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Changes in phytoplankton composition of the Golden Horn Estuary (Sea of Marmara) following remediation

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Abstract

Changes in phytoplankton composition of the Golden Horn Estuary were investigated following remediation, through seawater transfer from the Strait of Istanbul to the estuary. Average values of Secchi depth, salinity and dissolved oxygen increased during this study when compared with a previous study. The average number of species (S) and species diversity (H′) increased and they correlated positively with Secchi depth, salinity and dissolved oxygen. There was a similar phytoplankton group composition between this and a previous study, however, the species composition differed. A total of 127 taxa consisting of diatoms (66 taxa), dinoflagellates (49 taxa) and others (12 taxa) were identified. Abundance of dinoflagellates and phytoflagellates and their relative contribution to the total phytoplankton abundance increased during this study, however, the abundance of diatoms and their relative contribution decreased notably, as compared with the previous study. There was a significant positive correlation between salinity and dinoflagellates and phytoflagellates ($P < 0.01$), and also between Secchi depth and dinoflagellates ($P < 0.01$) in the upper estuary. Additionally, salinity and Secchi depth correlated positively with species diversity (H') and number of species (S) ($P < 0.01$). The increase in water transparency probably contributed to the increase in abundance of dinoflagellates and phytoflagellates. The results revealed that water transparency was one of the most important factors affecting phytoplankton composition in the study area. Changes in some environmental conditions following seawater transfer appear to have changed phytoplankton composition. As a result, phytoplankton species were confirmed as a very good indicator of changed environmental conditions.

Introduction

Phytoplankton are easily detectable indicators of ecological change, because they are sensitive to changing environmental conditions (Paerl *et al.*, 2007). Estuaries are a transition zone between freshwater environments (i.e. rivers or streams) and marine environments and are known to be highly productive ecosystems, often being nutrient-rich. Phytoplankton growth may be very low in river-dominated estuaries due to the high turbidity caused by river inputs. Light availability plays a fundamental role for phytoplankton growth in estuaries with high turbidity (Cloern, 1987, 1996; Barbosa *et al.*, 2010; Domingues *et al.*, 2010) especially in the middle and upper estuary (Domingues *et al.*, 2012). It is well known that in these systems light and nutrient availability are mainly controlled by river flow (Domingues *et al.*, 2011, 2012). Studies performed around the Mediterranean Sea revealed that estuaries are generally stratified in their middle and lower sections and well-mixed in the upper section (Trigueros & Orive, 2001; Cetinić *et al.*, 2006; Burić *et al.*, 2007; Lopes *et al.*, 2007; Barbosa *et al.*, 2010; Domingues *et al.*, 2012).

The rehabilitation project of the Golden Horn Estuary (GHE) was conducted by the Municipality of Istanbul between 1997 and 2000, in order to remove heavy pollution and improve water quality. During this rehabilitation project, surface discharges were gradually controlled, connected to collector systems, and then discharged into the lower layer of the Strait of Istanbul through two deep discharge systems; 4.25×10^6 m^3 of anoxic sediment from the upper part of the estuary was removed and the Valide Sultan Bridge floating on pontoons was partially opened. These processes resulted in rapid renewal and oxygenation of anoxic and highly polluted waters (Okus *et al.*, 2001; Tas *et al.*, 2009).

The depth of the upper part of the estuary declined to 2 m due to heavy sedimentation and water quality deteriorated rapidly from 2006. However, improvement of the ecosystem has become mandatory and to this purpose, a project was implemented by the General Directorate of Istanbul Water and Sewerage Administration of the Istanbul Municipality in October 2012. This new project included the transfer of seawater from the Strait of Istanbul with a pumping system via the Kagithane Streams into the GHE. Within the scope of this project, 260,000 m^3 day^{-1} from the top layer of highly oxygenated Black Sea water from the Strait of Istanbul was targeted to be pumped into the GHE. This pumping operation was interrupted during rainy months due to the risk of the Kagithane Stream overflowing. An average of 119,500 tons of seawater per day were transferred to the GHE during this study period.

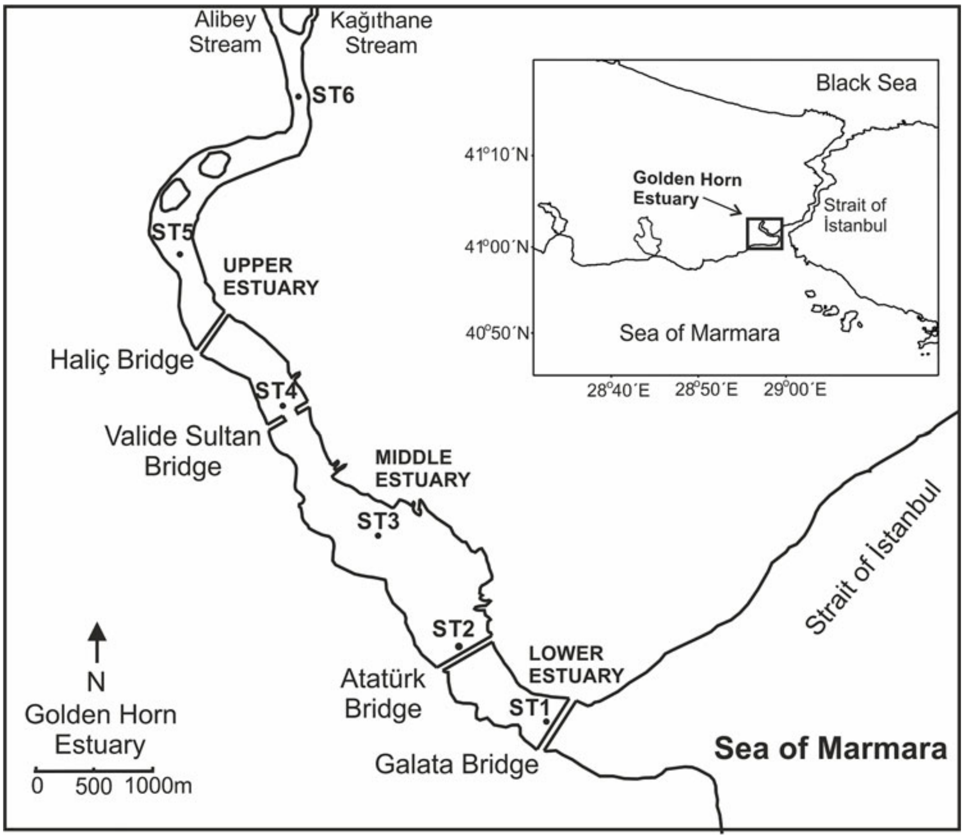

Fig. 1. Study area and sampling stations.

