Chapter 2

The phycology of phytoplankton

Phytoplankon are the assemblage of photoautotrophic microorganisms making up the first trophic level of pelagic food chains. All belong to the botanical categories grouped as algae and studied by "phycologists". Compared to the roughly quarter million species of plants in terrestrial habitats, we find very few species of algae in the plankton, about 5000 (Tett & Barton 1995). Similar comparisons hold for zooplankton and fish, and an explanation will be sought below in the treatment of pelagic biogeography. Planktonic algae are classified into both ecological and botanical groups, sets associated functionally or taxonomically. We will mix those groupings in our study of what these algae are like. Representatives of most of the major algal *divisions* live as plankton. Divisions, the largest units of algal and plant classification and equivalent to the zoologist's phyla, are defined on both morphological and biochemical criteria. We will focus on a few ecologically dominant groups, some of them exactly parallel to the botanical ones, others not (Box 2.1).

The relative importance of phytoplankton groups varies with the ecological situation. The importance of the smaller phytoplankton has only been recognized recently, and, because of their size, some were not discovered until after 1979. We will examine each of these groups in considerable biological detail because of their large role in oceanic ecology. Often biological oceanographers must take off their ocean ecologist caps and put on marine lab T-shirts to masquerade as marine biologists. We'll do that now.

Evolution of phytoplankton

Globally, cyanobacteria and microalgae dominate marine photosynthesis. Cyanobacteria evolved about 2.85 billion years ago (Falkowski *et al.* 2004) and are simple prokaryotic cells without a membrane-bound nucleus or other cellular organelles. The microalgae are also single-celled organisms but have a more complex cell structure. They were actually formed as symbioses of photosynthetic prokaryotes or eukaryotes with heterotrophic eukaryotic hosts (Plate 2.1; Parker *et al.* 2008). Microalgae comprising the various taxonomic groups were formed by three types of these endosymbioses. In a primary endosymbiosis, a cyanobacterium was acquired by a heterotrophic eukaryote. In a secondary endosymbiosis, a eukaryotic heterotroph acquired a photosynthetic eukaryote. In a tertiary symbiosis, a dinoflagellate host engulfed a secondary endosymbiont, that provided its chloroplast to the dinoflagellate. The clue to this is that pigments and genes of the transferred chloroplasts match those of extant eukaryotic algae (Keeling 2010). In all of these symbioses, some or all genes from the chloroplasts were eventually transferred to the host nucleus.

Biological Oceanography, Second Edition. Charles B. Miller, Patricia A. Wheeler.

Box 2.1 Ecologically distinct major groups of phytoplankton

GROUP	PHYCOLOGICAL TERMINOLOGY
Picoplankton	Photoautotrophs <2μm
Cyanobacteria	Photosynthetic prokaryotes, size <2μm, from division Cyanophyta (lately "cyanobacteria") *Synechococcus* and *Prochlorococcus*
Eukaryotic picoplankton	Very small, but structurally advanced forms
Microflagellates	An ecological assemblage of several divisions and classes: Cryptophyta, Haptophyta, Prasinophyceae, Prymnesiophyceae
Diatoms	Bacillariophyceae from division Heterokontophyta
Dinoflagellates	Dinophyceae from division Dinophyta

Main constituents of marine phytoplankton

Picoplankton – both prokaryotic and eukaryotic

Cyanobacteria

Bacteria are prokaryotes, organisms in which the macromolecules carrying genetic information, deoxyribonucleic acid (DNA), are not held in a nucleus ("karyon"), a specialized organelle surrounded by a membrane. The relatively simple DNA-bearing chromosomes disperse in the central region of the cell, which is the basis of the term "prokaryotic". Bacteria are more fully characterized in Chapter 5. One group of bacteria, called cyanobacteria, produce organic matter by photosynthesis. In that sense, they are algae as well as bacteria, so botanists classify them as the division Cyanophyta, the blue-green algae. Their photosynthetic pigments are arrayed in layers, thylakoids, around the cell periphery. Streams and ponds often support sizeable stocks of filamentous blue-green algae (*Nostoc*, *Anabaena*, and others), macroscopic forms familiar to many from introductory biology classes. It was not until surprisingly late that we realized cyanobacteria are important in marine, pelagic habitats.

Waterbury *et al.* and Johnson and Sieburth both reported in 1979 that very large numbers of photosynthetic bacteria can be counted in water samples from the ocean. These bacteria had been overlooked previously for several reasons, principally that they are small, all under 2μm and most about 1μm in diameter. Cyanobacterial thylakoids (Fig. 2.1) include alternate layers of phycobilisomes, particles of protein binding photosynthetic accessory pigments including phycoerythrin. This pigment has a characteristic orange fluorescence in blue light that is strong enough to mask the red fluorescence of chlorophyll. This makes cyanobacteria readily countable with an epifluorescence microscope, an important tool in the study of this group's ecology (see Box 2.2).

Now that oceanographers watch for *Synechococcus*, we find that it frequently constitutes half or more of the photosynthetic biomass in coastal and oceanic areas throughout the euphotic zone. Small size doubtless accounts for this importance. Because sinking (or buoyant rising) rate is proportional to the square of cell diameter (Stokes's Law), picoplankton sink or rise very slowly. Even very modest reproductive rates compensate for any net losses to depth at the *Synechococcus* sinking speeds of less than a centimeter per day (Raven 1985). Minuscule size also maximizes relative surface area for nutrient absorption and reduces grazing loss to suspension feeders. *Synechococcus* possesses a variety of phycocyanin and phycoerythrin pigments that allow the different strains to utilize the wide range in light quality naturally occurring over both horizontal (coastal–oceanic) and vertical gradients (Scanlan *et al.* 2009). Phycoerythrin is also abundant in the Rhodophyta, red algae, which are progressively more dominant with increasing depth in subtidal habitats nearshore. In fact, rhodophyte chloroplasts resemble cyanobacteria in the form of their thylakoids and the presence of phycobilisomes. It is likely that these chloroplasts have descended from an early, intracellular symbiosis of cyanobacterial cells in the macrophytes (Plate 2.1).

One genus of filamentous cyanophytes, *Trichodesmium*, lives in tropical seas. "Tricho", as it is known to students of its biology, can generate gas vacuoles and float at the sea surface. It may also regulate buoyancy to move up and

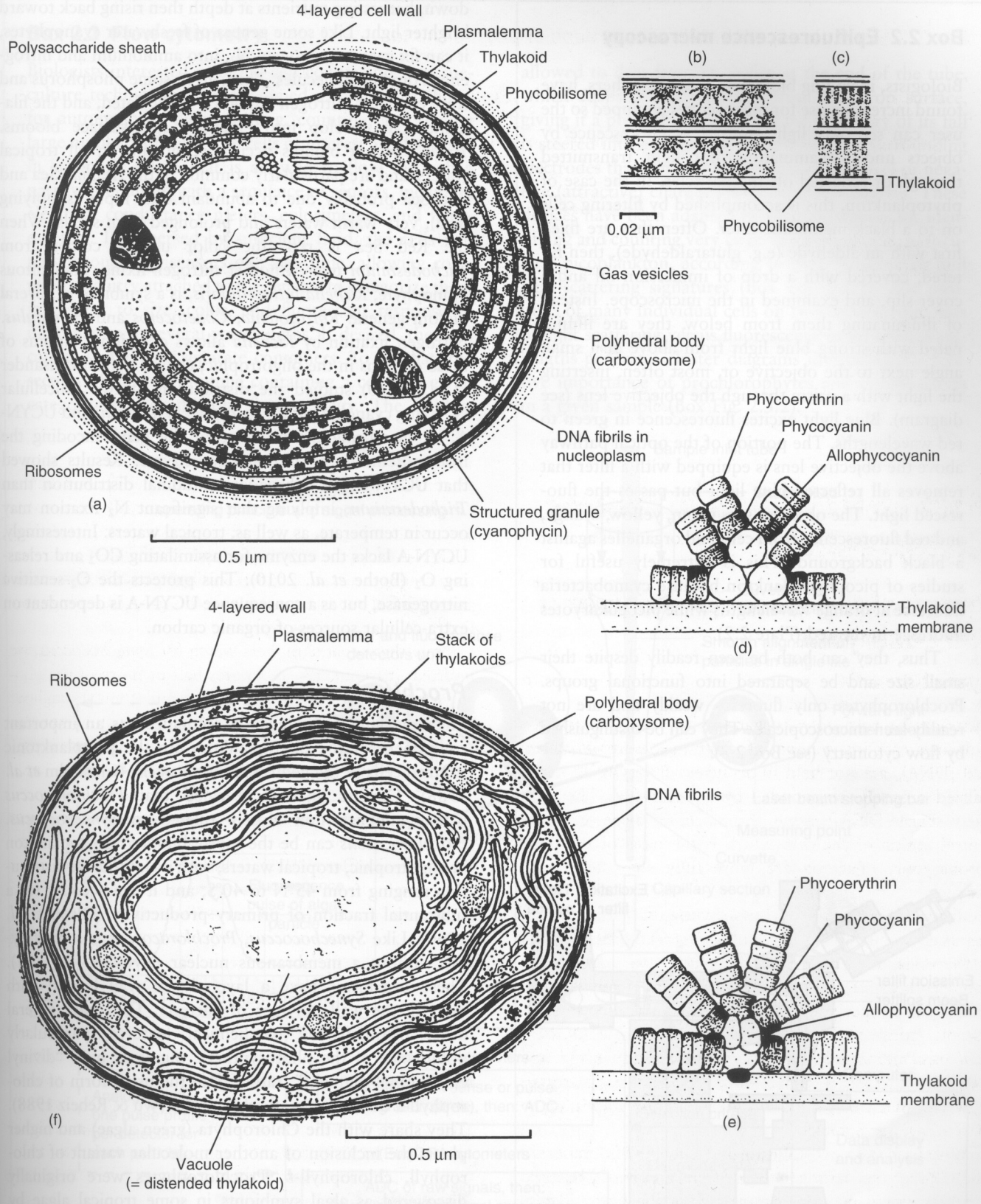
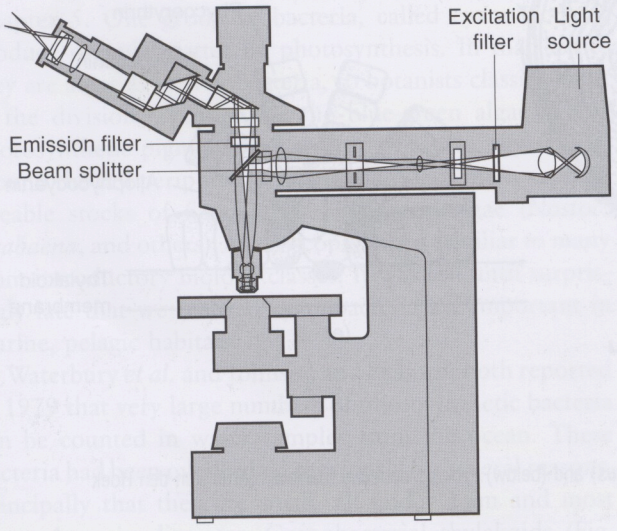


Fig. 2.1 Morphological plans of (above) single-celled cyanobacteria (*Synechococcus*) and (below) *Prochlorococcus marinus*. (After Van den Hoek *et al.* 1995.)

Box 2.2 Epifluorescence microscopy

Biologists, including biological oceanographers, have found increasing use for microscopes equipped so the user can see only light emitted as fluorescence by objects under examination, not light transmitted through or reflected off the particles. In the case of phytoplankton, this is accomplished by filtering cells on to a black membrane filter. Often they are fixed first with an aldehyde (e.g. glutaraldehyde), then filtered, covered with a drop of immersion oil and a cover slip, and examined in the microscope. Instead of illuminating them from below, they are illuminated with strong blue light from above at a small angle next to the objective or, most often, inserting the light with a prism through the objective lens (see diagram). Blue light excites fluorescence in green to red wavelengths. The portion of the optical pathway above the objective lens is equipped with a filter that removes all reflected blue light but passes the fluoresced light. The observer sees green, yellow, orange, and red fluorescence from cells and organelles against a black background. This is extremely useful for studies of picophytoplankton because cyanobacteria fluoresce at orange wavelengths while picoeukaryotes fluoresce in the red (Plate 2.2).

Thus, they can both be seen readily despite their small size and be separated into functional groups. Prochlorophytes only fluoresce weakly and are not readily seen microscopically. They can be distinguished by flow cytometry (see Box 2.3).



Box Fig. 2.2.1 Schematic of an epifluorescence microscope.

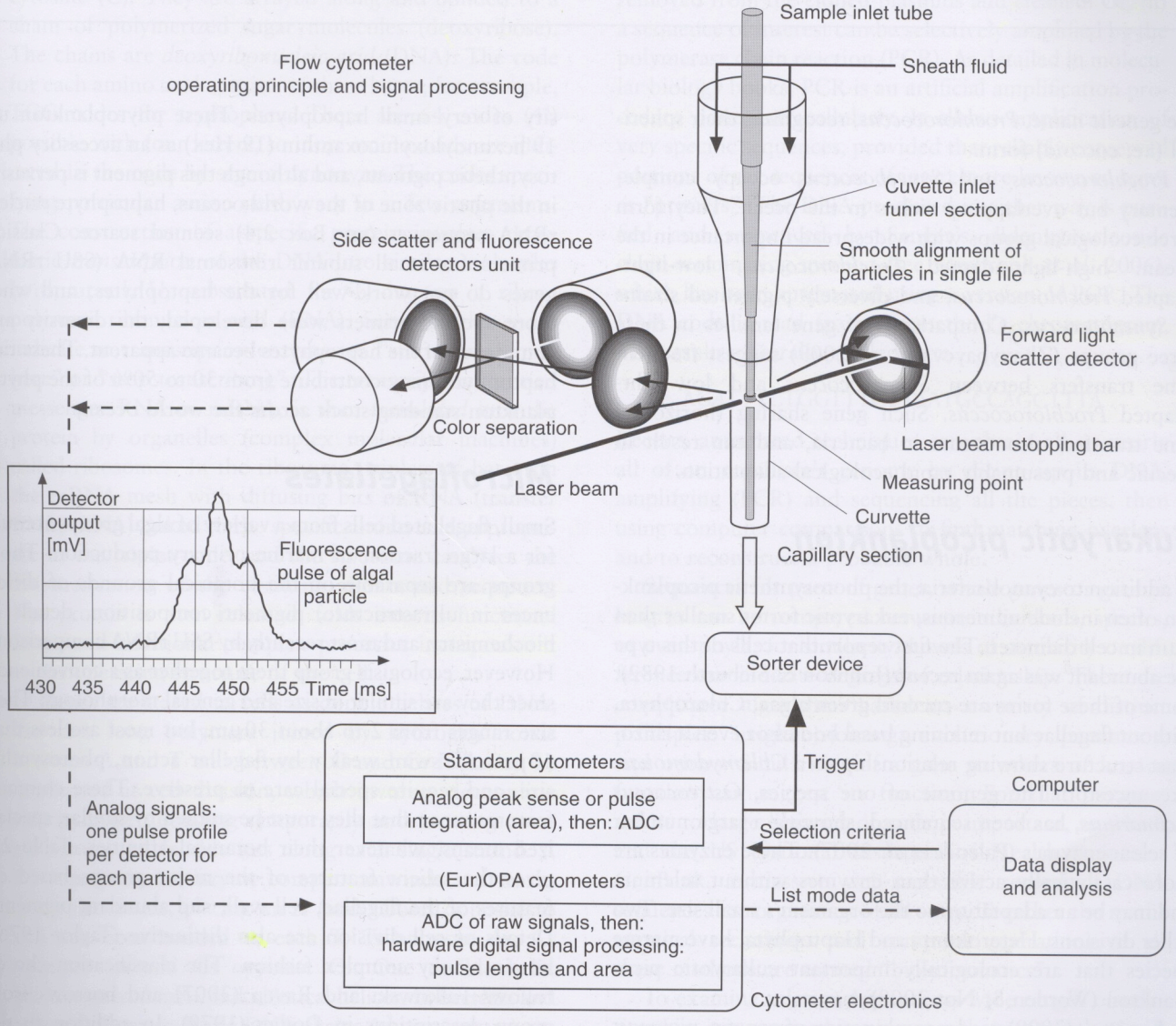
down, harvesting nutrients at depth then rising back toward brighter light. Like some genera of freshwater cyanophytes, it can fix gaseous nitrogen, N_2, into ammonium and nitrogenous organic molecules. Thus, given some phosphorus and trace elements, nitrogen limitation is alleviated, and the filaments can develop into extensive near-surface blooms. *Trichodesmium* blooms occur occasionally in all tropical waters, and are particularly common in the Arabian Sea and Red Sea probably due to abundant dust input (supplying iron), very warm waters and prolonged calm spells. When the Red Sea is red, the color usually comes from *Trichodesmium* mats. Another nitrogen-fixing, filamentous cyanophyte, *Richelia intracellularis*, is a symbiont of several diatom genera: *Rhizosolenia*, *Chaetoceros* and *Hemiaulus*. Nitrogen fixation by *Richelia* allows occasional blooms of these diatoms in the oligotrophic, central gyres. Moisander *et al.* (2010) studied the distribution of two unicellular N_2-fixing cyanobacteria, *Crocosphaera watsonii* and UCYN-A, by measuring the presence of the gene encoding the iron-protein in the nitrogenase enzyme. Results showed that UCYN-A has a broader latitudinal distribution than *Trichodesmium*, implying that significant N_2 fixation may occur in temperate, as well as, tropical waters. Interestingly, UCYN-A lacks the enzyme for assimilating CO_2 and releasing O_2 (Bothe *et al.* 2010). This protects the O_2-sensitive nitrogenase, but as a consequence UCYN-A is dependent on extra-cellular sources of organic carbon.

