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Role of Tau Acetylation in Alzheimer's Disease and Chronic Traumatic Encephalopathy: The Way Forward for Successful Treatment

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Abstract

Progressive neurodegenerative diseases plague millions of individuals both in the United States and across the world. The current pathology of progressive neurodegenerative tauopathies, such as Alzheimer's disease (AD), Pick's disease, frontotemporal dementia (FTD), and progressive supranuclear palsy, primarily revolves around phosphorylation and hyperphosphorylation of the tau protein. However, more recent evidence suggests acetylation of tau protein at lysine 280 may be a critical step in molecular pathology of these neurodegenerative diseases prior to the tau hyperphosphorylation. Secondary injury cascades such as oxidative stress, endoplasmic reticulum stress, and neuroinflammation contribute to lasting damage within the brain and can be induced by a number of different risk factors. These injury cascades funnel into a common pathway of early tau acetylation, which may serve as the catalyst for progressive degeneration. The post translational modification of tau can result in production of toxic oligomers, contributing to reduced solubility as well as aggregation and formation of neurofibrillary tangles, the hallmark of AD pathology. Chronic Traumatic Encephalopathy (CTE), caused by repetitive brain trauma is also associated with a hyperphosphorylation of tau. We postulated acetylation of tau at lysine 280 in CTE disease could be present prior to the hyperphosphorylation and tested this hypothesis in CTE pathologic specimens. We also tested for ac-tau 280 in early stage Alzheimer's disease (Braak stage 1). Histopathological examination using the ac tau 280 antibody was performed in three Alzheimer's cases and three CTE patients. Presence of ac-tau 280 was confirmed in all cases at early sites of disease manifestation. These findings suggest that tau acetylation may precede tau phosphorylation and could be the first "triggering" event leading to neuronal loss. To the best of our knowledge, this is the first study to identify acetylation of the tau protein in CTE. Prevention

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Conflict of Interest

The authors declared that they have no conflict of interest.

of tau acetylation could possibly serve as a novel target for stopping neurodegeneration before it fully begins. In this study, we highlight what is known about tau acetylation and neurodegeneration.

Keywords

Tau acetylation; Neurodegenerative diseases; Pathologic changes; Secondary injury cascades; Novel treatment target

Introduction

Tau hyperphosphorylation and progression have long dominated the underlying dogma behind disease progression in Alzheimer's disease (AD) and Chronic Traumatic Encephalopathy (CTE). Multiple treatment strategies have been employed to prevent tau hyperphosphorylation or tau deposition as well as preventing amyloid deposition in AD. Unfortunately, these flawed hypotheses have failed to produce meaningful treatment options that can benefit patients. Our group and others have shown that looking earlier in the disease process may be more valuable in finding a treatment solution that can be clinically successful. Secondary injury mechanisms such as oxidative stress, endoplasmic reticulum (ER) stress, and neuroinflammation play a more important role in disease onset than previously assumed [1–4]. These secondary injury mechanisms can be primed by genetic predisposition and triggered by insults such as neurotrauma, drug abuse, and cardiovascular disease [5–7]. Once activated, these cascades lead to persistent damage to neurons and surrounding glia causing distinct tau acetylation [8]. It is at this crucial stage that the pivot point occurs on whether the brain recovers or progresses to neurodegeneration. Tau acetylation has been shown to both disengage tau from the microtubule and also facilitate tau aggregation [9]. Because of this, preventing tau acetylation is critical for stopping disease onset. Salsalate and methylene blue have both been shown to reduce tau acetylation in pre-clinical models, however the exact mechanism has not been fully elucidated and warrants further investigation [10]. It is likely that these drugs are limiting the expansion of secondary injury cascades following insult. In this review, we highlight the relationship of tau acetylation to AD and CTE and then discuss the most effective strategy for reducing tau acetylation via pharmaceutical intervention.

Current Understanding of AD

On November 3, 1906, Alois Alzheimer presented the first definitive microscopic evidence of the tau tangle pathology that has become characteristic of degenerative AD [11]. By using the same stain that Max Bielschowsky used four years earlier, he also described neuritic plaques that sparked the amyloid hypothesis years later. The pathology was accompanied by detailed clinical reports of progressive dementia for several years prior to death. These findings would set the framework of defining AD. In 1984 the amyloid protein was identified as the core of the neuritic plaque and the amyloid cascade hypothesis was born. Over the past 33 years scientists have been attempting to find safe and effective treatments to remove amyloid from the brains of AD patients. Pharmaceutical companies have been testing agents to slow production of amyloid as well as administering antibodies or vaccines

to remove amyloid from the brains of AD patients. Sadly, every anti-amyloid study to date has failed [12].

Proponents of the amyloid hypothesis still hold out hope that if such agents are administered at the asymptomatic stage or at the very early mild cognitive impairment stage positive results with acceptable side effects might be achieved [13]. More recently there has been a shift to look at pathologic tau in the progression of AD. It is the location and amount of tau found on autopsy in AD subjects that correlates best with stage and severity of symptomatology, not the amyloid plaque [14]. As far back as 1963, such tangles were noted to be composed of filaments with a diameter of approximately 10 nm that have come to be known as paired helical filaments (PHF) [15]. In 1992, Dreschel et al. first reported on the structural importance of the micro-tubular associated protein, tau in disease pathology [16].

