

THE CYST OF THE CALCAREOUS DINOFLAGELLATE *SCRIPPSIELLA TRIFIDA*: RESOLVING THE FOSSIL RECORD OF ITS ORGANIC WALL WITH THAT OF *ALEXANDRIUM TAMARENSE*

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ABSTRACT—*Scrippsiella trifida* Lewis, 1991 ex Head, 1996 is a nontoxic marine calciodinelloidean dinoflagellate whose resting cyst has a distinctive wall containing large, erect, trifurcate, recurving calcareous processes that separate two organic layers. We show that the organic wall layers of living *Scrippsiella trifida* cysts are resistant to acetolysis and can therefore potentially fossilize, and we report on abundant *Scrippsiella trifida* cysts from latest Pleistocene and early Holocene marine sediments off eastern Canada, representing the first confirmed fossil discovery of this species in the North Atlantic. A reappraisal of late Quaternary palynological records now shows that the organic remains of *Scrippsiella trifida* cysts have been widely misidentified as cysts of *Alexandrium tamarense* (Lebour, 1925) Balech, 1985, a goniodomacean (and hence noncalcareous) dinoflagellate and major cause of paralytic shellfish poisoning in humans. The morphology of these two cyst types is contrasted, and the modern and fossil distribution of *Scrippsiella trifida* cysts in sediments of the North Atlantic and adjacent areas is now clarified. It is apparent from this distribution that *Scrippsiella trifida* favors neritic environments characterized by cool winters and relatively warm (14°–25°C) summers. Extremely high fluxes of *S. trifida* cysts in nearshore areas off Nova Scotia and southern Greenland during deglaciation and early postglacial time (14–7 ka) have no modern analog but may signal a reduction in salinity caused by meltwater discharge. In general, the organic walls of calcareous dinoflagellate cysts are more common components of palynological assemblages than hitherto realized.

INTRODUCTION

SCRIPPSIELLA TRIFIDA Lewis, 1991 ex Head, 1996 is an extant marine, autotrophic, nontoxic dinoflagellate belonging to the subfamily Calciodinelloideae Fensome et al., 1993. Its most distinctive feature is the presence of large, trifurcate, recurving, calcareous processes within the wall of its resting cyst. These calcareous processes occur between two organic layers: the inner layer forming an ovoidal central body, and the outer layer having an irregular topography corresponding to the underlying calcareous processes. The relationship between the two organic layers and intervening calcareous processes was clearly shown by Lewis (1991) who illustrated an empty cyst treated with HCl to remove the processes (Lewis, 1991, fig. 39). However, the organic lining was not tested for resistance to acetolysis, which would have given some indication of potential geological preservability.

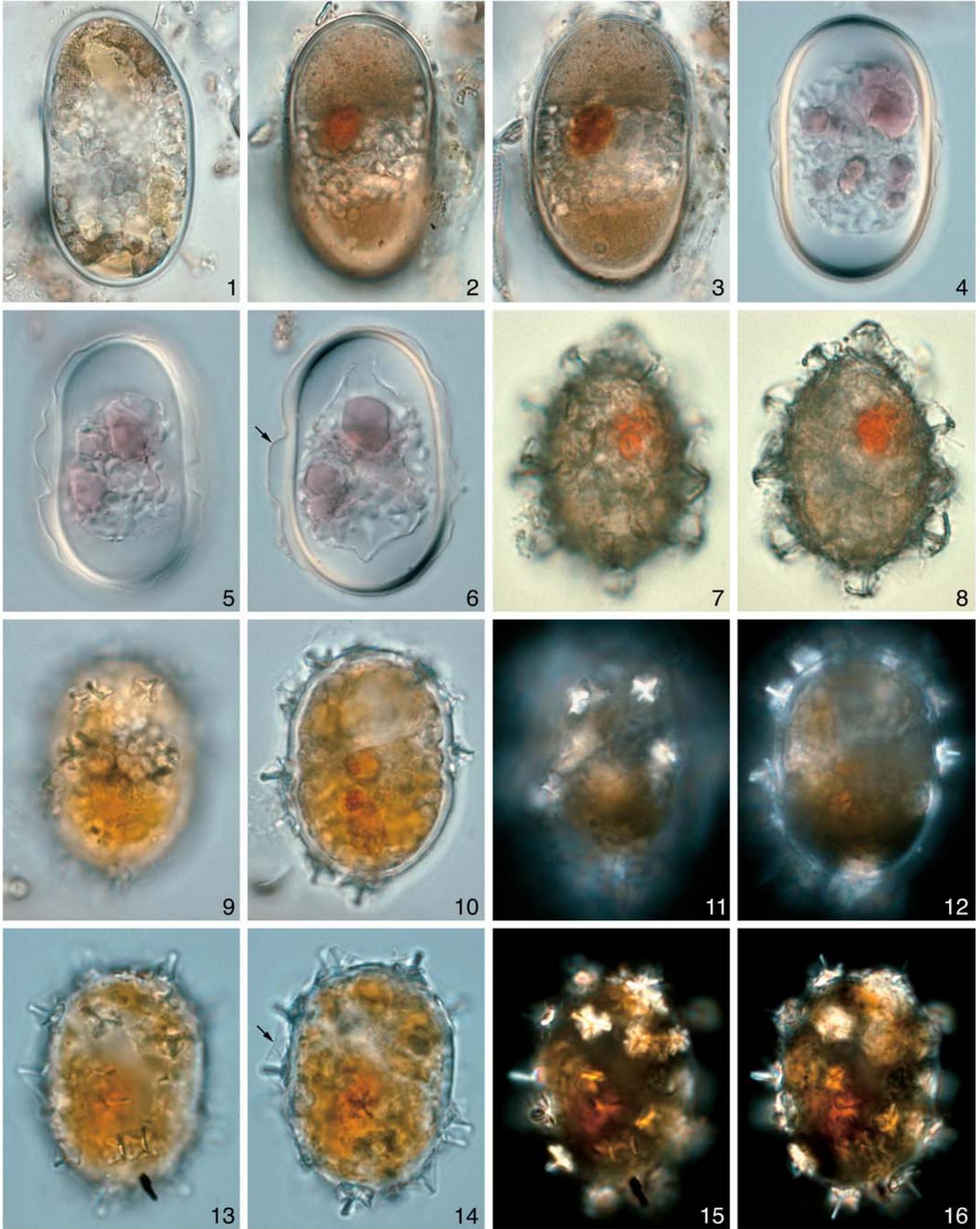
Alexandrium tamarense (Lebour, 1925) Balech, 1985 is a noncalcareous, toxic dinoflagellate belonging to the family Goniodomaceae Lindemann, 1928. It is a major cause of PSP (paralytic shellfish poisoning) in humans, and there is considerable interest in mapping the distribution of its cysts in modern sediments because these may give rise to future toxic blooms (e.g., Lewis et al., 1995). Researchers have also examined the fossil distribution of these cysts to explore possible linkages between cyst abundances and changing climatic and oceanographic regimes (Simard and de Vernal, 1998; Mudie et al., 2002). The resting cysts of *Alexandrium tamarense* are cylindrical in shape with rounded ends, are unornamented, and have a mucilaginous covering. Light microscopy (LM) reveals a two-layered wall comprising an outer layer that is resistant to acetolysis but very thin, and an inner layer that is thicker but probably made of cellulose (Anderson and Wall, 1978). The cysts therefore have some resistance to degradation but a generally low potential for geological preservation (Head, 1996, p. 1199). More detailed transmission electron microscopic (TEM) analysis shows the cyst wall to be composed of three layers (Fritz et al., 1989; Kennaway and Lewis, 2004) in which a thin inner layer and much thicker middle layer correspond to the inner layer in LM (see Systematic Paleontology).

The weak preservational potential of *Alexandrium tamarense* cysts is incompatible with frequent records given (sometimes under the synonym *Alexandrium excavatum* (Braarud, 1945) Balech and Tangen, 1985—see Systematic Paleontology) for the uppermost Pleistocene and throughout the Holocene of the North Atlantic region (e.g., Simard and de Vernal, 1998). Despite the claimed presence of these cysts, published illustrations appear not to be of *Alexandrium tamarense* but the insoluble organic walls from calcareous cysts of *Scrippsiella trifida*. This inconsistency challenges the validity of important claimed and inferred records of toxic blooms of *Alexandrium tamarense* in the geological past (e.g., Simard and de Vernal, 1998; Levac, 2001; Mudie et al., 2002).

The aims of the present paper are to test beyond doubt whether the organic remains of *Scrippsiella trifida* cysts have been mistaken for cysts of *Alexandrium tamarense* in the palynological record, and to highlight differences in morphology between cysts of the potentially toxic *Alexandrium tamarense*-type (Fig. 1.1–1.6) and those of the nontoxic *Scrippsiella trifida* (Figs. 1.7–1.16, 2, 3, 4.1–4.9). We then reassess the fossil record of *Scrippsiella trifida* cysts and the organic linings of other calcareous dinoflagellate cysts.

THE CALCIODINELLOIDEAE

The Calciodinelloideae Fensome et al., 1993 are an extant subfamily of dinoflagellates found commonly today in many tropical to temperate coastal and oceanic waters. All presently examined living species are marine and presumably autotrophic (Montresor et al., 1993, 1997). A characteristic feature of this subfamily is the production of a calcareous cyst within the life cycle. Such cysts are usually considered to be resting cysts, rather than temporary cysts, on account of their complex wall and other features such as storage products. Indeed, dormancy has been confirmed physiologically for the resting cyst of *Scrippsiella trochoidea* (von Stein, 1883) Loeblich, 1976 (Binder and Anderson, 1990). The production of resting cysts in dinoflagellates is generally regarded to result from sexual reproduction (Head, 1996), and this has been documented for some calciodinelloidean species, including



Scrippsiella hexapraeicingula Horiguchi and Chihara, 1983 (Horiguchi and Chihara, 1983), *Scrippsiella minima* Gao and Dodge, 1991 (Gao et al., 1989a, 1989b), and *Scrippsiella trochoidea* (Watanabe et al., 1982). However, it is evident for some others, such as *Leonella granifera* (Fütterer, 1978) Janofske and Karwath, 2000 and *Pernambugia tuberosa* (Kamptner, 1963) Janofske and Karwath, 2000, that the encystment process is not related to a sexual phase (Janofske and Karwath, 2000). It remains to be established whether the cysts of *L. granifera* and *P. tuberosa* are true resting cysts (a dormant stage in which normal life processes are severely reduced), but they refute any assumption that all calcareous cysts are hypnozygotes.

Extant calciodinelloideans are represented by the theca-defined genera *Ensiculifera* Balech, 1967 emend. Matsuoka et al., 1990, *Pentapharsodinium* Indelicato and Loeblich, 1986 emend. Montresor et al., 1993, and *Scrippsiella* Balech, 1959 emend. Janofske, 2000; and by numerous cyst-defined genera (Head, 1996; Meier et al., 2002; Streng et al., 2002, 2004, appendix 1), including some with long fossil records (Fensome et al., 1993). The Calciodinelloideae are presently diagnosed as “peridiniaceans in which the episomal tabulation is bipesoid and the Kofoid second anterior intercalary plate is hexa (i.e., six-sided). The cyst wall includes a calcareous layer or layers. The archeopyle is centred about the apical region” (Fensome et al., 1993, p. 133). The presence of a true transitional (t) plate in the cingulum is also regarded as an important character (Montresor et al., 1993; Fig. 5.4). The presence of a calcareous resting cyst is not regarded by all systematists as a unique diagnostic character of this subfamily, and *Pentapharsodinium dalei* Indelicato and Loeblich, 1986 is included despite having an organic-walled cyst (Indelicato and Loeblich, 1986; Montresor et al., 1993). Nonetheless, the production of a calcareous cyst is a dominant and exclusive feature of this subfamily.