Prochlorococcus

The organisms most recently recognized as an important component of the phytoplankton are the picoplanktonic (<2 μm) cells of the genus *Prochlorococcus* (Chisholm *et al.* 1988). Genetic analysis revealed that *Prochlorococcus* is a cyanobacterium closely related to *Synechococcus*. *Prochlorococcus* can be the most abundant phytoplankton in oligotrophic, tropical waters, with its latitudinal distribution ranging from 45°N to 40°S, and it can account for a substantial fraction of primary productivity (Olson *et al.* 1990). Like *Synechococcus*, *Prochlorococcus* is a prokaryote without a membranous nuclear envelope (Fig. 2.1). These organisms have a layered cell wall braced with murein, the wall polymer of bacteria. They have several other distinguishing biochemical features, particularly dominance of the photosynthetic pigments by a "divinyl chlorophyll pigment", rather than the usual form of chlorophyll-*a* dominant in all other plants (Wu & Rebeiz 1988). They share with the Chlorophyta (green algae) and higher plants the inclusion of another molecular variant of chlorophyll, chlorophyll-*b*. Prochlorophytes were originally discovered as algal symbionts in some tropical algae by Lewin (Lewin & Withers 1975). They are also extracellular symbionts in some attached tropical ascidians (tunicates or sea squirts). Discovery of them in pelagic habitats came from work with automated cell-counting machines (see Box 2.3) in use for study of *Synechococcus*. In pelagic habitats,

Box 2.3 Flow cytometry

Biologists interested in blood-cell counts and in tissue-culture techniques have developed a suite of machines for automatically distinguishing, counting, and sorting large numbers of cells. The cells of interest are dispersed in isosmotic saline that is allowed to flow through a narrow tube fitted with a sequence of sensors, including electrical conductance meters, colorimeters, and fluorometers. If dispersed by sufficient fluid, the cells will pass through the sensors one at a time, allowing computer circuitry attached to the sensors to categorize them in respect to size, color, and fluorescent response to illumination in several wavelengths. If desired, the cells can be sorted into separate containers. To do that, the section of fluid in the tube containing a cell of a given category is tracked from flow velocity. The fluid is allowed to drip from an orifice at the end of the tube, which strips a few electrons from the drop surface, giving it a positive charge. Then the trajectory of its fall is steered into an appropriate container by surrounding electrodes that can be given positive (repulsive) or negative (attractive) charges under computer control. These devices have been adapted by oceanographers for identifying and counting very large numbers of cells, particularly picoplankton, according to their fluorescence and light-scattering signatures (Box Fig. 2.3.1). The positions of many individual cells on two-dimensional plots of light scattering versus fluorescence at different wavelengths generates cluster diagrams illustrating the relative importance of prochlorophytes and cyanobacteria in a given sample (Box Fig. 2.3.2).



Box Fig. 2.3.1 Flow cytometer system layout (courtesy of Dr George Dubelaar, Cytobuoy, The Netherlands.)

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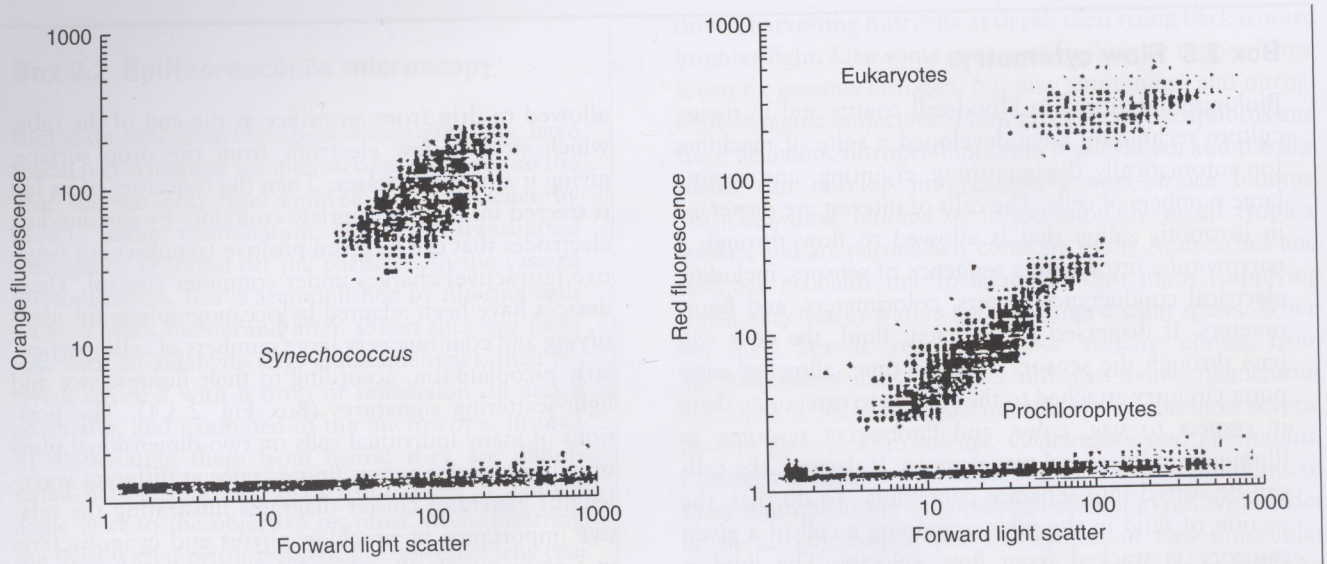

Box Fig. 2.3.2 Scatter plots of fluorescence vs. forward light scatter. (After Olson *et al.* 1990.)

the generic name, *Prochlorococcus*, recognizes their spherical (i.e. coccoid) form.

Prochlorococcus and *Synechococcus* occupy complementary but overlapping niches in the ocean. They form three ecological groups with widespread importance in the ocean: high-light-adapted *Prochlorococcus*, low-light-adapted *Prochlorococcus*, and diversely pigmented strains of *Synechococcus*. Comparisons of gene families in these three groups (Zhaxybayeva *et al.* 2009) suggest frequent gene transfers between *Synechococcus* and low-light-adapted *Prochlorococcus*. Such gene sharing (horizontal gene transfer) also occurs in bacteria, and can result in specific and presumably rapid ecological adaptation.

Eukaryotic picoplankton

In addition to cyanobacteria, the photosynthetic picoplankton often include numerous, eukaryotic forms smaller than 2 μm in cell diameter. The first report that cells of this type are abundant was again recent (Johnson & Sieburth 1982). Some of these forms are coccoid green algae, Chlorophyta, without flagellae but retaining basal bodies or even a rhizoplast structure showing relationship to a *Chlamydomonas*-like ancestor. The genome of one species, *Ostreococcus lucimarinus*, has been sequenced, showing a large number of selenoenzymes (Palenik *et al.* 2007). These enzymes are more catalytically active than enzymes without selenium and may be an adaptation to the organism's small size. Two other divisions, Heterokonta and Haptophyta, have marine species that are ecologically important eukaryotic picoplankton (Worden & Not 2008).

Liu *et al.* (2009) used a combination of genetic, pigment, and microscopy data to elucidate the abundance and diversity of very small haptophytes. These phytoplankton use 19′hexanoyloxyfucoxanthin (19-Hex) as an accessory photosynthetic pigment, and although this pigment is pervasive in the photic zone of the world oceans, haptophyte nuclear rRNA sequences (see Box 2.4) seemed scarce. Classical primers for small subunit ribosomal RNA (SSU rRNA) genes do not work well for the haptophytes, and when more specific primers were developed, the diversity and abundance of the haptophytes became apparent. These tiny haptophytes may contribute from 30 to 50% of the phytoplankton standing stock across the world ocean.

Microflagellates

Small, flagellated cells from a variety of algal groups account for a large fraction of marine primary production. Those groups are separable on the botanical grounds of differences in ultrastructure, pigment composition, details of biochemistry, and most recently by SSU rRNA comparisons. However, ecologists group them together as a convenience, since they are similar in size and general morphology. Their size ranges from 2 to about 30 μm, but most are less than 10 μm. All swim weakly by flagellar action, photosynthesize, and require special care to preserve. These common features mean that they must be studied by similar, specialized means, whatever their botanical affinities. Table 2.1 gives the salient features of the major groups based on features of the flagellae, cell wall, and abundant pigments. Details of cell division are also distinctive (Taylor 1976), but in a very complex fashion. The classification chosen follows Falkowski and Raven (2007) and borrows from group descriptions in Dodge (1979). In addition to the groups listed, a number of benthic algal groups have small,

Box 2.4 Molecular genetic classifications and phylogenetic reconstructions

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The formulas, *genes*, for construction of proteins from 20 amino acids are stored in cells as sequences of four, small distinctive compounds, nucleotides (or "bases"): thymine (T), adenine (A), guanine (G), and cytosine (C). They are arrayed along and bonded to a chain of polymerized sugar molecules (deoxyribose). The chains are *deoxyribonucleic acid* (DNA). The code for each amino acid consists of three bases; for example, TGG codes for tryptophan. There are 64 codes (4^3) possible with four letters, so most amino acids are indicated in the code by several synonyms.The codes specifying particular amino acids vary in a few organisms. The construction of a specific protein involves enzymatic *transcription* of the DNA into similar molecules (with uracil, U, substituted for T) but on a ribose polymer: ribonucleic acid (RNA). Some of the codes indicate to transcription enzymes to "start transcribing here" or "stop transcribing". The transcription, termed messenger RNA or mRNA, is then *translated* into the protein by organelles (complex molecular machines) called ribosomes. In the ribosome, triplets of bases on the mRNA mesh with diffusing bits of RNA (transfer RNA, tRNA) linked to the specific amino acid appropriate to the triplet code, and the ribosome catalyzes the polymerization of the resulting amino acid sequence. The protein is then released, folds into its functional form, and additional complex processes incorporate it in the operating structure of the cell.

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In the long process of revealing DNA structure, storage, replication and translation as proteins, molecular biologists learned how to "read" the code for long DNA sequences. Radically oversimplifying (see Sambrook *et al.* 2006), DNA from an organism is chemically extracted, and then selected portions are copied to generate readable quantities. There are two main ways: (i) The DNA is broken into bits that are installed in bacterial plasmids (closed loops of DNA), and those are multiplied through massive reproduction of carrier bacteria, usually *Escherichia coli*. DNA of specific interest is removed from the cloned plasmids and cleaned. Or, (ii) a sequence of interest can be selectively amplified by the polymerase chain reaction (PCR). As detailed in molecular biology books, PCR is an artificial amplification procedure done extracellularly. It allows amplification of very specific sequences, provided that reliably conserved portions of the sequences of interest are known to serve as "primers". The DNA produced in either way is purified and sequenced by Sanger's dideoxynucleotide chain-terminating method (see Sambrook *et al.* 2006), which has been automated using a version of PCR. The DNA code is read from the method's chromatogram. The result is literally spelled out:

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living things. Genes coding the RNA constituting ribosomes have been particularly important. This RNA comes in two subunits, termed large and small. Small subunit (SSU RNA) sequences have been particularly useful in identifying microbial relationships and very old phylogenetic connections generally. The terms 16S RNA and 18S RNA are commonly used, reflecting the molecular weights (in kilodaltons) of prokaryote and eukaryote SSU RNA, respectively. Conserved primers for SSU DNA have been known for ~30 years. Over the course of evolution to more complex life forms, ribosomal structure has become more complex and the code has gotten longer. This growing elaboration and the variations among elaborations on different phylogenetic branches are a basis for classification and reconstruction of evolutionary relationships. The conserved parts apparently are fundamental to ribosomal function, explaining their consistent sequences throughout the evolution of life on Earth. They provide starting places for both comparisons and for PCR amplification. Classification of life forms using SSU RNA was pioneered by C.R. Woese (e.g. Woese & Fox 1977), and then expanded as "exploratory systematics" by Norman Pace and colleagues (e.g. Olsen *et al.* 1986). That work has profoundly reorganized thinking about microbial forms in particular.

Because mitochondria and chloroplasts were acquired originally as internal symbionts of ancestral cells, they retain a modest part of the DNA (and the ribosomes) required to construct their proteins. That DNA is clonally reproduced without sexual recombination (leading to simpler patterns of inheritance than those of nuclear or bacterial genes), evolves relatively rapidly, and can provide somewhat more direct tracing of ancestry. Mitochondrial DNA, mtDNA, is often favored for studies of phylogeny in eukaryotes, particularly animals. Gene variants are termed "haplotypes" because of the haploid character of mitochondrial genes. Sequences of particular mtDNA genes are widely used for species and strain identification using either short sequences from genes or species-specific, mtDNA PCR primers. The gene for cytochrome oxidase I (COI) is popular in that regard for animals, sometimes termed a DNA "barcode", and has been widely applied in the recent Census of Marine Life (COML).

Additional specific details of molecular genetics will be supplied as needed throughout the book.

Table 2.1 Salient features of taxonomic groups of microalgae. Several groups seldom abundant in marine habitats are left out: Euglenophyta, Raphidophyceae, Eustigmatophyceae. Principal pigments for the major groups are shown in Table 2.2.

	FLAGELLAE	CELL WALL
Prokaryotes (Cyanobacteria)		
Synechococcus	0	Murein
Prochlorococcus	0	Murein
Chlorophyta	Two (or four) apical, equal, smooth	Naked or with cellulose sheath, sometimes calcified
Prasinophyceae	One or two unequal, or four equal and scaly	Organic scales or naked
Cryptophyta	Two, apical, equal, sometimes hairy	Naked
Haptophyta	Two, equal or not, hairy + haptonema emerging between	Chlorophylls *c*1, *c*2 Organic scales
Prymnesiales	Two, unequal, hairy, haptonema vestigial	Organic or calcite scales
Heterokontophyta Bacillariophyceae (Diatoms)	One in male gamete, hairy, no microtubules	Naked gametes
Chrysophyceae and relatives*	Two, unequal, posterior short, smooth, anterior long	Naked or scales (some opal)
Dinophyta	Two, one girdling, one posterior	Cellulosic plates or often naked
Rhodophyta	0	Cellulose

*A number of groups related to the Chrysophyceae have recently been separated within a new phylum Heterokontophyta. These include Synurophyceae, Dictyochophyceae (including the Dictyocales or silicoflagellates that bear opal scales) and the very tiny Pelagophyceae (the principal eukaryotic picoplankton). Heterokontophyta are characterized by flagellae of unequal length (anterior "tinsel" and posterior smooth), chlorophyll-*c*, and chrysolaminarin as a storage product. The Chrysophyceae and Bacillariophyceae (listed above) are Heterokontophyta, as are the brown algae Pheophyceae and Xanthophyceae.

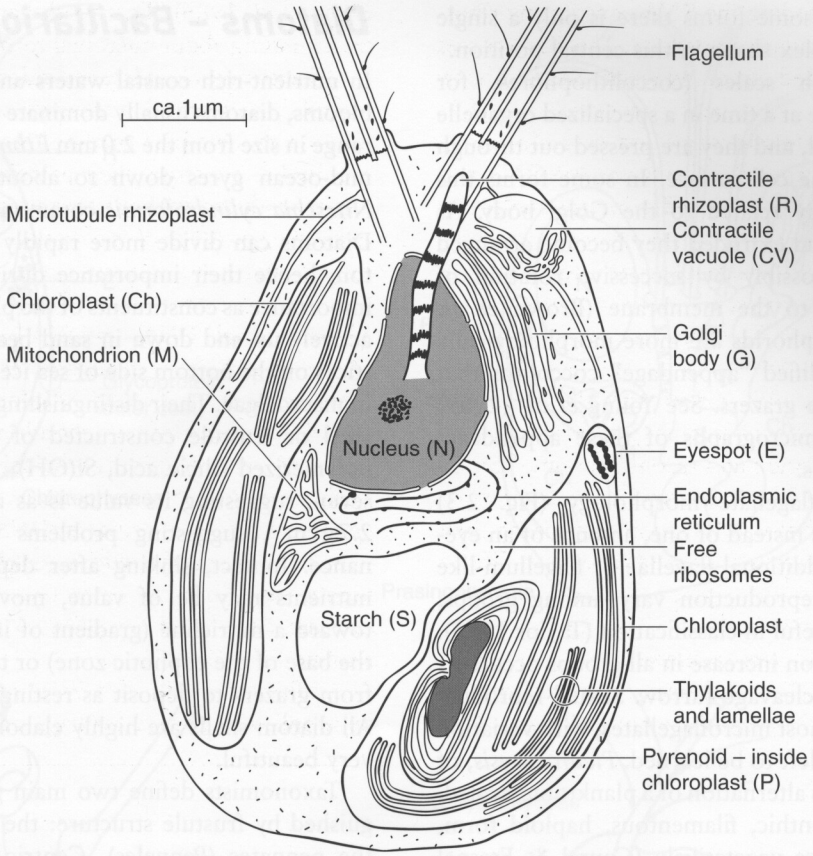


Fig. 2.2 General morphological plan of a number of microflagellate types.

flagellated, photosynthetic gametes that may be abundant in coastal phytoplankton from time to time. Those include the benthic diatoms, brown algae (Phaeophyceae), various Chlorophyta, Xanthophyceae and the Eustigmatophyta. Descriptions of the last two groups can be found in Van den Hoek *et al.* (1995).

