

Linskens and Heinen¹⁶, and perhaps offer some clue as to the meaning of the cytochemically detectable 'esterase' activity of the pellicle itself.

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- ¹ Knox, R. B., and Heslop-Harrison, J., *Nature*, **223**, 92 (1969).
- ² Knox, R. B., and Heslop-Harrison, J., *J. Cell Sci.*, **6**, 1 (1970).
- ³ Heslop-Harrison, J., Heslop-Harrison, Y., Knox, R. B., and Howlett, B., *Ann. Bot.*, **37**, 403 (1973).
- ⁴ Knox, R. B., Willing, R. R., and Ashford, A. E., *Nature*, **237**, 381 (1972).
- ⁵ Heslop-Harrison, J., Knox, R. B., and Heslop-Harrison, Y., *Theor. appl. Genet.* (in the press).
- ⁶ Dickinson, H. G., and Lewis, D., *Proc. R. Soc.*, **B183**, 21 (1973).
- ⁷ Linskens, H. F., and Kroh, M., *Handb. Pfl. Physiol.*, **18**, 506 (Springer Verlag, Berlin, 1967).
- ⁸ Lewis, D., *Genetics Today*, **3**, 657 (Pergamon, London, 1965).
- ⁹ Heslop-Harrison, J., and Heslop-Harrison, Y., *Stain Technol.*, **45**, 115 (1970).
- ¹⁰ Jensen, W. A., *Botanical Histochemistry* (Freeman, San Francisco, 1962).
- ¹¹ Pearse, A. G. E., *Histochemistry: Theoretical and Applied*, **2** (Churchill, London, 1972).
- ¹² Barka, T., and Anderson, P. J., *J. Histochem. Cytochem.*, **10**, 741 (1962).
- ¹³ Knox, R. B., and Heslop-Harrison, J., *J. Cell Sci.*, **9**, 239 (1971).
- ¹⁴ Knox, R. B., *J. Cell Sci.*, **12**, 421 (1973).
- ¹⁵ Green, J. R., *Ann. Bot.*, **8**, 225 (1894).
- ¹⁶ Linskens, H. F., and Heinen, W., *Z. Bot.*, **50**, 338 (1962).

Mitosis in the Cryptophyceae

In a recent issue of *Nature*¹ Oakley and Dodge are critical of my theory on the evolution of the algae². I believe that their criticisms are the result of some misconceptions and inaccuracies. In reference to my article² they state "the Cryptophyceae were thought to be ancestral and closely related to the Cyanophyceae". First, there must be some error in sentence structure, for to conclude that the eukaryotic Cryptophyceae are ancestors to the prokaryotic Cyanophyceae is a difficult assumption to make. Second, I never said that the Cyanophyceae were closely related to the Cryptophyceae but that the Cyanophyceae were closely related to the chloroplasts of the Cryptophyceae. Therefore one would not expect cell division in the Cyanophyceae to be similar to that in the Cryptophyceae as they infer.

Oakley and Dodge refer to McDonald's work on mitosis in the Rhodophyceae³ and state that "It (mitosis in the Cryptophyceae) is quite different from that in the Rhodophyceae" without expanding further. I believe that they have overstated their case, as the only differences in mitosis between the two classes are: (1) the presence of some heterochromatin referred to as a kinetochore in the Rhodophyceae with no such structure in the Cryptophyceae, and (2) the presence of basal bodies in the Cryptophyceae and their absence in the Rhodophyceae (which would be expected since the Rhodophyceae have no flagellated cells) although the polar ring³ in the latter may prove to be a derivative of a basal body. These differences between mitosis in the two classes are not as significant as Oakley and Dodge seem to believe and do not rule out an evolutionary link between the Cryptophyceae and Rhodophyceae.

Lastly, Oakley and Dodge infer that *Chroomonas* (a Cryptophyte with a chloroplast) is a primitive genus in my scheme on the phylogeny of the algae². This is not so as this type of organism is fairly advanced along the evolutionary pathway. If they were seeking a more primitive organism to investigate they should have chosen a Cryptophyte without chloroplasts or one with cyanelles (endosymbiotic Cyanophyceae).

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¹ Oakley, B. R., and Dodge, J. D., *Nature*, **244**, 521 (1973).

² Lee, R. E., *Nature*, **237**, 44 (1972).

³ McDonald, K., *J. Phycol.*, **8**, 156 (1972).

