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Electron microscopy in oceanographic research

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The phytoplankton of offshore waters is composed of unicellular species encompassing a broad size range. The larger forms are caught by fine-meshed nets, while the smaller forms are obtained by sedimentation of water samples and examined by means of inverted microscopes. After subsequent transfer to regular microscopes, the minute specimens can be examined with the best optical equipment available. Even then it is obvious that in many oceanic phytoplankton species the morphological details are too fine to be adequately observed in the light microscope. The taxonomical treatment of the nannoplankton component of oceanic populations, which includes representatives of several taxonomic groups, has, therefore, been inadequate. Recently the electron microscope has been introduced in this field and has shed light on many of the problems encountered.

As a rule diatoms are dealt with without difficulty in light microscopes, but in some cases they are so small that adequate descriptions cannot be produced. An example of this is *Fragilaria nana* Steemann Nielsen. This species forms an important part of the summer vegetation in oceanic waters of the North Atlantic, including the Norwegian Sea,(Steemann Nielsen, 1935; Halldal, 1953). Indentification of this species and similar small diatoms is now possible by means of electron microscopy (Halldal and Markali, 1955 a).

In the case of the " guinea pig " *Nitzschia closterium* f. *minutissima*, which has been used extensively for experimental work, electron microscope observations by Hendey (1954) have revealed that it does not belong to the diatoms at all. Consequently the numerous physiological observations on this species, now referred to as *Phaeodactylum tricornutum* Bohlin, can no longer be considered relevant for diatoms.

The extensive electron-microscopical studies of the thecal structure in diatoms (for literature see Helmcke and Krieger, 1953–54) will doubtless prove useful in future marine plankton research.

The systematic group which has represented the greatest obstacle in the study of oceanic phytoplankton is the chrysophyceans and especially the coccolithophorids.

Naked chrysophyceans have been cultured, and in the electron microscope interesting morphological details of importance for the systematics of the group, such as the structure of their flagella, have been observed (Parke, 1954).

In the study of coccolithophorids, which form such an important part of the phytoplankton in warm seas, electron microscopy has proved especially useful. The calcified coccoliths of these forms represent the most characteristic morphological feature of their cell structure, and from the time of the first description of species of this group they have been used in identification and systematical grouping. The first electron microscope pictures of coccoliths (Braarud and Nordli, 1952; Kamptner, 1952) made it evident at once that, for this group, electron microscopy would initiate a new era for the study of taxonomy, systematics and phylogeny.

During the last few years observations have been made on approximately 50 species (for literature see HALLDAL and MARKALI, 1955 B). A new terminology for the description of the coccolith morphology has been introduced, and the first outline has been drawn up for a coccolith typology based upon electron microscope observations (BRAARUD, DEFLANDRE, HALLDAL and KAMPTNER, 1954; HALLDAL and MARKALI, 1955 B). To a large extent the old coccolith types have had to be discarded. It has also been revealed that some of the old species actually include more than one taxonomical unit. It seems obvious that, in future, electron microscopy will be indispensable when describing new species of this group.