Despite the diversity in details, a general pattern of cellular organization can be described that applies to most microflagellates. The cell is round to ovoid, occasionally spindle-shaped. It has an "anterior" end at which are located the insertions of the flagellae, most often two in number. Movement of the cell usually is toward the flagellae, which pull. The outer surface may be the naked cell membrane, or the cell may be covered with a secondary organic wall and bear organic, siliceous, or calcareous scales. Internally, the structure is as shown in Fig. 2.2.

The nucleus is pear-shaped, with its narrow end located anteriorly beneath the roots of the flagellae. The root of one flagellum (sometimes both flagellae) attaches to a spray of striated fibers, the rhizoplast, which extends posteriorly into the cytoplasm around the nucleus. Rhizoplast fibers can contract, pulling intermittently on the root to generate the wave-like movements of the flagella. To one side of the nucleus and root structure of the flagellae sits a contractile vacuole. Located anteriorly is the Golgi body, a system of flattened membranous cisternae involved in secretory activity. In many forms it produces the scales that cover the cell externally.

There may be one or two chloroplasts. If there is just one, then it is cup-shaped and fills the posterior end of the cell. If two, they wrap the sides of the cell, one often extending farther posteriorly. Thylakoids, the layers of membrane inside the chloroplast bearing the photosynthetic pigment systems, are arranged parallel to the external surface of the cell. There may be two to many thylakoids, and thylakoids vary among groups in the number of membranes constituting them. In the center of the chloroplast cup is a *pyrenoid*, which organizes the formation of storage product, variously lipid or starch, in a mass below the nucleus. The anterior-most extension of the chloroplast often bears an eye-spot consisting of several layers of pigmented granules. This probably enables phototaxis or other responses to light and, thus, slight vertical migration. Mitochondria either scatter about between the chloroplast

and the nucleus, or in some forms there is only a single mitochondrion of complex shape in this central position.

Microflagellates with scales (coccolithophores, for example) form them one at a time in a specialized organelle in the interior of the cell, and they are pressed out through the membrane on to the cell surface. In some forms this scale organelle is clearly related to the Golgi body. As the scales are formed and extruded they become arranged on the cell surface, possibly by successive rotation of the cytoplasm relative to the membrane (Brown *et al.* 1973). Some coccolithophorids are more morphologically complex and have modified "appendage" coccoliths that may serve to discourage grazers. See Young *et al.* (2009) for scanning electron micrographs of these appendage-bearing coccolithophores.

Variations in microflagellate morphology (Fig. 2.3) include two chloroplasts instead of one, absence of an eye-spot, and presence of additional flagellae or flagellum-like organelles. Details of reproduction vary among groups, adding characteristics useful in classification (Taylor 1976). However, most population increase in all groups occurs by simple division along a cleavage furrow. Sexual reproduction has been found in most microflagellates, with variation in the details and much left to be learned. *Pleurochrysis*, of the Haptophyta, exhibits alternation of a planktonic, scaled, diploid form with a benthic, filamentous, haploid form, both of which reproduce vegetatively (Gayral & Fresnel 1983). The benthic form releases naked swarmers that fuse to form zygotes that develop into the scaled form. Haploid–diploid alternation of two distinct forms also occurs in most coccolithophores (de Vargas *et al.* 2007). In chlorophycean forms like *Chlamydomonas*, the vegetative cells are haploid, with sex as fusion of vegetative cells modified to + and – forms with complementary cell structures to enable fusion and nuclear transfer (Goodenough & Weiss 1978). Meiosis takes place in the "zygote" (Triemer & Brown 1977).

The Prymnesiophyta are particularly prominent in the oceanic phytoplankton. *Emiliania huxleyi* and other coccolithophores bear calcareous plates. They sometimes bloom in sufficient density to reflect light strongly back into the sky, and appear in satellite images as creamy outlines of eddies (Fig. 2.4).

Their cousins, the Pavlovales, retain the full function of a structure between the two flagellae called a haptonema, constructed of three concentric sheaths of membrane surrounding a core of seven microtubules. The outer sheath may bear small scales. The haptonema can bend and coil through activity of the microtubules, allowing it to serve as a feeding organelle (Kawachi *et al.* 1991). Particles adhere to it, are moved to the base, stuck together into a mass, and then moved back out to the tip. The tip then twists around to the base of the cell, and the surface of the cell forms a food vacuole around it (phagocytosis). Mixing of autotrophy and phagotrophy is quite widespread, even in extremely small cells.

Diatoms – Bacillariophyceae

In nutrient-rich coastal waters and during oceanic spring blooms, diatoms usually dominate the phytoplankton. They range in size from the 2.0 mm *Ethmodiscus rex* of the warm, mid-ocean gyres down to about 2 μm, as for example *Nitzschia cylindroformis* common in the subarctic Pacific. Diatoms can divide more rapidly than other phytoplankton, hence their importance during blooms. Beside their importance as constituents of the plankton, diatoms migrate actively up and down in sand beaches, grow in the interstices on the bottom side of sea ice, and live on the surfaces of macroalgae. Their distinguishing feature is a hard mineral shell or frustule constructed of opal, that is, hydrated, polymerized silicic acid, $Si(OH)_4$. Opal has a hardness of seven, suggesting its value is as armor, and a density of $2.7\,g\,cm^{-3}$, suggesting problems with buoyancy maintenance. In fact, sinking after depletion of water-column nutrients may be of value, moving the cell downward toward a nutricline (gradient of increasing of nutrients at the base of the euphotic zone) or taking them rapidly away from grazers to deposit as resting spores in the sediment. All diatom shells are highly elaborate microscopically, and very beautiful.

Taxonomists define two main groups, primarily distinguished by frustule structure: the centrics (Centrales) and the pennates (Pennales). Centric diatoms derive from a radially symmetrical primitive form with frustules shaped like petri dishes (Fig. 2.5). The upper and lower valves (*epitheca* and *hypotheca*) each consist of a flat plate (the valve) and a cylindrical rim, the girdle band, that wraps the curved edge. The valve and the girdle are sometimes loosely connected, sometimes fused. The girdle of the lower valve fits inside the girdle of the upper valve. As the cell grows, this sliding joint can expand, providing room for increase in cell content. In some genera (e.g. *Rhizosolenia*) the joint is not actually sliding, but is a set of pieces or girdle rings, more of which can be added as the cell grows until it is tubular. Cytoplasm is located along the inner surface of the shell, forming a hollow lining around a large vacuole in the cell center. The cytoplasmic layer contains several of the cell organelles, most obviously the chloroplasts and mitochondria. "Cell sap", a fluid not unlike seawater but with variations in specific ion content, fills the central vacuole. The nucleus of the cell is usually located against the center of one valve, but prior to division it slides into a central cytoplasmic island suspended in the vacuole by strands from the sides. Many variant shapes appear among the centrics through extension of the shell in one axis or another and by adding spines.

Pennate diatoms are bilaterally symmetric, not radially symmetric. On each valve there are openings in the form of slits along the surface, termed raphes (Fig. 2.6). Typically, the shell is elongate parallel to the raphes. Cytoplasm streams along the raphes, generating cell movement. The

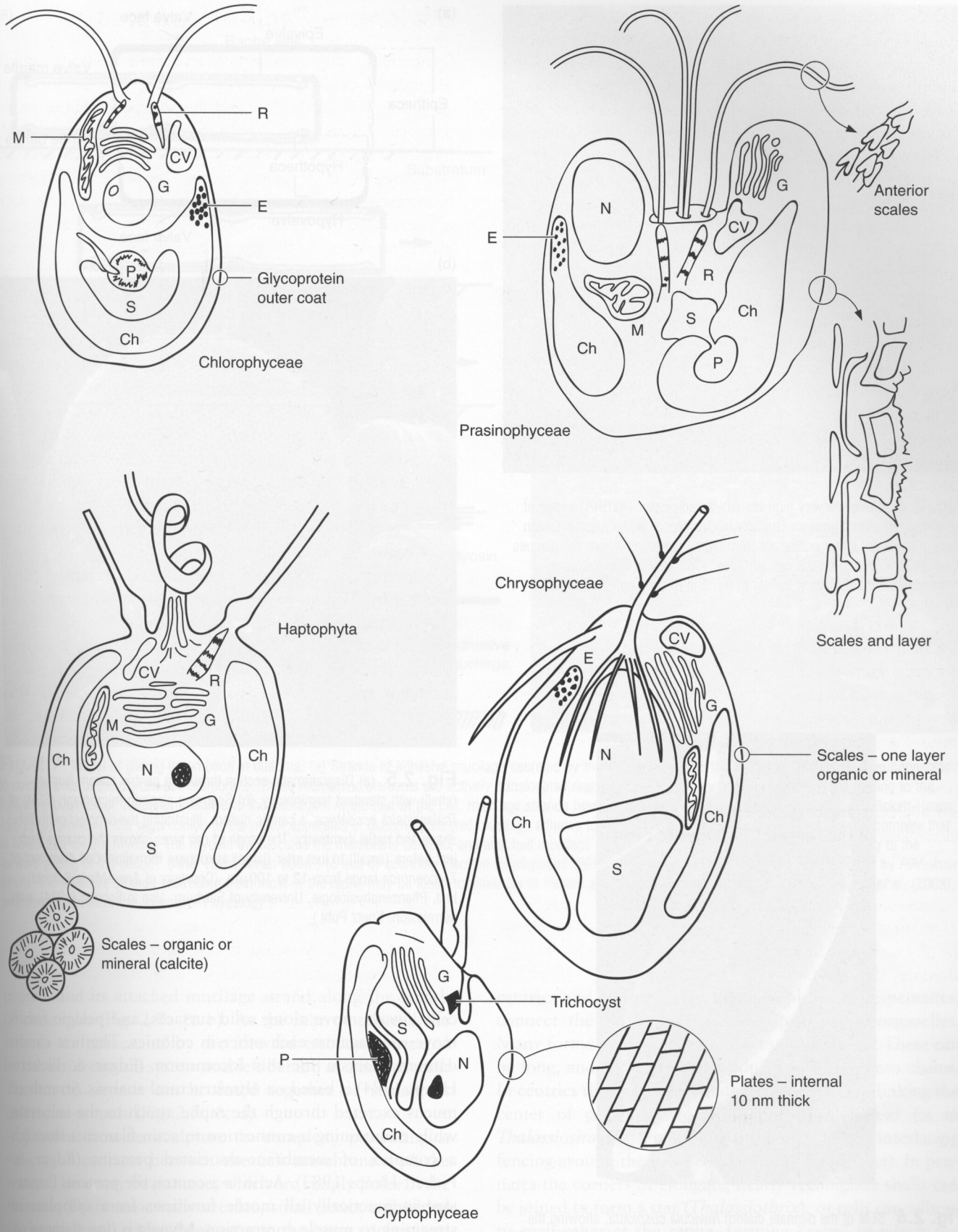


Fig. 2.3 Morphological variations among the more commonly occurring microflagellate groups. Refer to Fig. 2.2 for abbreviations.

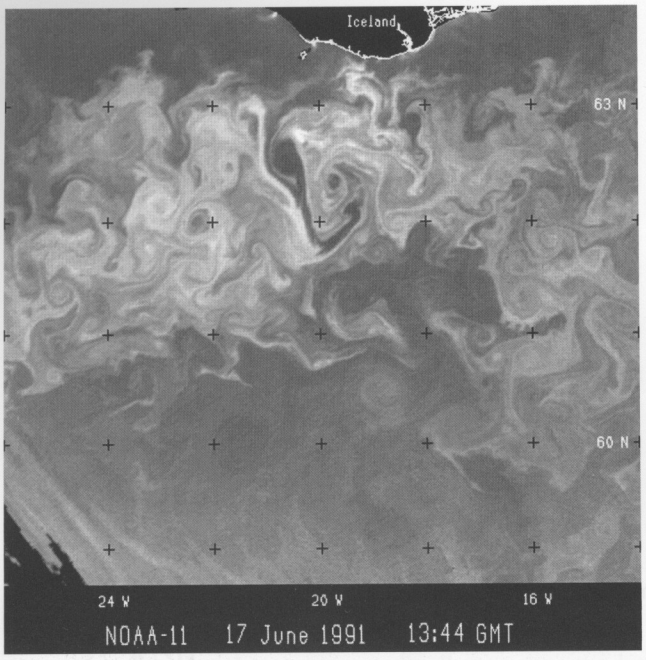


Fig. 2.4 Advanced very high-resolution radiometer (AVHRR) image of the visible reflections from an *Emiliana huxleyi* bloom in the Atlantic Ocean south of Iceland. Lighter colors are from higher reflectance from the plaques of calcite (coccoliths) on the cell surfaces. (Courtesy of Steve Groom, Plymouth Marine Laboratory, similar to fig. 2 in Robertson *et al*. 1994.)

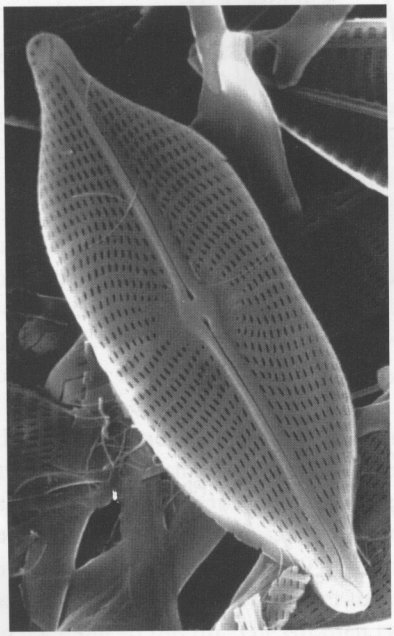


Fig. 2.6 SEM of the pennate diatom *Navicula cuspidata*, showing the central raphe. (Courtesy of E.G. Vrieling, Department of Marine Biology, Centre for Ecological and Evolutionary Studies, University of Groningen, The Netherlands, with permission from the *Journal of Phycology*.)

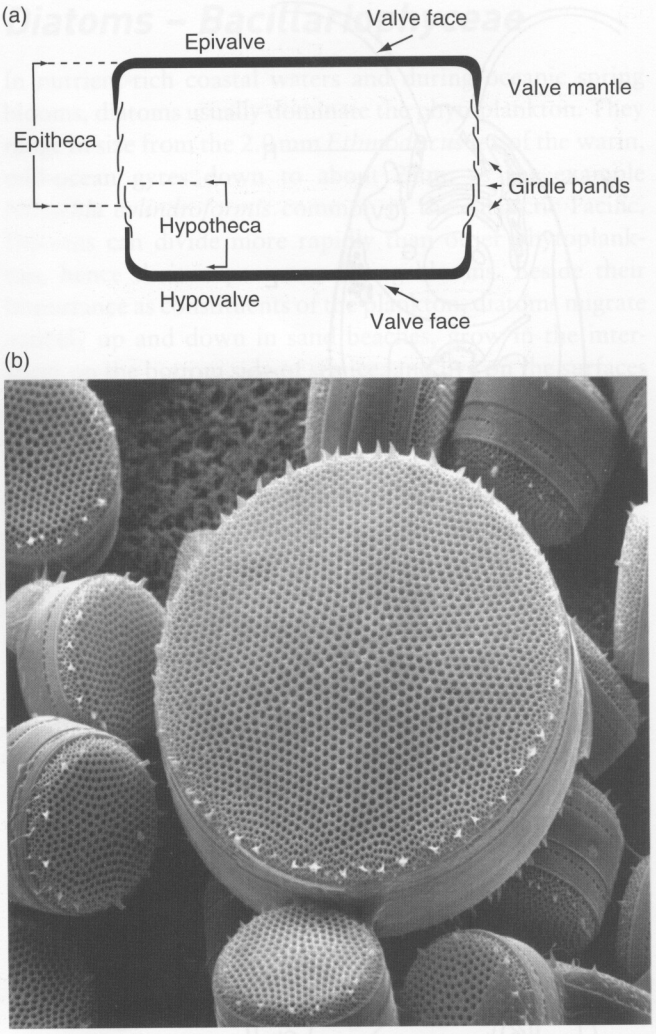


Fig. 2.5 (a) Diagrammatic section through a centric diatom frustule (shell) with standard terminology. (b) Scanning electron micrograph (SEM) of *Thalassiosira eccentrica*, a centric diatom, illustrating the simplest petri-dish shape and radial symmetry. The range of cell sizes shows the change from just before (small) to just after (large) auxospore formation. Cell diameters of *T. eccentrica* range from 12 to 100 μm. (Courtesy of Anne-Marie Schmid, Inst. Pflanzenphysiologie, University of Salzburg; also in Schmid (1984), with permission, Koetz Publ.)

