Neuroglia and the myelin-bearing cell: a symposium

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DYNAMIC ASPECTS OF THE MYELIN-GLIAL RELATIONSHIPS IN DEVELOPMENT AND REPAIR

RICHARD P. BUNGE (New York) An understanding of the mechanism of myelin breakdown required a detailed analysis of the structural and metabolic relationship between myelin and the myelin related cell—that is, the peripheral Schwann cell and the central oligodendrocyte. The present discussion emphasized the following aspects of this relationship.

The oligodendrocyte formed the central myelin sheath by the spiral disposition and compaction of its plasma membrane around the axon (often simultaneously myelinating a number of axons). After a period of active myelin formation the oligodendrocyte apparently retained continuity with the cytoplasmic constituents which had been demonstrated on the internal and external aspects of the mature sheath and at the region of the termination of the myelin lamellae near the node of Ranvier. The special junctions established between terminating myelin loops and the axolemma in the perinodal region might be significant for normal saltatory conduction; observations on pathological changes in these contacts during demyelination would be of interest.

The cytoplasmic constituent of myelin contained a prominent microtubule component. This component might provide a mechanism by which materials were conveyed from the perikaryon of the oligodendrocyte to the more distal parts of the myelin-glial complex. The ongoing activities of the mature myelin-related oligodendrocyte were emphasized by its active incorporation of RNA precursors and by evidence that certain molecular components of myelin might be in equilibrium with component pools, presumably within the oligodendrocyte perikaryon. Recent evidence that the demyelinating agent, diphtherial toxin, was a potent inhibitor of protein synthesis suggested that this ongoing metabolism might apply to protein as well as to lipid myelin components.

Dramatic changes in myelin structure (with apparent fluid accumulation between myelin lamellae) occurring in response to triethyl tin administration, in certain forms of spongy degeneration, and in a number of experimental situations (as well as the proclivity of the oligodendrocyte to swell acutely in a variety of disease conditions) suggested that the myelin related cell might be unusually active in ion (and secondarily water) transport. This process might be expected to require energy and had been little explored in myelin related cells.

Additional dynamic aspects of the myelin-glial cell system were evident in the variety of cytological reactions

observed in glial responses to disease. A number of workers now believed that small glial cells were capable of proliferation and provided a reserve supply of glial cells for the disposal of debris, for repair, or for scarring. The responses for repair included the potential for remyelination of demyelinated axons within the central nervous system. This had been observed in a number of situations and had involved either oligodendrocytes or apparent Schwann cells (the latter perhaps derived from contiguous peripheral nerve).

SOME ASPECTS OF THE KINETICS OF THE NEUROGLIA

J. B. CAVANAGH (London) A comparison was made between the rates of the responses of neuroglial cells and microglia after a brain wound and after chemical denervation of a spinal tract. A quantitative approach to cell proliferation around a brain wound showed that from the second day onwards there was an increase in cells which was greatest at the wound edge but extended for several hundred micra into the brain parenchyma. At the wound edge the mitotic cells might be, as Königsmark and Sidman (1964) have shown in the mouse, largely haematogenous in origin. Beyond the first 200 μ it was principally endogenous cells that divide. Results of this quantitative approach to cell proliferation disclosed the importance of rapid perfusion-fixation to show the true mitotic rates, and also showed that colchicine did not penetrate into the brain. This useful mitotic inhibitor could not, therefore, be used for the study of cell proliferation in the brain. As might be expected, proliferation rates fell off logarithmically with distance from the wound edge.

Contrary to previous views, neuroglia were quite susceptible to x-irradiation provided that their mitotic function was studied. Doses of more than 500 r profoundly affected the capacity of astrocytes and microglia to respond to injury. After 2,000 r the mitotic rates of these cells were significantly lowered. After 1,000 r the astrocytes showed an anomalous enhanced activity that might be due to failure of the vascular bed to reconstitute after injury (Cavanagh, 1968). The finding of Hopewell and Wright (1967) that the suppression of proliferation was as marked one year after irradiation as after one week suggested a use for this type of injury to determine the replacement rates of these cells.

Ultrastructural studies showed that the chief feature of this type of lesion was leakage of protein into brain (Blakemore, 1969). Astrocytes, as well as microglia, were active in taking this up and re-establishing the normal low protein condition of the brain extracellular space.

