

UNUSUAL DOMINANCE OF HARMFUL MICROALGAE *PSEUDO-NITZSCHIA DELICATISSIMA* CF. (CLEVE) HEIDEN IN THE COASTAL WATERS OF JEDDAH, CENTRAL RED SEA

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Abstract

Dominance of harmful pennate diatom *Pseudo-nitzschia delicatissima* was observed during a regular environmental monitoring study carried out in the coastal waters of Jeddah, Saudi Arabia. Neither any discoloration of water nor any fish kill was reported during the period of this specific phenomena. This diatom species dominated the area with a higher percentage (97.5%) of the total phytoplankton community encountered. Apart from these, the occurrence of harmful algal bloom causing heterotrophic dinoflagellates was also noticed in a few numbers. Phytoplankton biomass in terms of chlorophyll *a* obtained was 4.1 mg m⁻³ and the silicate values (1.92 μmol l⁻¹) were comparatively higher than the other inorganic nutrients. Zooplankton abundance was relatively low (705 ind. m⁻³) and were dominated mainly by copepods (79.9%) followed by mollusc and decapod larvae.

Key words: *Pseudo-nitzschia delicatissima*, Abundance, Oligotrophic, Jeddah, Red Sea.

Introduction

The coastal marine ecosystems worldwide are subjected to increasing anthropogenic influences (Smith, 2003; Polat, 2007) and thus making the regular monitoring of the coastal regions a mandatory in order to check the presence of harmful algal blooms (HABs). Such HABs can create chaos in the system by many ways and can cause financial loss as well as health related problems. The occurrence of *Pseudo-nitzschia* bloom is a common phenomenon both in the coastal and oceanic waters around the globe (Rhodes *et al.*, 1998; Adams *et al.*, 2000; Stehr *et al.*, 2002; Lundholm *et al.*, 2003; Anderson *et al.*, 2006; Naz *et al.*, 2012). It is often associated with the phenomenon of upwelling in the oceanic waters and in the coastal region it flourishes due to the anthropogenic nutrient loading (Parsons *et al.*, 2002). *Pseudo-nitzschia* blooms are known for its production of the neurotoxin domoic acid, which create harmful impacts such as shellfish poisoning and domoic acid poisoning. Domoic acid has been detected in strains of *Pseudo-nitzschia* from California, other US Pacific coast states and in multiple other locations around the world (Anderson *et al.*, 2006; Schnetzer *et al.*, 2007; Lelong *et al.*, 2012; Schnetzer *et al.*, 2012; Stauffer *et al.*, 2012). Studies suggest that the frequency and duration of such phenomenon are increasing due to the continuous alteration of marine ecosystems resulting in the increasing availability of nutrients, which enhance the proliferation of microalgae (Dortch *et al.*, 1997; Anderson *et al.*, 2002; Parsons *et al.*, 2002). The pennate diatom, *Pseudo-nitzschia delicatissima* (Cleve) Heiden is having a cosmopolitan occurrence ranging from the Atlantic to Pacific Ocean (Hasle, 2002). Many studies consider *Pseudo-nitzschia delicatissima* as a cryptic species complex, having both toxic (Fryxell *et al.*, 1997) and non-toxic strains (Fehling *et al.*, 2005). Triggering factors, which result in the formation of HABs and the subsequent toxin production varies according to the environmental conditions, ranging from the upwelling regions (Kudela *et al.*, 2005) to the areas of anthropogenic influences (Quijano-Scheggia *et al.*, 2008). Laboratory experiments

further confirmed the capability of *Pseudo-nitzschia delicatissima* to produce domoic acid under favorable conditions (Smith *et al.*, 1990).