cells always move along solid surfaces, and pelagic forms move only against each other in colonies. The best candidate mechanism for this locomotion (Edgar & Pickett-Heaps 1984) is based on ultrastructural analysis. Strands of mucus excreted through the raphe attach to the substrate while maintaining a connection to actin filaments through a complex of membrane-associated proteins (Edgar & Pickett-Heaps 1983). Actin is a contractile protein, important in practically all motile functions from cytoplasmic streaming to muscle contraction. Myosin is one element of the protein complex, and its movement against the actin filaments produces a rearward translocation of a membrane

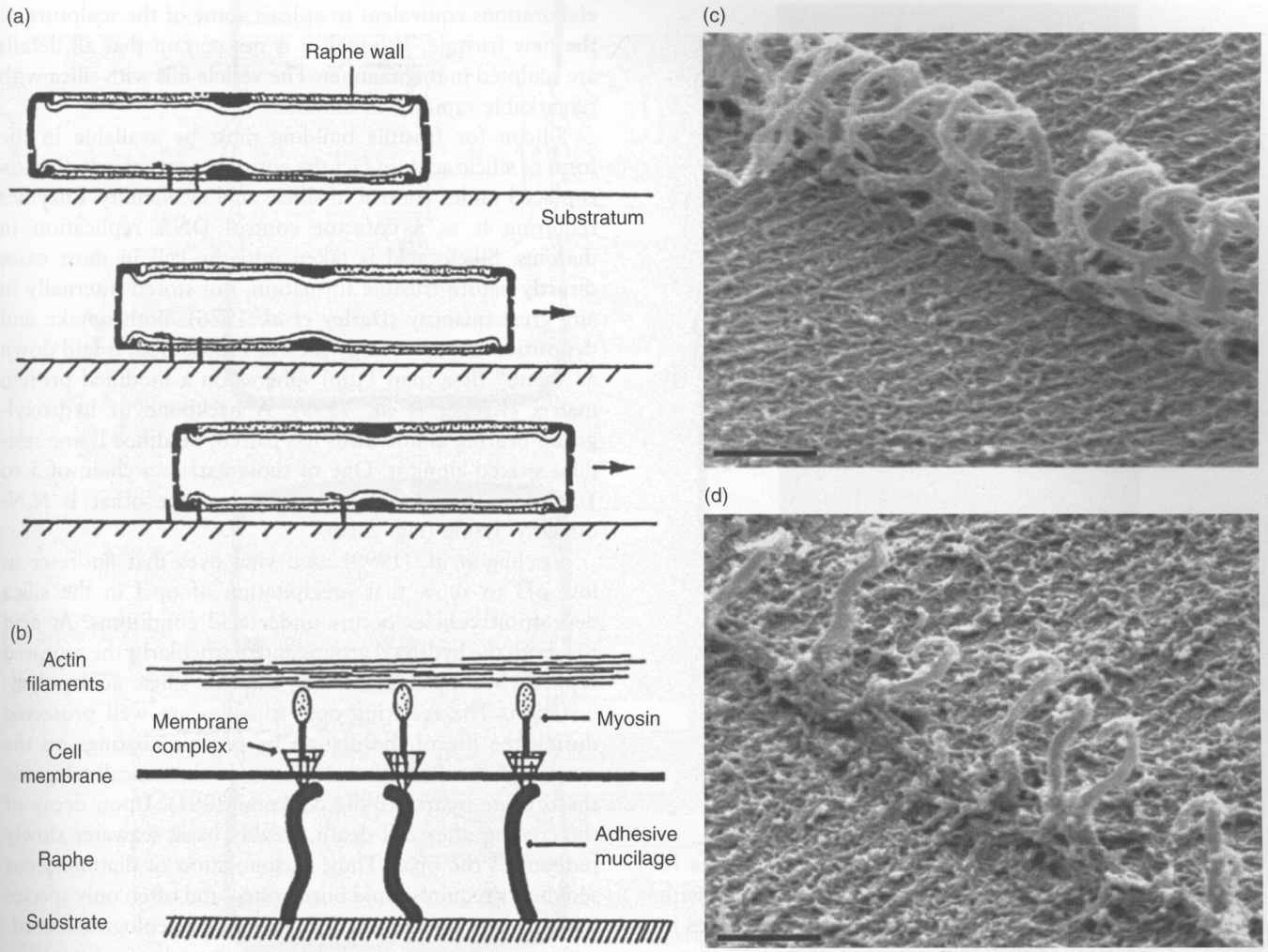


Fig. 2.7 Model of gliding locomotion in diatoms. (a) Strands of adhesive mucilage secreted by the diatom adhere to the substrate but also remain connected to components of the diatom cell membrane. These membrane elements are actively translocated rearward upon a framework of actin filaments, leading to the forward movement of the diatom relative to the substrate. At the rear of the cell, mucilage strands break and are deposited as a trail. (After Edgar & Pickett-Heaps 1983.) (b) Model for the organization of the motor apparatus in diatoms. Excreted musilage adheres to the substrate and to a membrane-associated complex that is linked to a diatom myosin. The myosin translocates the membrane complex and attached mucilage rearward along a tract of actin filaments leading to the forward gliding of the diatom. (After Heintzelman 2006.) (c and d) scanning electron micrographs (SEMs) of chemically fixed mucoid trails deposited by *Pinnularia viridis*. The adhesive strands protruding from most of the raphe may be fused together (c) or remain as individual strands (d). (c and d after Higgins *et al*. (2003), with permission from the *Journal of Phycology*.)

protein and its attached mucilage strand along the length of the raphe (Fig. 2.7a & b). At the rear of the cell the strands are broken off by the forward traction generated by the actin–myosin system and left behind as a mucus trail. The mucus strands can be fused together or remain as individual strands (Fig. 2.7c & d).

Diatom frustule sculpture is complex, producing a variety of contacts between the cell membrane and the water. There may be punctae, which are simply holes in the frustule. There are usually areolae, small boxes embedded in the shell wall with a large pore in the outer surface and a lacework with many small pores against the cell membrane. Some openings, such as the labiate process of both centrics and pennates and the pore plate of the pennates, connect the water outside with specific cell organelles. Many forms have spines and processes (Fig. 2.8). These can be long, and they sometimes connect the cells into chains. In centrics this may be a slender siliceous thread linking the center of one valve to the center of the next (as in *Thalassiosira*), or a complete ring of elaborate, interlacing fencing around the valve edges (as in *Skeletonema*). In pennates the corners of elongate, nearly rectangular shells can be joined to form a star (*Thalassiothrix*), or cells can adhere to each other at the raphe, forming "rafts" in which the cells slide back and forth along each other's length (*Bacillaria*). It is often assumed that the very long side

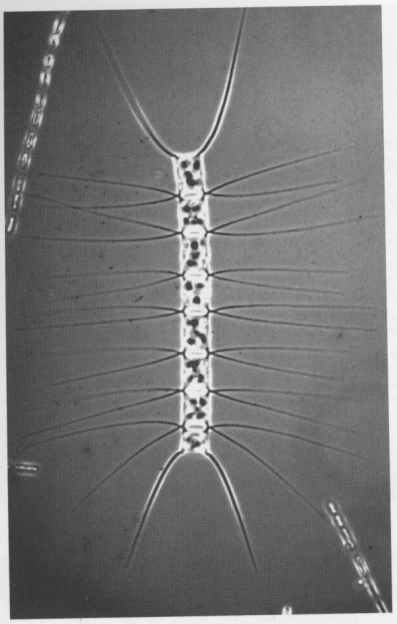


Fig. 2.8 Phase-contrast light micrograph of *Chaetoceros decipiens*, showing chaining of cells and fine siliceous spines (spines this fine are usually called setae). (Courtesy of J.D. Pickett-Heaps, University of Melbourne.)

spines either inhibit predation or enhance surface area to increase drag and inhibit sinking. However, for forms with siliceous spines, calculations show that the cost of spines in increased density exceeds the gain in increased drag. Moreover, Gifford *et al.* (1981) have shown that copepods more readily eat a form of *Thalassiosira weissflogii* with long, chitinous spines than they do a form without them, presumably because longer spines make the cell more readily detected and captured more effectively. The whole matter of spines is still not fully explained.

Diatom cells divide by an unusually elaborate process, since the cell not only must divide, but must fabricate new valves. Pickett-Heaps *et al.* (1990) have reviewed much of what is known about diatom mitosis. Division (Fig. 2.9) proceeds inside the old frustule until there are two protoplasts, one adjacent to each valve. Each has its own cell membrane at the central plane. New siliceous walls form inside and parallel to each of these membranes. Formation begins with the appearance of membranous vesicles, probably derived from one or more sets of Golgi apparatus (cell organelles involved in packaging of secretory products, particularly protein–carbohydrate complexes) in each cell. These vesicles aggregate in the center of the division plane, and coalesce to form a "silicalemma", a membrane surrounding a silica deposition vesicle (SDV). The two sides of the silicalemma may connect in a "donut hole" wherever there is to be a pore in the new frustule. There are other elaborations equivalent to at least some of the sculpture of the new frustule, although it is not certain that all details are sculpted in that manner. The vesicle fills with silica with remarkable rapidity, in minutes.

Silicon for frustule building must be available in the form of silicic acid; in fact the entire process of cell division is placed under control of silicic acid availability. Enzymes requiring it as a cofactor control DNA replication in diatoms. Silicic acid is taken into the cell in most cases directly before frustule formation, not stored internally in any great quantity (Darley *et al.* 1976). Both uptake and deposition require energy (Lewin 1955). Opal is laid down as "nano" (less than 1 μm) spheres on a modified protein matrix (Kröger *et al.* 1999). A backbone of hydroxyl-group-bearing amino acids has paired, modified lysine residues spaced along it. One of those carries a chain of 5 to 10 repeats of N-methyl-propylamine, the other is *N*,*N*-dimethyl lysine (Fig. 2.10).

Vrieling *et al.* (1999) used vital dyes that fluoresce at low pH to show that precipitation of opal in the silica deposition vesicles occurs under acid conditions. At acid pH, both the hydroxyl groups and, particularly, the repeated *N*-methyl-propylamine chains catalyze silicic acid polymerization. The resulting opal frustules are well protected during the life of the diatom by organic coatings on the outer surfaces. Bacteria colonize and enzymatically degrade the organic matrix (Bidle & Azam 2001). Upon decay of the coating after cell death, weakly basic seawater slowly redissolves the opal. Thus, accumulation of diatomaceous sediments requires rapid burial rates, and often only species with the very thickest shells remain in the geological record.

In many, but not all, diatom species, cell size diminishes progressively as division proceeds (MacDonald 1869). Since the hypotheca becomes the epitheca for one of the daughter cells, that cell is smaller than its sister. This process terminates by formation of an *auxospore*. This is usually, if not always, coupled with sexual reproduction. Drebes (1977) provides a thorough discussion of sexual processes in diatoms. In centric forms (an example of a life-history sequence is shown in Fig. 2.11) many species produce four flagellated sperm by meiosis. A sperm fertilizes a single oocyte produced inside the shell of the reduced vegetative cell. The resulting zygote drops the frustule, grows to a large size, and forms a heavy cell membrane of organic material containing siliceous scales (Edlund & Stoermer 1991), and finally develops a shell of the vegetative type internally. This new frustule, surrounding the so-called "initial cell", is produced in association with several mitoses, indicating a relationship between the control of frustule formation and the control of mitosis *per se*. All of the daughter nuclei, save one, simply degenerate. Pennates exhibit similar sexual processes, although motile sperm are not involved. Rather the gametes are "isogametes", cells of the same size (sometimes amoeboid) produced by meiosis that fuse to form a zygote when exchanged between touching parent frustules.

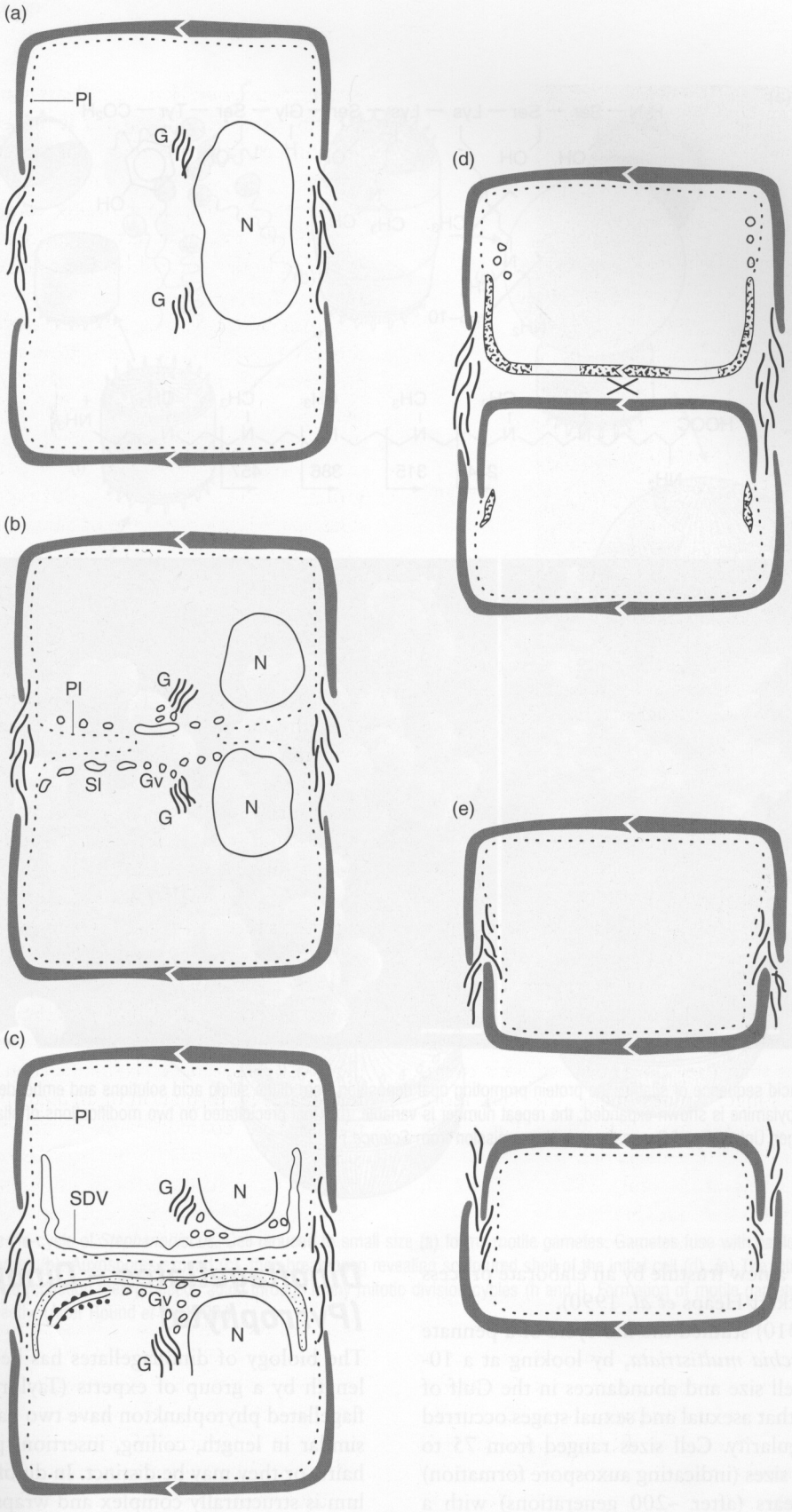


Fig. 2.9 Sequence of cell division and frustule formation events in the pennate diatom *Gomphonema parvulum*. (a) Cell elongates, additional volume covered by addition of more girdle bands, the nucleus enlarges with DNA duplication, and organelles, represented by Golgi body (**G**), are partitioned. (b) Nucleus divides, then protoplasts are separated by invagination of the plasmalemma (**Pl**) across the division plane, and vesicles (**Gv**) apparently derived from the Golgi body accumulate adjacent to it. (c) Formation is completed of silicalemma (**Sl**) around deposition vesicles (**SDV**) in which the valves will develop. (d) Opal is deposited in the SDV, then smaller SDV form at the sides for girdle band deposition (shown in lower cell). (e) Cells separate. (After Dawson 1973.)