After bromophenylacetylurea poisoning in the rat, severe denervation occurred in the posterior columns, especially in its upper parts (Cavanagh, Chen, Kyu, and Ridley, 1968). This began from the eighth day onwards and was complete by the third week, about 80% of the fibres being lost with the dose used. Cellular responses to axonal degeneration were rapidly initiated but only slowly completed. Phagocytosis of myelin debris took many weeks and astrocytes actively 'walled off' debris and other constituents of the damaged area by the growth of numerous processes. The absence of protein escaped from blood vessels thus profoundly modified the responsiveness of both microglia and astrocytes.

In neither lesion had oligodendrocytes been found unequivocally to respond to these stimuli and they showed only regressive changes. Moreover, the absence of irradiation damage in the nuclei of these cells suggested that they did not proliferate in these circumstances.

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BIOCHEMISTRY OF GLIA AND THE DEVELOPING BRAIN

A. N. DAVISON (London) Biochemical studies on the neuropil and on isolated glial cells (see Rose, 1968) indicated that in many ways the metabolism of glia resembled that of the neurone. In addition, the work of Hydén and his colleagues suggested a very special metabolic relationship between the two types of cell, but the exact rôle of glia in the partnership was not established. Even the presence in glia of high activities of pseudo-cholinesterase and carbonic anhydrase (Giacobini, 1962) remained unexplained, as did the significant absence of gangliosides.

There was, however, little doubt that glial cells had an important rôle in maintenance of the mature myelin sheath and in myelinogenesis. Thus, myelination in the central nervous system was effected by macroglial cells (Maturana, 1960; Peters, 1960) whose extruded plasma membrane could be traced from the cell to the newly synthesized myelin lamellae (Bunge, Bunge, and Ris, 1961). In the early steps of development the formative cell might have different properties from that of the oligodendroglial cell and had been variously identified as astrocyte or reactive macroglial cell. Certainly the myelinogenic cell was rich in enzymes required for lipid synthesis (Blunt and Wendell-Smith, 1967); RNA and lipid might also be seen to accumulate within its cytoplasm (Mickel and Gilles, 1968).

Shortly before myelination there was a multiplication of glial cells (Bensted, Dobbing, Morgan, Reid, and Payling Wright, 1957; Friede, 1961), so that much of the adult population was reached by the onset of the process. The stage had been referred to as 'myelination gliosis'.

If myelin originated from glial membrane it would therefore have been expected that typical myelin con-

stituents would be found in the brain before myelination occurred, but at that time only traces of the characteristic myelin lipid (cerebroside) could be detected (Cuzner and Davison, 1968). Subsequently, there was parallel increase of cholesterol and phospholipid with deposition of myelin but accretion of cerebroside remained delayed. Thus, a relatively low cerebroside content had been found in kitten optic nerve (Banik, Blunt, and Davison, 1968) and rat brain myelin isolated by centrifugation during early development. The newly formed myelin contained relatively more lecithin and was deficient in long chain fatty acid phosphoglycerides in comparison with that of mature optic nerve or isolated myelin. These various features were characteristic of typical cell membrane, including possibly those of the glial cell plasma membrane, so that it was proposed by Davison, Cuzner, Banik, and Oxberry (1966) that the 'early' myelin fraction contained a transition phase of myelinogenesis consisting of undifferentiated glial plasma membrane as well as mature type myelin. This correlated with morphological observations showing that during the initial phase of myelination the lamellae were loosely wound around the axon (Caley and Maxwell, 1968) and that only later was compact adult type myelin formed. Similarly, changes had been noted in tissue culture in the appearance of newly formed myelinated fibres during early stages of myelinogenesis.

In later work it proved possible to separate two fractions from 'early' myelin (from 10 to 25-day-old rat brain), one with a composition comparable with that of adult myelin and a second similar to that of cell membranes (Norton, Davison, and Spohn, 1968). The hypothesis was supported by further evidence based on the enzymatic composition of both membrane fractions and from isotope experiments suggesting a product-precursor relationship for myelin and the second membrane fraction. Current work suggested that 'early' myelin might be metabolically more active than mature myelin, for 7-dehydrocholesterol and 24-dehydrocholesterol incorporated into myelin at this time could undergo reduction to cholesterol, whereas preliminary experiments suggested that this did not happen in older animals (Banik and Davison, unpublished observation).

It was proposed that during the period of myelinogenesis glial plasma membrane was converted to myelin by the insertion of cerebroside and loss of lecithin molecules. This might, therefore, be an example of the transition (Benson, 1966; Lucy, 1968) of one type of membrane—for example, subunit structure—to another—for example, the unit membrane.

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