mutations cause frontotemporal dementia with parkinsonism-chromosome 17 type by affecting multiple alternative RNA splicing regulatory elements.[10]

In addition to examining Tau's primary structure, the secondary and tertiary structures are equally important in understanding Tau's significance in neurodegenerative disorders. Tau is an intrinsically disordered protein, meaning that it lacks a well-defined three-dimensional structure. Yet, a secondary structure exists; it retains a flexible conformation important to its role in cellular processes.[11] Furthermore, intrinsically disordered proteins undergo order-to-disorder or disorder-to-order transitions as part of their normal biology. Their structure and function may be modulated by protein chaperones, post-translational modifications, and degradation processes. Tau's secondary structure is largely transient but consists of α-helices, β-pleated sheets, and a polyproline II helix.[8]

A "paper-clip" structure for some molecules of Tau monomers may have been indicated through nuclear magnetic resonance and small-angle X-ray scattering. A "paper-clip" structure may suggest that the N and C termini work together closely.[12] However, when Tau is bound to microtubules, the two terminals are disjointed, with the N-terminal facing away from the microtubules.[13] The presence of a "paper-clip" structure may suggest the presence of intramolecular interactions between at least two different regions of the Tau protein. If one of those regions is involved in Tau self-interaction, the opening of the "paper-clip" structure could be what facilitates self-aggregation in Tau pathology.[13]

Furthermore, the elastic and bendable structure of the Tau protein enables interaction with multiple partners, implying its involvement in many signaling pathways.[14] However, being intrinsically disordered allows tau to interact with other Tau molecules to form oligomers and filaments, which are the root cause of the gain of toxic function.[14] These neurofibrillary tangles cause degeneration of neurons and glial cells, displaying as a group of neurodegenerative disorders termed 'tauopathies.'

Phosphorylation of Tau:

An important feature of Tau is the presence of various types of post-translational modifications that it can undergo. These modifications include phosphorylation, acetylation, deamidation, methylation, O-Glycylation, and ubiquitination.[15] Of these modifications, a critical one to investigate is phosphorylation. The phosphorylation of proteins involves adding a phosphate group to three types of amino acids: serine, threonine, and tyrosine. Phosphorylation regulates tau's functioning by neutralizing its positive charge, reducing its affinity for microtubules, thereby detaching Tau from microtubules.[16] In an intact cell, Tau is constantly phosphorylated and dephosphorylated to regulate microtubule assembly.[16] There are eighty-five potential phosphorylation sites (45 serine, 35 threonine, and five tyrosine residues) scattered on the longest Tau isoform; specifically, they are located in regions around the repeat microtubule-binding regions on the C-terminal region.[17] According to Bramblett *et al*,[18] Tau's ability to stabilize microtubules inversely correlates with its phosphorylation, meaning that the more Tau gets phosphorylated, the less it can stabilize microtubules.

Furthermore, the amount of phosphorylation in Tau is directly tied to the amount of active protein kinase. This enzyme catalyzes the chemical reaction between ATP and Tau protein and phosphatase. This enzyme removes a phosphate group from a protein.[19] In addition, O-linked glucosamine modifications occur on serine and threonine residues and block phosphorylation. When glucosamine is removed, tau can be phosphorylated, making the enzymes that remove O-linked glucosamines a regulator of tau phosphorylation and a target in clinical trials.[20] Lastly, an enzyme called Glycogen synthase kinase 3 (GSK3β) is the most effective Tau kinase in the human brain, and it is directly linked to phosphorylation levels in Tau.[21] In other words, as GSK3β increases, so does the amount of Tau phosphorylation. This can have potentially detrimental effects on the brain, such as the onset of tauopathies.

