

Neurofibrillary Degeneration in Progressive Supranuclear Palsy and Corticobasal Degeneration: Tau Pathologies with Exclusively “Exon 10” Isoforms

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Abstract: Pathological tau proteins that constitute the basic matrix of neuronal inclusions observed in numerous neurodegenerative disorders are disease specific. This is mainly the consequence of the aggregation of specific sets of tau isoforms according to the diseases, i.e., six isoforms in Alzheimer’s disease (AD) and exclusively the three tau isoforms lacking the corresponding sequence of exon 10 (E10–) in Pick’s disease (PiD). By using antibodies specific to the different tau isoforms and one- and two-dimensional gel electrophoresis followed by western blots, we demonstrate herein a third group of neurodegenerative disorders characterized by intraneuronal inclusions exclusively constituted of tau isoforms containing the sequence corresponding to exon 10, progressive supranuclear palsy (PSP) and corticobasal degeneration (CBD). Together, tau isoforms with exon 10 clearly differentiate three groups of neurodegenerative diseases: AD, PiD, and PSP/CBD. For each group, the neuropathological and clinical phenotypes are most likely related to specific sets of tau isoforms expressed by the vulnerable neuronal populations. The recently described mutations of the tau gene responsible for familial frontotemporal dementias also support this hypothesis. **Key Words:** Neurofibrillary degeneration—Tau proteins—Progressive supranuclear palsy—Corticobasal degeneration—Alzheimer disease—Antibodies—Tau gene. *J. Neurochem.* **72**, 1243–1249 (1999).

Paired-helical filaments (PHFs) are the hallmark of degenerating neurons in Alzheimer’s disease (AD). Abnormally phosphorylated tau proteins, named PHF-tau (Goedert et al., 1995) or pathological tau proteins (Delacourte and Buée, 1997), form these pathological filaments. In many other neurodegenerative diseases, tau-positive inclusions are also observed, including corticobasal degeneration (CBD), progressive supranuclear palsy (PSP), amyotrophic lateral sclerosis/parkinsonism-dementia complex of Guam, myotonic dystrophy, postencephalitic parkinsonism, and Pick’s disease (for review, see Delacourte and Buée, 1997). Aggregates of tau proteins are the basic biochemical substrates for neurofibrillary degeneration. Most important, the presence of neurofibrillary degeneration in neocortical asso-

ciation areas correlates to cognitive impairment, whatever the diseases (Bierer et al., 1995; Delacourte and Buée, 1997; Duyckaerts et al., 1997; Delacourte et al., 1998b).

Tau are phosphoproteins belonging to the family of microtubule-associated proteins. In human adult brain, alternative mRNA splicing of a single gene transcript gives six tau isoforms (Goedert et al., 1995; Delacourte and Buée, 1997). These isoforms differ by the insertion of 29 or 58 amino acids, corresponding to the exon 2 or exons 2 and 3 sequences, near the amino-terminus, and/or by the insertion at the proximal half of the carboxy-terminus of an additional microtubule-binding motif corresponding to the exon 10 sequence. The sequence corresponding to exon 10 increases the affinity of tau toward tubulin 40 times (Panda et al., 1995) and is thus considered as playing an important role in the neuronal microtubule network.

The electrophoretic profiles of pathological tau proteins differ from normal tau and these profiles are disease specific, as revealed by phosphodependent tau antibodies. In particular, we have shown in AD that abnormally phosphorylated tau from PHFs of neurofibrillary tangles, named PHF-tau, have an electrophoretic profile characterized by three main bands (tau 55, 64, and 69) and an additional 74-kDa band, resulting from a specific distribution of the six tau isoforms (Sergeant et al., 1997). In Pick’s disease, phosphorylated tau from Pick bodies are resolved as two major bands (tau 55 and 64 kDa) and a minor 69-kDa band, which are corresponding to the three tau isoforms without the exon 10 corresponding se-

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Abbreviations used: AD, Alzheimer’s disease; CBD, corticobasal degeneration; CPK, creatine phosphokinase; 1-D and 2-D, one- and two-dimensional, respectively; PHF, paired-helical filament; PSP, progressive supranuclear palsy.

