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Short Communication

Tight Junctions of Oligodendrocytes

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Summary. Freeze-fracture replicas of the rat corpus callosum revealed prominent junctional strands in fractured cell membranes of the somata of oligodendrocytes. The junctional strands were characterized by an elaborate system of straight or slightly undulating rows of linear aggregates of particles or ridges in the P face and furrows in the E face.

Key words: Oligodendrocytes – Tight junction – Freeze-fracture.

Introduction

Junctional strands, characteristic of tight junctions, were reported in oligodendroglia, particularly at the internal mesaxon, between adjoining paranodal loops (Schnapp et al., 1973; Reale et al., 1975; Schnapp and Mugnaini, 1975), at the junction of the outer loop with the outermost myelin and in corresponding areas of deeper myelin layers (Dermietzel, 1974a; Mugnaini and Schnapp, 1974; Reale et al., 1975; Schnapp and Mugnaini, 1975; Rosenbluth, 1976). The observation of tight junctions in human oligodendroglioma (Tani, 1976) led to the examination of cell membrane structures in normal oligodendrocytes.

Material and Methods

Brains of adult rats were fixed by intravascular perfusion with 2.5% glutaraldehyde buffered with 0.1 M phosphate (pH 7.4). Small pieces of fixed specimens were dissected from the corpus callosum, immersed overnight at 4° C in a 30% glycerol solution after rinsing in phosphate buffer, then rapidly frozen in liquid Freon 12 and subsequently transferred to liquid nitrogen. The frozen specimens were fractured in vacuo in a HFZ-I freeze-replica apparatus and replicated by platinum and carbon in a HUS-4 vacuum evaporator. The replicas were cleaned in a commercial bleaching solution, washed twice in distilled water, and finally placed on copper grids for examination in a HU-12 electron microscope.

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Results and Discussion

Prominent junctional strands (Fig. 1) were arranged parallel to each other, following a straight or gently undulating course. The strands were composed of linear aggregates of particles or ridges in the P face and furrows in the E face, suggesting tight junctions. The furrows were occasionally studded with a few particles. The width of the individual linear aggregates of particles or ridges measured approximately 8 nm. The distance from one strand to the other varied between 30 and 90 nm. Adjacent strands occasionally fused to form a single strand.

Membrane particles were few in the P face where the junctional strands were present, whereas they appeared scattered or aggregated in the P face where the junctional strands were absent (Fig. 1). The E face, intercalated between adjacent

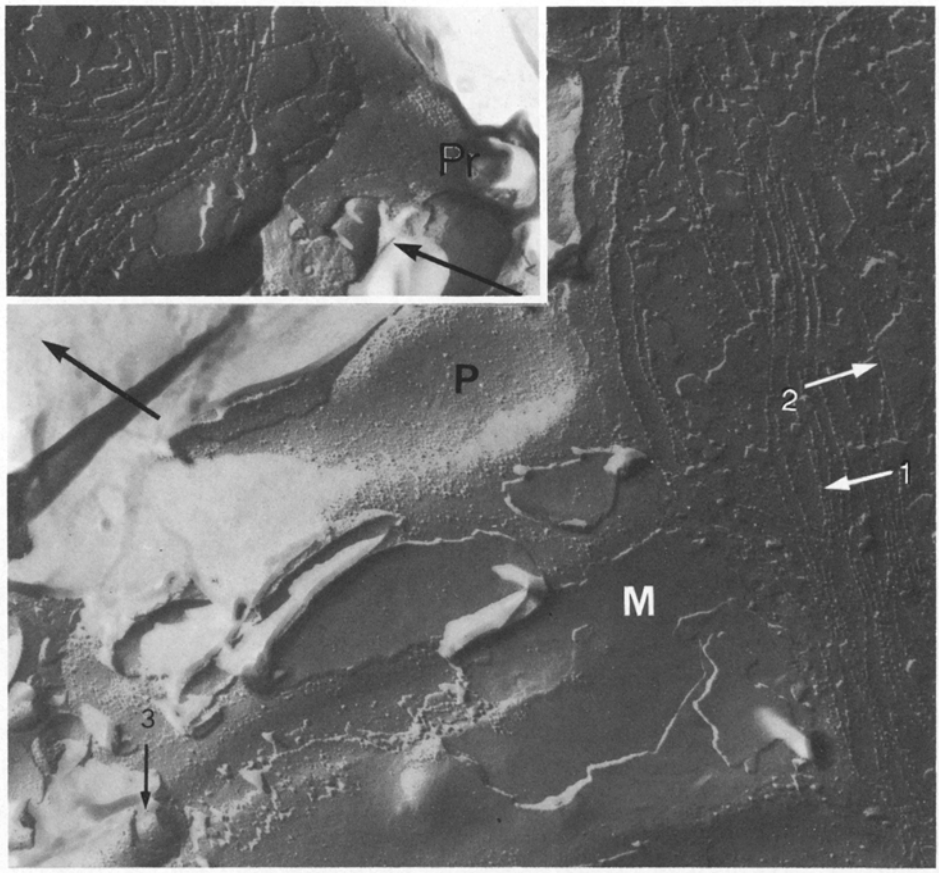

Fig. 1. Freeze-fracture replica of the rat corpus callosum. Conspicuous parallel rows of linear aggregates of particles or ridges (*arrow 1*) and occasional furrows (*arrow 2*). The P face (*P*) in areas devoid of junctional strands shows broad and shallow depressions as well as randomly distributed membrane particles. Myelin sheets (*M*) and myelinated fibers (*arrow 3*) are located on the broad depressions. The P face studded with junctional strands (*inset*), shows a small process (*Pr*). The long arrows in the corners indicate the direction of shadow casting. × 32,000; *inset* × 28,000

junctional strands, was usually smooth (Fig. 1). It is suggested that the P face most likely does not represent the myelin sheet but rather the axolemma or a glial cell membrane due to the fact that the myelin sheet was usually devoid of membrane particles (Bischoff and Moor, 1967a and b; Branton, 1967; Tani et al., 1973a). The P face was quite large, about 12 μm in length, sometimes showing a small process (Fig. 1). Consequently, the fractured membrane appears to be too extensive to be considered as an unmyelinated axolemma and apparently does not represent an axolemma of the node of Ranvier, since the presence of tight junctions has not been reported here (Livingston et al., 1973; Schnapp et al., 1973; Dermietzel, 1974b; Mugnaini and Schnapp, 1974; Schnapp and Mugnaini, 1975; Rosenbluth, 1976).

