



# Possible Mechanisms of Tau Spread and Toxicity in Alzheimer's Disease

Huiqin Zhang, Yu Cao, Lina Ma, Yun Wei\* and Hao Li\*

*Institute of Geriatrics, Xiyuan Hospital, China Academy of Chinese Medical Sciences, Beijing, China*

## OPEN ACCESS

### Edited by:

Zhifang Dong,  
Chongqing Medical University, China

### Reviewed by:

Vladimir I. Titorenko,  
Concordia University, Canada  
Chandrasekar Raman,  
Joslin Diabetes Center and Harvard  
Medical School, United States

### \*Correspondence:

Yun Wei  
weiyun\_0913@163.com  
Hao Li  
xyhplihao1965@126.com

### Specialty section:

This article was submitted to  
Molecular and Cellular Pathology,  
a section of the journal  
Frontiers in Cell and Developmental  
Biology

**Received:** 09 May 2021

**Accepted:** 09 July 2021

**Published:** 28 July 2021

### Citation:

Zhang H, Cao Y, Ma L, Wei Y and  
Li H (2021) Possible Mechanisms of  
Tau Spread and Toxicity in Alzheimer's  
Disease.  
*Front. Cell Dev. Biol.* 9:707268.  
doi: 10.3389/fcell.2021.707268

Tau is a protein that associates with microtubules (MTs) and promotes their assembly and stability. The protein loses its ability to bind MTs in tauopathies, and detached tau can misfold and induce the pathological changes that characterize Alzheimer's disease (AD). A growing body of evidence indicates that tauopathies can spread between cells or connected regions. Pathological tau transmission in the brain of patients with AD and other tauopathies is due to the spread of various tau species along neuroanatomically connected regions in a "prion-like" manner. This complex process involves multiple steps of secretion, cellular uptake, transcellular transfer, and/or seeding, but the precise mechanisms of tau pathology propagation remain unclear. This review summarizes the current evidence on the nature of propagative tau species and the possible steps involved in the process of tau pathology spread, including detachment from MTs, degradations, and secretion, and discusses the different mechanisms underlying the spread of tau pathology.

**Keywords:** tau, tauopathy, spread, toxicity, mechanism, Alzheimer's disease

## INTRODUCTION

The microtubule (MT)-binding protein tau is mainly expressed in the cytoplasm of neurons (Pérez et al., 2016) and plays key roles in regulating MT dynamics, axonal transport, and neurite outgrowth (Johnson and Stoothoff, 2004). Tau protein changes affect its MT-binding ability and consequently alter its normal physiological functions. For example, the phosphorylation of tau protein in and around its microtubule-binding domain (MBD) may neutralize its positive charges (Jho et al., 2010), alter MBD conformation, and lead to its detachment from MTs (Fischer et al., 2009). Once detached, tau accumulates in neurites and neuronal cell bodies, where it forms insoluble intracellular aggregates or inclusion bodies such as neurofibrillary tangles (NFTs), which are one of the major pathological features of Alzheimer's disease (AD) (Lee et al., 2001; von Bergen et al., 2005; Zhang et al., 2009). Following detachment from MTs, tau can undergo structural transition, misfolding, and degradation (Frost et al., 2009). Tau can also be secreted into the extracellular space (Riemenschneider et al., 2003; Barthélemy et al., 2016) either in its naked form (Chai et al., 2012) or packaged in exosomes or other membranes (Saman et al., 2012; Simón et al., 2012; Polanco et al., 2016) following neuronal activity in mature neurons (Pooler et al., 2013; Dujardin et al., 2014b), neuron death (Gómez-Ramos et al., 2006), and/or when accumulated tau reaches a certain

level in non-neuronal cells. In agreement with these findings, exogenous misfolded tau protein can be internalized by cells (Guo and Lee, 2011; Wu et al., 2013), a process that is mediated by heparin sulphate proteoglycans (HSPGs) and cell membrane receptors such as muscarinic (M1, M3) and  $\alpha$ -amino-3-hydroxy-5-methyl-4-isoxazolepropionic acid (AMPA) receptors, as well as via endocytosis (Gómez-Ramos et al., 2008; Holmes et al., 2013; Tian H. et al., 2013). Once internalized, pathogenic misfolded tau proteins act as “seeds” that recruits soluble endogenous tau into larger aberrant conformations (Jucker and Walker, 2013) that slowly propagate across interconnected brain regions, as shown in various animal models (Clavaguera et al., 2009, 2013; Lasagna-Reeves et al., 2012). Fibrillar tau species can also transfer between cells and then recruit endogenous tau proteins onto their ends (Kfoury et al., 2012), a mechanism that may be responsible for the intracerebral spread of tau pathology (de Calignon et al., 2012; Iba et al., 2013).

Tau pathology spreading between neuronal cells and adjacent brain regions is a complex process involving many physiological and pathological aspects of tau protein, including its degradation, secretion, transmission, and toxicity. However, the exact mechanism underlying the spread of tau pathology after its release from cells remains unclear, and understanding these processes is the focus of an increasing number of studies (Le et al., 2012; Mohamed et al., 2013). There is some evidence that progressive accumulation of tau pathology in affected brain regions during AD development is due to the spread of aggregated tau along anatomically connected pathways (Hanger et al., 2014; Clavaguera et al., 2015; Lewis and Dickson, 2016). Accumulation of aggregates leads to neuronal loss and trans-synaptic spread of tau aggregates to more distal regions of the brain (Liu et al., 2012; Croft et al., 2017). The spread of extracellular species is the main pathway propagating neurofibrillary lesions and tau toxicity throughout different brain regions in neurodegenerative diseases (Iba et al., 2013; Pérez et al., 2018). A better understanding of the precise molecular mechanisms underlying tau propagation will contribute to the development of new therapeutic approaches for halting this process and provide new perspectives for the early diagnosis and prevention of tau pathologies (Fuster-Matanzo et al., 2018; Pérez et al., 2018). This review covers the most recent advances in our understanding of tau-spreading mechanisms, as well as the underlying implications of tauopathy-associated toxicity in AD. We further outline the possible mechanisms involved in pathology propagation including tau protein detachment from MTs; tau cleavage; tau degradation; and the release, uptake, and movement of pathogenic tau among synaptically connected neurons (Usenovic et al., 2015).

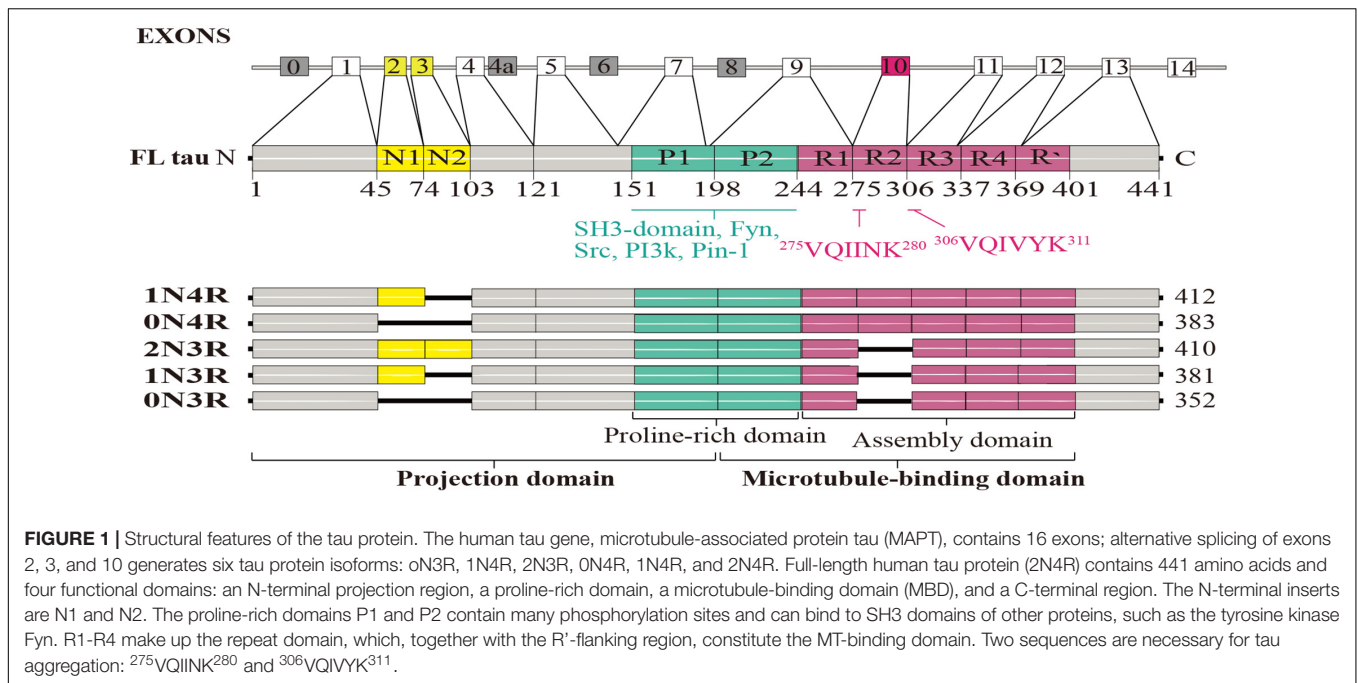
## PHYSIOLOGICAL CHARACTERISTICS AND DISSOCIATION OF TAU PROTEIN FROM MTs

Tau protein can be divided into four functional domains: an N-terminal projection region, a proline-rich domain, an MBD, and a C-terminal region (Goedert and Spillantini, 2011). Tau can

bind to the outside—and possibly also the inside—of MTs with the N- and C-terminal regions projecting out (Kar et al., 2003; Santarella et al., 2004). The N-terminal region can associate with the cell membrane and may be as a part of a membrane-associated complex; it also regulates the spacing between MTs (Maas et al., 2000; Al-Bassam et al., 2002). The proline-rich domain includes multiple phosphorylation sites (Augustinack et al., 2002) and can bind to Src homology 3 (SH3) domains of other proteins (Reynolds et al., 2008) such as the tyrosine kinase Fyn (Lee et al., 1998; **Figure 1**). Tau protein not only plays a crucial role in regulating MT dynamics but also promotes MT assembly and stabilization, processes that are required for morphogenesis and axonal transport in the nervous system (Johnson and Hartigan, 1999). However, the ability of tau to stabilize MTs is due in large part to its MBD (Gustke et al., 1994). Tau is thought to directly bind MTs through positively charged tandem repeat sequences within its MBD that are attracted to tubulin's negatively charged residues (Kar et al., 2003; Jho et al., 2010).

The human tau gene, microtubule-associated protein tau (MAPT), is located on chromosome 17q21 and comprises 16 exons. Alternative splicing of exons 2, 3, and 10 generates six isoforms of the tau protein (Goedert et al., 1989). They are equally expressed in central nervous system neurons of a healthy adult brain (Goedert et al., 1989; Garcia and Cleveland, 2001), and can be grouped into tau-3R class members, which contain three MT-binding repeats (MTBRs), and tau-4R class members contain four MTBRs (**Figure 1**). Because of the extra repeat, 4R isoforms have a higher affinity for MTs and can therefore bind and stabilize MTs more efficiently (Goedert and Spillantini, 2011; Morris et al., 2011; Chen and Jiang, 2019).

Tau protein functions are regulated by complex post-translational modifications including phosphorylation, glycation, isomerization, sumoylation, nitration, acetylation, and truncation (Morris et al., 2011). Moreover, tau contains numerous serine and threonine residues, so almost 20% of the protein has the potential to be phosphorylated (Wang and Mandelkow, 2016). The phosphorylation state of tau and its MT-binding affinity are controlled by a balance between kinase and phosphatase activity (Brandt et al., 1995; Shackelford and Yeh, 1998). Tau phosphorylation is mediated by MT affinity-regulating kinases (also known as PAR1 kinases), cyclic AMP-dependent protein kinase A, calcium ( $\text{Ca}^{2+}$ ), or calmodulin-dependent protein kinase II (CaMKII), and tyrosine kinases like Src family members (Hanger et al., 2009). The activation of tau phosphorylation-associated kinases (e.g., CDK-5 and GSK-3 $\beta$ ) can induce tau hyperphosphorylation, which drives dissociation of tau protein from MTs (Hanger et al., 2009). Dissociated tau can misfold and become toxic seeds that are secreted from the cell. In contrast, fully dephosphorylated tau binds to MTs with high affinity (Shackelford and Yeh, 1998). Tau dephosphorylation is mediated by protein phosphatases 1, 2A, 2B, 2C, and 5 (Hanger et al., 2009; Pérez et al., 2018). In addition, detached tau can accumulate in neurites and neuronal cell bodies, first forming insoluble filaments and eventually NFTs (Lee et al., 2001; von Bergen et al., 2005; **Figure 2**). Abnormal tau phosphorylation decreases MT binding and likely increases tau-tau interactions (Morris et al., 2011). Physiologically,

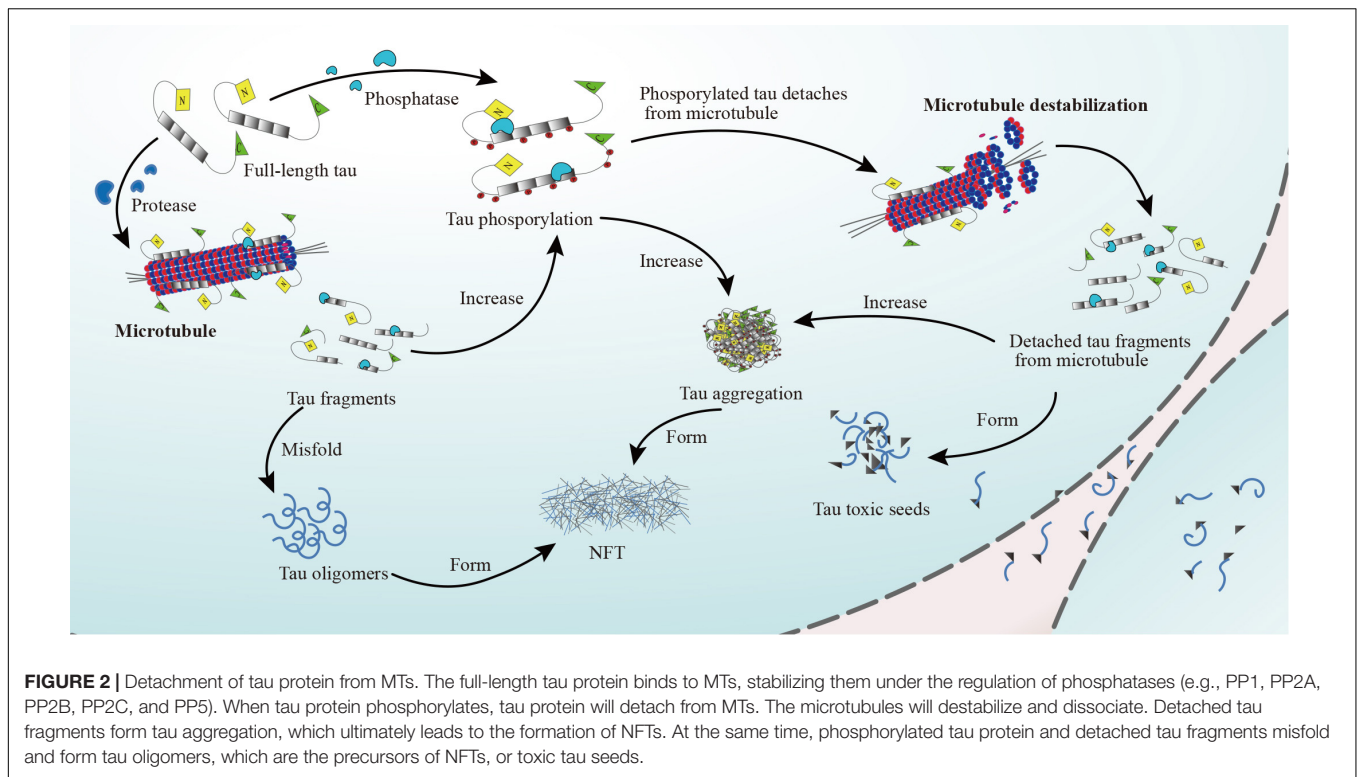


tau is continuously phosphorylated and dephosphorylated to ensure its proper function; however, when the balance shifts toward phosphorylation, tau affinity for MTs decreases (Alonso et al., 1997), resulting in higher cytosolic tau levels, which facilitates tau aggregation (Wegmann et al., 2018). Additionally, aberrantly phosphorylated tau protein appears to sequester other microtubule-associated proteins (MAPs), further destabilizing MTs (Alonso et al., 1997). Similarly, aberrant tau phosphorylation and self-aggregation lead to the formation of oligomers and higher-order aggregates that can lead to tau detachment from MTs and disturb the binding of other MAPs to MTs (von Bergen et al., 2000).