quence (E2-, E3-, E10-; E2+, E3-, E10-; E2+, E3+, E10-) (Delacourte et al., 1998a). Also, we demonstrated that PSP (Flament et al., 1991; Vermersch et al., 1994) and CBD (Buée-Scherrer et al., 1996b; Delacourte et al., 1997) possess a specific tau signature, with two main tau 64- and 69-kDa bands and a minor 74-kDa band. These diseases are not completely well characterized from a clinical point of view, because they are considered as Parkinson "plus" diseases (Litvan et al., 1997a,b) or, for others, as frontotemporal dementias of the Pick complex (Kertesz and Munoz, 1996). The relationship between PSP and CBD, if any, is also poorly understood from a clinical and etiopathogenic point of view (Litvan et al., 1997a,b). Moreover, recently described mutations on the exon 10 of tau gene are directly responsible for familial neurodegenerative disorders (Murrell et al., 1997; Hutton et al., 1998). In the present study, we have addressed the problem of the isoform content of tau proteins aggregating in PSP and in CBD, to have a better understanding of the basic mechanisms of NFD.

MATERIALS AND METHODS

Patients

AD patients met the NINCDS-ADRDA criteria and were histopathologically confirmed for the definite diagnosis of AD, as already reported (Sergeant et al., 1997; Delacourte et al., 1998a,b). The PSP and CBD cases studied here have already been presented elsewhere (Flament et al., 1991; Vermersch et al., 1994; Buée-Scherrer et al., 1996b; Delacourte et al., 1997). Brain tissues were obtained at autopsy in accordance with the local ethics committee and the French Caillavet Law no. 76-1181 (December 22, 1976); postmortem delays were <48 h and the PSP and CBD cases studied had clear-cut clinical features, as described by Litvan et al. (1997a,b).

Monoclonal and polyclonal antibodies

AD2 is a protein A-purified monoclonal antibody obtained by immunizing mice with a crude PHF preparation purified from an AD brain. It specifically recognizes phosphorylated serines 396 and 404 (numbering according to the longest human brain tau isoform) in the carboxy-terminal part of tau (Buée-Scherrer et al., 1996a). AD2 was used at 0.2 µg/ml in Tris-buffered saline containing 0.05% (vol/vol) Tween 20, as described by Sergeant et al. (1997).

Phosphodependent tau antibodies were also used such as S199P (Delacourte et al., 1998a), 12E8 (Seubert et al., 1995), and AT100 (Hoffmann et al., 1997), which bind to tau proteins phosphorylated at serine-199, serine-262, threonine-212, and serine-214.

Isoform-specific tau polyclonal antibodies Tau-E2, Tau-E3, and Tau-E10 were obtained by immunizing New Zealand rabbits with synthetic peptides (Neosystem, France) corresponding to the first 16 amino acids encoded by exon 2 and exon 3 and to the first 10 amino acids encoded by exon 10. These polyclonal antibodies are phosphorylation independent; their conditions of use and their specificity have been described previously in detail (Sergeant et al., 1997; Delacourte et al., 1998a).

One-dimensional (1-D) and two-dimensional (2-D) gel electrophoresis

For 1-D and 2-D gel electrophoresis, frontal brain tissue samples were freshly homogenized in Laemmli sample buffer (1:10) containing 5% (wt/wt) sodium dodecyl sulfate. The protein concentration was determined using the bicinchoninic acid protein assay (Pierce), and brain tissue homogenates were aliquoted and frozen at -80°C until used. Each aliquot was used not more than twice. A same amount of total brain protein (90 µg) was loaded, and 1-D and 2-D gels were performed as already described (Sergeant et al., 1997; Delacourte et al., 1998a). Tau isoform distribution was investigated by using 7.5–15% polyacrylamide gradient gels. For 2-D gels, isoelectric points (pI) were determined by using the carbamylated creatine phosphokinase (CPK) (molecular mass = 45 kDa; pI range, 4.9–7.1) of the Carbamylate 2-D gels calibration kit (Pharmacia).

