



Seasonal Diversity of Planktonic Ciliates in Relation to Environmental Variables from Coastal Waters of Pakistan (Northern Arabian Sea)

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ABSTRACT

Ciliates are an essential component of the microzooplankton and occupy a significant role in the microbial food web. In the present study, seawater samples were collected in four seasons from the Gadani shipbreaking area and Sandspit coasts for one year. The seasonal diversity of planktonic ciliates and physicochemical characteristics of seawater were determined from samples collected on board using Niskin bottles. The ciliates diversity and abundance display variations in different seasons and vary from station to station. In Sandspit and Gadani, the maximum abundance and diversity of ciliates were recorded in the Southwest Monsoon. The total number of 128 species of ciliates classified into 56 genera from Gadani and 83 species of ciliates classified into 37 genera from Sandspit were recorded. In Sandspit, the most dominant ciliate species were *Leprotintinnus simplex*, *Salpingacantha ampla*, *Salpingella acuminata*, *Spirostomum minus* and *Strombidium conicum*. However, in Gadani, the most dominant species of ciliates were *Salpingella acuminata*, *Tintinnopsis beroidea* and *Tintinnopsis gracilis*. The present research on the dynamics of ciliate species diversity, abundance and standing stocks would provide information on the functioning of marine ecosystems. Ciliate communities are vulnerable to changes in their environment, the pollution in the coastal waters and changing climatic conditions trigger HAB-forming species, which is hazardous for fish and shellfish.

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NS designed the study. NS, TH data collected. RY and NS performed the experiments, analysed the data and wrote the article.

Key words

Gadani, Microorganisms, Ciliates, *Tintinnopsis*, *Leprotintinnus*, *Strombidium*

INTRODUCTION

Ciliates are unicellular, free-living aquatic characterized by cilia on their body surface (Hausmann and Hulsmann, 1996). Their size ranges from 20-200 μm are heterotrophic, although they also comprise mixotrophic forms (Stoecker *et al.*, 1987). Mostly, they are holozoic and feed on algae, detritus, protists and bacteria. Some are carnivores and depend on small metazoans. Ciliates are an essential component of the microbial food web (Pierce and Turner, 1992). Mixotrophic ciliates are found simply in the aloricate sub-group (Stoecker *et al.*, 1987) in the order Oligotrichida. According to the group division by Flynn *et al.* (2019) mixotrophic ciliates are part of

non-constitutive mixotrophs, which are grazers that can keep their prey chloroplasts and can perform photosynthesis. Ciliates are microplankton and dominate marine microzooplankton communities in species abundance and number (De Vargas *et al.*, 2015). They consume phytoplankton and serve as prey for metazoans therefore, they are an intermediate link in energy transfer in food webs (Fenchel, 2008). Worldwide studies on the diversity and distribution of the ciliates have been reported (Gomez, 2007; Yang *et al.*, 2020).

Ciliates are extremely widespread across various habitats and environmental conditions (Wang *et al.*, 2021). They are capable of turning into cryptobiotic forms when facing unfavourable conditions (Foissner *et al.*, 2005), among which cyst formation is a common way to engage in resting and resistant stages and to support cell dispersion (Farmer, 1980). Red-coloured blooms of *Mesodinium rubrum* are reported in coastal waters occasionally in connection with upwelling. They are considered microzooplankton, their photosynthetic activity represents 70% of the contribution towards overall primary productivity (Crawford, 1989). Due to the blooms of *Mesodinium rubrum*, oxygen depletion occurs, which leads to fish kill in the coastal areas (Pau *et al.*, 2017).

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Ciliate communities are highly influenced by environmental factors such as salinity, nutrients, temperature, pH and biotic factors (predators) (Sun *et al.*, 2017). Ciliates are found growing in some extreme environmental conditions, having sufficient vital energy to endure it (Lynn, 2008). Anaerobic ciliates are reported from anoxic environments, including marine, freshwater sediments, deep basins of estuaries and the anoxic hypolimnia of lakes (Finlay, 1982; Finlay *et al.*, 1991; Fenchel *et al.*, 1995; Xu *et al.*, 2013). Ciliates are sporadically reported living in hot springs at temperatures greater than 40°C (Kahan, 1972). They are frequently found in several submarine hydrothermal vents. Twenty species of ciliates on the East Pacific Rise hydrothermal vents were reported by Small and Gross (1985). Ciliates were also reported in salt lakes having a pH value of 9.5 (Wilbert, 1995).

Hauer and Rogerson (2005) reported heterotrophic protozoan from hypersaline environments. According to their study, thirty species were identified from high salinity (>15‰) waters. They elucidate that with an increase in salinities, species number of ciliates tends to decrease. In harsh Arctic and Antarctic environments, a number of research studies have been carried out to reveal the ecological role of planktonic marine ciliates (Roberts *et al.*, 2004). However, spatial factors (dispersal) can also be considered in the case of ciliate community assemblage. The limitation in dispersal could lead to a reduction in community resemblance with distance (Pan *et al.*, 2020). Studies have shown that the influence of environmental factors and spatial variables on ciliates mainly depends on the types of ecosystems and study scale (Zhang *et al.*, 2018). The ciliate community structure in the middle pelagic zone is controlled by geographic distance and depth (Sun *et al.*, 2019). In contrast, environments have a significant influence on ciliates than spatial factors in intertidal areas at the continental scale (Pan *et al.*, 2020).