## TAU PROTEIN CLEAVAGE AND DEGRADATION

Tau cleavage and degradation are closely related to its pathological transmission and aggregation. The cleavage of tau generates seeds that promote tau aggregation (Wang et al., 2014), alter tau clearance, and can impair cognition and motor ability (Bondulich et al., 2016). Tau cleavage occurs at its N- and C-terminals (García-Sierra et al., 2008) and depends on associated proteases, mainly caspase-3, calpain, and cathepsin L (Wang et al., 2009; de Calignon et al., 2010; **Table 1**). Tau truncation can be initiated by caspase-3, which cleaves tau at residue D421, its predominant target (Chung et al., 2001). Caspase-3 can be activated by amyloid-beta (A $\beta$ ) and caspase-2 (Gamblin et al., 2003), following which caspase-3 can cleave tau at D25-Q26 (Corsetti et al., 2008) and D421-S422 (Gamblin et al., 2003). Cleavage at D421-S422 produces the N-terminal fragment (NTF) tau<sub>1-421</sub> (Tau-C) (Nicholls et al., 2017). Caspase-2 is a protease that initiates activation of other caspases; it cleaves tau

at D314-L315 to produce a soluble, toxic NTF (tau<sub>1-314</sub>;  $\Delta$ tau<sub>314</sub>; Zhao et al., 2016). Tau<sub>1-314</sub> levels were elevated in the brains of mice with mild cognitive impairment (MCI) and in the brains of AD patients compared with healthy controls (Zhao et al., 2016). Moreover, memory deficits were ameliorated following the application of anticaspase-2 morpholino oligonucleotides (Zhao et al., 2016). However, *in vitro* experiments using recombinant tau preparations suggested that caspase-2 preferentially cleaves tau at D421-S422 (Zhao et al., 2016). Another effector caspase, caspase-6, and puromycin-sensitive aminopeptidase (PSA) were reported to cleave recombinant human tau<sub>441</sub> at D13-H14 (Sengupta et al., 2006), which was sufficient to cause axonal degeneration (Sokolowski et al., 2014). Caspase-6 cleavage at D402-T403 produces the NTF tau<sub>1-402</sub>, a cerebrospinal fluid (CSF) biomarker for AD (Ramcharitar et al., 2013). In addition, calpain-1 and -2 can both cleave tau (Chesser et al., 2013) and play opposing roles in regulating synaptic plasticity and promoting neurodegeneration (Baudry and Bi, 2016). The 17-kDa tau<sub>45-230</sub> fragment is generated through cleavage by calpain-1 at K44-E45 (Yang and Ksiezak-Reding, 1995) or via calpain-1 (Park and Ferreira, 2005) or -2 (Garg et al., 2011) action at R230-T231. Tau is also cleaved by calpain-1 at R242-L243 to produce the 24-kDa C-terminal fragment (CTF) tau<sub>243-441</sub> (Matsumoto et al., 2015). The levels of the tau<sub>243-441</sub> fragment increase with aging in a tauopathy mouse model (Tg601 mice expressing wild-type human tau), and CTFs with sizes ranging from 20 to 28 kDa are present in brain samples from patients with AD and familial frontotemporal dementia (Matsumoto et al., 2015). Tau<sub>243-441</sub> can proficiently propagate to other tau-expressing cells, leading to further seeding and tau<sub>441</sub> phosphorylation (Matsumoto et al., 2015). Interestingly, tau<sub>441</sub> build up can activate calpain-2, which leads to the degradation of nicotinic acetylcholine receptor subunit 4 (Yin et al., 2016), a crucial



component of cholinergic signaling. Calpain-2 activation by  $\tau_{441}$  creates a positive feedback loop that enhances neurotoxic tau fragment generation. Cathepsins B, D, and L can also proteolytically cleave tau. One study reported that cathepsin B was associated with intracellular NFTs, and its expression was elevated around A $\beta$  plaques (Li et al., 1993). Cathepsin D can cleave recombinant tau at F8-E9, M419-V420, and L436-A437; there is another potential cleavage site at either T427-L428 or L428-A429 and additional cleavage sites in D34-G161, P200-K257, and K267-D358 (Kenessey et al., 1997). In Neuro-2A murine cells, cathepsin L can cleave  $\tau_{244-372}$  (lacking K280;  $\tau_{RD\Delta K}$ ), which is a mutated version of the aggregation-prone MBD tau fragment (García-Sierra et al., 2008).  $\tau_{RD\Delta K}$  cleavage by cathepsin L depends on an initial cleavage at K257-S258 by an unknown cytosolic protease that generates  $\tau_{258-372}$ ; this fragment is further cleaved by cathepsin L at V363-P364 to produce  $\tau_{258-363}$  that is subsequently cleaved at I360-T361 to produce  $\tau_{258-360}$  (García-Sierra et al., 2008).  $\tau_{258-360}$  and  $\tau_{258-363}$  induce aggregation of intact  $\tau_{RD\Delta K}$  and full-length tau, which is coincident with lysosomal leakage (García-Sierra et al., 2008). Asparagine endopeptidase-mediated tau cleavage occurs at both N255-V256 and N368-K369 and produces five tau fragments. Among them,  $\tau_{1-368}$  and  $\tau_{256-368}$  were the only critical drivers of enhanced apoptosis in rat primary neurons, while only  $\tau_{1-368}$  has been found in the brains of patients with AD (Zhang et al., 2014; Quinn et al., 2018).

Intracellular tau degradation mainly involves two major proteolytic systems: ubiquitin-proteasome and autophagy-lysosomal (Chesser et al., 2013; Lee et al., 2013; Guo et al., 2017). Full-length tau is cleared via the former system (Liu et al., 2009;

Dolan and Johnson, 2010), whereas its mutated and truncated forms appear to be degraded through the latter pathway (García-Sierra et al., 2008; Fernández-Montoya and Pérez, 2015). Moreover, tau phosphorylation can exacerbate its proteolytic degradation (Kenessey et al., 1997), while hyperphosphorylation is associated with impaired tau degradation via the ubiquitin-proteasome system (Dickey et al., 2007) and tau secretion. Tau can undergo natural self-degradation at cysteine residues by acetyl-coenzyme A-induced autoacetylation (Cohen et al., 2016) following its dissociation from MTs (Cohen et al., 2013, 2016). Tau accumulation can also result from increased expression or decreased degradation of the protein (Barton et al., 1990; Zhang et al., 2014), and degradation is impaired by a modified form of tau (Zhang et al., 2014). Thus, tau acetylation may both inhibit and facilitate its degradation and also suppress its phosphorylation and aggregation (Min et al., 2010; Cook et al., 2014). Acetylated tau has been found in brains from patients with AD and other tauopathies. For example, Lys174 acetylation was recently described in AD brains and may be a critical determinant for tau-induced toxicity by delaying tau turnover (Min et al., 2015). This result indicates that targeting tau acetylation could be a novel therapeutic option for AD and other human tauopathies.

## SECRETION AND RELEASE OF TAU FRAGMENTS

Although tau is intracellular, recent studies have indicated that it is also present in the extracellular space both *in vitro*

**TABLE 1** | Tau protein cleavage at different sites by known proteases.

Protease	Cleavage site	Cleavage domain	Tau fragment	Effect on AD	References
Caspase-2	D314-L315	MBD	Tau <sub>1-314</sub> ( $\Delta$ Tau <sub>314</sub> )	Caspase-2 preferentially cleaves recombinant tau at D421-S422 compared to D314-L315. Detached from MTs, tau invades healthy dendritic spines. This impaired synaptic transmission and drove hippocampal neuronal loss, but spatial memory deficits and toxicity were only observed when tau <sub>1-314</sub> promoted mislocalization of full-length tau to dendritic spines.	Zhao et al., 2016
Caspase-3	D421-S422	C-terminus	Tau <sub>315-441</sub> Tau <sub>1-421</sub> (Tau-C)	Unclear Linked to other tauopathies	Zhao et al., 2016 Gamblin et al., 2003; Corsetti et al., 2008; Zhao et al., 2015
Caspase-3	D25-Q26	N-terminus	Tau <sub>1-25</sub>	No toxicity to neurons	Corsetti et al., 2008
Caspase-3 Calpain-1	D25-Q26 K44-E45	N-terminus	Tau <sub>26-44</sub>	Caused NMDAR-mediated cell death in rat CGCs	Gamblin et al., 2003; Park and Ferreira, 2005; Corsetti et al., 2008; Garg et al., 2011; Zhao et al., 2015
Caspase-3 Calpain-1 Calpain-2	D25-Q26	N-terminus	Tau <sub>26-230</sub> (20–22kDa fragment)	Enriched in synaptic mitochondria; binds to A $\beta$ peptides and exacerbates mitochondrial dysfunction. Induced NMDAR-mediated death of rat CGCs.	Gamblin et al., 2003; Park and Ferreira, 2005; Corsetti et al., 2008; Garg et al., 2011; Zhao et al., 2015
Caspase-6	R230-T231 D13-H14	MBD N-terminus	Tau <sub>1-13</sub> Tau <sub>14-441</sub>	Caused axonal degeneration Possible role in tangle maturation	Sengupta et al., 2006; Sokolowski et al., 2014 Sengupta et al., 2006; Sokolowski et al., 2014
	D402-T403	C-terminus	Tau <sub>1-402</sub> (Tau $\Delta$ Casp6)	Serves as a CSF biomarker of neurodegeneration in AD	Ramcharitar et al., 2013
Caspase-1, -3, -6, -7, -8	D421-S422	C-terminus	Tau <sub>403-441</sub> Tau <sub>422-441</sub>	Unclear Unclear	Ramcharitar et al., 2013 Quinn et al., 2018
	D421-S422	C-terminus	Tau <sub>151-421</sub> ( $\Delta$ tau)	Led to tau aggregation and disrupted axonal transport, mitochondrial function, Golgi apparatus, and synaptic protein levels	
PSA	K150-I151 D13-H14	N-terminus N-terminus	Tau <sub>1-13</sub>	Caused axonal degeneration	Sengupta et al., 2006; Sokolowski et al., 2014
Calpain-1	K44-E45	N-terminus	Tau <sub>1-44</sub> Tau <sub>45-441</sub>	Caused NMDAR-mediated cell death in rat CGCs Unclear	Park and Ferreira, 2005; Garg et al., 2011 Yang and Ksiezak-Reding, 1995
	R242-L243	MBD	Tau <sub>243-441</sub> (24kDa CTF)	Accelerated the propagation to other tau-expressing cells, causing further seeding of aggregates and tau <sub>441</sub> phosphorylation; reduced capacity for promoting MT assembly compared with tau <sub>441</sub>	Matsumoto et al., 2015
Calpain-1 and thrombin, Calpain-1 and -2	K44-E45	N-terminus	Tau <sub>1-242</sub> Tau <sub>45-230</sub> (17kDa fragment)	Unclear Caused synapse loss and behavioral abnormalities; impaired organelle transport	Matsumoto et al., 2015 Yang and Ksiezak-Reding, 1995; Park and Ferreira, 2005; Garg et al., 2011; Quinn et al., 2018
Calpain-2 and thrombin, Calpain-1 and -2	R230-T231 A2-E3	MBD N-terminus	Tau <sub>3-230</sub>	Unclear	Quinn et al., 2018
	R230-T231 R230-T231 Q124-A125	MBD MBD Projection domain	Tau <sub>125-230</sub>	Not toxic	

(Continued)