The wall of modern calcareous cysts contains one or more organic layers that are resistant to HCl (Wall and Dale, 1968; Wall et al., 1970; Dale, 1983; Lewis, 1991; Montresor et al., 1993). Some extinct species of calciodinelloideans have a demonstrably fossilizable organic lining, such as *Pithonella? organica* Hultberg, 1985a from the Lower Paleocene of Sweden (Hultberg, 1985a, 1985b). Fensome et al. (1993, p. 133) assumed the organic layers to consist usually of the chemically resistant polymer dinosporin (rather than cellulose), which implies geological preservability. This assumption has since been supported by demonstration that

the organic wall layer of the calcareous cyst of *Pentapharsodinium tyrrhenicum* (Balech, 1990) Montresor et al., 1993 is resistant to acetolysis treatment and therefore not made of cellulose (Montresor et al., 1993). Palynological residues hence should frequently include the organic remains of calcareous cysts in modern and fossil marine sediments, despite the removal of calcite during acid treatment. Although there are occasional reports of actual or presumed organic linings in the fossil record (e.g., Barremian of Canada, Lentini, 1985; Albian and Cenomanian of West Africa, Verreussel, 1997; Paleocene of Scandinavia, Hultberg, 1985a, 1985b; Eocene–Miocene of the North Atlantic region, Damassa, 1998; Pliocene of Belgium, Louwey et al., 2004), such remains are seldom noted in the palynological literature and may have been overlooked.

HYPNOZYGOTIC STATUS OF *SCRIPPSIELLA TRIFIDA* CYSTS

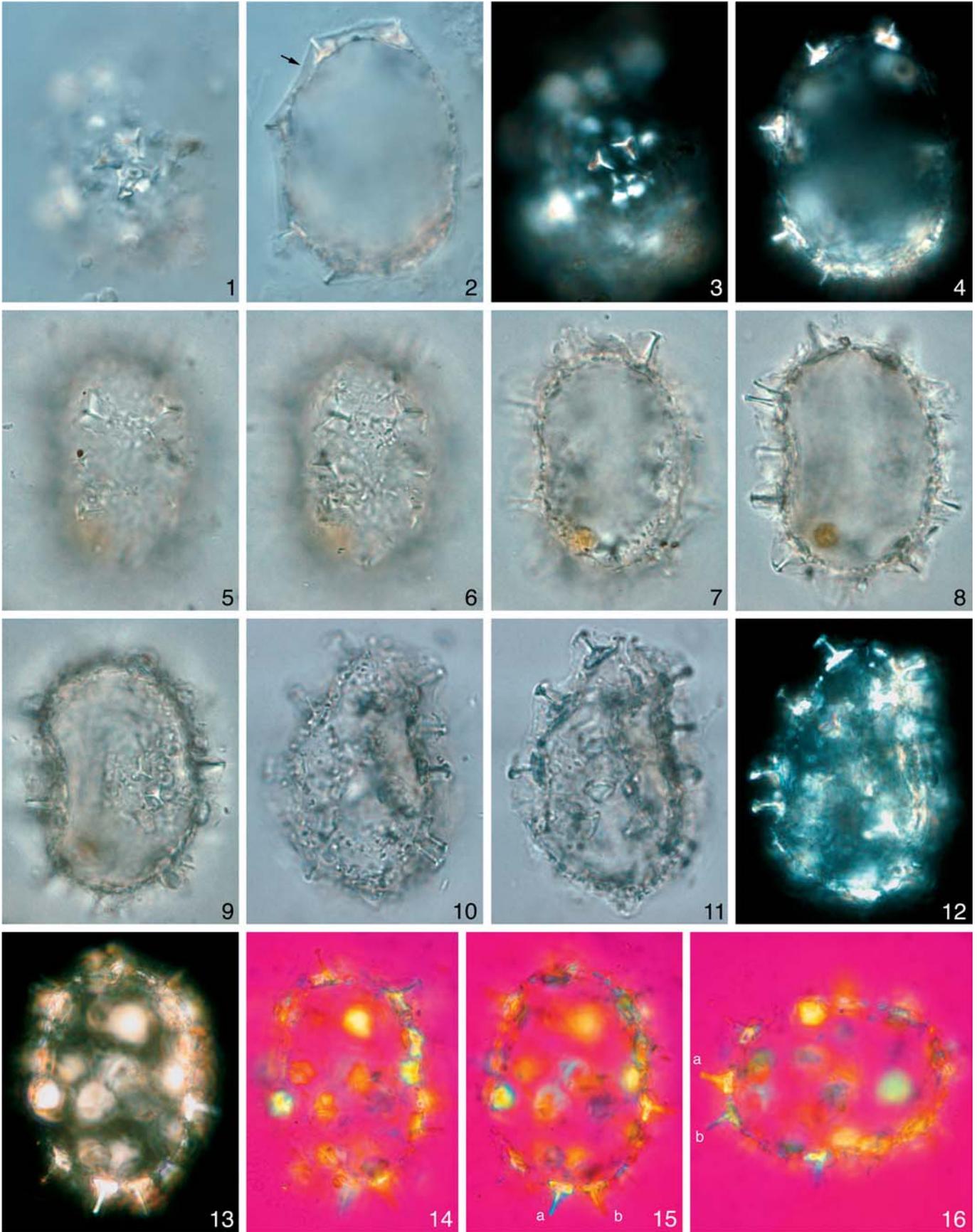
Laboratory observations show *Scrippsiella trifida* cysts to have a minimum dormancy period of about one week (R. Ahmed, personal commun.), implying that they are resting rather than temporary cysts. This fairly short duration is similar to that observed in *Scrippsiella trochoidea* (Binder and Anderson, 1987, 1990; Nuzzo and Montresor, 1999) where dormancy lasts between 2 and 5 weeks; although in *Scrippsiella rotunda* Lewis, 1991 ex Head, 1996 dormancy takes rather longer, between 17 and 24 weeks (Nuzzo and Montresor, 1999).

No direct observations of sexual reproduction, i.e., documenting the mating process through to cyst formation, are known for *S. trifida*. However, the variety of motile forms seen in culture is suggestive of gametes and planozygotes, and cell behaviors have been observed (JL, personal data) that are reminiscent of mating in other species, such as *Scrippsiella minima* (Gao et al., 1989a). Furthermore, the overall appearance of the cyst (including a red spot) is comparable with other *Scrippsiella* species where sexuality has been documented. For these reasons we consider our cysts of *S. trifida* to be hypnozygotes, i.e., resting cysts produced by sexual reproduction.

MODERN ECOLOGY OF *SCRIPPSIELLA TRIFIDA*

There are no reports of the motile stage of *Scrippsiella trifida* in the wild to our knowledge, and this is probably a reflection of its relatively indistinct morphology. However, resting cysts have been clearly documented from modern sediments in the following temperate neritic areas: west coast of Scotland (Lewis, 1991),

FIGURE 1—*Alexandrium tamarense*-type cysts from modern sediments (1–6) and cysts of *Scrippsiella trifida* Lewis, 1991 ex Head, 1996 from culture (7–16). Various magnifications. Micrographs are in bright field (1–3, 7–10, 13, 14), interference contrast (4–6), or cross-polarized light (11, 12, 15, 16). All cysts in equatorial view. An England Finder reference follows the slide number. 1–3, *Alexandrium tamarense*-type cysts from bottom sediments of Bedford Basin, Nova Scotia. Note closely appressed inner and outer wall layers. 1, Cyst in glycerine jelly mount at midfocus, showing cell contents mostly of clear starch and lipid granules, but with darkening of polar zones; Bedford Basin, Slide 1, D34/3; cyst length, 54 μm . 2, 3, Upper and midfocus of living cyst in water mount, close to hatching; note surrounding layer of mucilage with mineral inclusions; thick, hyaline, organic cyst wall; median zone of clear starch and lipid granules; polar zones of dense, brown, granular material in Brownian-like motion; and centrally located eye spot; cyst length, 53 μm . 4–6, *Alexandrium tamarense*-type cyst from bottom sediments off Orkney, North Sea. Cysts in glycerine jelly mount, after treatment with cold HCl and cold HF, 30 seconds sonification, and staining with safranin-o. Note slight separation of thin outer wall layer (indicated by an arrow in 6) and thick hyaline inner wall layer, and preservation of cell contents. 4, Midfocus, Orkney slide 4, L31/0; maximum length, 46 μm . 5, 6, Upper and midfoci, Orkney slide 4, L31/1; maximum length, 45 μm . 7, 8, Living cyst of *Scrippsiella trifida* from culture; in water mount. Note trifurcate, recurved, calcareous processes that separate the two organic wall layers, an ovoidal inner layer and an undulating outer layer; and centrally positioned red body. 7, Upper focus; 8, midfocus; central body length, 36 μm ; length including outer wall layer, 46 μm . 9–16, Living cysts of *Scrippsiella trifida* from culture; mounted in glycerine jelly. Note calcareous processes with blunt or weakly trifurcate tips, arising from discrete platforms often with ridges radiating from process base and forming a Y-shaped pattern; calcified parts of the cysts show as brightly illuminated areas (owing to strong birefringence) in cross-polarized light. 9–12, Upper and midfocus, respectively, in bright field and cross-polarized light; central body length, 33 μm ; length including outer wall layer, 38 μm ; slide 3, W33/4. 13–16, Upper and midfocus, respectively, in bright field and cross-polarized light; with arrow in 14 indicating outer organic wall layer; central body length, 32 μm ; length including outer wall layer, 36 μm ; slide 3, T25/1.



Scapa Flow, Orkney, Scotland (Joyce, 2004), Danish coastal waters of the Kattegat (Ellegaard et al., 1994, fig. 3), the German Bight (Nehring, 1994, fig. 3; 1997) and Wadden Sea areas (Hoppenrath et al., 2004) of the southern North Sea, and the more open waters of the south-central North Sea (Nehring, 1995). The cysts have not been reported to occur abundantly in these sediments (e.g., up to 18 cysts cm^{-3} in Nehring, 1994; up to 50 cysts ml^{-1} and up to 6.7% of the total cyst assemblage in Joyce, 2004), which suggests that the optimal areas for their growth have not yet been investigated by modern plankton workers.

Godhe et al. (2001) recorded *S. trifida* cysts in sediment trap samples from the western coast of Sweden, where the mean number of cysts found in traps was 1,885 cysts $\text{m}^{-2} \text{day}^{-1}$. This number is some 3% of the abundance of the most commonly occurring species found in the traps, again implying that *S. trifida* is not a common component of the dinoflagellate population here. Hamer et al. (2001) have recorded cysts of *S. trifida* in the ballast water of two ships entering English and Welsh ports. Rubino et al. (2000) recorded *Scrippsiella* cf. *trifida* cysts in modern sediments from the North Adriatic. However, it is not clear from their figure (Rubino et al., 2000, fig. 3e) that this is a true record of this species. Moscatello et al. (2004) listed but did not illustrate *Scrippsiella trifida* cysts in modern sediments off the Salento Peninsula, southeast Italy.

The above limited records indicate a neritic and broadly temperate distribution for *Scrippsiella trifida*. In particular, the record from Danish coastal waters of the Kattegat (Ellegaard et al., 1994), where salinities are about 20, shows that reduced salinity is tolerated. However, this species is not known from the Baltic Sea proper where salinities are about 10 and less.