It was at this point that research shifted towards understanding tau post-translational modifications. Normally tau has 83 potential sites of phosphorylation at various serine, threonine, and tyrosine sites of the 441 amino acid long tau protein. Excess p-tau is as high as 4–5× the normal level in the brain homogenates of AD brains when compared to control brains [17]. Because tau hyperphosphorylation was the first characterized, it garnered the most attention. In 1994, Khalid Iqbal was first to report that tau is a marker of neurofibrillary tangles in AD patients upon autopsy [18]. He and others noted that an excess phosphorylation (p-tau) of the human tau protein could be pathological [17]. In subsequent studies of AD and related “abnormal” tau associated disorders such as corticobasilar degeneration, progressive supranuclear palsy, Picks disease and chronic traumatic encephalopathy, excess of tau phosphorylation have been consistently reported [19]. Recently however evidence has emerged that p-tau is not pathogenic and is not responsible for the loss of neuronal function in AD [20].

Current Understanding of CTE

CTE is a devastating neurodegenerative disease triggered by head injury. Like other tauopathies, it is progressive in nature and contributes to both cognitive and functional decline. Omalu et al. described the modern version of CTE in retired professional athletes (21), which have been validated by McKee et al. [22]. The players had extensive histories of repetitive concussions followed by a series of impulsive events, cognitive decline, and frequently suicide. This original characterization of CTE sparked huge controversy and met resistance by sports organizations and scientists alike. Over the next decade, other groups validated the findings proposed by Omalu and began describing a very distinct pathologic paradigm [23]. Similar to Alzheimer’s disease, the disease correlation was most in line with the tau progression but not the amyloid hypothesis.

Our group and others began looking towards secondary injury mechanisms that can contribute to tauopathy. Promising pre-clinical data pointed to the role of endoplasmic reticulum stress and oxidative stress in the pathophysiology of neurodegeneration following neurotrauma [2,24]. The exact mechanism by which this occurs however was not completely elucidated. We found that ER stress is increased in cells that are undergoing apoptosis as well as those that develop tauopathy [25]. The key markers that were increased were markers of tau hyperphosphorylation AT100, PHF, and CP-13 [26]. It is apparent that tau

hyperphosphorylation is an end-stage marker. How these markers got increased and what maintained the progressive tauopathy is still under investigation. Kondo and colleagues proposed that cis tau might play a role [27]. Kaye et al. assert that it is more likely the tau oligomers [28]. We however have discovered an earlier contributor to the disease process in tau acetylation.

The Tau Pathology Catalyst

The lack of correlation between tau phosphorylation and functional decline sparked interest in understanding which tau modification actually does contribute to pathology. Tau is a very soluble hydrophilic protein [29]. Full-length tau remains soluble in solution up to 10M before it can aggregate. The tau repeat domain needs a concentration in solution of 4M to aggregate. High p-tau expressed in Sf9 cells (high phosphate) requires 0.2M before p-tau can aggregate. Since the concentration of p-tau in the CSF is 20 attomolar, 1 picomolar in the interstitial fluid, and 1.0 micromolar in the neurons, it would seem quite impossible to expect p-tau at the levels seen in human AD brains to aggregate without a catalyst [30]. Thus, there must be a nucleating factor that acts as the “seed” which leads to the phosphorylation seen in the neurofibrillary tangle. A neurofibrillary tangle is composed of a “fuzzy coat” or “soft polymer brush” [31]. This coat surrounds the core of tau fibers and can bind multiple cell components. Inside the tangle is a rigid fibril core. There are spokes emanating out of the core where two post translationally modified protein motifs exist known as hexapeptide PHF6* and PHF6. PHF6* has the following amino acid sequence: V Q I N N K whereas PHF6 has the following amino acid sequence: V Q I V Y K. Both hexapeptides are in the microtubular binding domain (MTBD) portion of the tau protein and are thought to be essential for the proper binding of tau onto the alpha and beta tubulin subunits of the microtubule [32].

Recent evidence suggests that a posttranslational acetylated modification of lysine at position 280 of the hexapeptide of the PHF6 can lead to pathological aggregation of tau [33]. Post mortem mass spectrometry analysis of AD brains has shown such acetylation occurs most specifically at lysine position 280. However, acetylation of lysine at positions 174, 274 and 281 has also been reported in other human tauopathies [34]. This acetylation may be due to the overactivity of the acetyltransferase enzyme p-300, which acts specifically on PHF6. Gorsky et al showed that even pseudo-acetylation of the single K280 residue by p-300 was able to exacerbate hTau neurotoxicity in vivo, which is suggestive that acetylated tau contributes to the pathology seen in neurodegenerative diseases [35].