Drs Oakley and Dodge reply: In regard to Dr R. E. Lee's correspondence, our letter to *Nature*¹ was not intended to be particularly critical of Dr Lee's previous article on endosymbiosis and the evolution of the algae². In fact Dr Lee's theory was only mentioned very briefly. Nevertheless we should like to respond to his comments. Of course when we stated that "the Cryptophyceae were thought to be ancestral and closely related to the Cyanophyceae", there was no intentional implication that the Cryptophyceae were ancestral to the Cyanophyceae but rather that they were ancestral algae. Since Dr Lee's scheme regards them as ancestral to every group of algae other than the Cyanophyceae we feel that it is fair to regard them as ancestral. Although Dr Lee does not state explicitly that the Cryptophyceae are closely related to the Cyanophyceae, his chart (Fig. 1) giving phylogenetic relationships places two groups of cryptophytes, the colourless cryptophytes and those with cyanelles, closer to the Cyanophyceae than any other groups of algae. We do not feel, therefore, that our statement was unfair.

This point, however, is of little importance. A much more important point, and the point we were making, is that Dr Lee's scheme implies that the Pyrrophyta with very little histone³, extremely unusual base pair composition⁴, and an apparently primitive mitotic apparatus⁵ has evolved from the Cryptophyceae which show no such apparent primitiveness or uniqueness. Similarly, the mitotic apparatus of the Euglenophyceae seems to be much more primitive than the cryptophytes from which they have evolved according to Dr Lee's scheme.

In fairness, Dr Lee does not feel that the cryptophytes with chloroplasts are ancestral to the Pyrrophyta. He feels that the colourless cryptophytes and those with cyanelles are more primitive than those with chloroplasts. With regard to the colourless cryptophytes, our results were so similar to the light

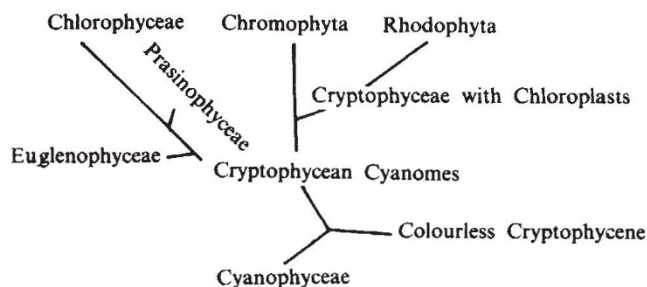


Fig. 1 Fig. 2 from ref. 2. The origin of plastids from a Cyanophyceae alga involved in an endosymbiosis with a colourless Cryptophyte. The remainder of the eukaryotic algae then evolved from this Cryptophyceae cyanome (endosymbiotic Cyanophyceae alga plus the host cell).

microscopic studies on *Chilomonas paramecium*^{6,7} that there was no reason to believe that there might be any difference between the coloured and colourless cryptophytes. In addition, there is recent evidence that the colourless cryptophytes possess degenerate chloroplasts⁸ and may not be ancestral to the cryptophytes with chloroplasts at all; the reverse being the case.

In regard to *Cyanophora paradoxa* which Dr Lee regards as a cryptophyte with a cyanelle, though this organism certainly seems to possess a cyanelle, there is some question as to its being a cryptophyte. Moreover, mitosis has been studied in this organism⁹ and it shows no primitive characteristics.

Finally, Dr Lee does not feel that the presence of kinetochores and the lack of basal bodies in the Rhodophycean alga previously studied¹⁰ are particularly significant, but to us these seem to be quite significant differences. The presence of the unique polar ring and the near complete retention of the nuclear envelope during mitosis in the Rhodophyceae also constitute striking differences. These differences suggest that the mitotic apparatus of the Cryptophyceae is more nearly like that of several other groups of algae than that of the Rhodophyta.

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- ¹ Oakley, B. R., and Dodge, J. D., *Nature*, **244**, 521 (1973).
- ² Lee, R. E., *Nature*, **237**, 44 (1972).
- ³ Rizzo, P. J., and Nooden, L. D., *Science*, *N. Y.*, **178**, 796 (1972).
- ⁴ Rae, P. M. M., *Proc. natn. Acad. Sci. U.S.A.*, **70**, 1141 (1973).
- ⁵ Leadbeater, B., and Dodge, J. D., *Arch. Mikrobiol.*, **57**, 239 (1967).
- ⁶ Nigler, K., *Arch. Protistenk.*, **25**, 295 (1912).
- ⁷ Hollande, A., *Archs. Zool. exp. gén.*, **83**, 1 (1942).
- ⁸ Sepsenwol, S., *Expl. Cell Res.*, **76**, 395 (1973).
- ⁹ Pickett-Heaps, J., *New Phytol.*, **71**, 561 (1972).
- ¹⁰ McDonald, K., *J. Phycol.*, **8**, 156 (1972).

