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# An Unusual Structure in Kuru Brain

A. Peat and E. J. Field

Medical Research Council Demyelinating Diseases Unit, New Castle General Hospital
Westgate Road, Newcastle upon Tyne

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*Summary.* Within the cytoplasm of otherwise normal Purkinje cells barred structures have been found in cases of Kuru. They are also present in small numbers in axoplasm of nerve fibres, both in human and chimpanzee material. They present a discrete punctate structure and are closely associated with intracellular membranous profiles.

*Zusammenfassung.* In Fällen von Kuru wurden im Cytoplasma von ansonst normalen Purkinjezellen Strukturen mit strahlendichten Balkenmustern gefunden. Solche sind auch in geringerer Zahl im Axoplasma von Nervenfasern sowohl im menschlichen wie auch im Schimpansenmaterial vorhanden. Sie weisen eine zarte Punktstruktur auf sind und eng mit intracellulären Membranprofilen verbunden.

**Key-Words:** Electron Microscopy — Kuru — Inclusion — Cytoplasmic.

Prolonged further examination of the Kuru material (human and chimpanzee) which formed the basis of the first electron microscopic study of this condition (Field *et al.*, 1968) has shown the presence of a structure not previously seen, and this is here described.

## Material and Methods

Formalin fixed human Kuru brain (obtained through the courtesy of Dr. J. D. Mathews in Okapa, New Guinea) and a block of second passage chimpanzee cerebellum (kindly given by Drs. C. D. Gajdusek and C. J. Gibbs, Jr., of National Institute of Health, Bethesda, U.S.A.) together with non-Kuru brain from New Guinea and normal chimpanzee cerebellum, formed the basis of this study. Preparation of the material was as described by Field *et al.* (1968) — post fixation in Dalton's osmic acid fixative and inclusion in Epon. Ultrathin sections were cut using an LKB Ultrotome and doubly stained with uranyl acetate and lead citrate. They were examined in a Philips EM 300 equipped with goniometer stage.

## Results

As a consequence of formalin fixation ultrastructural preservation was poor in the human cerebellar material. Numerous densely barred structures were however found in association with mitochondria in the granule cell layer of the cerebellar cortex. Although they appeared intracytoplasmic it was impossible to identify with any certainty the cell type (Fig. 1). However, prolonged search uncovered similar bodies in glutaraldehyde fixed second passage Kuru chimpanzee cerebellum, where they were clearly located within the cytoplasm of Purkinje cells, the general structure of which was well preserved with normal mitochondria and other organelles (Figs. 2 and 3). On rare occasions similar barred structures were found within nerve fibre axoplasm (Fig. 4). Here they were orientated along the length of the fibre.

The densely barred structures were apparantly formed from parallel aggregates of rough endoplasmic reticulum with which were associated ribosome like particles, 250 Å in diameter (Fig. 5). Where the cisternae were closely apposed to each other the intervening spaces appeared dense and amorphous at low magnification (Fig. 5). At higher magnification, however, and with tilting, the dense region could be