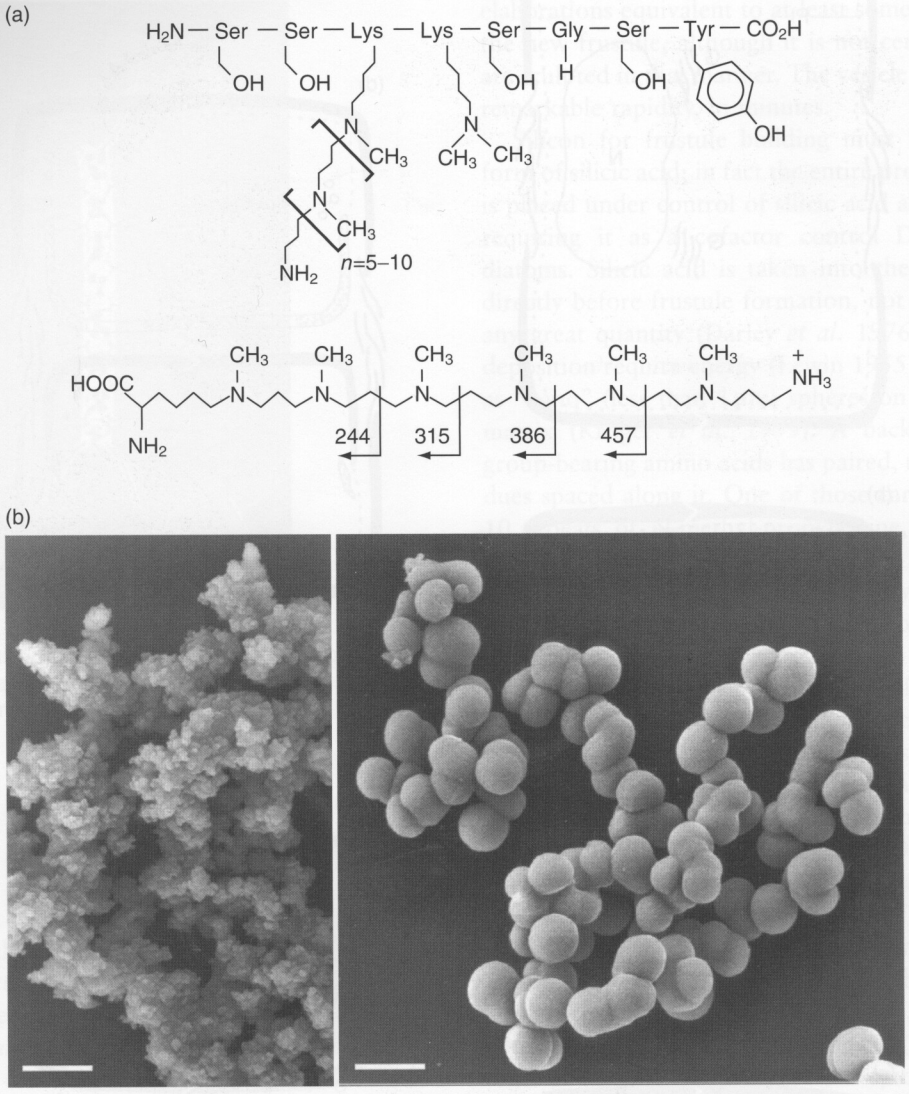


Fig. 2.10 (a) Amino acid sequence of silaffin, the protein promoting opal deposition from dilute silicic acid solutions and embedded in diatom opal; the side chain of poly-*N*-methyl-propylamine is shown expanded; the repeat number is variable. (b) Opal precipitated on two modifications of silaffin out of dilute silicic acid. (Courtesy of Nils Kröger, University of Regensburg, with permission from *Science*.)

The zygote produces a new frustule by an elaborate process (see Mann 1984; Pickett-Heaps *et al.* 1990).

D'Alelio *et al.* (2010) studied the life-cycle of a pennate diatom, *Pseudo-nitzschia multistriata*, by looking at a 10-year time series of cell size and abundances in the Gulf of Naples. They found that asexual and sexual stages occurred with remarkable regularity. Cell sizes ranged from 75 to 30 µm, and large cell sizes (indicating auxospore formation) occurred every 2 years (after ~200 generations) with a subsequent decrease in size over the following period. This is the first report of such regulation of cell division and sexual reproduction across a regional population in a diatom. The mechanism of this regulation is unknown.

Dinoflagellates – Dinophyceae (Pyrrophyta)

The biology of dinoflagellates has been reviewed at book length by a group of experts (Taylor 1987). Most of the flagellated phytoplankton have two flagellae. These may be similar in length, coiling, insertion, presence of scales or hairs, or they may be distinct. In dinoflagellates, one flagellum is structurally complex and wraps around the equator of the cell in a groove, the *cingulum*. Its wave-like movements serve to rotate the cell in the water as it swims, a characteristic from which the name of the group derives: διηοσ, "whirling". The other, simpler flagellum originates

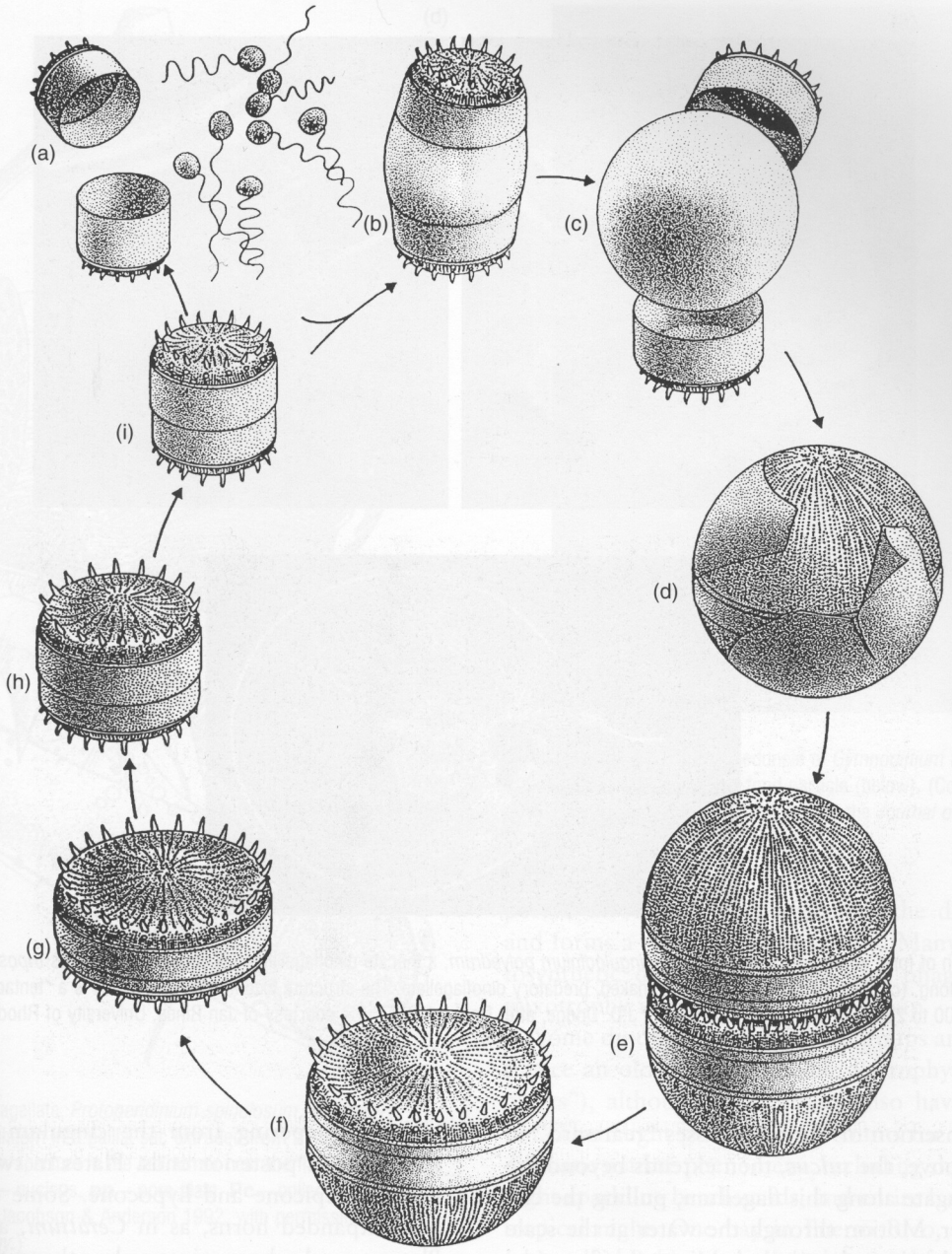


Fig. 2.11 Events in the life-cycle of *Stephanodiscus*. Cell reduced to small size (a) forms motile gametes. Gametes fuse with haploid oogonial protoplasts, which form enlarged auxospores (b). Auxospores expand (c), then break open revealing sculptured shell of the initial cell (d). (e) The initial cell divides, producing vegetative cells (f then g). Size reduction then proceeds again through many mitotic division cycles (h and i). Formation of motile gametes is only supposed for this genus, not observed directly. (After Round *et al.* 1990.)

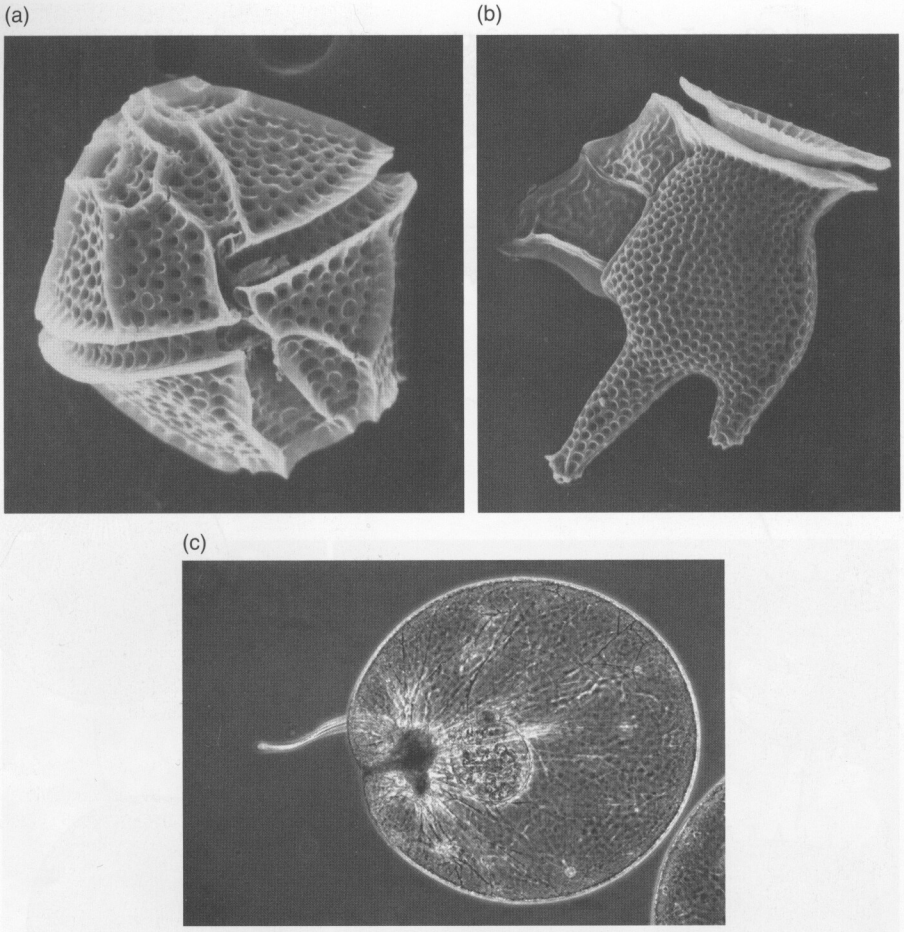


Fig. 2.12 Variation of form among dinoflagellates. (a) *Lingulodinium polyedrum*, a thecate dinoflagellate, ~30 μm. (b) *Dinophysis tripos*, a tropical flagellate with a "sail" ~100 μm long. (c) *Notiluca scintillans*, a large, naked, predatory dinoflagellate. The structure extending from the cell is a "tentacle", not a flagellum; diameter varies from 200 to 2000 μm (a and b – courtesy of J.D. Dodge, after Dodge (1985); c – courtesy of Jan Rines, University of Rhode Island.)

just behind the insertion of the first, passes "rearward" in a longitudinal groove, the *sulcus*, then extends beyond the cell. Waves propagate along this flagellum, pulling the cell through the water. Motion through the water at the scale of a flagellum is dominated entirely by viscous effects (see Chapter 1 for an explanation).

Dinoflagellates divide into three broad groups: the unarmored Gymnodiniales and the armored Peridiniales and Dinophysiales. Dodge and Crawford (1970), however, have shown that the species really form a gradual series. All forms have a pellicle constituted of a cell membrane externally and underlain by flattened vesicles. The vesicles are filled in armored forms with cross-linked cellulose, forming plates. The pellicle in both groups divides into an *epicone* ahead of the cingulum and a *hypocone* behind. Plates of armored forms are arranged in a heritable pattern over the epi- and hypocones, and the patterns distinguish subgroups and species. These morphological species designations do not always match up with SSU rRNA designations. A range of forms is illustrated in Fig. 2.12. Peridiniales are roughly biconical, tapering from the cingulum toward rounded anterior and posterior ends. Plates in two rows surround both the epicone and hypocone. Some of the plates can bear expanded horns, as in *Ceratium*, a common genus. Plates may also bear spines or be otherwise elaborated. The Dinophysiales have much smaller individual plates, fused into anterior and posterior valves, with the cingulum and sulcus bordered by thin expansions or crests arising from the edges of the grooves. The pattern varies between species by differences in expansion and sculpture of crests. Many of the resulting forms are reminiscent of the intergalactic cruisers of science-fiction movies. Dinophysiales are mostly tropical and exclusively marine, rarely if ever a major constituent of the plankton.

Some members of both naked and armored groups can prey upon smaller organisms such as diatoms, ciliated protozoa even copepod nauplii. Some forms are not photosynthetic; they lack chloroplasts, and depend exclusively upon prey for nutrition. In *Protoperidinium* and the *Diplopsalis* group, a membranous sac, called a pallium, is produced by

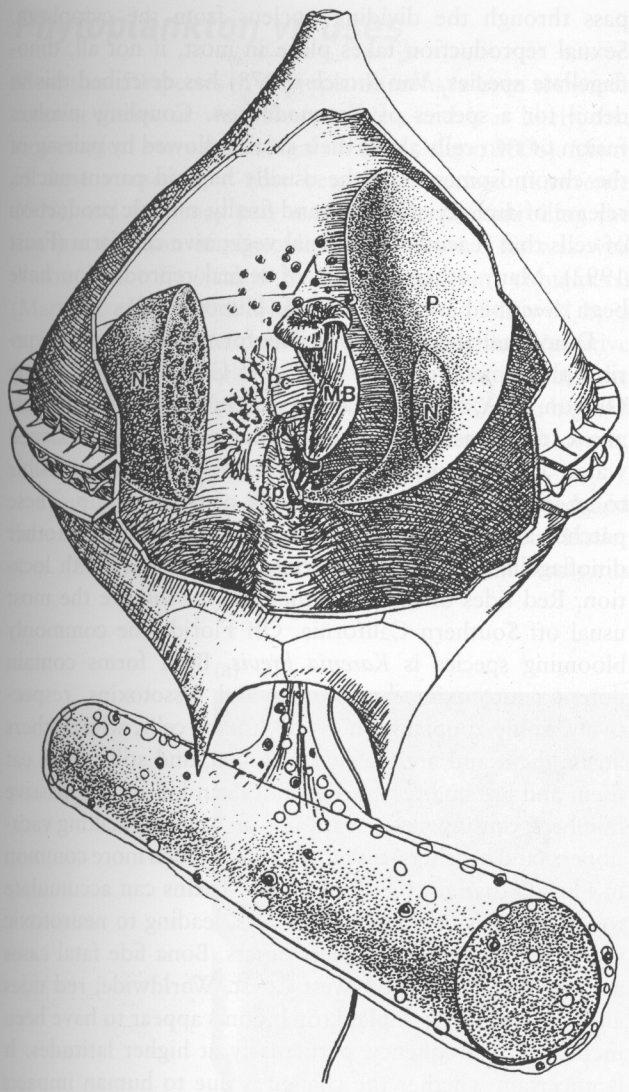


Fig. 2.13 A dinoflagellate, *Protoperidinium spinulosum*, externally digesting a cylindrical diatom in a pallial sac extruded from a microtubular basket (**MB**) through an aperture in the cell armor near the flagellar pore. Other abbreviations: **N** – nucleus, **pp** – pore plate, **Pc** – collecting pusule, **P** – sac pusule. (After Jacobson & Anderson 1992, with permission from the *Journal of Phycology*.)

an elaborate apparatus (Fig. 2.13) internal to a "mouth", or pallial pore, located in the sulcus. In *Protoperidinium spinulosum*, prey become attached first to a slender pseudopod strengthened with protein microtubules called a tow filament. The pallial membranes are then extruded around it and digestion proceeds externally in a pallial sac (Jacobson & Anderson 1992).

The naked, non-photosynthetic *Gymnodinium fungiforme* attacks prey by insertion into their tissue of a tubular peduncle (Fig. 2.14; Spero 1982). From a cell about 15 µm in diameter, the peduncle can extend at 3 µm diameter for 12 µm, supported by microtubules derived from an internal microtubular "basket" similar to that seen in forms secreting pallial sacs. Cytoplasm from the prey is actively transported through the peduncle into the dinoflagellate cell, and forms a mass of food vacuoles. Many (if not all) photosynthetic dinoflagellates are also phagotrophs, so that mixotrophy is a hallmark of the group.