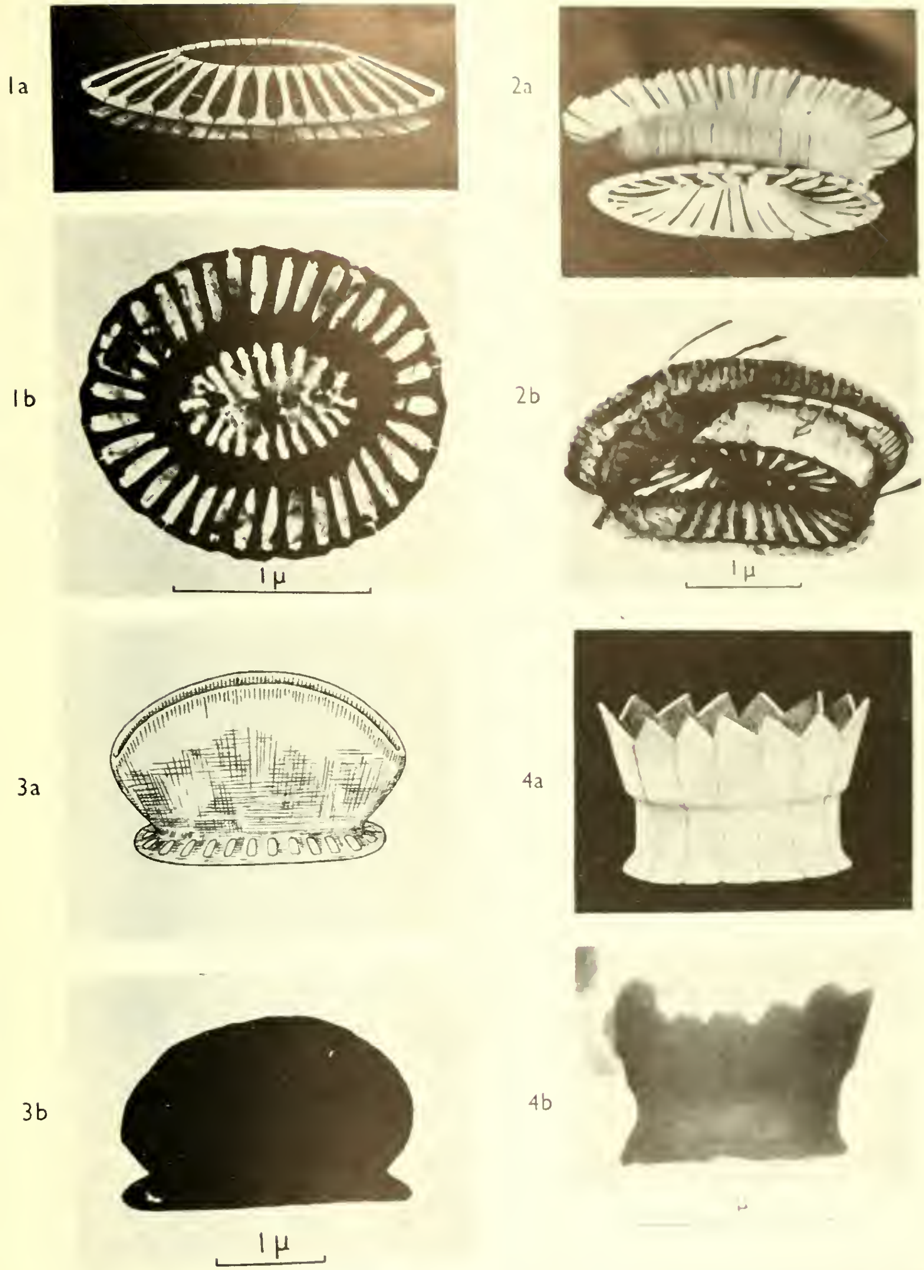
Electron microscope observations have disclosed an unforeseen variety in the microstructure of the coccoliths, and definite taxonomic groups based upon clear-cut differences in the microstructure of the coccoliths can already be distinguished. In Pl. I and II are shown examples of some coccolith types which illustrate the striking diversity in their architecture. Species which, because of their coccolith microstructure, must be assumed to be closely related, may exhibit increasing differentiation in the coarser features of their coccolith morphology. As an example Pl. II, 1–6, shows coccoliths from four species, all of which have coccoliths of the holococcolith type, ranging from *Crystallolithus hyalinus* with its very simple coccoliths to *Homozygosphaera tholifera* with its elaborate " mitras ". In this case the microstructure of the coccoliths, which can be revealed only through electron microscopy, must form the basis for a revised systematical treatment of the group. The extremely different microstructure of such coccoliths as shown in Pl. I and II may, in time, give proof of a more complex origin of the coccolithophorids than hitherto suspected.

The study of this group with the electron microscope is at its beginning. Much work is still ahead before full advantage can be taken of this new tool, but already one may point to useful applications of these results in other fields, as for instance the study of coccoliths in rocks and in oceanic sediments of different ages.

Coccoliths form a part of sediments in the oceans, and of calcareous rocks from various periods (DEFLANDRE, 1952). In most cases the coccoliths are found detached, but since observations on the planktonic forms of today have established the specific nature of the coccolith structures, their identification is now possible through electron microscopy. Considerable work has already been done on fossil forms (for literature see DEFLANDRE and FERT, 1954). In some cases their coccoliths have been found to be identical with those of species living in the oceans today, while in other cases they are different from any yet observed in living plankton.

In examining stratification in oceanic sediments the study of the small coccoliths, which hitherto have been unidentifiable, may become most useful.