The highest average amount of seawater (220,500 tons day^{-1}) was pumped in October 2013, while the lowest (84,200 tons day^{-1}) was pumped in September 2014 (Koyuncu, 2018).

Previous studies performed in the GHE demonstrated that extreme pollution limited phytoplankton (Uysal & Unsal, 1996; Tas & Okus, 2003; Tas *et al.*, 2006). Improved water quality resulted in a rapid renewal and oxygenation of anoxic and highly polluted water, and this was followed by consecutive blooms of phytoplankton (Tas *et al.*, 2009). Recently, potentially harmful microalgae and algal blooms (Tas, 2015; Tas & Yilmaz, 2015), and diatom composition in the GHE were investigated (Tas, 2017).

The aim of this study is to explain changes in phytoplankton composition following seawater transfer to the GHE through the Kagithane Stream. This study was designed to test two hypotheses. The first hypothesis is that seawater transfer to the GHE would improve water quality and change some environmental conditions (e.g. increase in water transparency, dissolved oxygen and salinity) particularly in the upper estuary. The second hypothesis is that such improvement in environmental factors would increase phytoplankton abundance and diversity.

Materials and methods

Study area and sampling strategy

The Golden Horn, a ria-type (drowned river valley) estuary (Irvalı & Çağatay, 2008), is located south-west of the Strait of Istanbul, extending in a north-west–south-east direction, 7.5 km long and up to 0.7 km wide, with an area of 2.5 km^2. The study area was separated into three hydrographic sections (Figure 1): the lower estuary (LE) with a depth of 40 m, decreasing rapidly to 14 m in the middle estuary (ME) and to 5 m in the upper estuary (UE). The LE is characterized by a two-layered water mass. The upper layer (0–15 m) is the Black Sea-originated less saline waters (S = 18–21), and following a transition zone at 15–25 m depth, Mediterranean-originated saline waters (S = 38) constituted the lower layer (from 25 m to the bottom) (Ergin, 1990). At present, rainfall is the main source of freshwater input to the GHE (Sur *et al.*, 2002).

Water samples were taken biweekly (autumn–winter) and weekly (spring–summer) intervals from three depths (0.5, 2.5 and 5 m) at six sampling stations using 5-litre Niskin bottles. The hydrographic sections (LE, ME and UE) were represented by sampling stations ST1, ST3 and ST5, respectively (Figure 1). Temperature, salinity, pH and dissolved oxygen (DO) were measured by a multi-parameter probe (YSI Professional Pro Plus, USA). Water transparency was measured using a Secchi disc. Chlorophyll *a* (Chl-*a*) analyses were carried out by an acetone extraction method according to Parsons *et al.* (1984). Inorganic nutrient concentrations were estimated using an auto-analyser (Bran + Luebbe AA3 Auto-Analyser, USA) according to standard methods (APHA, 1999). Data of environmental variables belonging to the previous study performed between 2009 and 2010 were taken from Tas & Yilmaz (2015).

Phytoplankton analysis

Water samples (250 ml) for phytoplankton quantitative analysis were preserved with acidic Lugol's solution (2%) (Throndsen, 1978). Sub-samples (10, 25 or 50 ml) were left to settle in Utermöhl sedimentation chambers (Utermöhl, 1958). The settling chamber was examined under an inverted light microscope (Leica DM IL LED) equipped with phase contrast optics at the magnification required. Cells in size 2–20 µm (nanoplankton) were counted at magnification of 400×, while cells in size >20 µm (microplankton) were counted at magnification of 100× or 200×. At least 300 cells in two or more transects in the sedimentation chamber were counted and then cell density per litre was calculated. All data were averaged over the three depths.

Net samples for phytoplankton qualitative analysis were collected using a Nansen plankton net (57 cm diameter, 55 µm

Table 1. Comparison of mean annual values in environmental variables in this study and the previous study

Sections	Period	Temp. (°C)	Salinity	Secchi (m)	pH	DO	Chl-*a*	DIN	PO_4	SiO_2	N:P
LE	2009–2010	17.1	17.8	5.5	8.2	8.1	3.5	9.2	0.39	6.2	26.2
	This study	16.4	**18.4**	**6.6**	8.0	8.1	2.8	8.9	0.42	2.6	12.5
ME	2009–2010	17.7	17.3	2.4	8.1	7.7	10.4	14.9	0.99	10.0	24.1
	This study	16.9	**17.9**	**2.8**	7.8	7.2	**10.7**	**20.4**	**1.15**	4.3	15.4
UE	2009–2010	19.3	15.2	0.5	7.5	3.0	34.6	48.5	3.54	37.1	35.3
	This study	17.7	**16.4**	**1.6**	7.4	**3.4**	22.4	42.8	3.40	11.2	18.4

Bold numbers indicate values increased in this study compared with the previous study. Units for dissolved oxygen (DO), chlorophyll *a* (Chl-*a*) and nutrients are 'mg l^{-1}', 'µg l^{-1}', 'µM', respectively.