In tauopathies, Tau is hyperphosphorylated at specific sites, forming aggregates and neurofibrillary tangles and making it a potential target for therapy. Changes in Tau conformation could result in (1) increased phosphorylation because of altered binding to kinases and (2) decreased binding to microtubules. Both of these can cause tau-mediated neurodegeneration.[22]

According to Hanger *et al.*,[21] Tau hyperphosphorylation occurs when there is an increased activity of Tau kinases (specifically GSK3β) and a decreased activity of Tau phosphatases. Another study has shown that hyperphosphorylation can occur when Tau is exposed to proteins such as β-amyloid, Fyn kinase, Pin1, heat shock cognate Hsc70, and heat shock protein Hsp90, immunophilins FKBP51 and FKBP52, α-synuclein or actin interacting protein PACSIN1.[22] Moreover, hyperphosphorylation causes many complications regarding tau's functional capabilities. When Tau gets hyperphosphorylated, the affinity of Tau to microtubules is lessened, causing microtubule instability and disassembly and then promoting Tau self-aggregation, which leads to neurofibrillary tangles

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made up of paired helical filaments.[6] Therefore, hyperphosphorylation is the critical event at the onset of Tau pathology.

Moreover, according to Goedert *et al.*,[23] stress-activated protein kinases also contribute to tau phosphorylation. They may also explain several observations demonstrated in a study on rat brains. Cold-water stress induces a relatively spontaneous (30–90 min) two to three times increase in Tau phosphorylation.[24]

Several animal models depict the influence of hyperphosphorylation in tauopathies. For instance, Ishihara *et al.*,[25] designed a transgenic mouse model in which three wild-type Tau transgenic mouse lines expressing different levels of the shortest tau isoform were used. Ishihara and colleagues found clusters of phosphorylated tau at several phospho-epitopes where there was an increased level of the Tau kinase, GSK-3β.[25] Furthermore, the authors found a substantial correlation between the specific phosphorylation changes and the aggregation levels of tau. In the *hTau* strain mice specifically, there were increased levels of Tau kinases such as p38, p35, and p25, which later caused an increase in phosphorylated tau. Changes in Tau kinases in the *hTau* mice were directly linked with how much Tau was present as toxic, insoluble aggregates.[25] In summary, this illustrates that Tau is subject to varying levels of phosphorylation with respect to the differential activities of kinases.

Tau Seeding and Aggregation:

Evidence suggests that hyperphosphorylated tau can spread through the brain in the form of "seeds" that contaminate neurons in a prion-like fashion, meaning they can transmit their misfolded shape onto the normal variant of the same protein. This causes Tau to lose its normal function and gain a toxic function model (Figure 1). In specific animal models, Tau aggregates are shown to actively spread from neuron to neuron, illustrating Tau seeding and how that causes Tau aggregates to propagate.

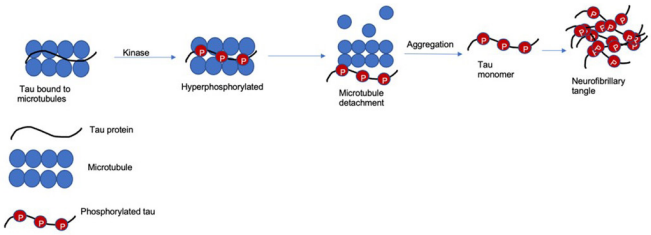

Figure 1: Tau aggregation and loss of microtubule affinity.

Braak *et al.*,[26] and colleagues used transgenic mouse models with localized Tau expression to express mutant Tau in the entorhinal cortex. After the Tau protein was injected into the mice, it demonstrated similar behaviors as it does in humans, such as hyperphosphorylation, abnormal Tau folding, and accumulation of aggregates. They found that Tau progressively spreads across the brain to neuroanatomically connected regions, demonstrating the possibility that this spreading could be due to Tau seeding. However, this study did not provide conclusive evidence that Tau seeding was causing aggregation to occur.[26]

In 2009, Clavaguera *et al.*,[27] conducted a vital experiment with respect to Tau seeding and spreading, which eventually paved the way for studies that utilized patient-derived Tau by injecting it into mouse models. They injected Tau filament containing Tau fibrils from a Transgenic mouse model that expressed the 0N4R human Tau isoform with the FTD-linked P301S mutation into the hippocampus and overlying cerebral cortex of Transgenic mice overexpressing a single WT human Tau isoform (2N4R). Through their results, this study contributed the first evidence that injected Tau fibrils induce the onset of pathological Tau aggregates, which progressively propagate to parts of the brain anatomically associated with the injection sites. Tau pathology was observed in different cell types, with aggregates present in the form of neurofibrillary tangles and neuropil threads. Moreover, further analysis suggested that the induced aggregates were composed of insoluble, phosphorylated tau.[27]

