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LIFE HISTORY AND *IN SITU* GROWTH RATES OF *ALEXANDRIUM TAYLORI* (DINOPHYCEAE, PYRROPHYTA)[1]

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ABSTRACT

Alexandrium taylori *Balech is a phototrophic marine dinoflagellate. It produced recurrent blooms during the summer months (July and August) of 1994 to 1997 in La Fosca beach (NW Mediterranean). In addition to a motile vegetative form,* A. taylori *had two benthic forms: temporary cysts and resting cysts. Temporary cysts were a temporally quiescent stage produced from the ecdysis of the vegetative cell in both natural populations and laboratory cultures. Temporary cysts may divide to form motile cells. Resting cysts had a thicker wall than the temporary cysts and had a red accumulation body. Gametes and planozygotes were also observed in laboratory cultures.* Alexandrium taylori *showed* in situ *diurnal vertical migration with an increase of vegetative cells in the water column in the morning through midday, with concentrations peaking in the afternoon followed by lower levels at night. Most vegetative cells lost their thecae and flagella, and with them their motility, turning into temporary cysts that settled in the early evening. The number of temporary cysts in the water column rose in the evening and at night. The temporary cysts gave rise to motile cells the following morning. Synthesis of DNA occurred in vegetative cells at night, and a preferential period of cell division occurred at sunrise. The estimated division rate in the field was 0.4–0.5 vegetative cells·day^{-1}. Temporary cysts had twice the DNA of a G_1 vegetative cell. The minimum* in situ *division rate of the temporary cysts was 0.14 day^{-1}. The role of the resting and temporary cyst population in the annual recurrence and maintenance of the* A. taylori *bloom is discussed.*

Key index words: Alexandrium, in situ *growth rate, life history, planozygotes, resting cyst, temporary cyst*

Many physical, chemical, and biological factors contribute to the development and persistence of dinoflagellate blooms. Research into growth rates and life cycles is needed to explain the population dynamics of dinoflagellates, including the onset and end of blooms, survival over extended periods, and recurrence of blooms. The measurement of species-specific growth rates *in situ* is important in prediction of red tides.

Alexandrium taylori is a little-known dinoflagellate, first described in Arcachon (France) (Balech 1994). The first reported blooms attributed to *A. taylori* in the Mediterranean Sea occurred in relatively open shores along the coast of Catalonia in the warmer months of the year (July and August) (Garcés 1998). Recurrent blooms have been reported since 1994. The first studies on *A. taylori* reported morphological differences between Atlantic and Mediterranean populations and the occurrence of two kinds of cysts in the life cycle: resting and temporary (Delgado et al. 1997). The formation of durable resting cysts in some dinoflagellates permits over-wintering (Anderson and Wall 1978, Doucette et al. 1989, Fritz et al. 1989). Temporary cysts have been described in *Alexandrium hiranoi* (Kita et al. 1985, Kita and Fukuyo 1988) and *A. pseudogonyaulax* (Montresor 1995), and both of these species have been included in the subgenus *Gesnerium* as *A. taylori* (Balech 1994). In the present study the term ''resting cyst'' is used for a dormant zygote and the term ''temporary cyst'' for a temporally quiescent stage. The temporary cyst also has been called an ecdisal or pellicle cyst because the cell sheds its theca and emerges as a nonmotile cell surrounded by a pellicle. This cyst is produced asexually and is fully active metabolically and reproductively (Taylor 1987).

The aim of this paper is to describe the morphological life stages of *A. taylori* and the division cycle of the vegetative cells as well as the temporary cysts. In studying algal life cycles, it is important to investigate the alternation of nuclear phases. Haploid and diploid cells can be distinguished reliably with microfluorometric techniques for the determination of DNA content (Carpenter and Chang 1988, Cetta and Anderson 1990, Yamaguchi 1992, Veldhuis et al. 1997). This study used microfluorometry to quantify the DNA of individual cells of *A. taylori* and investigate the cell cycle by analyzing the diel pattern of DNA synthesis. The species-specific *in situ* growth rate of the vegetative cells was estimated by cell cycle analysis (Carpenter and Chang 1988). The *in situ* growth rate was defined as a potential growth rate estimated from cell divisions without taking population losses into account. The mitotic phase of the temporary cysts was used to estimate the *in situ* growth rate of this cellular stage.

