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

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#### **ABSTRACT**

moment of the microzooplankton and occupy a significant role in the microbial<br>tudy, seawater samples were collected in four seasons from the Gadani<br>stigs it coast for one year. The seasonal diversity of planktonic ciliates 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.

# **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](#page-11-0) *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](#page-10-0)). Mixotrophic ciliates are found simply in the aloricate sub-group ([Stoecker](#page-11-0) *et al*., 1987) in the order Oligotrichida. According to the group division by Flynn *et al*. (2019) mixotrophic ciliates are part of

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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](#page-9-1)). 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 [\(Crawford, 1989](#page-9-2)). Due to the blooms of *Mesodinium rubrum*, oxygen depletion occurs, which leads to fish kill in the coastal areas (Pau *et al*[., 2017\)](#page-10-1).

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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](#page-11-1)). Ciliates are found growing in some extreme environmental conditions, having sufficient vital energy to endure it [\(Lynn, 2008\)](#page-10-2). Anaerobic ciliates are reported from anoxic environments, including marine, freshwater sediments, deep basins of estuaries and the anoxic hypolimnia of lakes [\(Finlay, 1982](#page-9-3); Finlay *et al*[., 1991](#page-9-4); [Fenchel](#page-9-5) *et al*., 1995; Xu *et al*., 2013). Ciliates are sporadically reported living in hot springs at temperatures greater than 40°C [\(Kahan,](#page-10-3) [1972](#page-10-3)). They are frequently found in several submarine hydrothermal vents. Twenty species of ciliates on the East Pacific Rise hydrothermal vents were reported by [Small](#page-10-4) [and Gross \(1985\)](#page-10-4). Ciliates were also reported in salt lakes having a pH value of 9.5 (Wilbert, 1995).

**Examp[le](#page-11-3) 1** is a the th[r](#page-10-5)ee that in the states were also reported in salt lakes<br>
to Strickland and Parsons (1972)<br>
5 (Wilbert, 1995).<br>
Son (2005) reported heterotrophic *Abundance and diversity of the casaline environments.* [Hauer and Rogerson \(2005\)](#page-9-6) 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](#page-10-5)). 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\)](#page-11-2). 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](#page-8-0); [Caron](#page-9-7) *et al*., 1985). Ciliates have multifaceted ecological roles due to their morphological, trophic, genetic and metabolic diversity [\(Caron, 2016](#page-8-1)). These varied qualities shape the interspecific (parasitism, predation, etc.) interactions and speciesenvironment and have a significant role in the assembling of marine ciliate communities ([Fuhrman](#page-9-8) *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](#page-2-0)**-**[III](#page-4-0)). Ciliates species diversity in Gadani was more significant

<span id="page-2-0"></span>**Table I. Seasonal abundance of ciliates (cells/L) recorded from Gadani.**

