

**Red tides of the
dinoflagellate *Noctiluca
scintillans* associated with
eutrophication in the
Sea of Marmara (the
Dardanelles, Turkey)***

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Abstract

This investigation focused on weekly variations in cell density and volume of the dinoflagellate *Noctiluca scintillans* between March 2001 and January 2004 in the Dardanelles. March–June and October–December periods were excessive bloom periods. During the bloom periods the density of *N. scintillans* reached 2.20×10^5 cells L^{-1} with a volume of $1.32 \times 10^{12} \mu m^3 L^{-1}$. In addition to the high surface density, there was an increase in subsurface waters during the blooms. The bloom of

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The complete text of the paper is available at <http://www.iopan.gda.pl/oceanologia/>

N. scintillans, like that of diatom and other dinoflagellate blooms, was associated not only with eutrophication, but also with stable temperatures and salinities.

1. Introduction

Noctiluca scintillans (Macartney, 1810) Kofoid, 1920, an unarmoured marine planktonic dinoflagellate and bioluminescent in some parts of the world, is one of the most important and abundant red tide organisms. It has a worldwide (cosmopolitan) distribution in cold and warm waters. It is known that *N. scintillans* occurs in two forms. Red *Noctiluca* is heterotrophic and acts as a microzooplankton grazer in the food web. However, green *Noctiluca* contains the photosynthetic symbiont *Pedinomonas noctilucae* (Subrahmanyam) Sweeney, 1976 (prasinophyte), but it also feeds on other plankton when the food supply is abundant. Red *Noctiluca* occurs widely in the temperate to sub-tropical coastal regions of the world. It occurs over a wide temperature range of about 10 to 25°C and at higher salinities (generally not in estuaries). It is particularly abundant in high productivity areas such as upwelling or eutrophic areas where diatoms dominate, since they are its preferred food source. Green *Noctiluca* is much more restricted to a temperature range of 25–30°C and occurs mainly in the tropical waters of south-east Asia, Bay of Bengal (east coast of India), in the eastern, western and northern Arabian Sea and the Red Sea; recently it has become very abundant in the Gulf of Oman. Red and green *Noctiluca* overlap in their distribution in the eastern, northern and western Arabian Sea with a seasonal shift from green *Noctiluca* in the cooler, winter convective mixing, higher productivity season, to red *Noctiluca* in the more oligotrophic, warmer summer season (Dodge 1982, Fukuyo et al. 1990, Hallegraeff 1991, Taylor 1993, Taylor et al. 1995, Steidinger & Tangen 1996, Harrison et al. 2011).

Toxic blooms of *N. scintillans* have been linked to massive fish and marine invertebrate kills. Although this species does not produce a toxin, it has been found to accumulate toxic levels of ammonia, which is then excreted into the surrounding waters, possibly acting as the killing agent in blooms (Okaichi & Nishio 1976, Fukuyo et al. 1990). Extensive toxic blooms have been reported off the east and west coasts of India, where it has been implicated in the decline of fisheries (Aiyar 1936, Bhimachar & George 1950). *N. scintillans* red tides frequently form in spring to summer in many parts of the world, often resulting in a strong pinkish red or orange discolouration of the water (tomato soup). Blooms have been reported from Australia (Hallegraeff 1991), Japan, Hong Kong and China (Huang & Qi 1997), the Turkish Straits System (TSS) including the Bosphorus, Sea of Marmara and Dardanelles (Unsal et al. 2003, Turkoglu et al. 2004a,

Turkoglu & Buyukates 2005, Turkoglu 2010a, Turkoglu & Erdogan 2010) and the Black Sea (Porumb 1992, Turkoglu & Koray 2002, 2004), where the water is discoloured red. However, not all blooms associated with *N. scintillans* are red. The colour of *N. scintillans* is in part derived from the pigments of organisms inside the vacuoles of *N. scintillans*. For instance, green tides result from *N. scintillans* populations containing green-pigmented prasinophytes (Chlorophyta) that are living in their vacuoles (Housmann et al. 2003). Recent pink blooms with cell concentrations as high as 1.9×10^6 cells L^{-1} were reported from New Zealand (Chang 2000). In Indonesia, Malaysia and Thailand (tropical regions), however, the water colour is green due to the presence of green prasinophyte endosymbionts (Sweeney 1978, Dodge 1982, Fukuyo et al. 1990, Hallegraeff 1991, Taylor 1993, Taylor et al. 1995, Steidinger & Tangen 1996). This large cosmopolitan species is phagotrophic, feeding on phytoplankton (mainly diatoms and other dinoflagellates), protozoans, detritus and fish eggs (Dodge 1982, Fukuyo et al. 1990, Hallegraeff 1991, Taylor 1993, Taylor et al. 1995, Steidinger & Tangen 1996).

In order to discover the circumstances and reasons for the blooms of the dinoflagellate *N. scintillans* in given environmental conditions, the cell density and volume of *N. scintillans* were monitored for about three years between March 2001 and January 2004. These were then analysed in connection with eutrophication parameters such as nutrient and chlorophyll *a* in the system. Moreover, in order to discover the real extent of eutrophication in the system in the context of its biological characteristics, the contribution of *N. scintillans* to Dinophyceae and total phytoplankton was calculated.

2. Material and methods

2.1. Study area

The Dardanelles, a part of the Turkish Straits System (TSS), is located between the Aegean Sea and the Sea of Marmara. The sampling station ($40^{\circ}12'42.12''N-26^{\circ}26'29.18''E$) is located to the north of Cape Nara (Figure 1). The station has a depth of ca 80 m. The presence of Cape Nara makes this a topographical downwelling area. The Strait varies in width from 1.35 to 7.73 km. Its average depth is approximately 60 m with the deepest part reaching more than 100 m (Unsal et al. 2003, Turkoglu et al. 2004a, 2006, Baba et al. 2007). Its general NE/SW direction is interrupted by a north-south bend between Eceabat and Çanakkale. This bend is also the narrowest part of the Dardanelles. In addition, there is a second bend referred to as 'Cape Nara'. The narrowing of the Dardanelles leads to

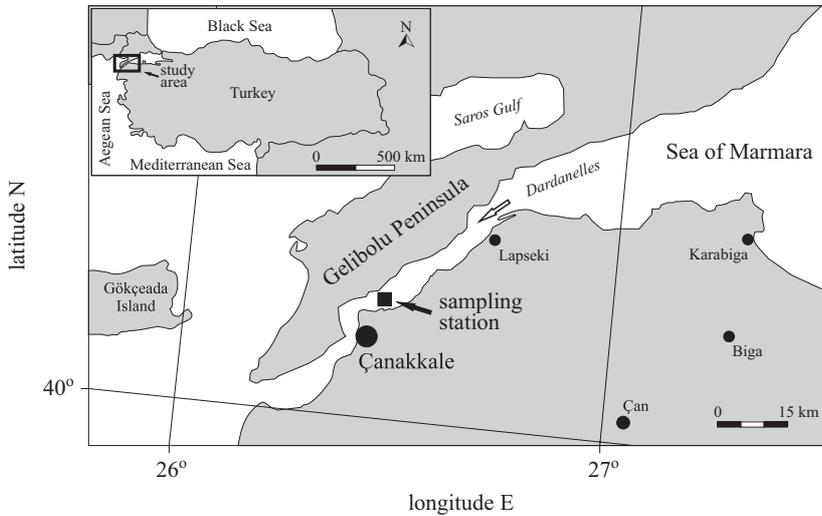


Figure 1. Dardanelles (Çanakkale Strait, Turkey) and study area (sampling station)

different surface temperatures and salinities in the NE and SW parts of Cape Nara. The surface waters in the southern part of the Dardanelles are also more saline, especially in the spring and winter (Unsal et al. 2003, Turkoglu et al. 2004a, 2006).