After tau is acetylated, there is dislodgement of the microtubular binding domain from the tubulin due to the neutralization of charges between tau and tubulin molecules. This exposes the previously unphosphorylated serines/threonines/tyrosines to kinases leading to the robust phosphorylation so often seen in brain homogenates of AD and CTE patients [36]. It is likely that the hyperphosphorylation will only occur in the context of tau acetylation. A shift in favor of the tau kinases over the phosphatases most likely occurs [37]. In Irwin’s seminal paper, there is co-localization of acetylated K280 with multiple p-tau epitopes in post-mortem AD brains [38]. Their study showed that acetylated K280 occurred early in the pathogenesis of neurodegeneration. Originally Braak and Braak indicated that

phosphorylated tau in AD patients starts in the entorhinal and transentorhinal cortex and spreads up through the neocortex [39]. In 2012 Senanarong et al. reported a very early occurrence of AD-related cytoskeletal changes of p-tau in brainstem nuclei with likely spreading in a prion like manner (prionoid) up the white matter tracks to the neo/allocortex [40] (Figure 1). We highlight below that tau acetylation occurs in these exact same regions.

Methods

Human paraffin embedded specimens were collected from post-mortem samples of the entorhinal cortex for CTE brains (N = 3) and from the putamen, caudate, thalamus, brain stem, and cerebral white matter of AD brains (N = 3). Control samples were selected from age and gender matched controls that succumbed to non-neurologic diseases. Brain slices were cut to 10 μ m thickness with a Leica RM2265 microtome (Leica Biosystems). The slides were soaked in 99% formic acid for 10 minutes. A tau acetylation antibody for K280 was utilized. (Anaspec, rabbit polyclonal antibody). Staining of AD brain slices was conducted by Udo Rub and Kay Seidel at their laboratory of Goethe-University, Frankfurt/Main, Germany. The antibody was used at a concentration of 1:200. The standardized method of staining with primary and secondary antibodies was performed as has previously been published [2]. CTE brains were co-stained with MC1, which was kindly gifted from Dr. Peter Davies. The CTE images were analyzed with the Just another Co-localization plugin from Image J. An overlap coefficient was generated for each overlay. $R > 0.8$ indicates strong correlation, $R = 0.6-0.8$ equals moderate correlation, $R = 0.4-0.6 =$ weak correlation, and $R < 0.4$ is minimal overlay.

Results

AD Brains

Three Braak stage 1 brains were stained with the tau K280 acetylation antibody as well as matched controls. Significant staining was seen in the brain stem, caudate, and putamen (Figure 2). These regions were specifically chosen because of their importance for disease progression [41]. Slightly less staining was seen in the thalamus. Such early staining likely indicates the initial pathology in disease progression, which proceeds tau hyperphosphorylation. Similarly, staining was positive in the brainstem but was not seen in controls (Figure 3). Therefore, a strategy to slow tau acetylation seems plausible as a method to prevent disease onset and progression.

CTE Brains

Three CTE brains were stained and compared to age-matched controls. An overlay between acetylated tau K280 and MC1 was done. MC1 is an early marker of tau hyperphosphorylation seen in neurodegeneration. Figure 4 shows an overlap coefficient of $R = 0.33$, which indicates minimal co-staining for tau pathology in control brains. Figure 5 shows an overlap coefficient of $R = 0.97$, which indicates strong correlation for tau acetylation and hyperphosphorylation in the entorhinal cortex. The results show that tau acetylation is at least present at time of tau phosphorylation, but more likely precedes it. This is important in that tau acetylation may serve as a potential pharmacologic target.

Conclusions

Targeting tau phosphorylation has yielded little in terms of viable treatments for patients with neurodegeneration therefore urging a new strategy. Min and Gan from the Gladstone Institute tested the non-steroidal anti-inflammatory agent salsalate in pseudo-acetylated mice at a dose of 2.25 grams per day. The results indicated significant improvement in cognitive function due to reduction in p-300 induced tau acetylation and reduced hippocampal atrophy [42]. Lagraoui et al. further tested salsalate in a traumatic brain injury mouse model. Their report revealed that when salsalate was given post traumatic brain injury (TBI) there was a significant reduction in neuroinflammation, improved functional ability, as well as an upregulation of genes that are associated with neuroprotection and neurogenesis [43]. The likely mechanism is salsalate reducing ER stress and thereby limiting p-300 activity.

Going forward it is imperative to examine what actually drives disease pathogenesis. In this paper, we have outlined how tau acetylation plays a critical role in the process of neurodegeneration. Furthermore, we have shown that tau acetylation was increased both in human AD and CTE brain specimens. Future studies are warranted for how to target tau acetylation in pre-clinical models of AD and CTE in order to prevent disease progression and advance towards clinical trials. It is likely that ER stress, oxidative stress, and neuroinflammation are the triggers that activate the tau acetylation process. Pre-clinical work should flush out the mechanisms at play in order to advance towards clinical trials for potential treatments. An interdisciplinary team of clinicians and scientists will be necessary to tackle this important work and target the injury mechanisms as early as clinically detectable. Re-examining the tau hypothesis might be the paradigm shift needed for making headway on discovering new treatments for neurodegeneration.

