

The coexistence of Alzheimer's disease and Creutzfeldt-Jakob disease in a patient with dementia of long duration

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Received March 13, 1992/Revised, accepted May 23, 1992

Summary. We report here a 75-year-old-male with a slowly progressive dementia of 5-year duration along with a rapid exacerbation of symptoms in the terminal 3 months. Neuropathological examinations revealed findings consistent with conspicuous Alzheimer's disease and mild Creutzfeldt-Jakob disease (CJD). The plaque amyloid was exclusively composed of β -protein. The immunohistochemistry of prion protein using hydrolytic autoclaving pretreatment showed diffuse gray matter stainings in the sections of both the cerebral and cerebellar cortices. This method was thus considered useful in confirming the diagnosis of CJD for this case.

Key words: Alzheimer's disease – Creutzfeldt-Jakob disease – Prion protein – Immunohistochemistry

The coexistence of Alzheimer's disease (AD) and Creutzfeldt-Jakob disease (CJD) is a rare phenomenon [3, 11]. We report here a patient who clinically demonstrated a slowly progressive dementia of 5-year duration as well as a rapid exacerbation of symptoms in the terminal 3 months, and neuropathologically suffered from both AD and CJD.

Case history

The patient was a 75-year-old man whose memory was beginning to fail during the autumn of 1986. In the next year, he often lost his way in familiar surroundings. Outpatient medical evaluations in March 1988 revealed a moderate defect in cognitive function, and a head computerized tomography (CT) showed moderate diffuse cerebral atrophy. He had no family history of dementing illnesses. In the winter of 1990, he was placid, hypokinetic, and could not distinguish right from left with certainty. In August 1991, his feeding declined, his gait became unstable, and he was restless when sitting. Within the subsequent 2 months his gait disturbance

and poor feeding habits gradually became aggravated until he could no longer walk or eat independently.

Upon admission to Imazu Red Cross Hospital in October 1991, he was described as irritable and confused with a striking startle response to any acoustic or touch stimuli and demonstrated myoclonic jerks in his arms. His condition continued to deteriorate in the hospital, with the spread of myoclonic jerks to the legs and the evolution of his mental deterioration to a stupor.

A head CT scan showed moderate to marked diffuse cerebral atrophy. An electroencephalogram showed a marked generalized background slowing (3–5 Hz) and repetitive, bilaterally synchronous sharp and slow wave complexes with high voltage. The patient died in November 1991.

