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Full Length Research Paper

Carrying capacity of *Chaetoceros gracilis* in Homa Lagoon and the bay of Izmir

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Marine diatom *Chaetoceros gracilis* has been investigated for its potential use as food in mariculture. In this study, we investigated the effects of temperature, salinity and nutrient on the growth of *C. gracilis*. The possibility for nutrient limitation to affect *C. gracilis* was assessed from two different ecosystems (Izmir Bay and Homa Lagoon). Our goal was to determine the growth rate of all nutrients and the maximum levels of the *C. gracilis* phytoplankton biomass (the maximum biomass carrying capacity) on the extent of its full growth and the level at which the nutrient restrictive growth keeps the biomass (biomass carrying capacity) and the nutrient(s) that sustain its year long growth. Nutrients significantly increased Izmir Bay and Homa Lagoon water's carrying capacity throughout the year and it was found out that the nutrients which restricted the carrying capacity and the growth rate were nitrogenous compounds.

Key words: *Chaetoceros gracilis,* mariculture, nutrient effects, biomass, marine diatom.

INTRODUCTION

The increase of human activities in coastal systems affects nutrient loading and consequently, the phyto-plankton response (McComb, 1995). This worldwide phenol-menon has led to extensive research on the effects of nutrients on primary producers (Granéli et al., 1990, 1999; Cottingham et al., 1998). Published results have shown that nutrient availability could control algal growth (DiTullio et al., 1993; Sakka et al., 1999; Gobler and Sañuado-Wilhelmy, 2001), biomass (Graziano et al., 1996; Caron et al., 2000) and species composition (Berdalet et al., 1996; Carlsson and Granéli, 1999; Duarte et al., 2000), but there is no general consensus as to which element (N or P) limits phytoplankton. Primary producers are traditionally considered to be N limited in marine environments like the South central Pacific (Dufour and Berland, 1999). There is growing evidence that P may be the main limiting nutrient in other ecosystems like the western and eastern Mediterranean Sea (Thingstad and Rassoulzadegan, 1995; Thingstad et al., 1998; Zohary and Robarts, 1998; Diaz et al., 2001). Phytoplankton in the North Atlantic were previously considered to be N limited (Graziano et al., 1996), but recent studies reported a P deficiency in the Atlantic Ocean (Ammerman et al., 2003; Vidal et al., 2003). Karl et al. (1995) claimed that the subtropical North Pacific could shift from N limitation to P limitation. Other authors have demonstrated that a combination of several nutrients (N, P and Si) rather than one alone limit the primary producers in marine (Sakka et al., 1999; Vrede et al., 1999; Caron et al., 2000) and freshwater (Dodds et al., 1989; Axler et al., 1994) systems. In theory, the nature of the nutrient limitation in an ecosystem depends on the internal and external hydrographic processes (for example, vertical mixing of the water column, advection, fresh water discharge, natural rainfall, anthropogenic loading) that affect the ambient N: Si: P ratio (Smith, 1984; Lignell et al., 1992). Algal growth is inherently complex in general, showing non-linear responses to various environmental parameters such as temperature, light and several nutrients, as well as demonstrating poorly understood interactions among these separate factors (Bowie et al., 1985; Thomann and Mueller, 1987). Site-specificity also makes extrapolation from lab or other field studies inherently problematic. Finally, the diverse multi species algal community may resist being treated as a single homogeneous unit. Thus, in spite of the

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existence of simple, well validated models for nutrient limited growth (Droop, 1983), accurately treating the kinetics of algal growth in water quality models remains a significant challenge.

Of the phytoplankton groups, diatoms play a very important role in the primer production (Sarthou et al., 2005; Nelson et al., 1995; Tre´guer et al., 1995; Mann, 1999; Smetacek, 1999; Tre´guer and Pondaven, 2000). It is estimated that diatoms contributed to the primer production in the world by 20 to 25% because of their dominant roles in productive areas such as upwelling areas and continental shelves (Hendey, 1964). Therefore, it is not surprising that there have been a lot of studies on diatoms among the other phytoplankton groups. Growth kinetics determined in the laboratory can be considered “fundamental” measurements that tell a great deal about physiological and community ecology (Tilman et al., 1982).

Of plankton sea diatoms, *Chaetoceros* is the genus, along with Thalassiosira and Coscinodiscus, which is spread over the greatest and widest area. It densely populates neritic areas and is very common even in the high seas (Rines and Hargraves, 1988). The usage of both some other types of the *Chaetoceros* genus and the *C. gracilis* genus (artemia and penaeus, some other crustacea species, some bivalve and mollusc species) that were used in our trial in breeding enhances the importance of studies in this field. *C. gracilis* is commonly used as live feeds for all growth stages of bivalve mollusc (oysters, scallops, clams and mussels), for crustacean larvae and for zooplankton used as feed for larvae (Brown and Farmer, 1994; Brown and Miller, 1992; Lombardi and Wangersky, 1995; Lombardi and Wangersky, 1991; Sanchez-Saavedra and Voltolina, 1994; Sanchez-Saavedra and Voltolina, 1995; Parrish and Wangersky, 1990; Napolitano et al., 2007). The fact that phytoplankton cultures are used in kinetic studies and the individuals of the same species have the same features on constant environmental conditions (size, morphology, chemical composition, etc.) having a distinctive quality, is that they respond to the environmental conditions that they are supposed to be subjected to in the same way. A lot of studies have been done on nutrient intake and kinetic studies for the earlier mentioned reasons.

