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Distribution and morphology of the diatom genus *Chaetoceros* in the Lagoon of Nador, Morocco

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ABSTRACT

The genus *Chaetoceros*, is known for its abundance and diversity among the planktonic diatoms. Diatoms have long been used in aquaculture, especially as food for bivalves and juvenile gastropods. The general morphology and ultra- structural characters are presented for each species with special emphasis on the distinctive features with respect to species. The diversity and distribution of *Chaetoceros* (Bacillariophyceae) were studied at monthly intervals, throughout the experimental period (2018-2019) in the Nador Lagoon. A total of 24 *Chaetoceros* species were identified by inverted light microscopy. The highest recorded abundance of *Chaetoceros* species was 29320 cell/L, whereas *Chaetoceros compressus* (16000cell/L), *Chaetoceros danicus* (7200cell/L) and *Chaetoceros descipiens*(4120 cell/L) showed the highest dominance. The present work was constructed to determine the distribution of *Chaetoceros* at the lagoon of Nador in relation to the physicochemical parameters of the environment.

INTRODUCTION

Phytoplankton form the basis of food webs in aquatic ecosystems, representing an important biological component **(Sin *et al.*, 1999)**. Planktonic organisms can be grouped according to their size, nature, the biological characteristics of their developmental cycle, their vertical distribution on the water column or the type of environment they inhabit **(Rossi, 2008)**.

Phytoplankton have been extensively studied in the northern lagoons of the western Mediterranean **(Nuccio *et al.*, 2003; Aubry *et al.*, 2004; Pulina *et al.*, 2012; Daoudi *et al.*, 2013; Turki *et al.*, 2014)**. Diatoms are important primary producers in the ecosystems that have crucial roles in the global carbon cycle **(Nelson *et al.*, 1995; Falkowski *et al.*, 1998; Field *et al.*, 1998)**.

Diatoms are one of the most diverse and ecologically important groups of phytoplankton **(Rimet, 2020)**. They are important primary producers in ecosystems that have crucial roles in the global carbon cycle **(Nelson *et al.*, 1995; Falkowski *et al.*, 1998; Field *et al.*, 1998)**. In addition, the complexe volutionary history of diatoms as secondary endosymbionts is providing new insight in the host–endosymbiont relationships **(Prihoda *et al.*, 2012)**.

Diatoms are photosynthetic unicellular microorganisms. They comprise the most widespread phytoplanktonic group in the marine environment and can be found in isolated cells or grouped in colonies **(Zitouni, 2017; Bouchia *et al.*, 2018; Hayek *et al.*, 2020)**. Diatoms have been widely studied in the Mediterranean basin **(Caroppo *et al.*, 2005; Sahraoui *et al.*, 2009; Solak *et al.*, 2018)**.

Chaetoceros is one of the most abundant and diverse marine genera among the planktonic diatoms throughout the oceans **(Malviya *et al.*, 2016)**. This genus is found with a cosmopolitan distribution and is often dominant in plankton **(Cupp, 1943; Jensen & Moestrup, 1998; Bérard-Therriaut *et al.*, 1999; Hoppenrath *et al.*, 2009)**. Members of the genus are mainly marine, with hundreds of species **(Hasle & Syvertsen, 1997)**. A limited number of these species, specifically in inland waters, live in saline areas such as saltwater and saline lakes **(Rushforth & Johansen, 1986)**.

Chaetoceros sp. has been used as larval feed for shellfish and crustaceans in aquaculture due to their high nutritional qualities **(Brown *et al.*, 1997)**.

Additionally, *Chaetoceros* sp has been used in other applications for its antibacterial properties **(Seraspe *et al.*, 2012)** as well as being used as a sutainable biofuel producer **(Tokushima *et al.*, 2016)**.

The genus *Chaetoceros* was identified by Ehrenberg, who described two species, *Chaetoceros dichaeta* Ehrenberg (1844: 200) and *C. tetrachaeta* Ehrenberg (1844: 200) in the Southern Ocean (64ºS, 160ºW). The genus *Chaetoceros* is placed with the genus *Bacteriastrum* Shadbolt (1854: 13) in the family Chaetocerotaceae Ralfs in Pritchard (1861: 758, 860) **(Round *et al.*, 1990)**.

Some authors have described 400 species belonging to the genus *Chaetoceros*, but only 175 species have been confirmed **(Ferrario *et al.*, 2004)**. Their two main characteristics are the formation of chains and the presence of bristles **(Lee & Lee, 2011)**. Nevertheless, a significant proportion of *Chaetoceros* is currently unrecognized, and only about one-third to one-half is recognized taxa **(VanLandingham, 1968; Sundstrom, 1973; Rines *et al.*, 1990)**.

