Living Inside a Glass Box – Silica in Diatoms

Franz Brümmer

1 Introduction

Silicon (Si; name originates from the Latin, silicis = flint) is present in all living organisms and is required for the production of stable structural materials. This takes place not only in single-celled organisms, e.g. diatoms, but also in lower Metazoa like sponges as well as in higher plants. Silicon is one of the most abundant elements in the earth's crust, second only to oxygen. Some geologists estimate that nearly 90% of all known minerals are silicates in combination with other elements such as aluminium (andalusite, kyanite), calcium (wollastonite), iron (fayalite), magnesium (fosterite), zinc (willemite) or zirconium (zircon), just to mention a few.

Silica, or more precisely hydrated silica, often referred to as opal, is the second most abundant mineral type formed by organisms. Only carbonate minerals are distributed in a wider range and are more abundant.

The basic chemical unit of silicates is the (SiO_4) tetrahedron-shaped anionic group. The overall charge condition leaves the oxygens with the option of bonding to another silicon ion and therefore linking one (SiO_4) tetrahedron to another and another, and so on. Regrading the numbers of tetrahedrons and the different shapes results in the silicates being divided into subclasses, not by their chemistries, but by their structures, e.g. nesosilicates (single tetrahedrons), sorosilicates (double tetrahedrons), inosilicates (single and double chains) or cyclosilicates (rings).

The soluble form of silica is a monomer, orthosilicic acid, which is a silicon atom also tetrahedrally coordinated to four hydroxyl groups with the formula $Si(OH)_4$. Only this soluble form is biologically assimilable.

Several groups of marine organisms as well as their representatives in freshwater habitats such as diatoms, "radiolarians", choanoflagellates, sponges and higher plants take up $Si(OH)_4$ from water to build their opal – amorphous hydrated silica – skeletons.

Biologisches Institut, Abteilung Zoologie, Universität Stuttgart, Pfaffenwaldring 57, 70569 Stuttgart, Germany

Progress in Molecular and Subcellular Biology, Vol. 33
W. E. G. Müller (Ed.)

2 Silica in Protozoa, Sponges and Higher Plants

2.1 Phaeodaria

In the artificial grouping of radiolarians (Radiolaria), the Phaeodaria are oceanic protists encased with porous skeletons composed of biogenic opal with organic substances and traces of Mg, Ca and Cu (Lee et al. 2000). They have been observed near the water surface of the oceans down to greater depths of up to several thousands of meters. From a quantitative viewpoint, they are only important in the Pacific equatorial belt.

2.2 Choanoflagellates

The choanoflagellates are nowadays included within the protozoa as the order Choanoflagellida (Lee et al. 2000). They are a well-defined group of free-living, colourless monads. They are uninucleated and small, seldom more than 10 µm in size. Their most distinctive feature is the uniform appearance of the protoplast with a single anterior flagellum. Choanoflagellates are ubiquitously distributed in aquatic habitats. The 50 genera and more than 100 species are included in 3 families, one with basket-like siliceous loricae (Acanthoecidae) found only in marine or brackish water (for review see Leadbetter 1991).

2.3 Silicoflagellates

The silicoflagellates (Silicoflagellata) are known in botanical literature as the Dictyochophyceae inside the alga class of Heterokontophyta (van den Hoek et al. 1995). The known 30 extant species form a clade that is most closely related to diatoms. Silicoflagellates are small to medium-sized unicellular protists and are common in marine planktonic and benthic habitats. Silicoflagellates sensu stricto form complex siliceous external skeletons (Lee et al. 2000), for example, *Dictyocha speculum*. The skeleton takes the form of a flat basket composed of hollow, but robust tubes of silica.

Among the protists, there are several other groups including species forming tests of siliceous plates, e.g. testate amoebae, or showing siliceous endoskeletal elements, for example, the heterotrophic flagellate *Hermesium adriaticum* (Lee et al. 2000).