CYSTS OF *ALEXANDRIUM TAMARENSE*-TYPE

The goniodomacean genus *Alexandrium* Halim, 1960 contains at least 29 extant species (Balech, 1995), of which about 16 are known to produce resting cysts (Head, 1996; Yoshida et al., 2003). Of these, only three species produce cysts morphologically indistinguishable from *Alexandrium tamarense*. These species are *Alexandrium catenella* (Whedon and Kofoid, 1936) Balech, 1985 (Fukuyo, 1980, 1985; Yoshimatsu, 1981; Hallegraeff and Bolch, 1992; Fukuyo et al., 2003), *Alexandrium acatenella* (Whedon and Kofoid, 1936) Balech, 1985 (Yoshida et al., 2003), and *Alexandrium fundyense* Balech, 1985 (Anderson and Wall, 1978; White and Lewis, 1982, both as *Gonyaulax excavata* [Braarud, 1945] Balech, 1971). Of these, *Alexandrium catenella* is probably conspecific with, and hence junior to, *Alexandrium tamarense* based on molecular evidence (Scholin, 1998), although it is usually treated as a separate morphospecies (e.g., Steidinger and Tangen, 1996). *Alexandrium fundyense* likewise is probably also conspecific with, and junior to, *Alexandrium tamarense* judging from mating experiments (Anderson et al., 1994). Indeed, molecular

studies of *Alexandrium catenella*, *A. tamarense*, and *A. fundyense* all cast doubt on whether these are truly distinct species, and are accordingly known as the *A. tamarense* species complex (John et al., 2003). *Alexandrium acatenella* is very similar to *Alexandrium tamarense* in thecal morphology and was assigned by Balech (1995) to the same species group (his “*tamarense* group”) within the subgenus *Alexandrium*. We provisionally accept all these closely related taxa as individual morphospecies within the *A. tamarense* species complex, but also note that the identical appearance of their cysts reveals the potential of cyst morphology to elucidate phylogenetic relationships within the genus *Alexandrium*. An illustration of this potential is provided by *Alexandrium tamiyavanichii* Balech, 1994, which is shown by molecular data to be the most closely related species to the *A. tamarense* species complex (Usup et al., 2002; John et al., 2003). Despite this close relationship, *A. tamiyavanichii* does not have a cyst of the *Alexandrium tamarense*-type (Nagai et al., 2003), reflecting its evolutionary divergence.

Our cysts are referred to as *Alexandrium tamarense*-type because they are morphologically identical to cysts of *Alexandrium tamarense*, although we did not incubate them to confirm their identity.

MODERN ECOLOGY OF *ALEXANDRIUM TAMARENSE* SPECIES COMPLEX

The *Alexandrium tamarense* species complex (*A. tamarense*, *A. catenella*, *A. fundyense*, and *A. acatenella*) has a widespread distribution in neritic waters today, ranging from the subarctic through temperate to tropical zones (Steidinger and Tangen, 1996; Anderson, 1998). Scholin (1998) showed isolates identified as *A. tamarense* and *A. catenella* having broad geographic ranges (subarctic to tropical) with *A. catenella* favoring Pacific margins and *A. tamarense* favoring Atlantic margins. Only *A. fundyense* appears to have a restricted distribution, being present solely in the Bay of Fundy on the southeastern coast of Canada and the northeastern coast of the United States where it extends as far south as New York State (Balech, 1995). *Alexandrium tamarense* has not been found in the Bay of Fundy, which is the type locality of *A. fundyense*. Balech (1995) described *A. acatenella* as also having a widespread Pacific distribution. As mentioned previously, the distinction between these strains (as determined by molecular markers) is more closely modeled on geography than on morphology and in this respect their autecology is also influenced. It is evident that the physiology of different geographical isolates differs markedly (Gallagher, 1998; Lewis, 2002) and clearly fits regional strains to their particular environments. The *Alexandrium tamarense* species complex is clearly a highly adaptable and adapted suite of organisms that “have proven to be remarkably resilient and capable of colonising a spectrum of habitats and hydrographic regimes” (Anderson, 1998, p. 44).

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FIGURE 2—Cysts of *Scrippsiella trifida* from culture (1–4) and from the uppermost Pleistocene of core MD95–2033 (depth 455–456 cm), off Newfoundland, Canada (5–16); in glycerine jelly mount. Note calcareous processes separating inner from outer organic wall layers, and arising from discrete platforms. Various magnifications. Micrographs are in interference contrast (1, 2), bright field (5–11), or cross-polarized light (3, 4, 12–16) with gypsum plate inserted (14–16). All cysts in equatorial view. An England Finder reference follows the slide number. 1–4, Empty cyst with blunt processes, at upper and midfocus respectively, in bright field (1, 2) with outer organic wall layer indicated by arrow; and cross-polarized light (3, 4) showing highly birefringent calcareous processes; central body length, 32 μm ; length including outer wall layer, 36 μm ; slide 3, T38/0. 5–9, Upper through successively lower foci; note yellow “accumulation body” at lower end of cyst, possibly representing the red body in living cysts; central body length, 35 μm ; length including outer wall layer, 44 μm ; slide 1, W27/1. 10–12, Specimen with calcareous processes bearing stunted distal trifurcations; 10, upper focus and 11, 12, midfocus, with 12 showing highly birefringent calcareous processes; central body length, 37 μm ; length including outer wall layer, 42 μm ; slide 1, O13/4. 13–16, Specimen bearing calcareous processes that are either blunt or bearing stunted distal trifurcations, at upper (14) and midfocus (13, 15, 16); with the gypsum plate inserted in 14–16 to show processes as blue in the first and third quadrants and yellow in second and fourth quadrants, revealing their tangential crystallographic orientation. The specimen in 16 is rotated 90° clockwise relative to 15, with corresponding processes labeled “a” and “b” to show their change in color upon rotation; central body length, 37 μm ; slide 1, J41/0.

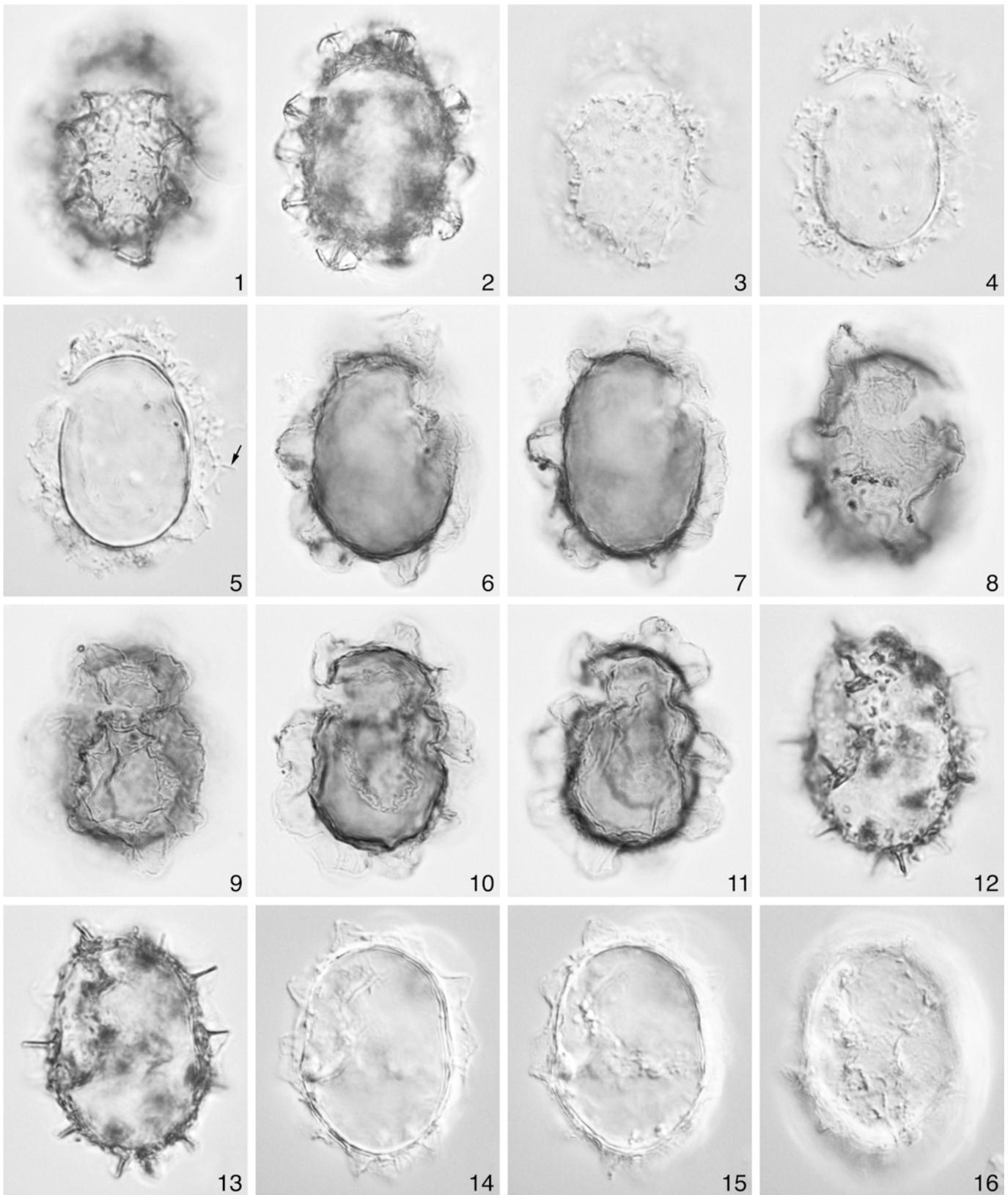


FIGURE 3—Cysts of *Scripsiella trifida* from culture (1–11), and from the uppermost Pleistocene of core MD95-2033 (depth 455–456 cm), off Newfoundland, Canada (12–16). Various magnifications. Micrographs are bright field (1, 2, 6–13) or interference contrast (3–5, 14–16) images. All cysts in equatorial view. An England Finder reference follows the slide number. 1, 2, Empty cyst in water mount showing archeopyle and calcareous processes; 1, upper, and 2, midfocus; central body length, 35 μm ; length including outer wall layer, 43 μm . 3–5, Same specimen as in

MATERIAL AND METHODS

Live cysts of *Alexandrium tamarense*-type were obtained from surface sediments of Bedford Basin, Nova Scotia, Canada. They are probably indeed those of *Alexandrium tamarense* on the basis of their internal and external physical characteristics and because this species is known to bloom along the southern coast of Nova Scotia (e.g., Cembella et al., 2000, 2002). However, they might belong to a closely related morphospecies of *Alexandrium* (see above, and Systematic Paleontology). The sediment was wet sieved at 20 μm to concentrate the *Alexandrium tamarense*-type cysts, which were photographed (Fig. 1.2, 1.3) while still alive under a coverslip using a $\times 100$ oil objective. Additional sediment was mounted onto microscope slides using glycerine jelly for a more permanent record (Fig. 1.1).

Surface sediment off Orkney, North Sea, containing *Alexandrium tamarense*-type cysts, was processed using cold HCl and cold HF. The residue was sonicated for 30 seconds, stained using safranin-o, sieved at 20 μm , and mounted onto microscope slides using glycerine jelly (Fig. 1.4–1.6). *Alexandrium tamarense* is known to occur in the waters off Orkney (Medlin et al., 1998).

Live cysts of *Scrippsiella trifida* were obtained from a unialgal culture (UW426) maintained at the University of Westminster. The culture originates from a single motile cell picked from the incubation of sediment slurry collected from Loch Creran, western Scotland, the type locality for this species. The live cysts were sonicated for 30 seconds, transferred in a drop of water onto a microscope slide, and covered with a coverslip. The live cysts were observed using a $\times 100$ oil objective, and photographed (Fig. 1.7, 1.8).

An additional microscope slide mount of *Scrippsiella trifida* cysts was made, and the empty wall of a hatched cyst located and photographed under a $\times 100$ oil objective (Fig. 3.1, 3.2). Then, while observing this cyst under a $\times 10$ objective, a drop of 7% HCl was placed along one side of the coverslip and drawn under it by applying a tissue from the opposite side. The HCl front moved across the field of view, and the calcite processes within the cyst wall dissolved in a few seconds. The cyst was kept in view by simultaneously moving the microscope stage. When the cyst had stopped moving, a $\times 100$ oil objective was used to observe and photograph the organic remains of the cyst wall (Fig. 3.3–3.5). This procedure was repeated for several other empty cysts in separate mounts.