The aim of the present study was to characterize the different species of *Chaetoceros* according to its distribution, morphology and water characteristics over different seasonal intervals and locations in the Nador Lagoon (Fig. 1).

MATERIALS AND METHODS

1. Study area and sampling procedure

Coastal lagoons are shallow water bodies **(Chauvet, 1988; Karim, 2005; Trut *et al.*, 2014)**, separated from the sea by a sedimentary barrier beach "Lido" that was formed by the action of the swells above the tidal level, and lie parallel to the coastline **(Ludovic, 2006; Andreu-boussut *et al.*, 2008; Vianello 2013)**.

The lagoon of Nador, still known as Sebkha BouAreg, is also called Marchica **(Najih *et al.*, 2015)**. This site is located on the Moroccan North-East coast between the Cape of Water and the Cape of Three Forks. It extends between 35°05'N - 35°14N and 2°44'W - 2°56'W.

The lagoon of Nador is the largest lagoon in Morocco, with a length of 25 km, a width of 7.5 km and an estimated area of 115 km^2. It is the second largest lagoon complex in North Africa **(Zerrouqui *et al*., 2013)**, connecting with the Mediterranean Sea through a pass locally called "Boccana", which allows water exchange between them.

The location of this one has varied over time, according to **Erimesco (1961)** and **Tesson (1977)** in **(Guelorget *et al*., 1987; Lefebvre *et al*., 1997; Raji *et al*.,2013).**

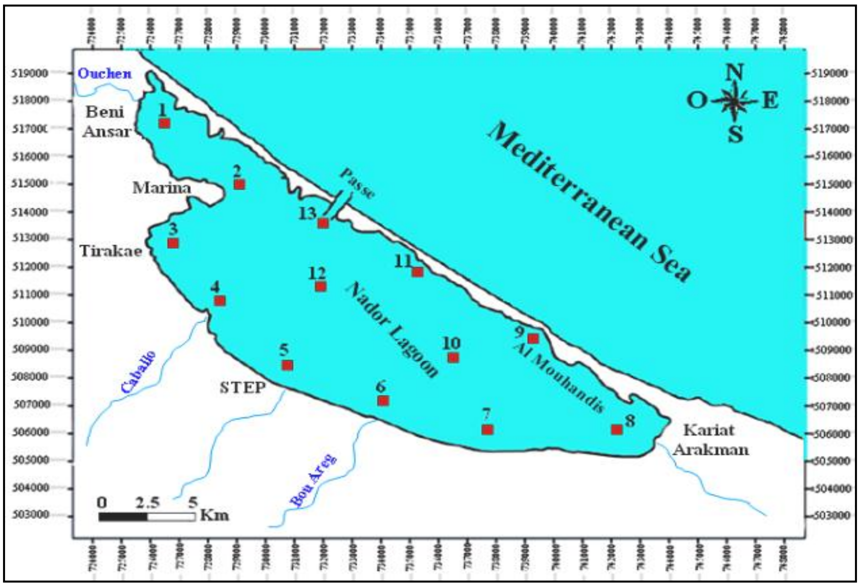

Fig. 1. Area of study and sampling stations

The Sampling was conducted in the Nador Lagoon, during four seasons and samples were collected throughout 10 months from March 2018 to February 2019 (except April and August 2018). One sampling point was collected each month, (26 water samples were collected respectively during the months) (Fig. 1). At each station, water samples were collected to perform the qualitative and quantitative study of *Chaetoceros* species as well as the physico-chemical analysis of the water, covering the whole Nador Lagoon. Samples of phytoplankton were collected using a planktonic net (diameter of 24 cm, mesh of 20µm).

2. Physical and chemical analyses

The physico-chemical parameters of water (temperature, salinity and hydrogen potential) were measured *in situ* using a specific probe type (ORION STAR A122) and pH meter (IONOMETER-EUTECH-INSTRUMENTS-CYBERSCAN-PH-510).

3. Morphological Analyses

To conduct a qualitative study, each sample was divided into three groups in the laboratory: one was preserved with neutral formaldehyde (4%); another was preserved with an acidic solution of Lugol's (2%); and the third was preserved as a living organism and analyzed directly by light microscopy (Leica "DM-IRB" inverted optical microscope) for general characteristics such as shape, size and color of the chloroplast.

The quantitative study was performed according to the Utermöhl sedimentation chamber method **(Utermöhl, 1958)**; the sample (25ml) was sedimented for 24 hours in the sedimentation chambers, and then quantified directly by the Leica "DM-IRB" inverted light microscope.

