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# Proliferation of mesaxons in the optic nerve of hamsters infected with the Echigo-1 strain of Creutzfeldt-Jakob disease

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**Abstract**. The Echigo-1 strain of CJD was isolated by Mori and colleagues from a case of 33-years-old female with a panencephalopathic type of CJD. An incubation period following intracerebral inoculation of hamsters with 10 % cleared suspension of the Echigo -1-affected brain was approximately six months. We report here ultrastructural changes in the optic nerves. Vacuoles developed within myelinated axons: within axoplasm or within the myelin sheath and these were accompanied by exuberant reaction of macrophages and hypertrophied astrocytes. Axons underwent Wallerian degeneration and dystrophic neurites were also seen. Most important, we observed proliferation of inner mesaxons. Cross-sectional profiles of innumerable myelinated fibers contained membranous organelles which were continuous with the inner lamellae of the oligodendroglial cells. These unusual proliferations of inner mesaxons formed whorls and elaborated loops. In some axons, proliferation was so severe that loops of mesaxon filled the whole cross-section of the axon. Occasionally, we observed intrusion of the membranous tongue of the inner mesaxon into axoplasm.

Short communication

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Creutzfeldt-Jakob disease (CJD) is a slow neurodegenerative disease occurring worldwide in a sporadic, infectious (iatrogenic) or familial manner and it is caused by a poorly defined infectious agent known as a prion (Prusiner 1998). CJD is classified as a transmissible spongiform encephalopathy (TSE) and it is regarded as a polioencephalopathy where, in addition to the grey matter change, there is severe white matter damage (Liberski et al. 1989, DeArmond and Prusiner 1997). From a standpoint of molecular neuropathology, CJD is characterized by the accumulation of disease-specific protein $PrP^{Sc}$, which is an abnormal conformer of a normal precursor protein, $PrP^{c}$ (Budka et al. 1997, DeArmond and Prusiner 1997).

We report here proliferation of the inner mesaxons in the optic nerve of hamsters infected with the Echigo-1 strain of CJD (Mori et al. 1989) in addition to the more typical pathological alterations of spongiform change, axonal degeneration and astrocytosis. Ten six-week-old inbred hamsters were inoculated with the Echigo-1 strain of CJD which had been passaged previously seven times including the primary isolation. The incubation period following intracerebral inoculation with 10% cleared suspension of the Echigo-1-affected brain was approximately six months. Brains from infected animals revealed spongiform changes, astrocytosis and widespread accumulation of abnormal isoform of $PrP^{Sc}$ (Liberski et al. 1999). Optic nerves were dissected at the terminal stage of disease and routinely processed for transmission electron microscopy. All experimental procedures have been described elsewhere (Liberski et al. 1993).

Optic nerves from sham-inoculated control animals demonstrated no obvious alterations. Optic nerves from the Echigo-1 strain of CJD-infected hamsters demonstrated intramyelin vacuoles, robust astrocytic processes and numerous dystrophic neurites. Cross-sectional profiles of innumerable myelinated axons showed intracytoplasmic membranous organelles which were continuous with the inner lamellae of the oligodendroglial cell (Fig. 1). We noted abnormal proliferation of inner mesaxons which formed whorls and elaborate loops. In some axons, proliferation was so severe that loops of the mesaxon filled whole cross-section of the axon. Occasionally, we observed intrusion of the membranous tongue of the inner mesaxon into axoplasm.

The Echigo-1 strain of CJD was isolated by Mori and colleagues (1989) from a case of 33-years-old female with a panencephalopathic type of CJD. The inoculum prepared from the patients brain passaged CJD to guinea pigs with the incubation period (IP) of 728 days at the primary and approximately 400 days at the subsequent passages. From an animal which exhibited active running and an excessive response to external stimuli, a substrain was isolated with substantially reduced IP (254 days). This strain was re-isolated in hamsters with an IP of 141 days at the 3rd passage and it was available for us at the 6th passage.

In a series of previous studies we evaluated the only existing model of panencephalopathic type of CJD, the Fujisaki strain of CJD passaged in mice (Tateishi et al. 1987, Liberski et al. 1989, 1990, 1997). In that model, we described two types of vacuoles and suggested that intramyelin vacuoles are brought about by lymphokines secreted from activated microglial cells and astrocytes (Liberski et al. 1997). Indeed, we subsequently showed that both TNF-$\alpha$ peptide and mRNA are upregulated toward the terminal stage of disease (Kordek et al. 1996) and that this cytokine injected into the anterior chamber of an eye, produces lesions in the optic nerve indistinguishable from those seen in the panencephalopathic type of CJD (Liberski et al. 1993).

Thus study presents a second panencephalopathic model of CJD available in small laboratory rodents. It is important because this is the only such model in hamsters (the Fujisaki strain of CJD was passaged in mice) and it may be used for comparative studies of different strains of agent in the same host (namely - hamster). Thus far only mouse and hamster model have been available for comparative studies (Liberski 1992a, b).