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References

1. Lucke-Wold BP, Naser ZJ, Logsdon AF, Turner RC, Smith KE, Robson MJ, et al. Amelioration of nicotinamide adenine dinucleotide phosphate-oxidase mediated stress reduces cell death after blast-induced traumatic brain injury. *Transl Res.* 2015; 166(6):509–528.e1. DOI: 10.1016/j.trsl.2015.08.005 [PubMed: 26414010]
2. Lucke-Wold BP, Turner RC, Logsdon AF, Nguyen L, Bailes JE, Lee JM, et al. Endoplasmic reticulum stress implicated in chronic traumatic encephalopathy. *J Neurosurg.* 2016; 124(3):687–702. DOI: 10.3171/2015.3.JNS141802 [PubMed: 26381255]
3. Turner RC, Lucke-Wold BP, Robson MJ, Lee JM, Bailes JE. Alzheimer's disease and chronic traumatic encephalopathy: Distinct but possibly overlapping disease entities. *Brain Inj.* 2016; 30(11):1279–1292. DOI: 10.1080/02699052.2016.1193631 [PubMed: 27715315]
4. Remondelli P, Renna M. The Endoplasmic Reticulum Unfolded Protein Response in Neurodegenerative Disorders and Its Potential Therapeutic Significance. *Front Mol Neurosci.* 2017; 10:187.doi: 10.3389/fnmol.2017.00187 [PubMed: 28670265]
5. Armstrong RA, Mckee AC, Stein TD, Alvarez VE, Cairns NJ. A quantitative study of tau pathology in 11 cases of chronic traumatic encephalopathy. *Neuropathol Appl Neurobiol.* 2017; 43(2):154–166. DOI: 10.1111/nan.12323 [PubMed: 26998921]

6. Asken BM, Sullan MJ, Snyder AR, Houck ZM, Bryant VE, Hizel LP, et al. Factors Influencing Clinical Correlates of Chronic Traumatic Encephalopathy (CTE): a Review. *Neuropsychol Rev.* 2016; 26(4):340–363. [PubMed: 27561662]
7. Lucke-Wold BP, Turner RC, Logsdon AF, Simpkins JW, Alkon DL, Smith KE, et al. Common mechanisms of Alzheimer's disease and ischemic stroke: the role of protein kinase C in the progression of age-related neurodegeneration. *J Alzheimers Dis.* 2015; 43(3):711–24. DOI: 10.3233/JAD-141422 [PubMed: 25114088]
8. Marcelli S, Corbo M, Iannuzzi F, Negri L, Blandini F, Nistico R, et al. The Involvement of Post-Translational Modifications in Alzheimer's Disease. *Curr Alzheimer Res.* 2017; doi: 10.2174/1567205014666170505095109
9. Trzeciakiewicz H, Tseng JH, Wander CM, Madden V, Tripathy A, Yuan CX, et al. A Dual Pathogenic Mechanism Links Tau Acetylation to Sporadic Tauopathy. *Sci Rep.* 2017; 7:44102.doi: 10.1038/srep44102 [PubMed: 28287136]
10. Panza F, Solfrizzi V, Seripa D, Imbimbo BP, Lozupone M, Santamato A, et al. Tau-Centric Targets and Drugs in Clinical Development for the Treatment of Alzheimer's Disease. *Biomed Res Int.* 2016; 2016:3245935.doi: 10.1155/2016/3245935 [PubMed: 27429978]
11. Zilka N, Novak M. The tangled story of Alois Alzheimer. *Bratisl Lek Listy.* 2006; 107(9–10):343–5. [PubMed: 17262985]
12. Ricciarelli R, Fedele E. The amyloid cascade hypothesis in Alzheimer's disease: it's time to change our mind. *Curr Neuropharmacol.* 2017; 15(6):926–935. DOI: 10.2174/1570159X15666170116143743 [PubMed: 28093977]
13. Okazawa H. Ultra-Early Phase pathologies of Alzheimer's disease and other neurodegenerative diseases. *Proc Jpn Acad Ser B Phys Biol Sci.* 2017; 93(6):361–377. DOI: 10.2183/pjab.93.022
14. Sabbagh MN, Lue LF, Fayard D, Shi J. Increasing Precision of Clinical Diagnosis of Alzheimer's Disease Using a Combined Algorithm Incorporating Clinical and Novel Biomarker Data. *Neurol Ther.* 2017; 6(Suppl 1):83–95. DOI: 10.1007/s40120-017-0069-5 [PubMed: 28733959]
15. Kurt MA, Davies DC, Kidd M, Duff K, Howlett DR. Hyperphosphorylated tau and paired helical filament-like structures in the brains of mice carrying mutant amyloid precursor protein and mutant presenilin-1 transgenes. *Neurobiol Dis.* 2003; 14(1):89–97. [PubMed: 13678670]
16. Drechsel DN, Hyman AA, Cobb MH, Kirschner MW. Modulation of the dynamic instability of tubulin assembly by the microtubule-associated protein tau. *Mol Biol Cell.* 1992; 3(10):1141–1154. [PubMed: 1421571]
17. Khatoun S, Grundke-Iqbal I, Iqbal K. Levels of normal and abnormally phosphorylated tau in different cellular and regional compartments of Alzheimer disease and control brains. *FEBS Lett.* 1994; 351(1):80–4. [PubMed: 8076698]
18. Tatebayashi Y, Haque N, Tung YC, Iqbal K, Grundke-Iqbal I. Role of tau phosphorylation by glycogen synthase kinase-3beta in the regulation of organelle transport. *J Cell Sci.* 2004; 117(Pt 9):1653–63. [PubMed: 15075227]
19. Lee VM, Brunden KR, Hutton M, Trojanowski JQ. Developing therapeutic approaches to tau, selected kinases, and related neuronal protein targets. *Cold Spring Harb Perspect Med.* 2011; 1(1):a006437.doi: 10.1101/cshperspect.a006437 [PubMed: 22229117]
20. Lai RY, Harrington CR, Wischik CM. Absence of a Role for Phosphorylation in the Tau Pathology of Alzheimer's Disease. *Biomolecules.* 2016; 6(2) pii: E19. doi: 10.3390/biom6020019
21. Omalu BI, Dekosky ST, Minster RL, Kamboh MI, Hamilton RL, Wecht CH. Chronic traumatic encephalopathy in a National Football League player. *Neurosurgery.* 2005; 57(1):128–34. discussion 128–34.
22. Mckee AC, Cairns NJ, Dickson DW, Folkerth RD, Keene CD, Litvan I, et al. The first NINDS/NIBIB consensus meeting to define neuropathological criteria for the diagnosis of chronic traumatic encephalopathy. *Acta Neuropathol.* 2016; 131:75–86. [PubMed: 26667418]
23. Mckee AC, Cantu RC, Nowinski CJ, Hedley-Whyte ET, Gavett BE, Budson AE, et al. Chronic traumatic encephalopathy in athletes: progressive tauopathy after repetitive head injury. *J Neuropathol Exp Neurol.* 2009; 68(7):709–35. DOI: 10.1097/NEN.0b013e3181a9d503 [PubMed: 19535999]