4. Statistical treatment

The XLStat-Pro® software was used which leads to a graphical representation where, as in PCA; the circle of correlations represents the factorial plane containing the initial observations and on the other hand the projection of the classes in the system of discriminating factorial axes which allows visualizing the quality of the discrimination. This analysis was used to detect the similarities or dissimilarities between the *Chaetoceros* data and the environmental parameters.

RESULTS AND DISCUSSION

1. **Physicochemical parameters**

The temperature is an ecological factor of primary importance with a great influence on the physicochemical properties of aquatic ecosystems **(Sigg *et al*., 2001)**. In macroalgae, temperature has a significant impact on metabolic rates as well as the overall dependence of respiration and photosynthesis **(Lüning, 1990)**.

The average temperature of the waters of Nador Lagoon, recorded at the level of the 13 stations under study during the 10 campaigns (March 2018-February 2019), fluctuated between a maximum of 27.3°C noted during the campaign of September 2018 and a minimum of 16.3°C observed during the campaign of January 2019. The recorded average temperature was around 20.4±2.79 (Fig. 2). These results were comparable to those found by **Mostareh (2017),** and the results measured at the level of the lagoon of Mellah by **Draredje *et al.* (2019).**

The average salinity of the waters of the lagoon of Nador recorded during this work, oscillated between a minimum of 34.6 observed during the campaign of December 2018 and a maximum of 38.1 noted during the campaign of January 2019. This value is comparable with that found by **EL Madani (2012).** The average recorded was about 36.2±0.82 (Fig. 2).

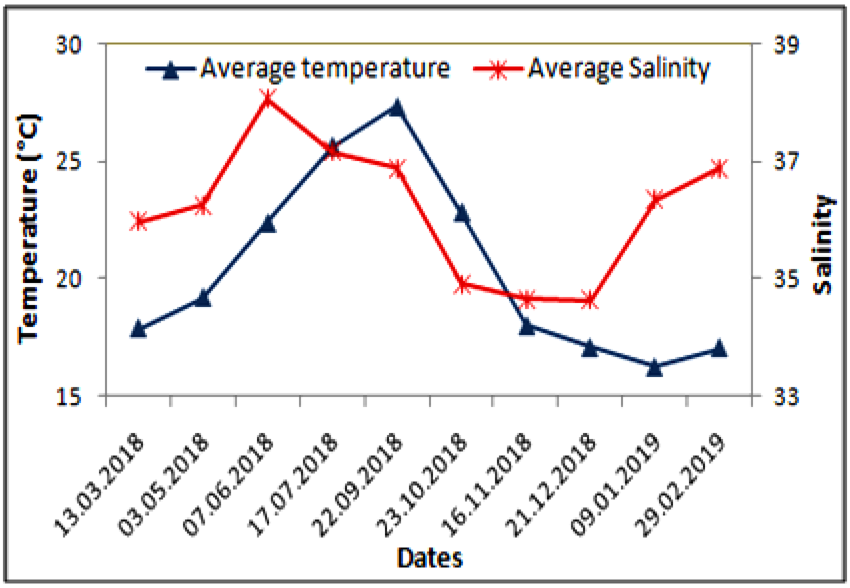

Fig. 2. Variation of temperature and salinity measured in the lagoon of Nador during the experimental period (2018-2019)

The average pH of the Nador Lagoon measured at the level of the 13 stations sampled oscillated between a minimum of 7.79 during the campaign of December 2018 and a maximum of 8.41 recorded during that of January 2019. The average recorded was about 8 ±0,13. Our results coincide with those of **Mostareh (2017)** and **Riouchi *et al*. (2021)** (Fig. 3).

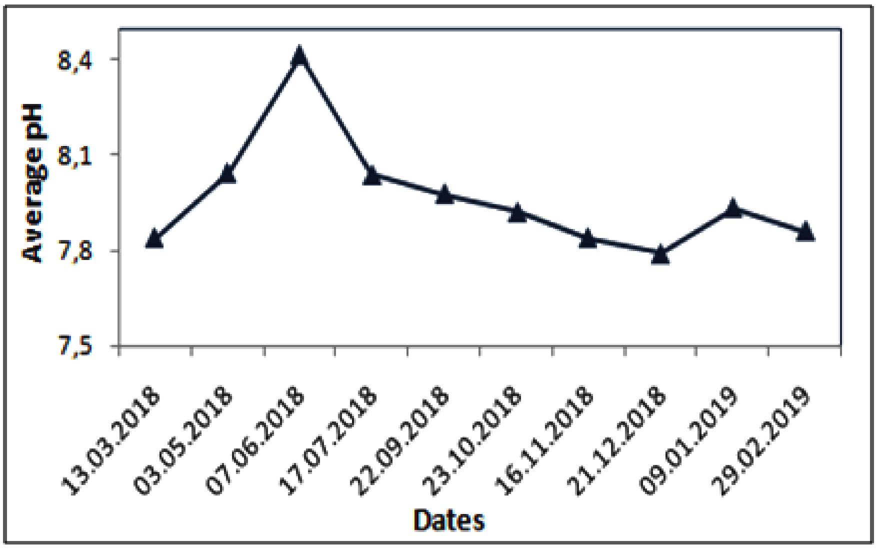

Fig. 3. Variation of pH value in the Nador Lagoon during the study (2018-2019)