The Red Sea is well known for its oligotrophic nature due to the lack of riverine input and minimal deep water regeneration of nutrients (Morcos, 1970; Edwards, 1987). The primary production in the region entirely depends either on the nutrient rich Indian Ocean water that intrude through the Gulf of Aden or on the winter convection that brings deep water nutrients towards the surface (Wafar *et al.*, 2016). The low nutrient nature of the particular ecosystem normally affects the phytoplankton community, which always show low abundances (Halim, 1969; Ismael, 2015) with an exception to the diazotrophic cyanobacterium, *Trichodesmium erythraeum* that flourishes in the surface waters during particular time periods (Böttger-Schnack & Schnack, 1989; Post *et al.*, 2002). For the past two decades, some parts of the coastal waters of the Red Sea is witnessing many changes in the status of phytoplankton abundance and distribution and are now known for many sporadic events of higher abundances of phytoplankton, mainly diatoms (Devassy *et al.*, 2017). The occurrence of dinoflagellate species that can create HABs are also increasing (Mohamed & Al-Shehri, 2011, 2012; Kürten *et al.*, 2015; Banguera-Hinestroza *et al.*, 2016; Devassy *et al.*, 2017), which further point towards the shifting nature of the ecosystem. The present study, which carried out in the coastal waters of Jeddah, a fast growing metropolitan city in the central Red Sea provides further insights into the consequences of anthropogenic influences. The city receives human interferences in many ways, resulting in the formation of severely polluted coastal waters and subsequent formation of 'eutrophication' in many of its areas (Behairy & Saad, 1984; El-Rayis, 1998; El-Sayed, 2002a, 2002b; Peña-García *et al.*, 2014; Al-Harbi & Affan, 2016). The present report of the unusual dominance of *Pseudo-nitzschia delicatissima* in the coastal phytoplankton community can be considered as a warning sign for the authorities in order to create awareness in maintaining the ecological balance of the system.

Materials and Methods

As a part of the coastal biodiversity monitoring program, both the plankton collection and seawater sampling were carried out from the coastal waters of Jeddah, Central Red Sea (21°41.37' N, 39°2.13' E) on 31 May 2015 (Fig. 1). The surface sea water temperature was recorded with the help of a bucket thermometer, while the surface salinity with the help of a micro salinometer (RBR MS-310). A Niskin water sampler (Hydrobios 5L) was used to collect the water samples from a depth of 1 meter below the surface and were collected in clean 20L carbuoys. On reaching the shore laboratory, about 6-8 liters of seawater were filtered for chlorophyll *a* using a Whatman GF/F filter paper (0.7 µm) and kept in a deep freezer (-80°C) until further analysis. The chlorophyll *a* estimation was then carried out with the help of a UV spectrophotometer (Shimadzu) following Parsons *et al.*, (1984). Water samples for the nutrient analysis were also filtered through a Millipore membrane filter paper (0.45 µm) and the determination of nutrients were carried out by following the standard procedures of Parsons *et al.*, (1984). Both the phytoplankton and zooplankton samples were collected with the help of Hydrobios plankton nets of 20 µm and 150 µm respectively. In order to find the volume of water filtered (VWF), both the nets were equipped with a flowmeter (Hydrobios). Immediately after the collection, phytoplankton sample was preserved with Lugol's iodine solution along with a few drops of concentrated formalin (Kürten *et al.*, 2015), while the zooplankton sample was preserved with 4% formaldehyde. The enumeration of phytoplankton species was carried out according to the standard procedures of LeGresley and McDermott (2010) with the help of an inverted microscope (Leica DMI 3000B) in a Sedgewick Rafter counting chamber. The taxonomical identification of the phytoplankton species was performed by following Taylor (1976) and Tomas (1997). All the phytoplankton species obtained were then cross-checked with WoRMS (World Register of Marine Species) (www.marinespecies.org) as well as www.algaebase.org and named according to the latest taxonomical nomenclature. The Light micrographic images of the *Pseudo-nitzschia delicatissima* species were taken with a Leica DFC 450°C camera, while the scanning electron micrographs (SEM) were performed with the help of Hitachi S-3500N. The enumeration of zooplankton organisms was carried out with a stereo zoom microscope (Wild Heerbrugg M3B) and identification following Giesbrecht (1892); Heron and Bradford-Grieve (1995) and Conway *et al.*, (2003).

