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Applications and uses of diatoms: prologue

JOHN P. SMOL AND EUGENE F. STOERMER

This book is about the uses of diatoms (Class Bacillariophyceae), a group of microscopic algae abundant in almost all aquatic habitats. There is no accurate count of the number of diatom species; however, estimates on the order of 10^4 are often given (Guillard & Kilham, 1977), although Mann & Droop (1996) point out that this number would be raised to at least 10^5 by application of modern species concepts. Diatoms are characterized by a number of features, but are most easily recognized by their siliceous (opaline) cell walls, composed of two valves, that, together with the girdle bands, form a frustule (Figure 1.1). The size, shape, and sculpturing of diatom cell walls are taxonomically diagnostic. Moreover, because of their siliceous composition, they are often very well preserved in fossil deposits and have a number of industrial uses.

The main focus of this book is not the biology and taxonomy of diatoms, although Julius & Theriot (this volume) provide a “primer” on this subject, and a number of chapters touch on these topics. Other books (e.g. Round *et al.*, 1990) and the review articles and books cited in the following chapters, provide introductions to the biology, ecology, and taxonomy of diatoms. Instead, our focus is on the applications and uses of diatoms to the environmental and earth sciences. Although this book contains chapters on practical uses, such as uses of fossilized diatom remains in industry, oil exploration, and forensic applications, most of the book deals with using these indicators to decipher the effects of long-term ecological perturbations, such as climatic change, lake acidification, and eutrophication. As many others have pointed out, diatoms are almost ideal biological monitors. There are a very large number of ecologically sensitive species, which are abundant in nearly all habitats where water is at least occasionally present.

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Importantly, diatom valves are typically well preserved in the sediments of most lakes and many areas of the oceans, as well as in other environments.

Precisely when and how people first began to use the occurrence and abundance of diatom populations directly, and to sense environmental conditions and trends, is probably lost in the mists of antiquity. It is known that diatomites were used as a palliative food substitute during times of starvation (Taylor, 1929), and Bailey’s notes attached to the type collection of *Gomphonopsis herculeana* (Ehrenb.) Cleve (Stoermer & Ladewski, 1982) indicate that masses of this species were used by native Americans for some medicinal purpose, especially by women (J. W. Bailey, unpublished notes associated with the type gathering of *G. herculeana*, housed in the Humboldt Museum, Berlin). It is interesting to speculate how early peoples may have used the gross appearance of certain algal masses as indications of suitable water quality (or contra-indications of water suitability!), or the presence of desirable and harvestable fish or invertebrate communities.

However, two great differences separate human understanding of higher plants and their parallel understanding of algae, particularly diatoms. The first is direct utility. Anyone can quite quickly grasp the difference between having potatoes and not having potatoes. It is somewhat more difficult to establish the consequences of, for example, *Cyclotella americana* Fricke being extirpated from Lake Erie (Stoermer *et al.*, 1996).

The second is perception. At this point in history, nearly any person living in temperate latitudes can correctly identify a potato. Some people whose existence has long been associated with potato culture can provide a wealth of information, even if they lack extended formal education. Almost any university will have individuals who have knowledge of aspects of potato biology or, at a minimum, know where this rich store of information may be obtained. Of course, knowledge is never perfect, and much research remains to

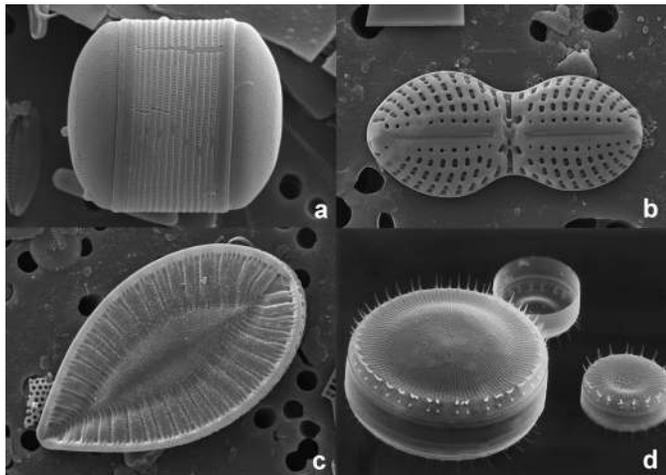


Figure 1.1 Scanning electron micrographs of some representative diatoms: (a) *Hyalodiscus*; (b) *Diploneis*; (c) = *Surirella*; and (d) *Stephanodiscus*. Micrographs a, b, and c courtesy of I. Kaczmarek and J. Ehrman; micrograph d courtesy of M. B. Edlund.

be done before our understanding of potatoes approaches completeness.