					42	Geleia sp
S. No.	<b>Species</b>	ST1	ST2	ST <sub>3</sub>	43	Gruberia foissneri
$\mathbf{1}$	Acanthostomella minutissima	$\boldsymbol{0}$	$\boldsymbol{0}$	20	44	Gruberia lanceolata
$\overline{2}$	Acanthostomella norvegica	20	$\boldsymbol{0}$	$\boldsymbol{0}$	45	Helicostomella edentata
3	Amphorella brandti	20	$\overline{0}$	$\overline{0}$	46	Helicostomella longa
4	Amphorella minor	40	$\overline{0}$	20	47	Helicostomella subulata
5	Amphorellopsis acuta	40	$\mathbf{0}$	$\boldsymbol{0}$	48	Holosticha sp
6	Anigsteinia clarissima	140	180	40	49	Kentrophoros sp
7	Ascampbelliella retusa	$\boldsymbol{0}$	20	20	50	Laackmanniella sp
$\,$ 8 $\,$	Chaenea teres	160	20	80	51	Leprotintinnus simplex
9	Clevea melchersi	$\boldsymbol{0}$	$\overline{0}$	20	52	Leprotintinnus nordqvistii
10	Codonella aspera	$\boldsymbol{0}$	$\boldsymbol{0}$	40	53	Leprotintinnus pellucidum
11	Codonella daday	20	$\mathbf{0}$	40	54	Litonotus fasciola
12	Codonella galea	20	20	40	55	Mesodinium rubrum
13	Codonella nationalis	20	40	100	56	Metacylis sp
14	Codonellopsis morchella	$\boldsymbol{0}$	$\boldsymbol{0}$	20	57	Opercularia sp
15	Codonellopsis schabi	$\boldsymbol{0}$	$\mathbf{0}$	20	58	Parallelostrombidium janko
16	Cyclotrichium gigas	$\boldsymbol{0}$	$\overline{0}$	40	59	Paramecium sp
17	Cyclotrichium sp	40	$\overline{0}$	$\boldsymbol{0}$	60	Pelagacineta interrupta
18	Cyrtophorid sp	140	$\boldsymbol{0}$	$\boldsymbol{0}$	61	Petalotricha ampulla
19	Cyrtostrombidium longisomum	20	20	0	62	Petalotricha major
20	Cyttarocylis brandti	40	40	40	63	Poroecus curtus
21	Cyttarocylis conica	20	20	20	64	Protorhabdonella simplex
22	Cyttarocylis magna	40	20 <sup>2</sup>	$\boldsymbol{0}$	65	Ptychocylis obtusa
23	Daturella sp	40	$\overline{0}$	$\boldsymbol{0}$	66	Rhabdonella amor
24	Dictyocysta elegans	40	80	$\boldsymbol{0}$	67	Rhabdonella sp
					68	Salpingacantha ampla
25	Didinium nasutum	20	$\boldsymbol{0}$	20	69	Salpingacantha nana
26	Dysteria compressa	20	60	$\boldsymbol{0}$	70	Salpingacantha pellucidum
27	Epiplocyloides reticulata	$\boldsymbol{0}$	$\overline{0}$	40	71	Salpingacantha undata
28	Euplotes patella	$\boldsymbol{0}$	$\boldsymbol{0}$	20	72	Salpingacantha unguiculata
29	Eutintinnus apertus	$\boldsymbol{0}$	20	20	73	Salpingella acuminata
30	Eutintinnus attemtor	20	$\mathbf{0}$	$\boldsymbol{0}$	74	Salpingella ampla
31	Eutintinnus attenuatus	$\boldsymbol{0}$	$\boldsymbol{0}$	20	75	Salpingella attenuata
32	Eutintinnus colligatus	20	$20\,$	20	76	Salpingella costata
33	Eutintinnus fraknoii	20	$\overline{0}$	20	77	Salpingella regulata
34	Eutintinnus lususundae	$\overline{0}$	40	$\boldsymbol{0}$	78	Salpingella rotundata
35	Eutintinnus rectus	20	20	20	79	Spirostomum minus
36	Eutintinnus rugosus	$\overline{0}$	$\overline{0}$	40	80	Spirostomum sp
37	Eutintinnus sp	$\overline{0}$	40	$\boldsymbol{0}$	81	Steenstrupiella intumescens
38	Eutintinnus stramentus	20	20	60	82	Stentor polymorphus
39	Favella azorica	$\boldsymbol{0}$	20	$\boldsymbol{0}$	83	Stentor sp
					84	Strobilidium spiralis
40	Favella campanula	20	$\overline{0}$	20	85	Strombidinopsis sp
	Table continues on next column		Table contil			