Similar to Clavaguera and colleagues, Ahmed *et al.*,[28] injected brain extracts of five-month-old transgenic mice for seeding into the brain of two-month-old mice from the same line. The result was an exponentially more rapid and immediate Tau pathology initiation than that demonstrated in the study performed before. They found the development of neuronal inclusions in the form of neuropil threads and neurofibrillary tangles beginning two weeks after the injection in the ipsilateral region and one month after in the contralateral region. Most notably, this series of experiments corroborated the hypothesis that pathological Tau can spread through connections between the synapses. These findings were additionally verified by the formation of Tau pathology in the white matter tracts linking regions with abundant toxic Tau aggregates.[28] To sum up, these studies demonstrate that Tau seeding is a mechanism that explains why Tau aggregation propagates throughout the brain.

Therapeutic Strategies to Treat Tauopathies:

A plenitude of Tau antibodies and vaccines have been tested in preclinical studies in the last two decades. Currently, eight Tau antibodies and two Tau vaccines have entered clinical trials for various tauopathies. Considering the failure of the clinical trials with amyloid targeting drugs, Tau therapy is manifesting as the frontrunner in the search for an effective treatment for Alzheimer's Disease. One such therapeutic is monoclonal antibodies (mABs) which are laboratory-produced molecules that act as substitute antibodies that can restore, enhance, or mimic the immune system's attack on cells. Since seeding mechanisms are driven by the passage of tau fibrils from cell to cell, antibodies are being designed to recognize these objects. Various mABs are being designed to target different Tau protein domains, demonstrating fruitful laboratory results.

It is well-known that Tau fibrils are hyperphosphorylated. Therefore, some mAbs have been designed to bind specific phosphorylation residues scattered along the Tau protein specifically. Research shows that using some mABs can successfully decrease the amount of Tau seeding when injected in transgenic AD Tau seeding mouse models. On a similar note, Dai *et al.*,[29] administered transgenic mice with injections of an mAB that targets the N-terminal region of Tau. The results

showed a decreased level of hyperphosphorylation and Tau seeding.

More recently, Courade *et al.*,[30] invented a screening tool to help classify the mAB that would be most effective against human Tau seeds and found mAB targeting the Tau mid-region to display the highest activity. This mAB was later shown to successfully halt the progression of Tau pathology following injection of human AD brain extracts in Tg mice expressing the human Tau P301L mutation. However, a concern that the authors of this paper had was that the specific mABs tested in their study after the screening tool did not thoroughly neutralize the seeding activity of the fibrillar tau in the AD extracts. This may be due to the conformation of Tau changing as it is being hyperphosphorylated, making it difficult for the mABs to bind to them. Furthermore, Gibbons *et al.*,[31] later identified two additional mABs, recognizing the abnormal conformation of tau fibrils, which can inhibit Tau pathology induced upon human AD brain extract inoculation in an aggressive amyloid pathology model (5xFAD mice). Confirming the efficacy of these treatments in models with better translational value, such as humanized Tau models injected with patient-derived Tau fibrils, is a crucial next step.[31]

Lastly, another potential therapeutic strategy to treat tauopathies is inhibiting Tau kinases from preventing hyperphosphorylation of tau. However, there is still uncertainty about which kinases are most relevant to Tau phosphorylation in neurons. Some ser/thr kinases have been proposed, such as glycogen synthase kinase-3β (GSK-3β), cell cycle-dependent kinase 5 (CDK5), MT-affinity regulated kinases (MARKs), protein kinase A (PKA), mitogen-activated kinases (MAPKs) and others.[32] Among these, the most well-studied and most endorsed are the GSK-3β and CDK5 kinases. Furthermore, there are convincing data demonstrating that changing the expression of GSK-3β or p25, an activator of CDK5, affects Tau pathology in transgenic mouse models. Lithium chloride and specific small synthetic molecules, which are inhibitors of the Tau kinase GSK-3β, have been shown to lower the amount of Tau phosphorylation and Tau deposits in transgenic mouse models of tauopathy.

