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Spatial and temporal variations of tintinnids (Ciliata: Protozoa) in the Bay of Mersin, Northeastern Mediterranean Sea

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Abstract

Seasonal variations in species composition and abundance of tintinnids were investigated in the eutrophic coastal and offshore waters of Mersin Bay. Twelve sampling cruises were performed from September 2008 to February 2011. A total of 85 tintinnid taxa were identified. Among the recorded genera, agglutinated *Tintinnopsis* had the largest number of species (12 species), followed by hyaline loricated *Eutintinnus* (9 species), and *Proplectella* (7 species). *Stenosemella ventricosa*, *Tintinnopsis beroidea*, *T. compressa*, *Favella azorica*, and *Helicostemella subulata* were the dominant species during the study period. There were clear seasonal variations in species composition and abundance of tintinnids. The highest number of tintinnid species occurred in January 2009 and November 2010 in the entire study area, while the lowest species number occurred in March 2009. Moreover, species numbers were higher in offshore stations than coastal stations. Some species were present in the coastal area of the bay, and other species were present offshore. The highest tintinnid abundance values were found in September 2008 (820 ind l⁻¹) and August 2009 (749 ind l⁻¹) at coastal stations. The high abundance values in the coastal stations indicate that an increase of tintinnids is closely related to trophic status of the marine environment. In connection with these results, correlation analysis revealed that chlorophyll-*a* had an important effect on tintinnid abundance. Tintinnid communities may be used as a potential bio-indicator for assessing the trophic state of the coastal seas and bays.

Keywords: Tintinnid; Ciliata; protozooplankton; species composition; Bay of Mersin; northeastern Mediterranean.

Introduction

Ciliated protozoa have a key position in the transfer of material and energy from small-sized phytoplankton to higher trophic levels and constitute an important link in the marine planktonic food web (Pitta *et al.*, 2001; Han-nachi *et al.*, 2008; Abboud Abi Saab *et al.*, 2012). Since, protozoans are important for both food quality and quantity in pelagic food webs (Montagnes *et al.*, 2010), their variabilities may effect food web dynamics and growth of higher organisms. Tintinnids, belonging to the ciliate protozoans, are ubiquitous and significant component of microzooplankton in marine systems (Dolan, 2010; Rakshit *et al.*, 2017). They are the best-known group of ciliates because of their hard loricae (Heneash *et al.*, 2015). In oligotrophic regions dominated by pico- and nano-planktonic fractions, tintinnids are one of the main consumers of the primary production (Zohary *et al.*, 1998; Sitran *et al.*, 2007), and they can ingest food particles up to 40-45% of the oral diameters of their loricae (Laybourn-Parry, 1992). Thus, the importance of ciliates as grazers of

the primary production is higher in the eastern Mediterranean Sea (Dolan *et al.*, 1999; Pitta *et al.*, 2001) which is characterized as one of the most oligotrophic areas in the world in terms of high transparency, low nutrient concentrations and primary production (Azov, 1986; Yacobi *et al.*, 1995).

Marine ecosystems are vulnerable to environmental changes due to the combined effects of natural conditions and anthropogenic activities. In recent years, environmental threats are increasing in the Mediterranean coastal areas. Industrialization and rapid population growth cause serious damage in the coastal ecosystems of the Mediterranean. Land-based inputs such as rivers, domestic and agricultural discharges cause eutrophic conditions in the NE Mediterranean coastal waters (Polat-Beken *et al.*, 2009; Tüfekçi *et al.*, 2013; Tuğrul *et al.*, 2011). Since, tintinnid communities are sensitive to changes in environmental factors, they are valuable tools for indicating hydrological conditions and trophic status, as well as for evaluating the water quality (Bojanic *et al.*, 2012; Kim *et al.*, 2012; Sivasankar *et al.*, 2018). There

are many studies on tintinnids in different regions of the Mediterranean Sea. The previous studies conducted on tintinnids were particularly concentrated in the western and central Mediterranean Sea, such as, in the Bay of Villefrance (Cariou *et al.*, 1999; Gomez & Gorsky 2003); the Gulf of Naples (Modigh & Castaldo, 2002); the Catalan Sea (Dolan & Marrase, 1995); the Tyrrhenian Sea (Sitran *et al.*, 2009) and the Ionian Sea (Sitran *et al.*, 2007). In the eastern Mediterranean Sea, previous studies were performed in the Lebanese coastal waters (Abboud Abi Saab *et al.*, 2012), the Tunisian coastal waters (Hannachi *et al.*, 2008; Rekik *et al.*, 2015), the Egyptian coastal waters (Heneash *et al.*, 2015), the Aegean Sea (Mihalatou & Moustaka Gouni, 2002), and the Adriatic Sea (Krsinic & Grbec, 2006; Monti *et al.*, 2012; Bojanic, 2001; Bojanic *et al.*, 2012). Local studies on tintinnids in Turkish coastal waters are also available (Koray & Özel, 1983; Polat *et al.*, 2001, 2003; Balkis, 2004; Balkis & Toklu-Alici, 2009; Yurga, 2012). However, there is no detailed study on tintinnids in the Mersin Bay. In this context, the aim of this study was to describe tintinnid community structure, and their seasonal and spatial variations in relation to environmental conditions in both of coastal and offshore waters in Mersin Bay (Fig. 1). The findings of this study could help to better understand the microzooplankton community and pelagic food web structure in the north-eastern Mediterranean.