A sample of live *Scrippsiella trifida* cysts was also subjected to standard acetolysis treatment which removes cellulose and polysaccharides by acetylation. This involved repeated washes in glacial acetic acid, immersion in a mixture of nine parts acetic anhydride and one part concentrated sulphuric acid for three minutes, followed by washes in glacial acetic acid and then water. The residue was stained with aqueous safranin-o, and mounted on microscope slides using glycerine jelly (Fig. 3.6–3.11).

Finally, live cysts from a separate population of the *Scrippsiella trifida* culture (UW426) were sonicated for 30 seconds, and

mounted with glycerine jelly on microscope slides for detailed morphological analysis (Figs. 1.9–1.16, 2.1–2.4).

Raw sediment was analyzed from core MD95-2033, located on the Laurentian Fan, continental slope south of Newfoundland, Canada, at 44°39.87'N, 55°37.21'W at a water depth of 1,412 m (Bertini et al., 1998; de Vernal et al., 2000; Figs. 7–9, Table 1). The core has a total length of 29.68 m. The sample is from the interval 455–456 cm and is about 11,000 calibrated yr BP (Pleistocene–Holocene transition), based on an unpublished age model using several ^{14}C measurements (see paleoceanographic database at www.geotop.uqam.ca). The sediment sample (a few grams) was disaggregated by warming for one hour in a 4.4% sodium pyrophosphate solution, sieved at 80 μm and 20 μm to concentrate any calcareous cysts, and rinsed in distilled water. Residue suspended in distilled water was placed onto a microscope slide and searched. Once *Scrippsiella trifida* cysts were found, each was transferred by micropipette onto a separate microscope slide and covered with a coverslip. Each cyst was photographed under a $\times 100$ oil objective (Fig. 3.12, 3.13), treated with HCl using the method described above, and the organic remains of the cyst wall again photographed using a $\times 100$ oil objective (Fig. 3.14–3.16). This procedure was repeated for several individually picked cysts in separate mounts. Additional quantities of residue were strewn-mounted with glycerine jelly on a microscope slide for more permanent study (Fig. 2.5–2.12). The crystallographic orientation of calcareous processes on several of these specimens was determined following the method of Janofske (1996) using a petrographic microscope equipped with a gypsum plate (Fig. 2.13–2.16). Finally, microscope slides of palynologically treated sediment from core MD95-2033 (Rochon et al., 1999) were re-examined for organic remains of *Scrippsiella trifida* cysts (Fig. 4.1–4.6). In order to explore more deeply the fossil record of *Scrippsiella trifida* cysts, a specimen from the Upper Pliocene of Belgium (as “cf. *Alexandrium tamarense*-type cysts of Rochon et al., 1999” in Louwye et al., 2004, fig. 10a, 10b) was reexamined (Fig. 4.7, 4.8). For completeness, SEM images of cysts of *Scrippsiella* species (from Lewis, 1991) are shown (Fig. 4.9–4.11).

All LM observations were made in Cambridge using a Leica DMR microscope and photographed using a Leica DC300 digital camera with zoom attachment, with the exception of Figure 2.13–2.16, which was taken using a Zeiss petrographic microscope at the Natural History Museum, London.

RESULTS

Examination of living cysts of *Scrippsiella trifida* confirms in detail the description given by Lewis (1991), including an ovoidal shape, a centrally positioned red body, and the presence of trifurcate, recurved, calcareous processes that separate inner from outer organic wall layers (Fig. 1.7, 1.8). Variation not previously highlighted includes the development of processes that are blunt rather than trifurcate. This morphotype dominated one population of living cysts of *Scrippsiella trifida* examined (Figs. 1.9–1.16,

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1, 2, but after introduction of 7% HCl; 3, upper surface, 4, slightly lower focus, and 5, midfocus. Note that HCl has removed the calcareous wall layer (including processes) while leaving the inner and outer organic wall layers unaffected. Sinuous filaments projecting from the cyst surface before and after acid treatment, e.g., as indicated by an arrow in 5, are spirochaete bacteria. 6–11, Cysts from culture after acetolysis, demonstrating the resistance of inner and outer organic wall layers; mounted in glycerine jelly. Note disappearance of spirochaete bacteria. 6, Midfocus, 7, slightly lower focus, 8, lower surface; central body length, 40 μm ; length including outer wall layer, 50 μm ; slide 3, J20/0. 9, Upper surface, 10, midfocus, 11, lower surface; central body length, 36 μm ; length including outer wall layer, 45 μm ; slide 3, J26/0. 12, 13, Fossil cyst in water mount showing calcareous processes and organic wall layers; 12, upper, and 13, midfocus; central body length, 35 μm ; length including outer wall layer, 40 μm . 14–16, Same specimen as in 12 and 13, but after introduction of 7% HCl. 14, Upper surface, 15, midfocus, and 16, lower surface; central body length, 33 μm ; length including outer wall layer, 42 μm . Note removal of the calcareous wall layer by HCl, and similarity of the remaining organic wall to the modern specimen (3–5) after similar treatment.

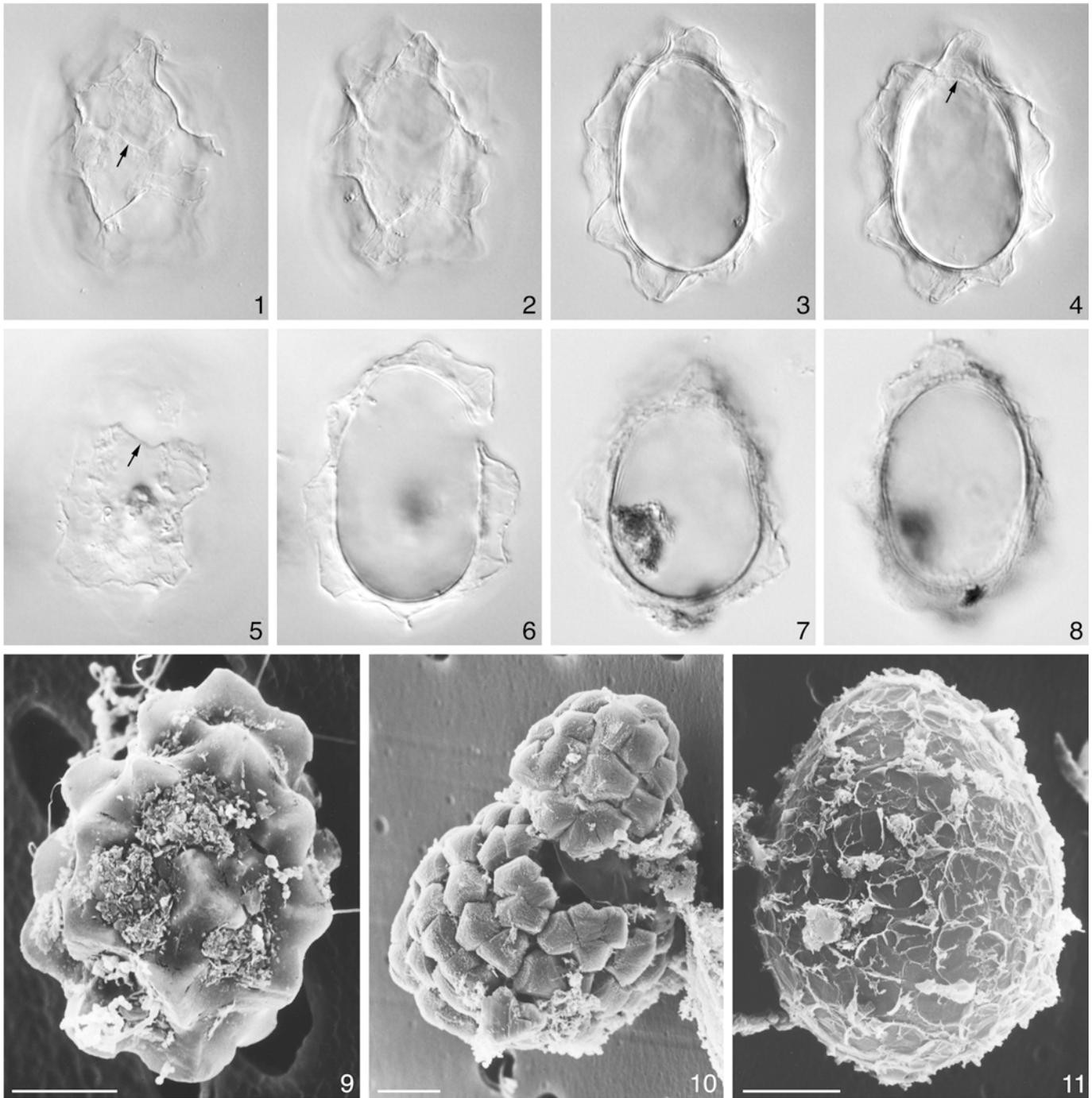


FIGURE 4—Modern and fossil cysts of the genus *Scrippsiella* Balech, 1959 emend. Janofske, 2000. Various magnifications. Scale bars = 10 μm . Micrographs are interference contrast (1–8) or SEM (9–11) images. All cysts in equatorial view. An England Finder reference follows the slide number. 1–6, Fossil Holocene cysts of *Scrippsiella trifida* from core MD95-2033, off Newfoundland, Canada. 1–4, Palynologically processed specimen originally illustrated as “cf. *Alexandrium tamarense*-type cysts” in Rochon et al. (1999, pl. 9, figs. 11–14), showing 1, upper surface, 2, slightly lower focus, 3, midfocus, and 4, slightly lower focus; with zigzag archeopyle suture indicated by an arrow in 1 and 4; central body length, 37 μm ; length including outer wall layer, 47 μm ; depth 205–206 cm, slide 7, O13/0. 5, 6, Palynologically processed specimen also showing zigzag archeopyle suture, indicated by an arrow in 5, and containing an accumulation body in the central part of the cyst; 5, upper surface, 6, midfocus; central body length, 34 μm ; length including outer wall layer, 42 μm ; depth 205–206 cm, slide 7, M24/0. 7, 8, Palynologically processed cyst of *Scrippsiella trifida* from the Upper Pliocene of Belgium, originally illustrated as “cf. *Alexandrium tamarense*-type cysts of Rochon et al., 1999” in Louwye et al. (2004, fig. 10a, b); 7, upper focus, 8, lower focus; central body length, 31 μm ; length including outer wall layer, 39 μm ; sample VBD 3.11 p2, X33/0. 9, Holotype of *Scrippsiella trifida*, a cyst from modern sediment of Loch Creran, Scotland, showing undulating outer organic wall layer supported by underlying calcareous processes (specimen also illustrated in Lewis, 1991, figs. 40, 41). 10, Modern hatched cyst of *Scrippsiella crystallina* Lewis, 1991 ex Head, 1996 from sediments of the west coast of Scotland showing blocky outer crystals, archeopyle, and inner wall layer that is resistant to HCl treatment and probably geologically preservable (from Lewis, 1991, fig. 22). 11, Modern cyst of *Scrippsiella crystallina* from sediments of the west coast of Scotland after HCl treatment, revealing, beneath the calcareous outer crystals, an organic wall layer with characteristic ornament (same specimen, at different angle of tilt, also illustrated in Lewis, 1991, fig. 19).

