REVIEW

Prions, Amyloid Precursor Protein, and Tau

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Prions, amyloid precursor protein (APP), and tau, and the diseases that result from them, share common characteristics. These include the manner in which the diseases progress, and the structures and physiology of the proteins. This paper discusses some common characteristics.

Proteinaceous infectious (prion) diseases result from the transformation of normal "cellular" prions – called PrP or PrP^{C} - to the disease form called PrP^{sc} where the sc refers to "scrapie", or PrP^{res} where the res refers to "protease resistant". This transformation is a post-translational process and involves conversion from the primarily α helices in normal prions to the β sheets of the disease form, which are resistant to proteases. This is a self-propagational process, as the disease form acts as a template into which the normal prions transform, and yet the amino acid sequence of both remains the same. Consisting only of protein, prions are the only known infectious agents that do not have nucleic acids, meaning they have no DNA or RNA (*1,2*).

Although located throughout the body, prions are most prevalent in the nervous system, and this is primarily where disease prions do harm. Amyloid precursor proteins (APP) and tau are also located throughout the body but primarily in the nervous system, the location where the disease forms of these proteins primarily do harm (*1,2*).

Background

Normal prions are encoded by chromosomes, and are known as PrP. The PrP protein contains approximately 209 amino acids. Disease-causing prions have been designated PrP^{sc}. The "sc" is from "scrapie", the disease-causing form that infects sheep and goats, and the first prion disease that was detected. Other prion diseases include bovine spongiform encephalopathy (BSE), which affects cows; feline spongiform encephalopathy, which affects cats; chronic wasting disease, which affects deer; transmissible mink encephalopathy, which affects ranch raised mink; and exotic ungulate encephalopathy, which affected zoo animals in Great Britain. The forms that affect humans include Creutzfeldt Jakob disease (CJD), variant Creutzfeldt Jakob disease (BSE contracted by humans), fatal familial insomnia, kuru (infection caused by cannibalism), and Gerstmann-Straussler-Scheinker syndrome (*2a*).

The amyloid precursor protein (APP) gene is located on chromosome 21. One mutation in the APP gene can cause a predisposition for Alzheimer's disease (AD) (*3*), and another mutation in the APP gene can protect against the disease (*4*). The entire APP protein contains approximately 639 to 770 amino acids, depending on the isoform. The disease form of APP results from its cleavage.

The gene that encodes tau protein is located on chromosome 17. Tau protein contains approximately 352 to 441 amino acids, depending on the isoform, of which there are six. The disease form of tau results from its improper phosphorylation. Besides AD, which is the most common, other diseases caused by tauopathy include corticobasal degeneration (CBD), Pick's disease, tangle only dementia, chronic traumatic encephalopathy (CTE), progressive supranuclear palsy (PSP), and argyrophilic grain disease (*5*).

Two Cell Membrane Proteins and One MAP

At least two types of prion and APP are cell transmembrane proteins, whereas tau is a microtubule associated protein (MAP).

PrP is anchored to the cell surface with a phosphatidylinositol glycolipid anchor. Accordingly, the cell membrane moiety is glycosylphosphatidylinositol (GPI) (6). The GPI anchor is necessary for PrP^{sc} to form inside cells. Without the anchor, PrP^{sc} forms outside the cell (*7*).

Like many other transmembrane proteins, prions have two domains – a globular domain and a long flexible unstructured tail (*8*). PrP contains a signal peptide at the amino (N) terminus. This peptide consists of a positively charged N terminus domain, a hydrophobic domain in the center, and a carboxyl (C) terminus domain. There is a cleavage site following the C terminus domain (*6*). Although located primarily in the nervous system, prions are also found in many other parts of the body (*8*).