MATERIALS AND METHODS

Alexandrium taylori blooms were observed on the coast of Catalonia, chiefly at La Fosca (Palamós). The beach of ''La Fosca'' is located in the northeastern Iberian Peninsula on the Catalan Costa Brava (41°51′30″ N 3°08′ E) 2 km south of Palamós (Fig. 1). It is approximately rectangular and measures 525 × 300 m, with the opening oriented toward the southeast. The average and maximum depths are 3 and 7 m, respectively, with a fairly uniform and gentle slope between 2 and 7 m. The beach is divided into two areas by the Roca Negra, which stretches 30 m into the sea (La Fosca beach and the St. Esteve beach). The mean sediment size in La Fosca ranged from 0.286 mm to 0.467 mm and con-

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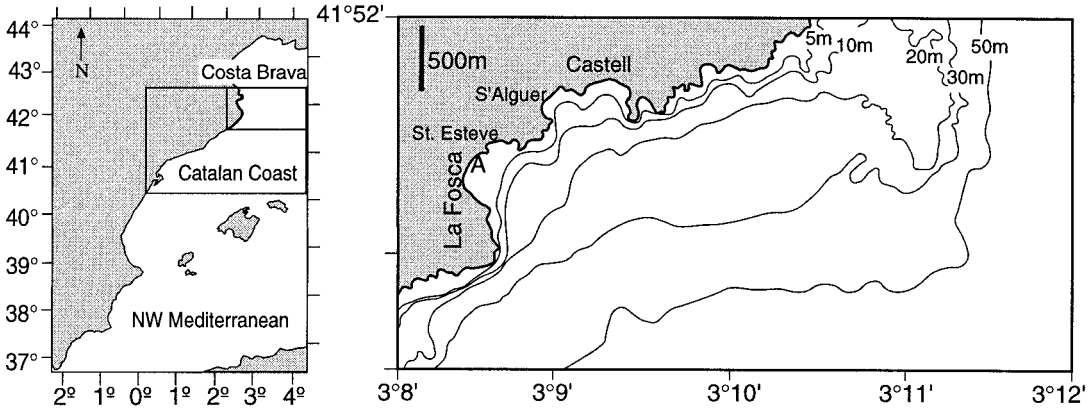

FIG. 1. Study area. Catalan Coast and the Costa Brava in the northwestern Mediterranean. St. A at La Fosca is the fixed station sampled from the beach.

sisted of 98% clean sand. Although the beach is relatively open, its waters tend to remain within the confines of the beach. The weak hydrodynamic conditions result in areas of preferential accrual of settlement. These conditions combine to produce a shallow beach where the water is warmed in summer to temperatures (24°–28° C) higher than normal values for neighboring areas.

Morphology. Cell morphology was observed both in the field samples and in laboratory cultures (*A. taylori* clone AV7), which were maintained in f/2-Si medium at 150 μmol photons·m^{-2}·s^{-1} irradiance with a 12:12 h LD cycle at 19° C. For morphological observations and quantification of vegetative cells and temporary cysts, water samples were fixed in formaldehyde and allowed to settle in 50-mL settling chambers. To distinguish vegetative cells from temporary cysts, samples were stained with calcofluor, which is specific for β-linked polysaccharides such as cellulose (Fritz and Triemer 1985); vegetative cells had a well-defined theca when stained with calcofluor, whereas temporary cysts were unstained. Cell stages were quantified under an inverted epifluorescence microscope.

Gametes and planozygotes. Morphology was observed in live laboratory cultures (*A. taylori* clone AV8), which were maintained in f/2-Si medium at 100 μmol photons·m^{-2}·s^{-1} irradiance and a 12:12 h LD cycle at 25° C.

Resting cysts. Seawater (5 L) from the sampling site, with sediment and the natural planktonic population of *A. taylori,* was maintained in the laboratory at 19° C and 50 μmol photons·m^{-2}·s^{-1} with a 12:12 h LD cycle. Cell stages in the water, in the sediment, and on the sides of the container were observed regularly every month for 1 year.

Diel changes in vegetative cells and temporary cysts. Initial sampling was performed at 3-day intervals during the warmest period of the year (June– September 1995 and 1996) to detect and monitor the presence of *A. taylori* in La Fosca beach. When cell density values were highest (water discoloration occurred), sampling was carried out every 2 h during a 24-h period (19–20 August 1996) to quantify and analyze diel changes in the population of vegetative cells and temporary cysts. Water samples (150 mL) were collected from La Fosca beach every 2 h starting at 0800 GMT at the surface and at a depth of 1 m. There were 14 h of daylight and 10 h of darkness at that time of year; sunrise was at 0600 GMT. The *in situ* irradiance was measured using a LI-COR photometer (LiCor, Lincoln, Nebraska), and the mean was 500 μmol photons·m^{-2}·s^{-1}, with a maximum of 1800 μmol photons·m^{-2}·s^{-1}.

