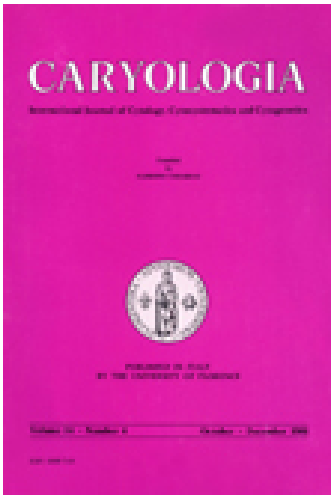

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PELLICLE ULTRASTRUCTURE OF SOME *EUGLENA* SPECIES *

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SUMMARY — The pellicle of five species of *Euglena*, belonging to three different groups (Catilliferae, Radiatae and Serpentes), was studied by transmission electron microscopy, in order to see if pellicular morphology could be taken as a valuable character to differentiate the groups. The data demonstrate that pellicular ultrastructure is not constant within the groups, and can therefore be considered a differential character only at the level of species.

INTRODUCTION

In the course of studies on the red neuston of two alpine water bodies (GEROLA *et al.* 1980*a* and 1980*b*), among other *Euglena* species we found one, identified as belonging to the group Catilliferae, whose pellicle had an ultrastructural aspect markedly different from that of all the other species described so far (WOLKEN and PALADE 1953; HALLER 1959; FREY WYSSLING and MUHLETHALER 1960; GIBBS 1960; LEEDALE 1964, 1967 and 1971; LEEDALE *et al.* 1965; MIGNOT 1965 and 1966; SOMMER 1965; ARNOTT and WALNE 1967; WOLKEN 1967; BUETOW 1968; SCHWELITZ *et al.* 1970; ROGERS *et al.* 1972; DODGE 1973; GEROLA *et al.* 1980*a* and 1980*b*). Since pellicle ultrastructure is thought to be a major differential character, we decided to examine the pellicle of two known species of Catilliferae, one of Radiatae and one of Serpentes, whose ultrastructure had not yet been described. Our aim was to see if differences in pellicle ultrastructure could be taken as a differential character among the groups. We did not consider any species of Rigidae, because half of them have already been studied (HALLER 1959; LEEDALE 1964, 1967 and 1971; LEEDALE *et al.* 1965; MIGNOT 1965 and 1966).

MATERIALS AND METHODS

The *Euglena* cells were collected from the Vezzena water body, as previously described (GEROLA *et al.* 1980*a*). This *Euglena* will be referred to as *Euglena sp.*,

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though for some characters it might be perhaps included in the group of *E. sanguinea* Ehrenb. The other species come from the collection of the Culture Centre of Algae and Protozoa, Cambridge, England: *E. polymorpha* Dangeard (No 1224/26) and *E. caudata* Hübner (No 1224/24b) (Catilliferae, sensu PRINGSHEIM 1956), *E. geniculata* var. *terricola* Dangeard (No 1224/4c) (Radiatae), *E. mutabilis* Schmitz (No 1224/9d) (Serpentes).

The samples were fixed in phosphate-buffered 3% glutaraldehyde, post-fixed in osmium tetroxide, dehydrated in ethanol and embedded in Epon-Araldite. Ultrathin sections were stained with uranyl acetate and lead citrate, and viewed in a Siemens Elmiskop 1A electron microscope.

RESULTS

Euglena sp. — This *Euglena* has been identified as belonging to the group Catilliferae on the basis of its chromatophores (Fig. 1), which have a central pyrenoid covered on both sides by a cup of paramylon. This type of chromatophore is peculiar to this group (PRINGSHEIM 1956; BOURELLY 1970).

The pellicle in transversal section appears as a series of rather long, protruding hooks arching over small knobs, slightly bent in opposite direction (Figs. 2 and 3). The periplast that contours the concave surface of the hooks, delimiting part of the groove, deepens inside the peripheral cytoplasm and ends as an enlarged flange below the knobs (Fig. 2). Both hooks and knobs contain 3 to 4 microtubules. Were the pellicle curves over to continue into the gullet, the pellicular hooks become erect, and are then bent in the opposite direction; the knobs become small protuberances at the base of the hooks, and the microtubular position changes from dorsal to ventral (Figs. 4 and 4, inset).

Elements of the subpellicular endoplasmic reticulum (SER) are numerous, and can sometimes be seen to penetrate characteristically inside each pellicular hook (Fig. 3).