Moreover, lithium chloride started to be clinically tested in AD patients. Unfortunately, no improvements in cognitive outcomes were observed in Phase 2 clinical study. Additionally, a non-competitive GSK-3β inhibitor (tideglusib) was recently evaluated in Phase 2 testing in PSP and AD patients, but it also failed to improve clinical outcomes.

The tertiary structure of a protein determines its function and what it can interact with. Because Tau in solution is a highly disordered protein and does not exhibit a stable tertiary structure, it remains a challenging protein target for structural analysis. The lack of Tau structure in the solution is not a barrier to understanding its function, just a barrier to being amenable to structure-function-based intervention. The lack of structural information about Tau limits the progress in neurodegeneration research and the development of effective therapeutic strategies. Some progress has been made in identifying a three-dimensional structure for Tau, such as the "paper clip" structure that was previously mentioned. However, this still does not provide sufficient information to design a therapeutic method because the "paper clip" structure was only briefly explored as a model for the Tau protein. Despite challenges regarding tau's tertiary structure, progress has been made in limiting Tau seeding and eventual aggregation through the use of monoclonal antibodies, refinement of antibody types through screening methods, and tau kinase inhibitors (see Table 1). This illustrates that novel strategies to overcome the gap in knowledge with respect to tau's structure are on the horizon.

Table 1: Pros and Cons of Recent Tauopathy Therapeutic Strategies.

Therapeutic Strategy	Pros	Cons
Monoclonal Antibodies (mAbs)	Decreased hyperphosphorylation and tau seeding in transgenic mouse models	Tau's structural changes as it gets hyperphosphorylated make it difficult for the mABs to neutralize the phosphorylation
Screening Tool	Classified the monoclonal antibody (mAB) that would be most effective against human tau seeds	
Tau Kinases Inhibitors	Certain tau kinases have been shown to decrease the amount of phosphorylation and tau deposits in transgenic mouse models	The tau kinase GSK-3β failed to improve clinical outcomes during Phase 2 clinical trials.

Conclusion

Hyperphosphorylation and fibrillization are linked to neurodegeneration and cognitive dysfunction in Alzheimer's Disease. Through the studies described in this paper, it is evident that Tau seeding makes aggregation more prevalent because the aggregates can spread from neuron to neuron through connections between the synapses. This demonstrates how one hyperphosphorylated Tau aggregate can propagate and spread throughout the brain. These discoveries have paved the way for new therapeutic strategies for neurodegenerative diseases that are primarily Tau targeted. For instance, by using monoclonal antibodies, phosphorylation sites on Tau can be specifically targeted to reduce Tau seeding. Another therapeutic strategy is using Tau kinase inhibitors to reduce phosphorylation. However, unfortunately, many of these strategies have not made it past Phase 2 clinical trials. Some argue that therapies that fail to demonstrate efficacy do so because it is too late to intervene when a patient has shown enough cognitive dysfunction to warrant inclusion in a tau therapy trial. Therefore, if the disease can be detected earlier, it would allow for earlier intervention and perhaps greater efficacy. Nevertheless, it is estimated that currently, 30 million people live with tauopathies, so it is incredibly pertinent to continue studying Tau aggregation and hyperphosphorylation to develop a therapeutic strategy that will hopefully advance the treatment and prevention of tauopathies in the future.

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References

1. Yiannopoulou, K. G.; Anastasiou, A. I.; Zachariou, V.; Pelidou, S.-H. Reasons for Failed Trials of Disease-Modifying Treatments for Alzheimer Disease and Their Contribution in Recent Research. *Biomedicines* **2019**, 7 (4), 97. https://doi.org/10.3390/biomedicines7040097.
2. Howard, J.; Hyman, A. A. Dynamics and Mechanics of the Microtubule plus End. *Nature* **2003**, 422 (6933), 753–758. https://doi.org/10.1038/nature01600.
3. Mitchison, T.; Kirschner, M. Dynamic Instability of Microtubule Growth. *Nature* **1984**, 312 (5991), 237–242. https://doi. org/10.