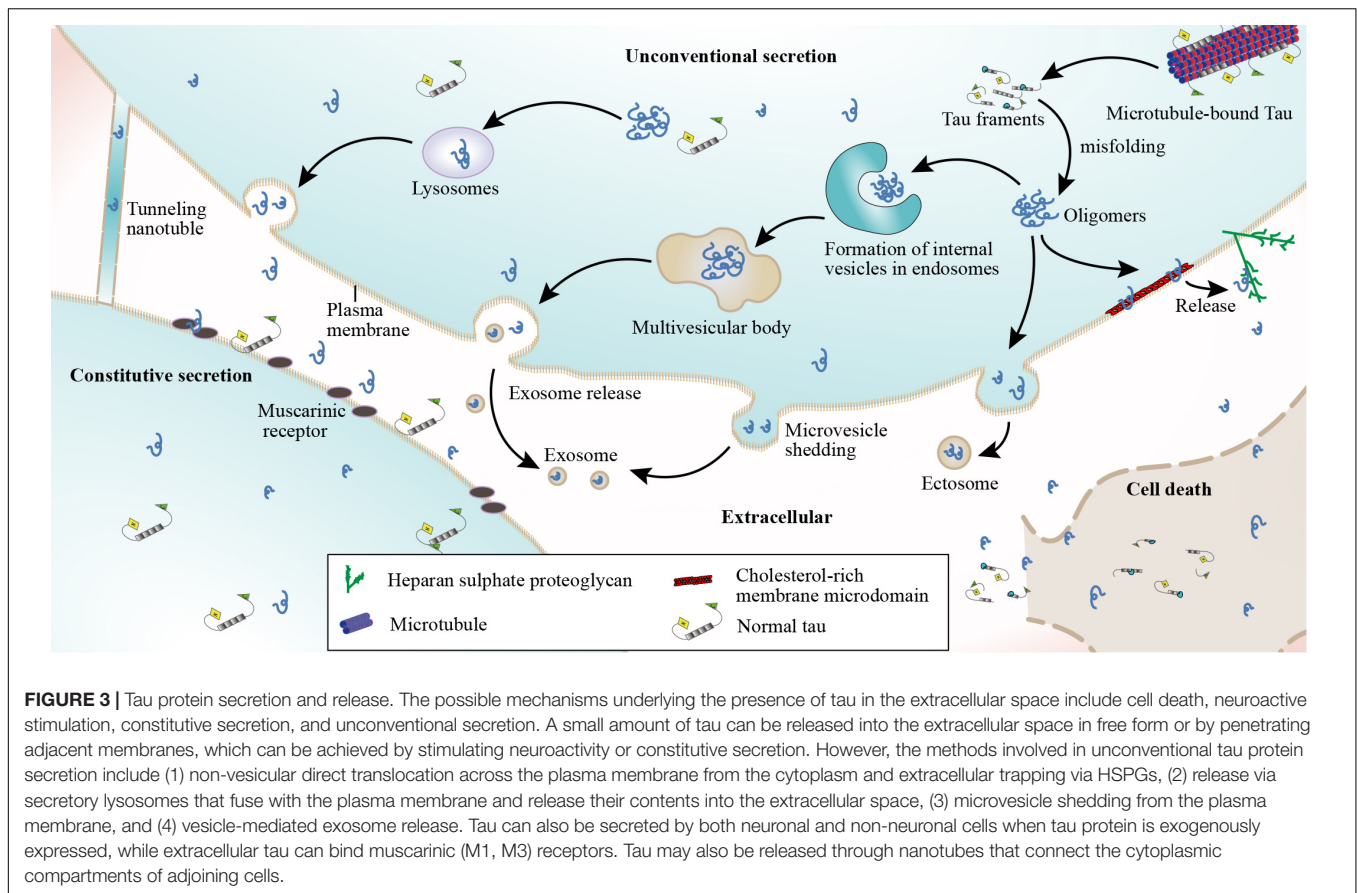
TABLE 1 | Continued

Protease	Cleavage site	Cleavage domain	Tau fragment	Effect on AD	References
Calpain-2	A2-E3	N-terminus	Tau <sub>3-124</sub>	Unclear	Quinn et al., 2018
	Q124-A125	Projection domain			
Cathepsin L	K257-S258	MBD	Tau <sub>258-372</sub>	Unclear	García-Sierra et al., 2008
	V363-P364	MBD	Tau <sub>258-363</sub>	Induced the aggregation of full-length tau and intact tau <sub>RD</sub> ΔK coincident with lysosomal leakage	
AEP	I360-T361	MBD	Tau <sub>258-360</sub>		Zhang et al., 2014; Quinn et al., 2018
	N255-V256	MBD	Tau <sub>1-255</sub>	Unable to promote MT polymerization or aggregation into PHFs, but tau <sub>1-255</sub> had strong AT8 immunoreactivity (at phosphorylation sites S202 and T205)	
	N255-V256	MBD	Tau <sub>256-441</sub>	Significantly reduced MT polymerization and showed increased propensity to aggregate into PHFs compared with tau <sub>441</sub>	
	N255-V256	MBD	Tau <sub>1-368</sub> , Tau <sub>256-368</sub>	Enhanced apoptosis; increased ability to aggregate into PHFs	
	N368-K369 N368-K369	MBD MBD	Tau <sub>369-441</sub>	Unable to cause MT polymerization and aggregation into PHFs	
Thrombin	R155-G156	Proline-rich domain	Tau <sub>156-441</sub>	Unclear	Quinn et al., 2018
	R155-G156	Proline-rich domain	Tau <sub>156-209</sub>	Unclear	
	R209-S210	MBD			
	R209-S210	MBD	Tau <sub>210-441</sub>	Unclear	
	R209-S210	MBD	Tau <sub>210-230</sub>	Unclear	
	R230-T231	MBD			
Chymotrypsin	R230-T231	MBD	Tau <sub>231-441</sub>	Unclear	Quinn et al., 2018
	Y197-S198	Projection domain	Tau <sub>1-197</sub>	Unclear	
ADAM10	Y197-S198	MBD	Tau <sub>198-441</sub>	Unclear	Quinn et al., 2018
	A152-T153	Proline-rich domain	Tau <sub>1-152</sub>	Unclear	
			Tau <sub>153-441</sub> (Tau-A)	Unclear	

Aβ, amyloid-beta; AD, Alzheimer's disease; ADAM10, a disintegrin and metalloprotease 10; AEP, asparagine endopeptidase; AMPA, α-amino-3-hydroxy-5-methyl-4-isoxazolepropionic acid; CGC, cerebellar granule cell; CSF, cerebrospinal fluid; CTF, C-terminal fragment; MBD, microtubule-binding domain; MT, microtubule; NFT, neurofibrillary tangle; NMDAR, N-methyl-D-aspartate receptor; NTF, N-terminal fragment; PHF, paired-helical filament; PSA, puromycin-sensitive aminopeptidase; PSP, progressive supranuclear palsy.

and *in vivo* (Kim et al., 2010a,b; Chai et al., 2012). Tau has been detected in both the CSF and interstitial fluid of tau transgenic mouse brains (Yamada et al., 2011; Barten et al., 2012). *In vitro* studies have shown that human tau is secreted by both neuronal and non-neuronal cell lines when the protein is overexpressed (Chai et al., 2012; Saman et al., 2012; Simón et al., 2012; Pérez et al., 2016). Extracellular tau may elicit toxicity (Gómez-Ramos et al., 2006; Díaz-Hernández et al., 2010) by binding to cellular receptors such as muscarinic receptors (Gómez-Ramos et al., 2008), but the mechanisms by which tau exits into the extracellular space remain unclear (Nickel and Rabouille, 2009). Another study showed that tau can be released into the extracellular space following neuronal death (Simón et al., 2012) and can subsequently be identified in the CSF (Iqbal et al., 2005). Secreted extracellular tau can be toxic to surrounding cells through interactions with specific

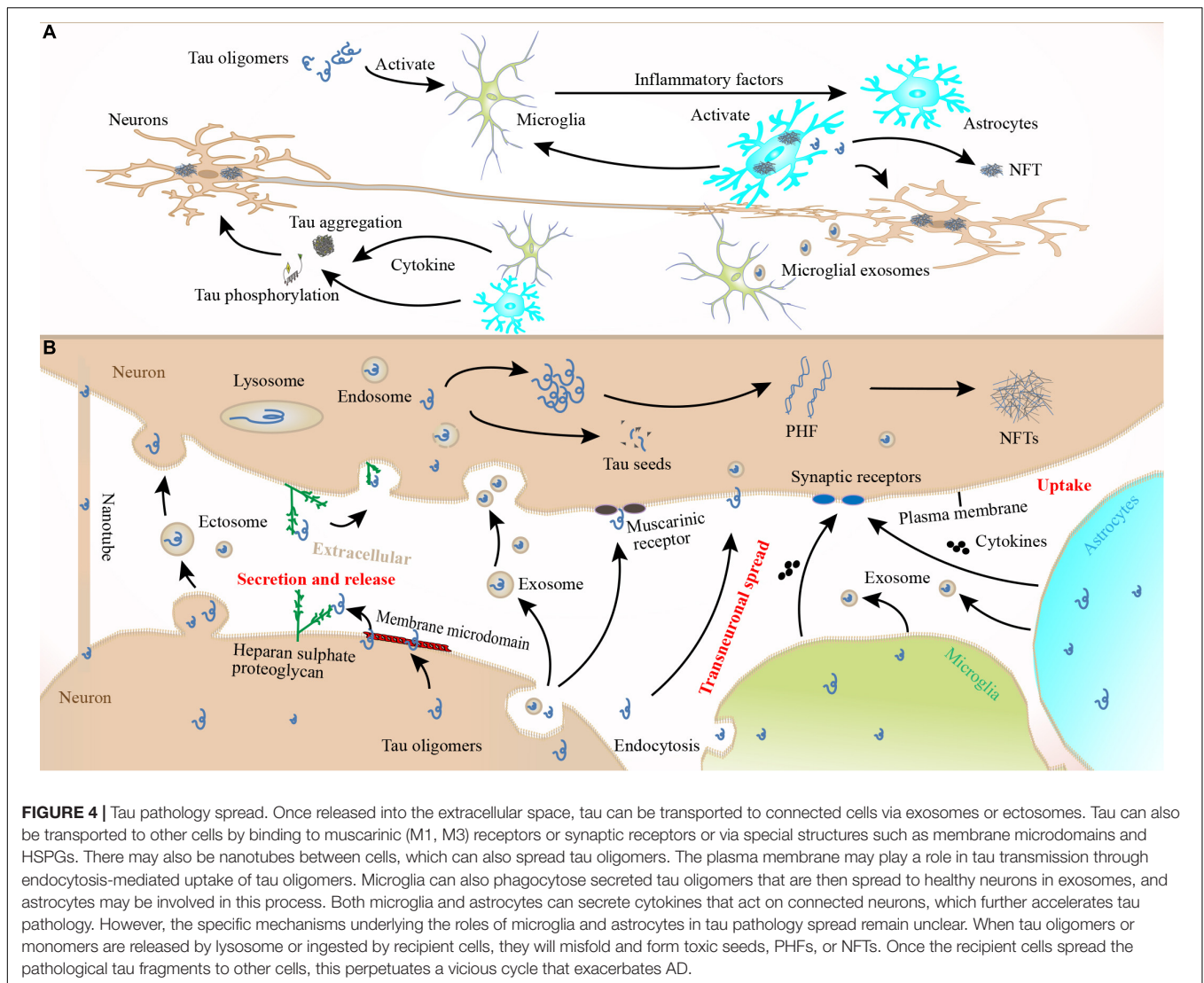
cell receptors (Gómez-Ramos et al., 2008; Díaz-Hernández et al., 2010; **Figure 3**). This toxic effect may result in cell death and the subsequent detection of tau in the CSF of patients with disorders such as AD (Iqbal et al., 2005; Yamada et al., 2011). Additionally, in affected regions such as the hippocampus, there is an inverse relationship between the numbers of surviving cells and extracellular tangles (Cras et al., 1995; Fukutani et al., 1995). This suggests that degenerating neurons containing fibrillar lesions might release NFT contents into the extracellular environment (Goedert, 1999). Meanwhile, several *in vitro* and *in vivo* studies reported that stimulation of neuronal activity can regulate the physiological secretion of endogenous tau by cortical neurons and enhance the release of pathological tau, a process that is Ca<sup>2+</sup>-dependent and modulated by phosphorylation (Sokolow et al., 2015; Fát et al., 2016; Wang et al., 2017). AMPA receptor stimulation promotes



tau release through a  $\text{Ca}^{2+}$ -dependent mechanism and the exocytosis of presynaptic vesicles. AMPA receptor stimulation generates action potentials that increase presynaptic  $\text{Ca}^{2+}$  concentrations, evoking vesicle release (Schmitz et al., 2009), and this plays a role in  $\text{Ca}^{2+}$ -mediated regulation of neuronal tau release (Pooler et al., 2013). The relationship between neuronal activity and tau release appears to be bidirectional; both extracellular tau and A $\beta$  perpetuate further neuronal tau release through feedback mechanisms (Bright et al., 2015). These results indicate that tau release partially occurs in a neuronal excitability-dependent manner in response to regional changes in the AD brain.

Tau can also be detected in the extracellular space before neurodegeneration, indicating that it can be released through mechanisms other than cell death (Yamada et al., 2011; Barten et al., 2012). Preliminary results demonstrated that tau can be released into the extracellular space via membrane vesicles in the absence of cell death (Simón et al., 2012). Tau secretion likely transpires via the unconventional vesicular- or non-vesicular-mediated secretory pathway since tau lacks an apparent endoplasmic reticulum-targeting sequence, which is necessary in the conventional secretory pathway (Yamada, 2017). Vesicle-mediated secretion might serve as a mechanism to regulate (proteostasis) cellular tau concentrations, maintaining them below a certain threshold level (Simón et al., 2012). Tau can also be transported through membrane vesicles

after lysosomal degradation (García-Sierra et al., 2008). Four different mechanisms have been proposed for the unconventional secretion of soluble, cytoplasmic tau: (1) non-vesicular direct translocation from the cytoplasm across the plasma membrane, (2) release via secretory lysosomes and release their contents into the extracellular space, (3) microvesicle shedding from the plasma membrane, and (4) vesicle-mediated exosome release (Nickel and Rabouille, 2009; Saman et al., 2012). In the last two scenarios, tau is surrounded by a membrane when it is released into the extracellular space (Nickel and Rabouille, 2009; Chai et al., 2012); **Figure 3**. It has also been proposed that tau may be released from cells through an exosome-independent pathway that requires heat shock cognate 70, its co-chaperone DnaJ (Hsp40), and synaptosomal-associated protein 23 (Fontaine et al., 2016). Additionally, tau secretion reportedly occurs through membrane vesicles when tau is overexpressed (Simón et al., 2012). Similarly, tau can be secreted in an exosome-dependent manner by Neuro2a cells overexpressing tau (Wang et al., 2017), as well as by microglia (Asai et al., 2015). Another vesicular-mediated mechanism involves large extracellular vesicles called exosomes that are directly shed from cells by plasma membrane budding (Théry et al., 2009; **Figure 3**). The third mechanism proposed to mediate tau release and spreading involves formation of thin membranous bridges called tunneling nanotubes (TNTs; Rustom et al., 2004). Moreover, cell depolarization was shown to induce



the release of a 20-kDa tau fragment from AD synapses (Sokolow et al., 2015). These four mechanisms appear to be temperature dependent and are likely to be less efficient at low temperatures (Nickel and Rabouille, 2009).