E. polymorpha. — As seen in transverse section, the pellicle forms a beak-like, larger ridge and a finger-like, smaller one, bent towards each

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Fig. 1. — Chromatophore of *Euglena sp.*, showing central pyrenoid (py), surrounded by two cups of paramylon (pa). × 15,000.

Fig. 2. — Pellicle of *Euglena sp.*, as seen in transverse section. Two kinds of ridges are visible: protruding, finger-like alternating with smaller, knob-like ones. The groove is outlined by a dense periplast band ending in an enlarged flange at the base of the smaller ridges. Three to four microtubules are visible in each ridge (arrows). × 60,000.

Fig. 3. — Pellicle of *Euglena sp.* Each finger-like ridge contains elements of the subpellicular endoplasmic reticulum which is abundant all along the cell surface. × 60,000.

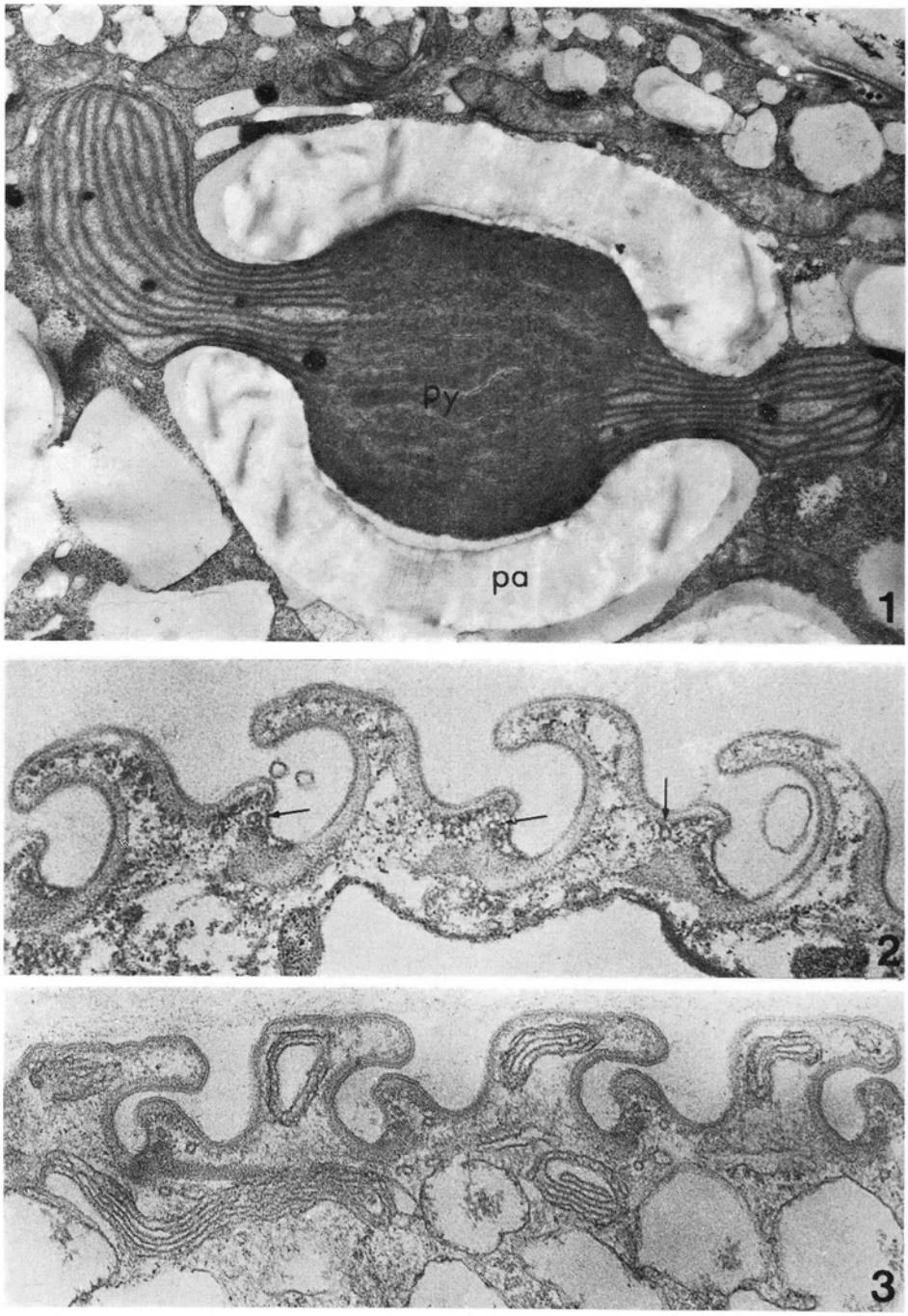
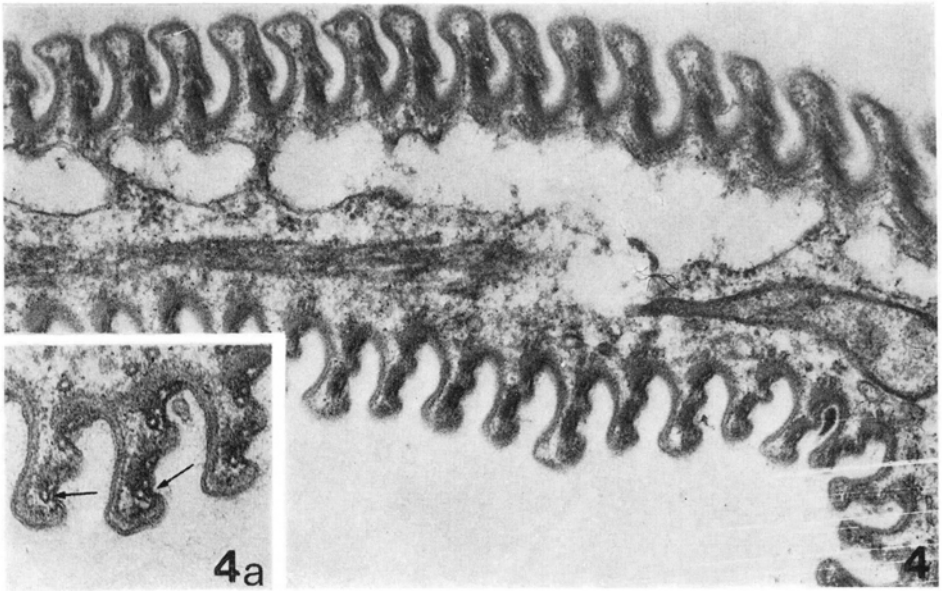
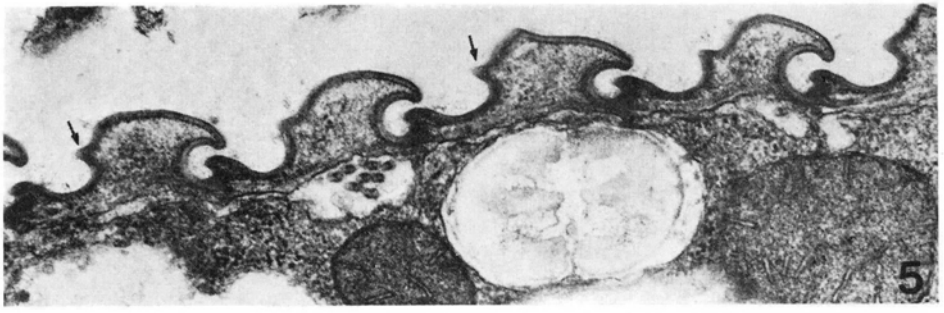
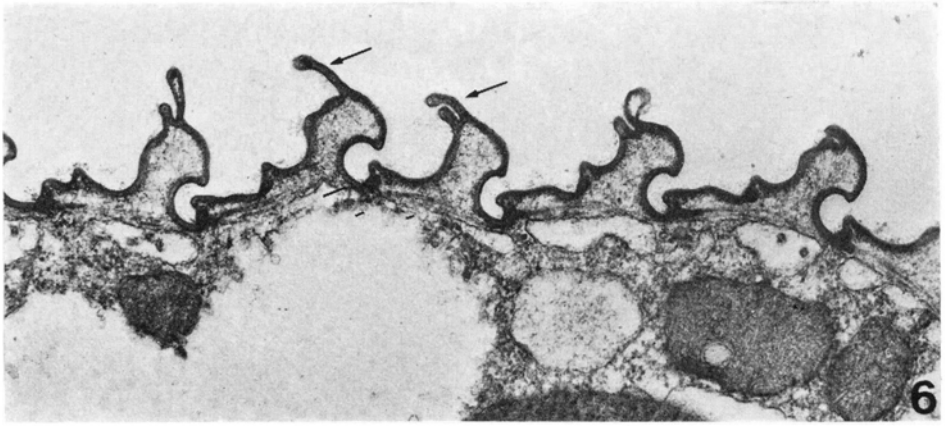


Fig. 4. — Pellicle of *Euglena sp.,* around the opening of the gullet. Here the ridges become erect and form uniform rows, showing clearly that the formations described previously as smaller ridges are actually protuberances at the base of the longer ones. Inside the gullet, as ridge inclination changes, the position of microtubules also changes, from dorsal to ventral (inset, arrows). × 50,000; inset × 80,000.