DOI: 10.36838/v5i1.21

38/312237a0.
4. Liu, Z.; Huang, Y. Advantages of Proteins Being Disordered. *Protein Sci. Publ. Protein Soc.* **2014**, 23 (5), 539–550. https://doi.org/10.1002/pro.2443.
5. Breuzard, G.; Hubert, P.; Nouar, R.; De Bessa, T.; Devred, F.; Barbier, P.; Sturgis, J. N.; Peyrot, V. Molecular Mechanisms of Tau Binding to Microtubules and Its Role in Microtubule Dynamics in Live Cells. *J. Cell Sci.* **2013**, 126 (Pt 13), 2810–2819. https://doi.org/10.1242/jcs.120832.
6. Goedert, M.; Spillantini, M. G.; Serpell, L. C.; Berriman, J.; Smith, M. J.; Jakes, R.; Crowther, R. A. From Genetics to Pathology: Tau and α-Synuclein Assemblies in Neurodegenerative Diseases. *Philos. Trans. Biol. Sci*. **2001**, 356 (1406), 213–227.
7. Lee, G.; Neve, R. L.; Kosik, K. S. The Microtubule Binding Domain of Tau Protein. *Neuron* **1989**, 2 (6), 1615–1624. https://doi. org/10.1016/0896-6273(89)90050-0.
8. Zabik, N. L.; Imhof, M. M.; Martic-Milne, S. Structural Evaluations of Tau Protein Conformation: Methodologies and Approaches. *Biochem. Cell Biol. Biochim. Biol. Cell.* **2017**, 95 (3), 338–349. https://doi.org/10.1139/bcb-2016-0227.
9. Goedert, M.; Spillantini, M. G.; Crowther, R. A. Cloning of a Big Tau Microtubule-Associated Protein Characteristic of the Peripheral Nervous System. *Proc. Natl. Acad. Sci. U. S. A.* **1992**, 89 (5), 1983 –1987. https://doi.org/10.1073/pnas.89.5.1983.
10. D'Souza, I.; Poorkaj, P.; Hong, M.; Nochlin, D.; Lee, V. M.-Y.; Bird, T. D.; Schellenberg, G. D. Missense and Silent Tau Gene Mutations Cause Frontotemporal Dementia with Parkinsonism-Chromosome 17 Type, by Affecting Multiple Alternative RNA Splicing Regulatory Elements. *Proc. Natl. Acad. Sci. U. S. A*. **1999**, 96 (10), 5598–5603.
11. Dyson, H. J.; Wright, P. E. Intrinsically Unstructured Proteins and Their Functions. *Nat. Rev. Mol. Cell Biol.* **2005**, 6 (3), 197–208. https://doi.org/10.1038/nrm1589.
12. Qi, H.; Cantrelle, F.-X.; Benhelli-Mokrani, H.; Smet-Nocca, C.; Buée, L.; Lippens, G.; Bonnefoy, E.; Galas, M.-C.; Landrieu, I. Nuclear Magnetic Resonance Spectroscopy Characterization of Interaction of Tau with DNA and Its Regulation by Phosphorylation. *Biochemistry* **2015**, 54 (7), 1525–1533. https://doi.org/ 10.1021/bi5014613.
13. Andronesi, O. C.; von Bergen, M.; Biernat, J.; Seidel, K.; Griesinger, C.; Mandelkow, E.; Baldus, M. Characterization of Alzheimer's-like Paired Helical Filaments from the Core Domain of Tau Protein Using Solid-State NMR Spectroscopy. *J. Am. Chem. Soc.* **2008**,130 (18), 5922–5928. https://doi.org/10.1021/ja7100517.
14. Shammas, S. L.; Garcia, G. A.; Kumar, S.; Kjaergaard, M.; Horrocks, M. H.; Shivji, N.; Mandelkow, E.; Knowles, T. P. J.; Mandelkow, E.; Klenerman, D. A Mechanistic Model of Tau Amyloid Aggregation Based on Direct Observation of Oligomers. *Nat. Commun*. **2015**, 6, 7025. https://doi.org/10.1038/ncomms8025.
15. Martin, L.; Latypova, X.; Terro, F. Post-Translational Modifications of Tau Protein: Implications for Alzheimer's Disease. *Neurochem. Int.* **2011**, 58 (4), 458–471. https://doi.org/10.1016/j.neuint.2010.12.023.
16. Jho, Y. S.; Zhulina, E. B.; Kim, M. W.; Pincus, P. A. Monte Carlo Simulations of Tau Proteins: Effect of Phosphorylation. *Biophys. J.* **2010**,99 (8),2387–2397.https://doi.org/10.1016/j.bpj.2010.06.056.
17. Hanger, D. P.; Gibb, G. M.; de Silva, R.; Boutajangout, A.; Brion, J.-P.; Revesz, T.; Lees, A. J.; Anderton, B. H. The Complex Relationship between Soluble and Insoluble Tau in Tauopathies Revealed by Efficient Dephosphorylation and Specific Antibodies. *FEBS Lett.* **2002**, 531 (3), 538–542. https://doi.org/10.1016/s0014-5793(02)03611-6.