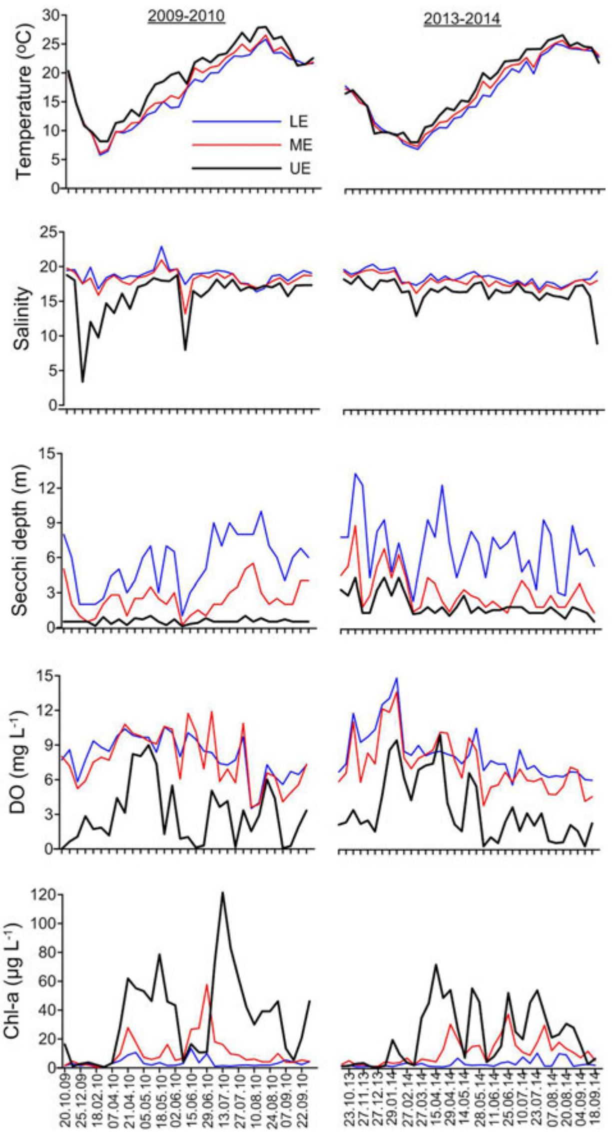

Fig. 2. Fluctuations in some environmental factors during this study and the previous study.

mesh size) (Hydro-Bios) by vertical tows (Tangen, 1978) at the stations ST1, ST2 and ST3. Then, samples were transferred to 250 ml opaque PVC jars and preserved with neutral formaldehyde (4%). A few drops of water from net samples were examined under a light microscope (Leica DM 2500 LM) at the magnifications required (100× to 400×). The following references were used for species identification: Cupp (1943), Delgado & Fortuna (1991), Dodge (1985), Drebes (1974) and Tomas (1997). For recent changes in taxonomic classification the WoRMs (World Register of Marine Species) database (http://www.marinespecies.org) was used. Definition of potentially toxic species was made according to the IOC Taxonomic Reference List of Toxic Plankton Algae (Moestrup *et al*., 2009).

Spearman rank correlation coefficients were used to analyse the relationships between the phytoplankton abundance and environmental parameters using the SPSS program. Shannon diversity index (H') was used to calculate species diversity in the phytoplankton community.

Results

Physical and chemical variables

Water temperature displayed a seasonal pattern and increased slightly from the LE to the UE. Mean annual temperatures were lower in this study (16.4–17.7 °C) than in the previous study (17.1–19.3 °C) at the LE and UE, respectively (Table 1, Figure 2). There was a typical decreasing salinity from the LE to the UE. Mean salinity values increased in this study, when compared with the previous study. Higher salinity was notable in the UE (15.2–16.4) in this study compared with the previous study (Table 1, Figure 2). Secchi depths decreased considerably from the LE to the UE in both studies, and they were higher in this study than in the previous study. Mean Secchi depths in the LE and UE were 6.6 and 1.6 m in this study, while they were 5.5 and 0.5 m in the previous study. The increase in the Secchi depth was 3-fold higher in the UE in this study than in the previous study (Table 1, Figure 2). Water transparency based on Secchi depth dropped considerably particularly in the upper estuary during rainy days.

Mean pH values were generally higher at the LE (8.0) than the UE (7.4) and they were slightly lower in this study than in the previous study (Table 1). DO values decreased from the LE to the UE. Mean DO values varied between 8.1 and 3.4 mg l^{-1} at the LE and UE during this study, while they were 8.1 and 3.0 mg l^{-1} in the previous study. They increased slightly in the UE during this study (Table 1, Figure 2). Chl-*a* values increased considerably from the LE to the UE due to higher phytoplankton abundance in both studies. Mean Chl-*a* values in the LE and the UE were 2.8 and 22.4 µg l^{-1} during this study, while they were 3.5 and 34.6 µg l^{-1} in the previous study (Table 1, Figure 2). Nutrient concentrations increased gradually from the LE to the UE in both studies. The most remarkable decrease was observed in SiO_2 values in this study which were 3-fold lower in this study compared with the previous study (Table 1). The average N:P ratio decreased nearly two-fold during this study compared with the previous study and it remained under the Redfield ratio (16:1) in the LE and ME (Table 1).

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Table 2. Comparison of phytoplankton group composition in this study and the previous study

Sections	Period	*N*-Dia	*N*-Dino	*N*-Pfla	*N*-Total	*C*-Dia	*C*-Dino	*C*-Pfla	*S*	*H′*
LE	2009–2010	1428	6.8	46.5	1481	96.4	0.5	3.1	8.4	1.29
	This study	159	**14.3**	43.5	217	73.3	**6.6**	**20.1**	**26.5**	**1.64**
ME	2009–2010	3486	29.7	425	3941	88.5	0.8	10.7	6.4	1.02
	This study	1046	**191.5**	**1024**	2261	46.3	**8.5**	**45.2**	**22.5**	**1.18**
UE	2009–2010	1350	50.6	1190	2591	52.1	2.0	45.9	2.3	0.45
	This study	1115	**865**	**2834**	**4815**	23.2	**18.0**	**58.8**	**7.1**	**0.63**

N-Dia, *N*-Dino, *N*-Pfla, *N*-Total: Abundance (10^3 cells l^{-1}) of diatom, dinoflagellate, phytoflagellate and total phytoplankton, respectively. *C*-Dia, *C*-Dino, *C*-Pfla: Relative contribution (%) of diatoms, dinoflagellates or phytoflagellates to the total phytoplankton, respectively. *S*, Number of species; *H′*, Shannon diversity.
Bold numbers indicate values increased in this study compared with the previous study.