Two-layer flow regimes in the TSS carry the brackish waters of the Black Sea into the Aegean basin of the north-eastern Mediterranean throughout the year. A counterflow in the TSS carries saline Mediterranean waters into the Black Sea via the Marmara deep basin. The annual volume inflow from the Black Sea to the Sea of Marmara upper layer is nearly twice the volume of saline waters exported from the Sea of Marmara to the Black Sea via the Bosphorus underflow. The brackish Black Sea inflow is relatively rich in nitrate and phosphate in winter, these nutrients dropping to minimum levels in late summer and autumn. Biologically labile nutrients of the Black Sea origin are utilized in photosynthetic processes in the Sea of Marmara and are partly exported to the Marmara lower layer. Ultimately, the brackish Black Sea waters reach the Dardanelles Strait with modified biochemical properties. On the other hand, the saline Mediterranean waters with low concentrations of nutrients enter the Marmara deep basin. During their 6–7 year sojourn in the Marmara basin, the saline waters become enriched in nitrate (DIN) and phosphate (DIP) as a result of the oxidation of planktonic particles sinking from the Marmara surface layer. The annual nutrient inputs from the Black Sea to the Marmara basin were estimated as 8.17×10^8 moles of DIN and 4.25×10^7 moles of DIP, which are much

less than the import from the Marmara lower layer via the Bosphorus undercurrent. Saline Aegean water introduces nearly 6.13×10^8 moles of DIN and 2.79×10^7 moles of DIP into the Marmara lower layer. The estimated outflow of DIP from the Aegean Sea is nearly half the quantity imported from the Sea of Marmara via the Dardanelles Strait (Tugrul et al. 2002).

2.2. Sampling period and collection of samples

This study took place between 05 March 2001 and 25 January 2004 in the Dardanelles. Water samples for nutrient analyses, phytoplankton enumeration and chlorophyll *a* estimation were collected with a Hydrobios Niskin Sampling Bottle (5.0 L) from different depths (0.5, 10, 25, 50 and 75 m) three times a month (approximately every 10 days).

2.3. Probe (CTD) measurements

CTD parameters (temperature, salinity, pH, and dissolved oxygen) were measured in situ using an YSI 6600 Model Multiple Probe System.

2.4. Nutrient measurements

Unfiltered water samples for nutrient analyses were kept frozen until analysis. Analysis for nitrite + nitrate ($\text{NO}_2^- + \text{NO}_3^-$), inorganic phosphate (PO_4^{-3}) and silicate (SiO_4) were measured using a Technicon model auto-analyser according to Strickland & Parsons (1972).

2.5. Chlorophyll *a*

Chlorophyll *a* samples were filtered through GF/F glass fibre filters. The filters were folded into aluminium foil and immediately frozen for laboratory analysis. Chlorophyll *a* was analysed spectrophotometrically after extraction by 90% acetone (Strickland & Parsons 1972).

2.6. Quantitative analysis of *N. scintillans* and other phytoplankton

For quantitative analysis of phytoplankton, samples were preserved with 2% buffered Lugol's solution. Microscopic analysis was conducted within a week of collection. Sampling glasses, sedimentation chambers and Sedgwick Rafter counting slides were used for the enumeration of *N. scintillans* (Guillard 1978, Hasle 1978, Venrick 1978).

The quantitative results of *N. scintillans* and other phytoplankton were calculated as both cell densities and cell volumes because of its huge volumetric capacity and, consequently, the important differences

in its numerical and volumetric contribution to dinoflagellates and total phytoplankton during sampling. On the other hand, volumetric estimates of *N. scintillans* and other phytoplankton were done using the results of new volume measurements at different sampling periods.

3. Results

3.1. Temporal variations in *N. scintillans* and other phytoplankton

The temporal distributions of *N. scintillans*, other dinoflagellates and total phytoplankton cell density are presented in Figure 2, and the bloom colour image of *N. scintillans* responsible for the excessive algal bloom in Dardanelles surface waters in late spring is presented in Figure 3. Other descriptive statistical results relating to the phytoplankton groups are given in Table 1.

While the cell density of *N. scintillans* varied between $0.00\text{E} + 00$ and $2.20\text{E} + 05$ cells L^{-1} , the cell volume varied between $0.00\text{E} + 00$ and $1.31\text{E} + 12$ $\mu\text{m}^3 \text{L}^{-1}$. Phytoplankton cell density varied between $1.00\text{E} + 05$ and $1.84\text{E} + 07$ cells L^{-1} , whereas their cell volume varied between $2.51\text{E} + 09$ and $1.44\text{E} + 12$ $\mu\text{m}^3 \text{L}^{-1}$ owing to excessive algal blooms caused by phytoplankton species with small cell sizes (Table 1).

There were two very excessive bloom periods of *N. scintillans* during the year in the Dardanelles. The first one was between March and June, and the second between October and December (Figure 2). Although cell

Table 1. Descriptive statistics of different data groups in the Dardanelles in the period between March 2001 and January 2004

	Descriptive statistics				
	<i>N</i>	Minimum	Maximum	Mean	Std. Dev.
Temperature [$^{\circ}\text{C}$]	83	6.75	26.00	16.00	6.18
Salinity [ppt]	83	22.28	32.80	26.56	2.06
pH	83	6.61	8.88	8.24	0.36
DO [mg L^{-1}]	83	5.91	12.40	8.80	1.17
$\text{NO}_2^- + \text{NO}_3^-$ [μM]	83	0.05	6.89	0.43	0.93
PO_4^{3-} [μM]	83	0.02	1.15	0.22	0.22
SiO_4 [μM]	83	0.35	7.23	2.49	1.34
Chlorophyll <i>a</i> [$\mu\text{g L}^{-1}$]	83	0.03	15.21	1.77	2.16
<i>N. scintillans</i> [Cell L^{-1}]	83	$0.00\text{E} + 00$	$2.20\text{E} + 05$	$1.59\text{E} + 04$	$3.01\text{E} + 04$
Dinophyceae [Cell L^{-1}]	83	$1.00\text{E} + 04$	$1.84\text{E} + 07$	$9.66\text{E} + 05$	$2.34\text{E} + 06$
Total Phyto [Cell L^{-1}]	83	$1.00\text{E} + 05$	$2.43\text{E} + 08$	$1.53\text{E} + 07$	$3.68\text{E} + 07$
<i>N. scintillans</i> [$\mu\text{m}^3 \text{L}^{-1}$]	83	$0.00\text{E} + 00$	$1.31\text{E} + 12$	$9.81\text{E} + 10$	$1.84\text{E} + 11$
Dinophyceae [$\mu\text{m}^3 \text{L}^{-1}$]	83	$1.50\text{E} + 08$	$1.40\text{E} + 12$	$1.16\text{E} + 11$	$1.94\text{E} + 11$
Total Phyto [$\mu\text{m}^3 \text{L}^{-1}$]	83	$2.51\text{E} + 09$	$1.44\text{E} + 12$	$1.43\text{E} + 11$	$2.14\text{E} + 11$

