

# Cryptic planktonic diatom challenges phytoplankton ecologists

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Planktonic diatoms are of major biogeochemical importance in the sea; their photosynthesis contributes 40% of the total annual marine production (1). Their distinguishing feature is an areolated cell wall (frustule) composed of hydrated silicon dioxide and a small amount of organic matter organized as two unequally sized, overlapping valves (thecae) similar to a Petri dish. Growth proceeds through progressive vertical separation of the thecae, culminating in an asexual division that produces two differently sized daughter cells, one of which is always smaller than the parent cell, resulting in a progressive diminution in cell size (1, 2). Cell size must be periodically restored through sexual reproduction or asexual cell enlargement (3) to prevent morbidity leading to extinction of the reproducing clone.

The coastal diatom *Skeletonema costatum sensu lato* (Fig. 1), first identified approximately 150 y ago (4), has been considered a keystone species. Its wide global distribution in coastal waters (it is absent in the high Arctic and Antarctic); its dominance of bloom cycles (abundances of  $10^7$  cells  $L^{-1}$  are common); its trophic importance to grazers; and its physiology (it is easy to culture) have been documented in thousands of publications. The genus *Skeletonema* is easy to recognize from its unique concatenation of sibling cells interconnected by a prominent marginal ring of tubular, silica rods reminiscent of a bird cage (Fig. 1). Light microscopy—traditionally used to identify species—led to the conclusion that *Skeletonema* is a depauperate genus. This conclusion was shattered recently by reports that *S. costatum s.l.* harbors at least eight cryptic species, distinguishable using a combination of electron microscopy and molecular techniques (4–6). This revelation has stunned ecologists: venerable *S. costatum s.l.* may not be the cosmopolitan, keystone species once thought. In fact, it may be absent throughout most of the global coastal ocean sites where it had been reported to dominate plankton dynamics, being replaced by *Skeletonema* taxa reported to be cryptic within the *S. costatum s.l.* morphospecies (7). The report of Hårnström et al. (8) in PNAS continues the “*Skeletonema* shock wave” to ecologists. Hårnström et al. report that dormant cells of *Skeletonema miranoi* (one of the cryptic species within morphospe-

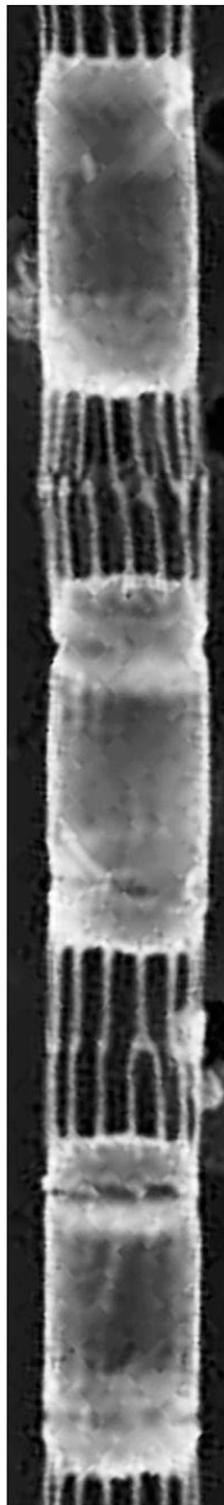


Fig. 1. Chain of cells of the diatom *Skeletonema*. Diameter  $\approx 10$   $\mu$ m.

cies *S. costatum s.l.*) buried deep (up to 22 cm) within <sup>210</sup>Pb-dated anoxic sediments in a sample cored from 30 m depth in a hypertrophic, anoxic Danish fjord retained their viability for >150 y and could be induced to resume photosynthesis and asexual, vegetative growth when re-suspended in slurry cultures exposed to light and supplied with nutrients. Also remarkable, microsatellite markers in the cultured strains suggest that the pelagic, vegetative population of *S. miranoi* in Mariager Fjord has remained genetically stable for a century or more and is reproductively isolated, failing to interbreed with *S. miranoi* strains that disperse into the fjord from contiguous Kattegat waters.

The formation of a benthic resting stage by planktonic, coastal diatoms, either as a resting spore or resting cell, is a common trait (2, 9, 10). Resting spores sink, accumulate, and can lie dormant for years in sediment seed banks, excysting under the stimulation of light and other improved growth conditions; the newly germinated vegetative cells are available as propagules to seed a new bloom (11). Unlike most coastal diatoms, *S. costatum s.l.* does not form a resting spore to escape and survive environmental conditions hostile to vegetative growth (12–15). Hårnström et al. (8) suggest that *S. miranoi* produces a physiological resting cell that explains its dormancy and capacity for physiological recovery, in agreement with the conclusion reached by McQuoid et al. (11), who germinated this cryptic species from cells buried in sediment laminae as deep as 35 cm in a Swedish fjord, which <sup>210</sup>Pb-dating suggests were deposited 55 y earlier. Morphological evidence for physiological resting cell formation in *Skeletonema* is limited to characterization of suspected resting cells as having a condensed cytoplasmic mass and deep brown chloroplasts but being otherwise morphologically similar to vegetative cells (12–15). In the diatom genus *Chaetoceros*, resting stage formation is species specific—some species form both resting spores and resting cells, others form only resting spores (9, 16, 17). That body of work also has

Author contributions: T.J.S. wrote the paper.