Phytoplankton community

A checklist of phytoplankton taxa identified in the study area during this study period and a comparison with the taxa detected in the previous study is given in Appendix I as Supplementary Material. A comparison of phytoplankton composition between this and the previous study is given in Table 2. A total of 127 taxa belonging to nine classes were identified during this study. The distribution of these taxa by groups is as follows: diatoms, 66 taxa (52%); dinoflagellates, 49 taxa (38.5%); phytoflagellates, 12 taxa (9.5%). There was a similar group composition between this and the previous study, however, the species composition differed. Twenty-three different taxa (14 diatoms, 7 dinoflagellates and 2 others) were observed only during this study period, while 82% of taxa in this study were the same as the previous study. The diatom *Chaetoceros* and heterotrophic dinoflagellate *Protoperidinium* were the most abundant genera in the phytoplankton community. The number of species was generally higher between April and June.

The increase in total phytoplankton abundance (*N*-Total) began from April and it lasted until August including dense phytoplankton blooms. *N*-Total increased 2-fold at the UE during this study, when compared with the previous study. *N*-Total was correlated positively with salinity, pH and DO ($P < 0.01$), and negatively with temperature and Secchi depth at the LE (Table 3).

Number of species (*S*) and the Shannon diversity index (*H′*) decreased generally from the LE to the UE. Mean *S* and *H′* increased substantially in this study, when compared with the previous study. *S* was 3-fold higher at the UE during this study than in the previous study, while *H′* increased 40% (Figure 3, Table 2). Spearman rank correlation coefficients (rho) were usually higher and more significant in the UE than in other parts. Temperature was the most important factor affecting *S* and *H′*. There was a significant negative correlation between temperature and *S* ($P < 0.01$), but a positive correlation with *H′* ($P < 0.05$). Variations in *S* and *H′* were well correlated with the increase in salinity, Secchi depth and DO (Table 3).

Diatom composition

A total of 66 diatom taxa in 34 genera were identified during this study period. The number of taxa identified in the previous study was 65. Diatoms comprised about 52% of the total number of species. The genus *Chaetoceros* constituted 32% of the diatom assemblage with 21 taxa. Fourteen additional diatom species, not observed in the previous study, were observed in this study. Two diatom species (*Pseudo-nitzschia calliantha* and *P. pungens*) are known as potentially toxic (Appendix I).

The contribution of diatoms to the total abundance of phytoplankton (*C*-Dia) was lower in this study than in the previous study (Table 2, Figure 4). Diatom abundance (*N*-Dia) decreased during this study when compared with the previous study (Table 2) and they were more common and in higher abundances particularly in the ME (Figure 5). The highest diatom abundance detected in this study period was 24.3×10^6 cells l^{-1} (Figure 5). The bloom-forming diatom species were *Cylindrotheca closterium* (2×10^6 cells l^{-1}, July 2014), *Pseudo-nitzschia calliantha* (1.6×10^6 cells l^{-1}, May 2014), *Skeletonema marinoi* (39×10^6 cells l^{-1}, June 2014) and *Thalassiosira minima* (13×10^6 cells l^{-1}, July 2014). Most of these species formed blooms during the previous study, as well.

Diatom species were found in a wide range of water temperatures from 5.7–27 °C and at salinities from 11.6–21.7. In general, their abundance was higher between May and July during this study period. However, *Pseudo-nitzschia* species were more abundant at temperatures below 10 °C. Diatoms formed blooms from late May to June (Figure 5). They were correlated positively with salinity, pH and DO ($P < 0.01$) (Table 3) and were generally scarce in the UE, when the Secchi depth was very low.

Dinoflagellate composition

A total of 49 dinoflagellate taxa in 19 genera were identified during this study. The number of taxa identified in the previous study was 52. Dinoflagellates comprised about 38.5% of the total number of species. The genus *Protoperidinium* represented 43% of the dinoflagellates comprising 21 taxa. Seven additional dinoflagellate taxa, not observed in the previous study, were observed in this study. 10 dinoflagellate species (*Dinophysis acuminata*, *D. acuta*, *D. caudata*, *D. fortii*, *D. tripos*, *Gonyaulax spinifera*, *Lingulodinium polyedra*, *Phalacroma rotundatum*, *Prorocentrum cordatum* and *Protoceratium reticulatum*) are known as the potentially toxic (Appendix I).

The contribution of dinoflagellate abundance to the total abundance of phytoplankton (*C*-Dino) increased in this study relative to the previous study (Table 2, Figure 4). The results showed that the contribution of dinoflagellates was highest in November and December, February April, and August, particularly in the ME and UE (Figure 4). The dinoflagellate abundance (*N*-Dino) value increased considerably during this study, when compared to the previous study. They displayed a seasonal pattern with highest abundances between April and July particularly in the ME and UE (Table 2). *N*-Dino was usually lower than 10^4 cells l^{-1} in autumn and winter. The highest *N*-Dino was 17.4×10^6 cells l^{-1} during this study, while it was 7.8×10^5 cells l^{-1} during the previous study (Figure 6). The bloom-forming dinoflagellate species were *Heterocapsa triquetra* (max. 21.8×10^6 cells l^{-1}, April 2014), and *Scrippsiella acuminata* (max. 1.8×10^6 cells l^{-1}, July 2014) and they caused water discolouration in the UE during blooms. Only *S. trochoidea* among these species formed blooms in the previous study.