Western blots

Proteins resolved on 1-D and 2-D gels were transferred onto nitrocellulose membranes (Hybond ECL, 0.45 µm pore size, Amersham) for 90 min at 0.8 mA/cm², using an LKB Multiphor II Nova Blot (Pharmacia). Proteins were reversibly stained with Ponceau Red, to check the quality of the resolution for the 1-D and 2-D blots and to visualize the carbamylated CPK on 2-D blots. Blocking was performed in Tris-buffered saline (0.05%, vol/vol) with Tween 20 containing 5% dry milk and blots were incubated for 2 h at room temperature with the antibodies. Monoclonal and polyclonal antibodies were detected with horseradish peroxidase-labeled sheep anti-mouse and anti-rabbit immunoglobulins both adsorbed with human serum proteins (Sigma Immuno Chemicals), respectively. Immunoreactive proteins were revealed using the ECL western blotting system (Amersham).

RESULTS

The phosphorylation state of pathological tau in CBD and PSP

We recently demonstrated that pathological tau proteins in Pick's disease are differently phosphorylated, when compared with those in AD. Thus, phosphorylated serine-262 detected by 12E8 antibody is absent in tau proteins from Pick bodies, whereas all of the pathological tau proteins engaged in the formation of Pick bodies are immunostained by AD2 (Delacourte et al., 1998a). In the same way, using various phosphodependent antibodies, we first verified that pathological tau protein profiles from PSP or CBD were not resulting from the presence or absence of specific phosphorylated sites.

As shown in Fig. 1, the immunostaining pattern was similar with phosphodependent antibodies such as S199P (serine-199), 12E8 (serine-262), AT100 (threonine-212 and serine-214), and AD2 (serine-396 and serine-404). The characteristic doublets of PSP and CBD (tau 64 and 69) were similarly immunostained by these phosphodependent antibodies. However, in all the cases analyzed so far (five PSP and five CBD), the immunostaining was always weaker in PSP cases. Moreover, in PSP cases, AT100 antibody showed the lowest immunoreactivity for pathological tau proteins (Fig. 1; PSP, lane AT100). Therefore, these results show that the specific tau profile in PSP and CBD is

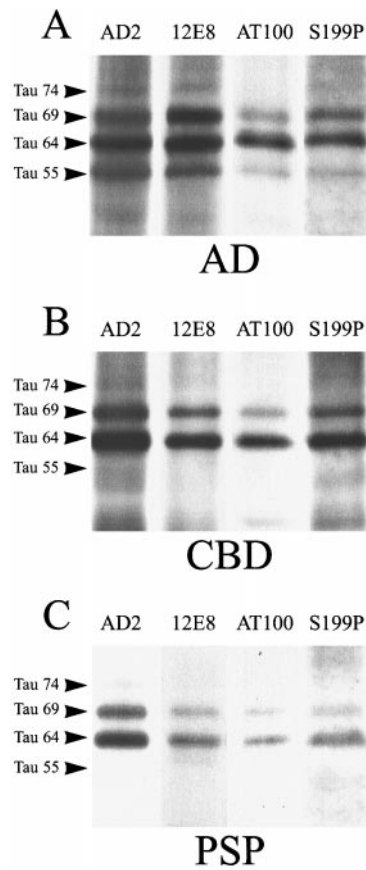


FIG. 1. Immunoblotting of pathological tau proteins in AD (A), CBD (B), and PSP (C). Total brain tissue homogenates from AD, CBD, and PSP patients were loaded for gel electrophoresis (90 μ g of protein/well). Phosphorylated tau proteins at serine-396 and serine-404 (numbering according to the longest tau isoform) were immunolabeled with AD2. In AD, they correspond to the typical four PHF-tau components at 55, 64, 69, and 74 kDa (arrowheads). These PHF-tau components were also detected by antibodies 12E8 (phosphorylated serine-262), AT100 (phosphorylated threonine-212 and serine-214), and S199P (phosphorylated serine-199). Note that the staining for all diseases is weaker with AT100. The 74-kDa PHF-tau band was not visible as well as the vertical smear. In both CBD and PSP, the phosphorylated tau components are made of two major components at 64 and 69 kDa and a less immunoreactive 74-kDa component very faintly detected in PSP samples. All these antibodies reacted with the main doublet of PSP and CBD (lanes AD2, 12E8, AT100, and S199P). In PSP, the phosphorylated tau components are less intensely labeled and the vertical smear is only observed in the S199P lane.