Diatoms occupy a place near the opposite end of the spectrum of understanding. Early peoples could not sense individual diatoms, and their only knowledge of this fraction of the world's biota came from mass occurrences of either living (e.g. biofilms) or fossil (diatomites) diatoms. Even in today's world, it is difficult to clearly and directly associate diatoms with the perceived values of the majority of the world's population. The consequences of this history are that the impetus to study diatoms was not great. Hence, many questions concerning basic diatom biology remain to be addressed. Indeed, it is still rather rare to encounter individuals deeply knowledgeable about diatoms even amongst university faculties. This, however, is changing rapidly.

What is quite clear is that people began to compile and speculate upon the relationships between the occurrence of certain diatoms and other things which were useful to know almost as soon as optical microscopes were developed. In retrospect, some of the theories developed from these early observations and studies may appear rather quaint in the light of current knowledge. For instance, Ehrenberg (see Jahn, 1995) thought that diatoms were animal-like organisms, and his interpretation of their cytology and internal structure was quite different from our modern understanding. Further, his interpretations of the origins of airborne diatoms (he thought they were directly associated with volcanoes) seem rather outlandish today. On the other hand, Ehrenberg did

make phytogeographic inferences which are only now being rediscovered.

As will be pointed out in chapters following, knowledge about diatoms can help us know about the presence of petroleum, if and where a deceased person drowned, when storms over the Sahara and Sub-Saharan Africa were of sufficient strength to transport freshwater diatom remains to the mid-Atlantic, and indeed to the most remote areas of Greenland, as well as many other applications and uses. As will be reflected in the depth of presentation in these chapters, diatoms provide perhaps the best biological index of annual to millennial changes in Earth's biogeochemistry. As it becomes increasingly evident that human actions are exercising ever greater control over the conditions and processes that allow for our existence (Crutzen & Stoermer, 2000), fully exercising all the tools which may serve to infer the direction and magnitude of change, and indeed the limits of change, becomes increasingly imperative. This need has fueled a considerable increase in the number of studies that deal with diatoms, particularly as applied to the problems alluded to above.

The primary motivation for this much-revised and expanded second edition is to compile this rapidly accumulating and scattered information into a form readily accessible to interested readers. A perusal of the literature will show that the authors of the different chapters are amongst the world leaders in research on the topics addressed.

The perceptive reader will also note that, despite their great utility, the store of fundamental information concerning diatoms is not as great as might logically be expected for a group of organisms that constitute a significant fraction of Earth's biomass. For example, readers will find few references to direct experimental physiological studies of the species discussed. Sadly, there are still only a few studies to cite, and practically none of those available was conducted on the most ecologically sensitive freshwater species. Readers may also note that there are some differences of opinion concerning taxonomic limits, even of common taxa, and that naming conventions are presently in a state of flux. These uncertainties are real, and devolve from the history of diatom studies.

As already mentioned, the study of diatoms started relatively late, compared with most groups of macroorganisms. Diatoms have only been studied in any organized fashion for about 200 years, and the period of effective study has only been about 150 years. It is also true that the history of study has been quite uneven. After the development of fully corrected optical microscopes, the study of microorganisms in general, and diatoms in particular, attracted immense

interest and the attention of a number of prodigiously energetic and productive workers. This grand period of exploration and description produced a very substantial, but poorly assimilated, literature. Diatomists who worked toward the end of this grand period of growth produced remarkably advanced insights into cytology and similarly advanced theories of biological evolution (Mereschkowsky, 1903). This, and the fact that sophisticated and expensive optical equipment is required for their study, gained diatoms the reputation of a difficult group of organisms to study effectively. Partially for this reason, basic diatom studies entered a period of relative decline beginning c. 1900, although a rich, if somewhat eclectic, amateur tradition flourished, especially in England and North America. The area that remained most active was ecology. As Pickett-Heaps *et al.* (1984) have pointed out, Robert Lauterborn, an exceptionally talented biologist who was well known for his studies of diatom cytology, could also be appropriately cited as one of the founders of aquatic ecology.

The people who followed often did not command the degree of broad recognition enjoyed by their predecessors, and many of them operated at the margin of the academic world. As examples, Friedrich Hustedt, perhaps the best known diatomist in the period from 1900 to 1960 (Behre, 1970), supported himself and his family as a high-school teacher for much of his career. B. J. Chohnoky (Archibald, 1973) was caught up in the vicissitudes of the Second World War, and produced his greatest works on diatom autecology, including his large summary work (Chohnoky, 1968), after he became an employee of the South African Water Resources Institute. Although many workers of this era produced notable contributions, they were peripheral to the main thrusts of academic ecological thought and theory, particularly in North America. Although this continent had numerous individuals who were interested in diatoms, and published on the group, most of them were either interested amateurs or isolated specialists working in museums or other non-university institutions. For example, when one of us (E. F. S.) decided to undertake advanced degree work on diatoms in the late 1950s, there was no university in the United States with a faculty member specializing in the study of freshwater diatoms.