The temperature parameter is the main factor determining the level of salinity of the Nador Lagoon water. In a non-polluted environment, the pH of marine water is around 8.2. This applies to the waters of both the Mideterranean and the Nador Lagoon.

Moreover, evaporation prevails over the contributions of fresh water because of the nature of the Mediterranean climate. This situation determines the salt content of the waters of the lagoon and consequently the biological processes taking place.

2. Morphological identification

Chaetoceros Ehrenberg, a diatom genus, is one of the most diverse and ubiquitous groups of marine phytoplankton. Its species are found all over the world, and they frequently dominate coastal habitats **(Hasle & Evensen, 1975; Guillard & Kilham, 1977; Rines & Hargraves,1990; Hernandez-Becerril, 1996; Hasle & Syvertsen, 1997; Bérard-Therriaut *et al*.,1999)**.

Results of investigating the species under study revealed morphological identifications described as follows:

***Chaetoceros Atlanticus*:** Inner oblong cells are straight and strong; the valve is slightly concave or flat with a central wave and the cap is raised. Oval cells. Terminal and incisive ends similar, long, thick, with a long basal part.

***Chaetoceros curvisetus*:** Tubular structure with a broadly convex primary valve and a nearly flat secondary valve **(Rines & Har-graves, 1988)**.

***Chaetoceros decepiens*:** Tubular structure and valve elliptical filaments, ciliated visible in the apical zones.

***Chaetoceros peruvianus*:** Solitary and heterogeneous cells. Cylindrical cells from within. Final stops and groups of dissimilar segments, long queues, ideas, limbs and base ground. The anterior node merges after a short base in the centre of the valve, and the posterior node appears near the corners, all directed vs. the posterior part of the cell.