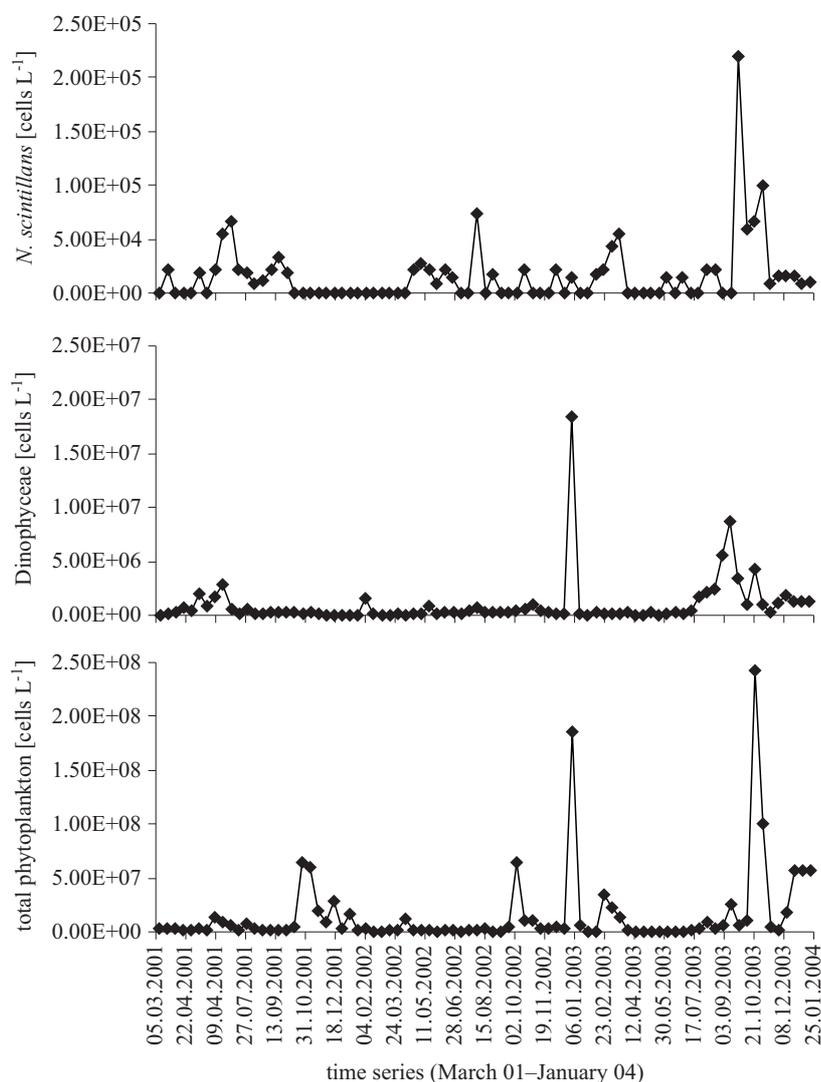


Figure 2. Temporal variations of *Noctiluca scintillans*, dinoflagellates and total phytoplankton cell density in the Dardanelles between March 2001 and January 2004

densities reached maximum values in November and December, the annual production potential in the period from March to June, especially in early May was higher than in the former period. On the other hand, *N. scintillans* did not proliferate significantly between July and October (Figure 2).

Except for early May 2003, there was no colour image in other early May periods from 2001 to 2004. However, May 2003 was the most important

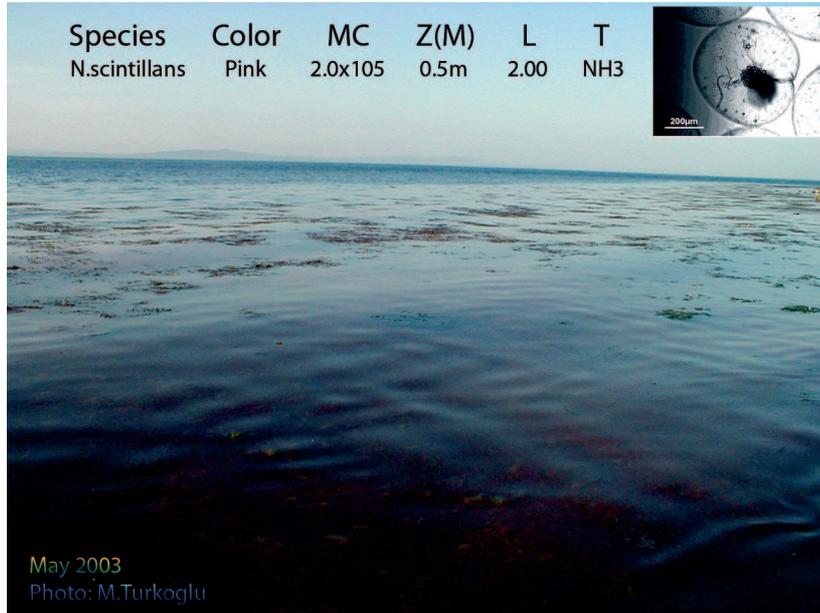


Figure 3. Heterotrophic dinoflagellate species *Noctiluca scintillans* responsible for excessive algal blooms (small picture) and its bloom color image (large picture) in the Lapseki coastal area of the Dardanelles in early summer (MC – maximum cell L^{-1} , Z – depth which shows maximum cell density, L – Lloyd cluster index, T – harmful effect type)

proliferation period ($2.20E + 05$ cells L^{-1}), just before the *E. huxleyi* bloom in the Dardanelles during June–July 2003 (Turkoglu 2008). In May 2003, the level of the algal bloom provoked by *N. scintillans* had a density of $2.20E + 05$ cells L^{-1} and this maximum density was responsible for the dominant orange colour of the Dardanelles surface waters (Figure 3).

3.2. Vertical distribution of *N. scintillans*

The vertical distribution of the *N. scintillans* cell density in excessive bloom periods in the Dardanelles between March 2001 and January 2004 is given in Figure 4.

The vertical distribution of the *N. scintillans* cell density in excessive bloom periods (late spring or May) revealed that excessive cell concentrations in the surface layer decreased with depth. However, in addition to the high density in the surface layer, there were secondary increases in subsurface waters (10–25 m) during the bloom periods, especially in May 2003 (Figure 4).

