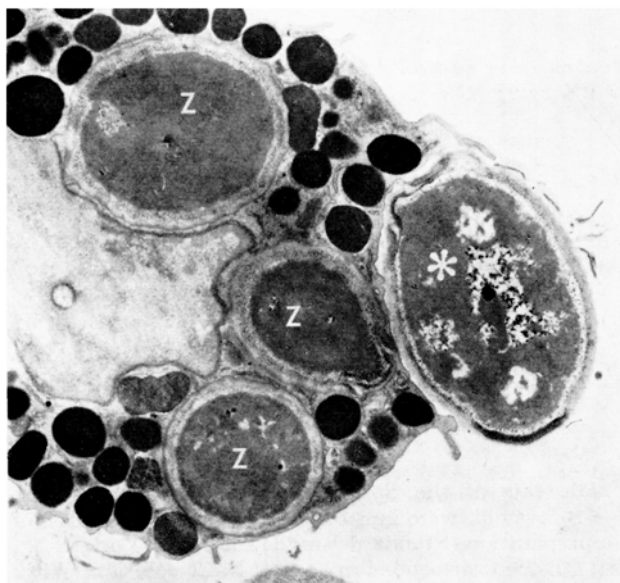


the electron microscope studies, peritoneal fluid was fixed for 3 h in Karnovsky's fluid diluted 1:2 with 0.1M cacodylate buffer, pH 7.2. While in the fixative, the cells were collected into a pellet by centrifugation. After washing the pellet 15–18 h in sucrose-cacodylate buffer at about 4°C, it was post-fixed for 1 h in 1% OsO₄ in veronal acetate buffer, pH 7.4, at room temperature. The pellet was dehydrated in ethanol at room temperature and was embedded in Epon according to the method of LUFT⁹. Thin sections were stained with solutions of either lead citrate or with both uranium acetate and lead citrate. An RCA EMU 3G microscope was used.

Some of the mast cells examined by light microscopy 1½ h after the injection of zymosan appeared to contain zymosan particles and, at 3 h after zymosan, this close association between zymosan particles and mast cells was even more common. The resolution of the light microscope was not sufficient to determine with certainty whether the zymosan particles had become intracytoplasmic or whether they were merely indenting the surface and creating this illusion. 1–3 h after the injection of zymosan, the peritoneal macrophages were filled with these particles. Many eosinophils contained some particles, and some mast cells were associated with one or several particles.



A mast cell from rat peritoneal fluid. Lead citrate stain. One zymosan particle (*) is adherent to the cell surface. Three zymosan particles (Z) are within vacuoles. $\times 7800$.

Electron micrographs supported the concept that mast cells took up zymosan particles by phagocytosis. The Figure shows a mast cell with several zymosan particles. One of these particles appears to be merely adhering to the cell whereas 3 particles are within vacuoles. One hour after the injection of zymosan, those mast cells associated with particles displayed large pseudopods, in contrast to their normal ovoid condition. More numerous and larger microvilli were often seen on the mast cell surface closest to the zymosan particles. Three hours after injection, neither pseudopods nor evidence of increased surface activity were noted. Mast cell granules, including those in the immediate vicinity of the ingested particles, appeared normal after both 1 and 3 h.

Experiments in progress indicate that zymosan particles are still visible in mast cells 8 days after injection and that, at the light microscope level, the particles still appear intact, contrary to their degraded condition in macrophages. The total number of peritoneal fluid mast cells decreased markedly following zymosan injection and remained low for at least 8 days.

This report demonstrates that mast cells can be phagocytic. Studies by others^{10–12} have implicated the mast cells in certain immunological reactions. It remains to be determined whether phagocytosis by mast cells is an integral part of such phenomena¹³.

Résumé. Des rats ont été injectés par voie péritonéale avec une suspension de zymosan, une préparation de parois de levure, *Saccharomyces cerevisiae*. Les cellules de la lymphe péritonéale furent étudiées après au microscope optique et électronique. Ces expériences ont démontré que les mastocytes changent d'aspect et que les particules de zymosan se retrouvent dans des vacuoles cytoplasmiques peu après l'injection. Les mastocytes sont donc des cellules capables de phagocytose.

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Small Myelinated Perikarya in the Cerebellar Granular Layer of Mammals Including Man

In the peripheral nervous system, myelinated neuronal perikarya have long been known to occur in spinal and some cranial ganglia, notably acoustic and vestibular, of a wide variety of vertebrates (cf. the review of ROSENBLUTH and PALAY¹). In the mouse cerebellum cultured in vitro, perikaryal myelin was frequently observed around small cells, ca. 7 μ in diameter, with round chromatin-rich nuclei and scanty cytoplasm (SUYEOKA and OKAMOTO²) (Figure 1).

The present investigation was undertaken to determine whether these myelinated perikarya exist in the mouse cerebellum in situ. Adult mice, over 3 months old, were perfusion-fixed according to the method of CAMMERMEYER³ and stained for myelin by WEIL's method. Small myelinated perikarya of similar size as observed in the cultures were also found not infrequently in the cerebellar granular layer⁴ (Figure 2). Although the isolated perikarya were not rare, they were often closely associated

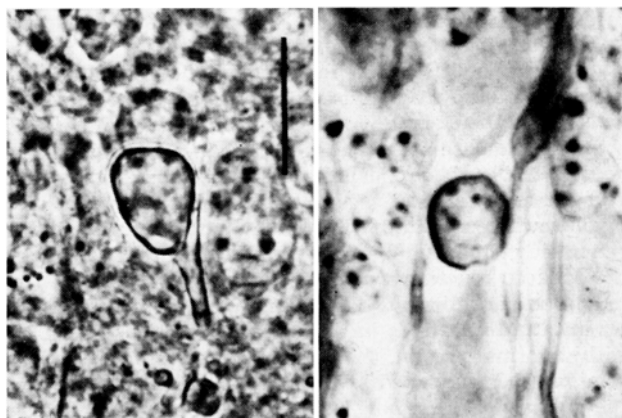


Fig. 1. A small myelinated perikaryon in the cultured mouse cerebellum in vitro. Note a round nucleus which lies in an eccentric position. This cell body is closely apposed to a myelinated axon, the portions of which are out of focus. New-born mouse cerebellum. 20 days in culture. Living. Phase-contrast. All the photographs in this paper are the same magnification and the bar indicates 10 μ .