The in situ *growth rate.* The *in situ* growth rates of vegetative cells in the field were estimated mainly on the basis of microfluorometry, measuring the relative nucleic acid content of cells (phases G_1, S, and G_2M of the cell cycle). The division rate of temporary cysts was also estimated from the proportion of mitotic cells observed. A mesh water sample (10-μm mesh) was collected from La Fosca beach at the surface every 2 h over 24-h periods on two occasions (2–3 August 1995 and 19–20 August 1996) starting at 1200 GMT and 0800 GMT, respectively. There were 14 h daylight and 10 h darkness at that time of the year; sunrise was at 0600 GMT. The *in situ* irradiance was measured using a LI-COR photometer, and the daily mean was 500 μmol photons·m^{-2}·s^{-1}, with a maximum of 1800 μmol photons·m^{-2}·s^{-1}.

The mesh sample (200 mL) was filtered through a 60-μm mesh to remove larger organisms. The filtrate was collected in chilled methanol and stored at −20° C for at least 8 h for pigment extraction. To measure the nucleic acid content of the vegetative cells, a subsample of 1 mL was centrifuged (21,000 × *g,* 2 min). The pellet was washed twice in PBS buffer, resuspended in the same buffer, and stained with DAPI to label the DNA at a final concentration of 1 μg·mL^{-1} for at least 1 h in the dark. The sample of stained cells was centrifuged to a minimum volume and transferred to a slide with coverslip. Observations and counts for estimating the cell phases were performed using a Nikon Optiphot epifluorescence microscope with a Nikon P1 microfluorometric system for measuring the intensity of fluorescence in a specific region. Passage of light through the photomultiplier was reduced to a pinhole large enough to admit the light from the entire nucleus of the cell being measured. The effect of nuclear orientation was tested by causing cells to roll into a new orientation for each measurement. There was no difference between means when the orientation was changed, but the standard deviation of the mean increased. Thus, since the U shape of the nucleus in *A. taylori* may be a major source of variation in the DNA measurements, all nuclei were measured with the U shape in an upright position in order to reduce variability.

Between 250 and 300 cells were measured in each sample with the assumptions of the model (proposed by Carpenter and Chang 1988, Chang and Carpenter 1990), and the measurements were used to construct a DNA histogram. DNA histogram analysis was performed with Multicycle software (Phoenix Flow Systems). The *in situ* growth rate from the different phases in the cell cycle was evaluated by the method of Carpenter and Chang (1988). Daily growth rates were estimated using the S and G_2M phases as the terminal event. Because it is difficult to sample at fixed intervals under field conditions, weighted means of phase fractions were used to calculate growth rates (Chang and Carpenter 1985):

$$\mu = \frac{1}{n(T_S + T_{G_2M})} \sum_{i=1}^{n} (t_s)_i \ln[1 + f_S(t_i) + f_{G_2M}(t_i)]. \quad (1)$$

The duration term (T) was measured by monitoring the diel change in the S and the G_2M phase fractions:

$$(T_S + T_{G_2M}) = 2(t_2 - t_1). \quad (2)$$

In equations 1 and 2, μ is the *in situ* growth rate (day^{-1}) from DNA synthesis, $f_{S(t)}$ is the frequency of cells in phase S, f_{G_2M} is the frequency of cells in phase G_2M, n is the number of samples, T_S is the duration of the S phase, T_{G_2M} is the duration of the G_2M phase, t_s is the interval in hours, t_1 is the time when the curve for

the number of cells in the S phase has attained its peak value, and t_2 is the time when the curve for the number of cells in the G_2M phase has attained its peak value. The values of t_1 and t_2 were estimated by fitting fourth-degree polynomial curves to the S and G_2M phase fraction values (Carpenter and Chang 1988).

Morphological observations indicated that temporary cysts could divide *in situ*; however, because no information about the duration of mitosis was available, the minimum *in situ* division rate for temporary cysts was estimated by the modified mitotic index (Vaulot 1992):

$$\mu_{min} = \ln(1 + f_{max}) \quad (3)$$

where f_{max} is the largest fraction of cells undergoing the terminal event and whose duration does not need to be known. Mitosis was the phase selected for the temporary cysts. The *in situ* division rate for temporary cysts considered in this paper was the minimum cell division rate.