Our goal was to determine the growth rate of all nutrients and the maximum levels of the *C. gracilis* phytoplankton biomass (the maximum biomass carrying capacity) on the extent of its full growth and the level at which the nutrient restrictive growth keeps the biomass (biomass carrying capacity) and the nutrient(s) that sustain its year long growth. By determining the hyperbolical relations between the maximum biomass levels (biomass carrying capacity) and nutrient concentrations obtained from *C. gracilis* growth graphs, a trial was made to apply flexible models which will restrict the levels that *C. gracilis* can reach.

MATERIALS AND METHODS

Izmir Bay

The bay of Izmir is located in the western part of Turkey and surrounded by a densely populated community. The bay is divided into the inner, middle and outer bay from the standpoint of topographical and hydrographical characteristics. The inner bay occupies a small area (57 km^2) and is shallow in depth (maximum 15 m) (Figure 1).

Homa Lagoon

This study was conducted in the Homa Lagoon area located at the outer part of Izmir Bay (northwest of Izmir, 38°31’10” north latitude and 26°49′50” east longitude). This is an important region for commercial fisheries including bivalve that is 35 km away from Izmir city in Aegean Sea (Figure 1).

The evaluation of water quality was based on physical and chemical parameters, such as: temperature, salinity, dissolved oxygen (Winkler method), pH and pHep-pH Electronic Paper (HANNA Ins). Water samples (1 L) were collected and preserved in cold, dark conditions. For laboratory analysis, dissolved nutrients (ammoniuma, nitrite, nitrate, silica and orthophosphates) were analysed by spectrophotometric method (Strickland and Parsons, 1972; Wood, 1975; Parsons et al., 1984). All of the spectrophotometric analyses were carried out by using Hach Dr-4000 UVD model spectrophotometer.

Cultures were maintained on a ratio of 12:120 light:dark cycle and the experiments were studied from Izmir Bay (Aegean Sea) and Homa Lagoon. For stock cultures and enrichment culture experiments, f/2 mediums described by Guillard (1975) were used. The experiments were carried out in 1 L Pyrex bottles initially containing 1 L of seawater. The experiments were carried out in 1 L Pyrex bottles initially containing 1 L of seawater. For the experiment, the concentrations of nutrients in f/2 medium were changed and thus, for every nutrient a different concentration was obtained. In the experiment groups, trace elements and vitamins were added to the seawater according to f/2 medium (Guillard, 1975). *In vivo chl-a* were measured on a daily basis and *in vivo* chlorophyll a were performed using Turner 10-AU model fluorometer.

The biomass carrying capacity of *C. gracilis* has been shown to be related to the concentration of substrate in the medium by the following equation:

$$\text{Chl } a_{max} = \text{P Chl } a_{max}(S/K_s+S),$$

where, chl a_{max} is the maximum chl a concentration, Pchl a_{max} is the potential maximum chl a unlimited by low concentrations of substrate (S) and K_s is half-saturation constant. From the growth graphics obtained (Chl a/ time), the maximum Chl a values reached were recorded (Chl a_{max}). Against each one of the limiting nutrient concentration, Chl a_{max} values were recorded in the graph. Growth rate and carrying capacity were calculated by formula 1 and 2, respectively:

$$\mu = \mu_{max}.S/(S+K_s) \quad (1)$$

$$\text{Chl-a}_{max} = P(\text{Chl-a}_{max}).\ S/(S+K_s') \quad (2)$$

RESULTS

The maximum potential reactive phosphate and the half-saturation constant (Ks^1) values for *C. gracilis* were found