2.4 Sponges

Two, the Demospongiae and the Hexactinellida, out of the three classes of the phylum Porifera contain siliceous spicules. Demospongiae contains more than 85% of all living sponges with about 6000 valid species already described; Hexactinellida include about 500 described species, 7% of all Porifera (Hooper and van Soest 2002). The specific spicule characteristics are used for taxonomic purposes. From one to eight types of spicules may be present within one single sponge species. The spicules stand for two major functions: they provide mechanical support to the soft part of the sponges and second, the presence of hard spicules also provides some kind of defence mechanism discouraging predators from eating sponges. The spicules are built by specialised cell types, the sclerocytes (for review see Simpson 1984). Up to now, the how and where the secretion of spicules take place – intra- or extracellularly– have been intensively discussed (Garrone et al. 1981; Simpson 1984; Uriz et al. 2000; Schönberg 2001).

Siliceous sponges proliferated well on the shelves of the Jurassic Thethys Sea, building up huge sponge reefs (Leinfelder 1993; Leinfelder et al. 1994).

2.5 Plants

Within higher plants, the Cyperaceae (e.g. horsetails, Arthrophyta) and Gramineae (grasses) contain up to 10–15% silica, which can be found in the cell walls, inside the cell lumen as well as extracellularly on the outer cuticle. The opaline silica deposits are most commonly in the form of particles called phytoliths (Perry and Keeling-Tucker 2000).

In higher animals, the role of silica is not known; in human blood, however, 138 µmol/l silicate (as SiO_2) has been reported (Wissenschaftliche Tabellen Geigy 1979).

3 Living in a Glass Box – the Diatoms

The diatom lineage contains the most beautiful, delicate eukaryotic organisms that are usually classified as algae (for reviews, see Round et al. 1990; van den Hoek et al. 1995.

Diatoms are extremely abundant in both freshwater and marine ecosystems. It is estimated that 20–25% of all organic carbon fixation on our "Blue Planet" is carried out by diatoms made possible by the chlorophyll they contain. Diatoms are thus a major food resource for all imaginable micro-organisms and animal larvae and diatoms are a major source of atmospheric oxygen.

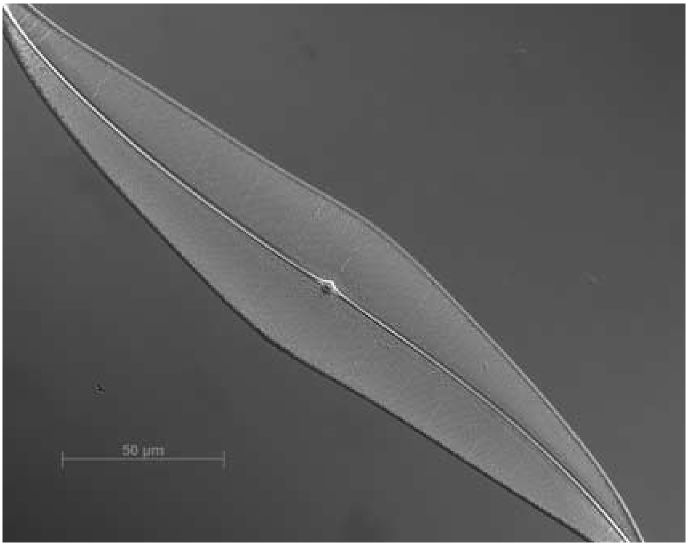

Fig. 1. *Pleursigma angulatum*, an example for a pennate diatom. In the middle of the cell, the raphe as well as the central nodule are visible

The oldest certain fossil diatoms are Lower Cretaceous (about 120 million years ago) in age. Most diatom fossils known are from Eocene and Miocene rocks, 35 and 5 million years old, respectively. The richest sources of diatom fossils are deposits of the skeleton known as diatomite or diatomaceous earth. This mineral was formed as ancient diatoms died and sank to the bottom of the oceans or lakes. Today, these large deposits of white chalky material are even mined and processed in cleansers, paints, filtering systems and abrasives. In addition, many toothpastes contain bits of fossil diatoms.