One of the most unfortunate aspects of separation of diatom studies from the general course of botanical research was substantial separation from the blossoming of new ideas. The few published general works on diatoms had a curious “dated” quality, and relatively little new understanding, except for descriptions of new species. The main impetus that kept this small branch of botanical science alive was applied ecology,

and this was the area that, in our opinion, produced the most interesting new contributions.

The above situation began to change in the late 1950s, partially as a result of the general expansion of scientific research in the post-Sputnik era, and partially as the result of technological advances, particularly in the area of electronics. The general availability of electron microscopes opened new orders of magnitude in resolution of diatom structure, which made it obvious that many of the older, radically condensed, classification schemes were untenable. This released a virtual flood of new, rediscovered, and reinterpreted entities (Round *et al.*, 1990), which continues to grow today. At the same time, the general availability of high-speed digital computers made it possible to employ multivariate statistical techniques ideally suited to objective analysis of modern diatom communities and those contained in sediments (Birks, this volume).

The history of ecological studies centered on diatoms can be roughly categorized as consisting of three eras. The first is what we might term the “era of exploration.” During this period (c. 1830–1900), most research focused on diatoms as objects of study. Work during this period was largely descriptive, be it the topic of the description of new taxa, discovery of their life cycles and basic physiology, or observations of their geographic and temporal distributions. One of the hallmarks of this tradition was the “indicator species concept.” Of course, the age of exploration is not over for diatoms. New taxa have been described at a rate of about 400 per year over the past four decades, and this rate appears to be accelerating in recent years. Basic information concerning cytology and physiology of some taxa continues to accumulate, although at a lesser rate than we might desire.

The second era of ecological studies can be termed the “era of systematization” (c. 1900–1970). During this period, many researchers attempted to reduce the rich mosaic of information and inference concerning diatoms to more manageable dimensions. The outgrowths of these efforts were the so-called systems and spectra (e.g. halobion, saprobion, pH, temperature, etc.). Such devices are still employed, and sometimes modified and improved. Indeed, there are occasional calls for simple indices as a means of conveying information more clearly to managers and the public.

We would categorize the current era of ecological studies focused on diatoms as the “age of objectification.” Given the computational tools now generally available, it is possible to determine more accurately which variables affect diatom occurrence and growth and, more importantly, do so quantitatively, reproducibly, and with measurable precision.



Figure 1.2 Some of the diverse ways that diatoms are collected for use in environmental and earth science applications. (a) Collecting a sediment core from Lake of the Woods. Photograph by K. Rühland. (b) Epilithic diatoms being sampled from a rock substrate from a High Arctic pond. Photograph by J. P. Smol. (c) Scuba divers collecting marine diatoms near Guam. Photograph by M. Scheffer.

Thus, applied studies based on diatoms have been raised from a little-understood art practised by a few extreme specialists, to a tool that more closely meets the general expectations of science and the users of this work, such as environmental managers.

The result is that we now live in interesting times. Diatoms have proven to be extremely powerful indicators with which to explore and interpret many ecological and practical problems. They are used in a variety of settings, using different approaches (Figure 1.2). The continuing flood of new information will, without doubt, make the available tools of applied ecology even sharper. It is also apparent that the maturation of this area of science will provide additional challenges. Gone are the comfortable days when it was possible to learn the characteristics of most freshwater genera in a few days and become



Figure 1.2 (cont.)

familiar with the available literature in a few months. Although we might sometimes wish for the return of simpler days, it is clear that this field of study is rapidly expanding, and it is our conjecture that we are on the threshold of even larger changes. The motivation for producing this updated volume is to summarize recent accomplishments and, thus, perhaps make the next step easier.