Results

During the particular period of collection, we hardly noticed any change in the color of the surface water in the region. The surface water temperature obtained at the sampling day was 26°C and the air temperature was 35°C. The surface sea water salinity obtained was 39.29. Regarding nutrient salts, the concentration of silicate was on the higher side, providing a value of 1.92 µmol l⁻¹. The other nutrients showed lower values with the nitrate displaying 0.33 µmol l⁻¹

, nitrite with 0.13 µmol l⁻¹, ammonia with 0.23 µmol l⁻¹ and the phosphate with 0.28 µmol l⁻¹. The phytoplankton biomass in terms of chlorophyll *a* obtained from the study region was 4.1 mg m⁻³. The total phytoplankton density obtained was 1237 X 10⁴ cells m⁻³ which was largely influenced by the abundance of the pennate diatom *Pseudo-nitzschia delicatissima* (1206 X 10⁴ cells m⁻³) that contributed almost 97.5 % of the total phytoplankton density. This high cell density, which can be considered as an almost a bloom condition for the oligotrophic waters eventually resulted in obtaining high chlorophyll *a* values for the region. The remaining portion was contributed by 63 different phytoplankton species, which were seems to be contributed in very low numbers towards the total density with an abundance of 0.12 cells X 10⁴ m⁻³ (Table 1). The dinoflagellates dominated the species diversity with 39 species with most of them known to produce HABs worldwide. The diatoms were represented by a total of 23 species (8 centrales, 15 pennales) and the blue green algae were represented by the native *Trichodesmium* sp. (Table 1).

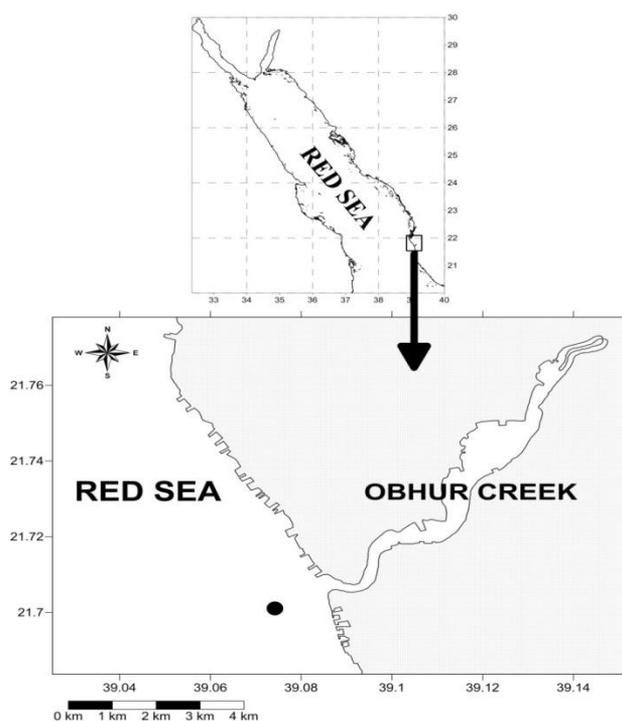


Fig. 1. Map showing the sampling site (black circle) in the coastal waters of Jeddah, central Red Sea.

From the microscopic analysis, the particular abundant pennate diatom species (*Pseudo-nitzschia delicatissima*) appeared to be seen aggregated together (Fig. 2a-b). The cells were in linear shape with slightly sigmoid and truncated in the girdle view (Fig. 2c) and spindle shaped with slightly rounded in the valve view (Fig. 2d). The cells have shown overlapping roughly about 1/9th of the entire length. The lengths of the individual cell varied with an average of 73.85 ± 4.76 µm and the measured of cell width were less than 3 µm. Each cell had a central interspace and one row of squarish poroids, with a multipartite hymen covering the poroids. The number of striae and fibulae obtained in 10 µm was 36 and 20 respectively (Fig. 2d).

Table 1. Other phytoplankton species observed during the study period. (* indicates the species that are known to produce HABs worldwide).