Different forms of tau protein might be secreted through different mechanisms or vectors. Endogenous tau released from primary cortical rat neurons under basal conditions is predominantly full length (Pooler et al., 2013), but truncated species have also been identified (Dujardin et al., 2014a). Tau fragments lacking the proline-rich region are either not secreted or are secreted in a manner different from that of the full-length molecule (Pérez et al., 2016). Monomeric and aggregated tau have been detected in CSF, suggesting that they may be released following axonal degeneration and neuronal death (Hampel et al., 2004). However, other studies found that unstimulated human and rodent neurons only secrete C-terminally truncated forms of endogenous tau (Kanmert et al., 2015). Cell culture studies revealed that tau is released via the unconventional secretory pathway and that tau mutations

influence the secretion rate, with 4R tau isoforms less abundant than 3R isoforms (Karch et al., 2012). Moreover, exogenously expressed hyperphosphorylated tau secreted by non-neuronal cells is cleaved at its C-terminus (Plouffe et al., 2012; Croft et al., 2017). For instance, tau cleavage at D421 can increase the secretion rate (Katsinelos et al., 2018). Aberrantly phosphorylated tau is secreted more efficiently than non-phosphorylated tau, at least in cultured cell lines (Plouffe et al., 2012; Katsinelos et al., 2018), possibly because abnormal phosphorylation impairs tau's ability to interact with its partners, therefore altering the protein's normal physiological properties (Yan et al., 2020). Meanwhile, endogenous tau is reportedly released either free and in a full-length, dephosphorylated form (Pooler et al., 2013) or as N-terminally truncated fragments (Bright et al., 2015; Kanmert et al., 2015). A small subset of tau released under these conditions is inside plasma membrane-derived vesicles called ectosomes (Dujardin et al., 2014a). Tau is also released from cells in association with exosomes, particularly when it is exogenously expressed or in a highly phosphorylated and misfolded state



(Plouffe et al., 2012; Saman et al., 2012). One study suggested that tau hyperphosphorylation in AD may induce a vicious circle that amplifies its secretion (Plouffe et al., 2012). In this case, tau hyperphosphorylation would enhance its secretion, which would subsequently increase the level of dephosphorylated tau in the extracellular space. Dephosphorylated extracellular tau would then induce an increase in intracellular  $\text{Ca}^{2+}$  concentrations, which has been linked with elevated tau hyperphosphorylation (Díaz-Hernández et al., 2010). This vicious circle then promotes tau pathology propagation in the brain and CSF accumulation (Hempel et al., 2010; Plouffe et al., 2012). Golgi dynamics were also proposed to modulate tau secretion from both HeLa cells and primary cortical neurons (Mohamed et al., 2017). Tau cleavage and hyperphosphorylation increase its secretion from HeLa cells. Mitochondrial damage might reduce tau secretion (Shafiei et al., 2017), whereas impaired lysosomal function may increase it (Mohamed et al., 2014). Pathological tau in animal models appears to be more localized to synapses compared to non-pathological tau (Sahara et al., 2014), and synaptosomes isolated from human AD brains were shown to contain more phosphorylated and aggregated tau than those isolated from healthy controls (Tai et al., 2014). In summary, different tau species and isoforms including mutated (Hempel et al., 2004), hyperphosphorylated (Plouffe et al., 2012; Bright et al., 2015), and truncated forms of tau appear to be released via distinct mechanisms (Plouffe et al., 2012).

## THE SPREAD OF PATHOLOGICAL TAU PROTEIN

Although the specific routes and mechanisms underlying the spread of pathological tau remain unclear (Braak and Braak, 1991), the propagation of cytosolic tau to connected neurons consists of at least four phases. First, tau must be secreted or released from donor neurons; second, it must undergo aggregation before or after being released; third, tau must be taken up into recipient neurons; and fourth, tau aggregation must be induced in recipient cells (Kanmert et al., 2015). After the release and secretion of tau to the extracellular space following cell death (Simón et al., 2012), stimulation of neuronal activity (Pooler et al., 2013), or other mechanisms (e.g., associated with vesicles, secretory lysosomes, or microvesicle shedding from the plasma membrane) (Nickel and Rabouille, 2009; Saman et al., 2012), pathological tau oligomers, monomers, or aggregates must enter other cells via endocytosis. Subsequently, pathological tau seeds might be degraded, resecreted, or mediate the misfolding of wild-type tau molecules in recipient cells (Wu et al., 2013). Recipient cells appear to favor the uptake of short, low-molecular-weight, tau fibrils over monomers and larger fibrils (Wu et al., 2013). Consequently, the potencies of various tau aggregate species on cellular propagation may be different (Frost et al., 2009). For example, tau uptake is closely related to both the size and conformation of tau aggregates (Kanmert et al., 2015). Intracellular tau accumulation is dependent on the isoform composition of the extracellular tau oligomers (Swanson et al., 2017). However, tau aggregate

uptake is not neuron specific; cell-to-cell transfer also occurs between glial cells and neurons. Tau aggregates can transfer between connected cells, induce templated misfolding, and be internalized from the extracellular space by a neighboring cell, which facilitates tauopathy propagation across different brain regions in a prion-like fashion (Jucker and Walker, 2013). Tau oligomers can be internalized by dynamin-dependent bulk endocytosis and are then transported through the endolysosomal pathway in recipient cells (Wu et al., 2013). One study demonstrated that neuronal uptake of  $\alpha$ -synuclein and tau aggregate seeds can occur through macropinocytosis, a form of fluid-phase bulk endocytosis that represents the most likely mechanism for tau uptake. This process begins when aggregated proteins bind to HSPGs, a family of core proteins with cell-surface glycosaminoglycan polysaccharides. Interestingly, the internalization process is only initiated by aggregated species, not by monomeric tau (Holmes et al., 2013). Another study also suggested that HSPGs can mediate exosome internalization (Christianson et al., 2013; Fuster-Matanzo et al., 2018). Before internalization via micropinocytosis, tau binds to plasma membrane HSPGs, which promotes membrane rearrangement before endocytosis (Holmes et al., 2013). Tau binding to HSPGs seems to be essential for internalization, and the 6-O-sulfation pattern on heparan sulfate sidechains is an important determinant for tau binding (Rauch et al., 2018; **Figure 4**). However, heparan-like glycosaminoglycan (GAG) mimetics can hide tau's HSPG binding site, which reduces cell-surface tau oligomer binding, uptake, and seeding (Holmes et al., 2013). HSPG-dependent macropinocytosis is instigated by small protein aggregates, and tau trimers were shown to be the smallest size able to initiate this mechanism (Mirbaha et al., 2015; Rauch et al., 2018). The results of a recent study also supported the hypothesis that different tau species could be internalized through different cellular mechanisms (Evans et al., 2018). For example, tau monomers and small oligomers are preferentially taken up by macropinocytosis, while dynamin-dependent endocytosis is the preferred route for larger aggregates. HSPGs and macropinocytosis may also play a role in the uptake of whole exosomes (Christianson et al., 2013), although exosomes are internalized because of this pathway's non-specificity. In short, endocytosis and/or pinocytosis might be favored over direct fusion to the plasma membrane as an exosome internalization route (Tian T. et al., 2013; Polanco et al., 2018). During this process, vesicles can be endocytosed by neighboring cells, which might be involved in the propagation of misfolded or aggregated tau proteins in different neurodegenerative disorders (Goedert et al., 2010; Guo and Lee, 2011). Meanwhile, micropinocytosis might be critical for pathological tau uptake both *in vitro* and *in vivo* (Holmes et al., 2013; Brunello et al., 2016), as well as the uptake of other pathologically misfolded proteins including  $\alpha$ -synuclein, TDP-43 (Zeineddine et al., 2015), and PrP (Hooper, 2011). Tau aggregates are reportedly internalized into primary neurons, where they are trafficked anterogradely and retrogradely along axons, and then spread to connected cells (Takeda et al., 2015; Wu et al., 2016; Wang et al., 2017), thereby propagating tau pathology (Calafate et al., 2015; Wu et al., 2016;

Nobuhara et al., 2017). They can also seed the aggregation of native monomers, thereby initiating more aggregates that are then released and spread to neighboring cells (Kundel et al., 2018; Brunello et al., 2020; **Figure 4**). Recent reports found that inoculation of preformed tau fibrils into tau transgenic mice quickly induced an AD-like NFT pathology in connected brain regions (Clavaguera et al., 2013; Ahmed et al., 2014; Boluda et al., 2015). Moreover, misfolded tau proteins spread through anatomically connected neurons, presumably via trans-synaptic tau aggregate transmission (Harris et al., 2012; Liu et al., 2012). However, preformed tau aggregates can also spread by means other than synaptic connections, suggesting the existence of alternative (non-synaptic) propagation pathways (de Calignon et al., 2012; Peeraer et al., 2015).

How neurons release tau that is then transmitted to recipient neurons to instigate tau propagation remains unclear. Although synaptic connections can facilitate tau's transcellular spread, other cellular uptake routes cannot be excluded. Once internalized, tau can be located in both early and late endosomes, which links these tau species to lysosomal vesicles in a retrograde axonal pathway and provides further evidence for a transsynaptic route of transmission (Frost et al., 2009; Wu et al., 2013). Extracellular vesicles play a major role in cellular communication and the transport of pathogenic proteins related to AD (Vingtdeux et al., 2012). Notably, exosomal tau protein levels are elevated in prodromal AD (Fiandaca et al., 2015; Gibbons et al., 2019). Microglia also contribute to tauopathy progression via exosome secretion, such that microglia depletion and inhibition of exosome synthesis can dramatically suppress tau propagation *in vitro* and *in vivo* (Asai et al., 2015). Additionally, astrocytes can internalize both fibrillar and monomeric tau, implying that these cells are also be involved in tau pathology spread (Martini-Stoica et al., 2018; Perea et al., 2019). However, one study found that oligomers and short fibrils that bind to the membrane can be internalized by neuronal cells via a receptor-independent mechanism, while tau monomers, long fibrils, and long filaments cannot (Wu et al., 2013). Thus, these structures (exosomes, ectosomes, or TNTs) mediate neuron-to-neuron transfer of pathological tau protein assemblages, which is considered a fast manner of tau spread that is prion like (Abounit et al., 2016; Tardivel et al., 2016); **Figure 4**). Tau protein modifications can also affect the spread of tau pathology. Tau hyperphosphorylation can enhance spread, while partial dephosphorylation slows it (Alonso et al., 1996). This observation indicates that tau hyperphosphorylation may be a potential target to prevent tau pathology progression in AD and other tauopathies (Hu et al., 2016). Tau fibrils can propagate by incorporating unphosphorylated tau monomers that undergo conformational changes and are then hyperphosphorylated (Goedert et al., 2017). The extracellular domain of the amyloid precursor protein might be involved in tau fibril uptake into cells (Takahashi et al., 2015). It is well known that increasing A $\beta$ 42 oligomerization can activate protein kinases (including GSK-3 $\beta$ ) that phosphorylate tau (Guo et al., 2013). A $\beta$  aggregation can promote tau hyperphosphorylation, suggesting that A $\beta$  might accelerate the spread of tau pathology (Pérez et al., 2018).

## TOXICITY ASSOCIATED WITH THE SPREAD OF TAU PATHOLOGY

Tau is normally enriched on MTs within axons. In tauopathies, tau is hyperphosphorylated and accumulates in the somatodendritic compartment of brain cells, which is one of the pathological hallmarks of AD (Lee et al., 2001). Tau aggregates into insoluble filaments, forming NFTs (Morris et al., 2011) that are associated with cognitive deficits (Braak and Braak, 1991; Bierer et al., 1995; Plouffe et al., 2012). Tau phosphorylation, mislocalization, and conformational changes can alter Ca<sup>2+</sup> homeostasis, induce dendritic spine loss, impair organelle trafficking (particularly mitochondria), and lead to cell death (Dixit et al., 2008; Zempel et al., 2010; Li et al., 2011; Spire-Jones et al., 2011). These phenotypes represent a pretangle stage, which is widely recognized as an early event in the pathological process of AD (Eckermann et al., 2007; Braak and Del Tredici, 2012). Three hypotheses have been proposed to underlie tau-mediated toxicity. First, insoluble NFTs may be toxic and lead to neuron death and cognitive dysfunction in AD; second, soluble species of misfolded, hyperphosphorylated tau may become toxic when they accumulate in inappropriate cellular compartments, whereas NFTs exert a protective effect by serving as a sink for these toxic species; and in the third view, soluble forms of pathological tau and insoluble NFTs are both toxic to cells in various ways and time scales (Kopeikina et al., 2012). We favor the third view. Neuronal transport disruption is considered an important form of tau toxicity, which is an early phenomenon and underlying cause of neurodegenerative conditions including AD (Lin and Beal, 2006; Morfini et al., 2009; Wang and Schwarz, 2009; Querfurth and LaFerla, 2010; Kopeikina et al., 2012). Recent studies have found that soluble tau species are related to synaptic or neuronal dysfunction (Berger et al., 2007; Polydoro et al., 2009; Hoover et al., 2010; Sydow et al., 2011), with results indicating that tau oligomers—but not monomers or fibrils—act as aggregation seeds in the brains of wild-type mice, leading to mitochondrial dysfunction, synaptic deficits, and memory impairment (Usenovic et al., 2015). Based on these results, the interneuronal spread of these soluble tau species might be involved in the spread of AD pathology through the brain (Braak and Del Tredici, 2012; de Calignon et al., 2012; Clavaguera et al., 2013). Furthermore, neurons can endocytose low-molecular-weight misfolded tau species (but not monomeric tau) that are transported anterogradely and retrogradely, resulting in endogenous tau pathology *in vivo*. However, they cannot endocytose fibrillar tau or brain-derived filamentous tau (Wu et al., 2013). This evidence strongly indicates that tau toxicity may be mediated by the cell-to-cell spread of trimeric and larger oligomeric forms in certain brain regions by endocytosis (Tian H. et al., 2013). Extracellular tau is neurotoxic (Gómez-Ramos et al., 2006) and contributes to the spread of AD pathology. When the extracellular level of free tau exceeds that inside the cell and reaches a critical concentration (Reynolds et al., 2005), tau protein will self-aggregate and induce extracellular toxicity. Paired-helical filament (PHF)-tau is less toxic than free tau. When tau interacts with muscarinic receptors, intracellular Ca<sup>2+</sup>

levels increase due to  $\text{Ca}^{2+}$  release from intracellular stores (Gómez-Ramos et al., 2006). Tau seeds can also cause toxicity and cell death via  $\text{Ca}^{2+}$  dysregulation (Querfurth and LaFerla, 2010; Tian H. et al., 2013; Hallinan et al., 2019); altered intracellular  $\text{Ca}^{2+}$  homeostasis results in tau phosphorylation, which is related to tau pathology progression in AD (Delacourte et al., 1999). Tau phosphorylation will increase tau detachment from MTs (Avila et al., 2004), increasing free tau levels. Free tau is then released during neurodegeneration or following cell death and binds to muscarinic receptors on surrounding cells, thereby inducing muscarinic toxicity and aggravating tau toxicity and transmission (Gómez-Ramos et al., 2008). In addition, different tau forms or isomers secreted into the extracellular space via different mechanisms may play varied roles in AD pathogenesis. Certain forms of tau released through cell death or neuroactive stimulation may be non-toxic, while misfolded tau fragments or seeds (induced by  $\text{A}\beta$ , kinases, and hydrolases) exert toxic effects on the extracellular space. Secreted vesicles containing tau protein inhibit tau binding to muscarinic receptors, thus reducing neurotoxicity.