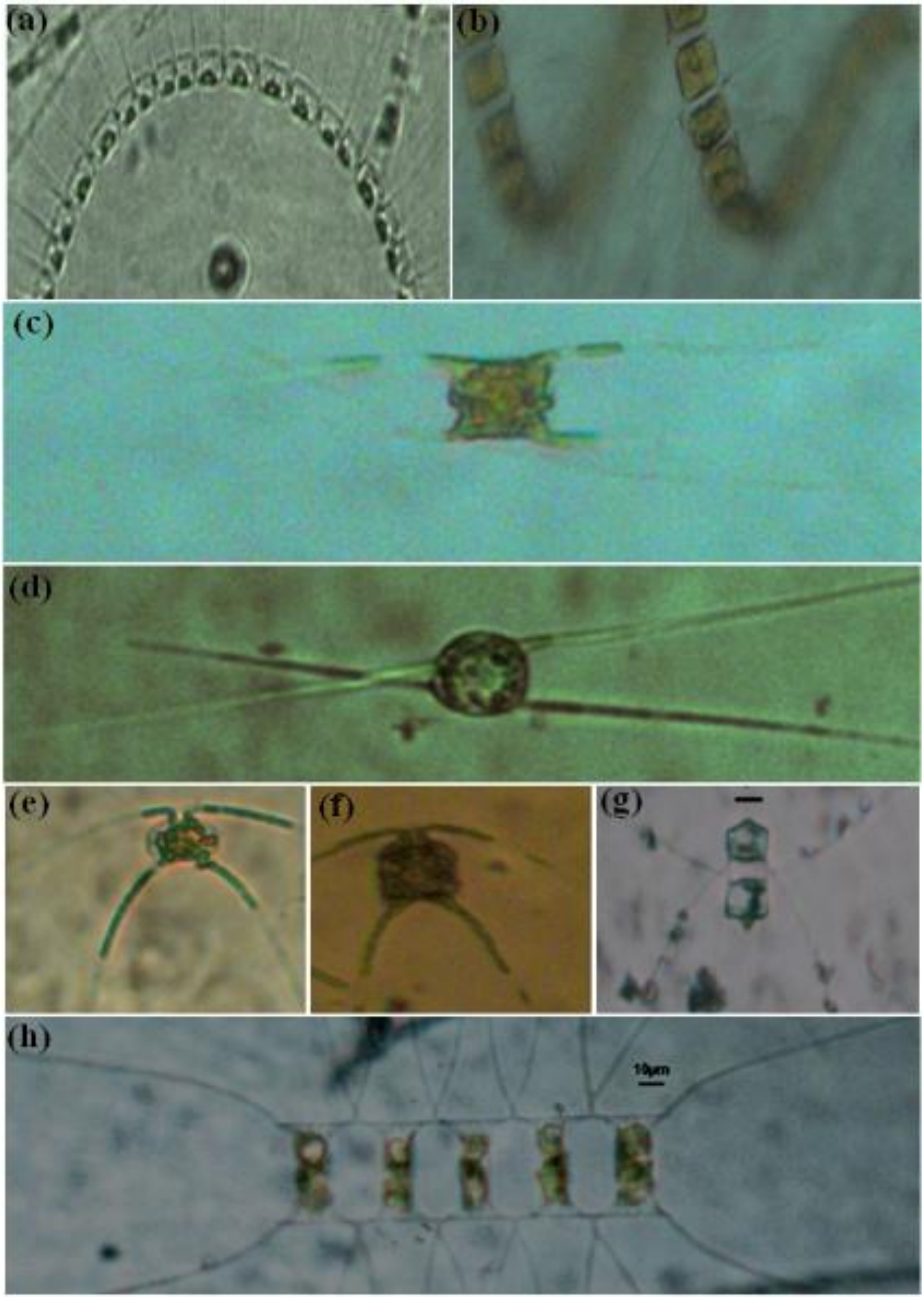

Fig.4. Morphological identification of *Chaetoceros* in the lagoon of Nador
a,b: *Chaetoceros curvisetus*. **c,d:** *Chaetoceros danicus*. **e,f:** *Chaetoceros peruvianus*.**g:** *Chaetoceros Atlanticus*. **h:** *Chaetoceros decepiens*. (10µm)

3. *Chaetoceros* distribution and abundance

During the study period, 24 species of *Chaetoceros* were detected in the Nador Lagoon (Table 1); namely, *Chaetoceros diversus* (Cleve, 1873), *Chaetoceros affinis* Var.Willei (Gran) (Hustedt, 1864), Chaetoceros *atlanticus* (Cleve, 1873), *Chaetoceros brevis* (F. Schütt, 1895), *Chaetoceros coarctatus* (Lauder, 1864), *Chaetoceros compressum* (Lauder, 1864), *Chaetoceros convolutus* (Castracane, 1886), *Chaetoceros curvisetus* (Cleve, 1889), *Chaetoceros danicus* (Cleve, 1889), *Chaetoceros diadema* (Ehrenberg) (Gran, 1897), *Chaetoceros descipiens* (Cleve, 1889), *Chaetoceros didymus* (Ehrenberg) (Gran, 1897), *Chaetoceros diversus* (Cleve, 1873), *Chaetocerosteres* (Cleve, 1896), *Chaetoceroslaciniosus* (F.Schütt, 1895), *Chaetocerossimplex* (Ostenfeld, 1902), *Chaetoceros laevis* (Leuduger- Fortmorel, 1892), *Chaetoceros mitra* (J.W. Bailey) (Cleve, 1896), *Chaetoceros lorenzianus* (Grunow, 1863), *Chaetoceros peruvianus* (Brightwell, 1856), *Chaetoceros pseudocurvisetum* (L. Mangin, 1910), *Chaetoceros sociales* (H.S. Lauder, 1864), *Chaetoceros tortissimus* (Gran, 1900) and *Chaetoceros* sp.

Table 1. Characterization of different *Chaetoceros* species found in the lagoon of Nador