APP is a Type I integral transmembrane (TM) glycoprotein. It has a hydrophobic single pass with its amide (NH_2) terminus outside the cell and carboxyl (COOH-) terminus inside the cell. APP is sent to the cell surface after post-translational processing in the Golgi body (*9*). It has a large ectodomain consisting of two subdomains – termed E1 and E2. These are connected by a negatively charged acidic domain – termed AcD, but there is no appreciable interaction between E1 and E2. E2 is directly connected to the transmembrane domain by JMR, a juxtamembrane region. E1 contains two subdomains – a copper binding domain (CuBD) and growth factor like domain (GFLD) (*10*). The TM domain is helical in structure (*11*). The APP intracellular domain (AICD) contains 15 to 20 residues that are unstructured (*12*). APP gets cleaved in the AB section by α-secretase or by β-secretase (BACE) in the amino terminal region, resulting in fragments that differ by only 17 amino acids. Each type of cleavage results in secretion of large ectodomains that are soluble. They are termed APPsa and APPsb. APPsa has been found to have neuroprotective and synaptotrophic roles, and APPsb was found to be far less active and possibly toxic (*13*).

Tau is an intracellular MAP, located primarily in neuronal axons, but also, to a lesser extent, in dendrites, oligo-

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dendrocytes, and in the lungs, kidneys, and testis. In its native form, it is unstructured and flexible, with a low proportion of secondary structures such as α-helices or β-strands. It has three domains – an N terminus which projects away from microtubules, a proline rich middle domain, and a C terminus which is the one that binds to microtubules. As with many multidomain proteins, the N terminus is acidic, and the C terminus is basic (*14*). The overall composition of tau is highly hydrophilic, which explains the fact that it is not compactly folded, as is common with most intracellular proteins (*15*). The disease form is characterized as having hyperphosphorylated inclusions that are insoluble and filamentous (*16*).

Normal Functions

Normal prions have a number of functions, including protecting cells from the oxidative stress that results from copper binding (*7,17,18),* and protecting N-methyl-D-aspartate receptors (*19*). Its transmembrane signaling function is involved in the development of neurons and in hematopoietic stem cell replication *(20)*. APPs are found throughout most tissues in the body, but are most prevalent in neurons, and most abundant in the central nervous system (CNS) where they protect against apoptosis. There is also antioxidant protection as well as signaling activity (*21,22*).

APP is essential to the development of neuromuscular and central nervous system (CNS) synapses and for the plasticity of CNS synapses (*23*). Evidence suggests it is involved with the development of neural stem cells, with the survival and repair of neurons, and with neurite outgrowth (*9*).

APP is also involved in cell matrix adhesion – the extracellular domain interacts with heparin, collagen, and laminin, which are parts of the extracellular matrix; cell to cell adhesion – by transcellular associations; neural and synaptic trophic functions – removing or reducing APP was shown to impair the viability of neurons, and to reduce synaptic activity; axon pruning and degeneration – APPsa was shown to protect against it, and APPsb was shown to promote it; intracellular signaling – cleavage at the cell membrane/ intracellular boundary produces the APP intracellular domain (AICD), which translocates to the nucleus where its proposed targets are KAI, GSK3Beta, EGFR, p53, LRP, APP, calcium regulation genes, and cytoskeletal activities; and apoptosis, the outcome when AICD is cleaved by caspases, resulting in two fragments, one of which appears to promote toxicity (*13*).

APP has been shown, in *C.elegans*, to interact with PIKfyve, an important endosomal protein. This interaction is essential for proper endosomal sorting, and improper PIKfyve functioning can cause extreme nerve degeneration, resulting in death in mice, and in Charcot-Marie-Tooth and Amyotrophic Lateral Sclerosis diseases in humans (*24*).

The primary function of tau is polymerization of tubulin, which results in the formation and stabilization of microtubules. Tau protein associates with neuronal microtubules, which it stabilizes by promoting the polymerization of tubulin (*25*). It is also involved in growth factor signaling (*14*). The main post translational modification of tau is phosphorylation. In fact, tau has 85 phosphorylation sites.

Disease Processes

The disease form of prions is characterized by a transformation from the primarily α-helical structure of PrP to the primarily β-sheet structure of disease prions (PrP^{sc} or PrP^{res}), resulting in amyloid plaques (*6*), and an increase in cell membrane permeability (*26*). The development of disease causing prions has been shown to be influenced by genetics, by transmission from another organism, or by neither (sporadic) (*2*). However, the majority of cases are sporadic (*8*).