24. Logsdon AF, Lucke-Wold BP, Nguyen L, Matsumoto RR, Turner RC, Rosen CL, et al. Salubralin reduces oxidative stress, neuroinflammation and impulsive-like behavior in a rodent model of traumatic brain injury. *Brain Res.* 2016; 1643:140–51. DOI: 10.1016/j.brainres.2016.04.063 [PubMed: 27131989]
25. Lucke-Wold BP, Logsdon AF, Turner RC, Huber JD, Rosen CL. Endoplasmic Reticulum Stress Modulation as a Target for Ameliorating Effects of Blast Induced Traumatic Brain Injury. *J Neurotrauma.* 2017; 34(S1):S62–S70. DOI: 10.1089/neu.2016.4680 [PubMed: 28077004]
26. Turner RC, Lucke-Wold BP, Logsdon AF, Robson MJ, Dashnaw ML, Huang JH, et al. The Quest to Model Chronic Traumatic Encephalopathy: A Multiple Model and Injury Paradigm Experience. *Front Neurol.* 2015 Oct 20;6:222.doi: 10.3389/fneur.2015.00222 [PubMed: 26539159]
27. Kondo A, Shahpasand K, Mannix R, Qiu J, Moncaster J, Chen CH, et al. Antibody against early driver of neurodegeneration cis P-tau blocks brain injury and tauopathy. *Nature.* 2015; 523(7561): 431–436. DOI: 10.1038/nature14658 [PubMed: 26176913]
28. Gerson J, Castillo-Carranza DL, Sengupta U, Bodani R, Prough DS, Dewitt DS, et al. Tau Oligomers Derived from Traumatic Brain Injury Cause Cognitive Impairment and Accelerate Onset of Pathology in Htau Mice. *J Neurotrauma.* 2016; 33(22):2034–2043. [PubMed: 26729399]
29. Zhang Y, Zhao J, Yin M, Cai Y, Liu S, Wang Y, et al. The influence of two functional genetic variants of GRK5 on tau phosphorylation and their association with Alzheimer’s disease risk. *Oncotarget.* 2017; 8(42):72714–72726. [PubMed: 29069820]
30. Fichou Y, Eschmann NA, Keller TJ, Han S. Conformation-based assay of tau protein aggregation. *Methods Cell Biol.* 2017; 141:89–112. DOI: 10.1016/bs.mcb.2017.06.008 [PubMed: 28882313]
31. Pickhardt M, Biernat J, Hubschmann S, Dennissen FJA, Timm T, Aho A, et al. Time course of Tau toxicity and pharmacologic prevention in a cell model of Tauopathy. *Neurobiol Aging.* 2017; 57:47–63. DOI: 10.1016/j.neurobiolaging.2017.04.022 [PubMed: 28600952]
32. Von Bergen M, Barghorn S, Li L, Marx A, Biernat J, Mandelkow EM, et al. Mutations of tau protein in frontotemporal dementia promote aggregation of paired helical filaments by enhancing local beta-structure. *J Biol Chem.* 2001; 276(51):48165–74. [PubMed: 11606569]
33. Cohen TJ, Guo JL, Hurtado DE, Kwong LK, Mills IP, Trojanowski JQ, et al. The acetylation of tau inhibits its function and promotes pathological tau aggregation. *Nat Commun.* 2011; 2:252.doi: 10.1038/ncomms1255 [PubMed: 21427723]
34. Min SW, Cho SH, Zhou Y, Schroeder S, Haroutunian V, Seeley WW, et al. Acetylation of tau inhibits its degradation and contributes to tauopathy. *Neuron.* 2010; 67(6):953–66. DOI: 10.1016/j.neuron.2010.08.044 [PubMed: 20869593]
35. Gorsky MK, Burnouf S, Dols J, Mandelkow E, Partridge L. Acetylation mimic of lysine 280 exacerbates human Tau neurotoxicity in vivo. *Sci Rep.* 2016; 6:22685.doi: 10.1038/srep22685 [PubMed: 26940749]
36. Gorsky MK, Burnouf S, Sofola-Adesakin O, Dols J, Augustin H, Weigelt CM, et al. Pseudo-acetylation of multiple sites on human Tau proteins alters Tau phosphorylation and microtubule binding, and ameliorates amyloid beta toxicity. *Sci Rep.* 2017; 7:9984. [PubMed: 28855586]
37. Lucke-Wold BP, Turner RC, Logsdon AF, Bailes JE, Huber JD, Rosen CL. Linking traumatic brain injury to chronic traumatic encephalopathy: identification of potential mechanisms leading to neurofibrillary tangle development. *J Neurotrauma.* 2014; 31(13):1129–38. DOI: 10.1089/neu.2013.3303 [PubMed: 24499307]
38. Irwin DJ, Cohen TJ, Grossman M, Arnold SE, Xie SX, Lee VM, et al. Acetylated tau, a novel pathological signature in Alzheimer’s disease and other tauopathies. *Brain.* 2012; 135(Pt 3):807–18. DOI: 10.1093/brain/aws013 [PubMed: 22366796]
39. Braak H, Braak E. Neuropathological staging of Alzheimer-related changes. *Acta Neuropathol.* 1991; 82(4):239–59. [PubMed: 1759558]
40. Senanarong V, Siwasariyanon N, Washirutmangkur L, Pongvarin N, Ratanabunakit C, Aoonkaew N, et al. Alzheimer’s disease dementia as the diagnosis best supported by the cerebrospinal fluid biomarkers: difference in cut-off levels from thai experience. *Int J Alzheimers Dis.* 2012; 2012:212063.doi: 10.1155/2012/212063 [PubMed: 22844634]