likely to arise from variables other than phosphorylation. Consequently, we undertook a western blot analysis using isoform-specific tau antibodies, to determine whether the isoform content of pathological tau in PSP and CBD could be responsible for the specific electrophoretic profile observed in these disorders.

Tau isoform content in AD, PSP, and CBD: 1-D analysis

Tau isoform content of aggregated tau in PSP and CBD was determined by using the same strategy as Sergeant et al. (1997) and Delacourte et al. (1998a).

Pathological tau isoforms in AD. Tau isoforms engaged in the formation of PHFs have already been described (Sergeant et al., 1997), but they were analyzed in parallel to our study of tau isoform distribution in PSP and CBD, for comparison and to precisely align the tau isoform electrophoretic profile. The typical AD PHF-tau components of 55, 64, and 69 kDa and the additional band of 74 kDa were immunolabeled by antibody AD2 (Fig. 2; AD, lane AD2). A vertical smear was always associated with the detection of PHF-tau in AD brain tissue homogenates, corresponding to undissociated tau polymers.

Tau isoform distribution was further investigated using isoform-specific tau antibodies. Antibody Tau-E10, specific for tau proteins with the exon 10 corresponding sequence, immunostained the 64, 69, and 74 PHF-tau bands and intensely stained the vertical smear (Fig. 2A; AD, lane Tau-E10). The Tau-E10-immunoreactive smear was very similar to the one observed with antibody AD2. Antibody Tau-E2, specific for tau proteins with the exon 2 corresponding sequence, immunolabeled PHF-tau 64, 69, and 74 kDa (Fig. 2A; AD, lane Tau-E2). Lower molecular weight tau-immunoreactive bands were also detected with antibody Tau-E2, corresponding to normal tau proteins and followed by catabolic products. Antibody Tau-E3, which specifically recognizes the corresponding sequence of exon 3, detected the PHF-tau bands at 69 and 74 kDa (Fig. 2A; AD, lane Tau-E3). It is interesting that no smear was observed with Tau-E2 and Tau-E3 antibodies and the 74-kDa band was less visible with the Tau-E2 antibody.

Pathological tau isoforms in CBD. AD2 immunolabeling showed the typical tau doublet (tau 64 and 69) and a faintly immunoreactive 74-kDa band (Fig. 2B; CBD, lane AD2), as already described (Delacourte et al., 1997a). A vertical smear was also visible. Antibody Tau-E10 immunodetection was similar to AD2 staining, and was characterized by the detection of the 64-, 69-, and 74-kDa pathological tau bands as well as the vertical smear (Fig. 2B; CBD, lane Tau-E10). Antibody Tau-E2 detected strongly the pathological tau 69 band, to a lesser extent the pathological tau 74 band, and four bands in the range of 45–65 kDa, but none of the latter bands comigrated with the pathological tau doublet (Fig. 2B; CBD, lane Tau-E2). As already described by (Delacourte et al. (1998a), the four bands correspond to four normal tau proteins with the exon 2 corresponding sequence and this was verified by 2-D electrophoresis (data not shown). Antibody Tau-E3 was able to clearly stain the pathological tau band at 74 kDa, but also two bands at 62 and 65 kDa, both of which comigrated with the two uppermost bands stained by Tau-E2 (Fig. 2B, Tau-E2 and Tau-E3 lanes).