The dinoflagellates, another important group of marine phytoplankton, for the most part are large enough to be studied adequately in the light microscope. However, some oceanic species are so small that essential morphological features escape observation. *Exuviaella baltica*, a species which has a wide distribution in the North Atlantic and which forms an important component of the summer populations (BRAARUD, GAARDER and GRØNTVED, 1953; HALLDAL, 1953), has been shown to have a spiny surface structure which was quite unexpected from observations in the light microscope (see Pl. II, 7). This morphological detail may be of considerable ecological importance in view of the fact that the spines increase the absorbing surface of the cell. In part, an explanation of the wide distribution of this species and its growth in



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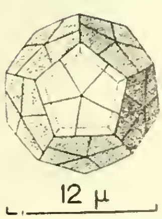
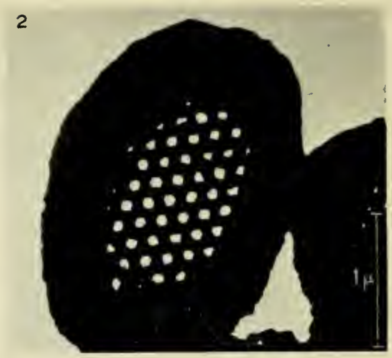
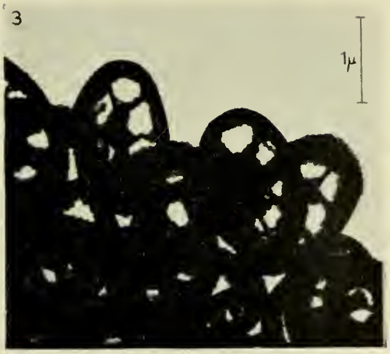
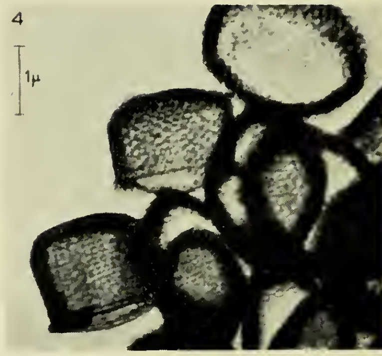
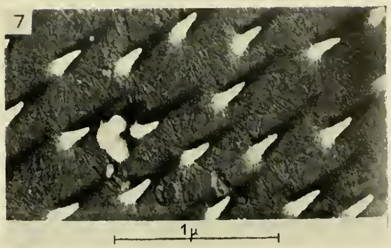


Pl. I 1. *Coccolithus huxleyi.* (*a*) drawing and (*b*) electron-micrograph of coccolith. (From Braarud, Gaarder, Markali and Nordli, 1952.)
2. *Syracosphaera mediterranea.* (*a*) model and (*b*) electron-micrograph of coccolith. (From Halldal and Markali 1954 b.)
3. *Anthosphaera robusta* (*a*) model and (*b*) electron-micrograph of coccolith. (From Halldal and Markali 1954 a.)
4. *Hymenomonas roseola* (*a*) drawing and (*b*) electron micrograph of coccolith. (From Braarud, 1954.)
5. *Braarudosphaera bigelowi* Drawing of cell with coccoliths (From Deflandre et Fert, 1954.)









Pl. 11 1. *Crystallolithus hyalinus.* Electron-micrograph of coccolith. (From GAARDER and MARKALI, 1955.)

2. *Sphaerocalyptra papillifera.* Electron-micrograph of coccolith. (From HALLDAL and MARKALI, 1954 A.)

3. *Homozygosphaera wettsteini.* Electron-micrograph of coccoliths. (By courtesy of J. MARKALI.)

4. *Calyptrosphaera oblonga.* Electron-micrograph of coccoliths. (From HALLDAL and MARKALI, 1955 B.)

5. *Homozygosphaera triarcha.* Electron-micrograph of coccolith. (From HALLDAL and MARKALI, 1955 B.)

6. *Homozygosphaera tholifera.* Electron-micrograph of coccolith. (From HALLDAL and MARKALI, 1955 B.)

7. *Exuviaella baltica.* Detail of electron-micrograph of theca. (From BRAARUD, MARKALI and NORDLI, 1955.)

waters poor in nutrients may be found in this feature (BRAARUD, MARKALI and NORDLI, 1955).

Electron microscopy has opened new vistas for the student of oceanic phytoplankton populations. It will lead to a revision of the taxonomy of the many minute forms, and a sound basis for ecological surveys may thus be established. It is not difficult to foresee that in experimental studies on the physiological and ecological characters of marine phytoplankton as well, the electron microscope will turn out to be a valuable tool.

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