Planktonic marine microbes have an important role in biogeochemical cycles and form a link between bacteria and higher trophic levels (Azam *et al.*, 1983; Caron *et al.*, 1985). Ciliates have multifaceted ecological roles due to their morphological, trophic, genetic and metabolic diversity (Caron, 2016). These varied qualities shape the interspecific (parasitism, predation, etc.) interactions and species-environment and have a significant role in the assembling of marine ciliate communities (Fuhrman *et al.*, 2015). The present research on the dynamics of ciliate species diversity, abundance and standing stocks would provide information on the functioning of marine ecosystems.

MATERIALS AND METHODS

For analysis of ciliates, seawater samples were

collected from two sites, Gadani ship-breaking area and Sandspit. The samples of water were collected in four seasons from two sites for the period of one year (October 2016 – September 2017). Triplicate water samples were collected each month using a water sampler (Niskin 1.7L) from a 1-meter depth analyzed for water quality and abundance diversity were recorded employing standard methods. Ecological parameters temperature, salinity and pH were recorded on the sampling site. The water quality parameters of the sampling site were analyzed using respective instruments. Water temperature (mercury thermometer), salinity (Refractometer; Atago, Japan) and pH (pH meter). Dissolved oxygen (DO), nutrients (nitrate, nitrite, ammonia and phosphate) were estimated according to Strickland and Parsons (1972) method.

Abundance and diversity of the ciliates

For diversity and abundance of ciliates, triplicate water samples were collected and preserved in acid Lugol's (1%) solution in 250 ml polycarbonated amber bottles. A sample volume of 50 ml was settled in a settling chamber (Hydrobios, Germany) for 24 h (Utermöhl, 1958). Ciliates were observed and counted under an inverted microscope (Olympus, IX-51 Japan). As a large number of species represented each genus, the counting was done for each species. The ciliates were identified on the basis of their characteristics for qualitative assessment.

Statistical analysis

Correlation coefficients (Pearson) between the ciliates density and the physiochemical parameters were determined. The data is analyzed using PRIMER 7.0. Changes in the ciliates community were examined using Shannon Weiner's diversity (H') and evenness (J).

RESULTS

The present investigation shows that the ciliate displayed a diverse species composition in Gadani and Sandspit in the Northern Arabian Sea. Seasonal abundance of ciliate was recorded in the present study. Seasons were categorized into Autumn Inter-Monsoon (October-November), Northeast Monsoon (December-February), Spring Inter-Monsoon (March-April) and Southwest Monsoon (May to September).

The ciliates diversity and abundance show variations in four seasons and vary from station to station. In Sandspit and Gadani, maximum abundance and diversity of ciliates was recorded in Southwest Monsoon (SWM) than in Spring Inter-Monsoon (SIM), Autumn Inter-Monsoon (AIM) and Northeast Monsoon (NEM) (Tables I-III). Ciliates species diversity in Gadani was more significant

Table I. Seasonal abundance of ciliates (cells/L) recorded from Gadani.

S. No.	Species	ST1	ST2	ST3
1	<i>Acanthostomella minutissima</i>	0	0	20
2	<i>Acanthostomella norvegica</i>	20	0	0
3	<i>Amphorella brandti</i>	20	0	0
4	<i>Amphorella minor</i>	40	0	20
5	<i>Amphorellopsiis acuta</i>	40	0	0
6	<i>Anigsteinia clarissima</i>	140	180	40
7	<i>Ascampbelliella retusa</i>	0	20	20
8	<i>Chaenea teres</i>	160	20	80
9	<i>Clevea melchersi</i>	0	0	20
10	<i>Codonella aspera</i>	0	0	40
11	<i>Codonella daday</i>	20	0	40
12	<i>Codonella galea</i>	20	20	40
13	<i>Codonella nationalis</i>	20	40	100
14	<i>Codonellopsiis morchella</i>	0	0	20
15	<i>Codonellopsiis schabi</i>	0	0	20
16	<i>Cyclotrichium gigas</i>	0	0	40
17	<i>Cyclotrichium</i> sp	40	0	0
18	<i>Cyrtophorid</i> sp	140	0	0
19	<i>Cyrtostrombidium longisomum</i>	20	20	0
20	<i>Cyttarocyliis brandti</i>	40	40	40
21	<i>Cyttarocyliis conica</i>	20	20	20
22	<i>Cyttarocyliis magna</i>	40	20	0
23	<i>Daturella</i> sp	40	0	0
24	<i>Dictyocysta elegans</i>	40	80	0
25	<i>Didinium nasutum</i>	20	0	20
26	<i>Dysteria compressa</i>	20	60	0
27	<i>Epiplocyloides reticulata</i>	0	0	40
28	<i>Euplotes patella</i>	0	0	20
29	<i>Eutintinnus apertus</i>	0	20	20
30	<i>Eutintinnus attemtor</i>	20	0	0
31	<i>Eutintinnus attenuatus</i>	0	0	20
32	<i>Eutintinnus colligatus</i>	20	20	20
33	<i>Eutintinnus fraknoi</i>	20	0	20
34	<i>Eutintinnus lususundae</i>	0	40	0
35	<i>Eutintinnus rectus</i>	20	20	20
36	<i>Eutintinnus rugosus</i>	0	0	40
37	<i>Eutintinnus</i> sp	0	40	0
38	<i>Eutintinnus stramentus</i>	20	20	60
39	<i>Favella azorica</i>	0	20	0
40	<i>Favella campanula</i>	20	0	20

Table continues on next column.....