41. Elahi FM, Marx G, Cobigo Y, Staffaroni AM, Kornak J, Tosun D, et al. Longitudinal white matter change in frontotemporal dementia subtypes and sporadic late onset Alzheimer's disease. *Neuroimage Clin.* 2017; 16:595–603. DOI: 10.1016/j.nicl.2017.09.007 [PubMed: 28975068]
42. Min SW, Chen X, Tracy TE, Li Y, Zhou Y, Wang C, et al. Critical role of acetylation in tau-mediated neurodegeneration and cognitive deficits. *Nat Med.* 2015; 21(10):1154–62. DOI: 10.1038/nm.3951 [PubMed: 26390242]
43. Lagraoui M, Sukumar G, Latoche JR, Maynard SK, Dalgard CL, Schaefer BC. Salsalate treatment following traumatic brain injury reduces inflammation and promotes a neuroprotective and neurogenic transcriptional response with concomitant functional recovery. *Brain Behav Immun.* 2017; 61:96–109. DOI: 10.1016/j.bbi.2016.12.005 [PubMed: 27939247]

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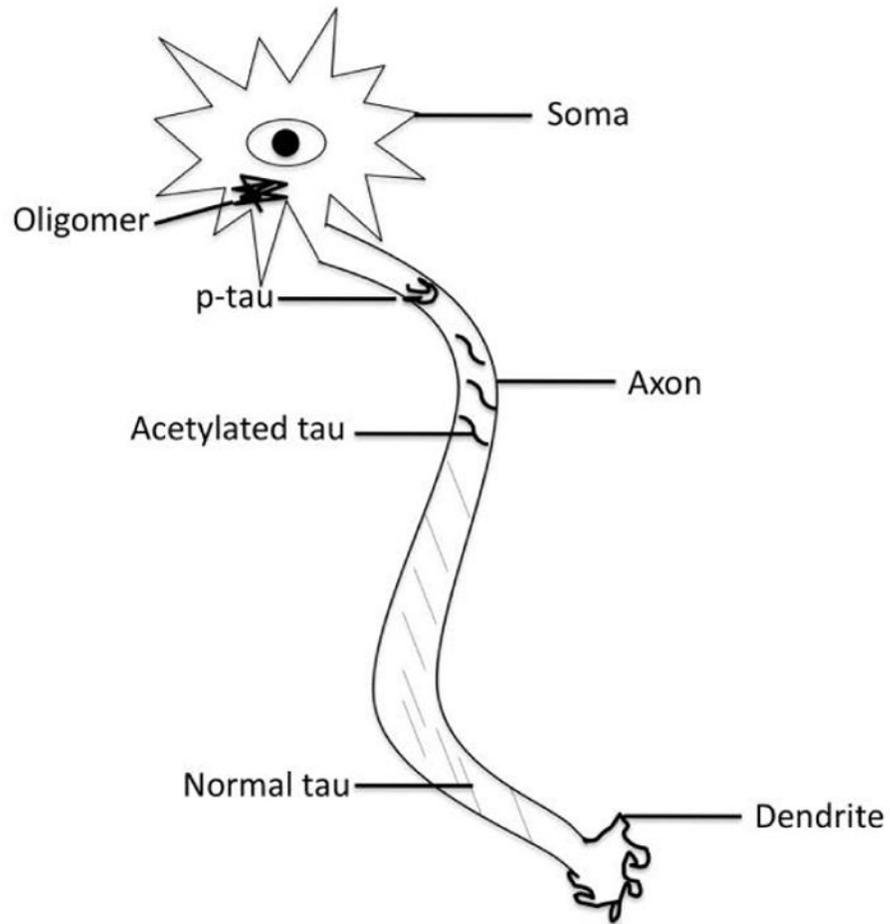