The neurodegenerative consequences of tau hyperphosphorylation include axonal transport impairment (Ittner et al., 2008), tau relocation to the somatodendritic compartment, and synaptic loss (Di et al., 2016). Synaptic dysfunction can occur both presynaptically, where it can interfere with the transport of phosphorylated tau via synaptic vesicles (Zhou et al., 2017), and postsynaptically via the downregulation of AMPA receptors (Hoover et al., 2010). In the context of the prion hypothesis, tau assemblies that enter the cytoplasm can seed native monomer aggregation, and these species can be released and spread to neighboring cells (Clavaguera et al., 2013; Kundel et al., 2018). Under pathological conditions, aberrant posttranslational modifications such as hyperphosphorylation, truncation, deamidation, and others (Avila et al., 2004) can induce tau detachment from MTs and promote their accumulation in a free form. When neurons degenerate and die, this free tau enters the extracellular space, where it is free to diffuse in every direction (Guo and Lee, 2011; Hu et al., 2016). This is in line with the observation that neuron loss is progressive within brain areas affected by degeneration, and tau species can seed misfolding in human brains without tangles (DeVos et al., 2018). This implies that tau seeds are released from intact neurons prior to neuronal death (Pickett et al., 2017). Another possible explanation of tau pathology patterns in the brains of AD patients is that extracellular NFTs or other substances released by degenerating neurons accumulate in the extracellular space and damage nearby cells (Avila, 2006). These toxic compounds can act like extracellular  $\text{A}\beta$  peptides (Gómez-Ramos et al., 2006). Thus, both soluble tau species and insoluble NFTs may contribute to the spread of tau toxicity.

## CONCLUSION AND PROSPECTS

The mechanism involved in the transcellular propagation of tau in neurodegeneration is still unclear. Studies on the molecular mechanisms underlying the release, propagation, and uptake

of tau are needed; improving the ability to detect secreted tau species is also important. Future research should focus on reducing the secretion and generation of extracellular tau in a soluble or aggregated form and inhibiting cell uptake. However, tau aggregate species are diverse, and it is currently unclear if certain species prefer certain secretion pathways. It will be important to clarify whether the physiologic secretion of non-pathological tau from neurons occurs via the same or overlapping mechanisms as those for the pathologic forms. Moreover, both forms of tau secretion are connected to neuronal activity, so blocking synapse-mediated tau propagation and boosting the clearance of tau aggregates that are internalized at the synapse are equally important (Calafate et al., 2015). More attention should also be given to glial cells and the glymphatic system, which might play a role in clearing and propagating pathological protein aggregates. Inhibition of donor cell release and recipient cell uptake are also novel therapeutic directions worthy of consideration. For example, it might be possible to block exosome secretion pathways and apply tau antibodies that can act on pathological tau fragments in the extracellular space and inhibit tau aggregation on membranous structures. Recent studies have shown that anti-tau antibodies can reduce tau hyperphosphorylation and aggregation in the transgenic mouse brain (Yanamandra et al., 2013; Sankaranarayanan et al., 2015). In addition, preventing tau from binding to HSPGs precludes recombinant tau fibrils from inducing intracellular aggregation and blocks transcellular aggregate propagation. *In vivo*, the heparin mimetic F6 prevents neuronal uptake of tau fibrils injected stereotactically (Holmes et al., 2013). Moreover, microglia disseminate tau via exosome secretion, and hampering exosome synthesis significantly reduces tau propagation *in vitro* and *in vivo*, which implies that exosomes and microglia contribute to tauopathy progression. It also suggests that targeting the exosome secretion pathway could be therapeutically useful. Depleting microglia dramatically suppressed tau propagation and reduced excitability in the dentate gyrus in a mouse model of AD (Asai et al., 2015). In summary, tau release mechanics can be explored to develop new treatments for AD and other tauopathies.

## AUTHOR CONTRIBUTIONS

HZ wrote the manuscript. YC and LM assisted in the manuscript writing. YW and HL assisted in ideas and modification of the manuscript. All authors contributed to the article and approved the submitted version.

## FUNDING

This review was funded by the National Natural Science Foundation of China (No. 81873350), the National Science and Technology Major Project for “Essential new drug research and development” (No. 2019ZX09301114), and the National Natural Science Foundation of China (No. 81904194).

## REFERENCES

- Aboutin, S., Wu, J., Duff, K., Victoria, G., and Zurzolo, C. (2016). Tunneling nanotubes: a possible highway in the spreading of tau and other prion-like proteins in neurodegenerative diseases. *Prion* 10, 344–351. doi: 10.1080/19336896.2016.1223003
- Ahmed, Z., Cooper, J., Murray, T., Garn, K., McNaughton, E., Clarke, H., et al. (2014). 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.* 127, 667–683. doi: 10.1007/s00401-014-1254-6
- Al-Bassam, J., Ozer, R., Safer, D., Halpain, S., and Milligan, R. (2002). MAP2 and tau bind longitudinally along the outer ridges of microtubule protofilaments. *J. Cell Biol.* 157, 1187–1196. doi: 10.1083/jcb.200201048
- Alonso, A., Grundke-Iqbal, I., and Iqbal, K. (1996). Alzheimer's disease hyperphosphorylated tau sequesters normal tau into tangles of filaments and disassembles microtubules. *Nat. Med.* 2, 783–787. doi: 10.1038/nm0796-783
- Alonso, A., Grundke-Iqbal, I., Barra, H., and Iqbal, K. (1997). Abnormal phosphorylation of tau and the mechanism of Alzheimer neurofibrillary degeneration: sequestration of microtubule-associated proteins 1 and 2 and the disassembly of microtubules by the abnormal tau. *Proc. Natl. Acad. Sci. U. S. A.* 94, 298–303. doi: 10.1073/pnas.94.1.298
- Asai, H., Ikezu, S., Tsunoda, S., Medalla, M., Luebke, J., Haydar, T., et al. (2015). Depletion of microglia and inhibition of exosome synthesis halt tau propagation. *Nat. Neurosci.* 18, 1584–1593. doi: 10.1038/nn.4132
- Augustinack, J., Schneider, A., Mandelkow, E., and Hyman, B. (2002). Specific tau phosphorylation sites correlate with severity of neuronal cytopathology in Alzheimer's disease. *Acta Neuropathol.* 103, 26–35. doi: 10.1007/s004010100423
- Avila, J. (2006). Tau phosphorylation and aggregation in Alzheimer's disease pathology. *FEBS Lett.* 580, 2922–2927. doi: 10.1016/j.febslet.2006.02.067
- Avila, J., Lucas, J., Perez, M., and Hernandez, F. (2004). Role of tau protein in both physiological and pathological conditions. *Physiol. Rev.* 84, 361–384. doi: 10.1016/j.tcb.2005.02.001
- Barten, D., Fanara, P., Andorfer, C., Hoque, N., Wong, P., Husted, K., et al. (2012). Hyperdynamic microtubules, cognitive deficits, and pathology are improved in tau transgenic mice with low doses of the microtubule-stabilizing agent BMS-241027. *J. Neurosci.* 32, 7137–7145. doi: 10.1523/jneurosci.0188-12.2012
- Barthélemy, N., Gabelle, A., Hirtz, C., Fenaile, F., Sergeant, N., Schraen-Maschke, S., et al. (2016). Differential Mass Spectrometry Profiles of Tau Protein in the Cerebrospinal Fluid of Patients with Alzheimer's Disease, Progressive Supranuclear Palsy, and Dementia with Lewy Bodies. *J. Alzheimers Dis.* 51, 1033–1043. doi: 10.3233/jad-150962
- Barton, A., Harrison, P., Najlerahim, A., Heffernan, J., McDonald, B., Robinson, J., et al. (1990). Increased tau messenger RNA in Alzheimer's disease hippocampus. *Am. J. Pathol.* 137, 497–502.
- Baudry, M., and Bi, X. (2016). Calpain-1 and Calpain-2: the Yin and Yang of Synaptic Plasticity and Neurodegeneration. *Trends Neurosci.* 39, 235–245. doi: 10.1016/j.tins.2016.01.007
- Berger, Z., Roder, H., Hanna, A., Carlson, A., Rangachari, V., Yue, M., et al. (2007). Accumulation of pathological tau species and memory loss in a conditional model of tauopathy. *J. Neurosci.* 27, 3650–3662. doi: 10.1523/jneurosci.0587-07.2007
- Bierer, L., Hof, P., Purohit, D., Carlin, L., Schmeidler, J., Davis, K., et al. (1995). Neocortical neurofibrillary tangles correlate with dementia severity in Alzheimer's disease. *Arch. Neurol.* 52, 81–88. doi: 10.1001/archneur.1995.00540250089017
- Boluda, S., Iba, M., Zhang, B., Raible, K., Lee, V., and Trojanowski, J. (2015). Differential induction and spread of tau pathology in young PS19 tau transgenic mice following intracerebral injections of pathological tau from Alzheimer's disease or corticobasal degeneration brains. *Acta Neuropathol.* 129, 221–237. doi: 10.1007/s00401-014-1373-0
- Bondulich, M., Guo, T., Meehan, C., Manion, J., Rodriguez Martin, T., Mitchell, J., et al. (2016). Tauopathy induced by low level expression of a human brain-derived tau fragment in mice is rescued by phenylbutyrate. *Brain* 139, 2290–2306. doi: 10.1093/brain/aww137
- Braak, H., and Braak, E. (1991). Neuropathological staging of Alzheimer-related changes. *Acta Neuropathol.* 82, 239–259. doi: 10.1007/bf00308809
- Braak, H., and Del Tredici, K. (2012). Alzheimer's disease: pathogenesis and prevention. *Alzheimers Dement.* 8, 227–233. doi: 10.1016/j.jalz.2012.01.011
- Brandt, R., Léger, J., and Lee, G. (1995). Interaction of tau with the neural plasma membrane mediated by tau's amino-terminal projection domain. *J. Cell Biol.* 131, 1327–1340. doi: 10.1083/jcb.131.5.1327
- Bright, J., Hussain, S., Dang, V., Wright, S., Cooper, B., Byun, T., et al. (2015). Human secreted tau increases amyloid-beta production. *Neurobiol. Aging* 36, 693–709. doi: 10.1016/j.neurobiolaging.2014.09.007
- Brunello, C., Merezko, M., Uronen, R., and Huttunen, H. (2020). Mechanisms of secretion and spreading of pathological tau protein. *Cell. Mol. Life Sci.* 77, 1721–1744. doi: 10.1007/s00018-019-03349-1
- Brunello, C., Yan, X., and Huttunen, H. (2016). Internalized Tau sensitizes cells to stress by promoting formation and stability of stress granules. *Sci. Rep.* 6:30498. doi: 10.1038/srep30498
- Calafate, S., Buist, A., Miskiewicz, K., Vijayan, V., Daneels, G., De Strooper, B., et al. (2015). Synaptic Contacts Enhance Cell-to-Cell Tau Pathology Propagation. *Cell Rep.* 11, 1176–1183. doi: 10.1016/j.celrep.2015.04.043
- Chai, X., Dage, J., and Citron, M. (2012). Constitutive secretion of tau protein by an unconventional mechanism. *Neurobiol. Dis.* 48, 356–366. doi: 10.1016/j.nbd.2012.05.021
- Chen, X., and Jiang, H. (2019). Tau as a potential therapeutic target for ischemic stroke. *Aging* 11, 12827–12843. doi: 10.18632/aging.102547
- Chesser, A., Pritchard, S., and Johnson, G. (2013). Tau clearance mechanisms and their possible role in the pathogenesis of Alzheimer disease. *Front. Neurol.* 4:122. doi: 10.3389/fneur.2013.00122
- Christianson, H., Svensson, K., Van Kuppevelt, T., Li, J., and Belting, M. (2013). Cancer cell exosomes depend on cell-surface heparan sulfate proteoglycans for their internalization and functional activity. *Proc. Natl. Acad. Sci. U. S. A.* 110, 17380–17385. doi: 10.1073/pnas.1304266110
- Chung, C., Song, Y., Kim, I., Yoon, W., Ryu, B., Jo, D., et al. (2001). Proapoptotic effects of tau cleavage product generated by caspase-3. *Neurobiol. Dis.* 8, 162–172. doi: 10.1006/nbdi.2000.0335
- Clavaguera, F., Akatsu, H., Fraser, G., Crowther, R., Frank, S., Hench, J., et al. (2015). Brain homogenates from human tauopathies induce tau inclusions in mouse brain. *Proc. Natl. Acad. Sci. U. S. A.* 110, 9535–9540. doi: 10.1073/pnas.1301175110
- Clavaguera, F., Bolmont, T., Crowther, R., Abramowski, D., Frank, S., Probst, A., et al. (2009). Transmission and spreading of tauopathy in transgenic mouse brain. *Nat. Cell Biol.* 11, 909–913. doi: 10.1038/ncb1901
- Clavaguera, F., Hench, J., Goedert, M., and Tolnay, M. (2015). Invited review: prion-like transmission and spreading of tau pathology. *Neuropathol. Appl. Neurobiol.* 41, 47–58. doi: 10.1111/nan.12197
- Cohen, T., Constance, B., Hwang, A., James, M., and Yuan, C. (2016). Intrinsic Tau Acetylation Is Coupled to Auto-Proteolytic Tau Fragmentation. *PLoS One* 11:e0158470. doi: 10.1371/journal.pone.0158470
- Cohen, T., Friedmann, D., Hwang, A., Marmorstein, R., and Lee, V. (2013). The microtubule-associated tau protein has intrinsic acetyltransferase activity. *Nat. Struct. Mol. Biol.* 20, 756–762. doi: 10.1038/nsmb.2555
- Cook, C., Carlomagno, Y., Gendron, T., Dunmore, J., Scheffel, K., Stetler, C., et al. (2014). Acetylation of the KXGS motifs in tau is a critical determinant in modulation of tau aggregation and clearance. *Hum. Mol. Genet.* 23, 104–116. doi: 10.1093/hmg/ddt402
- Corsetti, V., Amadoro, G., Gentile, A., Capsoni, S., Ciotti, M., Cencioni, M., et al. (2008). Identification of a caspase-derived N-terminal tau fragment in cellular and animal Alzheimer's disease models. *Mol. Cell. Neurosci.* 38, 381–392. doi: 10.1016/j.mcn.2008.03.011
- Cras, P., Smith, M., Richey, P., Siedlak, S., Mulvihill, P., and Perry, G. (1995). Extracellular neurofibrillary tangles reflect neuronal loss and provide further evidence of extensive protein cross-linking in Alzheimer disease. *Acta Neuropathol.* 89, 291–295. doi: 10.1007/bf00309621
- Croft, C., Wade, M., Kurbatskaya, K., Mastrandreas, P., Hughes, M., Phillips, E., et al. (2017). Membrane association and release of wild-type and pathological tau from organotypic brain slice cultures. *Cell Death Dis.* 8:e2671. doi: 10.1038/cddis.2017.97
- de Calignon, A., Fox, L., Pitstick, R., Carlson, G., Bacskai, B., Spire-Jones, T., et al. (2010). Caspase activation precedes and leads to tangles. *Nature* 464, 1201–1204. doi: 10.1038/nature08890