An experiment with transgenic mice supports the hypothesis that PrP^{sc} acts as a template that has the effect of transforming PrP to PrP^{sc} (*2*). The transformation takes place in the caveolae domains (CLDs) located in the proximity of cell surfaces (*27*). That is, PrP^{sc} aggregates into amyloid plaques outside the cell (*6*).

The amino acid sequences of normal prions are identical to the amino acid sequences of the disease-causing prions that are formed from them, but the conformation is different. PrPs have a structure that is approximately 40% α-helices with few β-sheets, and PrP^{sc}s have a structure that is approximately 30% α-helices and 40% β-sheets. This conformational difference results in prions that are resistant to proteases, resulting in the accumulation of inappropriate prions, which tend to be insoluble (*2,28*).

The beta amyloids of Alzheimer's disease (AD) are formed when APP is cleaved by β-secretase and then by γ-secretase to form C99, the APP carboxyl terminus domain of the transmembrane portion (*30*). Like prion diseases, AD can be caused by genetic factors, by transmission from one organism to another, or by sporadic causes. However, as with prion diseases, most cases are sporadic (*8*), and the only known cases caused by transmission are those done in experiments with transgenic mice, such as one that shows that natural and synthetic AD beta amyloids injected into transgenic mouse brains self-propagate (*29*).

Tau transforms into toxic neurofibrillary tangles (NFT), also known as tau fibrils, upon conformational changes that can occur with an imbalance between tau kinases and phosphatases. These conformational changes can result in over phosphorylation, which can result in a decrease in microtubule binding resulting in microtubule destabilization, and tau aggregation. The tau can then accumulate in the neuronal soma, where it can form neurofibrillary tangles, one of the hallmarks of AD, and can affect neuronal processes such as axonal transport, mitochondrial metabolism, post-synaptic functioning, and cell signaling. In AD the disease progresses from the locus coeruleus to the entorhinal cortex to the limbic regions, and then to the neocortical regions of the brain. The eventual outcome is cognitive decline (*14*). Like PrP^{sc}, NFTs are characterized by β-sheet-rich amyloid fibrils that are insoluble, in contrast to tau, which does not have significant secondary or tertiary structures (*31*).

Intracellular tau fibrils are secreted from an infected cell and taken up by other cells. Once inside another cell, the

tau fibrils act as seeds, in a manner similar to the behavior of prions, causing the fibrillization of the native tau in those cells (*32*). Through the use of transgenic mice it was shown that tau aggregation can be transmitted from one animal to another. Brain extracts from transgenic mice that had tau inclusions that were injected into transgenic mice that had native tau only (no inclusions) resulted in the transformation of the native tau into tau inclusions. Additionally, the tau inclusions spread into regions away from the site of injection (*33*). It was also shown that tau misfolding can be transmitted from misfolded tau seeds that act as a template (*5*). Additionally, cell free tau pathology can be heterotypically seeded by beta amyloid, in a prion like manner (*34*).

A positive correlation was found between beta amyloid formation and the amount of extracellular N terminal fragments of tau in the brain (*35*). Evidence points to the fact that tau is required for beta amyloids to form. However, it is not yet known if tau causes AD or it is caused by AD (*15*). Tau levels after traumatic brain injuries were found to be correlated with levels of normal prion protein (*36*), and tau levels were found to increase upon inoculation with disease prions (*37*). Like disease prions and beta amyloids, NFTs self-propagate when injected into murine brains (*31*).

Metals

Biometal homeostasis, or lack thereof, has been implicated in neurodegenerative diseases, including prion disorders and AD. Lack of biometal homeostasis is related to oxidative stress, which is implicated in AD (*38*). Copper has been implicated in prion diseases and in AD (*18*). PrP is a copper binding protein. Mice that lack the PrP gene were found to have one half the synaptosomal copper concentrations as normal mice (*39*). Copper has been found to cause proteinase resistance in PrP, and copper chelation has been shown to delay the onset of prion disease. Other experiments have resulted in contradictory conclusions. For instance, the binding of copper to PrP was shown to destabilize the primarily α-helix structure of PrP, resulting in an increase of the β-sheets characteristic of PrP^{sc}. Copper has also been shown to increase the infectiousness of PrP^{sc}. Seemingly contrarily, copper has also been shown to hinder the amplification of PrP^{res}, and it helps prevent reactive oxygen caused neuropathology. Lastly, seemingly contradictory to the fact that copper chelation has been shown to delay the onset of prion disease, another experiment shows that a diet high in copper also resulted in delays of the onset of prion disease (*40*).