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Table 3. Spearman rank correlation coefficients (*rho*) between some physical and chemical factors and the abundance of diatoms (*N*-Dia), dinoflagellates (*N*-Dino), phytoflagellates (*N*-Pfla), the number of species (*S*) and Shannon diversity index (*H′*)

Parameters		All stations	LE	ME	UE
Temperature	*N*-Dia	–	–	–	–
	N-Dino	–	–	–	–
	N-Pfla	–	–	–	–
	S	−0.248**	–	−0.254*	−0.291*
	H'	0.138*	0.323**	–	–
Salinity	*N*-Dia	0.184**	–	–	–
	N-Dino	0.176*	–	–	0.345**
	N-Pfla	–	–	–	0.327**
	S	0.670**	0.418**	0.567**	0.505**
	H'	0.451**	–	0.323**	0.304*
Secchi depth	*N*-Dia	–	−0.307*	–	–
	N-Dino	–	–	–	0.416**
	N-Pfla	−0.146*	–	–	–
	S	0.648**	–	0.391**	0.566**
	H'	0.570**	0.397**	0.383**	0.331**
pH	*N*-Dia	0.192**	–	–	–
	N-Dino	0.336**	–	0.606**	0.319**
	N-Pfla	0.159*	0.238*	0.420**	0.391**
	S	0.300**	–	–	–
	H'	0.261**	–	–	–
DO	*N*-Dia	0.269**	–	0.268*	–
	N-Dino	0.253**	–	–	0.443**
	N-Pfla	–	0.269*	0.253*	0.390**
	S	0.483**	–	–	0.372**
	H′	0.269**	−0.246*	–	–

Statistically significant correlations are indicated by symbols: – not significant; *$P < 0.05$; **$P < 0.01$.

Dinoflagellate species were found in the temperature range from 5.8–28 °C and salinity from 12.9–21.7 during the study period. They were more abundant between April and August and they reached the highest abundance during April in the ME and UE (Figure 6). Dinoflagellate abundances decreased at temperatures below 10 °C and salinities below 10. They were correlated positively with salinity, Secchi depth, pH and DO ($P < 0.01$) (Table 3). A considerable increase in dinoflagellates were observed in higher Secchi depths in the UE during spring and summer (Figure 6).

Phytoflagellate composition

Twelve phytoflagellate taxa belonging to seven classes were identified during this study period. One raphidophyte species, *Heterosigma akashiwo*, is known as potentially toxic (Appendix I). Phytoflagellate species comprised about 9.5% of the total number of phytoplankton in this study.

Phytoflagellate abundance (*N*-Pfla) and their contribution (*C*-Pfla) to the total abundance of phytoplankton was higher in April and July like the dinoflagellates (Table 2, Figures 4 and 7). Their abundance (*N*-Pfla) was more than 2-fold higher in the ME and UE during this study. The most abundant phytoflagellate species was *Heterosigma akashiwo* (31.2×10^6 cells l^{-1}, June 2014), *Plagioselmis prolonga* (62.4×10^6 cells l^{-1}, May 2014), *Pyramimonas grossi* (1.3×10^6 cells l^{-1}, July 2010) and *Eutreptiella marina* (30.2×10^6 cells l^{-1}, April 2014) causing water discolouration during dense blooms in the ME and UE. The same species formed dense blooms in the previous study as well. Phytoflagellate species were found in a wide range of water temperatures from 10.6–28 °C and salinities from 13.3–21.7 in this study period. The highest abundances of phytoflagellates were observed in mid-April at a temperature of 17 °C and salinity of 17.5. There was a significantly positive correlation between phytoflagellate abundance and salinity, pH and DO ($P < 0.01$) (Table 3).

Discussion

Phytoplankton studies carried out in estuaries worldwide indicate that the role of environmental forcing factors such as temperature, salinity, water transparency and nutrients are very important in variation of phytoplankton composition (Cetinić *et al.*, 2006; Burić *et al.*, 2007; Barbosa *et al.*, 2010; Jasprica *et al.*, 2012). The temporal variability of phytoplankton biomass in nutrient-rich estuaries is also related to variations in light availability (Cloern, 1987, 1999; Mallin *et al.*, 1999). Light intensity in the mixed layer is a major factor affecting phytoplankton interannual variability (Barbosa *et al.*, 2010). Light and nutrient availability are generally controlled by river flow (Domingues *et al.*, 2012).

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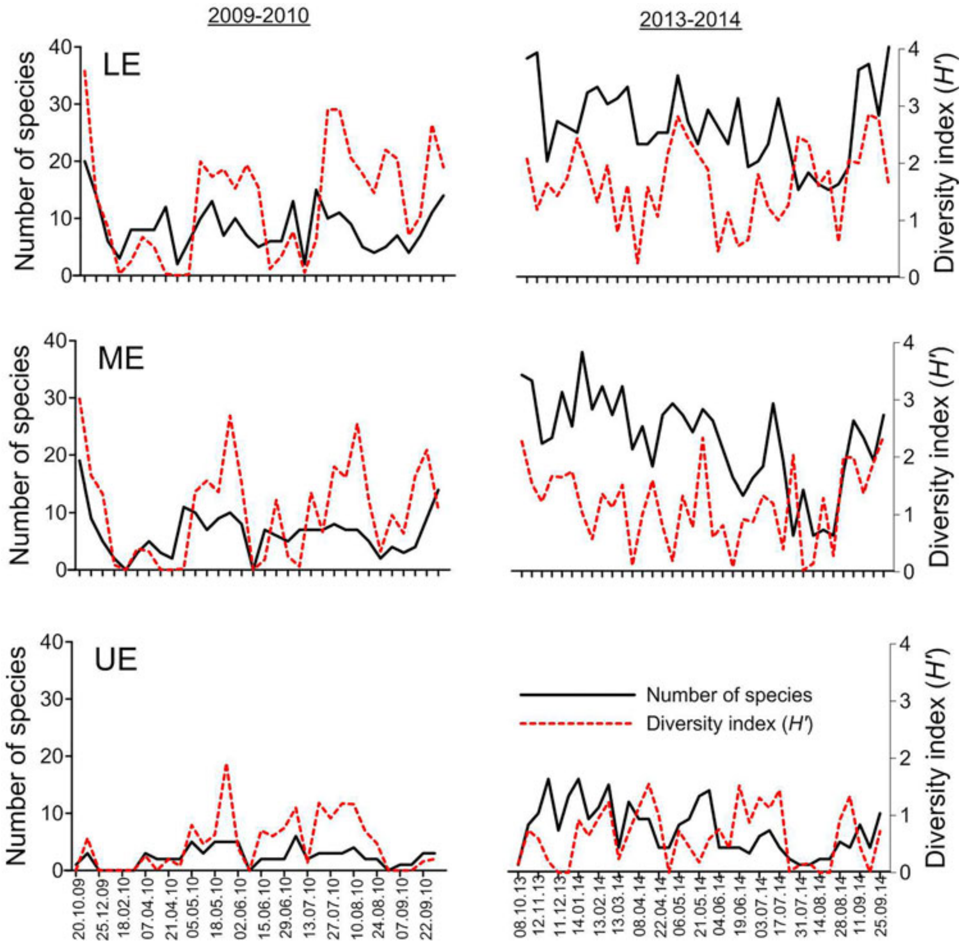

