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Transmission of Tau Pathology Induced by Synthetic Preformed Tau Filaments

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Review of Iba et al.

Neurodegenerative tauopathies encompass a large group of neurodegenerative disorders, including Alzheimer's disease (AD), that are characterized by the formation of tau inclusions in selective brain regions (Lee et al., 2001). The precise etiology of AD is not known in detail, but pathologically, it is characterized by progressive accumulation of abnormally modified tau in neurofibrillary tangles (NFTs), as well as formation of amyloid plaques composed mainly of β-amyloid peptides (Lee et al., 2001).

In the normal brain, tau is a highly soluble protein with limited secondary structure that binds and stabilizes the microtubule network. In the diseased state, however, tau undergoes a series of post-translational modifications that include phosphorylation (recognized by the AT8- and TG3-specific tau antibodies), abnormal conformations (MC1- or Alz-50-positive), and truncation events that lead to the formation of highly insoluble aggregates positive for Thioflavin S and resistant to proteinase K activity (Binder et al., 2005). Specifically, truncation events at the N- and C-terminal regions promote the formation of tau fragments composed mainly of the microtubule binding repeat region (MTBR) that is responsible for the aggregation and propagation of tau pathology (Binder et al., 2005; Frost et al., 2009).

In the AD brain, tau aggregates develop in a stereotypical pattern, first appearing in the transentorhinal cortex, and then spreading to the hippocampus and finally to neocortical regions (Braak and Braak, 1996). To mimic the progression of tau pathology of human AD, animal models, in which the expression of human tau harboring the P301L mutation associated with frontal temporal dementia and parkinsonism linked to chromosome 17 (FTDP-17) is restricted to the entorhinal cortex, have been generated (Liu et al., 2012; de Calignon et al., 2012). In these models, mutant tau spreads from the entorhinal cortex to interconnected hippocampal and cortical structures over time, suggesting tau aggregates propagate transsynaptically, similar to what occurs in human AD brains (Liu et al., 2012; de Calignon et al., 2012). Notably, mutant human tau recruited and coaggregated with endogenous murine tau, even in brain areas devoid of human tau expression, indicating that abnormal human tau serves as a seed to induce and propagate murine tau aggregation (de Calignon et al., 2012).

Several *in vitro* studies have shown that synthetic tau aggregates composed of specific tau isoforms can be taken up by cells and induce aggregation of endogenously expressed tau, a pathological process that can propagate between cells (Frost et al., 2009). To determine whether the tau pathology can propagate *in vivo*, Clavaguera et al. (2009) injected mouse brain lysates containing insoluble human tau proteins harboring the P301S mutation (associated with FTDP-17 and known to promote aggregation similar to P301L-tau) into brains of nonsymptomatic mice expressing human wild-type tau. Tau pathology developed at the injection site and then spread to neighboring brain regions, suggesting that insoluble brain lysates containing mutant tau proteins promote the spatial transmission of tau pathology (Clavaguera et al., 2009). A recent article published in *The Journal of Neuroscience* investigated this topic further.

To determine whether recombinant mutant (P301S) full-length or truncated (containing only the MTBR domain) tau preformed fibrils (pffs) generated *in vitro* also induce tau pathology *in vivo*, Iba et al. (2013) injected tau pffs unilaterally in young presymptomatic mice expressing human P301S-mutated tau. Tau pffs accelerated the formation of tau inclusions in young mice: inclusions appeared at 2–3 months, whereas they normally develop at 8–9 months. Tau pathology was first observed at the injection sites, but later spread to anatomically connected brain regions, a pathological process that was time and concentration dependent (Iba et al., 2013).

The tau pathology induced by synthetic tau pffs was further characterized using a panel of tau antibodies against ab-

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normal forms of tau identified in human AD brains. In contrast to the pathology observed in non-injected, old P301S mice (14 months), pff-induced tau aggregates displayed the abnormal MC1 conformation as well as hyperphosphorylated forms of tau (AT8- and TG3-positive). Moreover, these inclusions were acetylated, resistant to proteinase K treatment, and positive for Thioflavin S, thus resembling more closely the typical NFTs found in AD brains when compared with tau aggregates developed in old P301S transgenic mice. Remarkably, 2 weeks after injection, endogenous mouse tau was recruited to these inclusions, indicating that synthetic tau pffs function as seeds to recruit and induce murine tau aggregation even though murine tau has a lower propensity to aggregate than human tau (Ando et al., 2010).

Although few studies have identified the presence of endogenous mouse tau within human tau aggregates in transgenic mice, co-segregation of human and mouse tau is not surprising given that the MTBR is highly conserved between different species of tau. Alternatively, it is possible that the P301S mutation was responsible for this interspecies seeding effect, because this form of mutant tau has a higher propensity to aggregate *in vitro* and in animal models of tauopathy compared with wild-type tau (Yoshiyama et al., 2007; Combs and Gamblin, 2012).

As the P301S mice age, they display a progressive neurodegeneration along with motor and cognitive deficits (Yoshiyama et al., 2007). Strikingly, whereas pff-induced tau lesions resemble more closely the typical NFTs of AD brain than the aggregates observed in aged non-injected P301S mice (Iba et al., 2013), no neuronal loss was observed at 6 months postinjection. These findings suggest that the conformation of tau responsible for neurodegeneration may be different from the conformation that promotes the spread of tau pathology. This hypothesis is supported by the observation that although injections of P301S brain lysates promote the formation of tau aggregates in mice expressing wild-type human tau, they fail to induce any sign of neurodegeneration up to 15 months after injection (Clavaguera et al., 2009). A long-term study of the potential consequences of synthetic tau pff injection on the appearance of neurodegeneration and motor deficits remains to be reported.