S. No.	Species	ST1	ST2	ST3
41	<i>Favella ehrenbergii</i>	40	40	0
42	<i>Geleia</i> sp	140	60	20
43	<i>Gruberia foissneri</i>	20	0	0
44	<i>Gruberia lanceolata</i>	100	120	100
45	<i>Helicostomella edentata</i>	20	20	0
46	<i>Helicostomella longa</i>	20	0	40
47	<i>Helicostomella subulata</i>	0	20	0
48	<i>Holosticha</i> sp	0	100	0
49	<i>Kentrophoros</i> sp	0	40	0
50	<i>Laackmanniella</i> sp	20	0	0
51	<i>Leprotintinnus simplex</i>	220	160	20
52	<i>Leprotintinnus nordqvistii</i>	80	20	40
53	<i>Leprotintinnus pellucidum</i>	60	20	0
54	<i>Litonotus fasciola</i>	80	60	100
55	<i>Mesodinium rubrum</i>	0	20	0
56	<i>Metacylis</i> sp	0	20	20
57	<i>Opercularia</i> sp	0	0	20
58	<i>Parallelostrombidium jankowski</i>	40	0	20
59	<i>Paramecium</i> sp	40	0	40
60	<i>Pelagacinetia interrupta</i>	0	0	20
61	<i>Petalotricha ampulla</i>	0	0	20
62	<i>Petalotricha major</i>	0	20	0
63	<i>Poroecus curtus</i>	0	0	20
64	<i>Protorhabdonella simplex</i>	40	80	0
65	<i>Ptychocyliis obtusa</i>	0	0	20
66	<i>Rhabdonella amor</i>	0	0	20
67	<i>Rhabdonella</i> sp	0	20	0
68	<i>Salpingacantha ampla</i>	200	200	100
69	<i>Salpingacantha nana</i>	0	40	0
70	<i>Salpingacantha pellucidum</i>	40	0	0
71	<i>Salpingacantha undata</i>	20	0	20
72	<i>Salpingacantha unguiculata</i>	20	0	40
73	<i>Salpingella acuminata</i>	200	200	200
74	<i>Salpingella ampla</i>	20	0	60
75	<i>Salpingella attenuata</i>	100	60	80
76	<i>Salpingella costata</i>	0	0	20
77	<i>Salpingella regulata</i>	0	0	20
78	<i>Salpingella rotundata</i>	100	80	60
79	<i>Spirostomum minus</i>	20	0	80
80	<i>Spirostomum</i> sp	80	80	40
81	<i>Steenstrupiella intumescens</i>	0	20	40
82	<i>Stentor polymorphus</i>	0	0	20
83	<i>Stentor</i> sp	0	40	0
84	<i>Strobilidium spiralis</i>	40	100	0
85	<i>Strombidinopsis</i> sp	20	0	20

Table continues on next page.....

S. No.	Species	ST1	ST2	ST3
86	<i>Strombidium conicoides</i>	0	0	20
87	<i>Strombidium conicum</i>	120	40	40
88	<i>Strombidium diversum</i>	0	20	20
89	<i>Strombidium guangdongense</i>	0	0	20
90	<i>Strombidium</i> sp	60	60	0
91	<i>Stylicauda platensis</i>	0	20	20
92	<i>Suctorina acineta</i>	20	0	0
93	<i>Tintinnopsis amphistoma</i>	0	20	0
94	<i>Tintinnopsis aperta</i>	160	40	40
95	<i>Tintinnopsis balechi</i>	40	20	40
96	<i>Tintinnopsis baltica</i>	0	20	20
97	<i>Tintinnopsis beroidea</i>	220	200	120
98	<i>Tintinnopsis campanula</i>	0	20	0
99	<i>Tintinnopsis corniger</i>	40	20	60
100	<i>Tintinnopsis cylindrical</i>	120	80	160
101	<i>Tintinnopsis dadayi</i>	20	0	20
102	<i>Tintinnopsis directa</i>	20	0	0
103	<i>Tintinnopsis esturiensis</i>	0	40	0
104	<i>Tintinnopsis everta</i>	0	20	20
105	<i>Tintinnopsis fimbriata</i>	0	20	0
106	<i>Tintinnopsis gracilis</i>	80	300	160
107	<i>Tintinnopsis lobiancoi</i>	0	20	20
108	<i>Tintinnopsis major</i>	0	0	40
109	<i>Tintinnopsis nana</i>	40	40	60
110	<i>Tintinnopsis orientalis</i>	40	80	60
111	<i>Tintinnopsis parva</i>	100	20	80
112	<i>Tintinnopsis parvula</i>	80	0	20
113	<i>Tintinnopsis radix</i>	60	80	40
114	<i>Tintinnopsis rapa</i>	0	20	0
115	<i>Tintinnopsis rotundata</i>	20	20	40
116	<i>Tintinnopsis stenosemella</i>	0	40	0
117	<i>Tintinnopsis tocaninensis</i>	80	20	20
118	<i>Tintinnopsis compressa</i>	40	0	0
119	<i>Tokophrya</i> sp	0	0	20
120	<i>Tracheloraphis phoenicopterus</i>	0	0	20
121	<i>Undella globosa</i>	20	20	20
122	<i>Undella hemispherica</i>	0	0	20
123	<i>Undella hyalina</i>	40	100	20
124	<i>Undella pentagona</i>	0	20	0
125	<i>Undella subacuta</i>	20	20	0
126	<i>Undella turgida</i>	40	0	20
127	<i>Uroleptus</i> sp	0	0	20
128	<i>Zoothamnium elegans</i>	20	0	20
Genera: 56; Species: 128		4300	3780	3480

Rare = 1-150 * Common = 151-250** Dominant = 251-350< ***

Table II. Seasonal abundance of ciliates (cells/L) recorded from Sandspit.