Fig. 3. Fluctuations in the number of species and diversity index during this study and the previous study.

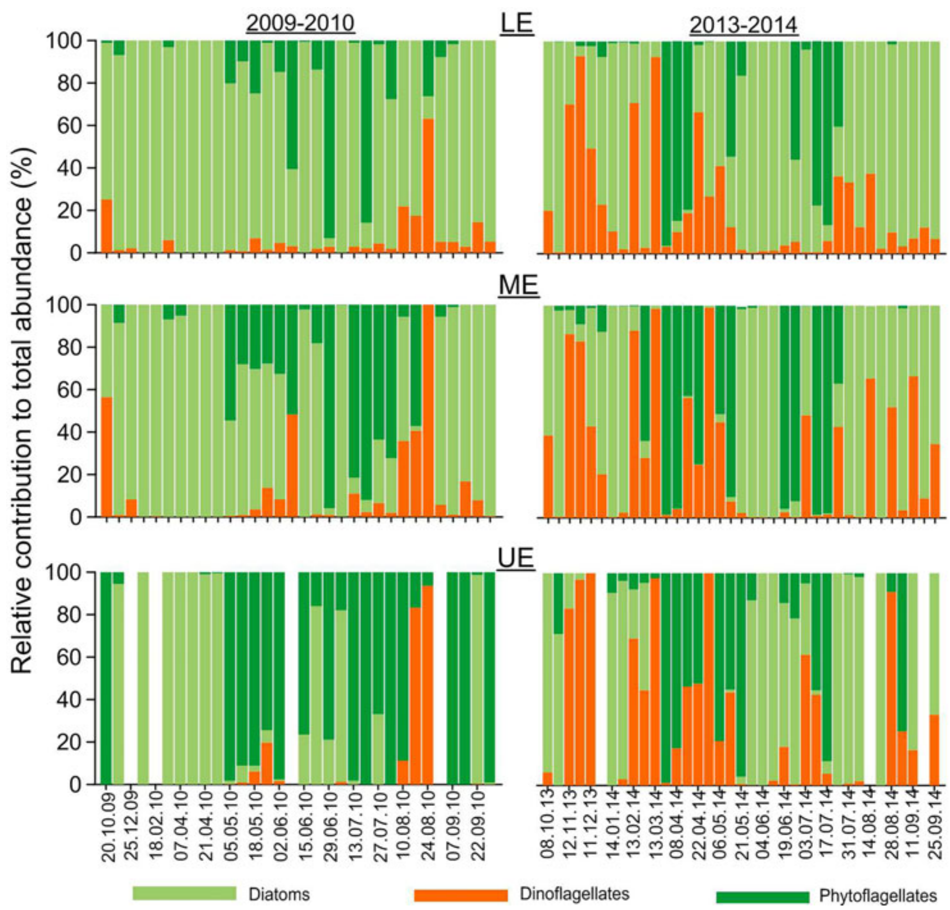

Fig. 4. The relative contribution of groups to total phytoplankton abundance during this study and the previous study (absence of a histogram = no species).

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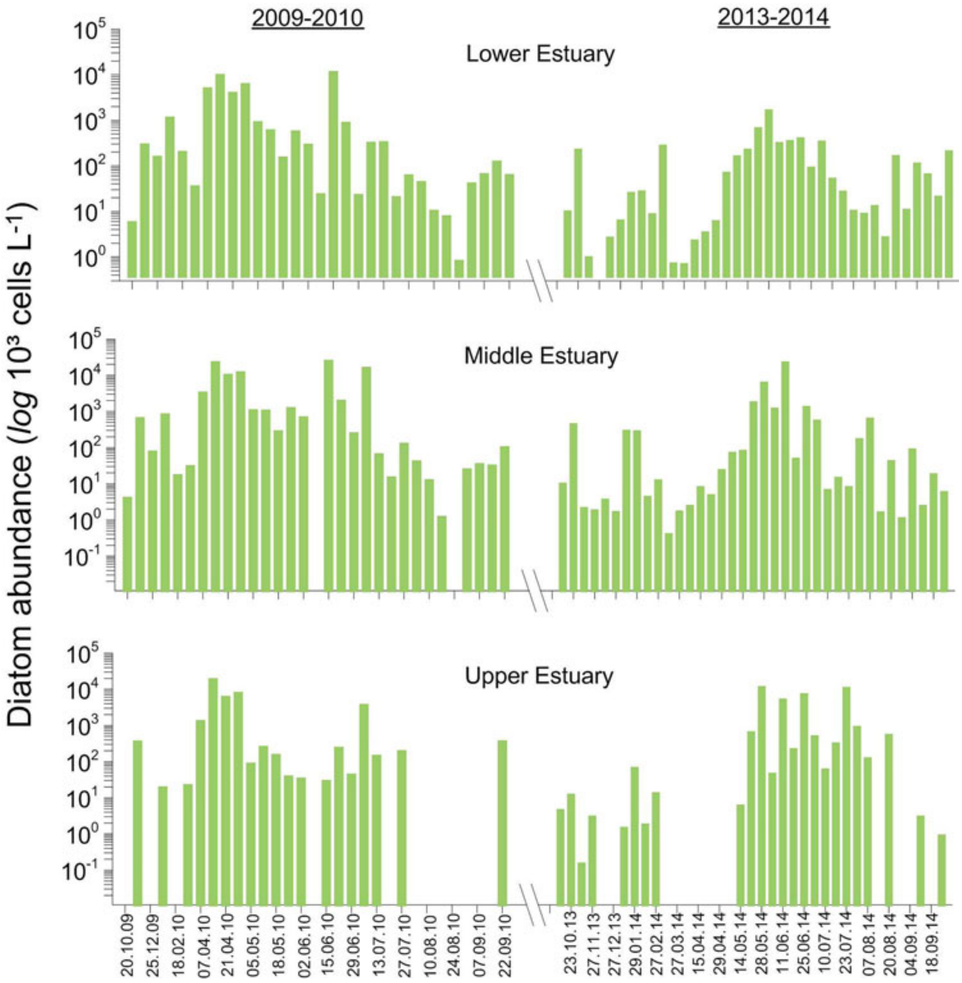