The proliferation of inner mesaxons is a rare, albeit already reported, phenomenon of unknown significance. All we know is that oligodendroglial cell proliferation is stimulated by contact with neurons but why the inner mesaxon wrinkles when the axons undergoing atrophy is still unclear. Plausibly, it is even the result of simple mechanical traction. Alternatively, it may be that an interruption occurs in the crucial physiologic interactions between the axon and oligodendroglial cell. Interestingly, proliferation of inner mesaxons has been reported in mice intoxicated with cuprizone (Hemm and Carlton 1971); a condition that mimics some aspects of experimental TSE (Kimberlin et al. 1976). The latter finding is of particular interest because copper ions seem to mediate transitions between different glycoforms of PrP (Wadsworth et al. 1999). Irrespective of the cause, proliferation of the mesaxon may lead to complex opening of either the intraperiod or the major dense line which re-

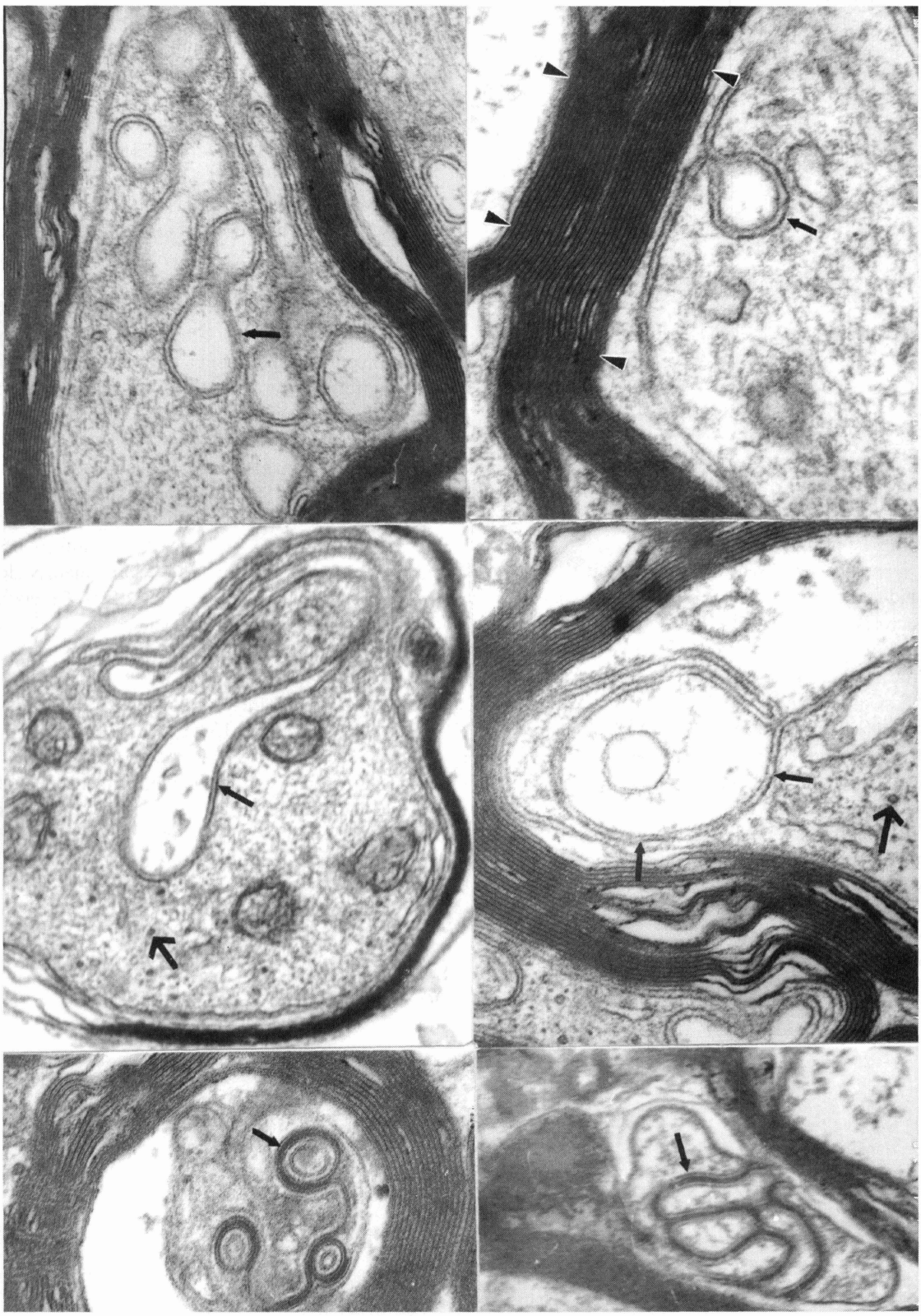

Fig. 1. Several examples of labirynth-like network of proliferating mesaxons (arrows). Note also compact myelin sheath (arrowheads), microtubules (large arrows) and neurofilaments. Original magnification, x 30 000.

sults in a formation of intramyelinic vacuoles as suggested by earlier observations (Liberski et al. 1990) and it further contributes to degeneration of the optic system. Further experiments are in progress which will at least partially, research this problem.

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