 $\overline{a}$ 

S. No.	<b>Species</b>	ST <sub>1</sub>	ST2	ST3		Table II. Seasonal abundano
86	Strombidium conicoides	0	$\overline{0}$	20		recorded from Sandspit.
87	Strombidium conicum	120	40	40		
88	Strombidium diversum	$\boldsymbol{0}$	20	20	S. No.	<b>Species</b>
89	Strombidium guangdongense	$\overline{0}$	$\mathbf{0}$	20	1	Amphorella brandti
90	Strombidium sp	60	60	$\boldsymbol{0}$	$\boldsymbol{2}$	Anigstenia clarissima
91	Stylicauda platensis	0	20	20	3	Chaenea teres
92	Suctoria acineta	20	$\overline{0}$	$\boldsymbol{0}$	4	Codonella galea
93	Tintinnopsis amphistoma	$\boldsymbol{0}$	20	$\boldsymbol{0}$	5	Codonella nationalis
94	Tintinnopsis aperta	160	40	40	6	Codonellopsis gaussi
95	Tintinnopsis balechi	40	20	40	7	Codonellopsis morchella
96	Tintinnopsis baltica	$\boldsymbol{0}$	20	20	$\,$ $\,$	Cyttarocylis magna
97	Tintinnopsis beroidea	220	200	120	9	Dadayiella sp
98	Tintinnopsis campanula	$\boldsymbol{0}$	20	$\boldsymbol{0}$	10	Epiplocylis blanda
99	Tintinnopsis corniger	40	20	60	11	Epiplocylis undella
100	Tintinnopsis cylindrical	120	80	160	12	Eutintinnus apertus
101	Tintinnopsis dadayi	20	$\overline{0}$	20	13	Eutintinnus elongatus
102	Tintinnopsis directa	20	$\overline{0}$	$\boldsymbol{0}$	14	Eutintinnus fraknoii
103	Tintinnopsis esturiensis	$\boldsymbol{0}$	40	$\boldsymbol{0}$	15	Eutintinnus rectus
104	Tintinnopsis everta	$\boldsymbol{0}$	20	20	16	Eutintinnus stramentus
105	Tintinnopsis fimbriata	$\boldsymbol{0}$	20	$\boldsymbol{0}$	17	Favella azorica
106	Tintinnopsis gracilis	80	300	160	18	Favella ehrenbergii
107	Tintinnopsis lobiancoi	$\boldsymbol{0}$	20	20	19	Favella markusouzkyi
108	Tintinnopsis major	$\boldsymbol{0}$	$\boldsymbol{0}$	40	20	Gruberia lanceolata
109	Tintinnopsis nana	40	40	60	21	Helicostomella subulata
110	Tintinnopsis orientalis	40	80	60	22	Laboea strobila
111	Tintinnopsis parva	100	20	80	23	Lacrymaria olor
112	Tintinnopsis parvula	80	$\overline{0}$	20	24	Leprotintinnus nordqvistii
113	Tintinnopsis radix	60	80	40	25	Leprotintinnus pellucidum
114	Tintinnopsis rapa	$\overline{0}$	20	$\boldsymbol{0}$	26	Leprotintinnus simplex
115	Tintinnopsis rotundata	20	20	40	27	Litonotus fasciola
					28	Mesodinium rubrum
116	Tintinnopsis stenosemella	$\boldsymbol{0}$	40	$\boldsymbol{0}$	29	Metacylis jorgensenii
117	Tintinnopsis tocantinensis	80	20	20	30	Paramecium sp
118	Tintinopsis compressa	40	$\bf{0}$	$\bf{0}$	31	Parundella aculeata
119	Tokophrya sp	0	$\boldsymbol{0}$	20	32	Petalotricha ampulla
120	Tracheloraphis phoenicopterus	$\overline{0}$	$\boldsymbol{0}$	20	33	Philasterides armatali
121	Undella globosa	20	20	$20\,$	34	Protorhabdonella striatura
122	Undella hemispherica	$\overline{0}$	$\boldsymbol{0}$	20	35	Ptychocylis obtusa
123	Undella hyalina	40	100	20	36	Ptychocylis sp
124	Undella pentagona	$\mathbf{0}$	20	$\boldsymbol{0}$	37	Salpingacantha ampla
125	Undella subacuta	20	20	$\boldsymbol{0}$	38	Salpingacantha perca
126	Undella turgida	40	$\overline{0}$	20	39	Salpingacantha undata
					40	Salpingacantha unguiculate
127	Uroleptus sp	0	$\theta$	20	41	Salpingella acuminata
128	Zoothamnium elegans	20	$\theta$	20	42	Salpingella attenuata
	Genera: 56; Species: 128			4300 3780 3480		Table conti

<span id="page-3-0"></span>**Table II. Seasonal abundance of ciliates (cells/L) recorded from Sandspit.**