Within the Heterokontophyta, diatoms form the class Bacillariophyceae (Diatomophyceae = the diatoms). They are subdivided into two main groups (classes or orders) – the centrate diatoms (Centrobacillariophyceae; Centrales) and the pennate diatoms (Pennatibacillariophyceae; Pennales). The former are generally radially symmetrical, the latter show a typical bilateral symmetry. There are over 250 genera containing around 100,000 living diatom species.

All species of diatoms are unicellular or colonial coccoid algae. The cells secrete intricate skeletons of silica. The skeleton of a diatom, the frustule, is made of very pure silica coated with a thin layer of organic material. The presence of silica in the cell walls means that these tiny organisms live in a "glass house" or a "glass box". The skeleton is divided into two parts, one of which (the epitheca) is larger and older and overlaps the other (the hypotheca) like the lid of a box. Therefore, a more accurate description is that they live in glass Petri dishes (frustules). The top of the frustules, the epitheca, is perforated with many holes, arranged in a pattern characteristic of the species. The holes permit close contact with the environment and allow the diffusion of materials into and out of the cell.

Diatoms reproduce through cell division – one cell divides into two cells. First, the nucleus divides via mitosis and, second, two new valves are formed within the cell wall. The parental epitheca and hypotheca separate and new valves are laid down between them. The older valves fit over the newly formed hypotheca. Thus, each new cell contains an old epitheca and a new hypotheca resulting in a decrease in the mean size of a dividing population of diatoms. They become smaller and smaller with time. The decrease in size of progeny cells is called diminution and occurs until a certain size is reached. Fortunately, diatoms can also reproduce sexually in order to regain maximum size.

Diatoms are, as already mentioned, part of the drifting community, the plankton. With their rather heavy silica cell walls, planktonic diatoms are faced with the problem of how to remain in the uppermost layers ensuring enough light for photosynthesis. First, diatoms are well protected by their glass box and, therefore, able to stay intact in the turbulent mixing of the upper layers. Second, several diatoms can reduce their densities and become more buoyant by excluding heavy ions from their cell sap, thereby reducing their density to even less than the density of the surrounding sea water. Third, they can bear long spines or other protrusions which slow down their sinking rate ("living snowflakes").

On the other hand, for benthic forms the heavy frustule guarantees their remaining at the bottom, where nutrient concentrations are usually higher than in the water column. Also, in this case, the lucent frustule does not block photosynthesis and may serve for collecting and amplifying light.

Diatoms have also been recognised to produce toxins which infect shellfish and also humans along the food chain. The poisoning is called amnesic shellfish poisoning (ASP) and was first recognised in 1987 in Canada (Bates et al. 1989). To date, reports of ASP are mainly from North America and Canada, while only very low and insignificant concentrations have been detected in other parts of the world (Hallegraeff 1995; Bates et al. 1998). The symptoms include abdominal cramps, vomiting, disorientation and memory loss (amnesia). Most unexpectedly, the causative toxin is produced by a diatom (*Pseudo-nitzschia* spp.) and not by a dinoflagellate. The toxin responsible is called domoic acid which is a naturally occurring compound belonging to the kainoid class. For the mode of action, it is assumed that domoic acid is absorbed in very low rates through the gastrointestinal mucosa and transferred to the brain tissue, acting as a glutamate agonist in the brain and central nervous system, where it strongly binds to a special type of glutamate receptor (Wright and Quilliam 1995).

4 Biosilicification in Diatoms

The cell wall of diatoms consists of polymerised silicic acid, an amorphous material without any crystalline structure. Each siliceous element, like the valves or girdles, is formed within its own flattened vesicle, the so-called silica