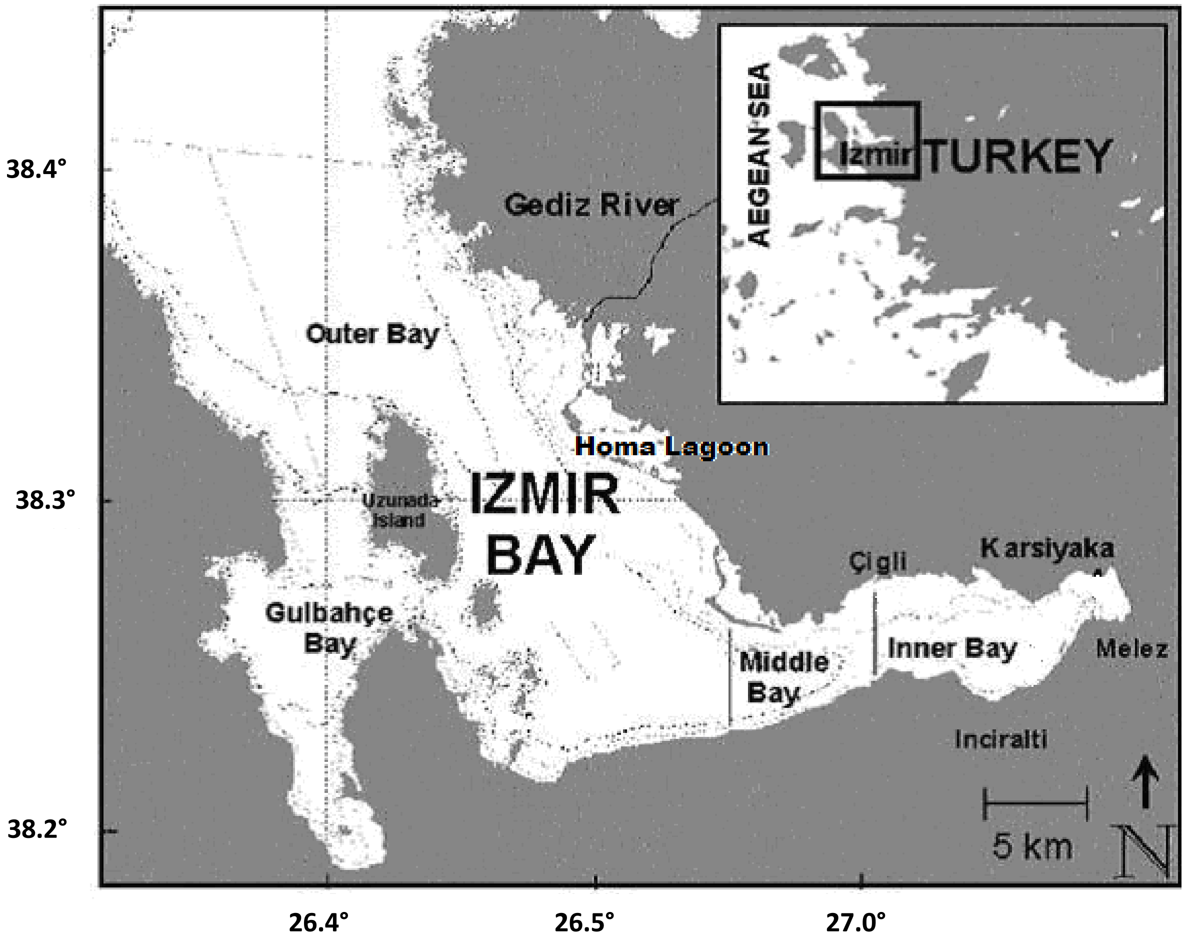

Figure 1. Map of the study area.

to be 661.6 day^{-1} and 23.52 µgat PO_4^{-3}-P/L, respectively in the study done in the bay of Izmir (Aegean Sea). The same values for *C. gracilis* were calculated as 18.063 day^{-1} and (Ks^1) 4.263 µgat PO_4^{-3}-P/L in the study done in the Homa Lagoon (Aegean Sea) (Figure 2).

The maximum potential silicate and the half-saturation constant (Ks^1) values for *C. gracilis* were found to be 39.735 day^{-1} and 1.784 µgat Si/L, respectively in the study done in the bay of Izmir (Aegean Sea). The same values for *C. gracilis* were calculated as 39.921 day^{-1} and 0.327 µgat Si/L and (Ks^1) in the study done in the Homa Lagoon (Aegean Sea) (Figure 3).

The maximum potential nitrate for chl-a values in the bay of Izmir and the Homa Lagoon were calculated as 401.05 and 34 day^{-1} and the half-saturation constants were calculated as (Ks^1) 118.16 and 6.923 µgat NO_3^--N/L, respectively. In the present study, the maximum chl-values were found to be higher in the bay of Izmir (Figure 4).

The maximum potential ammonium chl-a values in the bay of Izmir and the Homa Lagoon were calculated as 325.83 and 58.01 day^{-1} and the half-saturation constants were calculated as (Ks1) 80.24 µg at NH_4^+-N/L and 24.34 µg at NH_4^+-N/L, respectively. In this study, the maximum chl-a values were found to be higher in the bay of Izmir (Figure 5).

The uppermost curve in Figure 6 represents the maximum population growth for *C. gracilis.* The curve below explains the effect of temperature on the maximum growth rate. This implies that while the increase in temperature in the bay of Izmir is similar, it is very different in the months of April and August, and while an increase was observed throughout autumn, a decrease took place in winter. *C. gracilis* maximum carrying capacity was calculated in the Gulf of Izmir, where a decrease was observed at the end of winter and the beginning of spring. Although a rise in the temperature started with the beginning of summer, another decrease took place in August. It reached its maximum level in autumn and January. The Monod equation, which stands for the dependence of growth rates on nutrients, can also be used for carrying capacity (Figures 1 to 4, different parameter values and chl-a max, K_s^I).