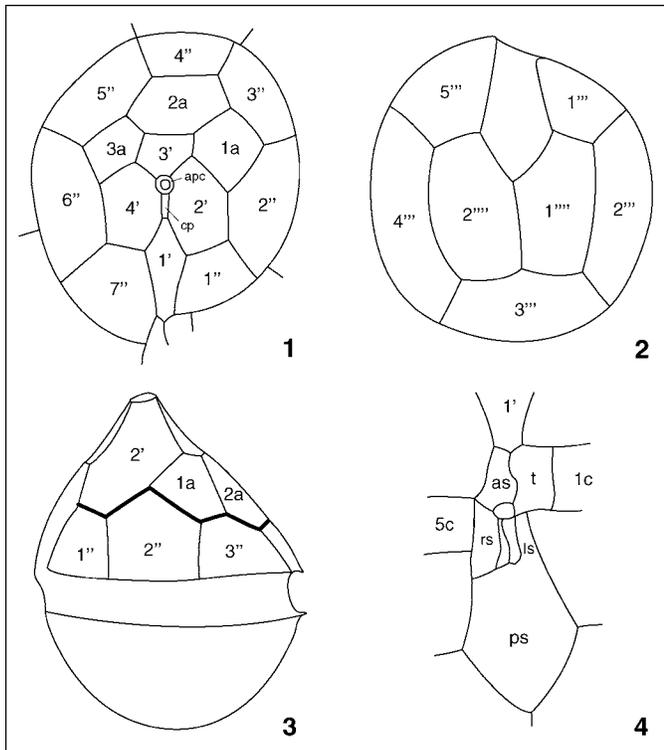


FIGURE 5—Thecal tabulation of *Scrippsiella trifida* using Kofoidian notation. 1, Epitheca; 2, hypotheca; 3, left lateral view of theca with heavy lines indicating interpreted position of principal archeopyle suture; 4, ventral view showing details of sulcus. apc = apical pore complex, cp = canal plate, as = anterior sulcal plate, ls = left sulcal plate, rs = right sulcal plate, ps = posterior sulcal plate, t = transitional cingular plate. Adapted from Lewis (1991, figs. 9c, 48a–c).

2.1–2.4; see Systematic Paleontology). Treatment of empty cysts with HCl to dissolve the calcareous processes (also performed by Lewis, 1991) revealed a distinctive morphology consisting of a smooth inner layer and an undulating outer layer (Fig. 3.1–3.5). Acetolysis of living cysts of *Scrippsiella trifida* removed all remains of the cysts except for these two distinctive organic wall layers, which were unaltered except for some slight distortion (Fig. 3.6–3.11).

Analysis of raw sediment from the interval 455–456 cm (uppermost Pleistocene) in core MD95-2033, located on the continental slope south of Newfoundland, revealed the presence of *Scrippsiella trifida* cysts. These cysts were fossils and lacked protoplasm. However, their ovoidal shape, and the presence of calcareous processes (mostly blunt but occasionally trifurcate and recurved) and inner and outer organic wall layers, confirmed their identification (Figs. 2.5–2.12, 3.12, 3.13). These cysts, upon dissolution with dilute HCl (Fig. 3.14–3.16), revealed an organic-layered morphology identical to that of *Scrippsiella trifida* cysts from culture (above), and of fossil cysts from palynological preparations (e.g., *Alexandrium excavatum*-type cysts in Simard and de Vernal, 1998; and “cf. *Alexandrium tamarense*-type cysts” in Rochon et al., 1999; Fig. 4.1–4.8).

Living cysts of *Alexandrium tamarense*-type were examined for comparison with those of *Scrippsiella trifida*. The cysts are noncalcareous and identical to the descriptions and illustrations of live *Alexandrium tamarense* cysts in Dale (1977, as *Gonyaulax excavata*) and Anderson and Wall (1978, as *Gonyaulax tamarensis* Lebour, 1925). The cylindrical shape with rounded

ends, and the wall comprising a thick (ca. 1.0–1.5 μm), hyaline inner layer and much thinner (<0.2 μm) outer layer often covered with mucilage, are characteristic features. The inner and outer wall layers are closely appressed in this live material (Fig. 1.1–1.3).

Cysts of *Alexandrium tamarense*-type from surface sediment samples off Orkney, North Sea, were observed after they had received gentle palynological treatment using cold HCl, cold HF, and 30 seconds of sonication. Cell contents were still present, but the mucilage was generally no longer visible, and the thin outer wall layer had, in most cases, separated from the thick, hyaline, inner wall layer (Fig. 1.4–1.6).

DISCUSSION AND CONCLUSIONS

Our results show that the two organic wall layers of calcareous cysts of *Scrippsiella trifida* are resistant to acetolysis and so are not made of cellulose. They are presumably made of dinosporin. This reveals the potential of the organic wall to enter the geological record. Calcareous process terminations vary from recurved trifurcate to blunt, with specimens bearing blunt processes dominating one of the two populations of cultured cysts examined. This and other observations (see Systematic Paleontology) extend the known morphological variability of this species, although it was noted by Lewis (1991) that cysts produced in culture have a generally less robust morphology than those in the wild.

Confirmation of fossil Scrippsiella trifida cysts.—The discovery of abundant calcareous cysts of *Scrippsiella trifida* in the latest Pleistocene of core MD95-2033 represents the first confirmed fossil record for this species in the North Atlantic region. Previous palynological investigations of the latest Pleistocene and Holocene of core MD95-2033 have recorded common “cf. *Alexandrium tamarense*-type cysts” (Rochon et al., 1999, pl. 9, figs. 11–14; MJH, personal observation; and as *Alexandrium excavatum* cysts in Bertini et al., 1998). Rochon et al. (1999, p. 42) had acknowledged that these cysts differ in detail from those of *Alexandrium tamarense*, and assigned them only questionably to the genus *Alexandrium*. Our study now shows that these are not cysts of an *Alexandrium* species, but the organic remains of *Scrippsiella trifida* cysts. This obviates the need to explain how *Alexandrium tamarense*-type cysts could have persisted widely in the geological record when they are known to have poor fossilization potential. A single *Scrippsiella trifida* cyst listed but not illustrated from a Holocene core in the eastern Mediterranean (Zonneveld et al., 2001) is the only previous fossil record of this species.

Modern distribution of organic remains of Scrippsiella trifida cysts.—The distribution of the organic remains of *S. trifida* cysts in modern sediments can also be clarified. We are now confident that this cyst type had been misidentified as *Alexandrium tamarense*-type cysts in the earlier literature, for example: de Vernal et al. (1994, 1997, 2001), Simard and de Vernal (1998), Rochon et al. (1999), Marret and Zonneveld (2003, in part); and, possibly, Mudie and Short (1985). Its modern distribution is documented from the palynological investigation of surface sediment samples collected at middle to high latitudes of the Northern Hemisphere to develop a reference dinoflagellate cyst database (de Vernal et al., 2001, 2005; Fig. 6). The organic remains of *S. trifida* cysts are scattered with a maximum of a few specimens per slide, which usually represent less than 1.5% of the total organic-walled dinoflagellate cyst assemblage. The distribution pattern nevertheless seems consistent, with frequent occurrences in neritic zones of circum-Atlantic areas, especially along the northeastern USA and southeastern Canadian coasts. The overall distribution of *S. trifida* cysts around Iceland and in the northeast North Atlantic up to the Norwegian and Barents Sea is compatible with a dispersal pattern across the ocean via the Gulf Stream and the North Atlantic Drift. The areas of *S. trifida* occurrence in sediment correspond to a

TABLE 1—Cores used to document the Upper Pleistocene and Holocene occurrence of cysts of *Scrippsiella trifida* Lewis, 1991 ex Head, 1996 by palynological analysis. The cores indicated in bold are characterized by a well-constrained chronostratigraphy and are used to reveal the occurrence of *S. trifida* through time (see Figs. 8, 9). In the occurrence column, x = none, o = occasional (less than 1%, and occurring in only a few samples). In the other cores, the occurrence is usually significant and the maximum percentages are indicated. Maximum occurrences for cores HU87-003-004 and HU95-030-024 are estimated from Mudie et al. (2002).

Cores	Latitude	Longitude	Water depth (in meters)	Occurrence of <i>S. trifida</i> cysts
HU91-039-008	77°16.00'N	74°19.90'W	663	x
HU91-039-012	76°48.30'N	71°51.50'W	823	x
HU95-030-024	43°46.00'N	63°43.00'W	256	21.5
DA89-007-016	49°42.80'N	61°56.91'W	258	9.4
DA89-007-021	49°31.28'N	60°48.13'W	281	5.6
DA89-007-036	50°06.92'N	58°43.59'W	300	45
DA89-007-111P	47°31.00'N	59°53.06'W	503	17.6
DA89-007-111T	47°31.00'N	59°53.06'W	503	o
HU76-029-033	71°20.00'N	64°16.00'W	2,207	x
HU77-027-013	68°26.90'N	63°31.73'W	1,889	x
HU84-030-021	58°22.06'N	57°30.42'W	2,853	o
HU85-027-016	70°30.78'N	64°31.24'W	2,091	x
HU87-028-069	55°28.60'N	77°57.80'W	165	x
HU87-003-004	43°53.10'N	62°56.52'W	247	32.9
HU87-033-007	57°25.20'N	65°24.01'W	823	x
HU87-033-008	62°38.91'N	53°53.07'W	2,424	x
HU87-033-009	62°30.99'N	59°26.82'W	1,437	x
HU90-013-012P	58°55.35'N	47°07.01'W	2,830	12.5
HU90-013-013P	58°12.59'N	48°22.40'W	3,379	4.5
HU90-031-019P	49°17.44'N	63°59.57'W	322	3.8
HU90-031-023P	47°55.43'N	65°12.31'W	73	2.6
HU90-031-044P	44°39.41'N	55°37.13'W	1,381	22.6
HU90-031-047P	45°51.14'N	57°35.56'W	473	29.9
HU91-045-006P	54°42.26'N	56°27.06'W	530	o
HU91-045-006T	54°42.26'N	56°27.06'W	530	o
HU91-045-052P	59°29.51'N	39°18.39'W	2,883	o
HU91-045-072P	58°56.45'N	28°44.32'W	2,237	3.9
HU91-045-080	53°04.40'N	33°31.78'W	3,024	x
HU91-045-082P	52°51.67'N	35°32.09'W	3,109	o
HU91-045-085P	53°58.51'N	38°38.25'W	3,603	o
HU91-045-091T	53°19.81'N	45°15.74'W	3,870	6.7
HU91-045-094P	50°12.26'N	45°41.14'W	3,448	4.9
MD95-2009	62°44.25'N	03°59.86'W	1,027	x
MD95-2010	66°41.05'N	04°33.97'W	1,226	o
MD95-2033	44°39.87'N	55°37.21'W	1,412	45.7
MD95-2220AH	48°38.33'N	68°37.82'W	324	o
MD95-2227	58°12.64'N	48°22.38'W	3,460	5.5
MD99-2254	56°47.78'N	30°39.86'W	2,440	x
PAR87-A10	54°21.80'N	148°28.00'W	3,664	x
AR92-P1	73°42.40'N	162°44.60'W	205	x

wide range of sea-surface temperatures, from 0° to 8°C in winter, and from 8° to 25°C in summer, with maximum occurrence above 14°C in August. The occurrence of *S. trifida* also corresponds to salinity ranging from 31 to 34.5 in summer.