Phytoplankton taxa/species
Bacillariophyceae (Centrales)
<i>Chaetoceros</i> sp.
<i>Coscinodiscus</i> sp.
<i>Coscinodiscus granii</i> Gough, 1905
<i>Leptocylindrus danicus</i> Cleve, 1889
<i>Proboscia alata</i> (Brightwell) Sundström, 1986
<i>Rhizosolenia setigera</i> Brightwell, 1858
<i>Thalassiosira subtilis</i> (Ostenfeld) Gran, 1900
<i>Triceratium</i> sp.
Bacillariophyceae (Pennales)
<i>Amphiprora</i> sp.
<i>Amphiprora gigantea</i> Grunow, 1860
<i>Asterionellopsis glacialis</i> (Castracane) Round, 1990
<i>Epithemia argus</i> W. Smith, 1853
<i>Gyrosigma fasciola</i> (Ehrenberg) J.W. Griffith & Henfrey, 1856
<i>Licmophora flabellata</i> (Grev.) C. Agardh, 1831
<i>Lyrella lyra</i> (Ehrenberg) Karajeva, 1978
<i>Navicula</i> sp.
<i>Navicula transitans</i> Cleve, 1883
<i>Pleurosigma directum</i> Grunow, 1880
<i>Pleurosigma elongatum</i> W. Smith, 1852
<i>Pleurosigma formosum</i> W. Smith, 1852
<i>Surirella ovalis</i> Østrup, 1897
<i>Synedra arcus</i> Kützing, 1844
Dinophyceae
<i>Alexandrium</i> sp.*
<i>Ceratocorys armata</i> (Schütt) Kofoid, 1910
<i>Ceratocorys horrida</i> Stein, 1883
<i>Ceratocorys</i> sp.
<i>Dinophysis acuta</i> * Ehrenberg, 1839
<i>Dinophysis caudata</i> * Saville-Kent, 1881
<i>Dinophysis dens</i> Pavillard, 1915
<i>Dinophysis hastata</i> Stein, 1883
<i>Gonyaulax spinifera</i> * (Claparède & Lachmann) Diesing, 1866
<i>Gonyaulax verior</i> Sourmia, 1973
<i>Ornithocercus quadratus</i> Schütt, 1900
<i>Ornithocercus splendidus</i> Schütt, 1895
<i>Ornithocercus thumii</i> (Schmidt) Kofoid & Skogsberg, 1928
<i>Phalacroma cuneus</i> F.Schütt, 1895
<i>Phalacroma doryphorum</i> Stein, 1883
<i>Phalacroma mitra</i> * F.Schütt, 1895
<i>Phalacroma rapa</i> Jørgensen, 1923
<i>Phalacroma rotundatum</i> * (Claparède & Lachmann) Kofoid & Michener, 1911
<i>Phalacroma</i> sp.
<i>Podolampas palmipes</i> Stein, 1883
<i>Prorocentrum lima</i> * (Ehrenberg) F.Stein, 1878
<i>Prorocentrum</i> sp.
<i>Protoperidinium bipes</i> (Paulsen, 1904) Balech, 1974
<i>Protoperidinium conicum</i> (Gran, 1900) Balech, 1974 †
<i>Protoperidinium divergens</i> (Ehrenberg, 1840) Balech, 1974
<i>Protoperidinium pallidum</i> (Ostenfeld, 1899) Balech, 1973
<i>Protoperidinium pellucidum</i> Bergh, 1881
<i>Protoperidinium pentagonum</i> (Gran, 1902) Balech, 1974
<i>Protoperidinium steinii</i> (Jørgensen, 1899) Balech, 1974
<i>Scrippsiella trochoidea</i> (Stein) Loeblich III, 1976
<i>Tripes candelabrus</i> (Ehrenberg) F.Gómez, 2013
<i>Tripes furca</i> (Ehrenberg) F.Gómez, 2013
<i>Tripes gibberus</i> (Gourret) F.Gómez, 2013
<i>Tripes lineatus</i> (Ehrenberg) F. Gómez, 2013
<i>Tripes lunula</i> (Schimper ex Karsten) F. Gómez, 2013
<i>Tripes macroceros</i> (Ehrenberg) F. Gómez, 2013
<i>Tripes symmetricus</i> (Pavillard) F. Gómez, 2013
<i>Tripes teres</i> (Kofoid) F. Gómez, 2013
<i>Tripes trichoceros</i> (Ehrenberg) Gómez, 2013
Cyanophyceae
<i>Trichodesmium</i> sp. Ehrenberg ex Gomont, 1892