To determine the concentration of DNA in vegetative cells and temporary cysts, culture samples were fixed at 13 h (5 h after light period) in 1% formaldehyde for 10 min, centrifuged, resuspended in chilled methanol, and stored at −20° C. Cell staining with DAPI followed the same protocol as described above for the microfluorometry samples. Human lymphocytes were stained under the same conditions and their fluorescence was measured as a reference for the DNA measurements. Fluorescence measurements were as described above.

RESULTS

Morphology. The wild vegetative cells of *A. taylori* had a mean length of 33.6 μm (range 26.3–42.7 μm, SD = 3.8 μm, n = 50), although in culture the variability increased significantly, with cells reaching 70 μm in length. There were several chloroplasts in the cytoplasm. The nucleus was U-shaped (Fig. 2A). Vegetative cells were dark green and contained reserves of starch and lipids. Starch granules were distributed throughout the cytoplasm and varied in size and shape. Lipid granules were present as droplets of variable size. The organism had a weak theca, which detached from cells in fixed samples.

Vegetative cells undergo ecdysis with complete loss of the theca. Ecdysis required opening of all amphiesmal membranes external to the theca, including the flagellum, whose membrane was continuous with the cell membrane; thus, several empty theca were found in the culture and field samples.

Vegetative division, or cytokinesis, in *A. taylori* occurred in the mother cell, giving rise to two daughter cells, which remain attached for a time and retain part of the wall of the mother cell (Fig. 2B). Vegetative cells produced temporary cysts in a few hours by ecdysis. The inner layer surrounding the resulting cells was the pellicle layer; therefore, these temporary forms were also called ecdisal or pellicle cysts (Fig. 2F). In turn, the temporary cysts, which were not motile, gave rise to motile vegetative cells in 1 day. Temporary encystment of the vegetative cells occurred in active, healthy cells, which passed into a state of temporary quiescence, although no physiological stress was discernible in new clonal cultures. Temporary cysts were cells enclosed by pellicles without thecae and flagella; consequently, they were nonmotile (Fig. 2F). The walls of temporary cysts were thicker than the thecae of vegetative cells and lacked ornamentation. Because calcofluor weakly stained the pellicle layer (devoid of cellulose), no surface plate pattern was discernible in the temporary forms. The temporary cysts formed in laboratory cultures had different shapes and sizes: coccoid (40–33 μm, n = 25), ovoid (42–26 μm, n = 25), spherical (35 μm, n = 25), and bilobate. Old temporary cysts in the field were mainly coccoid and bilobate. The organelles in the temporary cysts were the same as in the vegetative cells, with numerous chloroplasts distributed throughout the cell and a U-shaped nucleus in the same position as in the vegetative cells. Temporary cysts lost their red chlorophyll autofluorescence as they aged but retained brown coloration from other pigments.

Temporary cysts divided when the cytoplasm cleaved into two, four, or even eight cells (Fig. 2C). The pellicle layer was retained until the cells separated. In culture, the temporary cysts formed new thecae and flagella, giving rise to a vegetative cell without undergoing division, especially the bilobate forms, which retained the shape of the vegetative cell, except that the cingulum was not as prominent. In culture, the motile cells were formed from temporary cysts in a period of days without change in the environmental conditions. The pellicle layer remained attached to the chamber wall, with a round hole through which the cell protruded.

Temporary cyst formation in the field took place mainly at sunset and in the early evening (1–2 h before sunset) when the number of vegetative cells in the water column dropped appreciably as a result of marked migration (Fig. 3). The vegetative cells reached a maximum concentration of 10^6 cells·L^{-1} at the surface during the midday–afternoon period and diminished during the evening. The percentage of the total population at a temporary cyst stage in relation to vegetative cells before the dark period varied between 13% and 59%. The cell concentration of the samples taken from the bottom (1 m deep) tended to diminish during the night. At the same time, cluster formation near the sediment was observed, with cell concentrations of vegetative cells and temporary cysts greater than 10^7 cells·L^{-1}. The large cell concentrations that formed these clusters made representative sampling difficult, which means that quantification of the cells on the bottom was underestimated during the dark period for the temporary cysts.

Resting cysts were obtained in the laboratory from the vegetative population with sediment maintained for 1 year at 19° C and a 12:12 h LD cycle with an available irradiance of 50 μmol photons·m^{-2}·s^{-1}. Resting cysts were spherical, 40–45 μm diameter (n = 25), with globular organelles, a red accumulation body, a thicker wall than temporary cysts, and, unlike vegetative cells, large vacuoles (Fig. 2D). These forms were found clumped together and attached to the container walls in the laboratory cultures.

