Type 3—Cell Surface with Additional Intracellular Material in Vesicles

In this type of cell surface, the plasma is underlined by a system of flattened vesicles. An example is the complex outer region of dinoflagellates (Dinophyceae) termed amphiesma. Beneath the cell membrane that binds dinoflagellate motile cells, a single layer of vesicles (amphiesmal vesicles) is almost invariably present. The vesicles may contain cellulosic plates (thecal plates) in taxa that are thus termed thecate, or armored; or the vesicles may lack thecal plates, such taxa being termed athecate, or unarmored or naked. In athecate taxa, the amphiesmal vesicles play a structural role. In thecate taxa, thecal plates, one of which occurs in each vesicle, adjoin one another tightly along linear plate sutures, usually with the margin of one plate overlapping the margin of the adjacent plate. Cellulosic plates vary from very thin to thick and can be heavily ornamented by reticula or striae; trichocyst pores, which may lie in pits termed areolae, penetrate most of them.

A separate layer internal to the amphiesmal vesicles may develop. It is termed pellicle, though in the case of dinoflagellates the term "pellicle" refers to a surface component completely different from the euglenoid pellicle, hence with a completely different accepted meaning, and in our opinion its use should be avoided. The layer consists primarily of cellulose, sometimes with a dinosporine component, a complex organic polymer similar to sporopollenin that makes these algae fossilizable. In some athecate genera, such as *Noctiluca* sp., this layer forms reinforce the amphiesma, and the cells are termed pelliculate. This layer is also sometimes present beneath the amphiesma, as in *Alexandrium* sp., or *Scrippsiella* sp., and forms the wall of temporary cysts.

According to Dodge and Crawford (1970), the amphiesma construction falls into eight reasonably distinct categories (Figure 2.17): (1) simple membrane underlain by a single layer of vesicles 600–800 nm in length, rather flattened, circular, or irregular in shape, with a gap of at least 40 nm between adjacent vesicles that may contain dense granular material; beneath the vesicles are parallel rows of microtubules which lie in groups of three; this simple arrangement is present in *Oxyrrhis marina*; (2) simple membrane underlain by closely packed polygonal (generally hexagonal) vesicles 0.8–1.2 μm in length, frequently containing fuzzy material; these vesicles and the cell membrane are occasionally perforated by trichocyst pores; beneath the vesicles lie microtubules in rows of variable number; this type of amphiesma is found in *Amphidinium carterae*; (3) as in category (2), but with plug-like structures associated with the inner side of the vesicles; these plugs are cylindrical structures 120-nm long and are arranged in single lines between single or paired microtubules; an example of this arrangement is present in *Gymnodinium veneficum*; (4) as in category (2), but with thin (about 20 nm) plate-like structure in the flattened vesicles; this amphiesma characterizes *Aureodinium pigmentosum*; (5) in this group, the vesicles contain plates of medium thickness (60 nm), which slightly overlap; in *Woloszynskia coronata*, the plates are perforated by trichocyst pores; (6) the plates are thicker (up to 150 nm), reduced in number with a marked diversity of form; each plate has two or more sides bearing ridges and the remaining sides have tapered flanges; where