Late Quaternary distribution of *Scrippsiella trifida* cysts.—Fossil cysts reported as *Alexandrium excavatum*-type throughout the latest Pleistocene and Holocene of the Gulf of St. Lawrence (de Vernal et al., 1993, 1996; Simard and de Vernal, 1998; Levac, 2003) are clearly also the organic remains of *Scrippsiella trifida* cysts. Emerald and La Have basins, off Nova Scotia, eastern Canada, also include specimens that we attribute to *Scrippsiella trifida* (Mudie et al., 2002, fig. 8.5). These specimens show particularly high concentrations during the latest Pleistocene (10,000–13,000 ka) and early Holocene in apparent response to increased temperature and a rise in salinity to about 33.5 (Levac, 2001; Mudie et al., 2002). Other fossil records that we accept as representing *Scrippsiella trifida* are from the early Holocene of the southwest Greenland Rise (as *Alexandrium excavatum* in de Vernal and Hillaire-Marcel, 2000), latest Pleistocene and earliest Holocene of the southwest Greenland Rise (as *Alexandrium tamarense* in Solignac et al., 2004), and latest Pleistocene and earliest Holocene of the Orphan Knoll, northwest North Atlantic (as *Alexandrium tamarense* in Solignac et al., 2004). The Quaternary fossil record of

Scrippsiella trifida is therefore much more extensive than previously realized, and this species appears to extend at least into the Pliocene of Belgium (Louwey et al., 2004; Fig. 4.7, 4.8).

It is now possible to document the distribution of *S. trifida* during the Late Pleistocene and pre-modern Holocene from palynological analyses of cores collected at various locations of the northern North Atlantic and adjacent seas (Fig. 7, Table 1). The longest records available are from the southwest Greenland Rise (core HU90-013-013P) and Orphan Knoll in the northwest North Atlantic (core HU91-045-094P). They indicate an episodic occurrence of this species during interglacial and interstadial phases of the Late Pleistocene (Figs. 8, 9). Several records are available that represent the last 20,000 years. They show a distinct maximum occurrence peak at the transition from glacial to interglacial stage that approximately corresponds to the Pleistocene–Holocene boundary (Figs. 8, 9). This occurrence peak is marked by relatively high percentages of *S. trifida* in dinoflagellate cyst assemblages at many sites along the margins of southeast Canada and southern Greenland. The highest percentages, up to 47%, occur off Nova Scotia and Newfoundland (de Vernal et al., 1993; Simard and de Vernal, 1998; Mudie et al., 2002; and core MD95-2033, Fig. 8, Table 1). Extremely high fluxes of *S. trifida* are recorded regionally, with up to 10³–10⁴ cysts per cm² per year,

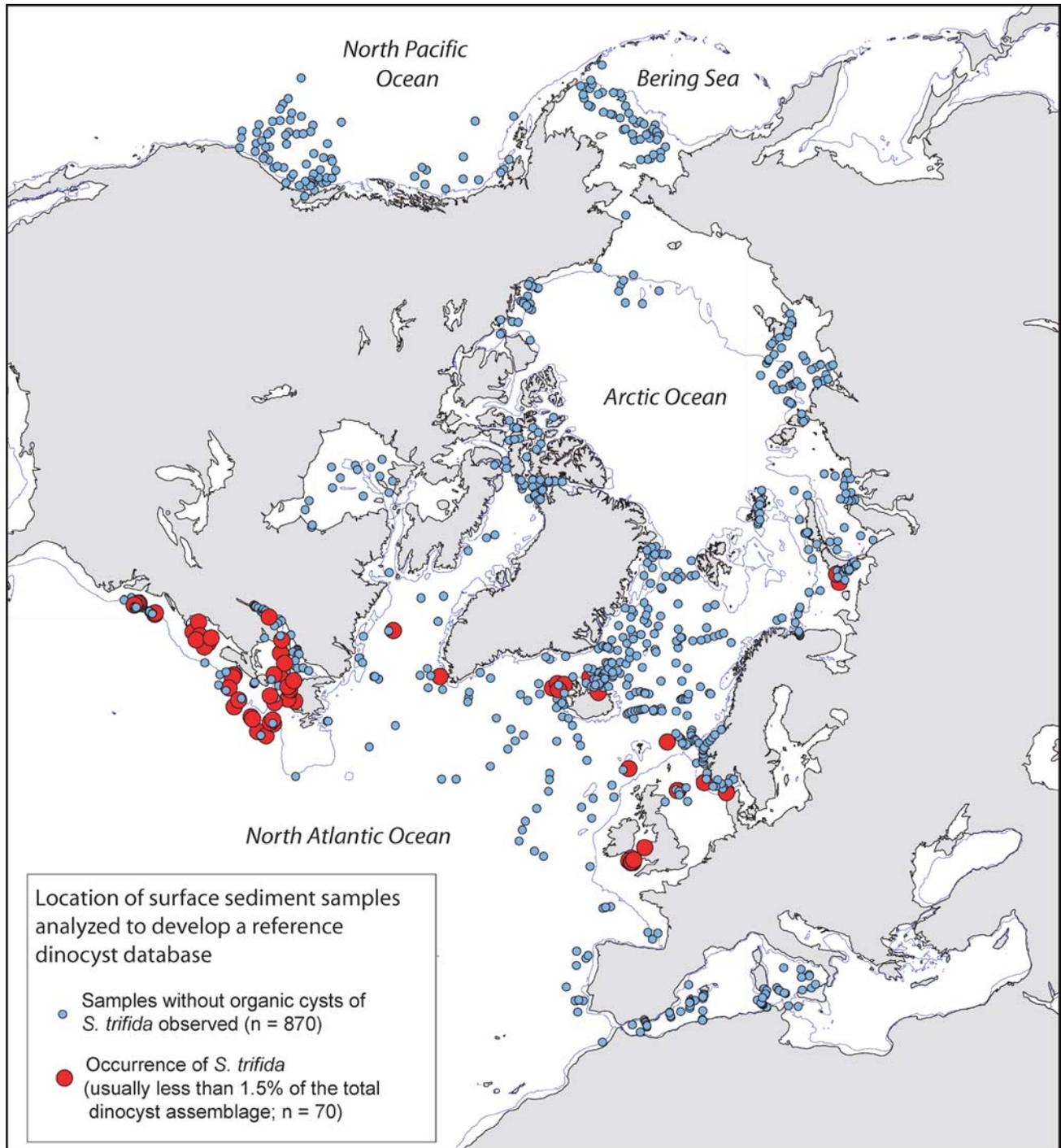


FIGURE 6—Map of the Northern Hemisphere showing the location of 940 surface sediment samples palynologically analyzed to develop a reference dinoflagellate cyst database (de Vernal et al., 2001, 2005). The red circles correspond to sites where organic-walled cysts of *Scrippsiella trifida* have been recovered, whereas the smaller blue circles correspond to sites where these cysts have not been reported.

suggesting that the continental shelf off Nova Scotia and Newfoundland was a main center of dispersal after about 12,500 ka (Fig. 9). The Nova Scotia shelf was then characterized by a relative sea level about 80 m lower than at present (Shaw et al., 2002), thus suggesting that shallow water may have been favorable for *S. trifida* blooms. Moreover, the conditions that prevailed during this interval were marked by maximum summer insolation

(Berger and Loutre, 2002) and by large amounts of freshwater discharge from the retreating ice sheets (de Vernal and Hillaire-Marcel, 2000). Reconstructions based on palynology and stable isotopes indicate that sea-surface waters were characterized by relatively low salinity in the range of 31.5–34.0 and warm summer temperatures ($>15^{\circ}\text{C}$), notably south of Nova Scotia (e.g., Levac, 2001) and at the southern Greenland margin (e.g., Solignac

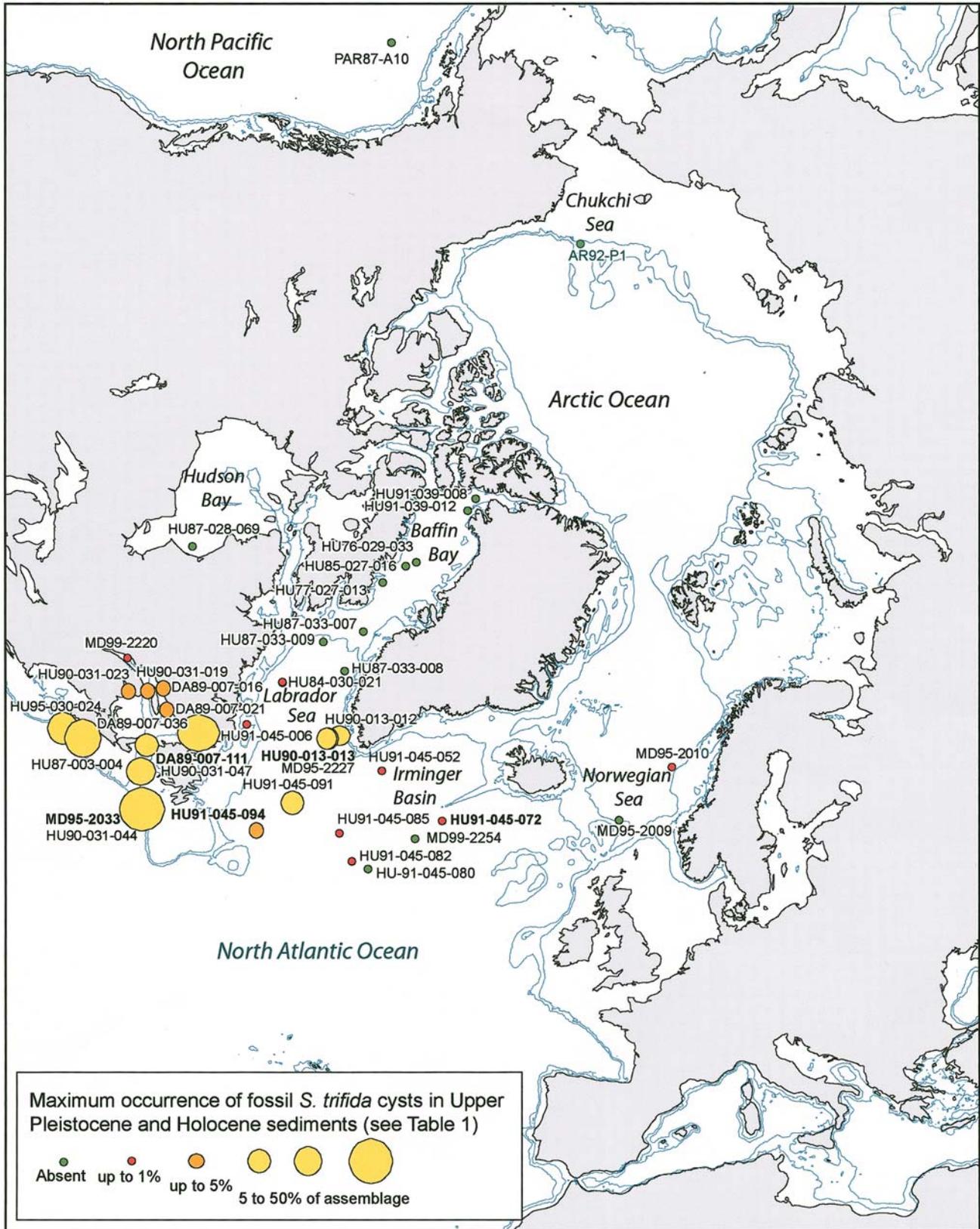


FIGURE 7—Location of cores containing a high-resolution palynological record of late Quaternary sedimentary sequences used to map the distribution of cysts of *Scrippsella trifida* through time (Table 1). Green dots correspond to cores where cysts of *S. trifida* have not been reported. Red, orange, and yellow symbols illustrate the location of cores containing cysts of *S. trifida*. At most of these sites, a maximum occurrence peak is recorded near the Late Pleistocene–Holocene transition. Cores indicated in bold are characterized by a well-constrained chronostratigraphy and are used to

et al., 2004). Hence it appears that extremely high fluxes of *S. trifida* cysts in nearshore areas off Nova Scotia and southern Greenland during deglaciation and early postglacial time (14–7 ka) may signal a reduction in salinity caused by meltwater discharge. The precise means by which meltwater runoff may have stimulated growth of *S. trifida* is not clear, but might be related to seasonal stratification of the water column and reduced competition from diatoms.