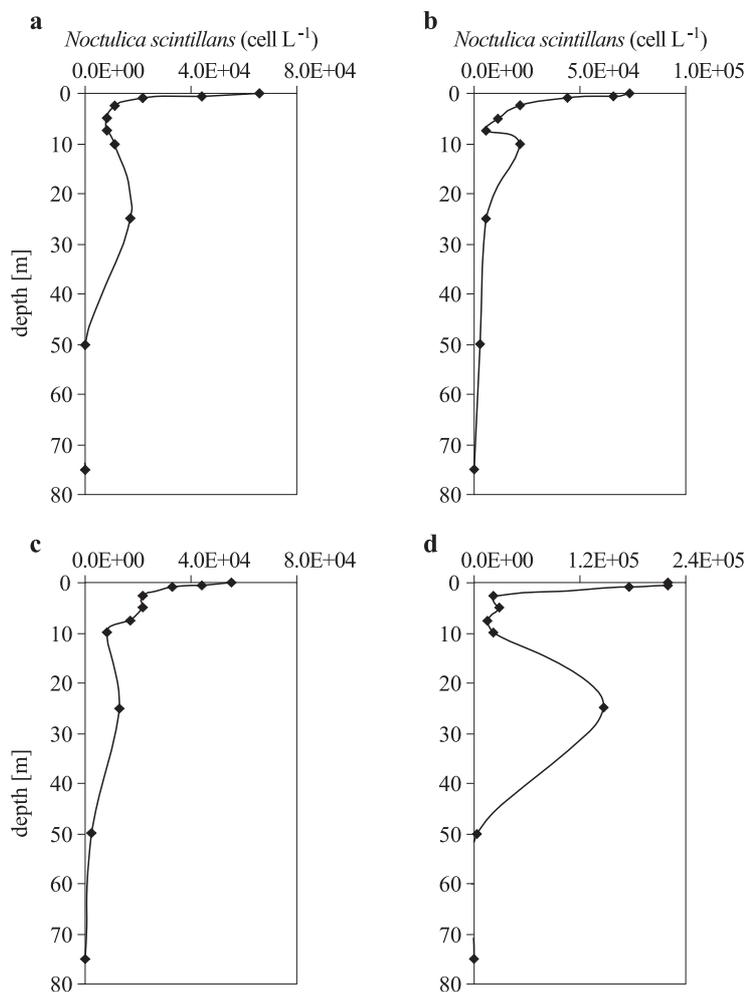


Figure 4. Vertical distribution of *Noctiluca scintillans* cell density in excessive bloom periods (a – 8 May 2001, b – 7 December 2001, c – 17 May 2002, d – 1 May 2003) in the Dardanelles between March 2001 and January 2004

3.3. Contributions of *N. scintillans* to the phytoplankton

Figure 5 shows the temporal distribution of the contributions of *N. scintillans* to Dinophyceae and total phytoplankton in surface waters, while Figure 6 gives the correlation coefficients R among *N. scintillans*, dinoflagellates and total phytoplankton with respect to cell density and cell volume.

The contribution of *N. scintillans* to the total dinoflagellate and phytoplankton cell density was less (0–33.30 and 0–3.70% respectively) than

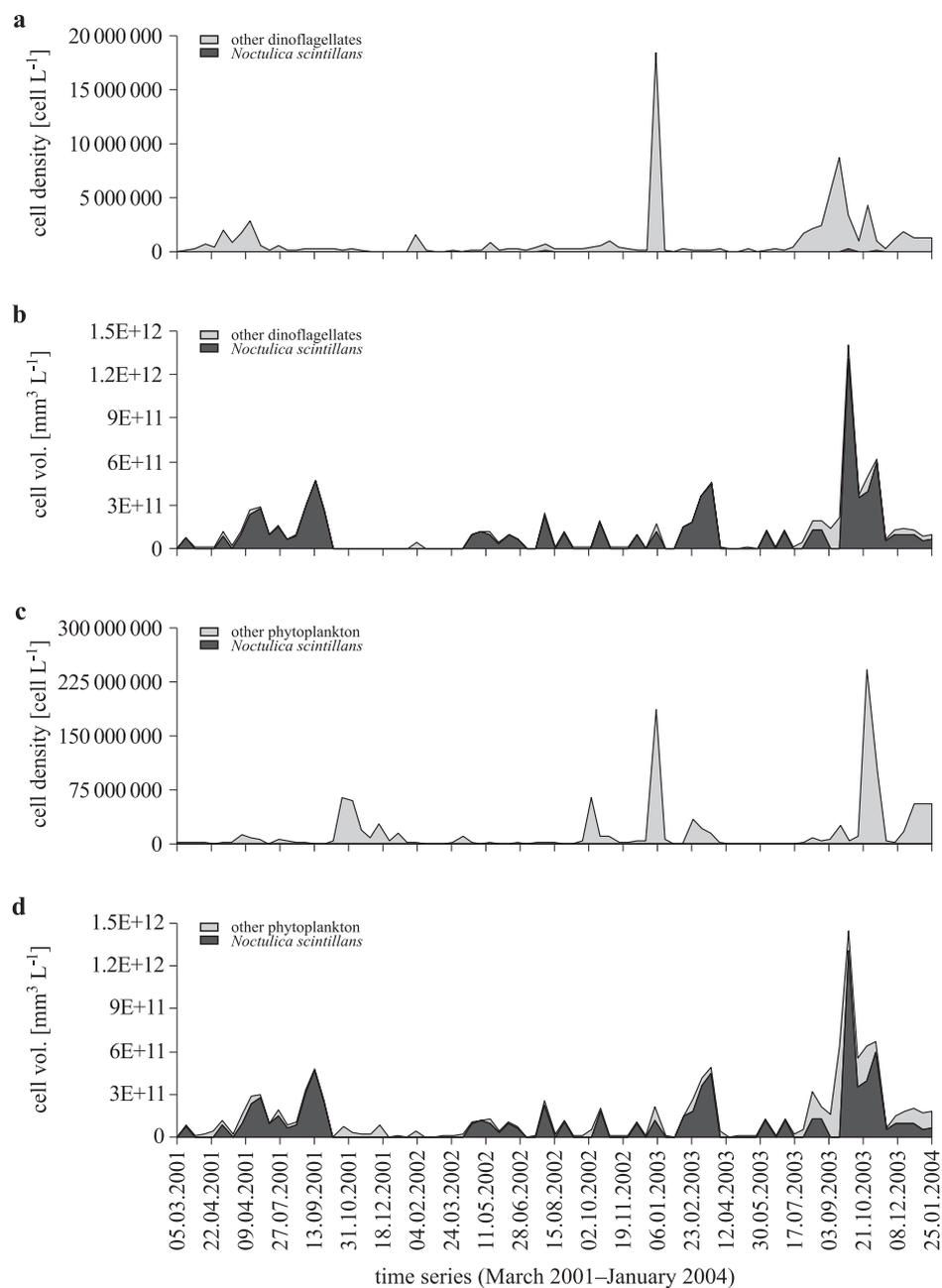


Figure 5. Temporal variations in the contributions of *Noctiluca scintillans* cell density (a) and biovolume (b) to the total dinoflagellate and total phytoplankton cell density (c) and biovolume (d) in the Dardanelles between March 2001 and January 2004