Pathological tau isoforms in PSP. As shown in Fig. 2C, results between PSP and CBD were very similar, but immunodetection was weaker in PSP with phosphodependent and Tau-E10 antibody. Indeed, tau 74 immunodetection by AD2, Tau-E2, and Tau-E3 (Fig. 2C; AD2,

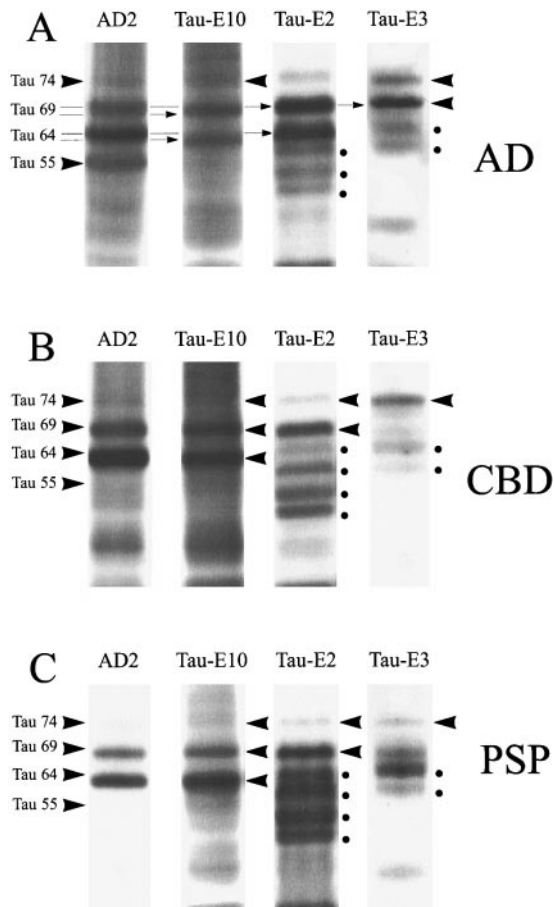


FIG. 2. Tau isoform distribution in PHF-tau of AD and pathological tau of CBD and PSP. On a single sodium dodecyl sulfate-polyacrylamide gel electrophoresis, similar amounts of proteins (90 μ g) from AD (A), CBD (B), and PSP (C) brain tissue homogenates were loaded and subjected to immunolabeling with isoform-specific tau antibodies. AD2 was used in the first lane to point the four PHF-tau components of AD (A: tau 55, 64, 69, and 74) and pathological tau components of both CBD and PSP (B and C: Tau 64 and 69) and the less immunoreactive 74-kDa component (indicated by arrowheads). Tau protein isoforms containing the exons 10, 2, and 3 corresponding sequences were labeled with the polyclonal antibodies Tau-E10, Tau-E2, and Tau-E3, respectively (A, B, and C: lanes Tau-E10, Tau-E2, and Tau-E3). Tau isoforms labeled by the isoform-specific tau antibodies and comigrating with the pathological tau components are indicated by arrowheads. Normal tau proteins are indicated by dots. Note that antibody Tau-E10 stained PHF-tau components of 64, 69, and 74 kDa in AD and pathological tau components 64 and 69 kDa in both CBD and PSP (A, B, and C: lanes Tau-E10, indicated by arrowheads). Note also that the staining was always associated with vertical smears, which were not visible with Tau-E2 and Tau-E3 antibodies.

Tau-E3) was extremely faint but visible with the naked eye.

2-D western blot analysis of AD PHF-tau

2-D gel electrophoresis allows the separation of normal tau from pathological tau, as pathological tau from autopsied brain tissue remains hyperphosphorylated and is thus acidic, even after a long postmortem delay. Con-

versely, normal tau are rapidly dephosphorylated during the same postmortem delay and are thus basic (Sergeant et al., 1997; Delacourte et al., 1998a). First, AD2 revealed the characteristic pattern of pathological tau of PSP and CBD, which is a pathological tau doublet as previously shown (Delacourte et al., 1997). We found also that pathological tau proteins from CBD were more intensely immunostained and more acidic than those of PSP (Fig. 3) but less acidic than those of AD (Delacourte et al., 1997). The 74-kDa spot was weak but readily detected. On similar blots, antibody Tau-E10 reacted strongly with the tau 64 and 69 doublet of PSP and CBD (Fig. 3) and the 74-kDa spot was weaker. Thus, the pattern of immunoreactivity obtained with Tau-E10 matched that obtained with AD2. Tau-E2 antibody immunodetected the pathological tau 69 and 74 spots, but antibody Tau-E3 only detected the pathological tau 74 spot (not shown).