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

S. No.	<b>Species</b>	ST1	ST <sub>2</sub>	Table III. Seasonal abundan
43	Salpingella decurtata	80	40	recorded from Gadani and San
44	Salpingella rotundata	40	20	Localities/ <b>Total No. of</b> Marga
45	Spirostomum ambiguum	80	40	No. of individuals richnes
46	Spirostomum minus	180	220	(Cells/L) index seasons
47	Steenstrupiella gracilis	160	$80\,$	Gadani
48	Steenstrupiella inteumescens	20	$\boldsymbol{0}$	<b>AIM</b> 88 0.4466
49	Steenstrupiella steenstrupii	40	$\boldsymbol{0}$	<b>NEM</b> 161 0.3935
50	Stenosemella sp	20	60	<b>SIM</b> 80 0.4564
51	Strobilidium spiralis	40	40	<b>SWM</b> 250 0.3622
52	Strombidium conicum	220	260	Sandspit 88 AIM
53	Strombidium elongatum	120	40	0.2233 <b>NEM</b> 118 0.2096
54	Strombidium oculatum	60	40	<b>SIM</b> 80 0.2282
55	Thuricola folliculata	20	20	0.1934 <b>SWM</b> 176
56	Tintinnopsis balechi	20	$\boldsymbol{0}$	For details of abbreviations, see Figure 2.
57	Tintinnopsis beroidea	160	$\boldsymbol{0}$	
58	Tintinnopsis campanula	180	120	Gadan
59	Tintinnopsis choroestrichids	20	$\boldsymbol{0}$	AIM ∙NEM
60	Tintinnopsis corniger	20	$\boldsymbol{0}$	
61	Tintinnopsis cylindrical	40	40	Temp °C
62	Tintinnopsis dadayi	180	100	Chlorophyll (µg/L) 30
63	Tintinnopsis everta	20	$\overline{0}$	
64	Tintinnopsis fistularis	20	20	
65	Tintinnopsis gracilis	100	40 <sup>°</sup>	Ammonia (ppm)
66	Tintinnopsis hemispiralis	$\bf{0}$	20	
67	Tintinnopsis karajacensis	20	$\boldsymbol{0}$	Phosphate (ppm)
68	Tintinnopsis kofoidi	20	$\boldsymbol{0}$	
69	Tintinnopsis lobiancoi	40	60	Nitrite (ppm)
70	Tintinnopsis nana	20	$\boldsymbol{0}$	
71	Tintinnopsis parva	120	200	
72	Tintinnopsis parvula	20	40	<b>Sandsp</b>
73	Tintinnopsis radix	120	140	
74	Tintinnopsis redixand	40	$\boldsymbol{0}$	∍AIM $\mathbin{{\multimap}}{\text{NEM}}$
75	Tintinnopsis rotundata	$80\,$	200	Temp °C
76	Tintinnopsis tocantinensis	100	60	40
77	Tintinnopsis ventricosoides	40	20	Chlorophyll (µg/L) 30
78	Trachelophyllum apiculatum	20	$\overline{0}$	
79	Trochilia sigmoides	20	60	Ammonia (ppm)
80	Undella hyalina	120	100	
81	Undella subacuta	20	$\boldsymbol{0}$	Phosphate (ppm)
82	Undella turgida	20	20	
83	Xystonella treforti	$\boldsymbol{0}$	20	Nitrite (ppm)
	Genera: 37, Species: 83	5080	4160	
	Rare= 1-150 * Common= 151-250** Dominant= 251-350< ***			Fig. 1. Physico-chemical parameter and Sandspit, For details of abbre

<span id="page-4-0"></span>**Table III. Seasonal abundance of ciliates (cells/L) recorded from Gadani and Sandspit.**







<span id="page-4-1"></span>Fig. 1. Physico-chemical parameters recorded from Gadani and Sandspit. For details of abbreviations, see [Figure 2](#page-5-0).

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,](#page-2-0) [II](#page-3-0)). The seasonal abundance of ciliates genera (cells/L) from Gadani and Sandspit are depicted in [Figures 2](#page-5-0) and [3.](#page-5-1) 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](#page-4-0) [III](#page-4-0)). The physicochemical parameters recorded from Gadani and Sandspit are shown in [Figure 1](#page-4-1).



<span id="page-5-0"></span>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](#page-6-0)) 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\)](#page-7-0) 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,



<span id="page-5-1"></span>Fig. 3. Ciliate genera (cells/L) in the seasonal abundance from Sandspit. For details of seasonal change, see [Figure 2](#page-5-0).