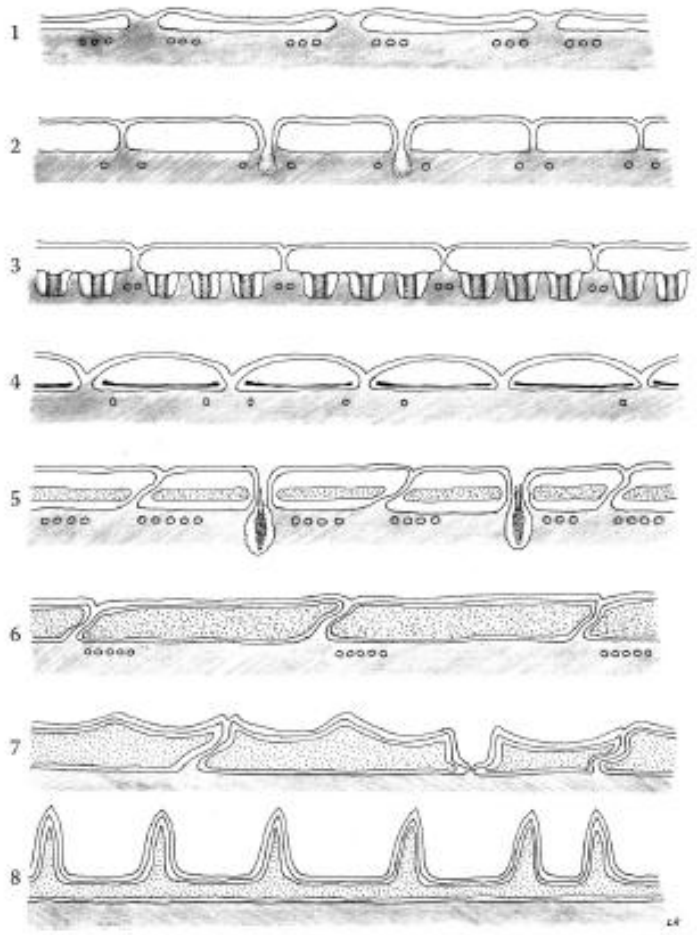

FIGURE 2.17 Diagram of the eight distinct category of the dinoflagellate amphiesma. See text for details.

the plates join, one plate bears a ridge and the opposite bears a flange; *Glenodinium foliaceum* belongs to this category; (7) the plates can be up to 25-μm large and up to 1.8-μm thick; they bear a corrugated flange on two or more sides, and a thick rim with small projections on the opposing edges; these plates may overlap to a considerable extent and their surfaces may be covered by a pattern of reticulations; a distinctive member of this category is *Ceratium* sp.; (8) amphiesma consisting of two large plates, with one or more small plates in the vicinity of the flagellar pores at the anterior end of the cell; plates can be very thin and perforated by two or three simple trichocyst pores as in *Prorocentrum nanum*, or thick and with a very large number (up to 60) of trichocyst pores as in *Prorocentrum micans*.

The arrangement of thecal plates is termed tabulation and is of critical importance in taxonomy of dinoflagellates. Tabulation can also be conceived of as the arrangement of amphiesmal vesicles with or without thecal plates. The American planktologist and parasitologist Charles Kofoid developed a tabulation system allowing reference to the shape, size, and location of a particular plate; plates were recognized as being in series relative to particular landmarks such as the apex, cingulum (girdle), and sulcus. His formulae (i.e., the listing of the total number of plates in each series) were especially useful for most gonyaulacoid and peridinioid dinoflagellates. Apart from some minor

changes introduced afterwards, the Kofoid system is still the standard in the description of new taxa. Plates are numbered consecutively from that closest to the midventral position, continuing around to the cell left. A system of superscripts and other marks are used to designate the plate series. Two complete

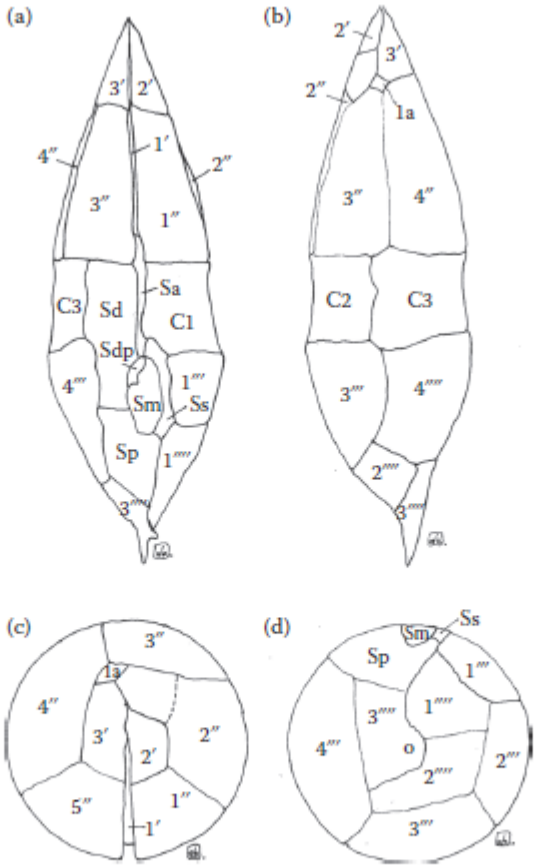

FIGURE 2.18 Line drawings of the thecal plate patterns of *Lessardia elongata* with the corresponding numeration: (a) ventral view; (b) dorsal view; (c) apical view; and (d) antiapical view. See text for details.

transverse series of plates are present in the epitheca: apical (′), and precingular (″), counted from the ventral side in a clockwise sequence. The hypotheca is also divided into two transverse series: postcingular (‴) and antapical (⁗). Some genera also possess an incomplete series of plates on the dorsal surface of the epitheca, termed anterior intercalary plates (a), and on the hypotheca, termed posterior intercalary plates (p). Cingular (C) and sulcal (S) plates are also identified (Figure 2.18). Thus, for example, the dinoflagellate *Proteperidinium steinii* has a formula 4′, 3a, 7″, 3C, 6S, 5‴, 2⁗, which indicates four apical plates, three anterior intercalary plates, seven precingular plates, three cingular plates, six sulcal plates, five postcingular plates, and two antapical plates.

