

ENDOCYTOSIS IN ADIPOKINETIC HORMONE (AKH)-PRODUCING CELLS OF THE MIGRATORY LOCUST, *LOCUSTA MIGRATORIA*, IN REST AND DURING FLIGHT.

J.H.B. Diederer, M. Dorland, W.F. Jansen and H.G.B. Vullings

Research Group for Histology and Cell Biology of Regulatory Systems, Zoological Laboratory, University of Utrecht, Padualaan 8, 3584 CH Utrecht, The Netherlands.

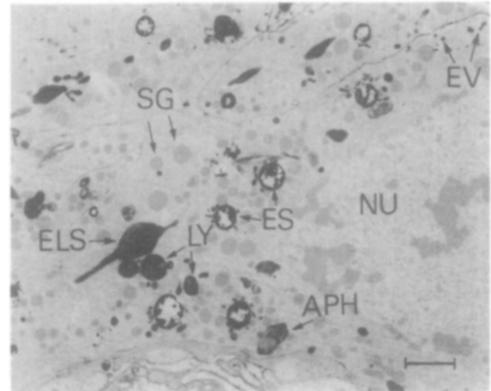
Intrinsic gland cells of the corpus cardiacum of *Locusta* synthesize AKH, and release it into the haemolymph shortly after onset of flight. AKH stimulates the release of diacylglycerol - the fuel for the flight muscles - from the fat body. The peak of haemolymph AKH is reached after about 30 min flight (1). Release of AKH mainly occurs from short processes of the gland cells, by means of exocytosis. It may be expected that this release is followed by compensatory re-uptake of membrane material by means of endocytosis near the site of exocytosis. To get information on the location of endocytotic activity and on the intracellular endocytotic pathway, the uptake of the tracer horse-radish peroxidase (HRP) by the gland cells in flying and in resting locusts was studied electron microscopically.

HRP (Sigma type II) was injected *in vivo* (resulting in $\pm 5 \mu\text{g}$ HRP per μl haemolymph) followed by a resting period of respectively 0.5 h, 1.0 h and 2.0 h, or by a resting period of 0.5 h followed by a flight period of 0.5 h. The corpora cardiaca were fixed in a mixture of 2.5% glutaraldehyde and 2% formaldehyde. For the cytochemical demonstration of HRP activity the method of Graham and Karnovsky (2) was used. The tissue was postfixed in 2% OsO_4 , embedded in Ladd plastic and ultrathin sectioned.

Signs of endocytosis were found frequently in cell bodies and proximal parts of cell processes, whereas they were only sporadically found in distal parts of cell processes, the sites of exocytosis of the secretory product. In resting animals 0.5 h after injection of HRP, endocytosed HRP was detected mainly in endocytotic pits and small vesicles, in larger vesicular structures called endosomes, in tubular structures associated with endosomes, and in some secondary lysosomes. The number of secondary lysosomes containing HRP had increased 1.0 h after injection of HRP. At that time some pre-existing, large, elongated, peculiarly structured bodies could be seen to contain HRP, whereas after 2.0 h all of them were filled with HRP. These organelles with still unknown function probably belong to the lysosomal system. Endocytotic activity seemed to be somewhat enhanced in flying animals; in these animals the HRP-filled tubular structures associated with the endosomes were particularly prominent. These structures are

thought to form together with the endosomes the prelysosomal compartment from which membrane recycling occurs (3). No endocytosed HRP was found in structures known to belong to the Golgi-GERL-complex or to other parts of the secretory pathway.

It may be concluded that the endocytotic pathway of HRP in the insect AKH-producing cells is similar to that described for mammalian cells (3). Most of the observed endocytotic activity is not coupled to the exocytotic activity of the gland cells, but is somehow connected with the uptake of nutritional or regulatory (macro)molecules by these cells.



Elements of the endocytotic pathway in cell bodies of locust AKH-producing cells, filled with HRP after 0.5 h rest followed by 0.5 h flight. The bar represents 1000 nm. APH-atrophagosome; ELS-elongated lysosomal structure; ES-endosomes associated with tubular structures; EV-endocytotic vesicles; LY-lysosomes; NU-nucleus; SG-secretory granules.

References

- (1) Goldsworthy, G.J. (1983). *Advances in Insect Physiology* **17**, 149-204.
- (2) Graham, R.C. and Karnovsky, M.J. (1966). *J. Histochem. Cytochem.* **14**, 291-302.
- (3) Van Deurs, B. and Christensen, E.I. (1984). *Eur. J. Cell Biology* **33**, 163-173.

Received: 13.3.85 Accepted: 9.4.85