The abundance of the mesozooplankton in the study area was relatively low (705 ind. m⁻³). Copepods were by far the most abundant taxon contributing 79.9 % of the total mesozooplankton abundance (Table 2). Molluscs contributed next towards the total abundance, forming 12.7 % of the total zooplankton abundance and were mostly represented by gastropod larvae (83.3%). Crab zoea (4.3 %) and chaetognaths (1.7 %) were the other groups, which also were present during the sampling period. Within the copepods, 20 species belonging to 13 different genera were recoded representing 63.8% of the total adult copepods. The most abundant copepod species were *Centropages mohamedi* (16.2%), *Clausocalanus farrani* (13.4%), *Paracalanus* spp. (12.5%), *Centropages orsinii* (9.2%), *Oithona plumifera* (8.9%) and *Acartia amboinensis* (7.2 %) (Table 2).

Discussion

As part of the industrial growth as well as rapid urbanization, the coastal marine ecosystems of Jeddah, Saudi Arabia are continuously subjected to various anthropogenic influences (Behairy & Saad, 1984; El-Rayis, 1998; El-Sayed, 2002a, 2002b; Peña-García *et al.*, 2014). The most important among them are the presence of 117 discharge points of both the treated and untreated sewage along the entire coastline resulting in the increased availability of inorganic nutrients in the water (Khomayis, 2002; Mudarris & Turkey, 2006). Such changes in the ecosystem can bring unprecedented consequences in the form of potential outbreaks of microalgae very similar to the present study, which witnessed the abnormal proliferation of a pennate diatom *Pseudo-nitzschia delicatissima*. The Saudi Arabian coastal waters of Red Sea is least known for the occurrence of HABs mainly because of its oligotrophic nature, which does not favor the proliferation of these species. Recently, the things have changed in this particular ecosystem and the occurrence of HAB causing phytoplankton, mainly dinoflagellates keeping on increasing (Mohamed & Al-Shehri 2011, 2012; Kürten *et al.*, 2015; Banguera-Hinestroza *et al.*, 2016; Devassy *et al.*, 2017). At present, none of the harmful species resulted in any outbreak in the region, but still leaves more chances of possible future eruptions and thereby creating chaos in the marine ecosystem. The change in environment due to various natural and anthropogenic influences may lead to more frequent outbursts in near future. The present study is the first of its kind to report the astounding growth of a potential HAB causing diatom species in the Red Sea with an abundance, which can be considered as a near-bloom phenomenon for an oligotrophic water body. The pennate diatom *Pseudo-nitzschia delicatissima*, which seen dominating the summer period (May) contributed almost 97% towards the total phytoplankton density observed during the particular period. The occurrence of *Pseudo-nitzschia* blooms are a common phenomenon in both the coastal and oceanic waters (Rhodes *et al.*, 1998; Adams *et al.*, 2000; Stehr *et al.*, 2002; Lundholm *et al.*, 2003). In the coastal waters, it is mainly triggered by the increasing amount of anthropogenic nutrient loading (Parsons *et al.*, 2002) and is same in the case of current observation too. It is well known that the *Pseudo-nitzschia delicatissima* species are capable of producing neurotoxic domoic acid,

which can result in lethal shellfish poisoning and domoic acid poisoning (Anderson *et al.*, 2006; Schnetzer *et al.*, 2012; Stauffer *et al.*, 2012). There was neither any change in the water color nor any fish kill reported during the dominance of this particular species in the region. Previous studies reveal that the *Pseudo-nitzschia delicatissima* is a cryptic species complex, having both the toxic strains (Fryxell *et al.*, 1997) and the nontoxic one (Fehling *et al.*, 2005). The lack of any damage to the coastal ecosystem by the growth of the current species revealed the possibility of the presence of a nontoxic strain of *Pseudo-nitzschia delicatissima* in the Red Sea coastal waters. It can be further concluded that the effect of anthropogenic influences in the region was unsuccessful in triggering the massive bloom formation and subsequent toxin production. Still, the density was in the brinks of explosion and leaves the chances for future sporadic events, if the system continues to receive changes in the form of anthropogenic influences. This can bring unexpected changes in the region and can result in the massive destruction of the ecosystem. The potential link between *Pseudo-nitzschia delicatissima* and organic nutrients loading in neritic water around Jeddah is supported by experimental results of Loureiro *et al.*, (2009), who pointed that *Pseudo-nitzschia delicatissima* can successfully utilize both the ammonium and urea as a nitrogen source for their growth. Also, Caroppo *et al.*, (2005) showed that *Pseudo-nitzschia delicatissima* bloom development was not restricted by inorganic nutrients,