Together, these observations led us to propose a synopsis representing the isoform content of pathological tau involved in PSP and CBD neurofibrillary degeneration (Fig. 4).

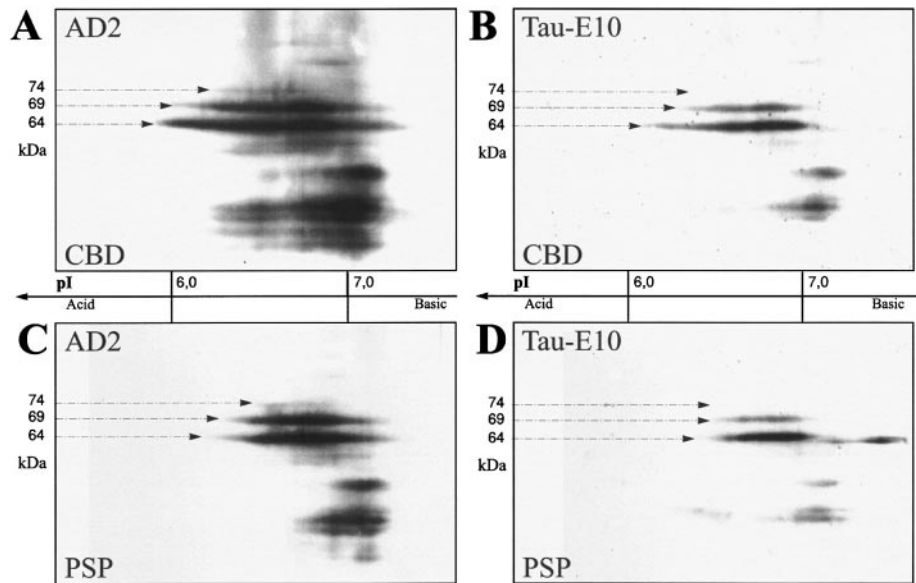
DISCUSSION

1-D and 2-D gel electrophoresis followed by western blotting is a powerful approach to analyze pathological tau proteins, which are the structural components of neuronal inclusions in AD and numerous other neurodegenerative disorders (Delacourte and Buée, 1997). Using specific antibodies against exons 2, 3, and 10 corresponding sequences of tau isoforms, we were able to determine the precise isoform content of the PHF-tau electrophoretic components in AD (Sergeant et al., 1997) as well as the pathological tau components of Pick's disease (Delacourte et al., 1998a). In the present study, we describe the tau isoforms that aggregate in PSP and in CBD, two neurodegenerative disorders that generally start with extrapyramidal signs and provoke severe cognitive impairments, with a concomitantly severe tau pathology in cortical and subcortical areas.

Phosphodependent antibodies did not differentiate between the phosphorylation sites of PHF-tau in AD and those in PSP and in CBD. The very specific probe AT100 that stains PHF-tau in AD also immunodetected pathological tau in CBD and with a lower immunoreactivity, pathological tau in PSP, as already described by Schmidt et al. (1996). These results show that this pathological epitope resulting from the phosphorylation of both threonine-212 and serine-214 (Hoffmann et al., 1997) is present in different neurodegenerative disorders.

Normal tau proteins derived from postmortem brain tissue samples have molecular masses ranging from 45 to 65 kDa, and thus, some of the tau isoforms seem to comigrate with pathological tau components (Sergeant et al., 1997; Delacourte et al., 1998a). 2-D gels enable us to distinguish normal tau from pathological tau, as normal tau are dephosphorylated during postmortem delay and thus have basic isoelectric points whereas PHF-tau are