Fig. 5. Temporal variations of diatom abundance during this study and the previous study (absence of a histogram = no species).

Despite high nutrients in estuaries, light limitation, salinity change and water circulation may limit the growth of phytoplankton (McLusky & Elliott, 2004). All these studies revealed that changes in light availability and salinity in marine ecosystems influence the spatio-temporal distribution and composition of phytoplankton.

Before rehabilitation of the GHE, the growth of phytoplankton was limited by weak water circulation and low water transparency due to heavy pollution (Saydam *et al*., 1986; Uysal, 1996; Tas & Okus, 2003). The improvement in water quality following the rehabilitation project resulted in an ecosystem shift including a change in phytoplankton composition (Tas *et al*., 2009). The increase in light availability caused an increase in phytoplankton abundance and diversity (Tas *et al*., 2009). The seawater transfer to the GHE through Kagithane Stream changed some physical parameters (e.g. Secchi depth and salinity) affecting the phytoplankton abundance and diversity, when compared with the previous study. One of the most important changes after seawater transfer to the GHE was a higher abundance of dinoflagellates and phytoflagellates particularly in the ME and UE.

The increase of the relative contribution of dinoflagellates and phytoflagellates to the total phytoplankton abundance showed that they are probably more competitive than the others. In contrast, the decrease of the relative contribution of diatoms may be related to a notable decrease in silicate values in this study when compared with the previous study. Domingues *et al.* (2012) reported that the relative abundance of the main phytoplankton groups in the Guadiana estuary changed after dam construction resulting in a decrease in nutrient concentrations, and increase in light availability. The findings obtained from this study are in agreement with the study performed by Domingues *et al.* (2012).

The increase in cell abundances together with the temperature rise in April are evidence that these conditions favoured phytoplankton. The significant positive correlation ($P < 0.01$) between dinoflagellates/phytoflagellates and salinity particularly in the UE implies that the increase in salinity may promote their growth. Thus, the fluctuations in salinity can play an important role in phytoplankton composition. Tas *et al.* (2009) reported that changes in salinity played a more significant role in phytoplankton composition than temperature, particularly in the UE. The effect of salinity on phytoplankton diversity also appears clearly in Spearman's correlation coefficients which correlated positively with salinity. It may be an indication that some species achieved optimal growth in these conditions. The increased light intensity also explains the high abundance of phytoplankton between April and August. These findings were also supported by the earlier study performed in the GHE following rehabilitation (Tas *et al*., 2009).

It is often reported that low water transparency due to high concentration of suspended particulate matter can limit phytoplankton growth (Saydam *et al*., 1986; Uysal, 1996; Tas & Okus, 2003; Tas *et al*., 2009). *Pseudo-nitzschia* species were rarely observed in the UE probably due to very low light availability (Tas & Lundholm, 2017). Phytoplankton productivity and composition are largely controlled by light availability (Cloern, 1987) and light is likely to be limiting to phytoplankton growth especially in the middle and upper estuarine zones (Domingues *et al*., 2010, 2012). The findings of this study are in agreement with all of these previous studies. Despite relatively decreasing

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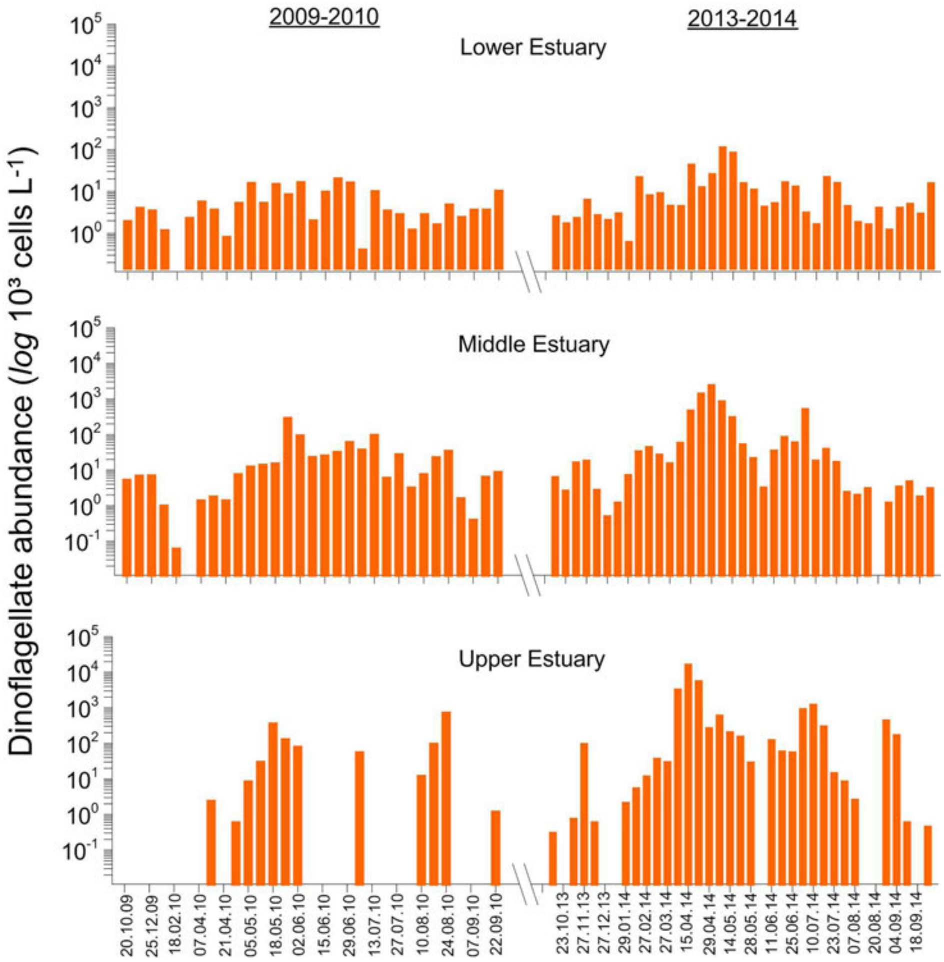