Type 4—Cell Surface with Additional Extracellular and Intracellular Material

Both the surface structure of Cryptophyceae (Cryptophyta) and Euglenophyceae (Euglenozoa) can be grouped under this type.The main diagnostic feature of the members of the Cryptophyceae is their distinctive kind of cell surface, colloquially termed periplast. Examples are *Chroomonas* (Figure 2.19) and *Cryptomonas*; in these algae, the covering consists of outer and inner components, present on both sides of the membrane but variable in their composition. The inner component is comprised of protein and may consist of fibril material, a single sheet or multiple plates having various shapes, hexagonal, rectangular, oval, or round. The outer component may have plates, heptagonal scales, mucilage, or a combination of any of these. The pattern of these plates can be observed on the cell surface when viewed with scanning electron microscope (SEM) and freeze-fracture transmission electron microscope (TEM), but it is not easily detectable under light microscopy view.

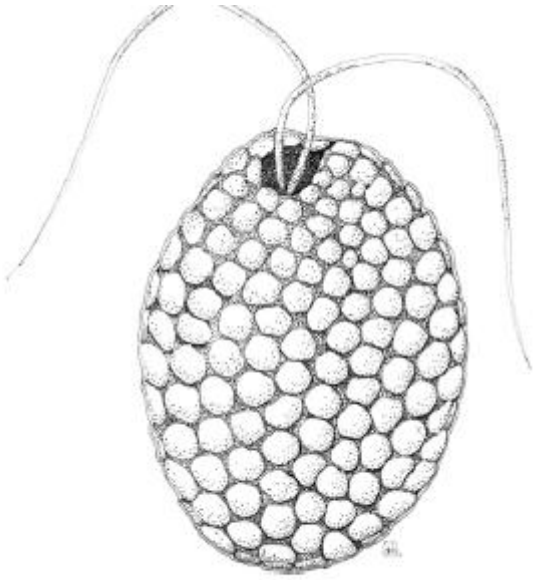

FIGURE 2.19 Periplast of *Chroomonas* sp.

These strips or striae can be described as long ribbons usually arising in the flagellar pocket and extending from the cell apex to the posterior. Each strip is curved at both its edges, and in transverse section it shows a notch, an arched, or a slightly concave ridge, a convex groove and a heel region where adjacent strips interlock and articulate. The strips can be arranged helically or longitudinally; the first arrangement, very elastic, is present in the "plastic euglenoids" (e.g., *Euglena*, *Peranema*, *Distigma*), either heterotrophic or phototrophic, where the strips are more than 16. Their relational sliding over one another along the articulation edges permit the cells to undergo "euglenoid movement" or "metaboly." This movement is a sort of peristaltic movement consisting of a cytoplasmic dilation forming at the front of the cell and passing to the rear. The return movement of the cytoplasm is brought about without dilation. The more rigid longitudinal arrangement is present in the "aplastic euglenoids" (e.g., *Petalomonas*, *Pleotia*, *Entosiphon*), all heterotrophic, where the strips are usually less than 12. These euglenids are not capable of metaboly.

The ultrastructure of the pellicular complex shows three different structural levels (Figure 2.20):

The plasma membrane with its mucilage coating (first level)
An electron-opaque layer organized in ridges and grooves (second level)
The microtubular system (third level)

First Level

A dense irregular layer of mucilaginous glycoproteins covers the external surface of the cell. It has a fuzzy texture that, however, has a somehow ordered structure of orientated threads. Mucilage bodies present beneath the cell surface secrete the mucilaginous glycoproteins. The consolidation of the secretory products and their arrangement at one pole or round the periphery of the cell leads to the formation of peduncles (stalks of fixation) and other enveloping structures homologs to the loricas of Chrysophyceae and Chlorophyceae. Peduncles are present in *Colacium*, an

euglenophyte that forms small arborescent colonies (Figure 2.21). Its cells, with reduced flagella, are attached by their anterior pole by a peduncle consisting of an axis of neutral polysaccharides and a cortex of acid polysaccharides. Loricas are present in *Trachelomonas* sp. (Figures 1.1bm and 2.22), *Strombomonas verrucosa* (Figure 2.23) and *Ascoglena*; they are very rigid, made up of mucilaginous filaments impregnated with ferric hydroxide or manganese compounds which confer an orange, brown to black coloration to