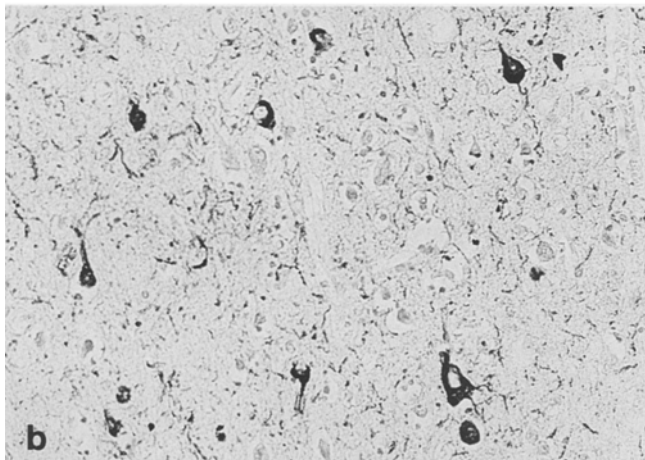
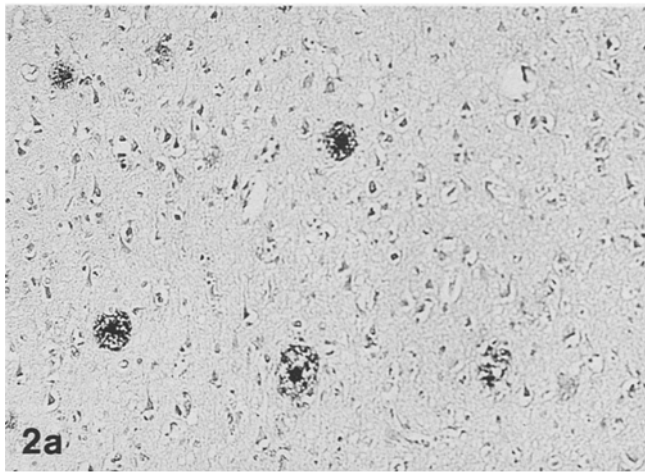
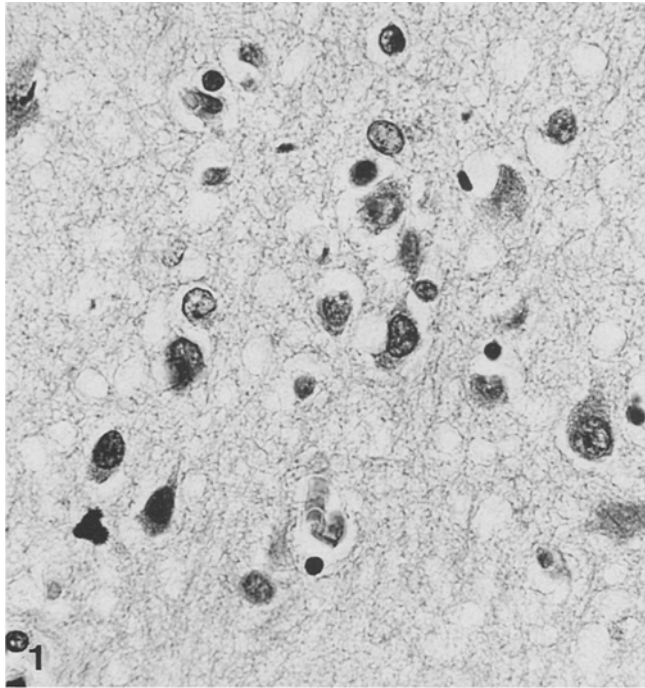
Results

An autopsy was performed 10 h after death. The brain weighed 1025 g and exhibited diffuse moderate cerebral atrophy, but cerebellar atrophy was less distinct. A diffuse mild shrinkage of whole cerebrum was noted on inspection of fixed slices. Microscopic examinations of the frontal, parietal, temporal, and occipital neocortex showed a mild to moderate neuronal loss, reactive astrocytosis and spongiform changes (Fig. 1).

Spongiform changes in the neocortex were mainly observed in the pyramidal cell layers. In the hippocampus, mild neuronal loss and astrocytosis were found, but no spongiform changes were apparent. Mild and focal spongiform changes and astrocytosis were found in the caudate nucleus, putamen, thalamus, and the molecular layer of the cerebellar cortex. A mild and focal thinning and astrocytosis were also observed in the granular cell layer of the cerebellar cortex. Numerous neuritic plaques and neurofibrillary tangles (NFTs) were seen in silver-stained sections of the frontal, parietal, occipital, and temporal cortex including the hippocampus. Various numbers of neuritic plaques were also found in the caudate nucleus, putamen, and thalamus. In addition, several NFTs were found in the nucleus basalis of Meynert.

Amyloid deposits in numerous neuritic plaques and fewer vessel walls were visualized in the neocortical

sections by immunostaining with an antiserum to a synthetic polypeptide of β -protein [10] and formic acid pretreatment [4] (Fig. 2a). Some β -protein amyloid deposits of the plaque type and superficial type [10] were



stained in a section of the cerebellar hemisphere (Fig. 3a). Immunostaining with an antiserum to paired helical filaments (PHFs) [12] disclosed numerous NFTs, dystrophic neurites in the periphery of neuritic plaques and neuropil threads in the sections of the neocortex and hippocampus (Fig. 2b). Immunostaining with an antiserum to a synthetic polypeptide of prion protein (PrP) [6] and hydrolytic autoclave pretreatment [7] revealed diffuse fine neuropil stainings in the neocortical gray