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2

The diatoms: a primer

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 EDWARD C. THERIOT

2.1 Introduction

Diatoms have long been lauded for their use as powerful and reliable environmental indicators (Cholnoky, 1968; Lowe, 1974). This utility can be attributed to their high abundance and species diversity, which are distributed among most aquatic environments. Additionally, their remains are highly durable and well preserved in accumulated sediments. Often, scientists exploiting the group simply as environmental proxies give little thought as to how and why the species diversity exists in these environments. This may be a by-product of how diatoms are collected and identified. Diatoms are most often recognized by the presence of a siliceous cell wall, the frustule. This structure varies considerably in shape and architecture among species (Figure 2.1) and virtually all taxonomic diagnosis of taxa is based upon frustular morphology. To properly observe diatom frustules for taxonomic identification, living and sedimentary collections are typically subjected to various “cleaning” techniques designed to remove all organic materials (e.g. Battarbee *et al.*, 2001; Blanco *et al.*, 2008), allowing unobstructed observation of the frustule in the microscope. This frequent observation of inorganic components of the cell without reference to the organic features allows observers to “forget” that the specimens seen in the microscope represent individual organisms competing in the selective environments driven by biotic and abiotic ecological pressures. The abundance and taxonomic diversity can be attributed to the extraordinary success of diatoms in the competitive ecological arena.

The casual observer frequently regards diatoms, like most protists, as primitive ancestral lineages to multicellular organisms. While some protists may fit this description, diatoms do not. Diatoms are a relatively recent evolutionary group with the common ancestor’s origin considered to be between 200

and 190 million years before present (Rothpletz, 1896, 1900; Medlin *et al.*, 1997). As a point of reference, the origin of this “first diatom” is approximately 60 to 70 million years younger than the specialized teeth found in mammals, including those in the reader’s mouth (Shubin, 2008). Dates for the origin of the diatom common ancestor are bracketed by molecular clock estimates (Sorhannus, 2007) and the oldest stratigraphic observation (Rothpletz, 1896, 1900). Both of these estimates are inherently biased. The temporal proximity of each estimate to one another does, however, suggest a certain degree of accuracy, given the complimentary nature of the biases. Molecular estimates represent an attempt to identify the absolute moment two populations diverged from one another. The oldest stratigraphic observation represents a period where fossil remains were sufficiently abundant to allow discovery. Given the expected disparity between the moment two populations diverged, and the time it would take divergent populations to develop sufficient numbers allowing paleontological discovery, the 10 million year gap between the two estimates does not appear to be overly large in context of other estimates in this temporal range.

Discussion of when diatoms originated begs the question: what did they originate from? Diatoms share ancestry with heterokonts. Heterokonts (or stramenopiles) are a group of protists with unequal flagella (Leedale, 1974; Hoek, 1978) that includes both chloroplast-bearing and non-chloroplast-bearing representatives (Patterson, 1989) whose common ancestor is thought to have arisen ~725 million years before present (Bhattacharya & Medlin, 2004). The group contains an array of morphologically diverse groups including giant kelps (>60 m) at the large end of the size spectrum and the Bolidomonads and Pelagomonads (1–2 μm) at the small end of the size spectrum (North, 1994; Andersen *et al.*, 1993; Guillou *et al.*, 1999). The heterokonts may be part of the larger “chromalveolate” evolutionary group, which includes

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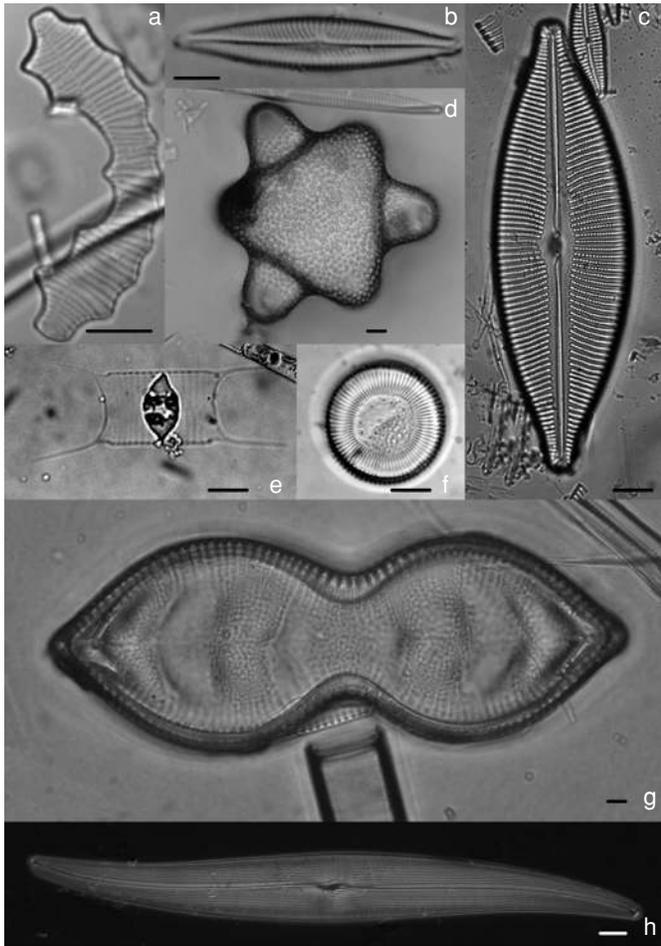