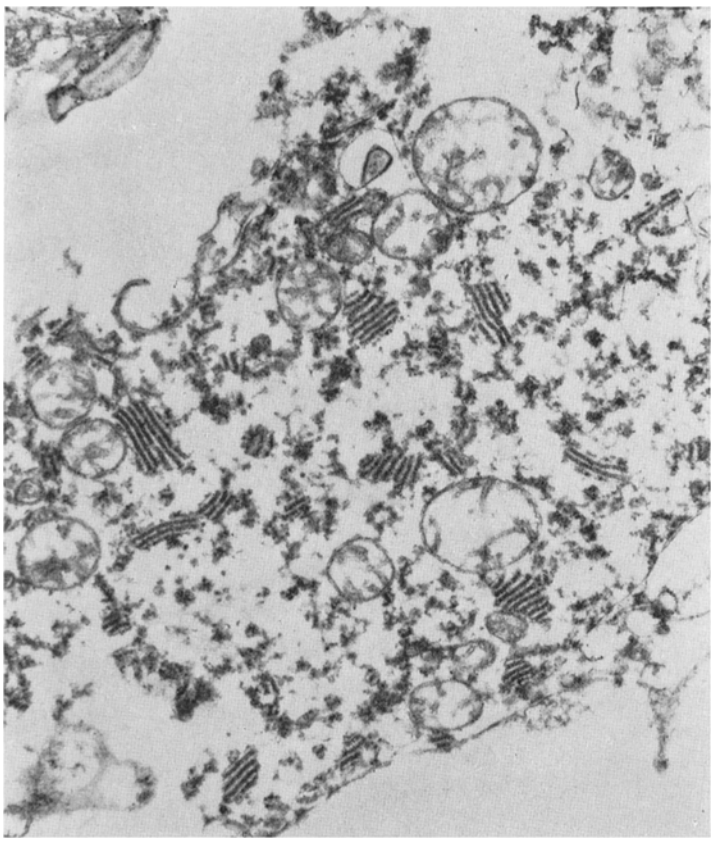

Fig. 1. Intracytoplasmic dense barred structures. ? Purkinje cell body. Human Kuru (formalin fixation) ×18,700

resolved into particles each around 200 Å in diameter. In some cases the particles from adjacent cisternae alternated with each other so that a "zip-like" appearance resulted. Whilst these were generally only 2 or 3 bands, sometimes they were more numerous and five, six or more might be present in series. These bands were remarkbly uniform in thickness: the dark bands being about 340 Å and the intervening lighter ones 300 Å. Stereo pictures did not materially facilitate interpretation, though there were occasional appearances suggestive of a spiral flamentous arrangement, the turns of which might have produced the discrete ribosome like particles (Fig. 6).

Nothing comparable has been seen in non-Kuru brain or in the single normal chimpanzee examined.

## Discussion

The present findings emphasize the vagaries of electron microscopic sampling, so subject to random chance. Even an extensive search must be limited to a very small total volume of tissue. Neither the study of Field *et al.* (1968) nor that of Lampert *et al.* (1969) noted the present structures, whose significance is uncertain. Herndon (1963) did not report their presence in normal rat cerebellum.