Fig. 5 and 6. — Pellicle of *E. polymorpha,* as seen in transverse section. Here too the pellicle seems to form two series of ridges: the larger ones have a beak-like outline, with a tiny tooth (Fig. 5, arrow), which may change to a ribbon-like appendix (Fig. 6, arrow). Elements of the SER run just under the ridges. Fig. 5: × 30,000; Fig. 6: × 25,000.



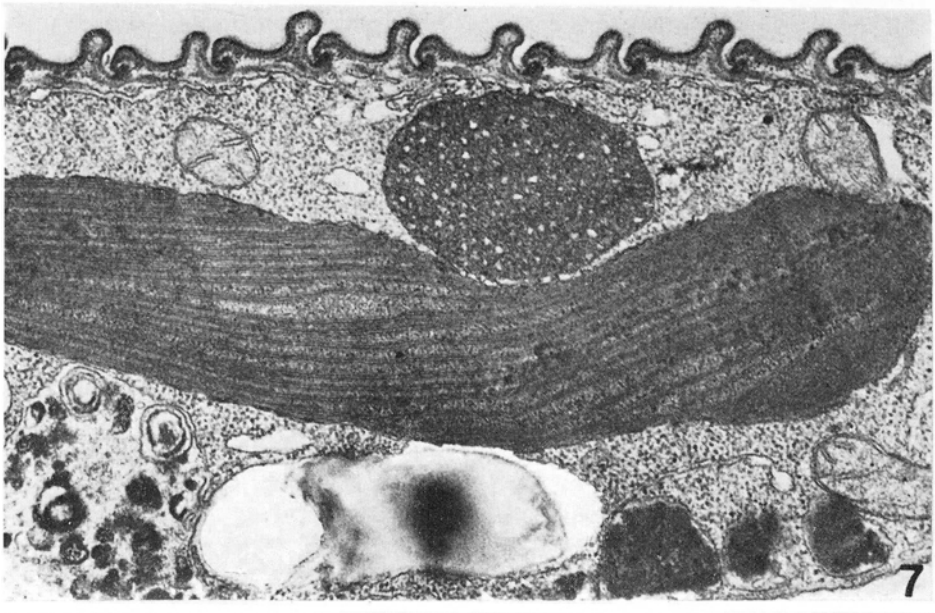


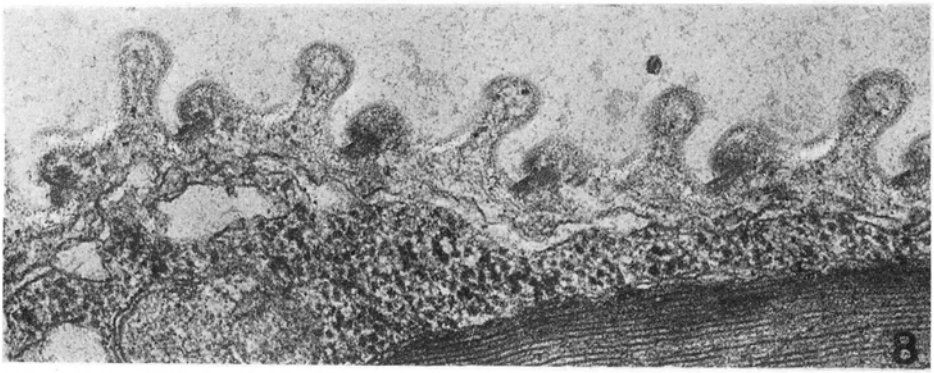


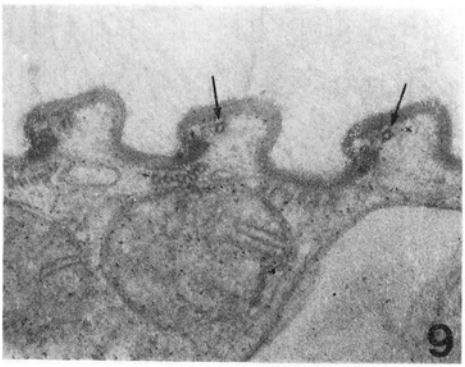
Figs. 7 and 8. — Pellicle of *E. caudata*, in transverse section. The ridges are rather flattened, and their outline shows two series of knob-like protuberances, one more protruding than the other. SER elements are visible. Fig. 7: × 24,000; Fig. 8: × 50,000.