The lower most curves show the maximum growth rate obtained by using the nutrient which, for the most part, decreases the growth rate of the phytoplankton population

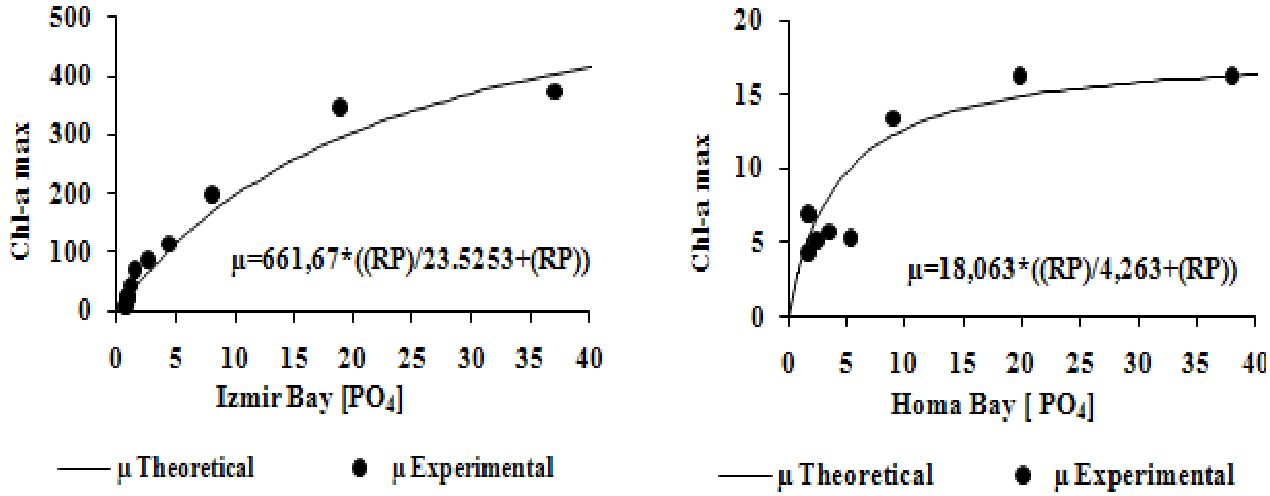

Figure 2. *C. gracilis* carrying capacity of Rp chl-a max in the bay of Izmir and the Homa Lagoon.

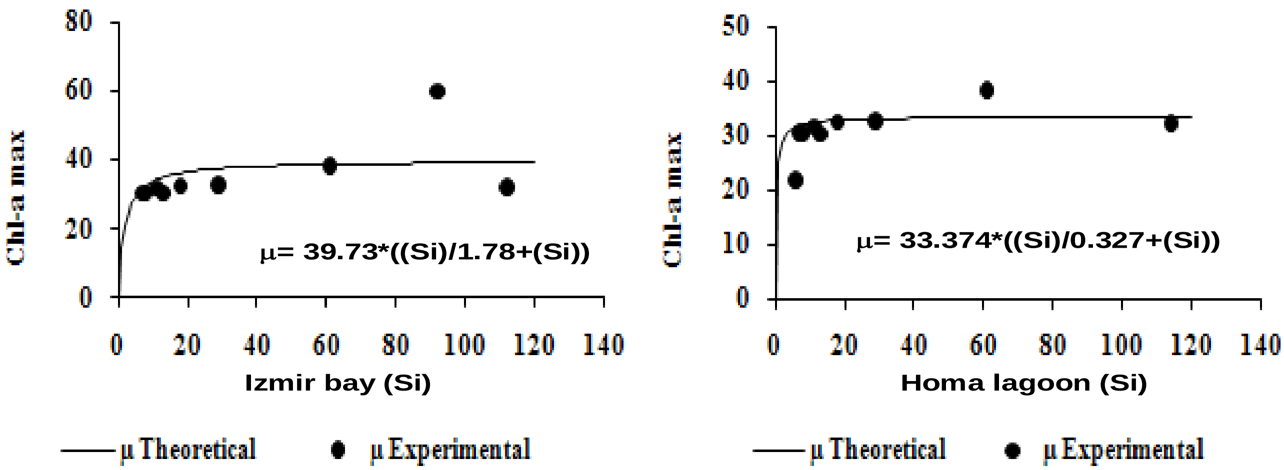

Figure 3. *C. gracilis* carrying capacity of Si in the bay of Izmir and the Homa Lagoon.