Figure 2.1 Variations in frustule morphology within diatoms species. (a) *Amphicampa mirabilis*, (b) *Navicula cryptocephala*, (c) *Cymbella inaequalis*, (d) *Hydrosera whampoensis*, (e) *Acanthoceras magdeburgensis*, (f) *Cyclotella striata*, (g) *Cymatopleura solea*, (h) *Gyrosigma acuminatum*. Scale bars equal 5 μm .

cryptophytes, dinoflagellates, ciliates, apicomplexans, and haptophytes (Yoon *et al.*, 2002; Cavalier-Smith, 2003; Harper & Keeling, 2003; Ryall *et al.*, 2003; Bachvaroff *et al.*, 2005; Harper *et al.*, 2005). This proposed relationship is controversial and highly debated (e.g., Falkowski *et al.*, 2004; Grzebyk *et al.*, 2004; Keeling *et al.*, 2004; Bachvaroff *et al.*, 2005; Bodyl, 2005). Within heterokonts, individual groups are well established and easily diagnosable, but the relationship between these groups has yet to be definitively identified (Saunders *et al.*, 1995; Sorhannus, 2001; Goertzen & Theriot, 2003). Molecular techniques utilizing multiple data sets have identified the bolidophytes (Goertzen & Theriot, 2003) as the heterokont most closely related to diatoms. The bolidophytes are a group of marine unicellular flagellates that were unknown to science prior to the late 1990s (Guillou *et al.*, 1999). This relatively

recent discovery of the diatoms sister group reflects how much discovery and description-level science remains uncompleted in heterokont biology.

This statement about heterokonts easily extends to diatoms. Once an understanding of origin is achieved, an appreciation should be given to the speed of diversification. Currently, >24,000 diatom species have valid scientific names (Fourtanier & Kociolek, 2009a, b). Many of these have only been illustrated in the literature with light microscopy, and few have yet been the subject of any other genetic, ecological, or physiological study. Mann and Droop (1996) conservatively estimated that there are 200,000 diatom species. If these numbers are taken at face value, 12% of the diatom flora is currently described. This means the modern diatom taxonomic community has a majority of the 24,000 described species to observe in the electron microscope, and an additional 176,000 species to describe. In addition, completing a phylogeny for these 200,000 species should also be an objective. Julius (2007a) demonstrated that the rate of species description in diatoms is approximately 183–185 per year and that this rate has remained constant for nearly a century. At this pace it will take approximately 951 years to describe diatom species completely!

With this in mind, it is easy to understand why the diatom systematics community is still grappling with the collection of detailed ultrastructural information for most species, description of species, and the proper way to analyze these data. Many modern diatom systematic studies deal with taxa at the generic or higher level, avoiding unresolved issues concerning ultrastructure and species concepts. Several recent studies suggest diatom diversity is much greater than previously imagined (Theriot & Stoermer, 1984; Bourne *et al.*, 1992), causing researchers to suggest continued emphasis on species description is most essential in developing phylogenetic hypotheses (Kociolek, 1997; Lange-Bertalot, 1997; Mann, 1997; Round, 1997). In many instances, researchers also continue to argue about what classes of data should be emphasized, valve morphology or cytological features, in classification systems (Round, 1996) without regard to any sense of evolution.

Systematic studies in the twenty-first century must incorporate all types of character information in some sort of an analysis emphasizing the similarity between evolved features (cladistic analysis is currently the most prominent system). This character information must be presented for individual species, not broad generic groups. This requires considerable additional descriptive work. Gradually, a classification more reflective of evolutionary history will develop. Simply put, there is a great deal of work to be done. We are gradually developing

a more structured way of handling the problem and, with luck, progress will be made. One indication of this activity is the distinctly non-linear trend seen in the rate of generic description over the last two decades (Fourtanier & Kociolek, 1999). Generic descriptions increased at an exponential rate during this time, contrasting with the linear rate seen in species. This may indicate that existing taxa are being placed into newly created higher taxonomic categories and a greater interest is being taken in the relationship between one species and another.