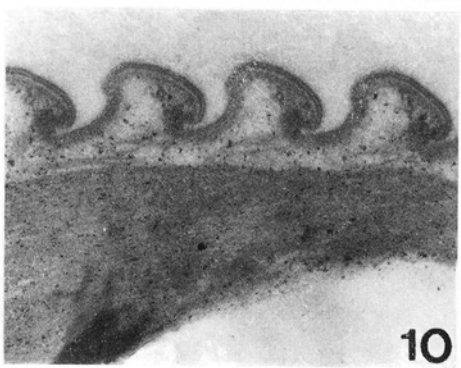
Fig. 9. — Pellicle of *E. geniculata*, in transverse section. The pellicle outline shows a single series of thick and rather distant ridges, each one containing a single microtubule in their middle outer portion and microtubules in the notch region of the groove (arrows). Five or six more microtubules are aligned just under the groove. × 40,000.

Fig. 10. — Pellicle of *E. mutabilis*, in transversal section. The outline of the pellicle is similar to that of *E. geniculata*, but the ridges are closer to each other. One microtubule is visible in the outer middle portion of the ridge. × 40,000.









other, the beak arching over the finger. The larger ridges bear also a tiny, tooth-like protuberance on the opposite side of the beak (Fig. 5). Often these tiny protuberances become thin, ribbon-like appendixes (Fig. 6). Microtubules are visible at the base of the groove, in the notch region. Elements of the SER run along the cytoplasmic surface.

E. caudata. — The pellicle, in transverse section, appears as two series of knobs: longer, protruding knobs with a neck and a round head and shorter, round ones without neck (Fig. 7). The longer knobs are generally erect or only slightly bent over the shorter ones (Fig. 8). Numerous elements of the SER run at the base of the ridges.

E. geniculata. — The pellicle forms regular, rather coarse ridges, with slightly flattened surface. Each ridge contains one microtubule, just under the outer surface, and microtubules along the notch region of the groove. A row of 5 to 6 microtubules is located in the peripheral cytoplasm, just under the groove (Fig. 9).

E. mutabilis. — The pellicular ridges are very regular, knob-like and slightly bent towards the notch of the groove. They contain one microtubule in their outer middle portion (Fig. 10).

DISCUSSION

The three *Euglena* species belonging to the group Catilliferae have similar pellicle ultrastructure, which distinguishes them from the other species belonging to different groups. At first sight, their pellicle seems to form a double series of strips, one more prominent than the other, but on closer examination of suitably cut sections, it can be deduced that they represent a temporary modification of the pellicle, because where the latter is less stretched, such as around the canal opening, it becomes apparent that the knob-like ridge is actually a side protuberance of the more prominent hook-like one. In other words, when the pellicular strips flatten during cell movement, their side protuberances may become so widely separated from the edge of the strips, as to appear as a self-standing series of less prominent strips.

Anyway, this feature, which might be taken as the expression of a particularly intense metabolic activity, cannot be considered as peculiar to *Euglena* belonging to the group Catilliferae, because it was found also in a species belonging to the group Rigidae (*E. spyrogyra*) (LEEDALE 1964), and, though present in another species of Catilliferae (*E. granulata*) (WALNE and ARNOTT 1964) it was never described in *E. gracilis*, the most studied member of the group.

A peculiarity of *Euglena sp.* is the presence of profiles of the SER in the interior of each pellicular ridge. Though elements of the SER are commonly found at the periphery of *Euglena* cells (BUETOW 1968), they have never been described inside the pellicular strips. The significance of this localization is obscure. The hypothesis that it might be related to mucilage secretion (*Euglena sp.* forms thick cysts) seems improbable because it was not observed in other cases of cyst formation (GEROLA *et al.* 1980*a* and 1980*b*).

To conclude, neither the general aspect of the pellicular strips nor the presence or absence of a flange or the number of pellicular microtubules are indicative of the group. Therefore, pellicle ultrastructure must be considered a valuable differential character only at the level of species. The pellicular aspect of *Euglena sp.* and of other *Euglena spp.* (*E. tonzigi* and *E. pinetana*) found in the red neuston of two alpine water bodies (GEROLA *et al.* 1980*a* and 1980*b*) gives support to our view, that a red neuston can be formed by intermingled different species, one prevailing over the other. This observation confirms the great importance of electron microscopy in systematic studies, and, on the other hand, explains the fact that, as Pringsheim says, the species *E. sanguinea* Ehrenb. « has caused more confusion than any other ».

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