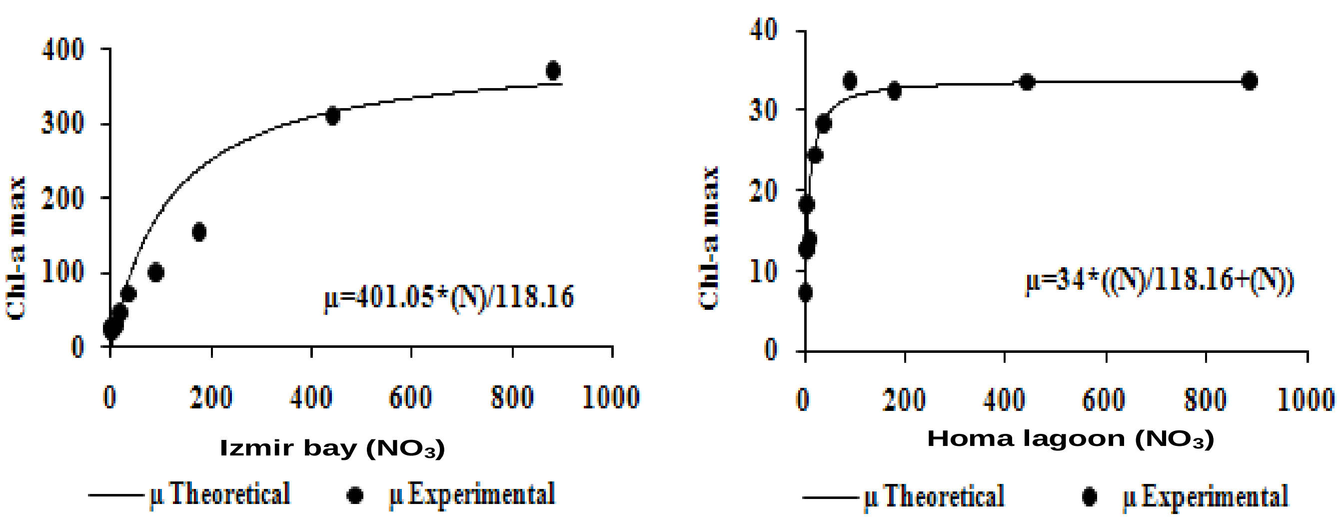

Figure 4. *C. gracilis* carrying capacity of NO_3 chl-a max in the bay of Izmir and the Homa Lagoon.

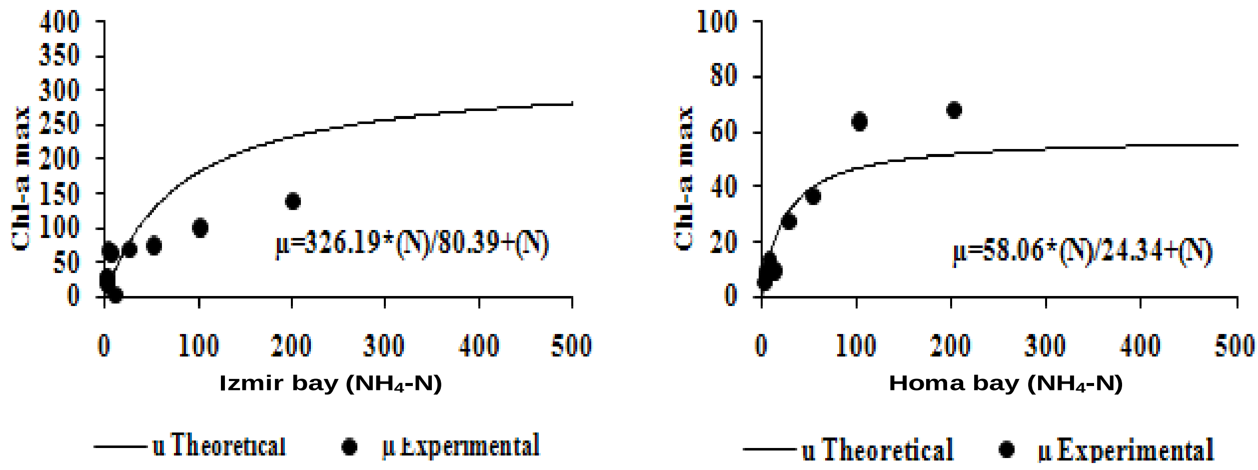

Figure 5. *C. gracilis* carrying capacity of NH_4 chl-a max in the bay of Izmir and the Homa Lagoon.

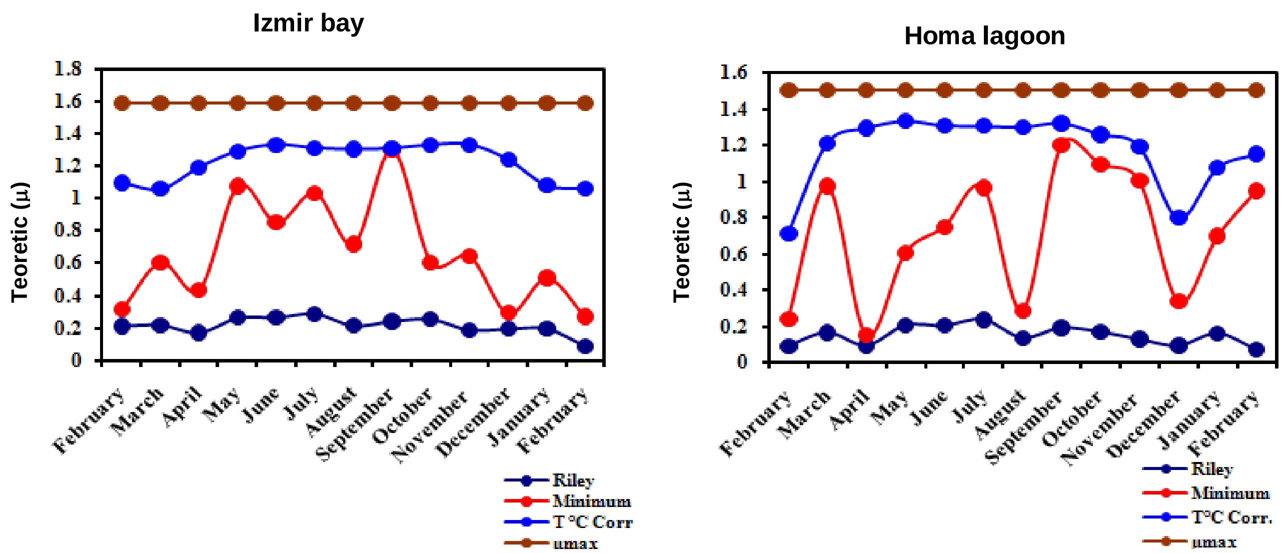

Figure 6. The effects of temperature and nutrient concentration on maximum growth rate.

according to Liebig's minimum rule (Table 1). A gradual increase with the onset of April and a radical decrease in August are observed in this curve; however, an increase in autumn and another decrease in mid-winter were still observed (Figure 6).