Gametes and planozygotes were observed in stationary phase of live cultures. Gametes were gym-

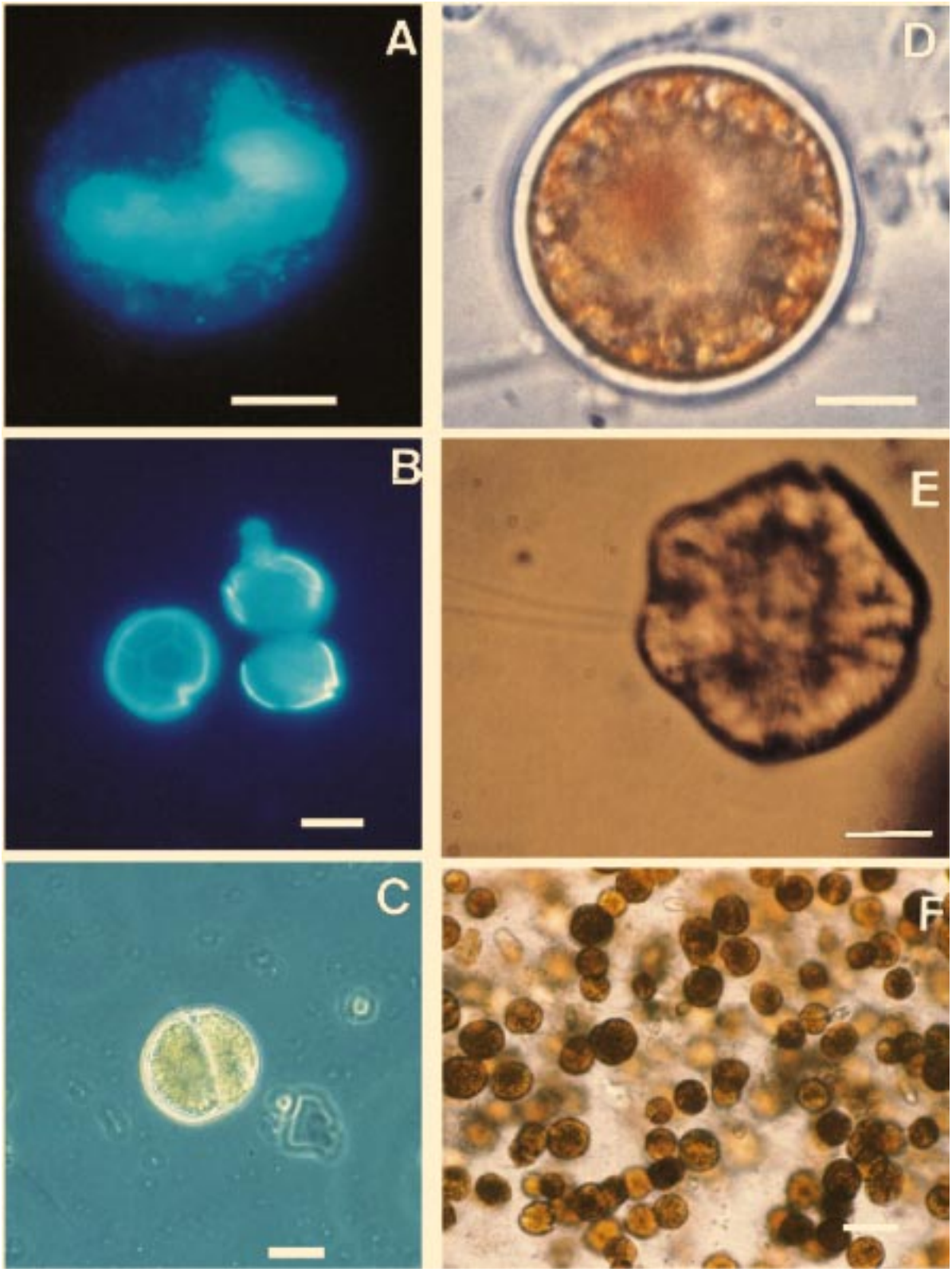

FIG. 2. (A–F) *Alexandrium taylori* from La Fosca. (A) Detail of the U-shaped nucleus of the vegetative cell with DAPI stain. (B) Detail of cell division of vegetative cell with calcofluor stain. (C) Division of temporary cysts in two cells. (D) Resting cyst. (E) Planozygote and detail of two trailing flagella. All scale bars for A–E are 10 μm. (F) Temporary cysts in culture showing size variability. Scale bar for F is 50 μm.