2.2 Classification

Modern systematics strives to achieve natural, or monophyletic, groups when designating categories above the species level. These natural groups contain an ancestral lineage and all of its descendants (monophyly). Many taxonomic groups were established prior to the acceptance of monophyly as a goal. Taxonomic designations for diatoms are no exception, and researchers have only recently begun attempts to test and adjust diatom taxonomic schemes to reflect monophyletic groupings. Not all individuals establishing genera and other categories for diatoms view monophyly as a goal, despite its widespread acceptance elsewhere in biology, and proceed in their endeavors in an evolutionary free context (Williams & Kociolek, 2007). Individuals utilizing taxonomic schemes for diatoms should be aware of the unstable status of many higher taxonomic categories (genus and above). Species are frequently moved in and out of categories and new categories are continually being established. This process, hopefully, reflects the gradual transition to a monophyletic taxonomic system and the overwhelming level of species description remaining incomplete. Individuals utilizing diatoms as indicator species often find this fluctuation in higher taxonomic categories frustrating. To circumvent this taxonomic instability, identifications should be made to the species level whenever possible, because species names can always be referenced back to the original population described no matter how many times a name is modified nomenclaturally.

Diatoms are traditionally classified as one of two biological orders, the Centrales (informally referred to as centrics) and the Pennales (or pennates). Diagnostic features cited supporting the two classes typically include (1) valve formation developing radially around a “point” in centrics, contrasted by deposition originating along a “plane” in pennates and (2) oogamous sex with relatively small motile flagella bearing sperm and a large non-motile egg in the centrics, contrasted by isogamous sex with ameboid gametes in the pennates. These features are not

distinctly distributed among bilaterally and radially symmetric morphologies on the diatom evolutionary tree, but are instead distributed along a gradient moving from basal radially symmetric groups to more recently diverged bilaterally symmetric groups.

Simonsen (1979) was the first to discuss a phylogeny for all diatoms in the context of a taxonomic system. While Simonsen presented an evolutionary tree for diatom families, he was reluctant to deconstruct class and order designations in a manner reflecting monophyly. Most notable is the presentation of centric diatoms as distinctly non-monophyletic while maintaining the traditional taxonomic category for the group. Round *et al.* (1990) presented a taxonomic system for genera and higher-level groups. This work treated the diatoms as a division with three classes consisting of the radially symmetric taxa, the araphid pennate taxa, and raphid pennate taxa, suggesting that evolution was along this line and that the centric diatoms preceded pennates. This text remains the most recent comprehensive coverage for diatoms, but the classification system was not developed in an evolutionary context and many of the taxonomic designations are being reconsidered and modified.

A comprehensive evolutionary tree for the diatoms is currently a popular research topic (summarized in Alverson *et al.*, 2006 and Theriot *et al.*, 2009). While molecular systematics has advanced rapidly in other areas of biology, only the small subunit of the nuclearly encoded ribosomal rDNA gene (SSU) has been used for comprehensive analyses of diatom phylogeny (other genes and morphology have generally just been employed selectively at the ordinal level or below). Trees produced using the SSU molecule have uniformly obtained a grade of multiple lineages of centric diatoms generally with radial symmetry of valve elements, then a series of lineages of centrics with generally bipolar or multipolar symmetry, then a series of araphid taxa and, finally, the clade of raphe-bearing pennates. The only exceptions to this are when only a few diatoms were included in the analysis or the analytical techniques used were improperly applied (Theriot *et al.*, 2009).

Goertzen and Theriot (2003) noted the effect of taxon sampling on topologies generated in phylogenetic analyses of heterokont taxa. Diatom diversity presents a challenge, in this context, to attempts at reconstructing phylogenies for the group using molecular data. All attempts to date have sampled <0.01% of the described diatom flora (and obviously have not included extinct taxa). A very small amount of morphological data has been formally analyzed and it supports the trees derived from the SSU gene (Theriot *et al.*, 2009). Again, however, results are weakly supported and the analyses did not

include extinct taxa. What can be conservatively concluded is that no investigation has produced a comprehensive and robust phylogenetic hypothesis for the group. Nevertheless, there seems to be mounting evidence that only raphid pennates are monophyletic among the major diatom taxa. Readers should understand that, like the terms “invertebrate” or “fish”, the words “centric” and “araphid pennate” are simply terms of convenience and do not reflect independent phylogenetic groups.

While a comprehensive diatom phylogeny is desirable, it is not a requirement for utilizing diatom species for environmental reconstruction and other applications. Reliable species identification is, however, essential. Two text collections represent the standards for freshwater species identification with the light microscope. Patrick and Reimer (1966, 1975) are English guides to the North American flora. Taxa are illustrated with line drawings. Unfortunately, the work was never completed and contains only a partial coverage of pennate diatoms. Some major pennate groups (e.g. keel-bearing genera such as *Nitzschia* and the order Surirellales) and centric diatoms were not treated. Volume 1 (Patrick & Reimer, 1966) is still available for purchase, but volume 2 (Patrick & Reimer, 1975) has become difficult to acquire. Krammer and Lange-Bertalot (1986, 1988, 1991a, 1991b) are German language guides to the central European flora. Taxa are illustrated with photographs. The texts have been used to identify species from around the world and represent a reasonably comprehensive coverage of species. All four volumes are currently available for purchase.