ST1	Abundance	<b>Temp</b>	<b>Salinity</b>	pН	Oxygen	<b>Nitrate</b>	<b>Nitrite</b>	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	$0.843$ <sup>ns</sup>					
Nitrate	$-0.077$	0.077	$0.825$ <sup>ns</sup>	$0.988$ <sup>ns</sup>	$0.810$ ns				
Nitrite	$-0.038$	0.083	$0.849^{ns}$	$0.951$ ns	$0.763^{ns}$	$0.985$ <sup>ns</sup>			
Phosphate	$-0.276$	$0.378$ <sup>ns</sup>	$0.304$ <sup>ns</sup>	$0.435^{ns}$	0.308 <sup>ns</sup>	$0.475$ <sup>ns</sup>	$0.509$ <sup>ns</sup>		
Ammonia	$-0.068$	$0.272$ <sup>ns</sup>	$0.708$ <sup>ns</sup>	$0.876^{ns}$	$0.689$ ns	$0.875$ <sup>ns</sup>	$0.854$ <sup>ns</sup>	$0.299^{ns}$	
Chlorophyll	$0.482$ <sup>ns</sup>	$0.405$ <sup>ns</sup>	$-0.205$	$0.021*$	$0.358$ <sup>ns</sup>	$0.026*$	0.07	$0.149^{ns}$	$0.157$ ns
ST <sub>2</sub>									
Temp	$-0.282$						HICIC		
Salinity	$-0.512$	$-0.169$							
pH	$-0.456$	0.101 <sup>ns</sup>	$0.791^{ns}$						
Oxygen	$-0.378$	0.061	$0.477$ ns	$0.843^{ns}$					
Nitrate	$-0.406$	0.077	$0.825$ <sup>ns</sup>	$0.988$ <sup>ns</sup>	$0.810^{\rm ns}$				
Nitrite	$-0.37$	0.083	$0.849$ <sup>ns</sup>	$0.951$ ns	$0.763^{ns}$	$0.985^{ns}$			
Phosphate	$-0.138$	$0.378$ ns	$0.304$ ns	$0.435^{ns}$	$0.308$ <sup>ns</sup>	$0.475^{ns}$	$0.509^{ns}$		
Ammonia	$-0.496$	$0.272^{ns}$	$0.708$ <sup>ns</sup>	$0.876$ <sup>ns</sup>	$0.689^{ns}$	$0.875$ <sup>ns</sup>	$0.854^{ns}$	$0.299^{ns}$	
Chlorophyll	$-0.12$	$0.405^{ns}$	$0.205$ $^{\rm ns}$	$0.021*$	$0.358^{ns}$	$0.026*$	0.07	$0.149^{ns}$	$0.157$ ns
ST <sub>3</sub>									
Temp	$-0.307$								
Salinity	$0.302$ $^{\rm ns}$	$-0.169$							
pH	$0.165$ <sup>ns</sup>	$0.101$ <sup>ns</sup>	0.791 <sup>ns</sup>						
Oxygen	$0.130$ $^{\rm ns}$	$0.061 -$	$0.477^{ns}$	$0.843$ <sup>ns</sup>					
Nitrate	$0.138^{ns}$	0.077	$0.825$ <sup>ns</sup>	$0.988$ <sup>ns</sup>	$0.810$ ns				
Nitrite	$0.128^{ns}$	0.083	$0.849$ <sup>ns</sup>	$0.951$ ns	$0.763^{ns}$	$0.985$ <sup>ns</sup>			
Phosphate	$-0.164$	$0.378$ <sup>ns</sup>	$0.304^{ns}$	$0.435^{ns}$	0.308 <sup>ns</sup>	$0.475$ <sup>ns</sup>	$0.509$ <sup>ns</sup>		
Ammonia	0.209 <sup>ns</sup>	$0.272$ <sup>ns</sup>	$0.708^{ns}$	$0.876$ <sup>ns</sup>	$0.689$ <sup>ns</sup>	$0.875$ <sup>ns</sup>	$0.854^{ns}$	$0.299$ <sup>ns</sup>	
Chlorophyll	$0.268$ <sup>ns</sup>	$0.405$ <sup>ns</sup>	$0.205$ <sup>ns</sup>	$0.021*$	0.358 <sup>ns</sup>	$0.026*$	0.07	$0.149$ ns	$0.157$ <sup>ns</sup>

<span id="page-6-0"></span>**Table IV. Pearson correlation coefficient between ciliates communities with environmental variables in Gadani.**

\*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 [\(Goes, 2005\)](#page-9-9). 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](#page-10-7)). 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 [\(Smurov](#page-10-8) *et al*., 2013). In high temperatures in the SWM, the organisms demand for oxygen increases, resulting in low dissolved oxygen retaining capacity of water ([Hussain](#page-9-10) *et al*., 2013). Ciliates are sensitive to changes in the concentration of oxygen in water ([Fenchel,](#page-9-11) [2012\)](#page-9-11). The diversity of the ciliates in marine waters also depends on the constancy of the oxygen gradients.