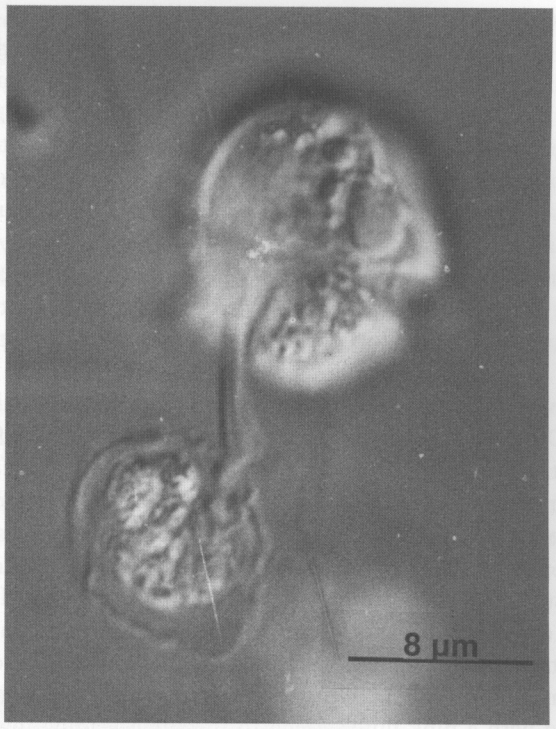


Fig. 2.14 The feeding peduncle of *Gymnodinium fungiforme* (above) extending into an unidentified food particle (below). (Courtesy of H. Spero, after Spero (1982), with permission from the *Journal of Phycology*.)

Some dinoflagellates from all groups are bioluminescent (hence an older division name, Pyrrophyta, meaning "fire plants"), although most groups also have forms that are not. Their light production accounts for a wide variety of dramatic marine phenomena, including shining boat wakes, night-sparkling beach sands, and entire bays glowing in the tropical night. One bay in Puerto Rico, Bahia Fosforente (also called Bahia Mosquito), is named for this glow, which is believed to arise from a complex scheme in which organic matter from mangrove trees along shore supports bacteria, which produce extracellular vitamin B_{12}, which then allows the vitamin-requiring and luminescent *Pyrodinium bahamense* to proliferate. Light is produced in some forms by a diffuse, soluble luciferin–luciferase system (a chemoluminescent compound and the enzyme moderating energy transfer to it), and in other forms by a similar but membrane-bound biochemistry. Dinoflagellates with membrane-bound luminescent organelles, called scintillons, are capable of remarkably intense, brief flashes, usually in response to mechanical stimulation of the cell. In many species, bright ambient light suppresses bioluminescence, presumably saving energy when luminescence could not be seen above the background. Experiments (Esaias & Curl 1972) make

clear that luminescent flashing set off by motion of the feeding limbs of planktonic herbivores shuts down their feeding activity, reducing rates of grazing on dinoflagellates. The mechanism certainly exploits the fact that a predator with a mass of glowing particles in its vicinity will attract its own visual predators. Thus, selection soon leaves only those individuals that stop feeding when they stimulate luminescent flashing.

Internal organization is complex in the dinoflagellates. The large nucleus of the cell is centrally located. Distinct chromosomes (and lots of them) are present throughout the cell cycle in the form of thick rings of DNA strands that are wound into secondary helices like twist donuts (Oakley & Dodge 1976). There are almost no proteins in dinoflagellate chromosomes, and those that do exist are different from chromosomal proteins in most eukaryotes (Rizzo & Nooden 1973). Mitochondria and Golgi bodies are scattered through the cytoplasm. Photosynthesizing chloroplasts are located around the periphery of the cell. They are constructed in fairly typical fashion with many layers of stacked membranes. Additional organelles occur in some forms but not others. One or more sacs, called *pusules* and filled with a pink fluid, attach to the canal leading inward from the cell surface at the root of the longitudinal flagellum. Pusules are believed to be involved in osmoregulation or possibly buoyancy adjustment (Dodge 1972). Proteinaceous *pyrenoids* involved in starch formation may be present. Some forms have a reddish stigma or eye-spot, like those in a variety of protozoan forms, mediating changes in rate or direction of movement in response to light. In dinoflagellates, eye-spots certainly permit the vertical migratory behavior (down at dusk, up at dawn) exhibited by many forms (e.g. Eppley *et al.* 1968). *Trichocysts*, organelles also found in ciliated protozoa like *Paramecium* and in some Chlorophyta, are located beneath pores in the theca and can explosively project a small protein thread into the surrounding medium. It is believed the thrust thus generated pushes the cell suddenly away from the disturbance causing the discharge. Sudden displacements of only a few body diameters may be enough to prevent predation and may also be involved in prey capture.

Before asexual reproduction in dinoflagellates, the pellicle splits and the cell emerges, naked. Presumably a new cell membrane forms beneath the vesicular layer. The cell swells into a globular form and divides into two, and sometimes four or eight daughter cells. Flagellar replication takes place prior to division generally, and each daughter receives a complement of two flagellae. Some organelles – pusules are an example – appear to be absent during reproduction, reforming later in the cell cycle. Nuclear division differs in many details from that in most eukaryotic forms. The nuclear membrane is present throughout and divides by formation of a progressively constricted waist. There are no centrioles, but the chromosomes migrate into the daughter portions of the nucleus along the inner surface of the nuclear membrane, following microtubules that form and pass through the dividing nucleus from the cytoplasm. Sexual reproduction takes place in most, if not all, dinoflagellate species. Von Stosch (1973) has described this in detail for a species of *Gymnodinium*. Coupling involves fusion of two cells along their sulci, followed by pairing of the chromosomes from the usually haploid parent nuclei, release of diploid swarmers, and finally, meiotic production of cells that reacquire the usual vegetative cell form (Faust 1992). Many other schemes of sexual reproduction have been described as well (Beam & Himes 1979).

Dinoflagellates are responsible for the seasonally recurring phenomenon of *red tides*. Off California, and rarely Oregon, USA, red tides can be seen in summer from bluffs above the coastal sea as irregular patches of reddened water. The intensity of the color ranges from barely visible to an impression of a massive spill of tomato soup. These patches are formed by intense blooms of one or another dinoflagellate. Species commonly involved vary with location. Red tides of *Lingulodinium polyedrum* are the most usual off Southern California. Off Florida the commonly blooming species is *Karenia brevis*. Both forms contain potent neurotoxins, brevetoxins and yessotoxins, respectively. Some zooplankton avoid "toxic" cells, while others ingest them and are harmed or killed, and still others eat them and are unaffected. Red tides can kill fish in massive numbers, causing messy wash-ups on beaches, making vacationers (and hoteliers) unhappy. That is much more common in Florida than on the West Coast. Toxins can accumulate to lethal levels in clams and oysters, leading to neurotoxic shellfish poisoning in careless diners. Bona fide fatal cases are very few for the US West Coast. Worldwide, red tides and other toxic phytoplankton blooms appear to have been increasing in frequency, particularly at higher latitudes. It is uncertain whether the change is due to human impacts upon coastal environments (e.g. hog farm effluents), but it is extremely likely (Glibert *et al.* 2005). Global warming, also a human impact, may play a part in the increase. Intense scientific and public interest surrounded the discovery of a highly toxic dinoflagellate, *Pfiesteria piscicida*, which produces a potent, fish-killing neurotoxin that can be transferred from the water to the air, affecting people directly (Burkholder & Glasgow 1997). Details of the life-cycle stages of *P. piscicida* have been described by Litaker *et al.* (2002).

In addition to their several roles as phytoplankton, microheterotrophs, illuminators of white caps at night and toxic bloom culprits, dinoflagellates are algal partners in a diverse array of symbioses with animals. Called *zooxanthellae* when symbiotic, they reside intracellularly in their animal hosts that harvest photosynthate from them. Such partnerships with zooxanthellae are found in several groups of pelagic protozoa (foraminifera, radiolaria), coral polyps, sea anemones, the giant clam (*Tridachna* sp.) and sundry nudibranch snails. Volumes of information about these relationships have never quenched the curiosity they evoke, so research goes on.

Phytoplankton viruses

More than 50 species of phytoplankton have been shown to contain viruses or virus-like particles, and it is likely that viruses infect every major algal division (Munn 2006). Many of these viruses have now been characterized (Lawrence 2008), and they may play a major role in controlling the size and duration of phytoplankton blooms. This was well illustrated for *E. huxleyi* in mesocosms (1 m^3 containers) (Martinez *et al.* 2007) where a bloom was followed by a rapid increase in virus particles. Exposure of the vegetative (coccolith-bearing diploid) stage to the virus induces the sexual stage of the life cycle (Frada *et al.* 2008). The haploid sexual stage of the coccolithophore is immune to the virus, thus allowing the population an escape mechanism.

Five viruses infecting diatom species have been studied, and these replicate either in the cytoplasm or in the nucleus. Eissler *et al.* (2009) studied the lytic cycle of an intranuclear virus infecting the diatom *Chaetoceros wighamii*. Following inoculation of cultures with the virus, rod-like arrays appear within the nucleus during the early stage of infection (Fig. 2.15).

These are replaced by virus-like particles over the following 24 hours, with about 20% of cells infected. Cell abundance then declines, and the free viral abundance increases.

Most algal viruses appear to be species-specific. Additional studies are under way to characterize the diversity of algal viruses, host specificity, and the ecological role of viruses. Eventually we may know how much phytoplankton mortality is due to viral lysis versus predation.

Phytoplankton standing stocks

Although many different types of phytoplankton can be present in seawater samples, it is often useful to have a measure of the total phytoplankton biomass or standing

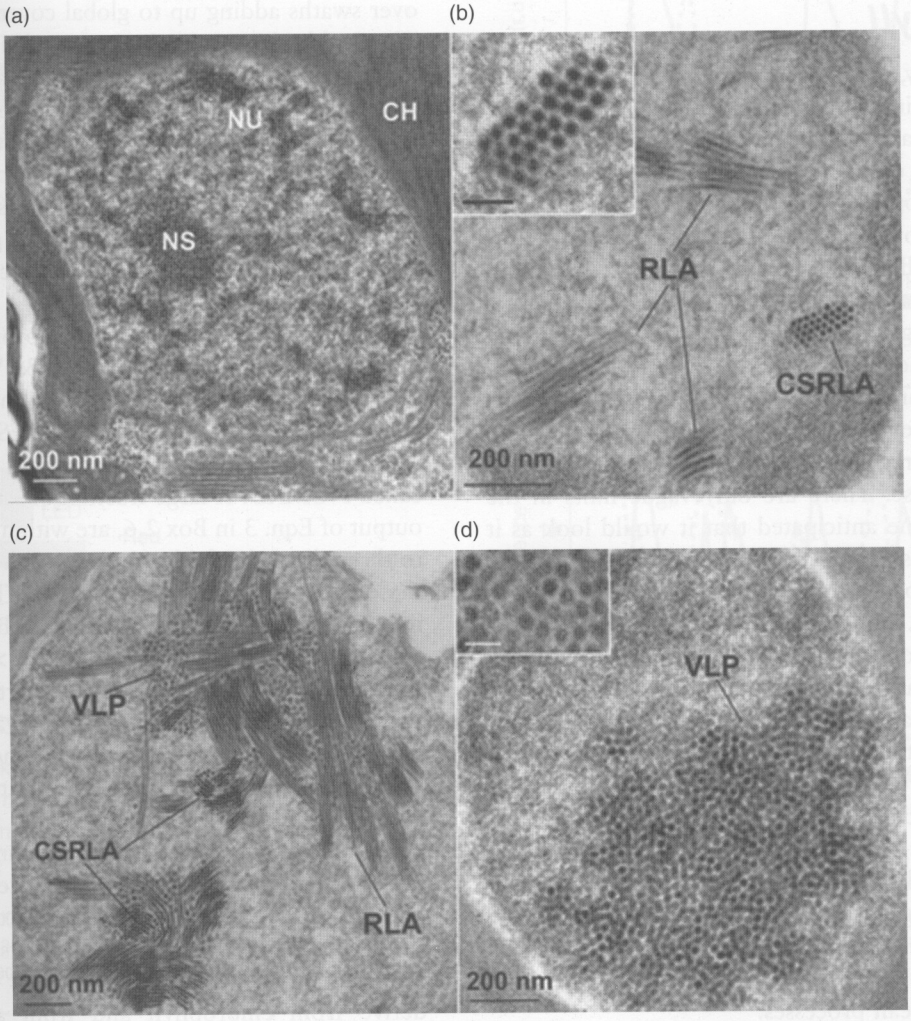


Fig. 2.15 Transmission electron micrographs of *Chaetoceros* cf. *wighamii*. (a) Section of a healthy nucleus: **NU** – nucleus, **NS** – nucleolus, **CH** – choroplast). (b) Section of a cell at early infection, showing rod-like arrays (**RLA**) and rod-like arrays in cross-section (**CSRLA**), including an insert for more detail (scale bar, 50 nm). (c) Thin section of a cell at mid-infection, showing virus-like particles (**VLP**), RLA, and CSRLA. (d) Thin section of a cell at late infection, showing VLP, including an insert for more detail (scale bar, 50 nm). (After Eissler *et al.* 2009, with permission from the *Journal of Phycology*.)

stock. Chlorophyll-*a* is present in almost all phytoplankton, and the amount of it in seawater is a reasonable, if imperfect, measure of phytoplankton standing stock. Moreover, chlorophyll concentration is relatively easy to determine. Phytoplankton are removed from a known volume of water with a suitably fine filter (glass-fiber mesh is most common), then chlorophyll is extracted with acetone and quantified by spectrometry, chromatography, or, since it shows red fluorescence in blue illumination, with a fluorometer. Parsons *et al.* (1984) give typical recipes for these techniques.

Greater spatial and temporal resolution of variations in phytoplankton stocks is often acquired with *in situ* fluorometers (Box 2.5). These can be deployed on moorings for time-series sampling, or lowered through the water column to determine vertical profiles of phytoplankton standing stocks. *In situ* fluorometric measurements are then calibrated with a set of extracted chlorophyll measurements.

Satellite-based estimates of chlorophyll

All of oceanography, particularly its physical and biological aspects, has been intensely challenged by satellite data. Snapshots from space of temperature distributions have challenged physical oceanographers, because earlier analyses of data taken from ships were blurred by widely spaced stations and the motion of patterns during sampling (low "synopticity"). Satellites swing across the Earth in minutes, gathering images from very wide swaths. Some sit in geostationary orbits and get instantaneous, nearly whole hemisphere images. In the mid-1970s, we were suddenly able to see the layout of variability on scales of a few kilometers. This reveals offshore jets and eddies, current meanders and surface ring structures. The dynamism of ocean processes was a major surprise. There are only vague hints in the literature that anyone anticipated that it would look as it does from this distant and instantaneous perspective. Physicists had dropped the necessary higher-order terms in their hydrodynamic models, thinking they were too small to have significant, observable effects. From the shipboard perspective, they had been right. From the instantaneous perspective they had blurred the picture dramatically. Biologists were surprised by similar data. The jets and swirls show up not only in satellite temperature maps (e.g. AVHRR), but in pictures based on ocean color, variation of which is mostly determined by chlorophyll content. Here, too, the swirls and jets, first seen in Coastal Zone Color Scanner images, are impressive. A picture (book cover) of the northwest Atlantic shows (albeit in "false" color) some of the dramatic features that changed the understanding of ocean processes.

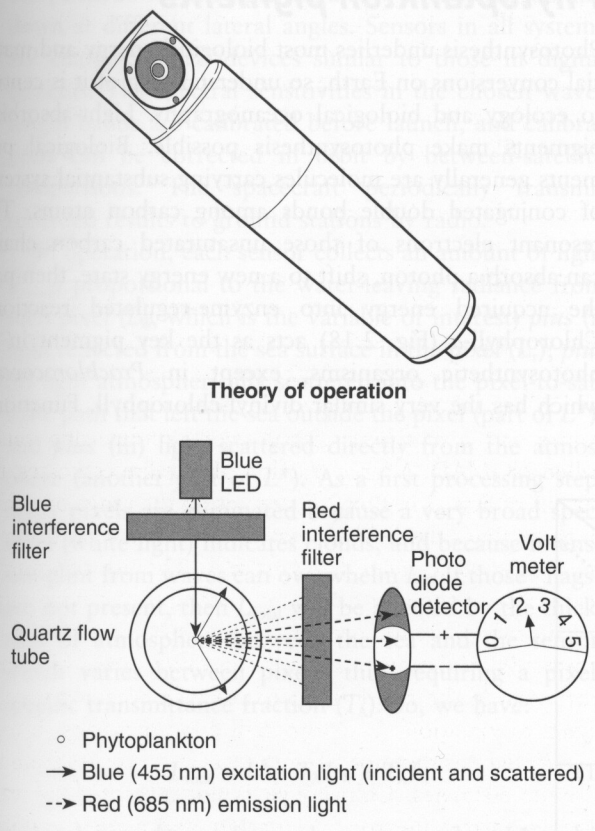
Just as impressive are the short-term changes in quantities and distribution from one picture date or season to the next. Satellites provide new pictures on time scales of less than a week, the intervals required for records from orbital swaths to accumulate as regional or global images. Short-term and seasonal comparisons become possible, and the general correlation between production rates and chlorophyll standing stock makes possible more-or-less convincing estimates of regional and global primary production rates. We can add up approximations of how much carbon is being incorporated in organic matter and get at large-scale biogeochemical transformation rates with moderate precision. Recently and currently active color sensors on satellites include SeaWiFS (now dead), MODIS-AQUA (fading), MERIS (European Space Agency), Oceansat (India), and FY1-D (China). You can find an archive of Level 3 (i.e. elaborately corrected, averaged, and mapped) SeaWIFS images at http://seawifs.gsfc.nasa.gov/SEAWIFS.html. Monthly global averages available there show the dominant features of seasonal cycling of phytoplankton stock in all ocean regions, except for the most polar areas and the two subpolar zones in their respective winters.