S. No.	Species	ST1	ST2
1	<i>Amphorella brandti</i>	0	20
2	<i>Anigstenia clarissima</i>	220	200
3	<i>Chaenea teres</i>	140	40
4	<i>Codonella galea</i>	60	0
5	<i>Codonella nationalis</i>	40	0
6	<i>Codonellopsis gaussi</i>	20	20
7	<i>Codonellopsis morchella</i>	20	0
8	<i>Cyttarocyliis magna</i>	40	40
9	<i>Dadayiella</i> sp	0	60
10	<i>Epiplocyliis blanda</i>	20	20
11	<i>Epiplocyliis undella</i>	20	0
12	<i>Eutintinnus apertus</i>	40	200
13	<i>Eutintinnus elongatus</i>	20	120
14	<i>Eutintinnus fraknoi</i>	0	20
15	<i>Eutintinnus rectus</i>	40	0
16	<i>Eutintinnus stramentus</i>	60	20
17	<i>Favella azorica</i>	20	20
18	<i>Favella ehrenbergii</i>	40	0
19	<i>Favella markusouzkii</i>	40	0
20	<i>Gruberia lanceolata</i>	120	100
21	<i>Helicostomella subulata</i>	0	20
22	<i>Laboea strobila</i>	20	20
23	<i>Lacrymaria olor</i>	20	40
24	<i>Leprotintinnus nordqvistii</i>	120	80
25	<i>Leprotintinnus pellucidum</i>	20	20
26	<i>Leprotintinnus simplex</i>	160	320
27	<i>Litonotus fasciola</i>	100	60
28	<i>Mesodinium rubrum</i>	80	40
29	<i>Metacyliis jorgensenii</i>	0	20
30	<i>Paramecium</i> sp	40	0
31	<i>Parundella aculeata</i>	20	20
32	<i>Petalotricha ampulla</i>	20	0
33	<i>Philasterides armatali</i>	20	0
34	<i>Protorhabdonella striatura</i>	0	20
35	<i>Ptychocyliis obtusa</i>	0	20
36	<i>Ptychocyliis</i> sp	60	0
37	<i>Salpingacantha ampla</i>	360	120
38	<i>Salpingacantha perca</i>	0	40
39	<i>Salpingacantha undata</i>	0	20
40	<i>Salpingacantha unguiculata</i>	20	0
41	<i>Salpingella acuminata</i>	260	260
42	<i>Salpingella attenuata</i>	140	40

Table continues on next page.....

S. No.	Species	ST1	ST2
43	<i>Salpingella decurtata</i>	80	40
44	<i>Salpingella rotundata</i>	40	20
45	<i>Spirostomum ambiguum</i>	80	40
46	<i>Spirostomum minus</i>	180	220
47	<i>Steenstrupiella gracilis</i>	160	80
48	<i>Steenstrupiella inteumescens</i>	20	0
49	<i>Steenstrupiella steenstrupii</i>	40	0
50	<i>Stenosemella sp</i>	20	60
51	<i>Strobilidium spiralis</i>	40	40
52	<i>Strombidium conicum</i>	220	260
53	<i>Strombidium elongatum</i>	120	40
54	<i>Strombidium oculatum</i>	60	40
55	<i>Thuricola folliculata</i>	20	20
56	<i>Tintinnopsis balechi</i>	20	0
57	<i>Tintinnopsis beroidea</i>	160	0
58	<i>Tintinnopsis campanula</i>	180	120
59	<i>Tintinnopsis choroestrichids</i>	20	0
60	<i>Tintinnopsis corniger</i>	20	0
61	<i>Tintinnopsis cylindrical</i>	40	40
62	<i>Tintinnopsis dadayi</i>	180	100
63	<i>Tintinnopsis everta</i>	20	0
64	<i>Tintinnopsis fistularis</i>	20	20
65	<i>Tintinnopsis gracilis</i>	100	40
66	<i>Tintinnopsis hemispiralis</i>	0	20
67	<i>Tintinnopsis karajacensis</i>	20	0
68	<i>Tintinnopsis kofoidi</i>	20	0
69	<i>Tintinnopsis lobiancoi</i>	40	60
70	<i>Tintinnopsis nana</i>	20	0
71	<i>Tintinnopsis parva</i>	120	200
72	<i>Tintinnopsis parvula</i>	20	40
73	<i>Tintinnopsis radix</i>	120	140
74	<i>Tintinnopsis redixand</i>	40	0
75	<i>Tintinnopsis rotundata</i>	80	200
76	<i>Tintinnopsis tocaninensis</i>	100	60
77	<i>Tintinnopsis ventricosoides</i>	40	20
78	<i>Trachelophyllum apiculatum</i>	20	0
79	<i>Trochilia sigmoides</i>	20	60
80	<i>Undella hyalina</i>	120	100
81	<i>Undella subacuta</i>	20	0
82	<i>Undella turgida</i>	20	20
83	<i>Xystonella treforti</i>	0	20
Genera: 37, Species: 83		5080	4160

Rare= 1-150 * Common= 151-250** Dominant= 251-350< ***

Table III. Seasonal abundance of ciliates (cells/L) recorded from Gadani and Sandspit.

Localities/ No. of seasons	Total No. of individuals (Cells/L)	Margalef richness index	Pielou's evenness index	Shanon diversity index
Gadani				
AIM	88	0.44669	0.99988	1.0985
NEM	161	0.39359	0.97338	1.0694
SIM	80	0.45641	0.93723	1.0297
SWM	250	0.36222	0.98401	1.081
Sandspit				
AIM	88	0.22335	0.99664	0.69082
NEM	118	0.20961	0.93925	0.65104
SIM	80	0.2282	0.97095	0.67301
SWM	176	0.19341	0.99767	0.69153

For details of abbreviations, see Figure 2.