The upper most curves in Figure 8 represent the maximum population growth for *C. gracilis*. The curve below explains the effect of temperature on the maximum growth rate. Temperature causes significant autumn in the maximum growth rate in the Homa Lagoon in the months of February and December 2006. The decline is especially evident in the periods when the temperature drops to 7 and 8°C (Table 2). The lower most curves is the maximum growth rate obtained by using the nutrient which most decreases the growth rate of the phyto-plankton population according to Liebig's minimum rule. An increase with the onset of spring and a decrease at the end of summer together with an increase in autumn and a decrease in mid-winter are observed in this curve. Thus, Yürür (2008) reported that the reactive-phosphate flow is from water towards sediment in Homa Lagoon. It can at least be said that reactive-phosphate is bound by adsorption in the oxygenated water and therefore, phosphate in the water above is restrictive for the diatom in question (Table 2).

C. gracilis was observed to have the maximum carrying capacity of chl-a in Homa Lagoon, while the rise which started in spring continued to increase until July, and the second rise started in September and reached its maximum in January. The lagoon water's carrying capacity of the species in question reaches its maximum level from September to January, but the obtained values (*in situ)* were significantly low (Figure 7). Besides, *in situ* chl-a concentrations explain community biomass. The

Table 1. The effects of temperature and nutrient concentration on maximum growth rate in the Izmir bay.

Days Izmir Bay	Ammonium µgat/lt	Nitrate µgat/lt	Phosphorus µgat/lt	Silica µgat/lt	T (ºC)	S (%)	TºC Corr.	Si Teoretic	NH_4 Teoretic	NO_2^- Teoretic	PO_4^{-3} Teoretic	µ Blackman	Minimum	Limiting nutrient
February	4.76	1.19	0.85	3.78	9	37.92	0.71	0.50	0.32	0.03	0.33	0.02	0.03	NO3
March	15.74	0.84	2.36	18.66	15	38.85	1.21	1.17	0.60	0.62	0.82	0.19	0.60	NH4
April	2.48	0.30	2.64	2.96	18	39.85	1.29	1.26	1.02	0.63	0.83	0.22	0.63	NO3
May	8.29	1.88	1.06	0.78	22	41.85	1.33	1.29	1.07	0.49	0.77	0.20	0.49	NO3
June	5.2	1.95	1.92	1.06	26.3	40.85	1.31	1.01	0.39	0.63	0.41	0.13	0.39	NH4
July	10.93	2.31	5.8	4.62	26.5	39.85	1.31	1.19	0.65	0.70	0.78	0.20	0.65	NH4
August	2.35	0.65	4.08	16	27	39.37	1.30	1.30	1.21	0.24	1.02	0.15	0.24	NO3
September	2.77	1.71	6.3	10.8	25	38.85	1.32	1.25	0.93	0.91	0.58	0.21	0.58	PO4
October	4.49	0.98	4.05	7.14	16.5	39.85	1.26	1.18	0.60	0.76	0.55	0.18	0.55	PO4
November	1.06	1.81	3.3	10.27	11.6	40.35	1.19	1.15	0.65	0.75	0.17	0.10	0.17	PO4
December	1.45	1.67	2.74	11.9	13	41.85	0.80	0.78	0.29	0.13	0.64	0.07	0.13	NO3
January	2.2	4.2	2.39	7.5	13	40.85	1.08	1.01	0.51	0.23	0.69	0.12	0.23	NO3
February	0.36	4.04	2.91	10.8	13.5	39.85	1.15	0.65	0.27	0.70	0.17	0.08	0.17	PO4

Table 2. The effects of temperature and nutrient concentration on maximum growth rate in the Homa bay.