suggesting a possible control by organic nutrients. Other field studies have demonstrated a significant relationship between *Pseudo-nitzschia delicatissima* and inorganic nitrate, supporting the capability of this species to utilize both inorganic and organic forms of nitrogen (Kaczmarek *et al.*, 2007; Quijano-Scheggia *et al.*, 2008). The zooplankton community from the study area seems to be unaffected by the occurrence of such higher abundances of *Pseudo-nitzschia delicatissima*. The presence of many larval stages of copepods, molluscs, crab zoea and fish larvae point towards a limited effect of this bloom on the zooplankton community. Proximity of the Jeddah international port to the study site reveals a possible ballast water introduction of this harmful species to the coastal waters of Jeddah. It clearly indicates that the fast growing anthropogenic activities around the coastal regions can be lethal to the marine environment and can expect this kind of sudden outbreaks of harmful algal blooms in the near future. Early warning and prediction of *Pseudo-nitzschia* blooms in the coastal waters of Jeddah need a continuous observation and monitoring to characterize species distribution in relation to different physico-chemical parameters. This type of information can then be used to understand differences in species-specific responses to different environmental conditions, information that is essential in developing models that relate *Pseudo-nitzschia* population dynamics and its effect on the coastal marine ecosystems.

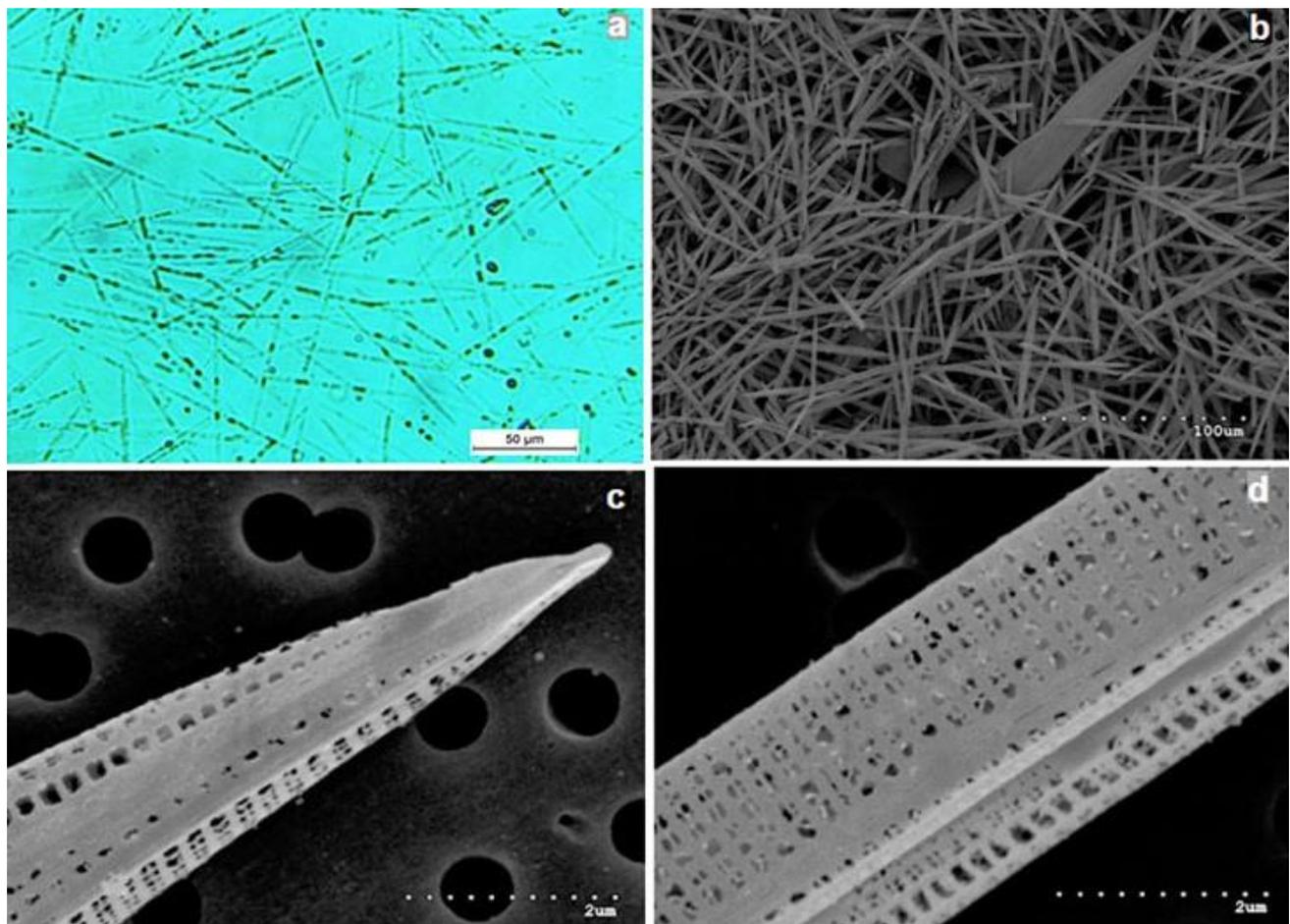


