

and viral proteins for their glycine/alanine ratio¹⁴. When taken together, the overall glycine-to-alanine ratio of cytochrome C (*Neurospora*), ferridoxin (*Clostridium*), tobacco mosaic virus, ribonuclease (*Aspergillus*), tryptophan synthetase (*E. coli*), and azurin (*Pseudomonas*) is 1.0. This ratio may be compared to the reported ratio of 60 for precambrian samples. The large difference between contemporary organisms and the microfossils cannot be explained by a difference in amino acid degradation rates. It is of interest to note that our synthetic (abiotic) glycine/alanine ratio of 14 (Table) is significantly closer to the fossil ratio.

The molecular mechanism involved in the formation of the complex thiocyanate structures described here is unclear. However, we have demonstrated that under plausible primitive earth conditions such formation is a spontaneous occurrence. Furthermore, we have observed morphology, chemical composition, and stability also found in geological microfossils. These results suggest

that the microfossils may actually be remnants of cell precursors or that the earliest cells possessed a biochemistry significantly different from that found in present day organisms. In any case, the need for more careful evaluation of the significance of these microfossils is clearly pointed out¹⁵.

Résumé. Les microsphères obtenues par irradiation UV du thiocyanate d'ammonium sont comparées du point de vue morphologique, chimique et de certaines propriétés physiques, à certains microfossiles précambriens.

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Amino acid analysis of irradiated NH₄SCN microspheres following hydrolysis

Time of irradiation (min)	Glycine μM/mM NH ₄ SCN	Alanine μM/mM NH ₄ SCN	Glycine alanine
75	0.14	0	—
195	3.22	0.23	14
315	0.38	0.17	2.23

¹³ P. H. ABELSON, in *Researches in Geochemistry* (Ed. P. H. ABELSON; John Wiley, New York 1959), p. 79.

¹⁴ R. V. ECK and M. O. DAYHOFF, *Atlas of Protein Sequence and Structure* (National Biomedical Research Foundation, Silver Springs, Md. 1966).

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Axo-Somatic Synapses in Procerebrum of Gastropoda

Central synapses of Gastropode molluscs are generally believed to be of axo-axonic type^{1,2}. In fact no axo-somatic synapses have been found electron-microscopically in ganglia of different Gastropode species³⁻⁶. In one case a synapse-like profile was described in trophosphonium of cerebral ganglia of *Glossodoris*⁷. Even if this was an axo-somatic synapse, it was a very unusual picture for this tissue, where synapses were reported to be mainly axo-axonic type⁷. So it is generally accepted that in Gastropodes the bodies of nerve cells do not play any role in the synaptic transmission, since the site of interneuronic contacts are restricted to the centrally located neuropile.

However, such organization is not a general rule: we were able to demonstrate in the procerebrum of the Pulmonates *Helix* and *Limax* abundant structures that were typical axo-somatic synapses so far as their morphology is concerned.

Procerebra of *Helix pomatia* and of *Limax cinerea-niger* were fixed in 1.5% OsO₄ buffered with collidine, embedded in Durcupan ACM, cut on the LKB Ultratome III. Sections were contrasted with lead citrate and examined in the Tesla BS 413A electron microscope.

Light microscopic studies have shown that the procerebrum differs in many respects from typical Gastropode ganglia: its neuropile has rather a lateral than central position and the cellular mass consists of very small (up to 10 μ in diameter) uniform, densely packed neurones¹. The submicroscopic organization of procere-

brum, a complete description of which we give elsewhere, is also peculiar in some respects. Axo-somatic synapses are one of these unusual features.

In both *Helix* and *Limax*, the presynaptic endings in the cellular mass of the procerebrum are of 2 types. The first type (Figure 1) contains dense-core vesicles of about 800–1200 Å in diameter, while the second type (Figure 2) contains clusters of empty vesicles with a diameter of 5–800 Å. The fibres giving origin to the first type endings, are usually thicker than those of the second type, though in some other respects there are similarities between them. Both types are varicose fibres consisting of thin segments and expansions with mitochondria and synaptic vesicles. Varicose expansions of both types are often contacted with more than 1 nerve cell, sometimes with 3–5 cells. Widening of synaptic cleft typical for mollusc

¹ T. H. BULLOCK and G. A. HORRIDGE, *Structure and Function in the Nervous Systems of Invertebrates* (Freeman, San Francisco and London 1965), vol. 2.

² L. TAUC, in *Physiology of Mollusca* (Academic Press, New York and London 1966), vol. 2.

³ H. M. GERSCHENFELD, *Z. Zellforsch. mikrosk. Anat.* 60, 258 (1963).

⁴ J. ROSENBLUTH, *Z. Zellforsch. mikrosk. Anat.* 60, 213 (1963).

⁵ D. A. SAKHAROV, V. L. BOROVYAGIN and I. Zs.-NAGY, *Z. Zellforsch. mikrosk. Anat.* 68, 660 (1965).

⁶ R. E. COGGESHALL, *J. Neurophysiol.* 30, 1263 (1967).

⁷ G. NICAISE, M. PAVANS DE CECCATTY and C. BALEYDIER, *Z. Zellforsch. mikrosk. Anat.* 88, 470 (1968).