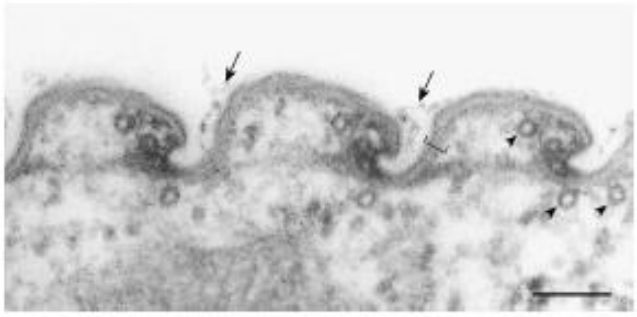

FIGURE 2.20 TEM image of the surface of *Euglena gracilis* in transverse section, showing the three different structural levels of the pellicle. Arrows point to the first level (mucus coating); a square bracket localizes the second level (ridges and grooves); arrowheads point the third level (microtubules). Scale bar, 0.10 µm.

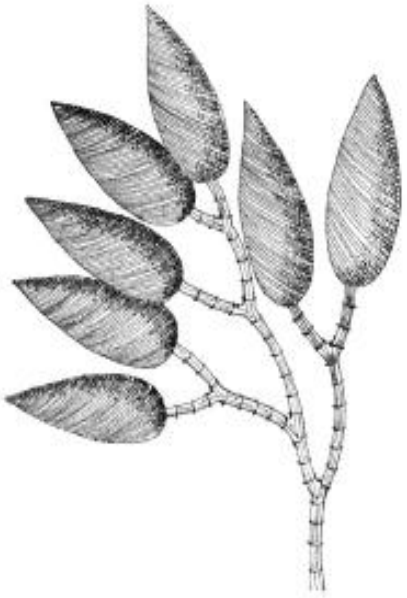

FIGURE 2.21 A small arborescent colony of *Colacium* sp. in which the cells are joined to one another by mucilaginous stalks.

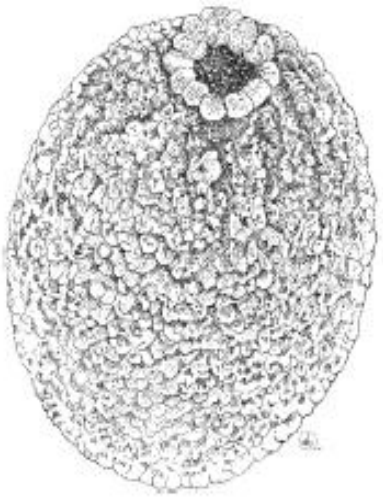

FIGURE 2.22 Lorica of *Trachelomonas* sp.

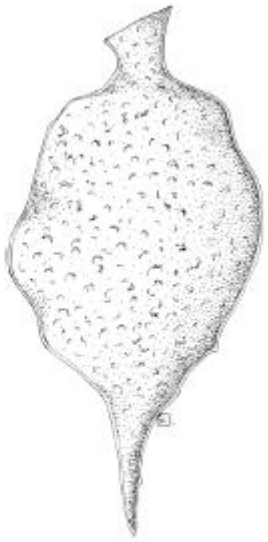

FIGURE 2.23 Lorica of *Strombomonas verrucosa*.

the structure. These loricas fit loosely over the body proper of the cell. They possess a sharply defined collar that tapers to a more or less wide apical opening, where the flagella emerge, or possess a wide opening in one pole and attached to a substrate at the other pole, as in *Ascoglena*.

Beneath the mucus coating, there is the plasma membrane (Figure 2.24). This cell membrane is continuous and covers the ridges and grooves on the whole cell and can be considered the external surface of the cell. The protoplasmic face (PF) of the plasma membrane shows that the strips are covered with numerous peripheral membrane proteins of about 10 nm.