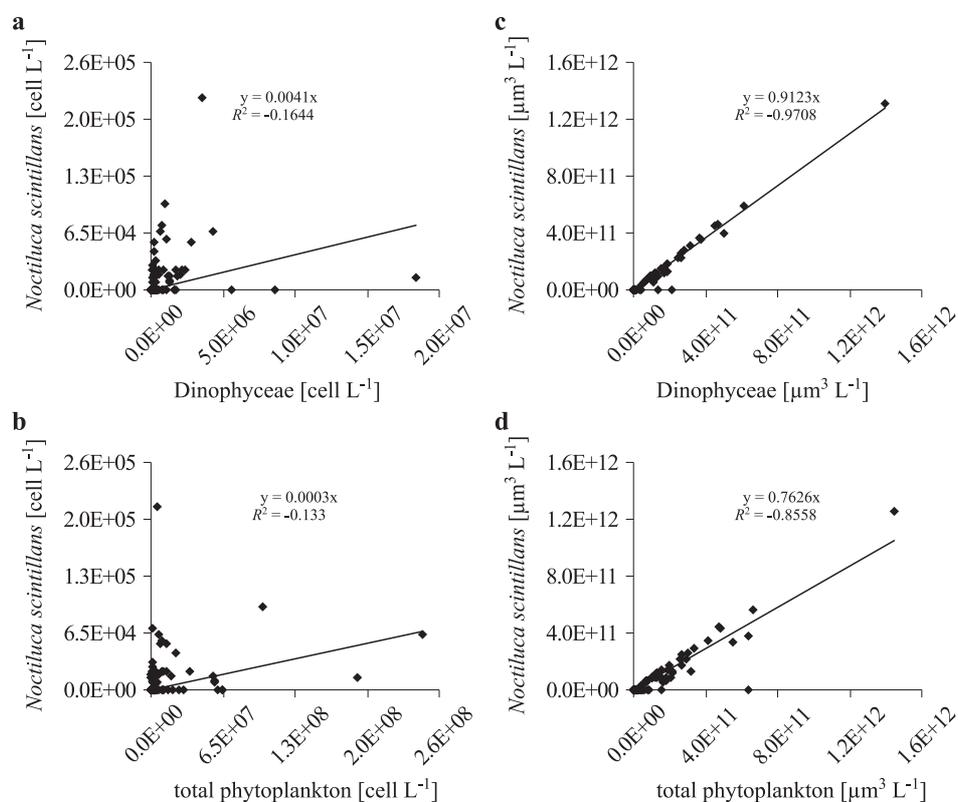


Figure 6. Relationships between *Noctiluca scintillans* cell density and dinoflagellates (a), total phytoplankton cell density (b) and between *N. scintillans* cell volume and dinoflagellates (c), total phytoplankton cell volume (d) in the Dardanelles between March 2001 and January 2004. The coefficients of determination (R^2) and the equating process (y) are shown for each regression

that to the total Dinophyceae and phytoplankton cell volume (0–99.50 and 0–99.00% respectively) (Figure 5). This contribution was endorsed by the correlations obtained. Although there was no significant correlation between *N. scintillans* and dinoflagellates ($R = -0.164$) or between *N. scintillans* and phytoplankton ($R = -0.133$) in respect of cell density, there was an important positive correlation between both dinoflagellates ($R = 0.971$) and total phytoplankton with *N. scintillans* ($R = 0.856$) with respect to cell volume (Figure 6).

3.4. Physical variations and bloom interactions

Vertical distributions of physical parameters such as temperature, salinity, pH and dissolved oxygen (DO) in the excessive bloom periods are

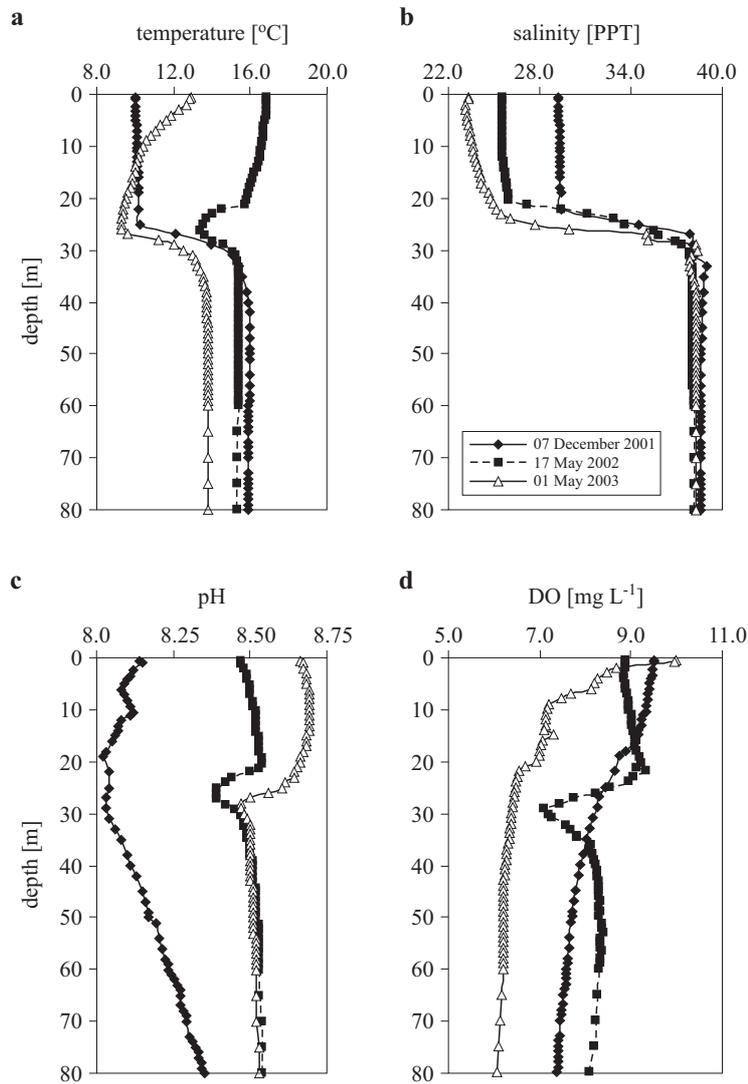


Figure 7. Vertical variations of temperature (a), salinity (b), pH (c) and dissolved oxygen (d) in the excessive bloom periods in the Dardanelles between March 2001 and January 2004

given in Figure 7. The descriptive statistics of the physical parameters are listed in Table 1.

Statistical analyses revealed that surface temperatures varied between 6.75 and 26.00°C (average: $16.00 \pm 6.18^\circ\text{C}$) during the study (Table 1). During two very excessive bloom periods of *N. scintillans*, the temperature was stable and phytoplankton was abundant. This temperature stability

and phytoplankton abundance were among the basic causes of *N. scintillans* blooms in the Dardanelles.

Although temperatures in the upper layer varied between 9.56 and 16.85°C in all the bloom periods, they were above 10.00°C (average 12.63 ± 1.06°C) in the upper surface layer during the largest bloom on 01 May 2003. On the other hand, surface salinities varied between 22.30 and 32.80 ppt (average 26.60 ± 2.06) during the study (Table 1). Because of the two opposing flow systems and the interactions between them, the salinity varied during the year in the Dardanelles. However, salinity variations in the excessive bloom periods of *N. scintillans* (Figure 2 and 6) were more stable than during any other period. Descriptive statistical analyses showed that temporal values of pH and DO varied from 6.61 to 8.88 (average 8.24 ± 0.36) and from 5.91 to 12.40 mg L⁻¹ (average 8.80 ± 1.17 mg L⁻¹) respectively during the study (Table 1).