18. Bramblett, G. T.; Goedert, M.; Jakes, R.; Merrick, S. E.; Trojanowski, J. Q.; Lee, V. M. Abnormal Tau Phosphorylation at Ser396 in Alzheimer's Disease Recapitulates Development and Contributes to Reduced Microtubule Binding. *Neuron* **1993**, 10 (6), 1089-1099. https://doi.org/10.1016/0896-6273(93)90057-x.
19. Yoshida, H.; Ihara, Y. Tau in Paired Helical Filaments Is Functionally Distinct from Fetal Tau: Assembly Incompetence of Paired Helical Filament-Tau - PubMed. *J Neurochem* **1993**, 61 (3). https://doi.org/10.1111/j.1471-4159.1993.tb03642.x.(1)
20. Hart, G. W.; Slawson, C.; Ramirez-Correa, G.; Lagerlof, O. Cross Talk Between O-GlcNAcylation and Phosphorylation: Roles in Signaling, Transcription, and Chronic Disease. *Annu. Rev. Biochem.* **2011**, 80, 825–858. https://doi.org/10.1146/annurev-biochem-060608-102511.
21. Hanger, D. P.; Byers, H. L.; Wray, S.; Leung, K.-Y.; Saxton, M. J.; Seereeram, A.; Reynolds, C. H.; Ward, M. A.; Anderton, B. H. Novel Phosphorylation Sites in Tau from Alzheimer Brain Suppor a Role for Casein Kinase 1 in Disease Pathogenesis. J. *Biol. Chem*. **2007**, 282 (32), 23645–23654. https://doi.org/10.1074/jbc.M703269200.
22. Mietelska-Porowska, A.; Wasik, U.; Goras, M.; Filipek, A.; Niewiadomska, G. Tau Protein Modifications and Interactions: Their Role in Function and Dysfunction. *Int. J. Mol. Sci*. **2014**, 15 (3), 4671-4713. https://doi.org/10.3390/ijms15034671.
23. Goedert, M.; Cuenda, A.; Craxton, M.; Jakes, R.; Cohen, P. Activation of the Novel Stress-Activated Protein Kinase SAPK4 by Cytokines and Cellular Stresses Is Mediated by SKK3 (MKK6); Comparison of Its Substrate Specificity with That of Other SAP Kinases. *EMBO J*. **1997**, 16 (12), 3563–3571. https://doi.org/10.1093/emboj/16.12.3563.
24. Phosphorylation of microtubule-associated protein tau by stress-activated protein kinases - Goedert - 1997 - FEBS Letters - *Wiley Online Library*. https://febs.onlinelibrary.wiley.com/doi/ full/10.1016/S0014-5793%2897%2900483-3 (accessed 2022-06-24).
25. Ishihara, T.; Hong, M.; Zhang, B.; Nakagawa, Y.; Lee, M. K.; Trojanowski, J. Q.; Lee, V. M.-Y. Age-Dependent Emergence and Progression of a Tauopathy in Transgenic Mice Overexpressing the Shortest Human Tau Isoform. *Neuron* **1999**, 24 (3), 751–762. https://doi.org/10.1016/S0896-6273(00)81127-7.
26. Braak, H.; Alafuzoff, I.; Arzberger, T.; Kretzschmar, H.; Del Tredici, K. Staging of Alzheimer Disease-Associated Neurofibrillary Pathology Using Paraffin Sections and Immunocytochemistry. *Acta Neuropathol. (Berl.)* **2006**, 112 (4), 389–404. https://doi. org/10.1007/s00401-006-0127-z.
27. Clavaguera, F.; Hench, J.; Lavenir, I.; Schweighauser, G.; Frank, S.; Goedert, M.; Tolnay, M. Peripheral Administration of Tau Aggregates Triggers Intracerebral Tauopathy in Transgenic Mice. A*cta Neuropathol. (Berl.)* **2014**, 127 (2), 299–301. https://doi.org/10.1007/s00401-013-1231-5.
28. Ahmed, Z.; Cooper, J.; Murray, T. K.; Garn, K.; McNaughton, E.; Clarke, H.; Parhizkar, S.; Ward, M. A.; Cavallini, A.; Jackson, S.; Bose, S.; Clavaguera, F.; Tolnay, M.; Lavenir, I.; Goedert, M.; Hutton, M. L.; O'Neill, M. J. A Novel in Vivo Model of Tau Propagation with Rapid and Progressive Neurofibrillary Tangle Pathology: The Pattern of Spread Is Determined by Connectivity, Not Proximity. *Acta Neuropathol. (Berl.)* **2014**, 127 (5), 667–683. https://doi.org/10.1007/s00401-014-1254-6.
29. Dai, C.-L.; Hu, W.; Tung, Y. C.; Liu, F.; Gong, C.-X.; Iqbal, K. K. Tau Passive Immunization Blocks Seeding and Spread of Alzheimer Hyperphosphorylated Tau-Induced Pathology in 3 x Tg-AD Mice. *Alzheimers Res. Ther.* **2018**, 10 (1), 13. https://doi.org/10.1186/s13195-018-0341-7.
30. Courade, J.-P.; Angers, R.; Mairet-Coello, G.; Pacico, N.; Tyson, K.; Lightwood, D.; Munro, R.; McMillan, D.; Griffin, R.; Baker, T.;