In AD, copper has been shown to promote the disease, and it has also been shown to cause improvements in cognitive function in transgenic AD mice who already had the disease (*41*). A recent study shows significantly higher free copper levels in patients with AD (*42*). APP has a copper binding domain (CuBD), which is located next to the growth factor domain, in the amino (N) terminal. It has a tertiary structure with one α-helix, three β-sheets, and a hydrophobic central region. Three disulfide bridges provide stabilization. The CuBD was found to affect APP metabolism, in addition to binding copper (*43*). Copper was found to increase tau phosphorylation and the copper chelator, tetrathiomolybdate, was found to decrease tau phosphorylation. It was also found that the addition of zinc lowered copper levels in the brain (*44*), and beta amyloids contain a disproportionately high level of copper (*45*).

PrP was shown to be a ferrireductase, an enzyme that reduces ferric iron (iron [III]) to ferrous iron (iron [II]). Genetically modified mice without the PrP gene acquire iron deficiency. Additionally, iron imbalances are common in Creutzfeldt-Jakob prion disease brains (*46*).

APP was found to be an active ferroxidase, an enzyme that oxidases iron II to iron III, thereby preventing oxidative stress. It also binds ferroportin, which exports iron from neuron cells, thereby helping to further prevent oxidative stress, and consequent neurodegenerative diseases, that are caused by iron (*47*). AD patients have been shown to have higher than normal iron levels in their brains, with the levels in beta amyloids being three times the levels in non-beta amyloid areas of the brain. An in vitro experiment shows an increase in the rate of formation of beta amyloids and tau tangles with an increase in iron levels (*48*).

PrP has been shown to regulate zinc in neurons, including its uptake into neuronal cells. In neurons, zinc binds to the N terminus region of the prion protein, at the same site where copper binds, where it has been shown to promote the PrP to PrP interface. In fact, it has been determined that ZIP (Zrt/Irt-like protein) transmembrane transporter genes are evolutionary ancestors of prion genes (*49*). The levels of zinc in the brain are greater than in any other tissue, and beta amyloids have a greater concentration than non-beta amyloid sections of the brain (*45*).

Ion transport with normal prions is increased with increases in calcium, similar to the increase in permeability that results with the transformation from PrP to PrP^{sc}. This increase in ion transport increases toxicity (*25*). The homeostasis of intracellular calcium is upset in AD brains, resulting in increased beta amyloid formation and an increased hyperphosphorylation of tau. In addition, beta amyloid can have an effect on signaling pathways that are involved in calcium buffering, thereby affecting the efficiency of neuronal responses to excitotoxic circumstances (*50*). Changes in Ca++ signaling have been shown to cause the commencement of the beta amyloid pathway of AD (*26*).

Cholesterol

As with most GPI transmembrane proteins, prions are associated with lipid rafts, which are cell membrane domains that have high levels of glycosylated sphinoglipids and cholesterol. It has been shown that a reduction in the synthesis of cholesterol disrupts this association, and prevents the transformation of PrP to PrP^{sc}. PrP can be transformed into PrP^{sc} without being bound to a membrane (and therefore a lipid raft), but this binding was shown to be necessary for the PrP^{sc} to be toxic. Treatment with mevinolin, a cholesterol inhibitor results in misfolded precursor (immature) PrP, which affects the development of mature PrP and therefore the production of PrP^{sc} (*51*).

High levels of cholesterol may also affect the onset and advancement of AD.

Cranial plasma membranes from AD patients have higher cholesterol levels than controls, and progressively higher levels of cholesterol as the disease progresses. In other words, there is more cholesterol in stage 6 (severe decline) patient brains than in stage 4 (moderate decline) patients, who have more brain cholesterol than stage 3 (mild decline) patients. Additionally, docosahexaenoic acid (DHA), a promoter of cholesterol homeostasis, is lower in AD patients than in controls (*51*).