Secretory discharge and microflora of milk gland in tsetse flies

THE uterine or milk glands in tsetse flies (*Glossina* spp.) are modified female accessory reproductive glands which elaborate and release a nutritive liquid of proteinaceous and lipid nature for the maturing intrauterine larva¹⁻³. The multi-branched tubules of the gland converge into a pair of efferent ducts which fuse inside the oviductal shelf and open into the lumen of the uterus just posterior to the opening of the oviduct^{4,5}. Cytological details of the milk gland and their modulations in relation to the state of pregnancy of the female have been described⁶ (W-C. M., D. L. D., D. S. Smith and U. Jarlfors, in preparation). Earlier work has suggested that milk is released directly into the lumen of the gland by apocrine secretion⁴. Our observations on the structure of the milk gland do not support such a mechanism, but rather, a novel type of exocrine discharge in which secretion is stored in an extracellular reservoir and released into the lumen through a dense cuticular network. At the points of milk release the lumen is frequently inhabited by bacteria which have not previously been described in milk glands. Our examination is based on milk glands from *G. morsitans morsitans* Westwood, but a comparative study using *G. austeni* Newstead and *G. longipalpis pallidipes* Austen has shown no essential differences among the three species.

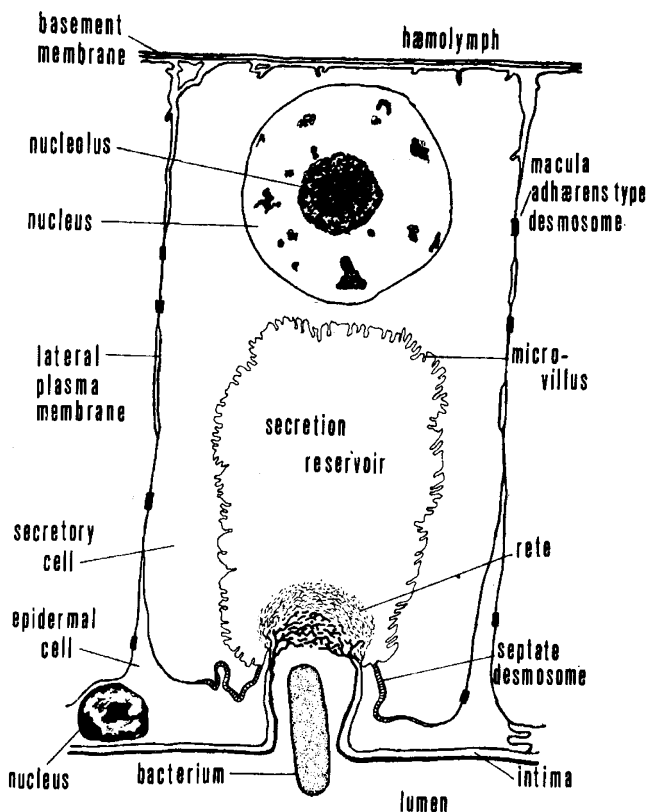


FIG. 1 Schematic representation of the structural organization of the milk gland tubules in *Glossina morsitans*.

Ultrastructural details were determined by fixation of the gland-fat body complex for 6 h in cold 5% glutaraldehyde buffered at pH 7.4 with 0.05 M sodium cacodylate buffer and post-fixation in buffered 1% OsO₄ for 1½ h. Sections from Epon-Araldite embedded materials were cut on a LKB Ultratome III using glass knives, mounted on unsupported copper grids, contrasted with saturated uranyl acetate in 50% ethanol followed by lead citrate, and examined with a Zeiss EM 9A electron microscope.

The walls of the gland tubules consist of two layers of cells: a single layer of squamous epidermal cells of ectodermal origin which lines the lumen, and a layer of secretory cells resting on a thin basement lamina. The epidermal layer is continuous with the non-glandular epithelium of the two efferent ducts and has a smooth unlaminate cuticular lining. The efferent ducts themselves have a helically thickened cuticle.

The accessory substances produced by the secretory cells are released by merocrine secretion into an extracellular storage space formed by invagination of the plasma membrane along the apical surface of the gland cell. One such storage reservoir is associated with each gland cell (Figs 1, 2). The stored secretion is channelled into the lumen through a cup-shaped invagination of the intima which projects into the reservoir. At the point of contact with the reservoir the intima is composed of 50–70 nm thick anastomosing strands forming a rete of 2.0–2.5 µm diameter. Underneath this coarse extracellular porous structure the cuticle is further ramified into a mass of extremely fine threads of about 3–6 nm diameter which has a wool-like appearance in the electron micrographs (Fig. 3). This layer is about 1.1 µm thick in the centre. There is no evidence that structural changes or deformations of the ductule occur during cycles of secretory activity. The cuticular wool however has a slightly blurred appearance in the electron micrographs when it becomes saturated with the