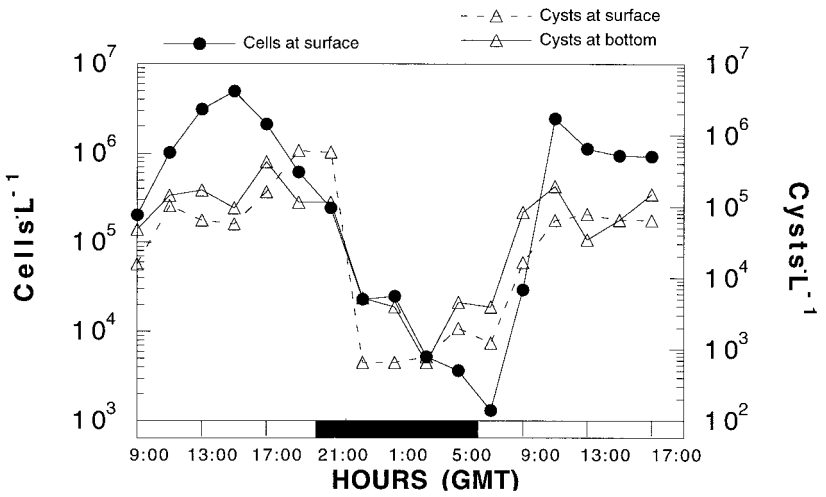

FIG. 3. Cell density of vegetative cells (at surface) and temporary cysts (at surface and bottom) of *A. taylori* in the water column over a diel cycle (19–20 August 1996). Horizontal black bar at bottom indicates the period between sunset and sunrise.

nodinoid small cells (20 μm mean length, n = 25) lacking thecal plates (unstained by calcofluor). Fused pairs of isogametes having one longitudinal flagellum each were occasionally observed. The couplets swam in a hellicoidal pattern. Planozygotes were numerically dominant in these cultures with regard to vegetative cells. Planozygotes were large cells (43–50 μm diameter, n = 25) having thecal plates and two trailing flagella (Fig. 2E).

Growth rate. The frequency of *in situ* cell division was estimated from the diel pattern of DNA content (Fig. 4a, c). The rhythm followed by the phases of cell division in *A. taylori* was appropriate for the application of the *in situ* growth model. *Alexandrium taylori* had a peak of DNA replication (S) between 4 and 5 h (1995 and 1996, respectively) after the dark period and a peak in cell division (G_2M) after sunrise in the 1995 cycle and few hours before sunrise in the 1996 cycle. The estimated duration of the S phase was 4.16 ± 0.8 h (n = 2) and that of the G_2M phase was 3.14 ± 1 h (n = 2). The *in situ* growth rate estimated from the frequencies of the S and G_2M phases was 0.4 day^{-1} in 1995 and 0.5 day^{-1} in 1996.

Like the vegetative cells, the temporary cysts presented an *in situ* cell division pattern with a peak frequency around sunrise (Fig. 4b). The division rate of the temporary cysts estimated from the mitotic index was 0.14 day^{-1}.

Data from microfluorometry showed that the relative concentration of DNA of temporary cysts was different from the vegetative cells (Table 1). DNA synthesis in vegetative cells represented a restricted segment of the photocycle, thus the cells in the G_2 phase had twice the concentration of DNA of those in G_1. The concentration of DNA in the temporary cysts is like that of the cells in G_2: almost double the concentration of DNA (1.8) in G_1 vegetative cells.

DISCUSSION

The two different nonmotile stages of the life history of *A. taylori* (temporary and resting cysts) are clearly characterized by their morphological features. Although few cytological differences are observed between the vegetative cell and the temporary cyst, the distinctive cell wall that surrounds such cell stages (theca and pellicle layer) differentiates the two cell types, especially in the field samples. The temporary cysts in *A. taylori* did not display a patterning of the outer cell wall, unlike those of *A. ostenfeldii* (Østergaard and Moestrup 1997), which can be stained using calcofluor and to reveal a surface pattern.

Sexual stages, such as gametes and planozygotes, were observed in the culture clone AV8 of *A. taylori.* Unfortunately, resting cysts from these planozygotes were not observed in 1 month. Resting cysts have been obtained from natural populations and are probably the result of sexual reproduction.