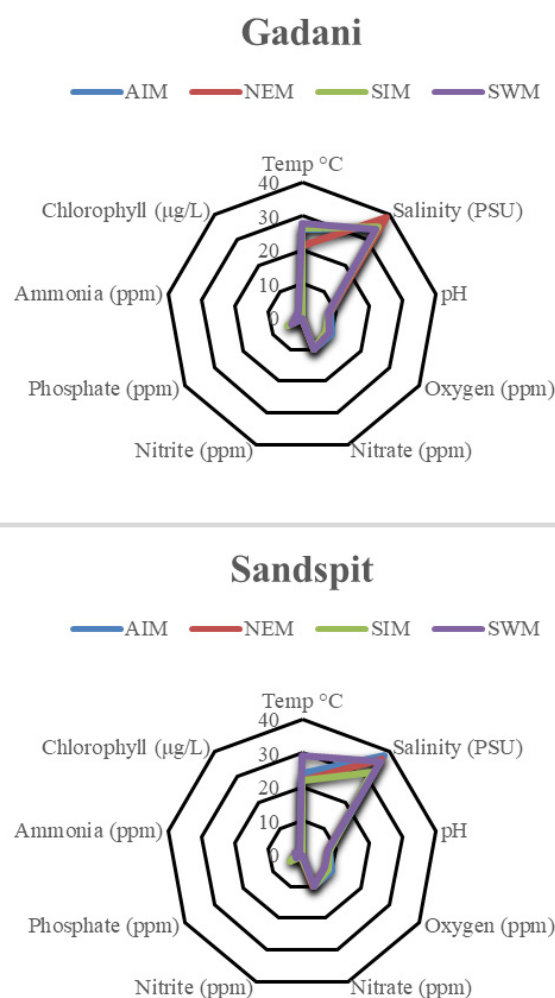


Fig. 1. Physico-chemical parameters recorded from Gadani and Sandspit. For details of abbreviations, see Figure 2.

as compared to Sandspit. The total number of 128 species of ciliates classified into 56 genera from Gadani and 83 species of ciliates classified into 37 genera from Sandspit were recorded. Twenty-six species of *Tintinnopsis* were observed in Gadani, whereas twenty-two species of *Tintinnopsis* were observed in Sandspit (Tables I, II). The seasonal abundance of ciliates genera (cells/L) from Gadani and Sandspit are depicted in Figures 2 and 3. The high values of the Shannon diversity index were 1.0985 in Gadani and 0.69153 in Sandspit. Richness was high in SIM on both sites. Evenness was high in AIM in Gadani while SWM in Sandspit (Table III). The physicochemical parameters recorded from Gadani and Sandspit are shown in Figure 1.

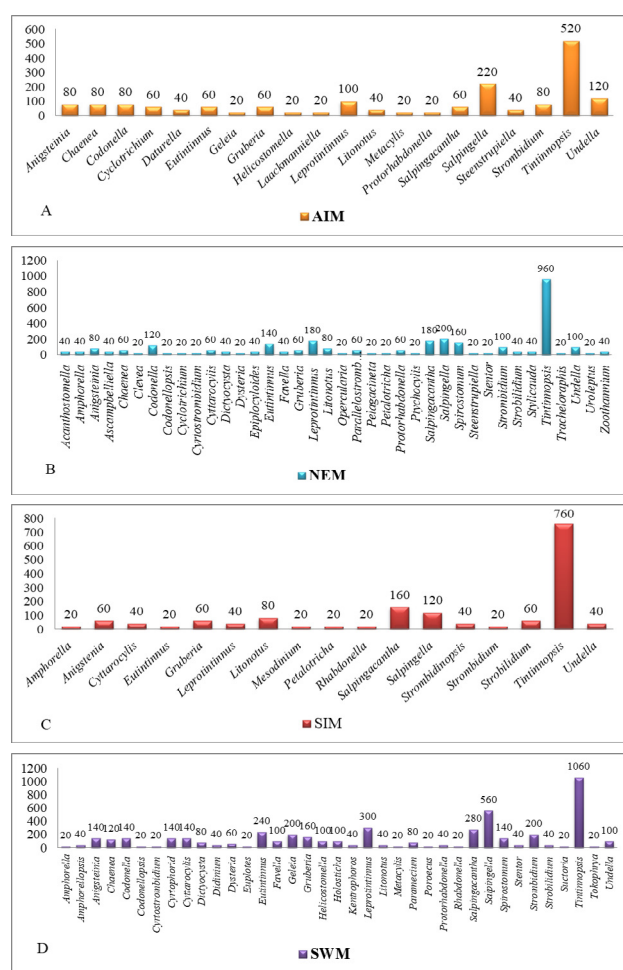


Fig. 2. Ciliate genera (cells/L) in the seasonal abundance from Gadani: A, autumn intermonsoon (AIM); B, northeast monsoon (NEM); C, spring intermonsoon (SIM); D, southwest monsoon (SWM).