Figure 1. Schematic showing tau pathology progression up the axonal tracts. Tau becomes acetylated thereby exposing more phosphorylation sites. Once hyperphosphorylated tau aggregates into paired helical filaments, which ultimately produce tau oligomers and neurofibrillary tangles.

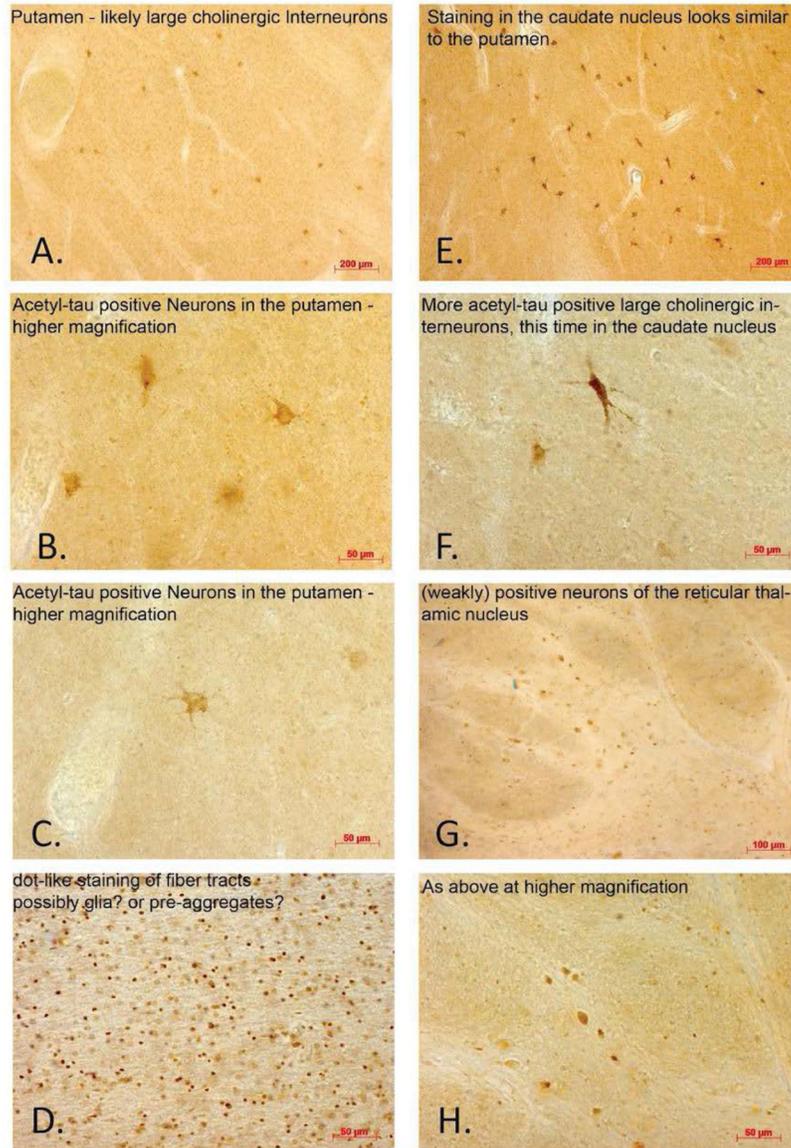


Figure 2. Tau acetylation at K280 in the putamen (A–D), caudate (E–F), and thalamus (G–H). These regions were chosen due to their known association with AD progression.