The vertical profiles of temperature (Figure 7a) and salinity (Figure 7b) suggest that there were two different water masses during the *N. scintillans* bloom periods (Figure 2a). Temperature variations in both the upper layer (9.56–16.85°C) and lower layer (12.96–15.99°C) were more variable than salinity variations during the bloom periods. The latter ranged from 23.10 to 29.38 ppt in the waters of the thin upper layer (0–20 m), but from 37.85 to 38.61 ppt in the much thicker lower layer (30–80 m). Both seasonal thermocline and halocline interfaces were distinct and formed between 20 and 30 m depth during the blooms (Figure 7a,b). However, while pH changed from 8.02 to 8.69 in the upper layer and from 8.04 to 8.53 in the lower layer (Figure 7c), DO values varied between 6.92 and 9.99 in the upper layer and between 6.06 and 8.40 mg L⁻¹ in the lower layer in the important bloom periods (Figure 7d). These highly saturated DO concentrations in the upper layer gradually decreased with depth during the blooms (Figure 7d).

3.5. Chemical variations and bloom interactions

Vertical distributions of nitrite + nitrate, phosphate, silicate and chlorophyll *a* in the excessive bloom periods are given in Figure 8. The descriptive statistics of these chemical variables are listed in Table 1.

Descriptive statistical analyses and temporal variations showed that nitrite + nitrate varied from 0.05 to 6.89 μM (average 0.43 ± 0.93 μM), phosphate from 0.02 to 1.15 μM (average 0.22 ± 0.22 μM) and silicate from 0.35 to 7.23 μM (average 2.49 ± 1.34 μM) (Table 1). Nutrient concentrations, especially nitrite + nitrate and silicate concentrations, increased with depth during the blooms. Therefore, their vertical profiles showed that the concentrations in the upper layer were lower than those in the lower layer during bloom conditions (Figures 8a,c). However, the phosphate profile was

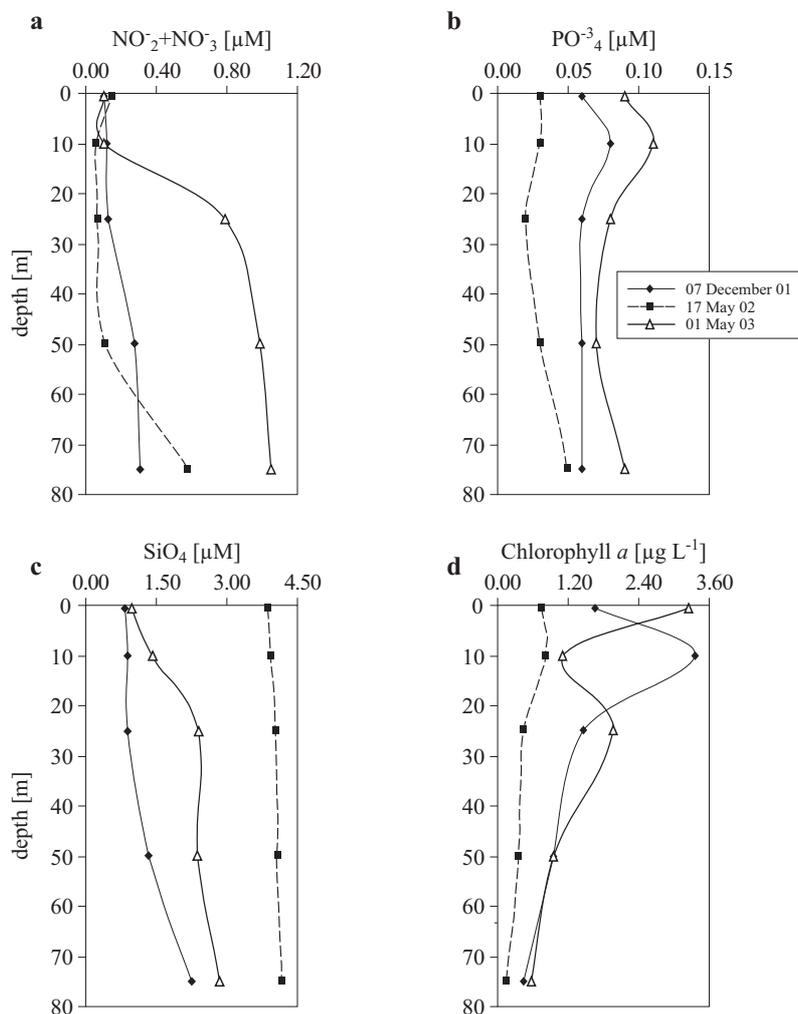


Figure 8. Vertical variations of nitrite + nitrate (a), phosphate (b), silicate (c) and chlorophyll *a* (d) in the excessive bloom periods in the Dardanelles between March 2001 and January 2004

different from the other nutrient profiles and generally decreased with depth just after the peak concentrations in the subsurface layer (10 m). In general, while nitrite + nitrate and silicate reached peak values at 75 m, phosphate reached peak values at 10 m (Figure 8b) during the bloom periods.

Chlorophyll *a* varied between 0.03 and 15.21 $\mu\text{g L}^{-1}$ (average $1.77 \pm 2.16 \mu\text{g L}^{-1}$) in Dardanelles surface water during the study (Table 1). Chlorophyll *a* concentrations ranged from 0.74 to 3.35 $\mu\text{g L}^{-1}$ in the upper layer during the important bloom periods (Figure 8d). The maximum

concentrations of chlorophyll *a* (Figure 8d) and phosphate (Figure 8a) were recorded at about 10 m depth in the bloom periods, except in May 2003, when the chlorophyll *a* maximum was at the surface.

4. Discussion

Carried out over a period of three years, this study has shown that there were both temporal and vertical variations in the abundance of *N. scintillans*. The *N. scintillans* cell density started to increase from early spring (March), reaching a maximum in late spring (May); this was the first proliferation period. However, there was an interruption in proliferation during the summer, especially in late summer. Thereafter the cell density again began to increase in mid-autumn (October), arriving at a maximum in early winter (December) – the second proliferation period. The cell density in May 2003 ($2.20\text{E} + 05 \text{ cell L}^{-1}$) was relatively high compared to the other years (Figure 2a). Annual variations in the abundance of *N. scintillans* in the German Bight over 20 years (Uhlig & Sahling 1990) showed that there was an annual oscillation at three-year intervals in the abundance of *N. scintillans*; these results resembled the findings of the present study. Those authors found that a year with relatively high abundance was followed by two years of relatively low abundance. On the other hand, in the three years from 1990 to 1992 in the coastal waters of Hong Kong, the abundance of *N. scintillans* in 1990 was relatively high compared with that in 1991 and 1992 (Huang & Qi 1997). In the Seto Inland Sea of Japan from 1995 to 1998, the abundance of *N. scintillans* in 1995 and 1998 was relatively high compared to other years (Pithakpol et al. 2000).