Starkie, D.; Nan, R.; Westwood, M.; Mushikiwabo, M.-L.; Jung, S.; Odede, G.; Sweeney, B.; Popplewell, A.; Burgess, G.; Downey, P.; Citron, M. Epitope Determines Efficacy of Therapeutic Anti-Tau Antibodies in a Functional Assay with Human Alzheimer Tau. *Acta Neuropathol. (Berl.)* **2018**, 136 (5), 729–745. https://doi.org/10.1007/s00401-018-1911-2.

31. Gibbons, G. S.; Kim, S.-J.; Wu, Q.; Riddle, D. M.; Leight, S. N.; Changolkar, L.; Xu, H.; Meymand, E. S.; O'Reilly, M.; Zhang, B.; Brunden, K. R.; Trojanowski, J. Q.; Lee, V. M. Y. Conformation-Selective Tau Monoclonal Antibodies Inhibit Tau Pathology in Primary Neurons and a Mouse Model of Alzheimer's Disease. *Mol. Neurodegener.* **2020**, 15 (1), 64. https://doi.org/10.1186/s13024-020-00404-5.
32. Lovestone, S.; Hartley, C. L.; Pearce, J.; Anderton, B. H. Phosphorylation of Tau by Glycogen Synthase Kinase-3 Beta in Intact Mammalian Cells: The Effects on the Organization and Stability of Microtubules. *Neuroscience* **1996**, 73 (4), 1145–1157. https://doi.org/10.1016/0306-4522(96)00126-1.

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