Preservation of *Alexandrium tamarense*-type cysts.—*Alexandrium tamarense*-type cysts are already known to be damaged by acetolysis, with only the thin outer wall being resistant. The thicker inner layer swells and then disappears, and is probably cellulosic in composition (Anderson and Wall, 1978). Cysts of *Alexandrium tamarense* may be rendered unrecognizable after standard palynological processing (Dale, 1976), although Cho and Matsuoka (2001) reported that gentle processing of modern sediments from Southeast Asia yielded abundant and well-preserved *Alexandrium* cysts, their identity being confirmed by primuline staining and fluorescence microscopy (Yamaguchi et al., 1995). Our own observations from surface sediments off Orkney show that most features of *Alexandrium tamarense* cysts will survive gentle treatment using cold HCl and cold HF. Moreover, the separation of the thin outer wall layer from the hyaline inner wall layer, and the removal of mucilage from the cyst, produce a morphology superficially similar to the organic wall of *Scrippsiella trifida* cysts. This probably explains the confusion between cysts of *Alexandrium tamarense* and *Scrippsiella trifida* in the palynological literature.

Fossil record of *Alexandrium tamarense*-type cysts.—Not all fossil records of *Alexandrium tamarense*-type cysts are attributable to *Scrippsiella trifida*. Some may indeed represent *Alexandrium tamarense* or a related species, as suggested by reported minor amounts of “smooth-walled oval *Alexandrium tamarense*-type cysts” from the latest Pleistocene of La Have Basin, off Nova Scotia (Mudie et al., 2002, p. 174). Likewise, reports of *Alexandrium tamarense*-type cysts from throughout the Holocene of the Bedford Basin in Nova Scotia, eastern Canada (as *Gonyaulax tamarensis* in Miller et al., 1982; Mudie and Harland, 1996, text-fig. 11), may represent *Alexandrium* because the sediments were deposited in an anoxic basin likely to have enhanced cyst preservation. The illustrated specimen in Miller et al. (1982, pl. 3, fig. 8) resembles an *Alexandrium tamarense*-type cyst but is too deformed to be certain. *Alexandrium tamarense* cysts have been recorded throughout the Holocene of Saanich Inlet, an anoxic basin on the west coast of Canada (Mudie et al., 2002). The younger records spanning the past decade or so might represent *Alexandrium tamarense* because an illustrated specimen from this interval broadly resembles this species (Mudie et al., 2002, fig. 8.1), but it is uncertain whether the older records represent cysts of *Alexandrium tamarense* or *Scrippsiella trifida*.

Organic remains of calcareous dinoflagellates.—*Scrippsiella trifida* is not the only calcareous dinoflagellate to leave discrete organic remains of its cyst wall (see the above section, The Calciodinelloideae, for an account of the fossil record). We now consider organic-walled cysts referred to as *Kallosphaeridium* sp. in Head and Westphal (1999) from the Pliocene of the Bahamas to be the organic linings of cysts of a calcareous dinoflagellate species, based on their similarity to the organic lining of a modern calcareous cyst illustrated by Dale (1983, figs. 27, 28, as an unidentified calcareous dinoflagellate cyst). As noted by Lewis

(1991), Dale’s cyst appears to be assignable to *Scrippsiella crystallina* Lewis, 1991 ex Head, 1996, based on both the calcareous morphology of the wall (Fig. 4.10) and that of its organic lining (Fig. 4.11). *Kallosphaeridium* sp. occurs with frequencies of up to at least 16% of assemblages in the Pliocene of the Bahamas (Head and Westphal, 1999). The thin wall, faint and variable ornamentation, weak expression of tabulation, and lack of awareness among palynologists that calcareous cysts can produce palynomorphs together presumably explain why the organic linings of calcareous cysts are so infrequently reported in the palynological literature.

SYSTEMATIC PALEONTOLOGY

Division DINOFLAGELLATA (Bütschli, 1885)

Fensome et al., 1993

Subdivision DINOKARYOTA Fensome et al., 1993

Class DINOPHYCEAE Pascher, 1914

Subclass PERIDINIPHYCIDAE Fensome et al., 1993

Order GONYAULACALES Taylor, 1980

Suborder GONIODOMINEAE Fensome et al., 1993

Family GONIODOMACEAE Lindemann, 1928

Subfamily HELGOLANDINOIDEAE Fensome et al., 1993

Genus ALEXANDRIUM Halim, 1960

Subgenus ALEXANDRIUM Autonym

ALEXANDRIUM TAMARENSE (Lebour, 1925) Balech, 1985—
type cyst

Figure 1.1–1.6

Cysts of the *A. tamarense* species complex, comprising:

Alexandrium tamarense (Lebour, 1925) BALECH, 1985, p. 38; BALECH, 1995, p. 38–41 (see for full synonymy); cysts illustrated in: DALE, 1977, fig. 1a–c (as *Gonyaulax excavata*); ANDERSON AND WALL, 1978, figs. 27–40, 43–50 (as *Gonyaulax tamarensis*); FRITZ, ANDERSON, AND TRIEMER, 1989, figs. 14, 16–23 (as *Gonyaulax tamarensis*).

Alexandrium catenella (Whedon and Kofoid, 1936) BALECH, 1985, p. 37; cyst documented in: FUKUYO, 1980, p. 51 [as *Protogonyaulax catenella* (Whedon and Kofoid, 1936) Taylor, 1979, unillustrated]; YOSHIMATSU, 1981, pl. 2, figs. 10–12 (as *Protogonyaulax catenella*); FUKUYO, 1985, p. 534 (questionably illustrated, as *Protogonyaulax catenella* or *Protogonyaulax tamarensis* (Lebour, 1925) Taylor, 1979, in fig. 20–p); HALLEGRAEFF AND BOLCH, 1992, fig. 29; FUKUYO, SAKO, MATSUOKA, IMAI, TAKAHASHI, AND WATANABE, 2003, fig. 3.16 (15).

Alexandrium acatenella (Whedon and Kofoid, 1936) BALECH, 1985, p. 37; cyst illustrated in YOSHIDA, MIZUSHIMA, AND MATSUOKA, 2003, fig. 2e–g.

Alexandrium fundyense BALECH, 1985, p. 37, fig. 18; cysts illustrated in: ANDERSON AND WALL, 1978, figs. 22–26, 41, 42 (as *Gonyaulax excavata*); WHITE AND LEWIS, 1982, fig. 2 (as *G. excavata*).

Description of cysts.—Cysts cylindrical with rounded ends; wall smooth, organic; cyst length about 45–55 μm . Wall with three closely appressed layers: inner layer not clearly seen in LM but evident in TEM as amorphous electron-dense layer of variable thickness up to about 0.3 μm ; middle layer, probably cellulose, about 1.0–1.5 μm thick, hyaline and structureless under LM but finely stratified under TEM; outer layer, electron-dense, probably dinosporin, about 0.1–0.2 μm thick, often covered with mucilage incorporating detritus. Cell contents consist of clear to granular cytoplasm with centrally located cluster of larger particles that include one or more orange-red pigmented accumulation bodies and pale granules and globules apparently of starch and lipid.

←

illustrate the occurrence of *S. trifida* cysts through time (see Figs. 8, 9). Note that cores with maximum occurrences of more than 5% have mostly neritic locations, and although core HU91-045-091 in the southern Labrador Sea would appear to be an exception at 6.7%, specimens were recorded in just one sample. Maximum occurrences for cores HU87-003-004 and HU95-030-024 are estimated from Mudie et al. (2002).

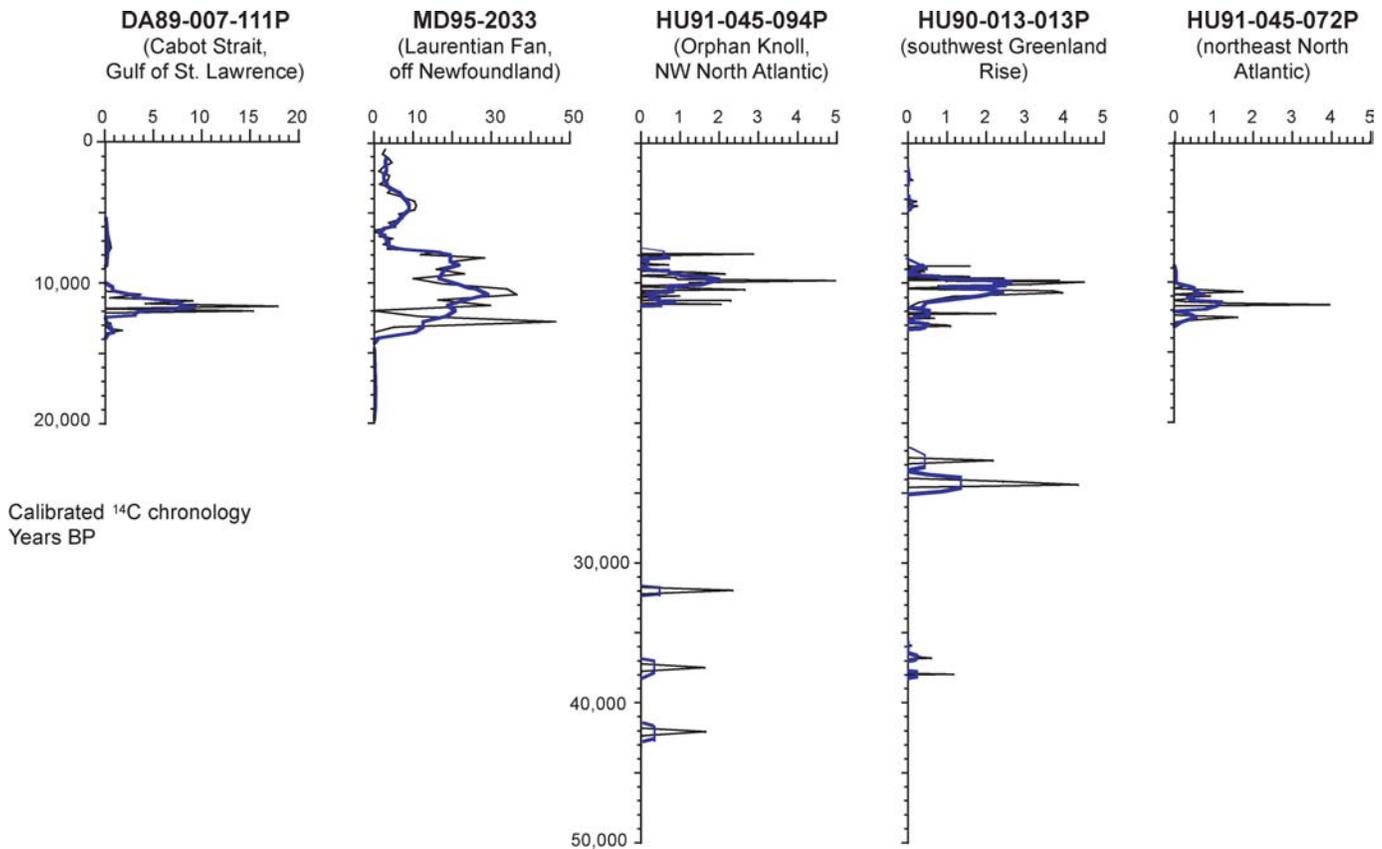


FIGURE 8—Percentage occurrences of *Scrippsiella trifida* cysts from selected well-dated cores of the North Atlantic (see Fig. 7 for location; see also Fig. 9), based on palynological analyses. Thin lines correspond to actual percentage values and the thicker gray curve corresponds to data smoothed according to a window of five samples. Full analyses of several of these cores are published elsewhere: DA89-007-111P (de Vernal et al., 1993), and HU91-045-094P and HU90-013-013P (de Vernal and Hillaire-Marcel, 2000; de Vernal et al., 2000; Solignac et al., 2004). Data from cores MD95-2033 and HU91-045-072P are not published, but are archived at GEOTOP and available upon request.