The author declares no conflict of interest.

See companion article on page 4252 in issue 10 of volume 108.

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demonstrated that silica and its concentrations determine whether resting spores or resting cells are produced once dormancy is induced; that differences and changes occur in protectant xanthophyll cycle pigments and in the biochemical composition of resting spores vs. vegetative cells—changes that affect metabolic activity, needs, and respiratory potential.

The germination of resting stages requires light, which mandates their water column resuspension or that light penetrates to the sediment–water boundary layer photostimulating *S. miranoi* to break out from its dormant stage. Deep burial of benthic resting stages within seed banks (8, 11) mitigates resuspension; their role in seeding blooms is expectantly superseded by propagules derived directly from pelagic vegetative cells or that have germinated from resting stages deposited at the sediment–water boundary layer. That expectation, the fact that many coastal bloom-forming diatoms do not form a resting stage, and the well-documented high growth rate, nutrient efficiency, and eurythermal and euryhaline traits of *S. costatum* s.l. challenge the presumption that the capacity of *Skeletonema* species to form resting cells is important to their blooms and succession. The remarkable dormancy reported for *S. miranoi* may be superfluous to its bloom initiation and provide some other, presently unrecognized ecophysiological need.

Phytoplankton species experience continuous change and variation in growth conditions at temporal and spatial scales that challenge their cellular growth and fitness of the evolved population. Rynearson et al. (18–20) have demonstrated the remarkable capacity of the coastal diatom *Ditylum brightwellii* to modify its population genetics and adapt

to temporal changes and spatial heterogeneity in habitat conditions by reassembling a genetically and physiologically diverse mix of distinct subpopulations. The low genetic diversity and reproductive isolation of the Mariager fjord population of *S. miranoi* and persistence of a genetically stable population for more than a century (11) contrast with the *Ditylum* model. However, heterozygote deficiency is reported to be generally characteristic of species growing under extreme selection pressures in marginal habitats (21). Anoxic Mariager Fjord is certainly ecologically marginal, beyond the norm of the habitat conditions characteristic of the distributional range of *S. miranoi* (7). Eutrophic since at least 1900, nutrient concentrations in this sill-fjord are among the highest reported for Danish coastal waters, supporting an annual primary production of  $800 \text{ g C m}^{-2} \text{ y}^{-1}$ —fivefold higher than in contiguous Kattegat waters—and when the surface layer periodically becomes anoxic and sulphidic the surface water is discolored milky-white (22). The sluggish hydrographic exchange with contiguous Kattegat waters—residence time of the inner fjord bottom water is  $\approx 3 \text{ y}$ ; the surface layer is replaced twice yearly—reduces advective loss, increasing generational exposure to the hypereutrophic conditions.

The hydrology and multidecadal chemical disturbance of Mariager fjord possibly override the habitat variations that otherwise might induce the genetic diversity such as reported for *D. brightwellii*, and foster the observed genetic erosion and reduced heterozygosity of the *S. miranoi* population. Asexual cell enlargement may also be contributory—consistent with the results—given that the *S. miranoi* population had to restore its cell size periodically to prevent morbidity leading to

extinction (3). The sustained low genetic diversity of *S. miranoi* in this chemically disturbed, slowly flushed fjord is extrapolated from the microsatellite loci in strains relict from past blooms buried in the sediment and induced to germinate. The low genetic diversity and apparent ability of the *S. miranoi* population to have out-competed other strains for >100 y suggest a genetic conservatism and competitiveness considerably beyond that usually attributed to diatoms. This raises the issue of whether other genetic strains of *S. miranoi* also bloom in Mariager Fjord but lack the capacity required to survive as physiological resting cells in the anoxic sediment seed banks.

Phytoplankton ecologists have expended great effort to explain the factors and mechanisms that determine the distribution, community assembly, blooms, and succession of species, applying macroecological and morphospecies approaches. Progress has been slow and marginal; the insights gained are more descriptive than quantitative, suggesting that the conceptual and practical approaches ecologists currently apply are not properly scaled to the ecophysiology and niche requirements of the phytoplankton phylogenetic groups and species present. The study of Härnström et al. (8), in combination with the insights into the speciation, genetic diversity, and ecophysiology being gained through molecular studies (refs. 18, 19, and 20, among others), suggests ecologists may need to redefine the aforementioned species behavior of classic interest and apply a deeper conceptual and applied level of inquiry—a microecological approach, and possibly even at the nano-level.

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