Although nuclear cyclosis may be a general feature of meiosis in dinoflagellates, it was not observed in *A. taylori.* This phenomenon has been described in various species of dinoflagellates, for example in *A. hiranoi* and *Amphidinium klebsii* (Barlow and Triemer 1988), especially during division of temporary cysts (Biecheler 1952, von Stosch 1972, 1973). If cyclosis is a morphological marker associated with meiosis, then the data do not permit association of the divisions of either vegetative cells, temporary cysts, or sexual stages with meiosis. The involvement of sexual processes in the divisions or in other cell stages in the life history of this species cannot be ruled out, because meiosis in dinoflagellates may involve many different stages in the cell cycle, namely, motile cells, temporary cysts, planozygotes, hypnozygotes, and planomeiocytes (von Stosch 1973). Knowledge of the ploidy level of such cell stages

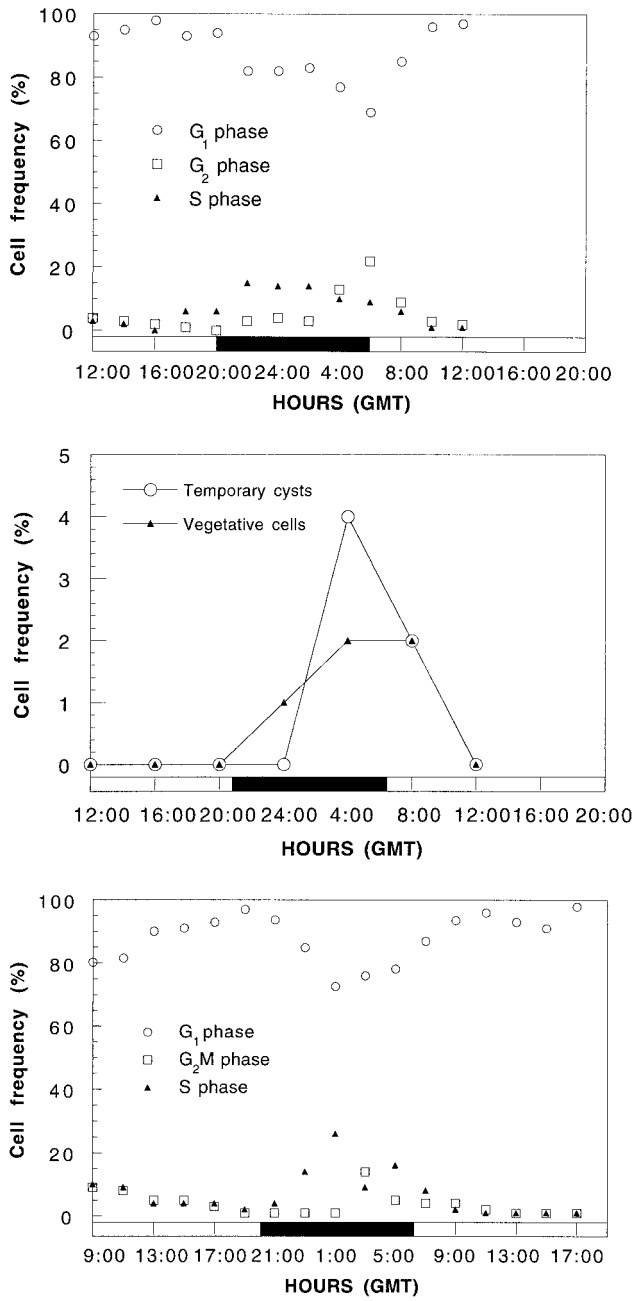

FIG. 4. Percentage values for the cell phases of *A. taylori*. (a) Phases G_1, S, and G_2M over a diel cycle on 2–3 August 1995. (b) Mitotic phase for the vegetative cells and temporary cyst over a diel cycle on 2–3 August 1995. (c) Phases G_1, S, and G_2M over a diel cycle on 19–20 August 1996. Horizontal black bar at bottom indicates period between sunset and sunrise.

such as temporary cysts, gametes, planozygotes, and resting cysts is required before we can understand meiosis in the life cycle of dinoflagellates. The relative concentration of DNA in the temporary cysts of *A. taylori* (1.8 times greater than the vegetative cells) could be interpreted as a duplication of DNA prior to cell division, such that the temporary cysts could divide and give rise to two vegetative cells. Unfortunately, no estimation of concentration of DNA

TABLE 1. *Relative DNA concentration of the vegetative cells (n = 240), the temporary cysts (n = 240) of* A. taylori *and the human lymphocytes (n = 50).*

	Vegetative cells	Temporary cysts	Lymphocytes
Minimum	73	150	7
Maximum	230	460	13
Mean	98	180	12
Median	95	168	12
SD	11	33	0.5
SE	1	3	0
CV	11	18	4

of the temporary cysts in a diel cycle is presented here, since the coefficient of variation of the DNA histograms (18%) was high.