Discussion

Disease prions and the proteins implicated in AD (APP and tau) have a number of similarities. All three proteins can result from transmission or inheritance, or can occur sporadically. The native forms of the three proteins or precursors are soluble, flexible, unstructured, and the disease forms are insoluble and structured. The disease forms for all three act as templates into which the native forms transform. All three are affected by the presence of metals. Disease forms are promoted with increases in cholesterol.

REFERENCES AND NOTES

1. F.E. Cohen, K.M. Pan, Z. Huang, M. Baldwin, R.J. Fletterick, S.B. Prusiner. Structural clues to prion replication. *Sci.* **264:5158**, 530-531 (1997).
2. S.B. Prusiner. Molecular biology and genetics of prion diseases. Phi. Trans:Bio. Sci. 343:1306, 447-463. (1994).
2. Prion Diseases. Centers for Dis. Cont'l and Prev. (updated 2017). https:// www.cdc.gov/prions/index.html.
3. J. Murrell, M. Farlow, B. Ghetti, M.D. Benson. A mutation in the amyloid precursor protein associated with hereditary Alzheimer's disease. *Sci.* **254:5028**, 97-9 (1991).
4. T. Jonsson, J.K. Atwal, S. Steinberg, J. Snaedel, P.V. Jonsson, S. Bjornson, H. Stefansson, P. Sulem, D. Gudbjartsson, J. Maloney, K. Hoyte, A. Gustafson, Y. Liu, Y. Lu, T. Bhangale, R.R. Graham, J. Huttenlocher, G. Bjornsdottir, O.A. Andreassen, E.G. Jonsson, A. Palotie, T.W. Behrens, O.T. Magnusson, A. Kong, U. Thorsteinsdottir, R.J. Watts, K. Stefansson. A mutation in APP protects against Alzheimer's disease and age-related cognitive decline. *Nature.* **488:7409**, 96-9 (2012.I.
5. A.L. Woerman, A. Aoyagi, S. Patel, S.A. Kazmi, I. Lobach, L.T. Grinberg, A.C. McKee, W.W. Seeley, S.H. Olson, S.B. Prusiner. Tau prions from Alzheimer's disease and chronic traumatic encephalopathy patients propagate in cultured cells. Proc. Natl. Acad. Sci. USA. 113:50, E8187-E8196 (2016).
6. K. Basler, B. Oesch, M. Scott, D. Westaway, M. Walchi, D.F. Groth, M.P. McKinley, S. B. Prusiner, C. Weissman. Scrapie and cellular PrP isoforms are encoded by the same chromosomal gene. *Cell.* **46:3**, 417-28 (1986).
7. T. Sonati, R.R. Reimann, J. Falsig, P.K. Baral, T. O'Connor, S. Hornemann, S. Yaganoglu, B. Lei, U.S. Herrmann, B. Wieland, M. Swayampakula, M.H. Rahman, D. Das, N. Kav, R. Riek, R. P. Liberski, M. N. G. James, A. Aguzzi. The toxicity of antiprion antibodiesis mediated by the flexible tail of the prion protein. *Nature.* **501**, 102-106 (2013).
8. S.B. Prusiner. Shattuck lecture – neurodegenerative diseases and prions. *N. Engl. J. Med.* **344:20**, 1516-26 (2001).
9. E.H. Koo, S.L. Squazzo, D.J. Selkoe, C.H. Koo. Trafficking of cell-surface amyloid beta-protein precursor. I. Secretion, endocytosis and recycling as detected by labeled monoclonal antibody. *J. Cell Sci.* **109:5**, 991-8 (1996).
10. I. Coburger, S.O. Dahms, D. Roeser, K.H. Guhrs, P. Hortschansky, M.E. Than. Analysis of the overall structure of the multi-domain amyloid precursor protein (APP). *PloS One*. **8:12**, e81926 (2013).