	St1	**St2**	**St3**	**St4**	**St5**	**St6**	**St7**	**St8**	**St9**	**St10**	**St11**	**St12**	**St13**
Chaetoceros diversus Cleve,1873	-	-	-	-	-	-	-	-	-	+	-	+	-
*Chaetoceros affinis*Var.Willei (Gran) Hustedt, 1864	-	-	-	-	-	+	-	-	-	-	-	-	-
Chaetoceros atlanticus Cleve,1873	-	-	-	-	-	-	+	-	-	-	-	-	-
Chaetoceros brevis F. Schütt, 1895	-	-	-	-	+	+	-	+	-	+	-	+	-
Chaetoceros coarctatus Lauder1864	+	-	-	-	-	-	-	-	-	-	+	+	-
Chaetoceros compressum Lauder1864	+	+	+	+	+	+	+	+	+	+	+	+	+
Chaetoceros convolutus Castracane,1886	-	+	+	+	+	-	-	-	-	-	-	+	-
Chaetoceros curvisetus Cleve,1889	+	+	+	+	+	+	+	+	+	+	+	+	+
Chaetoceros danicus Cleve,1889	+	+	+	+	+	+	+	+	+	+	+	+	+
Chaetoceros diadema (Ehrenberg) Gran,1897	-	-	+	+	-	-	-	+	-	-	+	+	+
Chaetoceros descipiens Cleve,1889	+	+	+	+	+	+	+	+	+	+	+	+	+
Chaetoceros didymus (Ehrenberg)Gran,1897	-	+	+	-	-	-	-	-	-	-	-	-	-
Chaetoceros diversus Cleve,1873	-	+	+	+	-	-	-	-	+	+	+	+	+
Chaetoceros teres Cleve,1896	-	-	-	-	-	-	-	-	-	-	-	+	-
Chaetoceros laciniosus F.Schütt, 1895	+	+	+	-	-	-	-	+	-	-	-	-	-
Chaetoceros simplex Ostenfeld 1902	-	-	-	-	-	-	-	-	-	+	+	+	-
Chaetoceros laevis Leuduger-Fortmorel 1892	-	-	-	-	-	-	-	-	-	+	-	+	+
Chaetoceros mitra (J.W.Bailey) Cleve 1896	+	+	+	+	-	-	-	-	-	-	+	-	-
Chaetoceros lorenzianus Grunow,1863	-	-	-	+	-	-	-	-	-	-	-	-	-
Chaetoceros peruvianus Brightwell,1856	+	+	+	+	-	+	+	-	+	+	+	+	+
Chaetoceros pseudocurvisetum L. Mangin,1910	-	+	-	-	-	-	-	-	-	-	-	+	-
Chaetoceros sociales H.S. Lauder, 1864	+	+	-	-	+	-	+	-	-	+	-	-	-
Chaetoceros sp	+	+	+	+	+	+	+	+	+	+	+	+	+
Chaetoceros tortissimus Gran, 1900	+	-	+	-	-	-	-	-	-	+	-	+	+

- **Note: + Present, - Absent.**

The present results (24 taxa) are stronger compared to other studies carried out in the Nador Lagoon, showing that the genus of *Chaetoceros* was represented by 14 taxa (**El madani, 2012)** and 17 taxa (**Mostareh, 2017)**. Furthermore, the study of **Anis *et al*. (2019)** conducted in the lagoon of Mellah of the southern Mediterranean, Algeria recorded 5 taxa of *Chaetoceros*. In Peter the Great Bay (Sea of Japan), a total of 33 taxa of *chaetoceros* were discovered in the study of **Shevchenko *et al*. (2006)**. *C. affinis*, *C. atlanticus*, *C. decipiens*, *C. didymus,* and *C. salsugineus* were subjected to deep studies identifying their morphology **(Orlova, 1987, 1988; Orlova & Selina 1993)**.

According to *Chaetoceros* species quantification (Fig. 5), our results showed that the most abundant total cell density was recorded at station 5 corresponding to the wastewater treatment plant, with a value of 29320 cell/L. This was followed by a density of the order of 25160 cell/L, noted at station 12 in the centre of the lagoon, whereas the least abundant was detected at station 9, corresponding to Al Mouhandis with a value of about 980 cell/L (Fig. 5).

The total cell density found during the present work was lower compared to that found in the study of **El madani (2012)**, recording a density varying between $1.19.10^5$ and $2.13.10^6$cell/l.