2.3 The diatom cell

The diatom cell has the same general pattern as many plastid-bearing protists. It has an outer cell wall lined internally with a plasma membrane containing the cytoplasm and a collection of organelles. The cell wall, as previously mentioned, is distinct and is composed of silicon dioxide (SiO_2). Generally, the cell wall consists of two halves, termed valves. The two valves have slightly different sizes. The larger valve is termed the epitheca and the smaller the hypotheca. The two valves fit together to form a box. Between the valves are a series of siliceous bands or belts termed girdle bands. The valves plus all girdle bands comprise a complete cell wall that is termed a frustule (Figure 2.2).

The frustule has many openings allowing the cell’s organic component to make contact with its surrounding environment. The openings can range from simple pores to specialized structures of extremely complex micro-architecture. The cell’s plasma membrane, like other eukaryotes, is rich with proteinaceous receptors (Scherer et al., 2007). Little work has been done

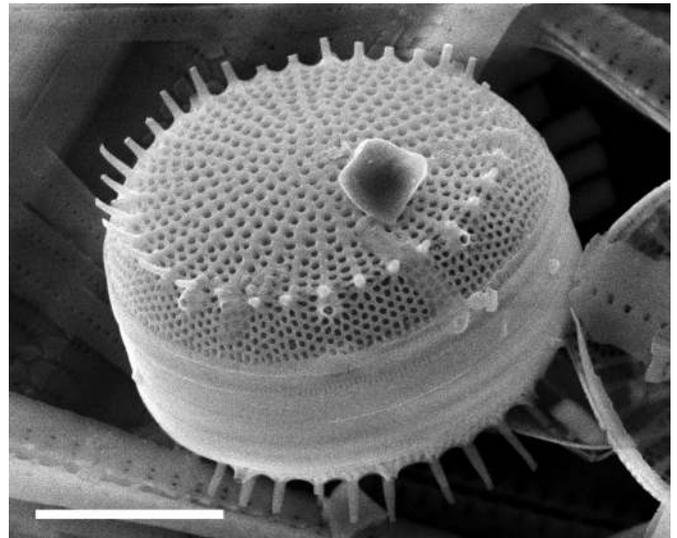


Figure 2.2 Complete *Stephanodiscus minutulus* frustule. Both valves and girdle bands comprising the “pill box” architecture are visible. Scale bar equals 5 μm .

concerning these receptors (Julius et al., 2007), but basic physical properties of extra cellular material emanating from the membrane can be altered quickly in response to environmental challenges (Higgins et al., 2003). Adhesion responses have been tied to specific plasma membrane receptors (Almqvist et al., 2004).

The cell interior (Figure 2.3) typically has a central cytoplasmic bridge containing the nucleus. This is bounded by a large vacuole, which can account for up to 70% of the total cell biovolume (Sicko-Goad et al., 1984). The vacuole is frequently associated with laterally positioned chloroplasts, which may deposit photosynthetic products in the vacuole. The silica deposition vesicles (SDVs) are closely associated with the golgi, and in some species mitochondria may be found in close proximity to both the SDVs and golgi. Little is understood about diatom physiology and most efforts focus on frustule formation or plastid function.

2.3.1 Reproduction

Diatoms, like many other unicells, reproduce primarily via asexual mitotic divisions with relatively rare instances of sexual reproduction. Cells are typically diploid with chromosome numbers varying considerably across species, having as few as 4 and as many as 68 (Kocielek & Stoermer, 1989). Some species exhibit high growth rates and may divide once per day in optimal environments (Rivkin, 1986). The frustule is involved in the divisional process to varying degrees with some taxa,