Fig. 6. Temporal variations of dinoflagellate abundance during this study and the previous study (absence of a histogram = no species).

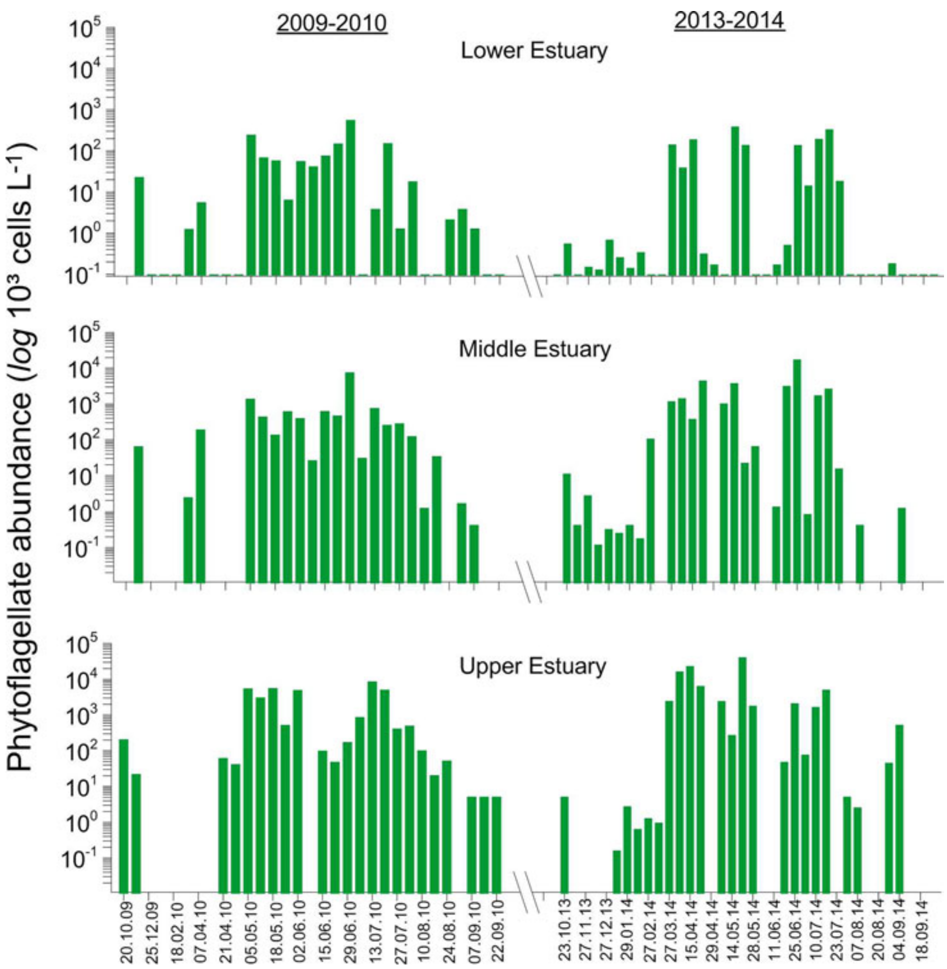

Fig. 7. Temporal variations of phytoflagellate abundance during this study and the previous study (absence of a histogram = no species).

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nutrient concentrations in the ME and UE, light availability is likely to be a more limiting factor for phytoplankton in this study area. A substantial increase in average Secchi depth (from 0.5–1.6 m, on average) has promoted the growth of dinoflagellates in the UE during this study period, as confirmed in the positive relationship between Secchi depth and *N*-Dino in the UE (Table 3). Light penetration through the water column might have increased particularly in the upper estuary after seawater transfer. Additionally, greater water transparency means a deeper euphotic layer and a more favourable environment for growth of dinoflagellates and other phytoflagellates probably due to their motility. This ability of dinoflagellates and other phytoflagellates gives them an advantage compared with other phytoplankton groups. Hence, light availability has been one of the most important factors for changes in phytoplankton composition in this study area. Diatom abundance and their relative contribution to total phytoplankton decreased probably due to the 3-fold decrease in silicate values (from 17.8 to 6 µM, in average) in this study when compared with the previous study.

In conclusion, the two hypotheses tested in this study were supported by the findings obtained. The first hypothesis is that the transfer of Black Sea water into the GHE increased water transparency and dissolved oxygen, particularly in the upper estuary. The second hypothesis is that changes in certain environmental factors affect phytoplankton composition that is evidenced in the relative increase of abundance of dinoflagellates and other phytoflagellates and their contribution to the total phytoplankton, and the increase in the number of species and species diversity. The results revealed that water transparency was one of the most important factors affecting phytoplankton composition in the study area. The findings confirm that phytoplankton species may be a good indicator for the changing environmental conditions in marine ecosystems.

Supplementary material. The supplementary material for this article can be found at https://doi.org/10.1017/S0025315420001058.

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