Fig. 2. Light microscopic (a) and scanning electron microscopic (b) images showing the dominance of *Pseudo-nitzschia delicatissima* along with (c) and (d) revealing the ultrastructure of the particular species.

Table 2. Zooplankton abundance (ind. m⁻³) and diversity recorded at the study area during the proliferation of *Pseudo-nitzschia delicatissima*.

Zooplankton taxa/species	Abundance (ind. m ⁻³)
Mollusca	
<i>Creseis acicula</i> (Rang, 1828b)	15
<i>Gastropod veliger larvae</i>	75
Copepoda	
Nauplii	28
Copepodites	176
Calanoida	
<i>Acartia (Odontacartia) amboinensis</i> Carl, 1907	26
<i>Calocalanus pavo</i> (Dana, 1852)	8
<i>Calocalanus styliremis</i> Giesbrecht, 1888	4
<i>Centropages mohamedi</i> El-Sherbiny & Al-Aidaros, 2015	58
<i>Centropages orsinii</i> Giesbrecht, 1889	33
<i>Clausocalanus farrani</i> Sewell, 1929	48
<i>Clausocalanus furcatus</i> (Brady, 1883)	19
<i>Clausocalanus minor</i> Sewell, 1929	2
<i>Labidocera pavo</i> Giesbrecht, 1889	2
<i>Nannocalanus minor</i> (Claus, 1863)	10
<i>Paracalanus</i> spp.	45
Cyclopoida	
<i>Copilia mirabilis</i> Dana, 1852	9
<i>Corycaeus (Onchocorycaeus) catus</i> Dahl, F., 1894	6
<i>Corycaeus (Corycaeus) speciosus</i> Dana, 1849	6
<i>Farranula gibbula</i> (Giesbrecht, 1891)	18
<i>Farranula rostrata</i> (Claus, 1863)	8
<i>Oithona plumifera</i> Farran, 1913	32
<i>Oncaea media</i> Sewell, 1947	6
<i>Oncaea scottodiarloii</i> Heron & Bradford-Grieve, 1995	8
Harpacticoida	
<i>Macrosetella gracilis</i> (Dana, 1847)	11
Decapods	
Crab zoaea	30
Chaetognatha	
<i>Sagitta</i> spp.	12
Pisces	
Fish larvae	10
Total zooplankton	705

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