Owing to variations in the vertical distribution of bio-physicochemical variables such as temperature, salinity, nutrients and other phytoplankton variables, there were important vertical distributions of *N. scintillans* abundance in the study area. The high concentrations in the surface water decreased with depth in the bloom periods. However, the vertical distributions showed that there was an increasing trend in the density of *N. scintillans* at subsurface depths (10–25 m) in some bloom periods (Figure 4). On the one hand, field studies in the upper Gulf of Thailand and Manila Bay of a red tide of green *N. scintillans* with the photosynthetic *Pedinomonas noctiluca* showed that vertical maximum of *N. scintillans* often occurred below the halocline at 10 to 15 m depths, suggesting that salinity influenced the vertical distribution of this organism (Miyaguchi et al. 2006, Thaithaworn et al. 2012). On the other hand, different locality demands of young and old cells based on feeding and proliferation behaviours generally affect the vertical variations of this heterotrophic species (Thomas & Titelman 1998). Therefore, the vertical distribution

profiles of *N. scintillans* (Figure 4) can reflect different bloom stages and feeding activities. Besides, although blooms of *N. scintillans* have been reported to be correlated with environmental factors, the initial trigger of large-scale bloom formations has not been specifically attributed to any special condition. During large-scale bloom formations in the Dardanelles, it is important wind systems like the poyraz and lodos (strong NE and SW winds respectively), especially predominant in the Aegean Sea, TSS, Black Sea and the Mediterranean Coast of Turkey all the year round, as well as variations in physicochemical variables such as temperature, salinity and nutrients that can initiate and retard phytoplankton blooms in the system. It is known that the lodos wind system carries African dusts rich in bioactive minerals such as Fe and Zn and ultimately generates heavy falls of rain. Although the relationships between trace metals and bloom formations are not very clear, trace metals, particularly bioactive ones, stimulate the growth of some phytoplankton species. Koray et al. (1996) showed that blooms of *N. scintillans* were obviously frequent and widespread when Fe concentrations were high in ambient waters, whereas blooms of this species were markedly inhibited at high Cu levels in Izmir Bay. However, the causes of large-scale bloom formations in such coastal systems are still complex.

In this study, *N. scintillans* blooms were observed as orange patches in the Dardanelles in May–July, when the weather was calm for the first time in the year. However, although *N. scintillans* blooms were also observed in October–December, the second calm weather period of the year, these blooms were not strong enough for the orange patches in the system to become visible. It is known that orange tides of *N. scintillans* were frequently observed in the TSS, including the Dardanelles, during ten years in such calm weather conditions (Unsal et al. 2003, Turkoglu et al. 2004a, Turkoglu & Buyukates 2005, Turkoglu 2010a, Turkoglu & Oner 2010). Excessive blooms like these have occurred in the various gulfs of the Aegean Sea. The blooms taking place in late spring in the Dardanelles, for instance, appeared in the cold periods of the year (February–March) in the Thermaikos Gulf. These massive blooms ($\geq 1.00E + 06$ cell L⁻¹) in the Thermaikos Gulf caused severe water discoloration in the bloom periods in 2000–2004 (Nikolaidis et al. 2005, Ignatiades & Gotsis-Skretas 2010). However, in the sampling period between 1990 and 1992 in Izmir Bay, *N. scintillans* was among the very important bloom species along with *Prorocentrum micans* Ehrenberg, 1834. Very large pink-orange patches of *N. scintillans* were observed between March and June in the Bay (Koray et al. 1996), at the same time as the first bloom period of this species was reported in the Dardanelles.

There was a large contribution of *N. scintillans* to the total dinoflagellate and total phytoplankton cell volume (Figure 5), which was corroborated by the correlations (Figure 6). Besides, extensive blooms of *N. scintillans* suggested that the Dardanelles is one of the most highly eutrophic and turbid aquatic systems in the world. In the Dardanelles and hence in the Sea of Marmara, high concentrations of phytoplankton (Figures 2 and 5), probably due to environmental conditions such as well-mixed nutrient-rich waters (Figure 8) and the seasonal circulation nature of physical factors such as temperature and salinity, encouraged blooms of *N. scintillans*, known as 'red tides' (Figure 3). It is known that the availability of phytoplankton as prey is also one of the important factors for the variation in abundance of *N. scintillans* (Elbrächter & Qi 1998). The correlation between *N. scintillans* and other phytoplankton groups (Figure 6) showed that there was an increase in proliferation levels of *N. scintillans* just after the diatom blooms in spring and autumn. Dela-Cruz et al. (2002) stated that *N. scintillans* blooms generally form after diatom blooms. Therefore, there are generally negative correlations between the abundance of phytoplankton, especially diatoms and *N. scintillans* (Huang & Qi 1997). Huang & Qi (1997) also reported that there was a negative relationship between the abundance of *N. scintillans* and chlorophyll *a* during the bloom events similar to the correlation results of this study. It is known that the contribution of diatoms to the total phytoplankton was particularly high in the period of peak chlorophyll *a* levels in the Dardanelles (Turkoglu 2010a, Turkoglu & Erdogan 2010, Turkoglu & Oner 2010).

Several researchers (Unsal et al. 2003, Turkoglu et al. 2004a, Turkoglu 2010a) reported that there were three diatom bloom periods in the Dardanelles and thus in the Sea of Marmara: the first one appeared from March to May, the second one from mid-July to late August, and the third one from November to January (Figure 5c). Microscopic examinations showed that the diatoms were composed mainly of *Chaetoceros* spp., *Dactyliosolen fragilissimus* (Bergon) G.R. Hasle, 1991, *Cylindrotheca closterium* (Ehrenberg) Reiman and Lewin, 1964, *Proboscia alata* (Brightwell) Sundström, 1986, *Rhizosolenia* spp., and *Thalassiosira* spp. in the study area. These diatoms were often observed in the cell body of *N. scintillans* and considered the main prey item of *N. scintillans*. They may be a factor contributing to the increase in the abundance of *N. scintillans* during the first phase of its bloom in spring. During this period, the cell density may have risen to some extent as a result of an increased growth rate under favourable food conditions. However, neither a positive nor a negative relationship was observed between the

abundance of *N. scintillans* and the chlorophyll *a* concentration in the present study ($R = -0.057$).

The orange colour appearing in the Dardanelles and in the Sea of Marmara is thought to be related to the contents of the food vacuole, which contains some phytoplankton species such as the dinoflagellate *Prorocentrum* spp., diatoms *D. fragilissimus* and *Leptocylindrus* spp., and *Thalassiosira* spp. in the study area. Sweeney (1978) reported that such photosynthetic symbionts were responsible for various colours such as pink, reddish, orange and green in the colour of the cytoplasm. Sometimes, in tropical regions like Indonesia, Malaysia and Thailand, the water was coloured green, owing to the presence of green prasinophyte endosymbionts with *N. scintillans* (Sweeney 1978, Dodge 1982, Fukuyo et al. 1990, Hallegraeff 1991, Taylor 1993, Taylor et al. 1995, Steidinger & Tangen 1996). The food vacuole content and hence its optical characteristics differed from each other because of the different phytoplankton species grazed by *N. scintillans* in different seasons and different ecosystems. Thanks to their optical characteristics, such algal blooms can be monitored by in situ and remote sensing methods in aquatic systems, especially in fisheries and aquaculture areas. Van Moll et al. (2007) found that intense blooms of *N. scintillans* were characterized by a high reflectance of wavelengths > 580 nm and a sharp increase in reflectance from 520 to 580 nm.