ST1	Abundance	<b>Temp</b>	<b>Salinity</b>	PH	Oxygen	Nitrate	<b>Nitrite</b>	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	$-0.214$	$-0.019$	$0.421$ <sup>ns</sup>	$-0.383$	$0.134$ <sup>ns</sup>						
Nitrite	$-0.344$	$-0.035$	$-0.053$	$0.379$ ns	0.089	0.478 <sup>ns</sup>					
Phosphate	$-0.103$	$-0.167$	$-0.297$	$0.448^{ns}$	$-0.171$	$-0.096$	$0.288$ $^{\rm ns}$				
Ammonia	$-0.117$	$0.243$ <sup>ns</sup>	$0.435$ <sup>ns</sup>	$-0.126$	$0.006**$	$0.539$ <sup>ns</sup>	$0.057*$	$-0.449$			
Chlorophyll	$-0.295$	$0.470^{ns}$	$0.118$ ns	$-0.61$	$0.237$ <sup>ns</sup>	$0.441$ <sup>ns</sup>	$-0.076$	$-0.335$	$0.301$ $^{\rm ns}$		
ST <sub>2</sub>											
Temp	$0.233$ <sup>ns</sup>										
Salinity	$0.102$ $^{\rm ns}$	$0.158$ <sup>ns</sup>									
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 <sup>ns</sup>	0.383	$0.134$ ns						
Nitrite	$0.422$ <sup>ns</sup>	$-0.035$	$-0.053$	$0.379$ ns	0.089	0.478					
Phosphate	$-0.35$	$-0.167$	$-0.297$	$0.448$ ns	$-0.171$	$-0.096$	$0.288$ ns				
Ammonia	$0.387$ $^{\rm ns}$	$0.243^{ns}$	$0.435^{ns}$	$-0.126$	$0.006**$	$0.539$ <sup>ns</sup>	$0.05*$	$-0.449$			
Chlorophyll	$-0.134$	$0.470$ ns	$0.118$ ns	$-0.61$	$0.237$ <sup>ns</sup>	$0.441^{ns}$	$-0.07$	$-0.335$	0.301 <sup>ns</sup>		
	*Represents significant at $p < 0.05$ , **represents significant at $p < 0.01$ and ns represents non-significant.										
	$\mathcal{D}$			8	microzooplankton may be linked to the high chlorophyll $a$ in the area. Chlorophyll $a$ distribution depends or 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						

<span id="page-7-0"></span>**Table V. Pearson correlation coefficient between ciliates communities with environmental variables in Sandspit.**



Fig. 4. Planktonic ciliates from Gadani (Baluchistan) and Sandspit (Karachi). 1, *Codonellopsis* sp.; 2, *Favella azorica*; 3, *Favella eherenbergii;* 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 tocantinenesis.* 

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, nutrientrich and less predation sites ([Cocheret de la Moriniere](#page-9-12) *et al*[., 2004](#page-9-12)). 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

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ever, previous studies that the diversity of ciliates reduced with an increase in distance from shore [\(Tamura](#page-11-4) *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 [\(Fenchel, 1988\)](#page-9-13). The abundance of ciliates in marine waters is controlled by zooplankton, especially filterfeeding copepods [\(Atikinson, 1996](#page-8-2)). Ciliates are primary grazers on bacterioplankton and nanoplankton ([Premke](#page-10-11)  [and Arndt, 2000\)](#page-10-11). 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\)](#page-11-1). 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](#page-10-12)) in the Northern Arabian Sea (Siddiqui *et al*[., 2000](#page-10-14); [Burhan](#page-8-3) *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 sixtyfive 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,](#page-10-15)  [2013\)](#page-10-15) that inhabit freshwaters and marine environments [\(McManus and Santferrara, 2013](#page-10-16)). 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 [\(Stoecker, 2013\)](#page-11-5). Tintinnids are used as bioindicators to assess environmental stress and anthropogenic impacts on marine ecosystems (Jiang *et al*[., 2011](#page-10-17); 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 [\(Ismail and Dorgham,](#page-10-18)  [2003\)](#page-10-18). 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*[. \(2011\)](#page-10-17) and Feng *et al*[. \(2015\)](#page-9-15). *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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