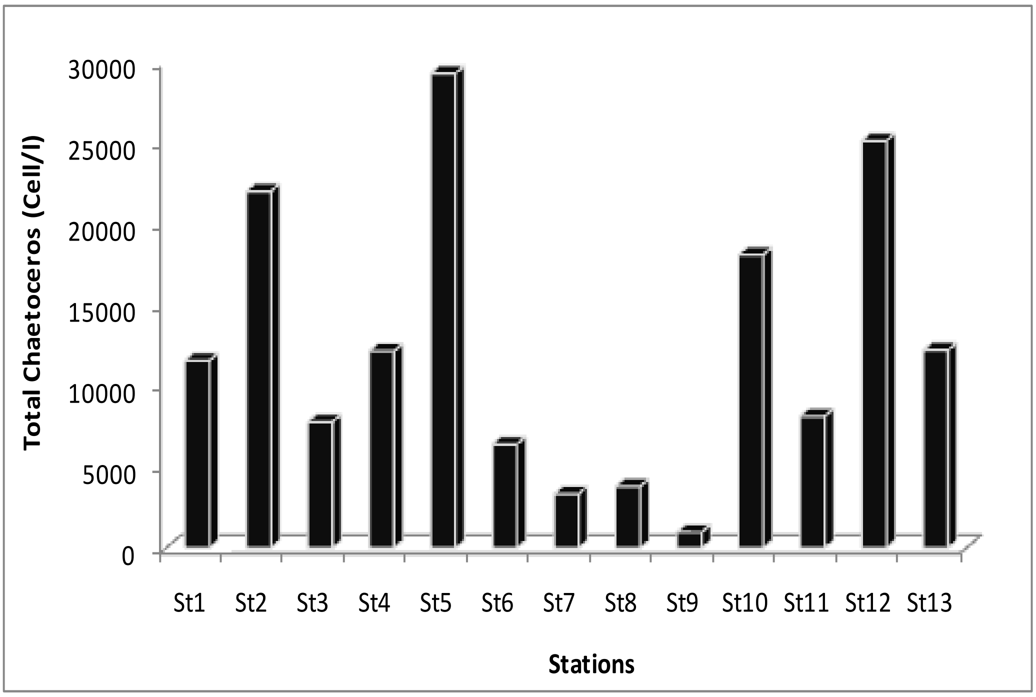

Fig. 5. Total number of *Chaetoceros* (Cell/L) for each station during the 10 sampling campaigns

During the sampling campaigns, the complex *Chaetoceros compressum* (Lauder, 1864) showed the highest cumulative density, ranging around 16000 cell/l during the month of October 2018. Concerning the complex *Chaetoceros danicus* (Cleve, 1889), it recorded the lowest cumulative density and was of the order of 280 cell/l during January of 2019 (Fig. 6).