It is worth noting that Iba et al. (2013), as well as Clavaguera et al. (2009), used sonication to dissociate insoluble tau filaments before injection. However, this method might play a role in promoting tau aggregation. Indeed, sonication has been shown to induce amyloid-like structures in disease and non-disease-associated proteins, thereby increasing their propensity to seed and self-propagate *in vitro* (Stathopulos et al., 2004). In fact, Stathopulos et al. (2004) determined that the cycles of sonication appear to coincide with an increase in β-sheet formation as shown by Thioflavin T fluorescence. Consequently, the sonication method may induce a tau conformation that is highly prone to aggregation and, thus, different from the pathological conformation present in human AD brains. Furthermore, sonicated proteins contain modifications that are induced by high heat and radical reactions generated during the breakup of water, which may lead to the formation of reactive oxygen species (O_2^- and H_2O_2) (Stathopulos et al., 2004). Therefore, artificially generated modifications to tau pffs following sonication cannot be ruled out. It is thus important to identify, within all the species present in sonicated tau pffs, the conformation of tau responsible for the propagation of tau pathology and determine its physiological relevance.

The fact that human α-synuclein pffs, which were injected as control (Iba et al., 2013), failed to induce tau pathology within the P301S model is notable given that cross-seeding mechanisms between tau and other proteins involved in neurodegenerative diseases have been described. More specifically, Waxman and colleagues demonstrated a cross-seeding effect between α-synuclein and human tau *in vitro* (Waxman and Giasson, 2011) and co-aggregation of these two proteins *in vivo* in a double transgenic mouse expressing mutant human α-synuclein (A53T) and P301L mutant tau (Giasson et al., 2003). The failure of α-synuclein pffs to induce tau aggregation in the study by Iba et al. (2013) may be explained by the quantity of pffs injected or the incubation periods required for α-synuclein pffs to promote NFT-like aggregation. It is also possible that mutant α-synuclein pffs may be more effective than wild-type α-synuclein in promoting the aggregation of P301S tau *in vivo*.

Although the tau conformation responsible for neurodegeneration has yet to be elucidated, these findings clearly indicate that specific isoforms of tau, when injected into the brain of transgenic mice overexpressing human tau proteins, have the ability to induce tau pathology that can spread from one area of the brain to another in a prion-like fashion.

References

Ando K, Leroy K, Heraud C, Kabova A, Yilmaz Z, Authelet M, Suain V, De Decker R, Brion JP (2010) Deletion of murine tau gene increases tau aggregation in a human mutant tau transgenic mouse model. Biochem Soc Trans 38: 1001–1005. CrossRef Medline

Binder LI, Guillozet-Bongaarts AL, Garcia-Sierra F, Berry RW (2005) Tau, tangles, and Alzheimer's disease. Biochim Biophys Acta 1739: 216–223. CrossRef Medline

Braak H, Braak E (1996) Evolution of the neuropathology of Alzheimer's disease. Acta Neurol Scand Suppl 165:3–12. Medline

Clavaguera F, Bolmont T, Crowther RA, Abramowski D, Frank S, Probst A, Fraser G, Stalder AK, Beibel M, Staufenbiel M, Jucker M, Goedert M, Tolnay M (2009) Transmission and spreading of tauopathy in transgenic mouse brain. Nat Cell Biol 11:909–913. CrossRef Medline

Combs B, Gamblin TC (2012) FTDP-17 Tau mutations induce distinct effects on aggregation and microtubule interactions. Biochemistry 51:8597–8607. CrossRef Medline

de Calignon A, Polydoro M, Suárez-Calvet M, William C, Adamowicz DH, Kopeikina KJ, Pitstick R, Sahara N, Ashe KH, Carlson GA, Spires-Jones TL, Hyman BT (2012) Propagation of tau pathology in a model of early Alzheimer's disease. Neuron 73:685–697. CrossRef Medline

Frost B, Jacks RL, Diamond MI (2009) Propagation of tau misfolding from the outside to the inside of a cell. J Biol Chem 284:12845–12852. CrossRef Medline

Giasson BI, Forman MS, Higuchi M, Golbe LI, Graves CL, Kotzbauer PT, Trojanowski JQ, Lee VM (2003) Initiation and synergistic fibrillization of tau and alpha-synuclein. Science 300:636–640. CrossRef Medline

Iba M, Guo JL, McBride JD, Zhang B, Trojanowski JQ, Lee VM (2013) Synthetic tau fibrils mediate transmission of neurofibrillary tangles in a transgenic mouse model of Alzheimer's-like tauopathy. J Neurosci 33: 1024–1037. CrossRef Medline

Lee VM, Goedert M, Trojanowski JQ (2001) Neurodegenerative tauopathies. Annu Rev Neurosci 24:1121–1159. CrossRef Medline

Liu L, Drouet V, Wu JW, Witter MP, Small SA, Clelland C, Duff K (2012) Trans-synaptic spread of tau pathology in vivo. PLoS One 7:e31302. CrossRef Medline

Stathopulos PB, Scholz GA, Hwang YM, Rumfeldt JA, Lepock JR, Meiering EM (2004) Sonication of proteins causes formation of aggregates that resemble amyloid. Protein Sci 13:3017–3027. Medline

Waxman EA, Giasson BI (2011) Induction of intracellular tau aggregation is promoted by alpha-synuclein seeds and provides novel insights into the hyperphosphorylation of tau. J Neurosci 31:7604–7618. CrossRef Medline

Yoshiyama Y, Higuchi M, Zhang B, Huang SM, Iwata N, Saido TC, Maeda J, Suhara T, Trojanowski JQ, Lee VM (2007) Synapse loss and microglial activation precede tangles in a P301S tauopathy mouse model. Neuron 53: 337–351. CrossRef Medline