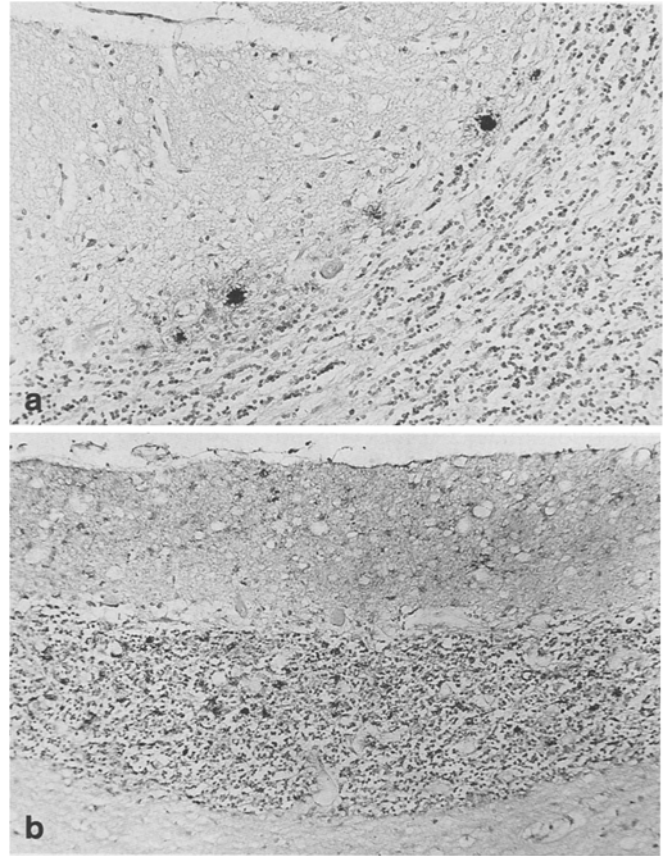


Fig. 3a,b. Immunoperoxidase staining in the sections of the cerebellum. **a** ABC staining with an antibody to β -protein and formic acid pretreatment. Some β -protein amyloid deposits of the plaque type are visible along the Purkinje cell layer of the cortex. Spongiform changes in the molecular layer are also noticed. **b** Peroxidase-antiperoxidase staining with an antibody to prion protein and hydrolytic autoclave pretreatment. Diffuse fine neuropil stainings in the molecular layer and coarse-dotted stainings in the granular cell layer of the cortex can be found. **a** $\times 125$, **b** $\times 106$

Fig. 1. A photomicrograph of the gray matter in the frontal cortex. Numerous small vacuoles in the neuropil represent mild spongiform changes. H&E, $\times 550$

Fig. 2a,b. Avidin-biotin-peroxidase complex (ABC) immunoperoxidase staining in the sections of the neocortex. **a** The staining with an antibody to β -protein and formic acid pretreatment, frontal cortex. Amyloid deposits in several neuritic plaques are stained. **b** The staining with an antibody to paired helical filaments without pretreatment, temporal cortex. Several neurofibrillary tangles (NFTs) and numerous neuropil threads are revealed. **a** $\times 100$, **b** $\times 200$

matter (data not shown) and the molecular layer of the cerebellar cortex and coarse-dotted stainings in the granular cell layer of the cerebellar cortex (Fig. 3b).

Protease-resistant PrP in the brain tissue was demonstrated using the protocol for its extraction and Western blot analysis as described (data not shown) [5]. PrP gene analysis was performed using peripheral blood lymphocytes and frozen brain tissue as described [8], and no substitution was found in codons 102, 117, 129, 178 and 200 of the PrP coding region.

Discussion

There are some difficulties in neuropathologically distinguishing AD from the normal aging process, because important hallmarks such as neuritic plaque or NFT can be found in both conditions. Our previous studies showed that some neuropathological findings could be used as reliable criteria to distinguish between them [10, 12]. These findings included β -protein amyloid deposits in the cerebellum, which are found in AD but not in the normal aging process [10]. Such deposits were also observed in this case. Moreover, an excessive number of neuritic plaques and NFTs were seen throughout the neocortex beyond the hippocampus. The diagnosis of AD is, therefore, indisputable in this case.