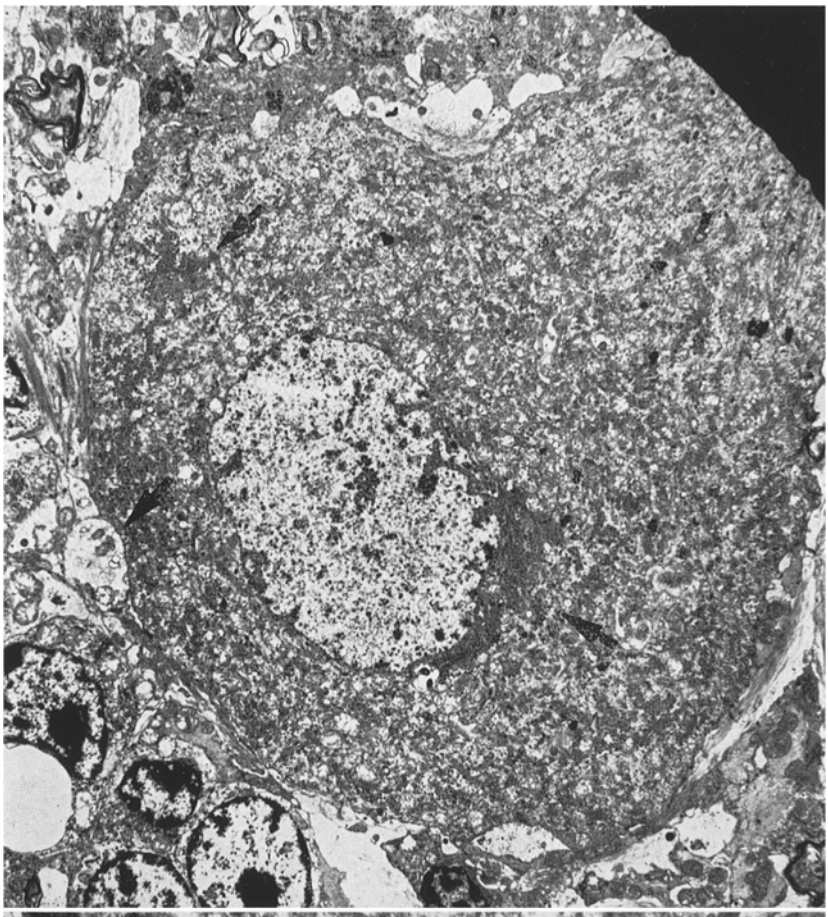

Fig. 2

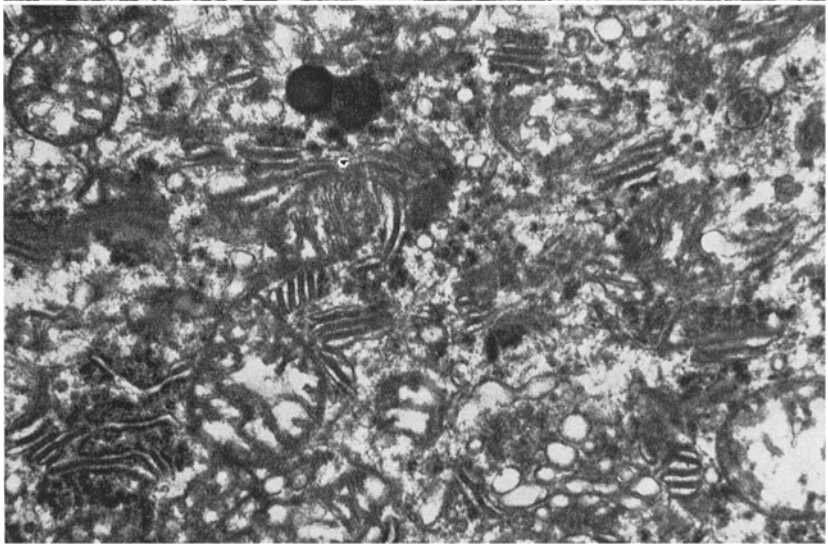

Fig. 3

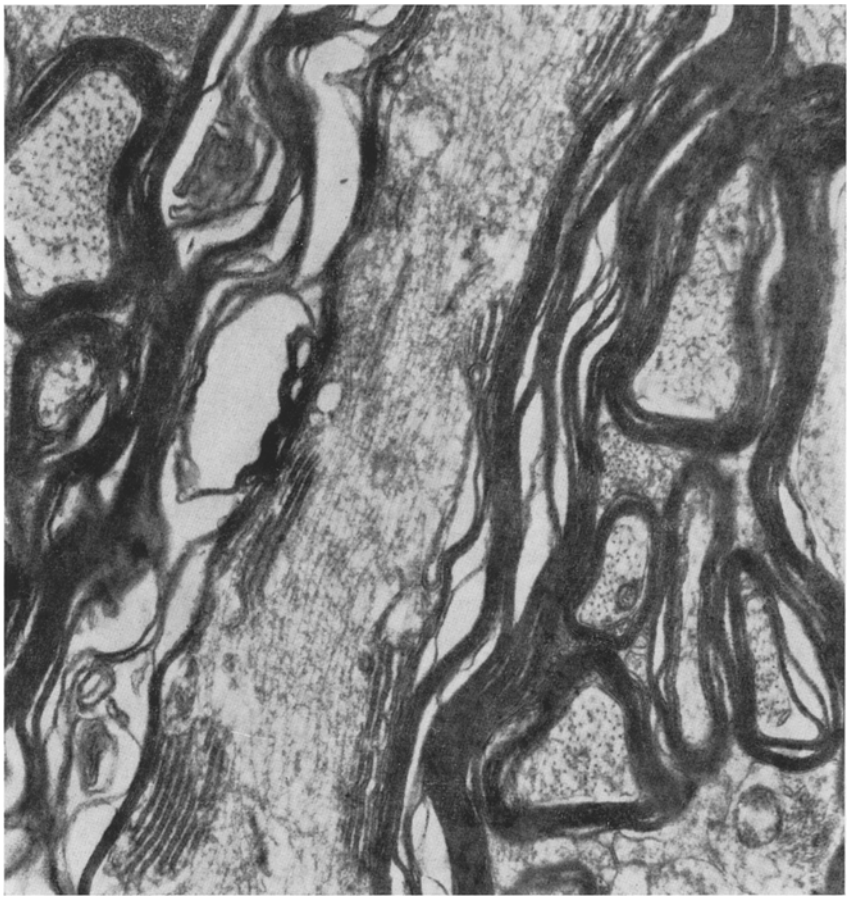

Fig. 4. Dense barred structures in axis cylinders of Kuru chimpanzee

Since the dense amorphous component may comprise discrete particles slightly smaller than those associated with normal endoplasmic reticulum they might represent modified ribosomes, though their presence within the axis cylinder would be highly exceptional.

In view of the similarities which Kuru bears to scrapie and aging (Field, 1967) an intensive search of cerebellum from scrapie infected or aged animals might prove rewarding. Even more interesting is the possibility that the changes are virus induced. It may be that the dense bands represent sites of some stage of viral replication. This is especially interesting because of the intimate association of scrapie agent (to which that of Kuru seems closely akin if not identical) with intracellular membranous structures (Gibbons and Hunter, 1967; Adams and Field, 1968; Adams *et al.*, 1970).