Pearson correlation coefficient (Table IV) was used to detect the association among ciliate communities

with hydrographical parameters and nutrients. Ciliates abundance was correlated with salinity, dissolved oxygen and chlorophyll *a* while negative correlation was detected with temperature, pH, nitrite, nitrate, phosphate and ammonia in station 1 at Gadani. Whereas in station 2, Ciliates abundance was negatively correlated with hydrographical parameters and nutrients. In station 3, ciliates abundance was correlated with salinity, pH, dissolved oxygen, nitrite, nitrate, ammonia and chlorophyll *a* while negative correlation was detected with temperature and phosphate. In Sandspit (Table V) in station 1, ciliates abundance was negatively correlated with hydrographical parameters and nutrients. While in station 2, ciliates abundance was positively correlated with temperature, salinity, pH,

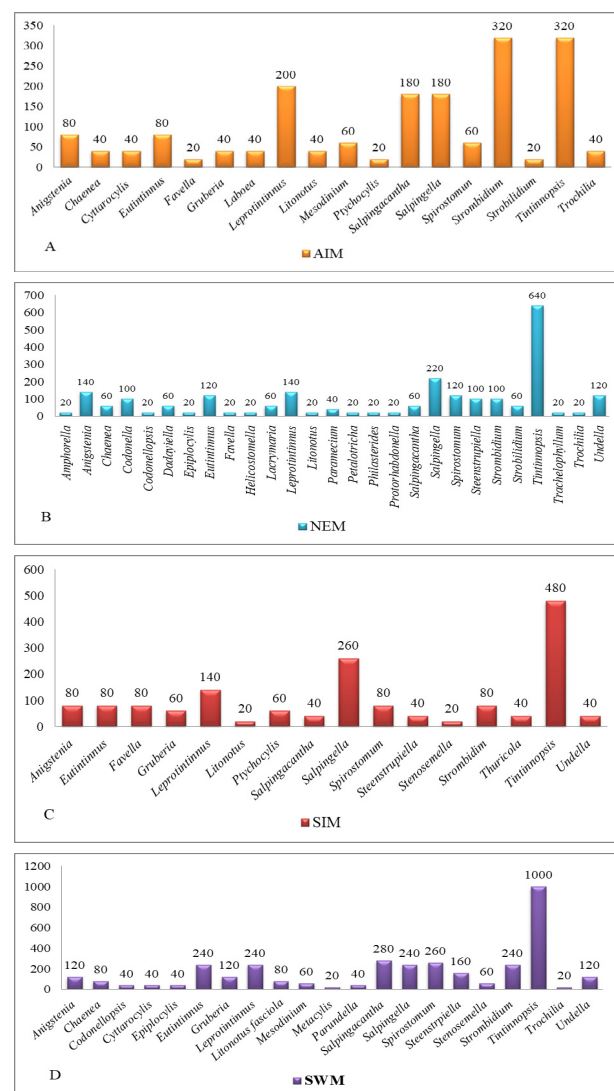


Fig. 3. Ciliate genera (cells/L) in the seasonal abundance from Sandspit. For details of seasonal change, see Figure 2.

Table IV. Pearson correlation coefficient between ciliates communities with environmental variables in Gadani.

ST 1	Abundance	Temp	Salinity	pH	Oxygen	Nitrate	Nitrite	Phosphate	Ammonia
Temp	-0.267								
Salinity	0.012**	-0.169							
pH	-0.079	0.101 ^{ns}	0.791 ^{ns}						
Oxygen	0.123 ^{ns}	0.061	0.477 ^{ns}	0.843 ^{ns}					
Nitrate	-0.077	0.077	0.825 ^{ns}	0.988 ^{ns}	0.810 ^{ns}				
Nitrite	-0.038	0.083	0.849 ^{ns}	0.951 ^{ns}	0.763 ^{ns}	0.985 ^{ns}			
Phosphate	-0.276	0.378 ^{ns}	0.304 ^{ns}	0.435 ^{ns}	0.308 ^{ns}	0.475 ^{ns}	0.509 ^{ns}		
Ammonia	-0.068	0.272 ^{ns}	0.708 ^{ns}	0.876 ^{ns}	0.689 ^{ns}	0.875 ^{ns}	0.854 ^{ns}	0.299 ^{ns}	
Chlorophyll	0.482 ^{ns}	0.405 ^{ns}	-0.205	0.021*	0.358 ^{ns}	0.026*	0.07	0.149 ^{ns}	0.157 ^{ns}
ST 2									
Temp	-0.282								
Salinity	-0.512	-0.169							
pH	-0.456	0.101 ^{ns}	0.791 ^{ns}						
Oxygen	-0.378	0.061	0.477 ^{ns}	0.843 ^{ns}					
Nitrate	-0.406	0.077	0.825 ^{ns}	0.988 ^{ns}	0.810 ^{ns}				
Nitrite	-0.37	0.083	0.849 ^{ns}	0.951 ^{ns}	0.763 ^{ns}	0.985 ^{ns}			
Phosphate	-0.138	0.378 ^{ns}	0.304 ^{ns}	0.435 ^{ns}	0.308 ^{ns}	0.475 ^{ns}	0.509 ^{ns}		
Ammonia	-0.496	0.272 ^{ns}	0.708 ^{ns}	0.876 ^{ns}	0.689 ^{ns}	0.875 ^{ns}	0.854 ^{ns}	0.299 ^{ns}	
Chlorophyll	-0.12	0.405 ^{ns}	0.205 ^{ns}	0.021*	0.358 ^{ns}	0.026*	0.07	0.149 ^{ns}	0.157 ^{ns}
ST 3									
Temp	-0.307								
Salinity	0.302 ^{ns}	-0.169							
pH	0.165 ^{ns}	0.101 ^{ns}	0.791 ^{ns}						
Oxygen	0.130 ^{ns}	0.061	0.477 ^{ns}	0.843 ^{ns}					
Nitrate	0.138 ^{ns}	0.077	0.825 ^{ns}	0.988 ^{ns}	0.810 ^{ns}				
Nitrite	0.128 ^{ns}	0.083	0.849 ^{ns}	0.951 ^{ns}	0.763 ^{ns}	0.985 ^{ns}			
Phosphate	-0.164	0.378 ^{ns}	0.304 ^{ns}	0.435 ^{ns}	0.308 ^{ns}	0.475 ^{ns}	0.509 ^{ns}		
Ammonia	0.209 ^{ns}	0.272 ^{ns}	0.708 ^{ns}	0.876 ^{ns}	0.689 ^{ns}	0.875 ^{ns}	0.854 ^{ns}	0.299 ^{ns}	
Chlorophyll	0.268 ^{ns}	0.405 ^{ns}	0.205 ^{ns}	0.021*	0.358 ^{ns}	0.026*	0.07	0.149 ^{ns}	0.157 ^{ns}