SeaWiFS, operational from September 1997, recorded over swaths adding up to global coverage every two days. The Goddard Space Flight Center of NASA produced processed images rapidly, with weekly averages as global images appearing a few days after the data were in, monthly averages right at the end of the month, and so on. The global picture (Plate 2.3) shows the layout of blue (low chlorophyll) and green to red (artificial colors indicating high and higher chlorophyll) regions across the globe. Note that the current satellite array also generates color pictures of the land, which have their own uses.

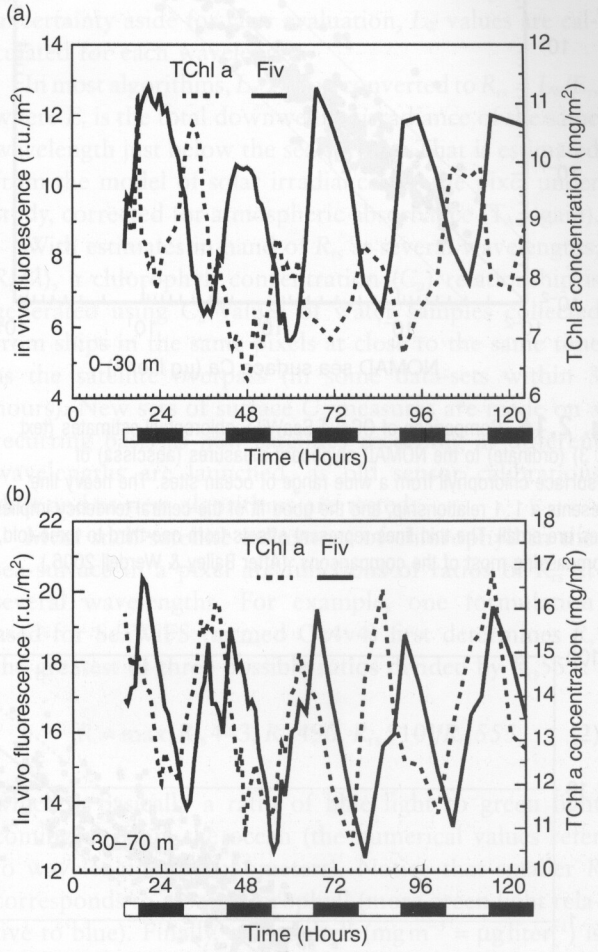
Satellite-based estimates of chlorophyll fit ship-based data in a general fashion (Bailey & Werdell 2006) (Fig. 2.16) over a range from 0.02 to ~20 $mg\,m^{-3}$. The comparison in Fig. 2.16 is between SeaWiFS OC4v4 (Box 2.6) values and a matching global set of surface-ship values called NOMAD. A large fraction of satellite estimates, the output of Eqn. 3 in Box 2.6, are within the range one-third to three times the field estimates, even somewhat better in the oligotrophic range (<0.3 $mg\,m^{-3}$). The use of logarithmic scales in the plot accommodates both the wide range of chlorophyll in the oceans (0.01 to >20 $mg\,m^{-3}$) and the substantial variation (essentially uncertainty for biological purposes) in the relationship of the water-leaving spectral signal to variations in chlorophyll concentrations. It must be pointed out that not all of the variation comes from the satellite estimates. Chlorophyll is not measured by water sampling without error or variation, and modest variation is expected across the range of pixels included in the patches averaged for comparison to the ship data. Furthermore, roughly equal amounts of in-water light attenuation, and thus water-leaving spectral variation, derive from chlorophyll and from accessory pigments of phytoplankton. At any given concentration, there is significant variation in the ratio of their concentrations and thus their effects, but the ratios examined (on log–log

Box 2.5 *In situ* fluorometry

Because they contain chlorophyll, phytoplankton can be quantified *in situ* by exciting, then measuring, their fluorescence. Water adjacent to a small window on an *in situ fluorometer* (Box Fig. 2.5.1) is flashed with a xenon lamp filtered at 455 nm, and the resulting fluorescence due to chlorophyll at 685 nm is measured with a photomultiplier circuit. Corrections for light-source variation are made by reporting the ratio of fluorescence to light intensity measured internally in the housing. Neveux *et al.* (2003) made periodic calibrations of this signal by filtering phytoplankton from the vicinity of the sensor and extracting chlorophyll for determination by spectrofluorometry. Fluorescence measured in this way for near-surface water samples varies strongly with external illumination. Daytime fluorescence is reduced by non-photochemical quenching where the excitation energy is transferred to photoprotective pigments or dissipated as heat. Maximum fluorescence is seen at night. This variation decreases with depth. Thus, there is artifactual day–night and depth variation (Box Fig. 2.5.2). Careful accounting for such effects must be incorporated in field studies with fluorometers. These instruments are deployed as vertical water-column profilers and as moored recorders.



Box Fig. 2.5.1 A commercial fluorometric chlorophyll *a* recording device with a diagram of its operating principle. Blue light is beamed into an observation space and the resulting red fluorescence is measured by a detector and recorded (Chelsea Instruments).



Box Fig. 2.5.2 Diel variations of integrated *in vivo* chlorophyll fluorescence (**Fiv** – solid line) in the (a) 0–30 m and (b) 30–70 m layers during a 5-day time series at the equator. Comparison with diel variations of integrated total extracted chlorophyll-*a* (**TChl** – a dashed line). (After Neveux *et al.* 2003.)

scales) over the entire range from severe oligotrophy to pea-soup green waters are well fitted by a 1:1 relationship (Trees *et al.* 2000). Those two aspects of chlorophyll and accessory pigment variation contribute both to the overall excellence of chlorophyll-*a* (C_a) estimates relative to *in situ* estimates and to the substantial variation around the general trend. While the satellite estimates of C_a are not always accurate for any given pixel, with sufficient areal averaging the data are very useful because the estimator shows no obvious bias. Regional and global totals benefit from massive averaging and should be reasonably accurate.

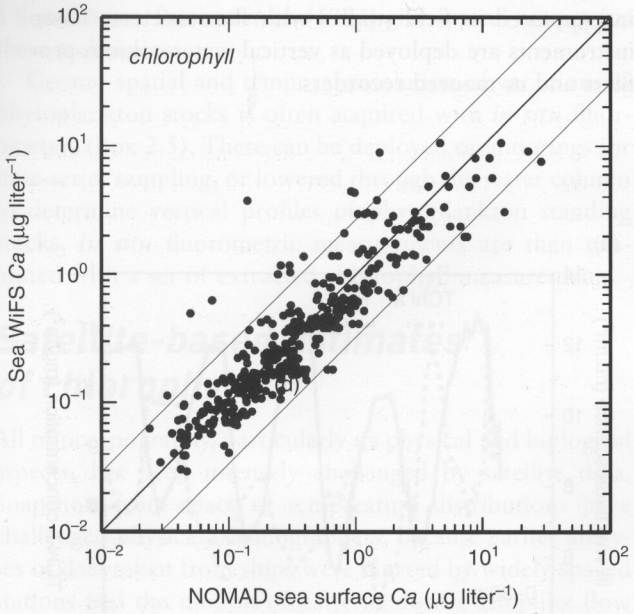


Fig. 2.16 Comparison of OC4v4 SeaWiFS chlorophyll estimates (text Eqn. 3) (ordinate) to the NOMAD shipboard measures (abscissa) of sea-surface chlorophyll from a wide range of ocean sites. The heavy line represents a 1:1 relationship, and the good fit of the central tendency implies biases are small. The thin lines represent offsets from one-third to three-fold, encompassing most of the comparisons. (After Bailey & Werdell 2006.)

On the other hand, the NOMAD (or any sea-surface-"truth" data-set) can be dissected into regional subsets, and those often show substantial offsets from the matching surface data, particularly in coastal regions. For example, a plot of surface chlorophyll concentration from NOMAD vs. SeaWiFS C_a estimates from Eqn. 3 (see Box 2.6) from coastal sites in the Atlantic Ocean (Fig. 2.17) shows substantial overestimates from the satellite algorithm for both moderately rich North American waters (near 1 mg Chl m^{-3}) and for the very oligotrophic Mediterranean. The match is better, at least apparently unbiased, for estimates from near the southwest coast of Africa. In contrast, C_a is generally underestimated for waters around the Palmer Peninsula in Antarctica (not shown).

Phytoplankton pigments

Photosynthesis underlies most biological energy and material conversions on Earth, so understanding of it is central to ecology and biological oceanography. Light-absorbing pigments make photosynthesis possible. Biological pigments generally are molecules carrying substantial systems of conjugated double bonds among carbon atoms. The resonant electrons of those unsaturated carbon chains can absorb a photon, shift to a new energy state, then pass the acquired energy into enzyme-regulated reactions. Chlorophyll-*a* (Fig. 2.18) acts as the key pigment in all photosynthetic organisms, except in *Prochlorococcus*, which has the very similar divinyl-chlorophyll. Functional

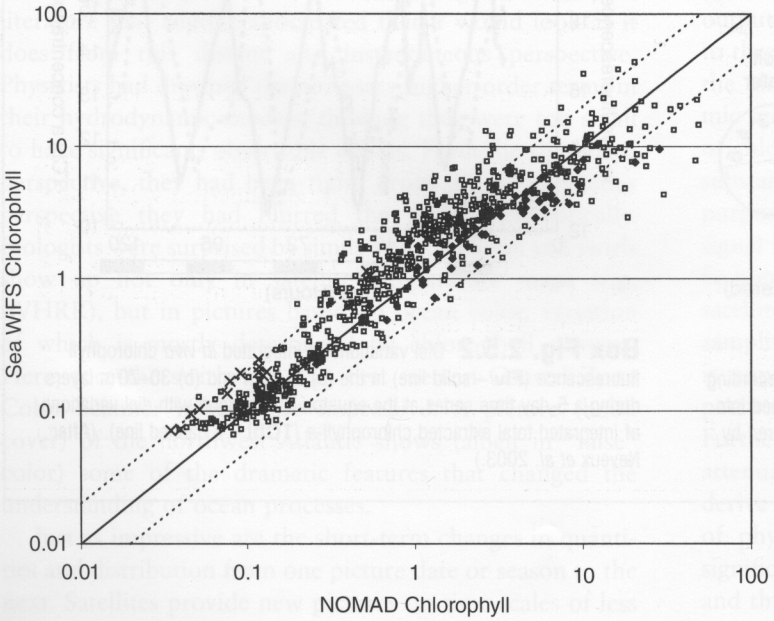


Fig. 2.17 Comparison of OC4v4 SeaWiFS chlorophyll estimates to NOMAD estimates from just Atlantic coastal sites. Lines represent the 1:1 relation and ½- to two-fold variation. Open squares are from along the North American coast; filled diamonds from off SW Africa; and **X**s from the Mediterranean. (Analysis and graph generously provided by Janet W. Campbell, University of New Hampshire.)

Box 2.6 Algorithms for satellite-based estimates of chlorophyll

The exact algorithms for SeaWiFS, MODIS, and MERIS data interpretation are not simple, and a sort of insiders-only technical haze covers the methodology. Outlines for many of the component algorithms are at:

http://oceancolor.gsfc.nasa.gov/DOCS/MSL12/master_prodlist.html/#prod11

Here is a general description for SeaWiFS; similar techniques are used for MODIS, MERIS and CZCS data. During daylight, a suite of spectral sensors looks down a telescope "folded" with prisms into a compact package. The image at the "eyepiece" of the telescope subtended a small (~1.2 × 1.2 km) sea-surface pixel. The telescope swung left and right across the path passing under the satellite, recording light from successive pixels. Dichroic beam-splitters and color filters divided light from each pixel into eight spectral bands, one for each recording sensor. Optical details differ among satellites; MERIS for example has a row of cameras looking down at different lateral angles. Sensors in all systems are charge-coupled devices similar to those in digital cameras. The spectral sensitivities in the chosen wavelength bands are calibrated before launch, and calibrations can be corrected in orbit by between-satellite comparisons. The spacecraft periodically transmit recorded results to ground stations by radio.

In operation, each sensor collects an amount of light (L_{total}) proportional to the water-leaving radiance from each pixel (L_W, which is the variable of interest) *plus* (i) light reflected from the sea surface in the pixel (L_r); *plus* (ii) light atmospherically scattered into the pixel-to-satellite path that left the sea outside the pixel (part of L^*); and *plus* (iii) light scattered directly from the atmosphere (another part of L^*). As a first processing step, many pixels are eliminated because a very broad spectrum (white light) indicates clouds, and because intense sun-glint from waves can overwhelm L_W. If those "flags" are not present, then L_{total} will be affected by the thickness of atmosphere between the sea and the sensor, which varies between pixels, thus requiring a pixel-specific transmittance fraction (T_A). So, we have:

$$L_{total} = L^* + T_A L_W + T_A L_r. \quad (1)$$

L_r is taken to be modest and proportional to L^* and is lumped with it, giving $L_{total} = L^* + T_A L_W$. L^* is estimated from solar irradiance, which is approximated from a model (see just below). Transmittance can vary with atmospheric conditions, and T_A is estimated from some ratios of received irradiance at several wavelengths known not to vary from each other in their L_W. L_W is <10% of L_{total}, and a great deal of uncertainty in the L_W estimate is incurred in approximating T_A. All of the calculations, including the incoming irradiance model, must account for the zenith angle of the sun, the zenith angle of the satellite from observed pixels, and other angles including the curvature of the Earth (usually ignored by only using swaths near the satellite's nadir). These angles vary with time-of-day, season, and latitude, and the solar input varies with the distance from the sun (greater in summer, less in winter). Setting possible uncertainty aside for later evaluation, L_W values are calculated for each wavelength.

In most algorithms, L_W is next converted to $R_{rs} = L_W/E_t$, where E_t is the total downwelling irradiance of the same wavelength just below the sea surface. That is estimated from the model of solar irradiance for the pixel under study, corrected for atmospheric absorbance (T_A, again).

With estimates in hand of R_{rs} at several wavelengths, $R_{rs}(\lambda)$, a chlorophyll concentration (C_a) relationship is generated using C_a values of water samples collected from ships in the same pixels at close to the same time as the satellite overpass (in some data-sets within 3 hours). New sets of surface C_a measures are made on a recurring basis as new satellites recording at different wavelengths are launched, as old sensor calibrations shift and as new algorithms are tested.

Most such satellite estimators of chlorophyll in the sea surface in a pixel are functions of ratios of R_{rs} at several wavelengths. For example, one formulation used for SeaWiFS (termed Oc4v4) first determines R, the greatest of three possible ratios divided by $R_{rs}555$:

$$R = \max[R_{rs}443, R_{rs}490, R_{rs}510]/R_{rs}555, \quad (2)$$

which is basically a ratio of blue light to green light coming up from the ocean (the numerical values refer to wavelengths in nanometers). Notice that smaller R corresponds to more chlorophyll (more green light relative to blue). Finally, chlorophyll ($mg\,m^{-3} = \mu g\,liter^{-1}$) is calculated from the fitted function:

$$C_a\ (mg\ m^{-3}) = \text{anti}\log_{10}(0.366 - 3.067R + 1.930R^2 + 0.649R^3 - 1.532R^4) \quad (3)$$

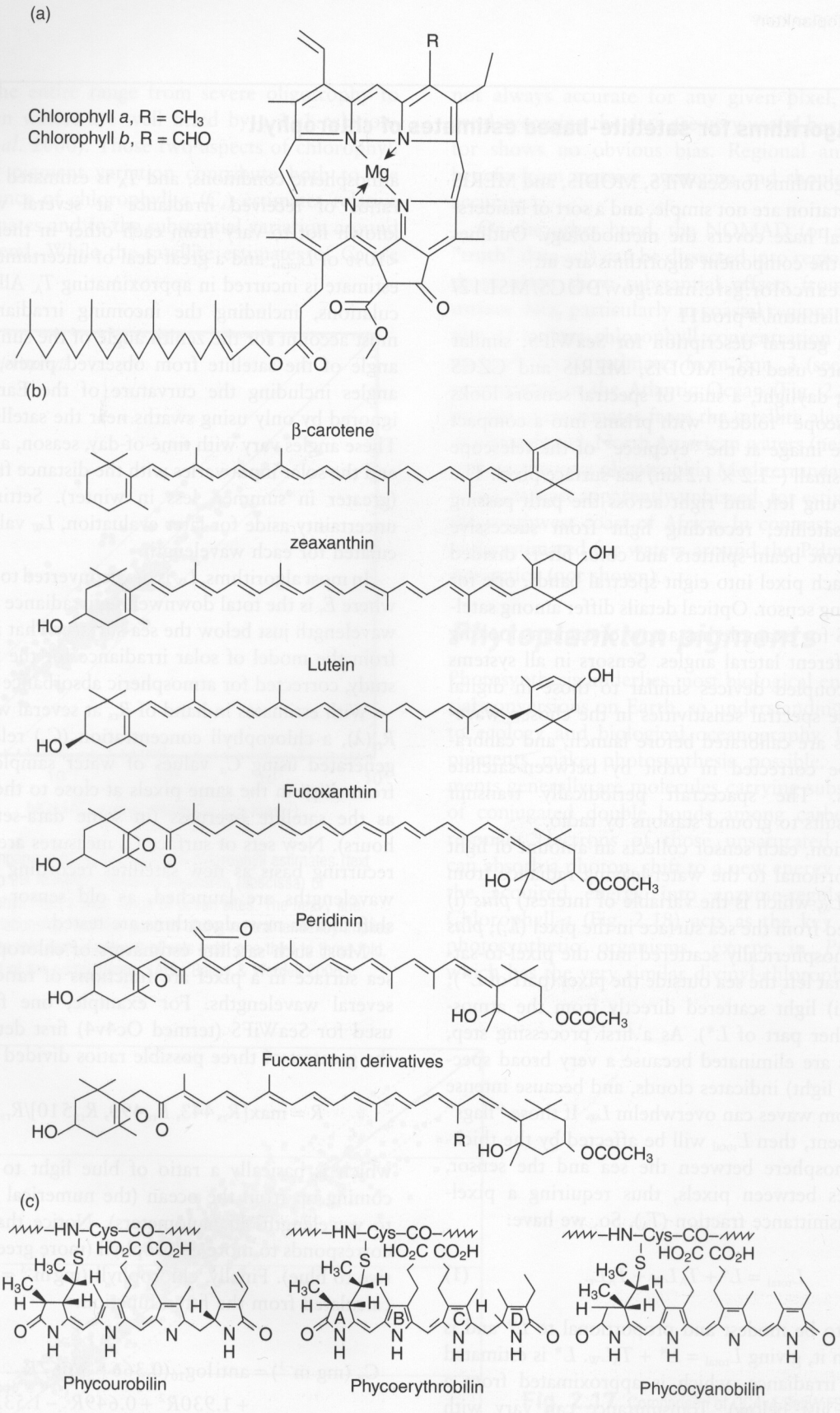


Fig. 2.18 Chemical structures of some abundant phytoplankton pigments: (a) Chlorophylls-*a* and *b*. (b) β-carotene, the most common carotenoid, and five xanthophyll derivatives. The R side chains for 19′ butanyloxyfucoxanthin and 19′-hexanyloxyfucoxanthin are: $CH_2\text{-}O\text{-}\overset{O}{\overset{\|}{C}}\text{-}C_3H_7$ and $CH_2\text{-}O\text{-}\overset{O}{\overset{\|}{C}}\text{-}C_5H_{11}$ respectively. (c) Several versions of phycoerythrin-like pigments based on open pyrrole structures.