Fig. 2. A similar cell in the granular layer of the adult mouse cerebellum. Perfusion-fixed. 3 μ paraffin section. WEIL stain.

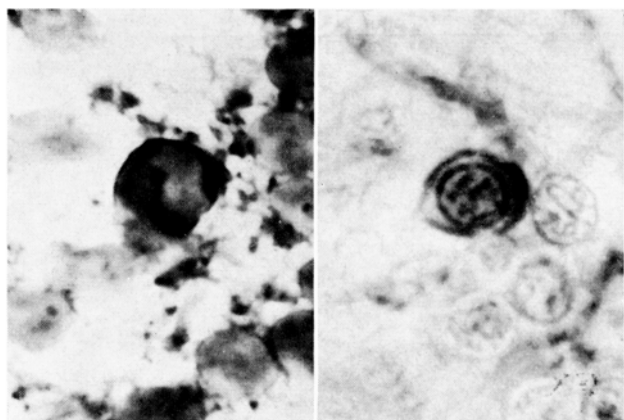


Fig. 3. The inner granular layer of the 8-day-old mouse cerebellum. Immersion-fixed. 3 μ paraffin section. SCHULTZE stain.

Fig. 4. A small myelinated perikaryon in the granular layer of the adult human cerebellum. Note the neighbouring nuclei of similar size, whose perikarya are not myelinated. 30 μ celloidin section. WEIGERT stain.

with myelinated axons. On some occasions the fine processes were myelinated thinly for a short distance (up to 10 μ), but it could not be ascertained whether these myelinated processes were portions of axons or dendrites. Thickness of the perikaryal myelin was variable among the cells, but was almost comparable to that of myelin sheaths around the nearby axons. Rough estimates indicated that the ratio of these myelinated perikarya to ordinary non-myelinated perikarya of similar size was 1/1000 or more. They were present exclusively in the deeper portion of the granular layer, though they were seen sporadically in the upper portion. In the meanwhile, there appeared no particular difference in regional distribution among the cerebellar folia. Since similar cells have not been encountered in the neighbouring white matter or in the brain stem, it is highly probable that they are identical with granule-cell neurons.

Ontogenetically, as examined in the SCHULTZE-stained preparations, they were noted in the mouse cerebellum as early as 8 days postnatally (Figure 3), though suggestive structures were observed already on the sixth day. These postnatal stages are concurrent with the onset of myelination around axons in the mouse cerebellum.

A comparative study was made with ordinary and phase-contrast microscopes on part of the extensive collection of myelin-stained, chiefly WEIGERT, preparations of a variety of mammalian species available in this laboratory. Phylogenetically, they were observed in all mammals thus far examined: mole, mouse, rabbit, racoon dog, dog, cat, badger, goat, Japanese monkey, and man⁵ (Figure 4). There was an impression that they were less frequent in larger animals than in smaller ones. ROSENBLUTH⁶, using an electron microscope, discerned similar myelinated perikarya in the toad cerebellum. It is evident, therefore, that they may exist widely throughout the vertebrate phyla, at least from amphibia to man, and that occurrence of these structures is a normal phenomenon not only in culture but also in situ.

Although the functional significance of these myelinated perikarya is not known, one possibility might be as follows: It is well established by CAJAL⁷ that granule cells in the deeper portion of the granular layer send their axons, i.e. parallel fibres, to the lower portion of the molecular layer. As a result of saltatory conduction, impulses via these myelinated granule cells may arrive slightly faster at some basket cells, the basal portions of some Purkinje-cell dendrites and also some Golgi-cell dendrites, and may induce a change in the postsynaptic membrane potentials of these neurons. Consequently such a change might exert delicate influence on the excitability of these neurons against other retarded impulses⁸.

Zusammenfassung. Kleinzellen mit perikarieller Marksheide wurden in der Körnerschicht des Kleinhirns verschiedener Säuger und des Menschen gefunden. Diese Zellen treten bei der Maus erst am 8. Tag nach der Geburt auf und wurden nur in der Körnerschicht, offenbar als Körnerzellen, beobachtet.

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- Identity of these structures as compact myelin sheaths was confirmed in electron micrographs of the adult mouse cerebellum by Dr. A. KONISHI of this laboratory, whose courtesy is gratefully acknowledged.
- They were more easily discerned in the perfusion-fixed preparations. In the preparations fixed by immersion, swellings of axonal myelin are frequent especially in smaller animals and thus it was often difficult to distinguish these small myelinated perikarya from such swellings of the myelinated axons.
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- Acknowledgment: The author is greatly indebted to Prof. Dr. M. OKAMOTO for permission to use the myelin-stained preparations in this study and for reviewing the manuscript, and to Dr. S. DEURA for discussion concerning the electrophysiological interpretation.