*Represents significant at $p < 0.05$, **represents significant at $p < 0.01$ and ns represents non-significant.

dissolved oxygen, nitrate, nitrite and ammonia, while a negative correlation was detected with phosphate and chlorophyll *a*.

DISCUSSION

In Sandspit and Gadani, maximum abundance and diversity of ciliates was recorded in the SWM than in the SIM, AIM and NEM. In the Southwest Monsoon, the more vigorous upwelling in the Northern Indian Ocean leads to high primary productivity (Goes, 2005). High primary productivity may be linked to temperature that increases during the SWM (May to September). Previous studies reported that the ciliate diversity is greatly influenced by

temperature and salinity (Xu *et al.*, 2018). Temperature affects the ciliates primarily by controlling their growth (Montagnes and Lessard, 1999). The salinity of the water increases with the rise in temperature. The ciliates can tolerate extreme changes in salinity, and some ciliates can withstand direct transfer from marine coastal areas to fresh waters (Smurov *et al.*, 2013). In high temperatures in the SWM, the organisms demand for oxygen increases, resulting in low dissolved oxygen retaining capacity of water (Hussain *et al.*, 2013). Ciliates are sensitive to changes in the concentration of oxygen in water (Fenchel, 2012). The diversity of the ciliates in marine waters also depends on the constancy of the oxygen gradients.

Table V. Pearson correlation coefficient between ciliates communities with environmental variables in Sandspit.

ST 1	Abundance	Temp	Salinity	PH	Oxygen	Nitrate	Nitrite	Phosphate	Ammonia
Temp	-0.607								
Salinity	-0.318	0.158							
pH	-0.107	-0.08	-0.107						
Oxygen	-0.007	-0.104	-0.109	0.096					
Nitrate	-0.214	-0.019	0.421 ^{ns}	-0.383	0.134 ^{ns}				
Nitrite	-0.344	-0.035	-0.053	0.379 ^{ns}	0.089	0.478 ^{ns}			
Phosphate	-0.103	-0.167	-0.297	0.448 ^{ns}	-0.171	-0.096	0.288 ^{ns}		
Ammonia	-0.117	0.243 ^{ns}	0.435 ^{ns}	-0.126	0.006**	0.539 ^{ns}	0.057*	-0.449	
Chlorophyll	-0.295	0.470 ^{ns}	0.118 ^{ns}	-0.61	0.237 ^{ns}	0.441 ^{ns}	-0.076	-0.335	0.301 ^{ns}
ST 2									
Temp	0.233 ^{ns}								
Salinity	0.102 ^{ns}	0.158 ^{ns}							
pH	0.217 ^{ns}	-0.08	-0.107						
Oxygen	0.127 ^{ns}	-0.104	-0.109	0.096					
Nitrate	0.190 ^{ns}	-0.019	0.421 ^{ns}	0.383	0.134 ^{ns}				
Nitrite	0.422 ^{ns}	-0.035	-0.053	0.379 ^{ns}	0.089	0.478			
Phosphate	-0.35	-0.167	-0.297	0.448 ^{ns}	-0.171	-0.096	0.288 ^{ns}		
Ammonia	0.387 ^{ns}	0.243 ^{ns}	0.435 ^{ns}	-0.126	0.006**	0.539 ^{ns}	0.05*	-0.449	
Chlorophyll	-0.134	0.470 ^{ns}	0.118 ^{ns}	-0.61	0.237 ^{ns}	0.441 ^{ns}	-0.07	-0.335	0.301 ^{ns}

*Represents significant at $p < 0.05$, **represents significant at $p < 0.01$ and ns represents non-significant.

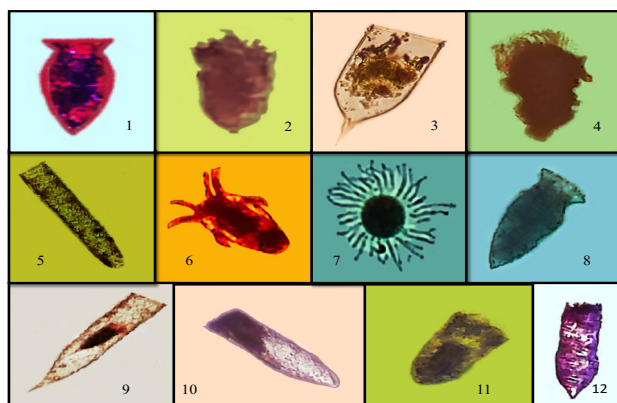


Fig. 4. Planktonic ciliates from Gadani (Baluchistan) and Sandspit (Karachi). 1, *Codonellopsis* sp.; 2, *Favella azorica*; 3, *Favella eherebergii*; 4, *Laboea strobila*; 5, *Leprotintinnus simplex*; 6, *Mesodinium rubrum*; 7, *Strobilidium spiralis*; 8, *Strombidinopsis* sp.; 9, *Tintinnopsis corniger*; 10, *Tintinnopsis cylindrical*; 11, *Tintinnopsis gracilis*; 12, *Tintinnopsis tocaninensis*.

