Looking for novel functions of tau

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Abstract
The lack or excess of the protein tau can be deleterious for neurons. The absence of tau can result in retarded neurogenesis and neuronal differentiation, although adult mice deficient in tau are viable, probably because of the compensation of the loss of tau by other MAPs (microtubule-associated proteins). On the contrary, the overexpression of tau can be toxic for the cell. One way to reduce intracellular tau levels can be achieved by its secretion through microvessicles to the extracellular space. Furthermore, tau can be found in the extracellular space because of the neuronal cell death occurring in neurodegenerative disorders such as Alzheimer’s disease. The presence of toxic extracellular tau could be the mechanism for the spreading of tau pathology in these neurodegenerative disorders.

Introduction
The protein tau was originally found together with other brain MAPs (microtubule-associated proteins) bound to isolated brain microtubules [1]. Tau, like other MAPs, stabilizes microtubule polymers [2], suppresses microtubule dynamics [3] and probably because of these effects is able to promote cytoplasmic extensions or neuritogenesis [4,5]. In addition, tau (which is a sticky protein) can bind to other proteins, some of which are components of the plasma membrane [2,6,7].

By dividing the protein tau into two halves, for example the molecule corresponding to the largest human tau isoform that is present in the central nervous system, two fragments are obtained. One part contains the N-terminal residue and 200 amino acids, whereas the other part comprises a fragment from amino acid 201 to the last residue at the C-terminus, amino acid 441. We know that the N-terminal fragment shows some variability from organism to organism, whereas the C-terminal fragment is a well-conserved region [8]. Tau binding to microtubules takes place through the C-terminal fragment [9], but the association of tau with the plasma membrane is through the N-terminal region [2,6,7,10].

Tau function
To decipher tau function(s), mice lacking or overexpressing tau were raised and their phenotypes were compared with wild-type mice. Tau-deficient mice are viable and no major phenotypic differences have been found with respect to wild-type mice [11,12]; however, tau-deficient mice show some minor specific characteristics.

Key words: calmodulin, extracellular tau, tau secretion, ventral dentate gyrus.
Abbreviations used: AD, Alzheimer’s disease; CaM, calmodulin; CSF, cerebrospinal fluid; MAP, microtubule-associated protein.
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Tau-deficient mice
Tau-deficient mice [12] showed a delayed differentiation and migration of new neurons generated at the dentate gyrus when compared with wild-type mice [13]. The mechanism for this delayed neurogenesis was analysed by looking at gene expression in the hippocampus, in the presence or absence of tau.

Tau deficiency leads to the up-regulation of BAF-57, a protein involved in neuron-specific gene repression [14]. A possible consequence could be the delayed axonogenesis observed in tau-deficient neurons that could explain the delayed differentiation observed in tau-deficient mice during dentate gyrus neurogenesis. On the contrary, tau-deficiency leads to the down-regulation of the neuronal gene calbindin [15]. Since tau is mainly a cytoplasmic protein, we have tested if the action of tau in the regulation of calbindin expression could be through another protein present in both compartments, cytoplasm and nucleus. Our results have shown that this particular protein could be CaM (calmodulin). Our data have indicated that, in the absence of tau, the amount of nuclear CaM decreases, whereas when the amount of tau increases nuclear CaM increases [15]. Since nuclear CaM is an activation co-factor of transcriptional factors such as the BAF complex, which is involved in neuron-specific gene expression [16], its nuclear presence could down-regulate the expression of genes, such as calbindin, which is regulated by the BAF complex [17].

Mice overexpressing human tau
Mice overexpressing human tau carrying the frontotemporal mutations G272V, P301L and R408W (tauFLW) were also generated [18]. These mice showed a decrease in the dentate gyrus ventral blade [19]. This degeneration was correlated with a change in behavioural mood, as determined in the Porsolt swim test [19]. The possible mechanism for ventral
dentate gyrus degeneration because of the tau overexpression is now under study. One of the possibilities, which are under study, is that neurodegeneration could be the consequence of tau phosphorylation. Thus, since GSK3 is a main tau kinase, a double transgenic mouse overexpressing GSK3 and human tauV19 was generated. However, in this mouse model, degeneration was mainly found first at the dorsal dentate gyrus [20]. Our studies suggested that tau modified by GSK3 is mainly toxic at the dorsal dentate gyrus, although the absence of tau, the overexpression of GSK3 could also produce some toxic effects, probably because of the additional toxicity of other substrates modified by GSK3 [21]. Later on, in aged double-transgenic (GSK3–tauV19) mice, the degeneration of the ventral dentate gyrus also takes place, as previously indicated. In summary, an overexpression of tau (or phospho-tau) could be toxic for newborn neurons in the dentate gyrus.

Proteostasis of tau protein
In the previous section, we have indicated that an overexpression of tau protein could be toxic for a developing neuron. Intracellular levels of a protein are the result of its synthesis, degradation or secretion. Protein secretion through microvesicles has been indicated to decrease intracellular levels of some proteins [22] and we therefore analysed if tau protein used this mechanism. Tau was expressed in cultured non-neuronal cells at different expression levels. When overexpressed, tau was detected extracellularly in the culture medium in association with microvesicles. These results suggest that tau overexpression can result in its secretion via membrane vesicles [23].

Extracellular tau and AD (Alzheimer’s disease)
AD is characterized by the presence of two aberrant structures, senile (amyloid) plaques and neurofibrillary tangles, present in the brain of the patients, together with a clear loss of neurons. It is well known that the main component of neurofibrillary tangles, present inside or outside of neurons, is the protein tau [24]. A possible mechanism for the presence of extracellular tau is neuronal death. During neuron loss, intracellular tau (in monomeric or aggregated form) can be found in the extracellular space. In this way, an inverse correlation was observed between the number of extracellular tau aggregates and the number of living neurons in the hippocampus of AD patients [25]. In previous studies, it was found that this extracellular tau can be toxic for neurons and that it can play a role in the spreading of tau pathology found in AD [26,27].

This tau-dependent toxicity occurs when extracellular tau interacts with cell membrane receptors identified as M1 and M3 muscarinic receptors [27]. As a consequence of this interaction, an increase in intracellular calcium, which could be toxic for the cell, takes place [28]. Recent results showed that tissue non-specific alkaline phosphatase promotes the neurotoxic effects of extracellular tau, since unphosphorylated tau is the toxic agent [29].

In addition to this possible mechanism for the spreading of tau pathology, another mechanism has been proposed. This involves the secretion through microvesicles of aggregated tau, which can be endocytosed by surrounding cells, suggesting a prion-protein-like behaviour, which can be toxic [30,31].

Recently, it has been indicated that during the progression of AD both mechanisms could take place. By looking at the CSF (cerebrospinal fluid) of AD patients at different stages of the disease, a higher proportion of vesicle-tau protein in the CSF of the patients at earlier stages of the disease was found, whereas the proportion of naked tau in CSF increases during the development of the disease in patients [32].

In summary, the lack or an excess of tau protein could be deleterious for a neuron.

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References