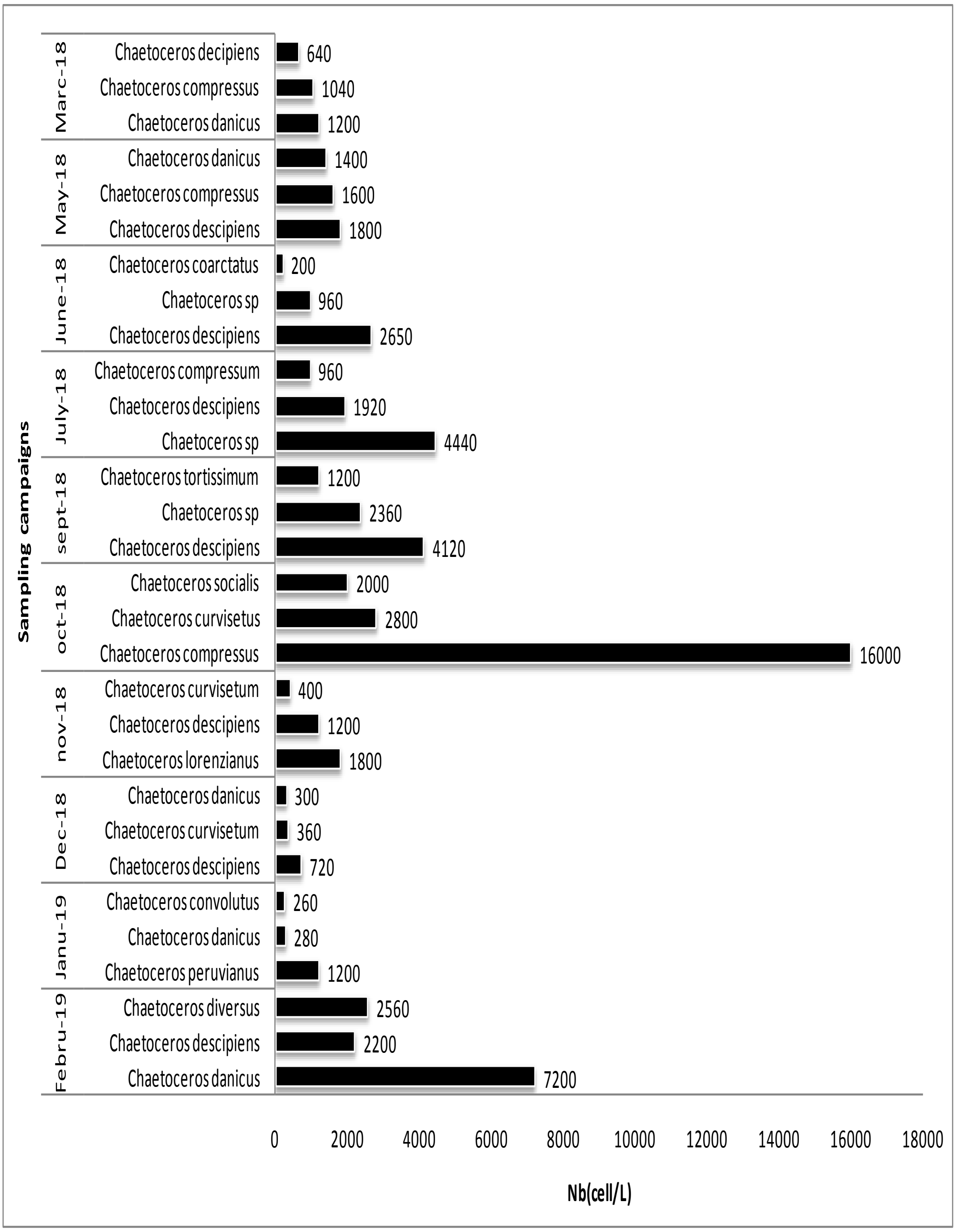

Fig. 6. Density of the two most dominant *Chaetoceros* species at the 13 stations during the sampling campaigns

4. Statistical analysis

The multivariate analysis (PCA) showed that the first two factorial axes provide nearly 72.24% of the information (Fig. 7). The F1 axis explains 41.82% of the total variation, and the F2 axis alone expresses a variation of 30.42%.

According to the two axes of F1 or F2; it is noticed that the average values of the environmental parameters (temperature, salinity and pH) are positively correlated with the average value of *Chaetoceros* taxon.

This multivariate analysis (PCA) indicates that the mean value of *Chaetoceros* is significantly correlated with environmental factors.

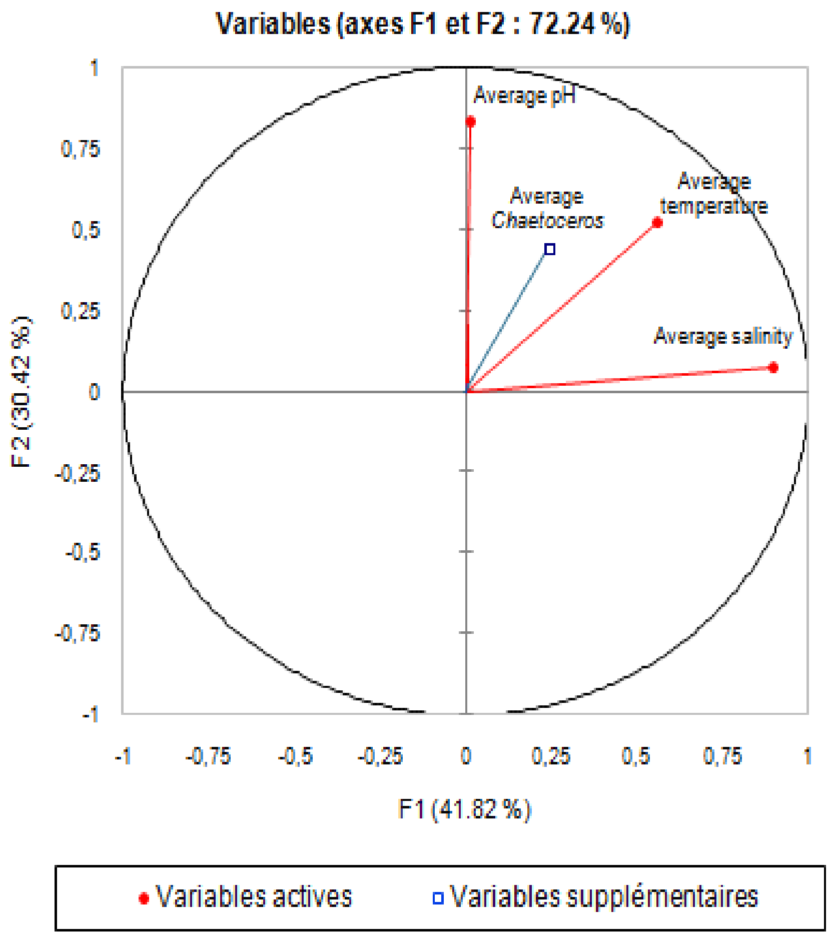

Fig. 7. Principal component analysis (PCA) of the average *Chaetoceros* as a function of environmental factors (T: temperature, Sal. salinity, pH) (axes F1 and F2 = 72.24%)

CONCLUSION

The diversity and distribution of the diatom genus *Chaetoceros* (Bacillariophyceae) were monthly studied in the Nador Lagoon. *Chaetoceros* was the most diverse genus in the diatom assemblage of the study area. Among the 24 species found in the lagoon, the three taxa; namely, *Chaetoceros Compressus, Chaetoceros danicus and Chaetoceros descipiens* were the most dominant species.

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