- de Calignon, A., Polydoro, M., Suárez-Calvet, M., William, C., Adamowicz, D., Kopeikina, K., et al. (2012). Propagation of tau pathology in a model of early Alzheimer's disease. *Neuron* 73, 685–697. doi: 10.1016/j.neuron.2011.11.033
- Delacourte, A., David, J., Sergeant, N., Buée, L., Wattez, A., Vermersch, P., et al. (1999). The biochemical pathway of neurofibrillary degeneration in aging and Alzheimer's disease. *Neurology* 52, 1158–1165. doi: 10.1212/wnl.52.6.1158
- DeVos, S., Corjuc, B., Oakley, D., Nobuhara, C., Bannon, R., Chase, A., et al. (2018). Synaptic Tau Seeding Precedes Tau Pathology in Human Alzheimer's Disease Brain. *Front. Neurosci.* 12:267. doi: 10.3389/fnins.2018.00267
- Di, J., Cohen, L., Corbo, C., Phillips, G., El Idrissi, A., and Alonso, A. (2016). Abnormal tau induces cognitive impairment through two different mechanisms: synaptic dysfunction and neuronal loss. *Sci. Rep.* 6:20833. doi: 10.1038/srep20833
- Díaz-Hernández, M., Gómez-Ramos, A., Rubio, A., Gómez-Villafuertes, R., Naranjo, J., Miras-Portugal, M., et al. (2010). Tissue-nonspecific alkaline phosphatase promotes the neurotoxicity effect of extracellular tau. *J. Biol. Chem.* 285, 32539–32548. doi: 10.1074/jbc.M110.145003
- Dickey, C., Kamal, A., Lundgren, K., Klosak, N., Bailey, R., Dunmore, J., et al. (2007). The high-affinity HSP90-CHIP complex recognizes and selectively degrades phosphorylated tau client proteins. *J. Clin. Invest.* 117, 648–658. doi: 10.1172/jci29715
- Dixit, R., Ross, J., Goldman, Y., and Holzbaue, E. (2008). Differential regulation of dynein and kinesin motor proteins by tau. *Science* 319, 1086–1089. doi: 10.1126/science.1152993
- Dolan, P., and Johnson, G. (2010). A caspase cleaved form of tau is preferentially degraded through the autophagy pathway. *J. Biol. Chem.* 285, 21978–21987. doi: 10.1074/jbc.M110.110940
- Dujardin, S., Bégard, S., Caillierez, R., Lachaud, C., Delattre, L., Carrier, S., et al. (2014a). Ectosomes: a new mechanism for non-exosomal secretion of tau protein. *PLoS One* 9:e100760. doi: 10.1371/journal.pone.0100760
- Dujardin, S., Lécolle, K., Caillierez, R., Bégard, S., Zommer, N., Lachaud, C., et al. (2014b). Neuron-to-neuron wild-type Tau protein transfer through a trans-synaptic mechanism: relevance to sporadic tauopathies. *Acta Neuropathol. Commun.* 2:14. doi: 10.1186/2051-5960-2-14
- Eckermann, K., Mocuano, M., Khlistunova, I., Biernat, J., Nissen, A., Hofmann, A., et al. (2007). The beta-propensity of Tau determines aggregation and synaptic loss in inducible mouse models of tauopathy. *J. Biol. Chem.* 282, 31755–31765. doi: 10.1074/jbc.M705282200
- Evans, L., Wassmer, T., Fraser, G., Smith, J., Perkinson, M., Billinton, A., et al. (2018). Extracellular Monomeric and Aggregated Tau Efficiently Enter Human Neurons through Overlapping but Distinct Pathways. *Cell Rep.* 22, 3612–3624. doi: 10.1016/j.celrep.2018.03.021
- Fá, M., Puzzo, D., Piacentini, R., Staniszewski, A., Zhang, H., Baltrons, M., et al. (2016). Extracellular Tau Oligomers Produce An Immediate Impairment of LTP and Memory. *Sci. Rep.* 6:19393. doi: 10.1038/srep19393
- Fernández-Montoya, J., and Pérez, M. (2015). Cathepsin D in a murine model of frontotemporal dementia with Parkinsonism-linked to chromosome 17. *J. Alzheimers Dis.* 45, 1–14. doi: 10.3233/jad-140456
- Fiandaca, M., Kapogiannis, D., Mapstone, M., Boxer, A., Eitan, E., Schwartz, J., et al. (2015). Identification of preclinical Alzheimer's disease by a profile of pathogenic proteins in neurally derived blood exosomes: a case-control study. *Alzheimers Dement.* 11, 600–607.e1. doi: 10.1016/j.jalz.2014.06.008
- Fischer, D., Mukrasch, M., Biernat, J., Bibow, S., Blackledge, M., Griesinger, C., et al. (2009). Conformational changes specific for pseudophosphorylation at serine 262 selectively impair binding of tau to microtubules. *Biochemistry* 48, 10047–10055. doi: 10.1021/bi901090m
- Fontaine, S., Zheng, D., Sabbagh, J., Martin, M., Chaput, D., Darling, A., et al. (2016). DnaJ/Hsc70 chaperone complexes control the extracellular release of neurodegenerative-associated proteins. *EMBO J.* 35, 1537–1549. doi: 10.15252/emboj.201593489
- Frost, B., Jacks, R., and Diamond, M. (2009). Propagation of tau misfolding from the outside to the inside of a cell. *J. Biol. Chem.* 284, 12845–12852. doi: 10.1074/jbc.M808759200
- Fukutani, Y., Kobayashi, K., Nakamura, I., Watanabe, K., Isaki, K., and Cairns, N. (1995). Neurons, intracellular and extracellular neurofibrillary tangles in subdivisions of the hippocampal cortex in normal ageing and Alzheimer's disease. *Neurosci. Lett.* 200, 57–60. doi: 10.1016/0304-3940(95)12083-g
- Fuster-Matanzo, A., Hernández, F., and Ávila, J. (2018). Tau Spreading Mechanisms; Implications for Dysfunctional Tauopathies. *Int. J. Mol. Sci.* 19:645. doi: 10.3390/ijms19030645
- Gamblin, T., Chen, F., Zambrano, A., Abrahá, A., Lagalwar, S., Guillozet, A., et al. (2003). Caspase cleavage of tau: linking amyloid and neurofibrillary tangles in Alzheimer's disease. *Proc. Natl. Acad. Sci. U. S. A.* 100, 10032–10037. doi: 10.1073/pnas.1630428100
- García, M., and Cleveland, D. (2001). Going new places using an old MAP: tau, microtubules and human neurodegenerative disease. *Curr. Opin. Cell Biol.* 13, 41–48. doi: 10.1016/s0955-0674(00)00172-1
- García-Sierra, F., Mondragón-Rodríguez, S., and Basurto-Islas, G. (2008). Truncation of tau protein and its pathological significance in Alzheimer's disease. *J. Alzheimers Dis.* 14, 401–409. doi: 10.3233/jad-2008-14407
- Garg, S., Timm, T., Mandelkow, E., Mandelkow, E., and Wang, Y. (2011). Cleavage of Tau by calpain in Alzheimer's disease: the quest for the toxic 17 kD fragment. *Neurobiol. Aging* 32, 1–14. doi: 10.1016/j.neurobiolaging.2010.09.008
- Gibbons, G., Lee, V., and Trojanowski, J. (2019). Mechanisms of Cell-to-Cell Transmission of Pathological Tau: a Review. *JAMA Neurol.* 76, 101–108. doi: 10.1001/jamaneurol.2018.2505
- Goedert, M. (1999). Filamentous nerve cell inclusions in neurodegenerative diseases: tauopathies and alpha-synucleinopathies. *Philos. Trans. R. Soc. Lond. B Biol. Sci.* 354, 1101–1118. doi: 10.1098/rstb.1999.0466
- Goedert, M., and Spillantini, M. (2011). Pathogenesis of the tauopathies. *J. Mol. Neurosci.* 45, 425–431. doi: 10.1007/s12031-011-9593-4
- Goedert, M., Clavaguera, F., and Tolnay, M. (2010). The propagation of prion-like protein inclusions in neurodegenerative diseases. *Trends Neurosci.* 33, 317–325. doi: 10.1016/j.tins.2010.04.003
- Goedert, M., Eisenberg, D., and Crowther, R. (2017). Propagation of Tau Aggregates and Neurodegeneration. *Annu. Rev. Neurosci.* 40, 189–210. doi: 10.1146/annurev-neuro-072116-031153
- Goedert, M., Spillantini, M., Jakes, R., Rutherford, D., and Crowther, R. (1989). Multiple isoforms of human microtubule-associated protein tau: sequences and localization in neurofibrillary tangles of Alzheimer's disease. *Neuron* 3, 519–526. doi: 10.1016/0896-6273(89)90210-9
- Gómez-Ramos, A., Díaz-Hernández, M., Cuadros, R., Hernández, F., and Avila, J. (2006). Extracellular tau is toxic to neuronal cells. *FEBS Lett.* 580, 4842–4850. doi: 10.1016/j.febslet.2006.07.078
- Gómez-Ramos, A., Díaz-Hernández, M., Rubio, A., Miras-Portugal, M., and Avila, J. (2008). Extracellular tau promotes intracellular calcium increase through M1 and M3 muscarinic receptors in neuronal cells. *Mol. Cell. Neurosci.* 37, 673–681. doi: 10.1016/j.mcn.2007.12.010
- Guo, J., and Lee, V. (2011). Seeding of normal Tau by pathological Tau conformers drives pathogenesis of Alzheimer-like tangles. *J. Biol. Chem.* 286, 15317–15331. doi: 10.1074/jbc.M110.209296
- Guo, J., Covell, D., Daniels, J., Iba, M., Stieber, A., Zhang, B., et al. (2013). Distinct  $\alpha$ -synuclein strains differentially promote tau inclusions in neurons. *Cell* 154, 103–117. doi: 10.1016/j.cell.2013.05.057
- Guo, T., Noble, W., and Hanger, D. (2017). Roles of tau protein in health and disease. *Acta Neuropathol.* 133, 665–704. doi: 10.1007/s00401-017-1707-9
- Gustke, N., Trinczek, B., Biernat, J., Mandelkow, E., and Mandelkow, E. (1994). Domains of tau protein and interactions with microtubules. *Biochemistry* 33, 9511–9522. doi: 10.1021/bi00198a017
- Hallinan, G., Vargas-Caballero, M., West, J., and Deinhardt, K. (2019). Tau Misfolding Efficiently Propagates between Individual Intact Hippocampal Neurons. *J. Neurosci.* 39, 9623–9632. doi: 10.1523/jneurosci.1590-19.2019
- Hampel, H., Blennow, K., Shaw, L., Hoessler, Y., Zetterberg, H., and Trojanowski, J. (2010). Total and phosphorylated tau protein as biological markers of Alzheimer's disease. *Exp. Gerontol.* 45, 30–40. doi: 10.1016/j.exger.2009.10.010
- Hampel, H., Teipel, S., Fuchsberger, T., Andreasen, N., Wiltfang, J., Otto, M., et al. (2004). Value of CSF beta-amyloid1-42 and tau as predictors of Alzheimer's disease in patients with mild cognitive impairment. *Mol. Psychiatry* 9, 705–710. doi: 10.1038/sj.mp.4001473
- Hanger, D., Anderton, B., and Noble, W. (2009). Tau phosphorylation: the therapeutic challenge for neurodegenerative disease. *Trends Mol. Med.* 15, 112–119. doi: 10.1016/j.molmed.2009.01.003
- Hanger, D., Lau, D., Phillips, E., Bondulich, M., Guo, T., Woodward, B., et al. (2014). Intracellular and extracellular roles for tau in neurodegenerative disease. *J. Alzheimers Dis.* 40, S37–S45. doi: 10.3233/jad-132054