FIG. 3. 2-D characterization of the exon 10 containing tau isoforms in CBD and PSP pathological tau components. Pathological tau components from CBD and PSP brain tissue homogenates were resolved by 2-D gel electrophoresis and immunolabeled with either AD2 (**A** and **C**) or the Tau-E10 polyclonal antibody (**B** and **D**). 2-D immunoblots are oriented with the basic origin on the right and the acid end on the left. pI 6.0 and 7.0 are indicated on the x-axis. The typical doublet of pathological tau components was detected in both CBD and PSP samples (dotted arrows in **A** and **C**). Note that in CBD, pathological tau were more acidic and that the 74 kDa was well visualized. Antibody Tau-E10 detected the two pathological tau components in CBD and PSP (**B** and **D**), which coincided with AD2 immunolabeling. Note that the 74-kDa band was almost not visible and that no other spots were detected by using Tau-E10 antibody.



more resistant to dephosphorylation and remain phosphorylated during postmortem delay. Thus, PHF-tau are acidic and may be resolved by the 2-D gel pH gradient (Delacourte and Buée, 1997; Sergeant et al., 1997; Delacourte et al., 1998a).

Isoform-specific tau antibodies used in 1-D and 2-D western blots demonstrated that tau isoforms aggregating in PSP and in CBD exclusively contain the exon 10 corresponding sequence. Therefore, three different isoforms are present, as shown in Fig. 4. The three E10+ tau isoforms were present, as they were detected by antibodies Tau-E2, Tau-E3, and our specific Tau-E10 antibody (Delacourte et al., 1998a). The only discrepancy is that antibody Tau-E2, which detected tau 69, weakly stained tau 74. In fact, tau 74 should be immunodetected, as exon 2 sequence is an obligatory component of the longest tau isoform and exon 3 is always spliced together with exon 2 (Goedert et al., 1995; Sergeant et al., 1997). However, we note that our antibody Tau-E2, as well as another antibody that recognizes an exon 2 corresponding sequence, named 304 (Goedert et al., 1992), bind with lower affinity to tau isoforms containing both exon 2 and 3 corresponding sequences compared with tau isoforms with the exon 2 sequence alone (Delacourte et al., 1998a). We conclude that exon 3 sequence is likely to provoke a conformational change that decreases the binding of Tau-E2 antibody and thus change the affinity for its epitope. Also, E10- tau isoforms are not present in the pool of pathological tau in PSP and CBD, as (1) the smallest tau 55 (E2-, E3-, E10-) was not detected with either phosphodependent or isoform-specific tau antibodies, which are phosphorylation-independent antibodies, (2) Tau-E2 did not label

the tau 64 band (E2+, E3-, E10-), observed in PHF-tau from AD, and (3) Tau-E3 did not label the tau 69 variant (E2+, E3+, E10-), which is also visible in AD (see Fig. 4).

Together, our results show that tau isoforms with or without the sequence corresponding to exon 10 can distinguish three types of neurodegenerative disorders. In AD, both E10+ and E10- isoforms are present (Sergeant et al., 1997). In PSP and in CBD, E10+ tau isoforms are exclusively present (the present study), but in Pick's disease, the only tau isoforms observed in Pick bodies lack the exon 10 corresponding sequence (Delacourte et al., 1998a).

The most likely explanation for the exclusive presence of E10+ tau isoforms in PSP and CBD lesions is that the subsets of neurons that degenerate in these diseases normally and exclusively express tau isoforms with the exon 10 corresponding sequence. When this specific group of neurons degenerate, E10+ tau isoforms aggregate to yield the characteristic immunohistochemical pattern containing the main doublet of tau 64 and 69. Indeed, it has been shown that the affected cells in PSP and in CBD are small pyramidal cells of layers II and III (Buée-Scherrer et al., 1996b), whereas the cells affected in Pick's disease are mainly smaller interneurons of layers II-III and granule cells of the dentate gyrus (Hof et al., 1994; Delacourte et al., 1998a).