Broad and shallow depressions were occasionally found on the P face (Fig. 1), the long axis being perpendicular to the axis of the junctional strands. If the depressions are generated by the paranodal loops, they can run parallel to the junctional strands between the paranodal loops (Schnapp et al., 1973; Reale et al., 1975; Schnapp and Mugnaini, 1975; Rosenbluth, 1976). One could, therefore, suggest that the depressions of the P face may be induced by adjacent myelinated fibers or other cytoplasmic processes. No paranodal loops or glial-axonal junctions (Livingston et al., 1973; Dermietzel, 1974b; Schnapp and Mugnaini, 1975) were evident around the fractured membranes in the present study (Fig. 1). Three myelin sheets which were located on the P face, as shown in Figure 1, did not completely surround the fractured face. It might be suggested that these myelin sheets belong to adjacent myelinated fibers.

Since no tight junctions have been reported in astrocytes (Dermietzel, 1973; Tani et al., 1973b, 1974; Landis and Reese, 1974), the observed junctional strands could be located in cell membranes of the oligodendroglia. The present study indicates that they are not located at the internal mesaxon, between adjoining paranodal loops, beneath the outer loops, or in the deeper myelin layers (Schnapp et al., 1973; Dermietzel, 1974a; Mugnaini and Schnapp, 1974; Reale et al., 1975; Schnapp and Mugnaini, 1975; Rosenbluth, 1976), but rather in the cell membranes of the somata of oligodendrocytes; the short cytoplasmic process also speaks in favour of this suggestion. The E face between the junctional strands was usually smooth and might represent the outermost myelin sheet adjacent to the oligodendroglia.

References

Bischoff, A., Moor, H.: Ultrastructural differences between the myelin sheaths of peripheral nerve fibers and CNS white matter. Z. Zellforsch. **81,** 303–310 (1967a)

Bischoff, A., Moor, H.: The ultrastructure of the 'difference factor' in the myelin. Z. Zellforsch. **81,** 571–580 (1967b)

Branton, D.: Fracture faces of frozen myelin. Exp. Cell Res. **45,** 703–707 (1967)

Dermietzel, R.: Visualization by freeze-fracturing of regular structures in glial cell membranes. Naturwissenschaften **60,** 208 (1973)

Dermietzel, R.: Junctions in the central nervous system of the cat. I. Membrane fusion in central myelin. Cell Tiss. Res. **148,** 565–576 (1974a)

Dermietzel, R.: Junctions in the central nervous system of the cat. II. A contribution to the tertiary structure of the axonal-glial junctions in the paranodal region of the node of Ranvier. Cell Tiss. Res. **148,** 577–586 (1974b)

Landis, D.M.D., Reese, T.S.: Arrays of particles in freeze-fractured astrocytic membranes. J. Cell Biol. **60,** 316–320 (1974)

Livingston, R.B., Pfenninger, K., Moor, H., Akert, K.: Specialized paranodal and interparanodal glial-axonal junctions in the peripheral and central nervous system: a freeze-etching study. Brain Res. **58,** 1–24 (1973)

Mugnaini, E., Schnapp, B.: Possible role of zonula occludens of the myelin sheath in demyelinating conditions. Nature (Lond.) **251,** 725–727 (1974)

Reale, E., Luciano, L., Spitznas, M.: Zonulae occludentes of the myelin lamellae in the nerve fibre layer of the retina and in the optic nerve of the rabbit: a demonstration by the freeze-fracture method. J. Neurocytol. **4,** 131–140 (1975)

Rosenbluth, J.: Structure of electrically excitable membrane at nodes of Ranvier in the frog brain. J. Cell Biol. **70,** 182a (1976)

Schnapp, B., Mugnaini, E.: The myelin sheath: Electron microscopic studies with thin sections and freeze-fracture. Golgi Centennial Symposium, pp. 209–233 (Santini, M., ed.). New York: Raven Press 1975

Schnapp, B., Peracchia, C., Mugnaini, E.: Freeze-fracture of Ranvier nodes. J. Cell Biol. **59,** 306a (1973)

Tani, E.: The membrane characteristics of human brain tumors as revealed by freeze-fracture technique. Progress in neuropathology, Vol. 3, pp. 129–172 (Zimmerman, H.M., ed.). New York-San Francisco-London: Grune & Stratton 1976

Tani, E., Ikeda, K., Nishiura, M.: Freeze-etching images of central myelinated nerve fibres. J. Neurocytol. **2,** 305–314 (1973a)

Tani, E., Ikeda, K., Nishiura, M., Higashi, N.: Specialized intercellular junctions and ciliary necklace in rat brain. Cell Tiss. Res. **151,** 57–68 (1974)

Tani, E., Nishiura, M., Higashi, N.: Freeze-fracture studies of gap junctions of normal and neoplastic astrocytes. Acta neuropath. (Berl.) **26,** 127–138 (1973b)

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