11. K.D. Nadezhdin, O.V. Bocharova, E.V. Bocharov, A.S. Arseniev. Dimeric structure of transmembrane domain of amyloid precursor protein in micellar environment. *FEBS Lett*. **586:12**, 1687-92 (2012). 013).
12. J. Radzimanowski, B. Simon, M. Sattler, K. Beyreuther, I. Sinning, K. Wild. Structure of the intracellular domain of the amyloid precursor protein in complex with Fe65-PTB2. *EMBO Rep.* **9:11**, 1134-40 (2008).
13. P.-H. Kuhn, H. Wang, D. Bastian, A. Colombo, U. Zeitschel, J.W. Ellwart, E. Kremmer, S. Robner, S.F. Lichtenthaler. ADAM10 is the physically relevant, constitutive α-secretase of the amyloid precursor protein in primary neurons. *EMBO J.* **29:17**, 3020-32 (2010).
14. A. Mietelska-Porowska, U. Wasik, M. Goras, A. Filipek, G. Niewiadomska. Tau protein modifications and interactions: their role in function and dysfunction. *Int. J. Mol. Sci.* **15:3**, 4671-713 (2014).
15. M.D. Mukrasch, S. Bibow, J. Korukottu, S. Jeganathan, J. Biernat, C. Griesinger, E. Mandelkow, M. Zweckstetter. Structural polymorphism of 441-residue tau at single residue resolution. *PLoS Biol.* **7: 2**, e1000034.
16. B. Falcon, A. Cavallini, R. Angers, S. Glover, T.K. Murray, L. Bernham, S. Jackson, M.J. O'Neill, A.M. Isaacs, M.L. Hutton, P.G. Szekeres, M. Goedert, S. Bose. *J. Biol. Chem.* **290:2**, 1049-65 (2015).
17. T. Voigtlander, S. Kloppel, P. Birner, C. Jarius, H. Flicker, S. Verghese-Nikolakaki, T. Sklaviadis, M. Guentchev, H. Budka. Marked increase of neuronal prion protein immunoreactivity in Alzheimer's disease and human prion diseases. *Acta. Neuropathol*, **101**, 417–423 (2001).
18. M. Hodak, R. Chisnell, W. Lu, J. Bernholc. Functional implications of multistage copper binding to the prion protein. *Proc. Natl. Acad. Sci. USA.* **106:28**, 11576-81 (2008).
19. H. You, S. Tsutsui, S. Hameed, T.J. Kannanayakal, L. Chen, P. Xia, J.D.T. Engbers, S.A. Lipton, P.K. Stys, G.W. Zamponi, AB neurotoxicity depends on interactions between copper ions, prion protein, and N-methyl-D-aspartate. *Proc. Natl. Acad. Sci. U.S.A.* **109:5**, 1737-1742 (2012).
20. C.C Zheng, A.D. Steele, S. Lindquist, H.F. Lodish. Prion protein is expressed on long-term repopulating hemato-poietic stem cells and is important for their self-renewal. *Proc. Natl. Acad. Sci. USA.* **103:7**, 2184-89 (2006).
21. Y. Bounhar, Y. Zhang, C.G. Goodyer, A. LeBlanc. Prion protein protects human neurons against Bax-mediated apoptosis. *J. Biol. Chem.* **276:42**, 39145-9 (2001).
22. X. Roucou, M. Gains, A C. LeBlanc. Neuroprotective functions of prion protein. *J. Neurosci. Res.* **75:2**, 153-61 (2004).
23. M. Klevanski, U. Herrmann, S.W. Weyer, R. Fol, N. Cartier, D.P. Wolfer, J.H. Caldwell, M. Korte, U.C. Muller. The APP intracellular domain is required for normal synaptic morphology, synaptic plasticity, and hippocampus-dependent behavior. *J. Neurosci.* **35:49**, 16018-33 (2015).