The vegetative cells produced ecdisal, pellicle, or temporary cysts by ecdysis. Ecdysis also occurs in certain species of gonyaulacoids and peridinoids (Taylor 1987) in response to environmental stress or encystment. Temporary cysts have been reported in the genus *Alexandrium* (Anderson and Wall 1978, Schmitter 1979, Kita et al. 1985, 1993, Doucette et al. 1989, Fritz et al. 1989, Montresor 1995, Østergaard and Moestrup 1997), and they have also been described in other genera such as *Ceratium* (Chapman et al. 1982), *Amphidinium* (Sampayo 1985, Barlow and Triemer 1988), and *Disodinium* and *Pyrocystis* (Elbrächter and Drebes 1978). However, although temporary stages are not uncommon in dinoflagellates, the factors triggering the formation of temporary cysts from motile cells, and vice-versa, are unclear. The formation of temporary cysts has been attributed to stressful conditions such as deficiencies in specific nutrients (Doucette et al. 1989), changes in temperature (Schmitter 1979, Grzebyk and Berland 1996), and aging of cultures (Østergaard and Moestrup 1997). In our study, formation of temporary cysts was observed toward evening, when cells descended in the water column. There is a cycle of temporary cyst formation, possibly related to light cycles, similar to that in other dinoflagellates (Lombard and Capon 1971), in which excystment and encystment appear to be controlled by light. Since the temporary cyst stage follows a diel cycle of formation and division, this stage in *A. taylori* may be related to cell division and the intrinsic growth of the organism.

Temporary cysts form aggregates that are found in the sediment in the beach. The formation of cell aggregates at the bottom must be caused by cell adhesion because no mucus or exopolymers are involved in these aggregates. Gregarious encystment of dinoflagellates (Lombard and Capon 1971, Horstmann 1980) and ciliates (Crawford and Purdie 1992, Jonsson 1994) occurs in places of strong dilution such as tidal areas, rock tide pools, and estuaries. The mechanistic basis for aggregate formation is largely unknown. In future work, it will be

interesting to correlate cell adhesion with factors such as growth stage and environmental conditions.

After ecdysis, the cells are surrounded by a pellicle. This layer is present in the vegetative cells of *A. taylori* because, during improper fixation, cells readily lose their theca and are enclosed by this inner wall. The pellicle may be present in different cell stages depending on the species. The pellicle could be formed in vegetative cells undergoing division (Morrill and Loeblich 1981) and after ecdysis of vegetative cells (Morrill and Loeblich 1984, Höhfeld and Melkonian 1992) and be present in the vegetative cells (Höhfeld and Melkonian 1992). The pellicle in temporary cysts of *A. pseudogonyaulax* (Montresor 1995) is characterized by low permeability. Also, in other species of dinoflagellates such as *Peridinium* spp. and *Heterocapsa* spp., it is characterized by resistance to acetolysis and low permeability, with some exceptions in *Alexandrium* and *Gonyaulax* species (Morrill and Loeblich 1981). In the case of *A. taylori,* high permeability of temporary cysts occurred following fixation in methanol. This permitted the use of nuclear fluorochromes and may be useful in studies of the temporary cysts in other dinoflagellates.

Cell division occurs in both vegetative cells and temporary cysts. The pattern of vegetative cell division has already been described for other species of the genus *Alexandrium,* such as *A. tamarense* (Turpin et al. 1978). Vegetative division in *A. taylori* is appropriate for application of the *in situ* growth model. Active growth of vegetative cells was shown in the field during the sampling period at high temperatures (26°–28° C). This is consistent with data of laboratory cultures, in which temperatures above 20° C were conducive to growth.

The divisions of temporary cysts observed in *A. taylori* have been described in other species (Sampayo 1985, Barlow and Triemer 1988, Montresor 1995, Silva and Faust 1995). Although the division rate of temporary cysts is lower than that of vegetative cells, the division of temporary cysts can contribute to population growth. Accordingly, population growth in *A. taylori* results from both vegetative cell divisions and the germination or division of temporary cysts (0.4–0.5 day^{-1} and 0.14 day^{-1}, respectively).

Theoretically, processes other than division of vegetative cells and temporary cysts take part in the growth of the population of *A. taylori*: namely encystment and excystment. Future work will focus on these processes, whose rates must also be known in order to quantify population dynamics *in situ.*

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