- Harris, J., Koyama, A., Maeda, S., Ho, K., Devidze, N., Dubal, D., et al. (2012). Human P301L-mutant tau expression in mouse entorhinal-hippocampal network causes tau aggregation and presynaptic pathology but no cognitive deficits. *PLoS One* 7:e45881. doi: 10.1371/journal.pone.0045881
- Holmes, B., DeVos, S., Kfoury, N., Li, M., Jacks, R., Yanamandra, K., et al. (2013). Heparan sulfate proteoglycans mediate internalization and propagation of specific proteopathic seeds. *Proc. Natl. Acad. Sci. U. S. A.* 110, E3138–E3147. doi: 10.1073/pnas.1301440110
- Hooper, N. (2011). Glypican-1 facilitates prion conversion in lipid rafts. *J. Neurochem.* 116, 721–725. doi: 10.1111/j.1471-4159.2010.06936.x
- Hoover, B., Reed, M., Su, J., Penrod, R., Kotilinek, L., Grant, M., et al. (2010). Tau mislocalization to dendritic spines mediates synaptic dysfunction independently of neurodegeneration. *Neuron* 68, 1067–1081. doi: 10.1016/j.neuron.2010.11.030
- Hu, W., Zhang, X., Tung, Y., Xie, S., Liu, F., and Iqbal, K. (2016). Hyperphosphorylation determines both the spread and the morphology of tau pathology. *Alzheimers Dement.* 12, 1066–1077. doi: 10.1016/j.jalz.2016.01.014
- Iba, M., Guo, J., McBride, J., Zhang, B., Trojanowski, J., and Lee, V. (2013). Synthetic tau fibrils mediate transmission of neurofibrillary tangles in a transgenic mouse model of Alzheimer's-like tauopathy. *J. Neurosci.* 33, 1024–1037. doi: 10.1523/jneurosci.2642-12.2013
- Ii, K., Ito, H., Kominami, E., and Hirano, A. (1993). Abnormal distribution of cathepsin proteinases and endogenous inhibitors (cystatins) in the hippocampus of patients with Alzheimer's disease, parkinsonism-dementia complex on Guam, and senile dementia and in the aged. *Virchows Arch. A Pathol. Anat. Histopathol.* 423, 185–194. doi: 10.1007/bf01614769
- Iqbal, K., Flory, M., Khatoun, S., Soininen, H., Pirttila, T., Lehtovirta, M., et al. (2005). Subgroups of Alzheimer's disease based on cerebrospinal fluid molecular markers. *Ann. Neurol.* 58, 748–757. doi: 10.1002/ana.20639
- Ittner, L., Fath, T., Ke, Y., Bi, M., Van Eersel, J., Li, K., et al. (2008). Parkinsonism and impaired axonal transport in a mouse model of frontotemporal dementia. *Proc. Natl. Acad. Sci. U. S. A.* 105, 15997–16002. doi: 10.1073/pnas.0808084105
- Jho, Y., Zhulina, E., Kim, M., and Pincus, P. (2010). Monte carlo simulations of tau proteins: effect of phosphorylation. *Biophys. J.* 99, 2387–2397. doi: 10.1016/j.bpj.2010.06.056
- Johnson, G., and Hartigan, J. (1999). Tau protein in normal and Alzheimer's disease brain: an update. *J. Alzheimers Dis.* 1, 329–351. doi: 10.3233/jad-1999-14-512
- Johnson, G., and Stoothoff, W. (2004). Tau phosphorylation in neuronal cell function and dysfunction. *J. Cell Sci.* 117, 5721–5729. doi: 10.1242/jcs.01558
- Jucker, M., and Walker, L. (2013). Self-propagation of pathogenic protein aggregates in neurodegenerative diseases. *Nature* 501, 45–51. doi: 10.1038/nature12481
- Kanmert, D., Cantlon, A., Muratore, C., Jin, M., O'malley, T., Lee, G., et al. (2015). C-Terminally Truncated Forms of Tau, But Not Full-Length Tau or Its C-Terminal Fragments, Are Released from Neurons Independently of Cell Death. *J. Neurosci.* 35, 10851–10865. doi: 10.1523/jneurosci.0387-15.2015
- Kar, S., Fan, J., Smith, M., Goedert, M., and Amos, L. (2003). Repeat motifs of tau bind to the insides of microtubules in the absence of taxol. *EMBO J.* 22, 70–77. doi: 10.1093/emboj/cdg001
- Karch, C., Jeng, A., and Goate, A. (2012). Extracellular Tau levels are influenced by variability in Tau that is associated with tauopathies. *J. Biol. Chem.* 287, 42751–42762. doi: 10.1074/jbc.M112.380642
- Katsinelos, T., Zeitler, M., Dimou, E., Karakatsani, A., Müller, H., Nachman, E., et al. (2018). Unconventional Secretion Mediates the Trans-cellular Spreading of Tau. *Cell Rep.* 23, 2039–2055. doi: 10.1016/j.celrep.2018.04.056
- Kenessey, A., Nacharaju, P., Ko, L., and Yen, S. (1997). Degradation of tau by lysosomal enzyme cathepsin D: implication for Alzheimer neurofibrillary degeneration. *J. Neurochem.* 69, 2026–2038. doi: 10.1046/j.1471-4159.1997.69052026.x
- Kfoury, N., Holmes, B., Jiang, H., Holtzman, D., and Diamond, M. (2012). Trans-cellular propagation of Tau aggregation by fibrillar species. *J. Biol. Chem.* 287, 19440–19451. doi: 10.1074/jbc.M112.346072
- Kim, W., Lee, S., and Hall, G. (2010a). Secretion of human tau fragments resembling CSF-tau in Alzheimer's disease is modulated by the presence of the exon 2 insert. *FEBS Lett.* 584, 3085–3088. doi: 10.1016/j.febslet.2010.05.042
- Kim, W., Lee, S., Jung, C., Ahmed, A., Lee, G., and Hall, G. (2010b). Interneuronal transfer of human tau between Lamprey central neurons in situ. *J. Alzheimers Dis.* 19, 647–664. doi: 10.3233/jad-2010-1273
- Kopeikina, K., Hyman, B., and Spire-Jones, T. (2012). Soluble forms of tau are toxic in Alzheimer's disease. *Transl. Neurosci.* 3, 223–233. doi: 10.2478/s13380-012-0032-y
- Kundel, F., Hong, L., Falcon, B., Mcewan, W., Michaels, T., Meisl, G., et al. (2018). Measurement of Tau Filament Fragmentation Provides Insights into Prion-like Spreading. *ACS Chem. Neurosci.* 9, 1276–1282. doi: 10.1021/acchemneuro.8b00094
- Lasagna-Reeves, C., Castillo-Carranza, D., Sengupta, U., Guerrero-Munoz, M., Kiritoshi, T., Neugebauer, V., et al. (2012). Alzheimer brain-derived tau oligomers propagate pathology from endogenous tau. *Sci. Rep.* 2:700. doi: 10.1038/srep00700
- Le, M., Kim, W., Lee, S., Mckee, A., and Hall, G. (2012). Multiple mechanisms of extracellular tau spreading in a non-transgenic tauopathy model. *Am. J. Neurodegener. Dis.* 1, 316–333.
- Lee, G., Newman, S., Gard, D., Band, H., and Panchamoorthy, G. (1998). Tau interacts with src-family non-receptor tyrosine kinases. *J. Cell Sci.* 111, 3167–3177.
- Lee, M., Lee, J., and Rubinsztein, D. (2013). Tau degradation: the ubiquitin-proteasome system versus the autophagy-lysosome system. *Prog. Neurobiol.* 105, 49–59. doi: 10.1016/j.pneurobio.2013.03.001
- Lee, V., Goedert, M., and Trojanowski, J. (2001). Neurodegenerative tauopathies. *Annu. Rev. Neurosci.* 24, 1121–1159. doi: 10.1146/annurev.neuro.24.1.1121
- Lewis, J., and Dickson, D. (2016). Propagation of tau pathology: hypotheses, discoveries, and yet unresolved questions from experimental and human brain studies. *Acta Neuropathol.* 131, 27–48. doi: 10.1007/s00401-015-1507-z
- Li, X., Kumar, Y., Zempel, H., Mandelkow, E., Biernat, J., and Mandelkow, E. (2011). Novel diffusion barrier for axonal retention of Tau in neurons and its failure in neurodegeneration. *EMBO J.* 30, 4825–4837. doi: 10.1038/emboj.2011.376
- Lin, M., and Beal, M. (2006). Mitochondrial dysfunction and oxidative stress in neurodegenerative diseases. *Nature* 443, 787–795. doi: 10.1038/nature05292
- Liu, L., Drouet, V., Wu, J., Witter, M., Small, S., Clelland, C., et al. (2012). Trans-synaptic spread of tau pathology in vivo. *PLoS One* 7:e31302. doi: 10.1371/journal.pone.0031302
- Liu, Y., Wei, W., Yin, J., Liu, G., Wang, Q., Cao, F., et al. (2009). Proteasome inhibition increases tau accumulation independent of phosphorylation. *Neurobiol. Aging* 30, 1949–1961. doi: 10.1016/j.neurobiolaging.2008.02.012
- Maas, T., Eidenmüller, J., and Brandt, R. (2000). Interaction of tau with the neural membrane cortex is regulated by phosphorylation at sites that are modified in paired helical filaments. *J. Biol. Chem.* 275, 15733–15740. doi: 10.1074/jbc.M000389200
- Martini-Stoica, H., Cole, A., Swartzlander, D., Chen, F., Wan, Y., Bajaj, L., et al. (2018). TFEB enhances astroglial uptake of extracellular tau species and reduces tau spreading. *J. Expe. Med.* 215, 2355–2377. doi: 10.1084/jem.20172158
- Matsumoto, S., Motoi, Y., Ishiguro, K., Tabira, T., Kametani, F., Hasegawa, M., et al. (2015). The twenty-four kDa C-terminal tau fragment increases with aging in tauopathy mice: implications of prion-like properties. *Hum. Mol. Genet.* 24, 6403–6416. doi: 10.1093/hmg/ddv351
- Min, S., Chen, X., Tracy, T., Li, Y., Zhou, Y., Wang, C., et al. (2015). Critical role of acetylation in tau-mediated neurodegeneration and cognitive deficits. *Nat. Med.* 21, 1154–1162. doi: 10.1038/nm.3951
- Min, S., Cho, S., Zhou, Y., Schroeder, S., Haroutunian, V., Seeley, W., et al. (2010). Acetylation of tau inhibits its degradation and contributes to tauopathy. *Neuron* 67, 953–966. doi: 10.1016/j.neuron.2010.08.044
- Mirbaha, H., Holmes, B., Sanders, D., Bieschke, J., and Diamond, M. (2015). Tau Trimers Are the Minimal Propagation Unit Spontaneously Internalized to Seed Intracellular Aggregation. *J. Biol. Chem.* 290, 14893–14903. doi: 10.1074/jbc.M115.652693
- Mohamed, N., Desjardins, A., and Leclerc, N. (2017). Tau secretion is correlated to an increase of Golgi dynamics. *PLoS One* 12:e0178288. doi: 10.1371/journal.pone.0178288
- Mohamed, N., Herrou, T., Plouffe, V., Piperno, N., and Leclerc, N. (2013). Spreading of tau pathology in Alzheimer's disease by cell-to-cell transmission. *Eur. J. Neurosci.* 37, 1939–1948. doi: 10.1111/ejn.12229
- Mohamed, N., Plouffe, V., Rémillard-Labrosse, G., Planel, E., and Leclerc, N. (2014). Starvation and inhibition of lysosomal function increased tau secretion by primary cortical neurons. *Sci. Rep.* 4:5715. doi: 10.1038/srep05715

- Morfini, G., Burns, M., Binder, L., Kanaan, N., Lapointe, N., Bosco, D., et al. (2009). Axonal transport defects in neurodegenerative diseases. *J. Neurosci.* 29, 12776–12786. doi: 10.1523/jneurosci.3463-09.2009
- Morris, M., Maeda, S., Vossell, K., and Mucke, L. (2011). The many faces of tau. *Neuron* 70, 410–426. doi: 10.1016/j.neuron.2011.04.009
- Nicholls, S., DeVos, S., Commins, C., Nobuhara, C., Bennett, R., Corjuc, D., et al. (2017). Characterization of TauC3 antibody and demonstration of its potential to block tau propagation. *PLoS One* 12:e0177914. doi: 10.1371/journal.pone.0177914
- Nickel, W., and Rabouille, C. (2009). Mechanisms of regulated unconventional protein secretion. *Nat. Rev. Mol. Cell Biol.* 10, 148–155. doi: 10.1038/nrm2617
- Nobuhara, C., DeVos, S., Commins, C., Wegmann, S., Moore, B., Roe, A., et al. (2017). Tau Antibody Targeting Pathological Species Blocks Neuronal Uptake and Interneuron Propagation of Tau in Vitro. *Am. J. Pathol.* 187, 1399–1412. doi: 10.1016/j.ajpath.2017.01.022
- Park, S., and Ferreira, A. (2005). The generation of a 17 kDa neurotoxic fragment: an alternative mechanism by which tau mediates beta-amyloid-induced neurodegeneration. *J. Neurosci.* 25, 5365–5375. doi: 10.1523/jneurosci.1125-05.2005
- Peeraer, E., Bittelbergs, A., Van Kolen, K., Stancu, I., Vasconcelos, B., Mahieu, M., et al. (2015). Intracerebral injection of preformed synthetic tau fibrils initiates widespread tauopathy and neuronal loss in the brains of tau transgenic mice. *Neurobiol. Dis.* 73, 83–95. doi: 10.1016/j.nbd.2014.08.032
- Perea, J., López, E., Diez-Ballesteros, J., Ávila, J., Hernández, F., and Bolós, M. (2019). Extracellular Monomeric Tau Is Internalized by Astrocytes. *Front. Neurosci.* 13:442. doi: 10.3389/fnins.2019.00442
- Pérez, M., Cuadros, R., Hernández, F., and Avila, J. (2016). Secretion of full-length tau or tau fragments in a cell culture model. *Neurosci. Lett.* 634, 63–69. doi: 10.1016/j.neulet.2016.09.026
- Pérez, M., Medina, M., Hernández, F., and Avila, J. (2018). Secretion of full-length Tau or Tau fragments in cell culture models. Propagation of Tau in vivo and in vitro. *Biomol. Concepts* 9, 1–11. doi: 10.1515/bmc-2018-0001
- Pickett, E., Henstridge, C., Allison, E., Pitsstick, R., Pooler, A., Wegmann, S., et al. (2017). Spread of tau down neural circuits precedes synapse and neuronal loss in the rTgTauEC mouse model of early Alzheimer's disease. *Synapse* 71:e21965. doi: 10.1002/syn.21965
- Plouffe, V., Mohamed, N., Rivest-Mcgraw, J., Bertrand, J., Lauzon, M., and Leclerc, N. (2012). Hyperphosphorylation and cleavage at D421 enhance tau secretion. *PLoS One* 7:e36873. doi: 10.1371/journal.pone.0036873
- Polanco, J., Li, C., Durisic, N., Sullivan, R., and Götz, J. (2018). Exosomes taken up by neurons hijack the endosomal pathway to spread to interconnected neurons. *Acta Neuropathol. Commun.* 6:10. doi: 10.1186/s40478-018-0514-4
- Polanco, J., Scicluna, B., Hill, A., and Götz, J. (2016). Extracellular Vesicles Isolated from the Brains of rTg4510 Mice Seed Tau Protein Aggregation in a Threshold-dependent Manner. *J. Biol. Chem.* 291, 12445–12466. doi: 10.1074/jbc.M115.709485
- Polydoro, M., Acker, C., Duff, K., Castillo, P., and Davies, P. (2009). Age-dependent impairment of cognitive and synaptic function in the htau mouse model of tau pathology. *J. Neurosci.* 29, 10741–10749. doi: 10.1523/jneurosci.1065-09.2009
- Pooler, A., Phillips, E., Lau, D., Noble, W., and Hanger, D. (2013). Physiological release of endogenous tau is stimulated by neuronal activity. *EMBO Rep.* 14, 389–394. doi: 10.1038/embor.2013.15
- Querfurth, H., and LaFerla, F. (2010). Alzheimer's disease. *N. Engl. J. Med.* 362, 329–344. doi: 10.1056/NEJMra0909142
- Quinn, J., Corbett, N., Kellett, K., and Hooper, N. (2018). Tau Proteolysis in the Pathogenesis of Tauopathies: neurotoxic Fragments and Novel Biomarkers. *J. Alzheimers Dis.* 63, 13–33. doi: 10.3233/jad-170959
- Ramcharitar, J., Albrecht, S., Afonso, V., Kaushal, V., Bennett, D., and Leblanc, A. (2013). Cerebrospinal fluid tau cleaved by caspase-6 reflects brain levels and cognition in aging and Alzheimer disease. *J. Neuropathol. Exp. Neurol.* 72, 824–832. doi: 10.1097/NEN.0b013e3182a0a39f
- Rauch, J., Chen, J., Sorum, A., Miller, G., Sharf, T., See, S., et al. (2018). Tau Internalization is Regulated by 6-O Sulfation on Heparan Sulfate Proteoglycans (HSPGs). *Sci. Rep.* 8:6382. doi: 10.1038/s41598-018-24904-z
- Reynolds, C., Garwood, C., Wray, S., Price, C., Kellie, S., Perera, T., et al. (2008). Phosphorylation regulates tau interactions with Src homology 3 domains of phosphatidylinositol 3-kinase, phospholipase Cgamma1, Grb2, and Src family kinases. *J. Biol. Chem.* 283, 18177–18186. doi: 10.1074/jbc.M709715200
- Reynolds, M., Berry, R., and Binder, L. (2005). Site-specific nitration differentially influences tau assembly in vitro. *Biochemistry* 44, 13997–14009. doi: 10.1021/bi051028w
- Riemenschneider, M., Wagenpfel, S., Vanderstichele, H., Otto, M., Wiltfang, J., Kretzschmar, H., et al. (2003). Phospho-tau/total tau ratio in cerebrospinal fluid discriminates Creutzfeldt-Jakob disease from other dementias. *Mol. Psychiatry* 8, 343–347. doi: 10.1038/sj.mp.4001220
- Rustom, A., Saffrich, R., Markovic, I., Walther, P., and Gerdes, H. (2004). Nanotubular highways for intercellular organelle transport. *Science* 303, 1007–1010. doi: 10.1126/science.1093133
- Sahara, N., Murayama, M., Higuchi, M., Sahara, T., and Takashima, A. (2014). Biochemical Distribution of Tau Protein in Synaptosomal Fraction of Transgenic Mice Expressing Human P301L Tau. *Front. Neurol.* 5:26. doi: 10.3389/fneur.2014.00026
- Saman, S., Kim, W., Raya, M., Visnick, Y., Miro, S., Saman, S., et al. (2012). Exosome-associated tau is secreted in tauopathy models and is selectively phosphorylated in cerebrospinal fluid in early Alzheimer disease. *J. Biol. Chem.* 287, 3842–3849. doi: 10.1074/jbc.M111.277061
- Sankaranarayanan, S., Barten, D., Vana, L., Devidze, N., Yang, L., Cadelina, G., et al. (2015). Passive immunization with phospho-tau antibodies reduces tau pathology and functional deficits in two distinct mouse tauopathy models. *PLoS One* 10:e0125614. doi: 10.1371/journal.pone.0125614
- Santarella, R., Skiniotis, G., Goldie, K., Tittmann, P., Gross, H., Mandelkow, E., et al. (2004). Surface-decoration of microtubules by human tau. *J. Mol. Biol.* 339, 539–553. doi: 10.1016/j.jmb.2004.04.008
- Schmitz, Y., Luccarelli, J., Kim, M., Wang, M., and Sulzer, D. (2009). Glutamate controls growth rate and branching of dopaminergic axons. *J. Neurosci.* 29, 11973–11981. doi: 10.1523/jneurosci.2927-09.2009
- Sengupta, S., Horowitz, P., Karsten, S., Jackson, G., Geschwind, D., Fu, Y., et al. (2006). Degradation of tau protein by puromycin-sensitive aminopeptidase in vitro. *Biochemistry* 45, 15111–15119. doi: 10.1021/bi061830d
- Shackelford, D., and Yeh, R. (1998). Dephosphorylation of tau during transient forebrain ischemia in the rat. *Mol. Chem. Neuropathol.* 34, 103–120. doi: 10.1007/bf02815073
- Shafiei, S., Guerrero-Muñoz, M., and Castillo-Carranza, D. (2017). Tau Oligomers: cytotoxicity, Propagation, and Mitochondrial Damage. *Front. Aging Neurosci.* 9:83. doi: 10.3389/fnagi.2017.00083
- Simón, D., García-García, E., Royo, F., Falcón-Pérez, J., and Avila, J. (2012). Proteostasis of tau. Tau overexpression results in its secretion via membrane vesicles. *FEBS Lett.* 586, 47–54. doi: 10.1016/j.febslet.2011.11.022
- Sokolow, S., Henkins, K., Bilousova, T., Gonzalez, B., Vinters, H., Miller, C., et al. (2015). Pre-synaptic C-terminal truncated tau is released from cortical synapses in Alzheimer's disease. *J. Neurochem.* 133, 368–379. doi: 10.1111/jnc.12991
- Sokolowski, J., Gamage, K., Heffron, D., Leblanc, A., Deppmann, C., and Mandell, J. (2014). Caspase-mediated cleavage of actin and tubulin is a common feature and sensitive marker of axonal degeneration in neural development and injury. *Acta Neuropathol. Commun.* 2:16. doi: 10.1186/2051-5960-2-16
- Spires-Jones, T., Kopeikina, K., Koffie, R., de Calignon, A., and Hyman, B. (2011). Are tangles as toxic as they look? *J. Mol. Neurosci.* 45, 438–444. doi: 10.1007/s12031-011-9566-7
- Swanson, E., Breckenridge, L., McMahon, L., Som, S., McConnell, I., and Bloom, G. (2017). Extracellular Tau Oligomers Induce Invasion of Endogenous Tau into the Somatodendritic Compartment and Axonal Transport Dysfunction. *J. Alzheimers Dis.* 58, 803–820. doi: 10.3233/jad-170168
- Sydow, A., Van Der Jeugd, A., Zheng, F., Ahmed, T., Balschun, D., Petrova, O., et al. (2011). Tau-induced defects in synaptic plasticity, learning, and memory are reversible in transgenic mice after switching off the toxic Tau mutant. *J. Neurosci.* 31, 2511–2525. doi: 10.1523/jneurosci.5245-10.2011
- Tai, H., Wang, B., Serrano-Pozo, A., Frosch, M., Spires-Jones, T., and Hyman, B. (2014). Frequent and symmetric deposition of misfolded tau oligomers within presynaptic and postsynaptic terminals in Alzheimer's disease. *Acta Neuropathol. Commun.* 2:146. doi: 10.1186/s40478-014-0146-2
- Takahashi, M., Miyata, H., Kametani, F., Nonaka, T., Akiyama, H., Hisanaga, S., et al. (2015). Extracellular association of APP and tau fibrils induces intracellular aggregate formation of tau. *Acta Neuropathol.* 129, 895–907. doi: 10.1007/s00401-015-1415-2