It is known that water circulations, temperature and salinity gradients are also the main physical forces affecting the population dynamics of many microzooplankton such as *N. scintillans* (Redden et al. 2009). Such mixing processes affect other bio-chemical processes such as the transport of nutrients and resuspensions of particulate organic matter (Redden et al. 2009). The study area, which is a part of the TSS, continuously receives less saline and also more polluted waters from Black Sea surface waters along with some other fresh waters from river inputs and underground waters (Turkoglu et al. 2006, Baba et al. 2007). Although the growth rate of *N. scintillans* was generally affected by temperature and salinity, it is known to be a eurythermal and euryhaline organism (Elbrächter & Qi 1998). Many previous research results showed that the optimum temperature and salinity demands of *N. scintillans* are different in each ecosystem (Huang & Qi 1997, Tada et al. 2004, Miyaguchi et al. 2006). Therefore, it is difficult to discover the effect of temperature and salinity alone on the bloom formation of *N. scintillans*. Despite the large-scale temperature and salinity variations (temperature – 10.00–16.85°C; salinity – 23.12–29.38 ppt) in the upper layer waters (0–20 m) of the Dardanelles in different bloom periods of *N. scintillans*, their variations were almost stable during each such period (Figures 7a,b). The optimum temperature and salinity

requirements for *N. scintillans* change not only in different ecosystems, but during different periods of time in the same ecosystem as well. It is known that the optimum temperature for *N. scintillans* growth is ca 20.0°C (Rissik & Suthers 2009). Various studies carried out in very different areas have shown that the temperature and salinity demands of *N. scintillans* are substantially different: temperature – 10–28°C, and salinity – 28–36 ppt (Huang & Qi 1997, Tada et al. 2004, Ignatiades & Gotsis-Skretas 2010). For instance, in Dapeng Bay (South China Sea), *N. scintillans* developed at temperatures from 15.8 to 28.6°C, and the highest population densities were found at temperatures between 19 and 25°C from March 1990 to June 1992. On the other hand, in Sagami Bay, *N. scintillans* blooms appear to prefer a lower temperature and a higher salinity compared to those found in previous studies (Miyaguchi et al. 2006). Huang & Qi (1997) and Tada et al. (2004) reported that there were important correlations between the high abundance of *N. scintillans* and temperature. However, temperature and salinity were not significantly correlated with the variation in abundance of *N. scintillans* in the present study, similar to correlations found in Sagami Bay of Japan (Miyaguchi et al. 2006).

In the light of the above information, we may state on the one hand that *N. scintillans* tolerates lower temperature levels due to the fact that the study area is a hypereutrophic system. On the other hand, the warming of coastal waters at a continually increasing rate due to global warming has caused an increase in the densities of opportunist and thermophilic dinoflagellate species such as *N. scintillans* in such eutrophic systems. However, there was relationship between *N. scintillans* blooms and some phytoplankton blooms in the Dardanelles, probably due to feeding behaviour of *N. scintillans*. Though classified among the dinoflagellates, a phytoplankton group, *N. scintillans* is known to have no photosynthetic pigments and to feed on other phytoplankton, small zooplankton and their eggs (Rissik & Suthers 2009).

The pH varied considerably, ranging from a minimum of 8.02 to a maximum of 8.69 in surface waters during the study. It is known that pH values were typically higher during bloom periods than in other sampling periods (Koray et al. 1996). However, there was neither a positive nor a negative significant correlation between pH and phytoplankton abundance in the present study ($R = -0.013$). On the other hand, there were very significant correlations (at the 0.05 level; $R = 0.225$) between DO and *N. scintillans* along with other phytoplankton. It is known that physical variables such as DO and pH are controlled by biological processes such as phytoplankton and micro-zooplankton blooms as well as other

physicochemical processes in surface waters in the system (Turkoglu et al. 2004a, Turkoglu 2005, 2010a, 2010b, 2010c, Turkoglu & Oner 2010).

Nutrient concentrations, except for PO_4^{-3} in surface layer waters, were lower than in deeper waters (Figure 8a-c) owing to the probably high phytoplankton assimilation. Therefore, although the surface waters of the Dardanelles originated from Black Sea surface waters, which generally have high nutrient levels, the concentrations in Black Sea surface waters in the Dardanelles decrease considerably in some periods, especially during bloom periods. On the other hand, the high nutrient concentrations in the Sea of Marmara due to the large nutrient inputs from both the Black Sea via the Bosphorus and from land-based sources in the Istanbul metropolis (Polat & Tugrul 1995, Tugrul & Polat 1995, Polat et al. 1998) decrease considerably as far as the Dardanelles during the Marmara surface flow (Basturk et al. 1990, Polat & Tugrul 1995). So nutrient levels, especially nitrite + nitrate and silicate concentrations, were lower in the upper layer waters than in the lower layer waters. However, the phosphate level was higher in the upper layer waters than in the lower layer waters (Figure 8b) owing to domestic waste water discharges rich in phosphate. Otherwise, the system is known to have very low elemental N:P ratios (3.21 ± 2.70), as given by the Redfield ratios in the surface waters of the Dardanelles (Turkoglu et al. 2004b, Turkoglu 2008, 2010b,c). Hence, nitrate generally limits algal bloom formations to a greater extent than phosphate in the Dardanelles as a result of the high phosphate input to the system.

High chlorophyll *a* concentrations (Table 1) in Dardanelles surface water during the study showed that there is a hypereutrophication syndrome in the system. In general, chlorophyll *a* maxima were observed at sub-surface depths in the bloom periods (Figure 8d). However, in the May 2003 bloom period, the chlorophyll *a* maximum was at the surface. It is known that this depth (25 m) is just above the lines of the upper thermocline and halocline in the Dardanelles (Turkoglu et al. 2004a, Turkoglu 2008). Although the temperature boundary area (25 m) in the Dardanelles does not have sufficient light for phytoplankton growth, this area usually has the best nutrient conditions and thereby sub-surface (10–25 m) chlorophyll *a* maxima are frequent, especially in summer (Unsal et al. 2003, Turkoglu et al. 2004a).

Consequently, the bio-physicochemical variables indicate that the Dardanelles were severely eutrophic due to the fact that it is a part of the TSS affected by the Black Sea, especially by its more polluted north-western part. The opportunistic *N. scintillans* blooms during the study clearly demonstrated that there were some fundamental disturbances in the Sea of Marmara, especially the Dardanelles part, as also in the north-western

part of the Black Sea (Porumb 1992). On the other hand, very excessive blooms of *N. scintillans* in the Dardanelles gave rise to some unpleasant consequences such as the appearance of gelatinous water and changes in water colour in some recreational swimming areas during late spring and early summer.

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