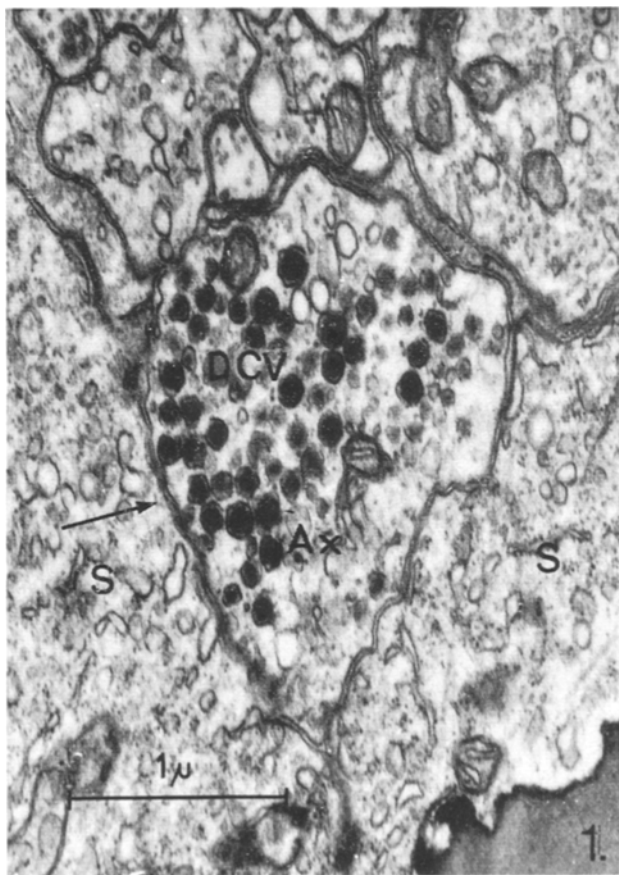


Fig. 1. Axo-somatic contact of the first type in procerebrum of *Helix pomatia*. Ax, axon, S, soma, DCV, dense-core vesicles.

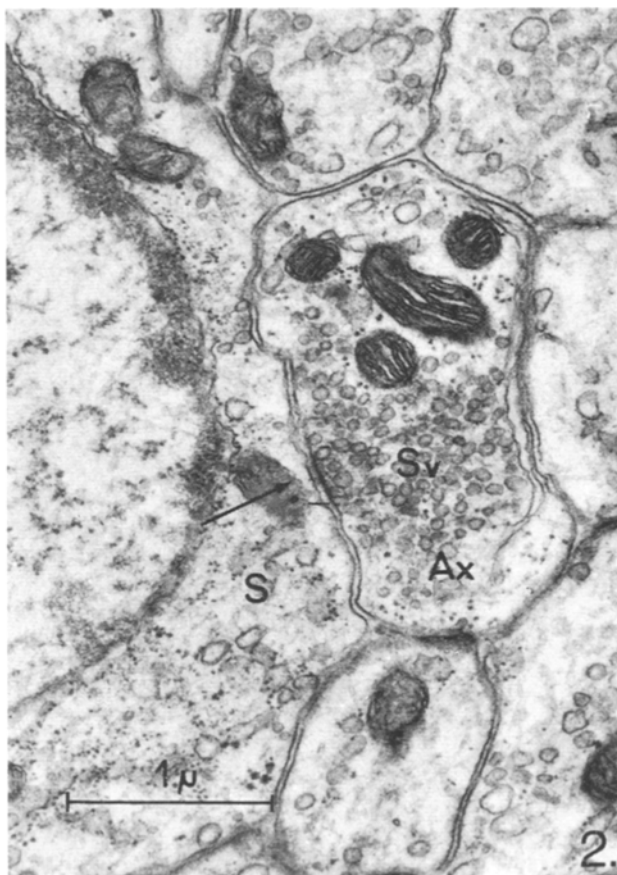


Fig. 2. Axo-somatic synapse of the second type in procerebrum of *Limax cinerea-niger*. Ax, axon, S, soma, Sv, synaptic vesicles.

synapses can be seen, and an invagination of the pre-synaptic ending similar to those described in axo-somatic synapses of the Pelecypode molluscs⁸ as well.

Axo-somatic synapses are much more abundant in *Limax* than in *Helix* procerebrum. The whole surface of a neurone is sometimes covered with presynaptic endings. Such pictures resemble the vertebrate nervous tissue and are strikingly different from the well-known picture of Gastropode ganglia. In both species the density of inter-neuronal contacts is maximal in the deepest layers of cells and progressively diminishes in the more superficial parts of cellular mass.

Dense-core vesicles are known to be connected with neurotransmitters of monoamine nature. We applied the sensitive and specific fluorescent histochemical method for revealing cellular monoamines to the *Helix* procerebrum. A simplified histochemical procedure based on fixation of tissue in an aqueous formaldehyde solution, was used⁹. It was shown that fine varicose fibres with green fluorescence were situated between non-fluorescing nerve cells in the internal third of the procerebral cellular mass. Such fibres can be seen in the neuropile as well, but generally the fibre matter of the procerebrum is very poor in monoamine-containing fibres as compared with the strongly fluorescing neuropile of metacerebrum. Thus, histochemical findings are in accordance with electron microscopic data and demonstrate the possible monoaminergic nature of axo-somatic synapses of the first type. Neither histochemical nor electron microscopic investigations gave evidence in favour of the presence

of monoamine-containing perikarya in procerebrum. Thus fibres of the first type are believed to be originated somewhere outside the procerebrum.

The second type of axo-somatic synapses is comparable morphologically to the cholinergic synapses of vertebrates, but in this respect we have had no physiological evidence as yet; thus at present it is difficult to speculate on the origin of these fibres. In the procerebrum nerve endings of similar morphology also form axo-axonic synapses.

Zusammenfassung. Es wurde gezeigt, dass im Procerebrum der Gastropoden axosomatische Synapsen von 2 Typen vorkommen. Diese Befunde sind ungewöhnlich, da axosomatische Synapsen in anderen Teilen des Zentralnervensystems nicht existieren.

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⁸ I. ZS.-NAGY, *Annales biol. Tihany* 31, 147 (1964).

⁹ A. V. SAKHAROVA and D. A. SAKHAROV, *Citologia* 10, 389 (1968).