Days Homa Lagoon	Ammonium µgat/lt	Nitrate µgat/lt	Phosphorus µgat/lt	Silica µgat/lt	T (ºC)	T (ºC) Corr.	Si Teoretic	NH_4 Teoretic	NO_2^- Teoretic	PO_4^{-3} Teoretic	µ Blackman	Minimum	Limiting nutrient
February	2.35	0.27	0.40	1.20	7	0.71	0.50	0.32	0.03	0.33	0.02	0.03	NO3
March	2.94	7.26	0.98	13.15	15	1.21	1.17	0.60	0.62	0.82	0.19	0.60	NH4
April	11.14	6.65	0.82	19.05	18	1.29	1.26	1.02	0.63	0.83	0.22	0.63	NO3
May	12.31	4.03	0.62	13.55	22	1.33	1.29	1.07	0.49	0.77	0.20	0.49	NO3
June	1.27	6.50	0.21	1.72	26.3	1.31	1.01	0.39	0.63	0.41	0.13	0.39	NH4
July	2.93	8.05	0.69	4.90	26.5	1.31	1.19	0.65	0.70	0.78	0.20	0.65	NH4
August	41.43	1.60	1.68	140.38	27	1.30	1.30	1.21	0.24	1.02	0.15	0.24	NO3
September	7.11	15.75	0.37	8.77	25	1.32	1.25	0.93	0.91	0.58	0.21	0.58	PO4
October	2.73	10.57	0.36	7.16	16.5	1.26	1.18	0.60	0.76	0.55	0.18	0.55	PO4
November	3.48	11.68	0.08	11.92	14.5	1.19	1.15	0.65	0.75	0.17	0.10	0.17	PO4
December	1.70	1.36	1.80	24.72	8	0.80	0.78	0.29	0.13	0.64	0.07	0.13	NO3
January	2.65	1.96	0.81	7.28	12	1.08	1.01	0.51	0.23	0.69	0.12	0.23	NO3
February	0.91	10.77	0.08	0.64	13.5	1.15	0.65	0.27	0.70	0.17	0.08	0.17	PO4

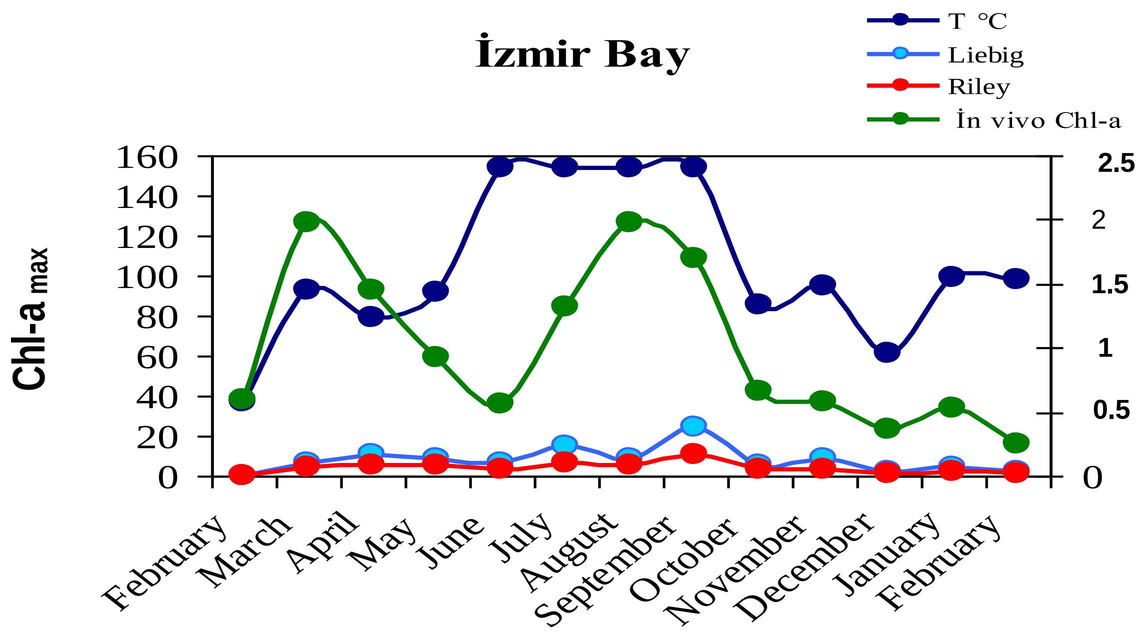

Figure 7. The effects of *in- vivo* chl-a, temperature on maximum growth rate.

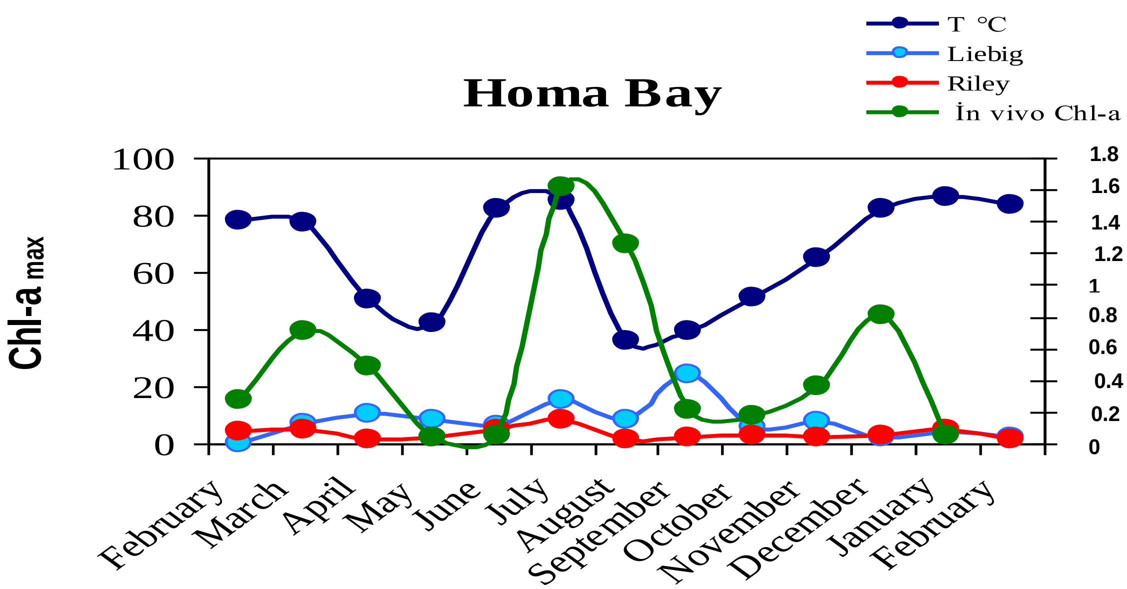

Figure 8. The effects of *in- vivo* chl-a, temperature on maximum growth rate.