24. Z. Balklava, C. Niehage, H. Currinn, L. Mellor, B. Guscott, G. Poulin, B. Hoflack, T. Wassmer. The amyloid precursor protein controls PIKfyve function. *PloS One.* **10:6**, e0130485 (2015).
25. D. G. Drubin, M.W. Kirschner. Tau protein function in living cells. *J. Cell Bio.* **103:6:2**, 2739-46 (1986).
26. S. Sorrentino, T. Bucciarelli, A. Corsaro, A. Tosatto, S. Thellung, V. Villa, M.E. Schinina, B. Maras, R. Galeno, L. Scotti, F. Creati, A. Marrone, N. Re, A. Aceto, T. Florio, M. Mazzanti. Calcium binding promotes prion protein fragment 90-231 conformational change toward a membrane destabilizing and cytotoxic structure. *Plos One.* **7:7**, e38314 (2012).
27. I.Y. Lee, D. Westway, A.F. Smit, K. Wang, J. Seto, L. Chen, C. Acharya, M. Ankener, D. Baskin, C. Cooper, H. Yao, S.B. Prusiner, L.E. Hood. Complete genomic sequence and analysis of the prion protein gene region from three mammalian species. *Genome Res.* **8:10**, 1022-37 (1998).
28. J. Stohr, N. Weinmann, H. Wille, T. Kaiman, L. Nagel-Steger, E. Birkman, G. Panza, S.B. Prusiner, M. Eigen, D. Reisner. Mechanisms of prion protein assembly into amyloid. *Proc. Natl. Acad. Sci. U. S. A.* **105:7**, 2409-2414 (2008).
29. J. Stohr, J.C. Watts, Z.L. Mensinger, A. Oehler, S. K. Grillo, S.J. DeArmond, S.B. Prusiner, K. Giles. Purified and synthetic Alzheimer's amyloid beta prions. *Proc. Acad. Sci. U. S. A.* **09:27**, 11025-11030 (2012).
30. P.J. Barrett, Y. Song, W.D. Van Horn, E.J. Schafer, A. Hadziselimovic, A.J. Beel, C.R. Sanders. The amyloid precursor protein has a flexible transmembrane domain and binds cholesterol. *Science.* **366:6085**, 1168-71 (2012).
31. J.L. Guo, S. Narasimhan, L. Changolkar, Z. He, A. Steiber, B. Zhang, R.J. Gathagan, M. Iba, J.D. McBride, J Q. Trojanowski, V.M. Lee. Unique pathological tau conformers from Alzheimer's brains transmit tau pathology in nontransgenic mice. *J. Exp. Med.* **213:12**, 2635-54 (2016).
32. N. Kfoury, B.B. Holmes, H. Jiang, D.M. Holtzman, M.I. Diamond. Trans-cellular propagation of tau aggregation by fibrillar species. *J. Biol. Chem.* **287:23**, 19440-51 (2012).
33. F. Clavaguera, T. Bolmont, R.A. Crowther, D. Abramowski, S. Frank, A. Probst, G. Fraser, A.K. Stalder, M. Beibel, M. Staufenbiel, M. Jucker, M. Goedert, M. Tolnay. Transmission and spreading of tauopathy in transgenic mouse brain. *Nat. Cell Biol.* **11:7**, 909-13 (2009).
34. B. Vasconcelos, I.C. Stancu, A. Buist, M. Bird, P. Wang, A. Vanoosthuyse, K. Van Kolen, A. Verheyen, P. Kienlen-Campard, J.N. Octave, P. Baatsen, D. Moechars, I. Dewachter. Heterotypic seeding of tau fibrillization by pre-aggregated Abeta provides potent seeds for prion-like seeding and propagation of tau-pathology in vivo. *Acta. Neuropathol.* **131:4**, 549-69 (2016).
35. J. Bright, S. Hussain, V. Dang, S. Wright, B. Cooper, T. Byun, C. Ramos, A. Singh, G. Parry, N. Stagliano, I. Griswold-Prenner. Human secreted tau increases amyloid-beta production. *Neurobiol. Aging.* **36:2**, 693-709 (2015).
36. R. Rubenstein, B. Chang, N. Grinkina, E. Drummond, P. Davies, M. Ruditzky, D. Sharma, K. Wang, T. Wishiewski. Tau phosphorylation induced by severe closed head traumatic brain injury is linked to the cellular prion protein. *Acta. Neuropathol. Comm.* **5:30** (2017).