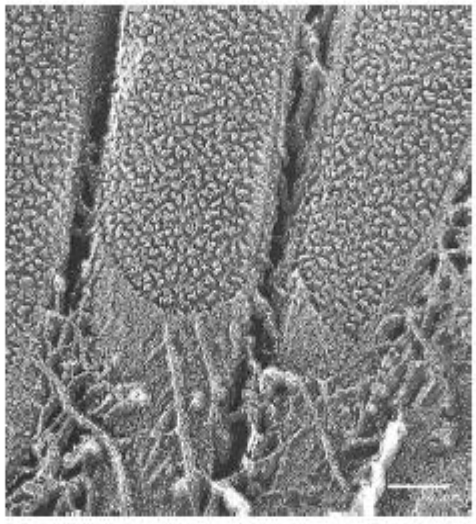

FIGURE 2.24 Deep-etching image of *Euglena gracilis* showing the mucus coating of the cell surface and the protoplasmic fracture of the cell membrane. Scale bar, 0.10 µm. (Courtesy of Pietro Lupetti.)

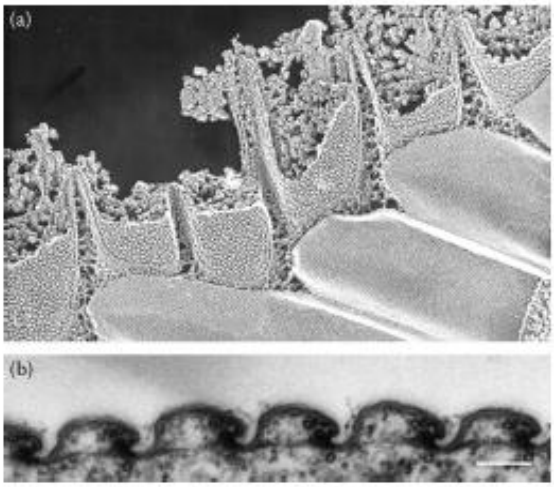

FIGURE 2.25 (a) Deep-etching image of *Euglena gracilis* showing the second structural level of the pellicular complex, showing the regular texture of the internal face of the pellicle stripes. (b) TEM image of the pellicle of *E. gracilis* in transverse section showing the transversal fibers connecting the edges of successive ridges. Scale bar, 0.10 µm.

Second Level

This peripheral cytoplasmic layer has a thickness that varies with the species. It consists of roughly twisted proteic fibers with a diameter from 10 to 15 nm arranged with an order texture or parallel striation (Figure 2.25a). The overall structure resembles the wired soul present in the tires, which gives the tire its resistance to tearing forces. Transversal fibers are detectable in some euglenoids, which connect the two longitudinal edges of the ridge of each strip (Figure 2.25b).

Third Level

There is a consistent number and arrangement of microtubules associated with each pellicular strip, which are continuous with those that line the flagellar canal and extend into the region of the reservoir. Within the ridge in the region of the notch, there are 3–5, usually 4, microtubules about 25-nm diameter running parallel along each strip. Two of these are always close together and are located immediately adjacent to the notch adhering to the membrane (Figure 2.20).

The lack of protein organization in the groove regions gives higher plasticity to these zones, and together with the presence of parallel microtubules in the ridge regions gives the characteristic pellucular pattern to the surface of euglenoids.

The solid structure of the pellicle confers a very high degree of flexibility and resistance to the cells. Our experience with *E. gracilis* allow us to say that this alga possesses one of the strongest covering present in these microorganisms. A pressure of more than 2000 psi (about 150 bar) is necessary to break the pellicular structure of this alga.

Flagella and Associated Structures

Flagella can be defined as motile cylindrical appendages found in widely divergent cell types throughout the plant and animal kingdom, which either move the cell through its environment or move the environment relative to the cell.

Motile algal cells are typically biflagellate, although quadriflagellate types are commonly found in green algae; it is generally believed that the latter have been derived from the former, and a

FIGURE 2.25 (a) Deep-etching image of *Euglena gracilis* showing the second structural level of the pellicular complex, showing the regular texture of the internal face of the pellicle stripes. (b) TEM image of the pellicle of *E. gracilis* in transverse section showing the transversal fibers connecting the edges of successive ridges. Scale bar, 0.10 μm.