Fig. 2. Purkinje cell from cerebellar cortex of second passage Kuru chimpanzee (glutaraldehyde immersion fixation). Numerous dense barred structures are present (arrows) ×2000

Fig. 3. Higher magnification of Fig. 2 showing dense barred structures and mitochondria. ×20,600

Fig. 5

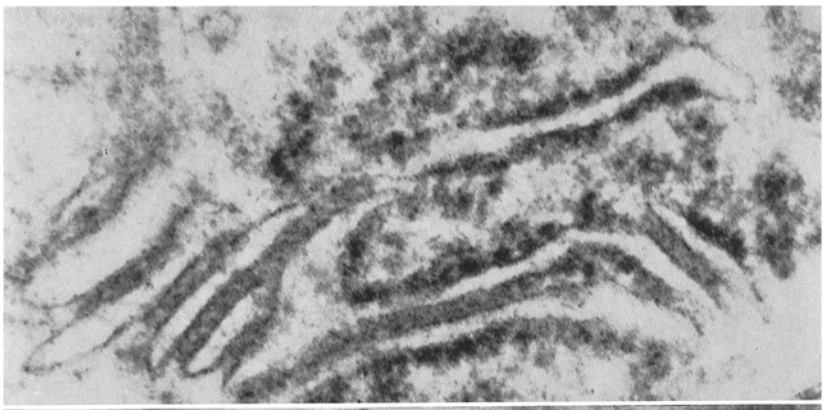

Fig. 6

Fig. 5. Association between granular endoplasmic reticulum and dense barred structures in Purkinje cells from Kuru chimpanzee. ×81,400

Fig. 6. Dense barred structure Kuru chimpanzee Purkinje cell cytoplasm. Suggestion of spiralization (arrows). EM stage tilted + 18°. ×105,250

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E. J. Field, M.D.
Medical Research Council Demyelinating Disease Unit
Newcastle General Hospital
Westgate Road
Newcastle upon Tyne NE4 6BE, England