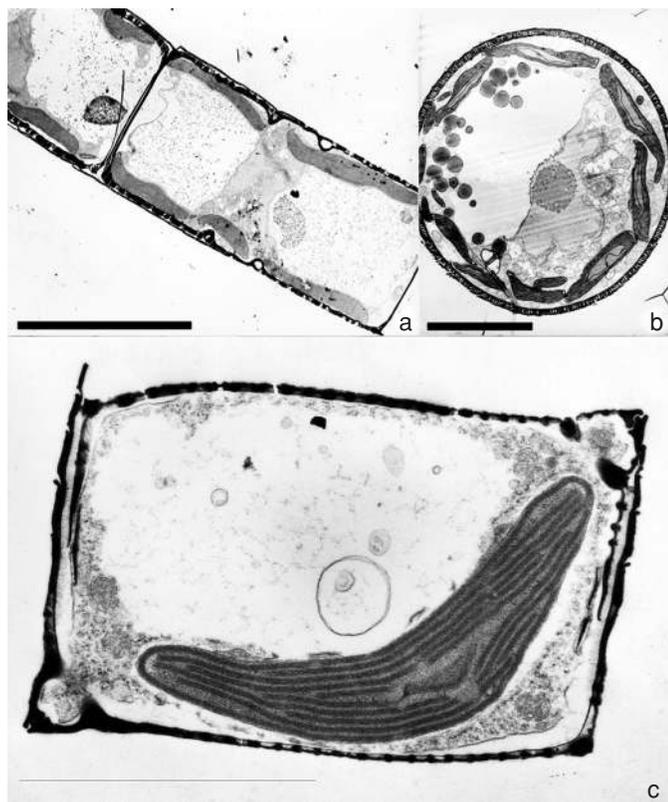


Figure 2.3 Transmission electron micrographs of *Aulacoseira ambigua* and *Nitzschia* sp. (a) *A. ambigua*, transmission electron micrograph (TEM) cross section through region of central cytoplasmic bridge. (b) *A. ambigua*, TEM longitudinal section showing fine structure of frustule and arrangements of cytoplasmic components. (c) *Nitzschia* sp. cross section: the hypothecal valve can be seen resting inside the epithecal valve. Scale bars equal 10 μm . Photographs courtesy Linda Goad.

specifically Thalassiosiraceae and Coscinodiscaceae, possessing dedicated structures for positioning and manipulating the nucleus (Schmid & Volcani, 1983; Chepurinov *et al.*, 2006). In most species, the arrangement of the epitheca and hypotheca influence mitotic products. Both the epitheca and hypotheca of the mother cells will become epitheca in the next generation, one for each daughter cell. The mitotic products result in one daughter cell equal in size to the mother cell and one daughter cell smaller than the mother cell. Repeated division results in a gradual decrease in population mean cell size over time. MacDonald (1869) and Pfitzer (1869, 1871) described this process over a century ago.

Cell size is restored via sexual reproduction, with a syngamy event producing a single maximally sized cell. Triggers for sexual reproduction are not well understood, but are generally thought to include both environmental factors and size cues

(Mann, 1993). A collection of genes that are expressed at the onset of sexual reproduction has been identified (Armbrust, 1999). The three genes identified in this collection were named Sig 1, Sig 2, and Sig 3 or sexually induced genes 1–3. Armbrust (1999) found that the genes encoded for a protein are similar in structure to an animal epidermal growth protein potentially involved in sperm and egg recognition. Sexual events are typically observed in cells approximately one third of the maximum cell size. A single cell cycle, the process of cell size reduction and restoration via sex, can take place over decades (Julius *et al.*, 1998). These factors must be accounted for when making decisions about the morphological variability of a population over time. Size ranges contained within a species category should have the largest individuals being at least three times the size of the smallest individuals. It may only be possible to observe this range in size over several seasons or numerous sediment intervals, depending upon where the population is in the size reduction/sexual cycle when observed.

Two basic forms of sexual reproduction are presented in reviews dealing with the subject (Mann, 1993; Edlund & Stoermer, 1997): oogamy in centric diatoms and isogamy in pennate diatoms. As mentioned in the introduction, these features are not distributed between radially and bilaterally symmetric diatoms in a strictly categorical fashion. Variety also exists and numerous modifications in sexual reproduction are known in the gradation ancestral to more recently diverged diatom lineages. Mann (1993) illustrates a transitional form of oogamy in an early pennate lineage in which flagella are absent in “sperm” but the gamete migrates toward a large egg to complete syngamy. All sexual events in diatoms result in auxospore production. The auxospore is a covering composed of siliceous scales or bands, which surrounds a newly formed zygote. The zygote and the auxospore expand together, ranging in shape from spheres to cigar-shaped structures. Within the auxospore a maximally sized frustule is formed, called the initial valve. Initial valves can appear slightly irregular in form as they are sometimes formed directly against the auxospore and resemble the inflated shape of this structure. These irregularities diminish as mitotic products are produced. Some taxa are capable of producing auxospores without sexual reproduction and will undergo automixis in which gametes form within a single individual and fuse with one another giving rise to an auxospore (Geitler, 1973). Parthenogenesis has also been described in some taxa (Geitler, 1982). These non-sexual mechanisms for regenerating size appear to be adaptations existing in addition to standard sexual processes producing auxospores and initial cells.