biomass of the species, *C. gracilis*, in the total biomass is even lower. The Monod equation, which stands for the dependence of growth rates on nutrients, can also be used for carrying capacity (different parameter values and chl-a max, $K_s^{'}$).

Nutrients significantly increased the lagoon's water carrying capacity throughout the year and it was found out that the nutrients which restricted the carrying capacity and the growth rate were nitrogenous compounds (Table 2). Nitrogen restrictiveness is remarkable for most parts of the year (Table 1). This may suggest the contribution of the water of Izmir Bay as a nutrient source.

DISCUSSION

The rapid development of setae, following the division of

C. gracilis, suggests that they play an important role in the life cycle of this organism. Maintenance of photo-autotrophic algae in the euphotic zone was obviously beneficial to photosynthetic phytoplankton and a role for setae in decreasing the sedimentation rate of *C. gracilis* Schütt. As a result, the nutrient of *C. gracilis* varies according to its location.

The Izmir Bay and Homa Lagoon, located in the Eastern Basin of the Aegean Sea, is an area with a very high hydrodynamics. A number of studies have shown a strong coupling among biological and geochemical structures and the hydrology in the Aegean Sea.

In coastal ecosystems, net phytoplankton primary production is regulated by the interaction of several abiotic (nutrient fluxes, light availability and physical variability) and biotic factors (grazing pressure and competition). In such systems, nutrients are highly influenced by the anthropogenic activity and climatic variability via coastal upwelling events or continental inputs.

The functioning of coastal ecosystems are closely linked to that of freshwater systems and the upstream of coastal systems (Howarth and Marino, 2006) as the nutrient concentrations and ratios of waters discharged to coastal areas largely depend on agricultural and industrial activities, but also on freshwater biological activity upstream to coastal areas, as well as on the nature of the sediment in the drainage basin. Although there is a treatment plant in the bay of Izmir, nutrient entry originating from sediments occurs. Nutrient entry in the Homa Lagoon originates from agricultural activities and the sediments of the coastal areas.

Phytoplankton growth may be limited by phosphorus concentrations in winter and spring as the N:P ratio is greater than the normal Redfield ratio (16:1). This is a rare situation also found in Ria Formosa coastal lagoon, as nitrogen is usually the limiting nutrient in temperate lagoons (Newton et al., 2003; Newton and Mudge, 2005). Similarly, an alternative interpretation is that there is an excess amount of nitrogen in the system within this period of time. Seasonal variations of nutrient and phytoplankton biomass in the surface layers were mainly driven by the hydrological features during each season and the higher nutrient concentrations that a phyto-plankton biomass observed during the summer time. Nutrients significantly increased the Homa Lagoon water's carrying capacity throughout the year and it was found out that the nutrients which restricted the carrying capacity and the growth rate were nitrogenous compounds.

Several approaches such as bioassays experiments, concentrations of dissolved inorganic nutrient and nutrient input have been used to evaluate the limitation of phytoplankton by nutrients (Howarth, 1988). The elemental N: Si: P ratio for the phytoplankton growing under optimal conditions is 16:16:1 (Redfield et al., 1963; Brzezinski, 1985). Deviations of these molar ratios have been used to infer to which of these nutrients could be potentially limiting for phytoplankton (Howarth, 1988). Despite the fact that this approach raise criticisms (Howarth, 1988), it has been used widely by many authors. There is an extensive list of publications in which nutrient concentrations, together with their molar ratios (N:P:Si), have been used to suggest/infer nutrient limitation, as well as changes in the phytoplankton community assemblage (Krom et al., 1991; Dortch and Whiteledge, 1992; Justic et al., 1995; Nedwell et al., 2002; Ortiz et al., 2002; Mountin et al., 2002; Bethoux et al., 2002; Dafner et al., 2003; Wang et al., 2003; Lane et al., 2004).

The Mediterranean Sea has been usually considered as P-limited basins, although a review of the published works on this matter shows that there are important discrepancies. Several studies have shown that some areas in the western and eastern Mediterranean Sea are P-limited (Krom et al., 1991; Thingstad and Rassoulzadegan, 1995). In contrast, N rather than P may be a limiting nutrient (Herut et al., 2000). Some authors have reported that N-limitation could be more probable than P-limitation in areas of the western Mediterranean (Owens et al., 1989; Karafistan et al., 1998; Ortiz et al., 2002). Likewise, the Homa bay of Izmir and more nitrates were preferred to *C. gracilis* in autumn, but the phosphate was required in two areas.

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