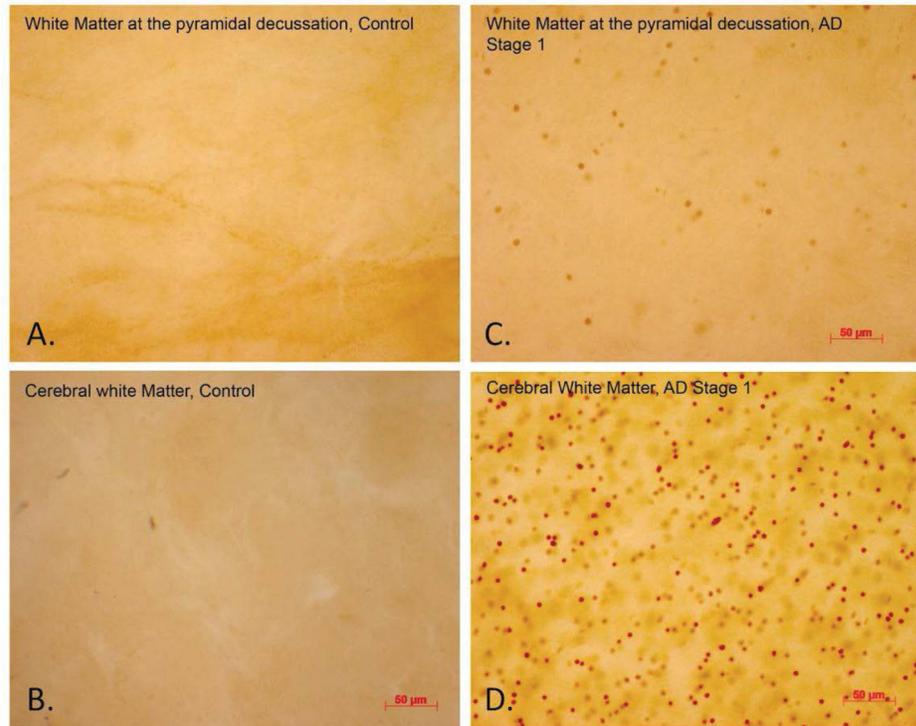


Figure 3. Tau acetylation at K280 in the brainstem of a control brain (A–B) and AD Braak Stage 1 (C–D). Tau acetylation was significantly more elevated in the AD brain versus the control.

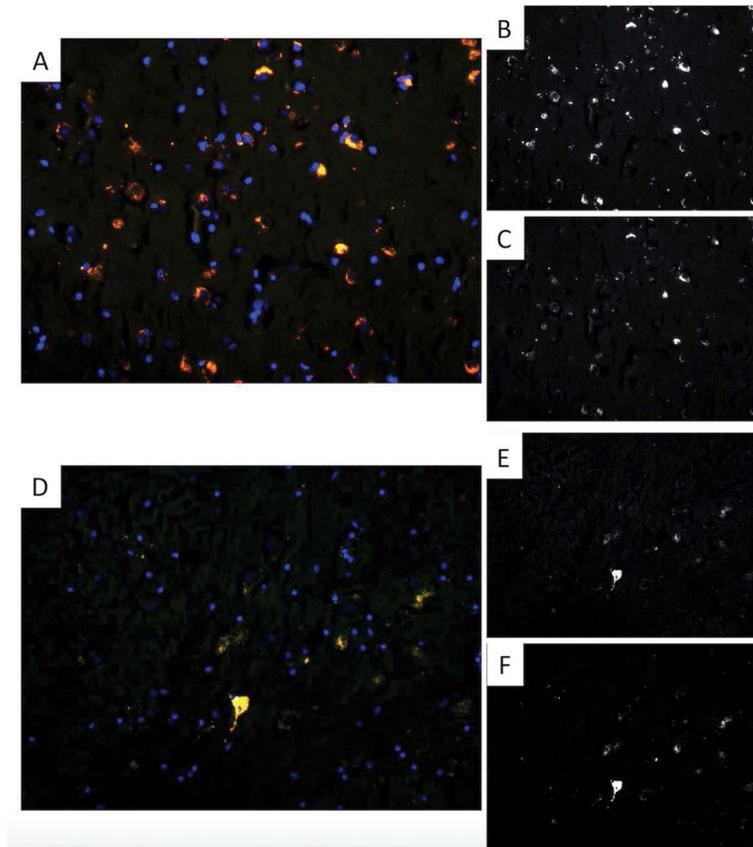


Figure 4. overlay of tau acetylation at K280 and phosphorylation (MC1) in entorhinal cortex of control brains. A) Overlay image at 20 \times , B) Acetylated tau K280 at 20 \times , C) MC1 at 20 \times . Overlay of tau acetylation at K280 and phosphorylation (MC1) in entorhinal cortex of CTE brains. D) Overlay image at 20 \times , E) Acetylated tau K280 at 20 \times , F) MC1 at 20 \times . Acetylation occurs at the same time or even prior to early tau phosphorylation.

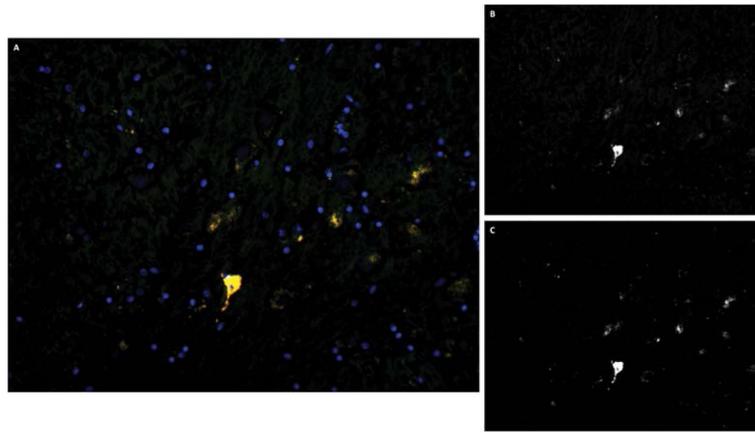


Figure 5. Overlay of tau acetylation at K280 and phosphorylation (MC1) in entorhinal cortex of CTE brains. A) overlay image at 20 \times , B) acetylated tau K280 at 20 \times , C) MC1 at 20 \times .