We found mild but typical spongiform changes in this case. In the neocortex, these were mainly observed in the pyramidal cell layers. Spongiform changes were also found in the caudate nucleus, putamen, and thalamus. These characteristics of distribution seem to be consistent with the characteristics of spongiform changes found in typical CJD cases. However, this is not conclusive evidence for a diagnosis of CJD without demonstrating protease-resistant PrP accumulation in the brain, for which a Western blot analysis is commonly used [1, 2].

We have recently developed a new pretreatment method for the immunohistochemistry of PrP, named hydrolytic autoclaving [7]. This method enabled us to show diffuse gray matter staining in sections of the central nervous system of CJD patients and CJD-infected mice [8, 9], and allowed us to show follicular dendritic cell staining in the sections of the lymphoid organs of CJD-infected mice [9]. Especially, the PrP

staining that was revealed in the cerebellar cortex showed most conspicuous patterns, fine-dotted stainings in the molecular layer and coarse-dotted stainings in the granular cell layer. These stainings were highly specific for CJD cases [7] and were found in all CJD cases without any PrP gene substitutions [8]. We think, therefore, that this method can be a tool as useful as Western blotting for confirming the diagnosis of CJD in such cases. In the present case we found the above-mentioned diffuse gray matter staining in sections of the cerebral and cerebellar cortices. This finding and the presence of protease-resistant PrP shown by the Western blotting were useful in confirming the diagnosis of CJD. With respect to the ease of performing the method and interpreting the results, Western blotting may be superior to our immunohistochemical method. However, this method has also allowed us to reveal several unique and distinct patterns of PrP accumulation in the brain sections of patients with Gerstmann-Sträussler syndrome (GSS), familial CJD, and other unclassified dementia, whose PrP gene had substitutions or an insertion [8]. Therefore, this method is worth applying to CJD and the allied diseases not only for diagnosis but also for further evaluation of their pathology.

The sensitivity of the immunohistochemistry using formic acid pretreatment was obviously lower than that of the method using hydrolytic autoclaving, because the former method stained plaque-type deposits of PrP in the brains of patients with CJD and GSS, but could not stain diffuse accumulation of PrP in the gray matter neuropil. In our experience with immunohistochemistry of other protein antigens using formalin-fixed brain sections, hydrated autoclaving was the most effective in enhancing the immunoreactivity of NFT or tau protein [13], and was also superior to hydrolytic autoclaving (unpublished data). In enhancing the immunoreactivity of β -protein, formic acid pretreatment was more effective than either hydrated or hydrolytic autoclaving (unpublished data).

There have been at least two neuropathologically proven reported cases of concomitant AD and CJD (Table 1) [3, 11]. These cases had, clinically, a very short duration of illness (10 and 15 months) and, neuropathologically, demonstrated marked spongiform changes and neuronal loss in the neocortex, suggesting CJD as a main pathological process in these cases. Our case was clinically unique in that it had a long period of slowly

Table 1. Reported cases with concomitant Alzheimer's disease (AD) and Creutzfeldt-Jakob disease (CJD)

Author	Sex	Age at onset	Duration of illness	Weight of brain	Presenting symptom(s)	Myoclonus	PSD	SP β /PrP	NFT ^c	SC ^c	Blotting ^a	Transmission ^b
Brown	F	73	10 mth	950 g	Memory loss	—	+	+/-	+++	+++	+	+
Powers	F	69	15 mth	930 g	Memory loss Ataxia	+	—	+/-	+	+++	+	NE
Muramoto	M	70	5 yr	1025 g	Memory loss	+	+	+/-	+++	+	+	Ongoing

SP, immunoreactivity of senile plaques to the antibody to β -protein or prion protein (PrP); SC, spongiform changes; NE, not examined

^a Positivity of protease-resistant PrP in Western blot analysis

^b Transmission of CJD to experimental animals

^c +++ or +, marked or mild changes respectively, in the original descriptions

progressive dementia, followed by rapid mental deterioration in addition to gait disturbance and myoclonus, and it was also unique pathologically in that it did not show such severe spongiform changes in the neocortex. These findings, therefore, suggest that the advanced AD might have been complicated by CJD in our case.

Acknowledgements. We thank K. Hatanaka for technical assistance, and B. T. Quinn for reviewing the manuscript.

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