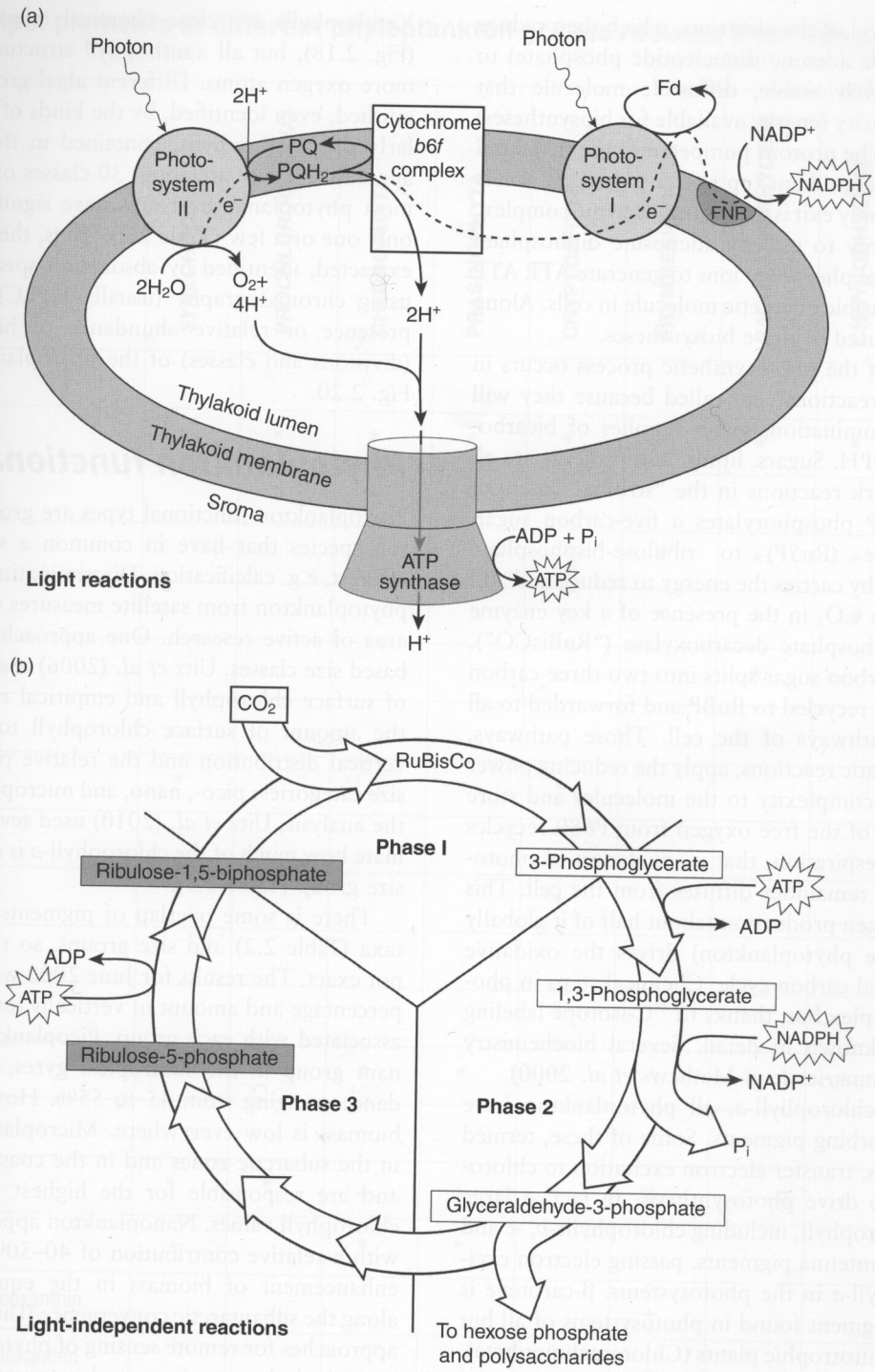


Fig. 2.19 Biochemical pathways of photosynthesis.

chlorophyll-*a* includes a large protein matrix that presents to the light a porphyrin ring with a magnesium atom held by central ligands. The ring has a tail, a linear carbon chain called phytol, by which it attaches to the protein portion of the system. Chlorophyll-*a* is associated with other protein-bound pigments (carotenoids, xanthophylls and several chlorophyll variants) in one of two types of "photosystem": Photosystem I, or P700 for its wavelength of maximum absorption and Photosystem II, or P680. P680 applies absorbed energy (Fig. 2.19) to capture electrons from water, releasing protons ($4H^+$) that are actively transported into the thylakoid lumen, and oxygen (O_2) that diffuses out of the thylakoids. The electrons move in a complex series of transfers to P700. More photic energy is absorbed there,

raising the energy level of the electrons, which then reduce $NADP^+$ (nicotinamide adenine dinucleotide phosphate) to NADPH, a moderately stable, diffusible molecule that carries reducing capacity (energy available for biosyntheses) into the cytoplasm. The protons pumped into the thylakoid generate an energetic gradient (up to 3.5 pH units) across the membrane. The only exit is through an enzyme complex, which uses the energy to esterify adenosine diphosphate (ADP) with additional phosphate ions to generate ATP. ATP is the principal, diffusible energetic molecule in cells. Along with NADPH, it is used to drive biosyntheses.

The remainder of the photosynthetic process occurs in "light-independent reactions", so called because they will proceed without illumination, given supplies of bicarbonate, ATP and NADPH. Sugars, lipids, and proteins are all produced by the dark reactions in the "stroma" layers of the thylakoids. ATP phosphorylates a five-carbon sugar, ribulose-5-phosphate (Ru5P) to ribulose-bisphosphate (RuBP), which thereby carries the energy to reduce and add a carbon atom from CO_2 in the presence of a key enzyme called ribulose bisphosphate decarboxylase ("RuBisCO"). The resulting six-carbon sugar splits into two three-carbon sugars that are both recycled to RuBP and forwarded to all the biosynthetic pathways of the cell. Those pathways, sequences of enzymatic reactions, apply the reducing power of NADPH to add complexity to the molecules and store more energy. Some of the free oxygen from P680 recycles within the cell to respiration, that is, oxidation of photosynthate, while the remainder diffuses from the cell. This photosynthetic oxygen production (about half of it globally comes from marine phytoplankton) drives the oxidative side of the ecological carbon cycle. Chemical steps in photosynthesis are complex but, thanks to ^{14}C-isotope labeling of substrates, are known in detail. Several biochemistry texts give good summaries (e.g. Mathews *et al.* 2000).

In addition to chlorophyll-*a*, all phytoplankton have accessory light-absorbing pigments. Some of these, termed "antenna" pigments, transfer electron excitation to chlorophyll-*a* in order to drive photosynthesis. In fact, a large fraction of the chlorophyll, including chlorophylls-*b*, -*c* and some -*a*, serves as antenna pigments, passing electron excitation to chlorophyll-*a* in the photosystems. β-carotene is another antenna pigment found in photosystems of all but one rare group of autotrophic plants (Chlorarachniophyta). Carotenes are hydrocarbon molecules with conjugated double bonds (Fig. 2.18) in chains long enough to retain and transfer photonic excitation. The Cyanophyta and Rhodophyta have distinctive pigments with this function, the phycobilins (Fig. 2.18), in which alternating double and single bonds occur in a tetrapyrrole resembling the porphyrin ring of chlorophyll, but not closed.

Other pigments, termed protective pigments, absorb photons to prevent photolytic damage to chlorophyll and the rest of the photosynthetic apparatus. Most of these belong to the class of pigments called xanthophylls. Xanthophylls are close chemical relatives of β-carotene (Fig. 2.18), but all xanthophyll structures include one or more oxygen atoms. Different algal groups can be distinguished, even identified, by the kinds of pigments, particularly the xanthophylls, contained in their photosynthetic apparatus. There are about 30 classes of xanthophylls, but most phytoplankton groups have significant quantities of only one or a few (Table 2.2). Thus, these pigments can be extracted, identified by absorption spectra and quantified using chromatography (usually HPLC) to determine the presence or relative abundance of higher-order groups (divisions and classes) of the phytoplankton, as shown in Fig. 2.20.

Phytoplankton functional types

Phytoplankton functional types are groups of phytoplankton species that have in common a specific function of interest, e.g. calcification. Discriminating distinct groups of phytoplankton from satellite measures of ocean color is an area of active research. One approach is to use pigment-based size classes. Uitz *et al.* (2006) used satellite measures of surface chlorophyll and empirical relationships linking the amount of surface chlorophyll to the pattern of its vertical distribution and the relative proportions in three size categories: pico-, nano, and microplankton. To simplify the analysis, Uitz *et al.* (2010) used seven pigments to estimate how much of the chlorophyll-*a* is associated with each size group (Table 2.3).

There is some overlap of pigments among the various taxa (Table 2.2) and size groups, so the relationships are not exact. The results for June 2000 were calculated as the percentage and amount of vertically integrated chlorophyll associated with each group. Picoplankton form the dominant group in the subtropical gyres, with relative abundances ranging from 45 to 55%. However, picoplankton biomass is low everywhere. Microplankton are dominant in the subarctic zones and in the coastal upwelling zones, and are responsible for the highest vertically integrated chlorophyll values. Nanoplankton appear to be ubiquitous, with a relative contribution of 40–50%, with a significant enhancement of biomass in the equatorial regions and along the subantarctic convergence (Uitz *et al.* 2010). Other approaches for remote sensing of phytoplankton functional types include use of spectral measurements to define six phytoplankton groups (Alvain *et al.* 2008) and spectral retrievals of satellite-measured backscattering to define three size classes of phytoplankton (Kostadinov *et al.* 2010). Each of these approaches lacks the specificity of actual species identification and enumeration, but does provide a synoptic view of regional, seasonal (Fig. 2.20), and interannual variability of some phytoplankton groups in the ocean. Availability of newer spectral sensors for future satellite deployment may provide higher resolution for discriminating specific groups.

Table 2.2 Principal pigments in different phytoplankton groups reduced from Van den Hoek *et al.* (1995).

	SYNECHOCOCCUS	*PROCHLOROCOCCUS*	CHLOROPHYTA	PRASINOPHYTA	CRYPTOPHYTA	PRYMNESIOPHYTA	BACILLARIOPHYCEAE	CHRYSOPHYCEAE	XANTHOPHYCEAE	DINOPHYTA
Chlorophylls										
a	*	*	*	*	*	*	*	*	*	*
b		*	*	*						
*c*1						*	*	*	+	
*c*2					*	*	*	*	+	*
*c*3							*			
Phycobilins										
phycocyanin	*				*					
allophycocyanin	*									
phycoerythrin	*				*					
Carotenes										
α					*			+		
β	*	*	*	*	*	*	*	*	*	*
Xanthophylls										
zeaxanthin	*	*	+	+		+		+		
violaxanthin			*	*						
19′-hexanoyloxyfucoxanthin						+				
19′-butanoyloxyfucoxanthin						+				
diatoxanthin						*	*	+	*	
diadinoxanthin						*	*	+	*	*
alloxanthin					*					
peridinin										*
neoxanthin							+	+	+	

*, important pigment; +, pigment present.

Table 2.3 Pigments used as biomarkers to estimate abundance of phytoplankton functional types from satellite-derived estimates of surface chlorophyll (Uitz *et al.* 2010).

PIGMENT	ABBREVIATION	PHYTOPLANKTON GROUP	SIZE CLASS
Fucoxanthin	Fuco	Diatoms	Microplankton
Peridinin	Perid	Dinoflagellates	Microplankton
19′-hexanoyloxyfucoxanthin	Hex-fuco	Prymnesiophytes	Nanoplankton
19′-butanoyloxyfucoxanthin	But-fuco	Prymnesiophytes	Nanoplankton
Alloxanthin	Allo	Cryptophytes	Nanoplankton
Chlorophyll-*b* + divinyl Chl *b*	TChlb	*Prochlorococcus*	Picoplankton
Zeaxanthin	Zea	Cyanobacteria	Picoplankton

Equations for calculating the fraction (f) of chlorophyll attributed to each size group:
$f_{micro} = (1.41[Fuco] + 1.41[Peri])/\Sigma DP_w$
$f_{nano} = (1.27[Hex\text{-}fuco] + 0.35[But\text{-}fuco] + 0.60[Allo])/\Sigma DP_w$
$f_{pico} = (1.01[TChlb] + 0.86[Zea])/\Sigma DP_w$
where ΣDP_w represents the chlorophyll-*a* concentration reconstructed from the knowledge of the seven other pigments.

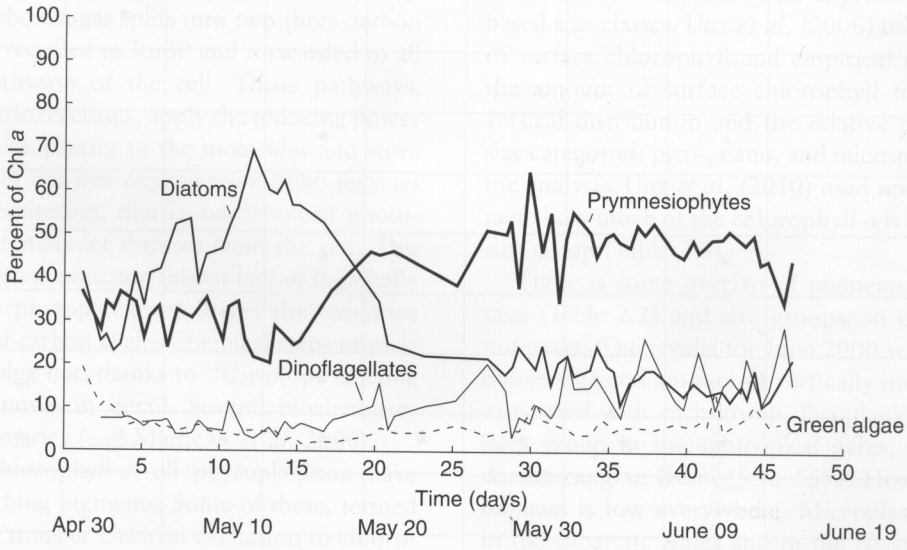


Fig. 2.20 Fractions of chlorophyll-*a* associated with accessory pigments from different phytoplankton groups (as labeled) during the course of the spring bloom in the northwest Atlantic (approximately 49°N, 20°W) in 1990. (After Barlow *et al.* 1993.)

In closing

This brief introduction covers just that part of phycology applicable to phytoplankton. Knowledge of algae is extensive and detailed. Because knowledge of algae is increasing rapidly with use of modern tools for microscopy, biochemistry, and molecular genetics, occasional hours in the library or on-line with recent issues of phycological journals are essential education maintenance for biological oceanographers. Refinements of techniques for *in situ* and remote sensing of phytoplankton distributions are providing important information for regional and global studies of variations in the standing stocks of the major phytoplankton groups. We address environmental factors affecting the algal growth rates and primary production in Chapter 3.