Immediately prior to excystment, cell contents become progressively browner, particularly from poles towards center (Fig. 1.2, 1.3), where microgranulation develops rapid Brownian-like motion. Archeopyle chasmic, formed by small split at one end of cyst. Based on Dale (1977), Anderson and Wall (1978), Fritz et al. (1989), Kennaway and Lewis (2004), and personal observation.

Material examined.—Specimens from surface sediments of Bedford Basin, Nova Scotia, Canada (Fig. 1.1–1.3); and from surface sediment off Orkney, North Sea (Fig. 1.4–1.6).

Discussion.—The illustrated cysts (Fig. 1.1–1.6) are of identical morphology to cysts of *Alexandrium tamarense*. Under light microscopy, the inner and outer wall layers of living and preserved specimens (Fig. 1.1–1.3) cannot be seen clearly, but specimens exposed to gentle palynological processing (Fig. 1.4–1.6) show a loose, membranous outer layer that represents the separation of middle and outer layers as recognized under TEM.

Anderson and Wall (1978) observed that the cysts of *Alexandrium tamarense* (as *G. tamarensis*) and the related species *Alexandrium fundyense* (as *G. excavata*) are identical. Although the cyst wall in *Alexandrium tamarense* is known to change slightly with maturity (Fritz et al., 1989), a report that the cyst wall of *Alexandrium fundyense* develops “a complex network of structures” (McKenzie et al., 1998, fig. 1) is here rejected on the grounds that the specimen described and illustrated by McKenzie et al. is clearly a bisaccate pollen, probably of pine.

Alexandrium excavatum is considered a taxonomic junior synonym of *Alexandrium tamarense* (e.g., A. R. Loeblich and L. A. Loeblich, 1979; Anderson et al., 1994; Balech, 1995, albeit with some reservations; and Steidinger and Tangen, 1996).

Order PERIDINIALES Haeckel, 1894
 Suborder PERIDINIINEAE Autonym
 Family PERIDINIACEAE Ehrenberg, 1831
 Subfamily CALCIODINELLOIDEAE Fensome et al., 1993
 Genus SCRIPPSIELLA Balech, 1959 emend. Janofske, 2000
 SCRIPPSIELLA TRIFIDA Lewis, 1991 ex Head, 1996
 Figures 1.7–1.16, 2, 3, 4.1–4.9, 5

Scrippsiella trifida LEWIS, 1991, p. 101, figs. 9c, 37–48; HEAD, 1996, p. 1229–1230; ELLEGAARD, CHRISTENSEN, AND MOESTRUP, 1994, figs. 3, 4; NEHRING, 1994, figs. 3, 5b; 1995, fig. 2g, tab. 3; 1997, p. 318, fig. 27, tab. 1; GODHE, NORÉN, KUYLENSTIERNA, EKBERG, AND KARLSON, 2001, fig. 2c, tab. 2; HAMER, LUCAS, AND MCCOLLIN, 2001, tab. 3; JOYCE, 2004, p. 177, fig. 9, tab. 1; HOPPENRATH, ELBRÄCHTER, HALLIGER, VAN BEUSEKOM, AND DREBES, 2004, tab. 1; MOSCATELLO, RUBINO, SARACINO, FANELLI, BELMONTE, AND BOERO, 2004, tab. 6.
 ? cf. *Scrippsiella trifida* Lewis, 1991 ex Head, 1996; RUBINO, BELMONTE, MIGLIETTA, GERACI, AND BOERO, 2000, fig. 3e, tab. 4.
Alexandrium excavatum-type cysts. DE VERNAL, GUIOT, AND TURON, 1993, figs. 2–5; DE VERNAL, TURON, AND GUIOT, 1994, fig. 3, tab. 2; SIMARD AND DE VERNAL, 1998, pl. 1, figs. 1–8, 11, 12.
Alexandrium excavatum (auct. non Braarud, 1945) cysts. DE VERNAL,

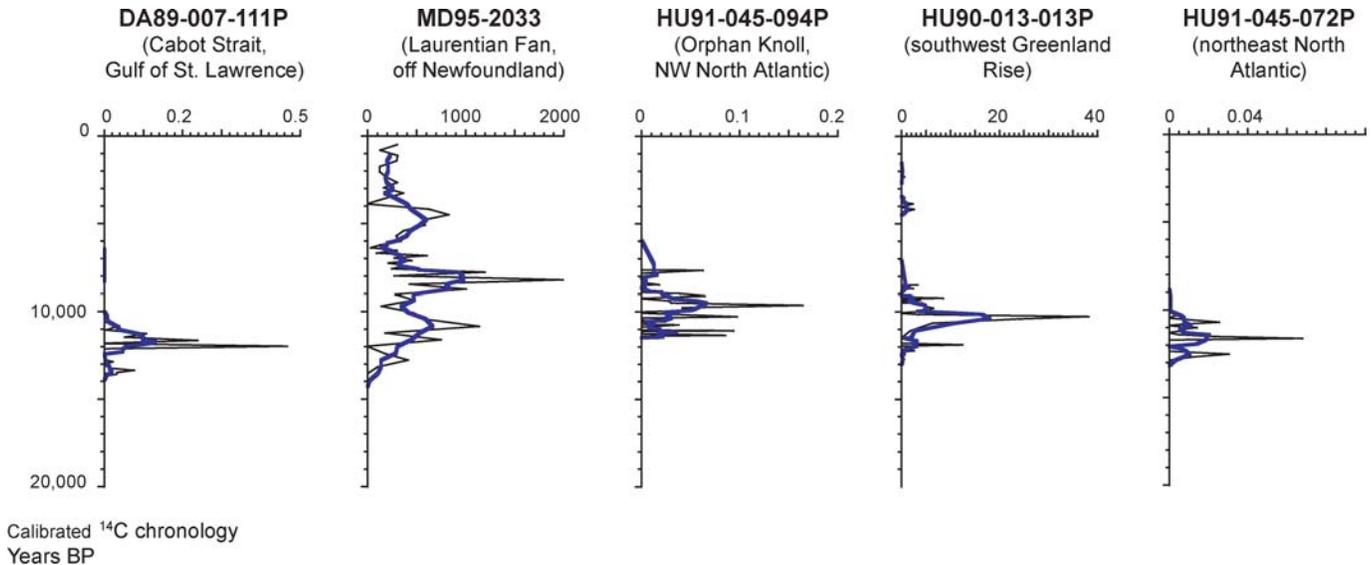


Figure 9—Fluxes of cysts of *Scrippsiella trifida* from selected well-dated cores of the North Atlantic (see Fig. 7 for location; see also Fig. 8), based on palynological analyses. The fluxes (cysts/cm²/year) were calculated from concentrations (in numbers of cysts per cm³) and sedimentation rates (cm per year) estimated from calibrated ¹⁴C ages. Thin lines correspond to actual flux values and the thicker gray curve corresponds to data smoothed according to a window of five samples. Note that flux units are different from one core to another. High fluxes are recorded in core HU90-013-013P, located off southwest Greenland. Extremely high fluxes are recorded in core MD95-2033 located on the Laurentian Fan south of Newfoundland. Such regionally high fluxes are consistent with those reported on the Nova Scotia shelf by Mudie et al. (2002). See also caption to Figure 8.

HILLAIRE-MARCEL, AND BILODEAU, 1996, fig. 2; DE VERNAL, ROCHON, TURON, AND MATTHIESSEN, 1997, figs. 4, 5; BERTINI, DE VERNAL, HILLAIRE-MARCEL, AND BILODEAU, 1998, p. 14, 15; DE VERNAL AND HILLAIRE-MARCEL, 2000, fig. 12; LEVAC, 2001, fig. 6, appendix A; LEVAC, 2003, text-fig. 5.

Alexandrium tamarense (auct. non Lebour, 1925) cysts. SOLIGNAC, DE VERNAL, AND HILLAIRE-MARCEL, 2004, fig. 4.

cf. *Alexandrium tamarense*-type cysts. ROCHON, DE VERNAL, TURON, MATTHIESSEN, AND HEAD, 1999, p. 40, 42, pl. 9, figs. 11–14; DE VERNAL, HENRY, MATTHIESSEN, MUDIE, ROCHON ET AL., 2001, fig. 7, tab. 1; MARRET AND ZONNEVELD, 2003, p. 22, 27, figs. 5–8, tab. 1 (in part); LOUWYE, HEAD, AND DE SCHEPPER, 2004, fig. 10a, b.

Alexandrium excavatum-type cysts of Simard and de Vernal, 1998. MUDIE, ROCHON, AND LEVAC, 2002, fig. 8.5 (lower of the two illustrated cysts).

Description of cysts.—Cysts ovoidal, apex slightly more pointed than antapex; containing centrally located red body. Cyst wall consists of smooth, inner organic layer supporting large, erect, usually trifurcate, recurved calcareous processes, over which lies outer organic layer with irregular topography (Fig. 4.9) that reflects underlying calcareous processes. Processes blunt or bear only rudimentary trifurcations, but shape of undulations in outer organic wall layer not noticeably affected by absence of fully developed trifurcations. Processes ca. 3.0–5.0 μm long, arise from discrete, polygonal to rounded platforms ca. 1.0–1.5 μm thick and up to ca. 5.0–7.0 μm in diameter. Ridges, usually three in number, may be present on upper platform surface, where they radiate outwards from base of process to platform margin in Y-shaped pattern. Adjacent basal platforms may, or may not, contact one another. There are always parts of wall not calcified, although small, scattered blebs of calcite often occur between adjacent process bases in these otherwise uncalcified regions. Crystallographic orientation of processes is tangential (Fig. 2.14–2.16). Processes apparently gonal in distribution. Outer organic wall layer contacts inner layer in places; both layers composed of dinosporin. Archeopyle theropylic, mesoepicystal (sensu Streng et al., 2004), involving plates 2'–4' and 1a–3a, has zigzag principal suture

(e.g., Fig. 4.1, 4.5); operculum polyplacoid, simple, usually adnate, remaining ventrally attached (Fig. 5.3). Based on Lewis (1991) and present study.

Material examined.—Living cysts from unialgal culture (UW426); fossil cysts from latest Pleistocene and early Holocene of core MD95-2033 on the continental slope south of Newfoundland.

Measurements.—Preserved cysts (without acid treatment) from unialgal culture: central body length, 29(33.0)36 μm . Eight specimens measured. Palynologically prepared cysts from the early Holocene of core MD95-2033: central body length, 29(35.4)41 μm , maximum length, 37(43.0)50 μm ; central body equatorial diameter, 19(22.9)26 μm , maximum equatorial diameter, 27(32.0)38 μm . Twelve specimens measured.

Discussion.—Most of the cultured specimens and those from core MD95-2033 have processes that lack well-developed trifurcations, with only the specimen on Figure 1.7 and 1.8 showing this feature clearly. Lewis (1991) also observed reduced trifurcations in cultured material.

Specimens from the uppermost Pleistocene of core MD95-2033 lack full cell contents, but undehisced specimens frequently contain an accumulation body (Fig. 2.5–2.9) which may derive from the red body in living cysts (Kokinov et al., 1998; Kennaway and Lewis, 2004). The zigzag archeopyle, with operculum either attached ventrally or removed completely, was observed frequently. An analysis of the crystallographic orientation (*c*-axis) of the processes, following the method of Janofske (1996, 2000), shows it to be tangential to the cyst surface for these specimens. This is a characteristic feature of the genus *Scrippsiella* (Janofske, 2000).

The name *Scrippsiella trifida* was not validly published in Lewis (1991) because the institution where the type material is conserved was not specified (ICBN Art. 37.6), this condition being fulfilled in Head (1996); and because two specimens, not one, were indicated as the holotype in Lewis (1991), as noted by Head (1996), who designated an illustrated cyst (Lewis, 1991, fig. 41; Fig. 4.9) as the holotype.

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