- Takeda, S., Wegmann, S., Cho, H., DeVos, S., Commins, C., Roe, A., et al. (2015). Neuronal uptake and propagation of a rare phosphorylated high-molecular-weight tau derived from Alzheimer's disease brain. *Nat. Commun.* 6:8490. doi: 10.1038/ncomms9490
- Tardivel, M., Bégard, S., Bousset, L., Dujardin, S., Coens, A., Melki, R., et al. (2016). Tunneling nanotube (TNT)-mediated neuron-to neuron transfer of pathological Tau protein assemblies. *Acta Neuropathol. Commun.* 4:117. doi: 10.1186/s40478-016-0386-4
- Théry, C., Ostrowski, M., and Segura, E. (2009). Membrane vesicles as conveyors of immune responses. *Nat. Rev. Immunol.* 9, 581–593. doi: 10.1038/nri2567
- Tian, H., Davidowitz, E., Lopez, P., Emadi, S., Moe, J., and Sierks, M. (2013). Trimeric tau is toxic to human neuronal cells at low nanomolar concentrations. *Int. J. Cell Biol.* 2013:260787. doi: 10.1155/2013/260787
- Tian, T., Zhu, Y., Hu, F., Wang, Y., Huang, N., and Xiao, Z. (2013). Dynamics of exosome internalization and trafficking. *J. Cell. Physiol.* 228, 1487–1495. doi: 10.1002/jcp.24304
- Usenovic, M., Niroomand, S., Drolet, R., Yao, L., Gaspar, R., Hatcher, N., et al. (2015). Internalized Tau Oligomers Cause Neurodegeneration by Inducing Accumulation of Pathogenic Tau in Human Neurons Derived from Induced Pluripotent Stem Cells. *J. Neurosci.* 35, 14234–14250. doi: 10.1523/jneurosci.1523-15.2015
- Vingtdaux, V., Sergeant, N., and Buée, L. (2012). Potential contribution of exosomes to the prion-like propagation of lesions in Alzheimer's disease. *Front. Physiol.* 3:229. doi: 10.3389/fphys.2012.00229
- von Bergen, M., Barghorn, S., Biernat, J., Mandelkow, E., and Mandelkow, E. (2005). Tau aggregation is driven by a transition from random coil to beta sheet structure. *Biochim. Biophys. Acta* 1739, 158–166. doi: 10.1016/j.bbadis.2004.09.010
- von Bergen, M., Friedhoff, P., Biernat, J., Heberle, J., Mandelkow, E., and Mandelkow, E. (2000). Assembly of tau protein into Alzheimer paired helical filaments depends on a local sequence motif ((306)VQIVYK(311)) forming beta structure. *Proc. Natl. Acad. Sci. U. S. A.* 97, 5129–5134. doi: 10.1073/pnas.97.10.5129
- Wang, J., Gao, X., and Wang, Z. (2014). The physiology and pathology of microtubule-associated protein tau. *Essays Biochem.* 56, 111–123. doi: 10.1042/bse0560111
- Wang, X., and Schwarz, T. (2009). The mechanism of Ca<sup>2+</sup>-dependent regulation of kinesin-mediated mitochondrial motility. *Cell* 136, 163–174. doi: 10.1016/j.cell.2008.11.046
- Wang, Y., and Mandelkow, E. (2016). Tau in physiology and pathology. *Nat. Rev. Neurosci.* 17, 5–21. doi: 10.1038/nrn.2015.1
- Wang, Y., Balaji, V., Kaniyappan, S., Krüger, L., Irsen, S., Tepper, K., et al. (2017). The release and trans-synaptic transmission of Tau via exosomes. *Mol. Neurodegener.* 12:5. doi: 10.1186/s13024-016-0143-y
- Wang, Y., Martinez-Vicente, M., Krüger, U., Kaushik, S., Wong, E., Mandelkow, E., et al. (2009). Tau fragmentation, aggregation and clearance: the dual role of lysosomal processing. *Hum. Mol. Genet.* 18, 4153–4170. doi: 10.1093/hmg/ddp367
- Wegmann, S., Eftekharzadeh, B., Tepper, K., Zoltowska, K., Bennett, R., Dujardin, S., et al. (2018). Tau protein liquid-liquid phase separation can initiate tau aggregation. *EMBO J.* 37:e98049. doi: 10.15252/embj.201798049
- Wu, J., Herman, M., Liu, L., Simoes, S., Acker, C., Figueroa, H., et al. (2013). Small misfolded Tau species are internalized via bulk endocytosis and anterogradely and retrogradely transported in neurons. *J. Biol. Chem.* 288, 1856–1870. doi: 10.1074/jbc.M112.394528
- Wu, J., Hussaini, S., Bastille, I., Rodriguez, G., Mrejeru, A., Rilett, K., et al. (2016). Neuronal activity enhances tau propagation and tau pathology in vivo. *Nat. Neurosci.* 19, 1085–1092. doi: 10.1038/nn.4328
- Yamada, K. (2017). Extracellular Tau and Its Potential Role in the Propagation of Tau Pathology. *Front. Neurosci.* 11:667. doi: 10.3389/fnins.2017.00667
- Yamada, K., Cirrito, J., Stewart, F., Jiang, H., Finn, M., Holmes, B., et al. (2011). In vivo microdialysis reveals age-dependent decrease of brain interstitial fluid tau levels in P301S human tau transgenic mice. *J. Neurosci.* 31, 13110–13117. doi: 10.1523/jneurosci.2569-11.2011
- Yan, X., Uronen, R., and Huttunen, H. (2020). The interaction of  $\alpha$ -synuclein and Tau: a molecular conspiracy in neurodegeneration? *Semin. Cell Dev. Biol.* 99, 55–64. doi: 10.1016/j.semdb.2018.05.005
- Yanamandra, K., Kfoury, N., Jiang, H., Mahan, T., Ma, S., Maloney, S., et al. (2013). Anti-tau antibodies that block tau aggregate seeding in vitro markedly decrease pathology and improve cognition in vivo. *Neuron* 80, 402–414. doi: 10.1016/j.neuron.2013.07.046
- Yang, L., and Ksiazek-Reding, H. (1995). Calpain-induced proteolysis of normal human tau and tau associated with paired helical filaments. *Eur. J. Biochem.* 233, 9–17. doi: 10.1111/j.1432-1033.1995.009\_1.x
- Yin, Y., Wang, Y., Gao, D., Ye, J., Wang, X., Fang, L., et al. (2016). Accumulation of human full-length tau induces degradation of nicotinic acetylcholine receptor  $\alpha$ 4 via activating calpain-2. *Sci. Rep.* 6:27283. doi: 10.1038/srep27283
- Zeineddine, R., Pundavela, J., Corcoran, L., Stewart, E., Do-Ha, D., Bax, M., et al. (2015). SOD1 protein aggregates stimulate macropinocytosis in neurons to facilitate their propagation. *Mol. Neurodegener.* 10:57. doi: 10.1186/s13024-015-0053-4
- Zempel, H., Thies, E., Mandelkow, E., and Mandelkow, E. (2010). Abeta oligomers cause localized Ca<sup>2+</sup> elevation, missorting of endogenous Tau into dendrites, Tau phosphorylation, and destruction of microtubules and spines. *J. Neurosci.* 30, 11938–11950. doi: 10.1523/jneurosci.2357-10.2010
- Zhang, Y., Tian, Q., Zhang, Q., Zhou, X., Liu, S., and Wang, J. (2009). Hyperphosphorylation of microtubule-associated tau protein plays dual role in neurodegeneration and neuroprotection. *Pathophysiology* 16, 311–316. doi: 10.1016/j.pathophys.2009.02.003
- Zhang, Z., Song, M., Liu, X., Kang, S., Kwon, I., Duong, D., et al. (2014). Cleavage of tau by asparagine endopeptidase mediates the neurofibrillary pathology in Alzheimer's disease. *Nat. Med.* 20, 1254–1262. doi: 10.1038/nm.3700
- Zhao, X., Kotilinek, L., Smith, B., Hlynialuk, C., Zahs, K., Ramsden, M., et al. (2016). Caspase-2 cleavage of tau reversibly impairs memory. *Nat. Med.* 22, 1268–1276. doi: 10.1038/nm.4199
- Zhao, Y., Tseng, I., Heyser, C., Rockenstein, E., Mante, M., Adame, A., et al. (2015). Apoptosis-Mediated Caspase Cleavage of Tau Contributes to Progressive Supranuclear Palsy Pathogenesis. *Neuron* 87, 963–975. doi: 10.1016/j.neuron.2015.08.020
- Zhou, L., McInnes, J., Wierda, K., Holt, M., Herrmann, A., Jackson, R., et al. (2017). Tau association with synaptic vesicles causes presynaptic dysfunction. *Nat. Commun.* 8:15295. doi: 10.1038/ncomms15295

**Conflict of Interest:** The authors declare that the research was conducted in the absence of any commercial or financial relationships that could be construed as a potential conflict of interest.

**Publisher's Note:** All claims expressed in this article are solely those of the authors and do not necessarily represent those of their affiliated organizations, or those of the publisher, the editors and the reviewers. Any product that may be evaluated in this article, or claim that may be made by its manufacturer, is not guaranteed or endorsed by the publisher.

Copyright © 2021 Zhang, Cao, Ma, Wei and Li. This is an open-access article distributed under the terms of the Creative Commons Attribution License (CC BY). The use, distribution or reproduction in other forums is permitted, provided the original author(s) and the copyright owner(s) are credited and that the original publication in this journal is cited, in accordance with accepted academic practice. No use, distribution or reproduction is permitted which does not comply with these terms.