37. R. Rubinstein, B. Chang, R. Petersen, A. Chiu, P. Davies. T-tau and P-tau in brain and blood from natural and experimental prion diseases. *PloS One*. **10:12**, e0143103 (2015).
38. I.M. Balmus, S. Strungaru, A. Ciobica, M. Nicoara, R. Dobrin, G. Plavan, C. Stefanescu. Preliminary data on the interaction between some biometals and oxidative stress statue in mild cognitive impairment and Alzheimer's disease patients. *Oxid. Med. Cell Longev.* **2017:7156928** C(2017).
39. H.A. Kretzschmar, T. Tings, A. Madlung, A. Giese, J. Herms. Function of PrP(C) as a copper-binding protein at the synapse. *Arch. Virol. Suppl.* **16**, 239-49 (2000).
40. O.M. Siggs, J.T. Cruite, X. Du, S. Rutschmann, E. Masliah, B. Beutler, M.B.A. Oldstone. Disruption of copper homeostasis due to a mutation of Atp7a delays the onset of prion disease. *Proc. Natl. Acad. Sci. USA*. **109:34**, 13733-38 (2012).
41. P.J. Crouch, L.W. Hung, P A. Adlard, M. Cortes, V. Lal, G. Filiz, K.A. Perez, M. Nurjono, S. Caragounis, T. Du, K. Laughton, I. Volitakis, A.I. Bush, Q.X. Li, C.L. Masters, R. Cappai, R.A. Cherry, P.S. Donnelly, A.R. White, K.J. Barnham. Increasing Cu bioavailability inhibits Abeta oligomers and tau phosphorylation. *Proc. Natl. Acad. Sci. USA*. **106:2**, 381-6 (2009).
42. G.J. Brewer. Alzheimer's disease causation by copper toxicity and treatment with zinc. *Front Aging Neurosci.* **6:92** (2014).
43. L. Spoerri, L.J. Vella, C.L. Pham. K.J. Barnham, R. Cappai. The amyloid precursor protein copper binding domain histidine residues 149 and 151 mediate APP stability and metabolism. *J. Biol. Chem*. **287:32**, 26840-53 (2012).
44. K. Voss, C. Harris, M. Ralle, M. Duffy, C. Murchison, J.F. Quinn. Modulation of tau phosphorylation by environmental copper. *Transl. Neurodegener.* **3:1**, 24 (2014).
45. M.A. Lovell, J.D. Robertson, W.J. Teesdale, J.L. Campbell, W.R. Markesbery. Copper, iron and zinc Alzheimer's disease senile plaques. *J. Neurol. Sci.* **158:1**, 47-52 (1998).
46. A. Singh, S. Haldar, K. Horback, C. Tom, L. Zhou, H. Meyerson, N. Singh. Prion protein regulates iron transport by functioning as a ferrireductase. *J. Alzheimers Dis.* **35:3**, 541-52 (2013).
47. J.A. Duce, A. Tsatsanis, M.A. Cater, S.A. James, E. Robb, K. Wikhe, S.L. Leong, K. Perez, T. Johanssen, M.A. Greenough, H.H. Cho, D. Galatis, R.D. Moir, C.L. Masters, C. McLean, R.E. Tanzi, R. Cappai, K.J. Barnham, G.D. Ciccotosto, J.T. Rogers, A.I. Bush. Iron-export ferroxidase activity of β-amyloid precursor protein is inhibited by zinc in Alzheimer's disease. *Cell.* **142:6**, 857-67 (2010).
48. M.A. Smith, P.L.R. Harris, L.M. Sayre, G. Perry. Iron accumulation in Alzheimer disease is a source of redox-generated free radicals. *Proc. Natl. Acad. Sci. USA*. **94:18**, 9866-68 (1997).
49. N.T. Watt, D.R. Taylor, T.L. Kerrigan, H.H. Griffiths, J.V. Rushworth, I.J. Whitehouse, N.M. Hooper. Prion protein facilitates uptake of zinc into neuronal cells. *Nat. Commun*. **3**, 1134 (2012).
50. A. Ghosh, K.P. Giese. Calcium/ calmodium-dependent kinase II and Alzheimer's disease. *Mol. Brain.* **8:1**, 78 (2015).
51. S. Hannaoui, S.Y. Shim, Y.C. Cheng, E. Corda, S. Gilch. Cholesterol balance in prion disease and Alzheimer's disease. *Viruses.* **6:11**, 4505-35 (2014).

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