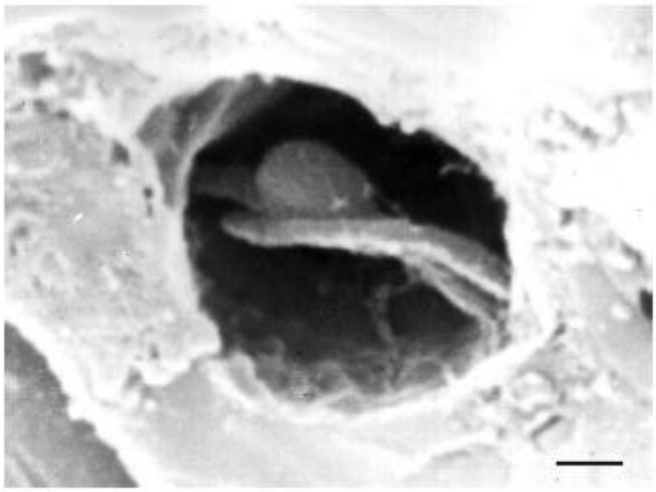

FIGURE 2.26 SEM image of the reservoir of *Euglena gracilis* in longitudinal section showing the locomotory emerging flagellum bearing the photoreceptor and the nonemerging flagellum reduced to a stub. Scale bar, 0.50 μm. (Courtesy of Franco Verni.)

convincing example of this derivation is *Polytomella agilis* (Chlorophyceae) from *Chlamydomonas* sp. (Chlorophyceae). A triflagellate type of zoospore such as that of *Acrochaete wittrockii* (Ulvophyceae) may have originated from a quadriflagellate ancestor by reduction, whereas the few uniflagellate forms are most likely descendant of biflagellated cells. Intermediate cases exist which carry a short second flagellum, as in *Mantoniella squamata* (Mamiellophyceae) or *Euglena gracilis* (Euglenophyceae), where one flagellum is reduced to a stub (Figure 2.26); in some species, one flagellum of the pair is reduced to a nonfunctional basal body attached to the functional one, as in the uniflagellate swarmer of *Dictyota dichotoma* (Phaeophyceae). A special case of multiflagellate alga is the naked zoospore of *Oedogonium* (Chlorophyceae), where the numerous flagella form a ring or crown around the apical portion of the cell (stephanokont zoospore).
The characteristics of the flagella in a pair, that is, relative length and surface features, have led to a specific nomenclature. When the two flagella differ in length and surface features, one being hairy and the other smooth, they are termed "heterokont." This term applies to all the members of the Ochrophyta. When the two flagella are equal in length and appearance, the term "isokont" is used (Figure 2.27), which applies to Haptophyta and to Chlorophyceae and Charopyceae. Within this group, there are few genera whose flagella differ in length, which are termed "anisokont."

Description of flagella anatomy will proceed from the outside to the inside, from the surface features and components to the axoneme and additional inclusions, to the structures anchoring the flagella to the cell.

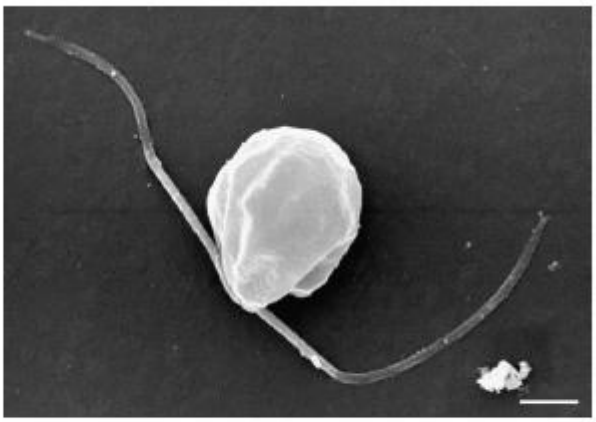

FIGURE 2.27 SEM image of an isokont cell (*Dunaliella* sp.). Scale bar, 3 µm.

How Algae Move

Cytoplasm, cell walls, and skeletons of algae have a density greater than the medium these organisms dwell in. The density of freshwater is 1.0 g cm−3 and that of seawater ranges from 1.021 to 1.028 g cm−3, but most cytoplasmic components have

a density between 1.03 and 1.10 g cm−3, the silica forming the diatom frustule and the scales of Chrysophyceae have a density of 2.6 g cm−3, and both calcite and aragonite of Haptophyta coccoliths reach an even higher value of 2.7 g cm−3. With this density values, algae must inevitably sink. Therefore, one of the problems planktonic organisms face (organisms that wander in the water and/or are carried about by the movements of the water rather than by their own ability to swim) is how to keep afloat in a suitable attitude between whatever levels are suitable for their life. The phytoplankton must obviously remain floating quite close to the surface because only there is a sufficient illumination for photosynthesis. There are broadly two solutions by which algae can keep afloat and regulate their orientation and depth: a dynamic solution, obtaining lift by swimming, and a static solution, by buoyancy control, or through adaptations reducing sinking rates. In many cases, the two solutions function together.