In conclusion, our results show for the first time that tau isoforms with and without the exon 10 corresponding sequence can precisely distinguish neuronal populations and neurodegenerative disorders. The polypeptide sequence corresponding to exon 10 plays an important role in maintaining the stability of microtubules, as it poten-

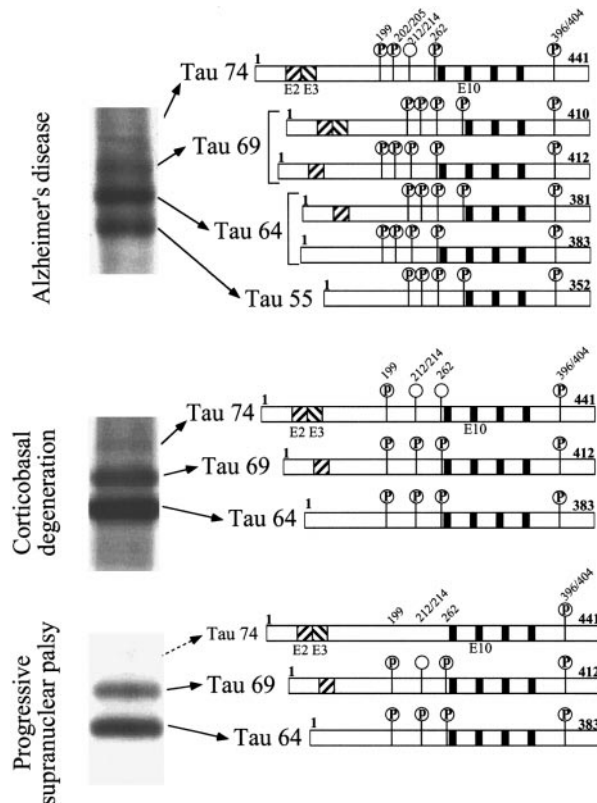


FIG. 4. Synopsis of the distribution of tau protein isoforms and their respective phosphorylated sites studied in PHF-tau of AD and pathological tau of both CBD and PSP. The six tau isoforms are engaged in formation of the PHF-tau electrophoretic profile of AD, whereas three tau isoforms with the exon 10 translated sequence are found in the doublet of pathological tau proteins from CBD and PSP. Isoform-specific anti-tau antibodies used in the present study enable us to precisely give the correspondence between each electrophoretic band of pathological tau proteins detected by phosphodependent anti-tau antibodies, such as AD2, and the tau isoform distribution (indicated by arrows). As already described (Sergeant et al., 1997; Delacourte et al., 1998a), tau 74 in AD corresponds to the longest tau isoform (exons E2+, E3+, E10+). Both tau isoform E2+, E10- and tau isoform E2+, E3-, E10- were found in the tau 69 component. The tau 64 was also made of two tau isoforms, tau isoform E2+, E3-, E10- and tau isoform E2-, E3-, E10+. The tau 55 component corresponded to the shortest tau isoform lacking exons E2, E3, and E10. Tau pathology in CBD and PSP results exclusively of E10+ tau isoforms. The tau 74 pathological tau component corresponds to the longest tau isoform, tau 69 corresponds to the tau isoform with exons 2 and 10, and the tau 64 is the tau isoform with exon 10 alone. Note that in PSP, the 74-kDa component is less represented and therefore indicated by a dotted arrow. Antibodies raised against specific phosphorylated sites on tau proteins were used: S199P for phosphorylated serine-199, AT100 for both phosphorylated threonine-212 and serine-214, 12E8 for phosphorylated serine-262, and AD2 for both phosphorylated serine-396 and serine-404. The phosphorylated sites detected on either PHF-tau of AD or pathological tau of CBD or PSP are represented by an encircled "P."

tiates the interaction of tau with tubulin (Panda et al., 1995). This crucial function of the exon 10 corresponding sequence could explain why mutations dysregulating the translation of exon 10 are responsible for severe

familial neurodegenerative disorders (Murrell et al., 1997; Hutton et al., 1998; Spillantini et al., 1998). The deleterious effect of such mutations is likely to be most dramatic in neurons that express exclusively E10+ tau isoforms. Indeed, as described by Murrell et al. (1997), these familial pathologies have a phenotype similar to PSP or to CBD, with the same straight filaments and the characteristic PSP/CBD electrophoretic doublet. Moreover, polymorphisms in the exon 10 region of tau gene seem to be linked to sporadic PSP (Conrad et al., 1997).

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