In the SWM, the peak abundance of ciliates was observed due to high chlorophyll *a* content in Gadani and Sandspit. Our results are in agreement with Soriede *et al.* (2010), who stated that the abundance of

microzooplankton may be linked to the high chlorophyll *a* in the area. Chlorophyll *a* distribution depends on physico-chemical concentrations, for example, nutrients and temperature (Lakkis *et al.*, 2003). Nutrients and climate changes (wind patterns and rainfall) in coastal areas also influence ciliate diversity and communities (Lopez-Abbate *et al.*, 2019; Zhu *et al.*, 2020). The diversity of species in Gadani was more remarkable as compared to Sandspit. The total number of 128 species of ciliates classified into 56 genera from Gadani and 83 species of ciliates classified into 37 genera from Sandspit were recorded. The abundance, diversity and survival of organisms initiate in favourable environments, nutrient-rich and less predation sites (Cocheret de la Moriniere *et al.*, 2004). Pollutants in marine environments are known to reduce species diversity and increase the population of tolerant species. In Sandspit, the most dominant ciliate species were *Leprotintinnus simplex*, *Salpingacantha ampla*, *Salpingella acuminata*, *Spirostomum minus* and *Strombidium conicum*. However, in Gadani, the most dominant species of ciliates were *Salpingella acuminata*, *Tintinnopsis beroidea* and *Tintinnopsis gracilis*.

In Sandspit and Gadani, we observed that the diversity of ciliates increases in near-shore waters as compared to shore and offshore waters. This is in agreement with

previous studies that the diversity of ciliates reduced with an increase in distance from shore (Tamura *et al.*, 2011). Ocean currents impact the waters from oceanic and neritic zones and are homogenous with the same hydrological characteristics, resulting in remarkable similarity of organisms. The importance of ciliates in energy transfer in marine food webs is well-known in ecological function (Fenchel, 1988). The abundance of ciliates in marine waters is controlled by zooplankton, especially filter-feeding copepods (Atkinson, 1996). Ciliates are primary grazers on bacterioplankton and nanoplankton (Premke and Arndt, 2000). Moreover, planktonic algae, bacteria and mesozooplankton have substantial effects on the diversity and abundance of ciliate communities (Yang *et al.*, 2020). Many scientists have confirmed that ciliate abundance can be influenced by environmental factors comprising nutrients, pH, salinity, temperature and biotic interactions, for example, predators (Gimmler *et al.*, 2016; Sun *et al.*, 2017). However, spatial factors have also been considered in the study of ciliate community assemblage. The limitation in dispersal would lead to a decrease in community similarity with distance (Pan *et al.*, 2020). The effect of environmental and spatial variables on ciliates depends on the study scale and kind of environment (Zhang *et al.*, 2018). The ciliate community structure in the mesopelagic zone is controlled by geographic distance and ocean depth (Sun *et al.*, 2019).

Planktonic ciliates are frequently dominated by aloricate ciliates (Leakey *et al.*, 1996). However, many studies are focused on tintinnids (Rakshit *et al.*, 2014) owing to difficulties in the identification which mislead the contribution of these aloricate ciliates. Previous studies reported aloricate ciliates numerically dominant than tintinnids in the central and western Arabian Sea (Leakey *et al.*, 1996) in the Northern Arabian Sea (Siddiqui *et al.*, 2000; Burhan *et al.*, 2018). However, in our studies, tintinnids were more dominant than aloricate in both Gadani and Sandspit. From Sandspit, eighteen species of aloricate ciliates are recorded, while loricate is sixty-five species in number. In Gadani, thirty-eight species of aloricate ciliates are recorded, while loricate is ninety species in number.

Tintinnids are unicellular loricate ciliates (Montagnes, 2013) that inhabit freshwaters and marine environments (McManus and Santferrara, 2013). They play an essential role in the food chain that feeds on bacteria and phytoplankton, they, in turn, serve as food for larger marine organisms, for example, copepods and fish larvae (Stoecker, 2013). Tintinnids are used as bioindicators to assess environmental stress and anthropogenic impacts on marine ecosystems (Jiang *et al.*, 2011; Xu *et al.*, 2011) and to monitor aquatic water quality (Wu *et al.*, 2016). Due to

their delicate pellicles and short life cycles, they respond quickly to environmental changes (Ismail and Dorgham, 2003). Twenty-six species of *Tintinnopsis* are present in Gadani, whereas twenty-two species of *Tintinnopsis* are present in Sandspit. Many ciliates are more resistant to extreme environmental conditions than macrofauna (Xu *et al.*, 2011). The dominant species of *Tintinnopsis* in Gadani and Sandspit coastal waters agrees with that reported by Jiang *et al.* (2011) and Feng *et al.* (2015). *Tintinnopsis* abundance may be related to their adaptive nature or sustaining in eurythermal and euryhaline aquatic environments. The ciliates are vulnerable to environmental variants, and the pollution arising from industrial units alongside the coastal areas is hazardous for fish. There is an essential requirement for monitoring of ciliates with respect to abundance, diversity, distribution and harmful algal bloom-forming species as it affects the fishery industry and marine environment.

DECLARATIONS

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Conflict of interest

The authors declare that there is no conflict of interest regarding the publication of this article.

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