Materials and Methods

Study site

Mersin Bay is located on the northeastern shelf zone of Cilician Basin, the eastern Mediterranean Sea of Turkey (Fig. 1). The coastline of the bay is heavily populated. There is a busy harbour, and there are many industries along the eastern coastline of the bay. Mersin Bay coastal waters are fed by freshwater inputs from the Seyhan River and the Berdan, Delicay, Muftu, and Lamas creeks (Karakaya & Evrendilek, 2011). Although oligotrophic conditions of the NE Mediterranean waters prevail in offshore area of the bay, the coastal waters of the Mersin Bay receive important amounts of nutrient and organic matter inputs from anthropogenic sources and river discharges. The effluents carry a significant amount of pollutants into the bay, influencing the basic bio-chemical properties of the bay waters (Yilmaz *et al.*, 1998).

Methods

A total of 12 cruises were performed between September 2008 and February 2011 in Mersin Bay. Seawater samples were collected at 17 stations using 5-l Nansen bottles attached on a Sea-Bird rosette sampler and a CTD probe system (Fig. 1). The stations in 0-50 m depth were evaluated as coastal and >50 m depth as offshore stations. Vertical samples were taken from surface and lower depths according to depths of the stations (surface and ~ 10,

Fig. 1: Study area with the sampling stations.

20, 50, 75, 100, 150 and 200 m depths). Salinity and temperature were simultaneously measured in situ with the CTD probe. For tintinnid enumeration, a 20 l sample for each depth was concentrated to 100 ml by gently filtering the water through an 18 μ m mesh. Samples for tintinnid investigation were preserved with acid Lugol solution. Then, buffered formaldehyde was added to the samples to a final concentration of 2%. The concentrated samples were allowed to settle for at least 48 hours in 50 ml settling chambers and analyzed using an inverted microscope, following the Utermohl method (Utermohl, 1958). Tintinnid species were identified by lorica morphology according to Kofoed & Campbell (1929), Tregouboff & Rose (1957), Balech (1959), Marshall (1969), Chihara & Murano (1997), Boltovsky (1999), and Abboud-Abi-Saab (2008). Seawater samples for dissolved nutrient analysis were stored in polyethylene bottles (pre-cleaned with 10% HCl). Dissolved nutrient concentrations (nitrate+nitrite, phosphate, silicate, and ammonium) were determined with a Bran Luebbe four-channel autoanalyzer (Strickland & Parsons 1972; Grasshoff *et al.*, 1983). Chlorophyll-*a* measurements were carried out by the conventional spectrofluometric method after digestion of filtered samples by 90% acetone solution, using a HITACHI model F-2500 Fluorescence Spectrophotometer (Strickland & Parsons, 1972; UNEP/MAP, 2005).

The Shannon-Wiener (H) diversity index ($\log_e$) commonly used in community structure investigations was computed by using Primer (v.7) software (Plymouth Routines in Multivariate Ecological Research, Plymouth Marine Laboratory, Plymouth, U.K.). The $\log x+1$ transformation was performed in order to normalize abundance of tintinnids. After the transformation, Welch Two Sample t-test was applied in order to compare tintinnid abundance between coastal (0-50 m) and offshore (>50 m) areas. The differences between sampling periods was also analyzed using one-way ANOVA (R Core Team, 2018). The relationships between tintinnid abundance and environmental variables were analyzed using Pearson correlation. The effect of environmental variables on distribution of tintinnid communities was explored using Constrained Correspondence Analysis (CCA). The CCA

is an easy ordination method to relate distribution of species along environmental variables (Ter Braak, 1987) and uses the chi-square distance among sites in the ordination diagram which is constrained by linear combinations of environmental variables (Ter Braak, 1986; Kindt & Coe, 2005). Methodology related to CCA analysis was obtained from Borcard *et al.* (2011). To eliminate the influence of extreme values on ordination scores, data were square-root transformed before CCA, and analyses were performed using the *Vegan R package* (Oksanen *et al.*, 2018). To avoid confusion in the CCA diagram, species having higher abundance than 1% of maximum species abundance were shown in the diagram.

Results

Environmental variables

The mean values of environmental variables recorded at sampling stations visited in coastal (0-50m) and off-

shore areas of the bay during the study are summarized in Table 1. Surface water temperature reached the peak values in summer months with a maximum mean value of 30.21°C in August 2009. Surface salinity was expectedly lower at the coastal stations fed by freshwater inflows. The mean surface concentrations of nutrients and chlorophyll-*a* were relatively high in the coastal waters due to riverine inputs (Table 1). The highest mean values of nitrate+nitrite and silicate were observed during the wet winter period, as recorded in February 2010. Since the regional rivers and rain water are poor in reactive phosphorus (Tuğrul *et al.*, 2018; Koçak *et al.*, 2010) phosphate concentrations were low almost throughout the entire study period, slightly increasing during the wet period (Table 1). Surface chlorophyll-*a* concentrations were higher in the coastal stations due to terrestrial nutrient inputs which increased during the wet winter-early spring period and the highest mean value was measured in February 2010. The mean values of surface chlorophyll-*a* were relatively low at the offshore stations, increasing during the winter period due to enhanced inputs of

Table 1. Mean values of environmental variables in the surface water of coastal (0-50 m) and offshore areas (>50 m) of Mersin Bay during the study period.

Coastal	T (°C)	Salinity (psu)	NO ₂ +NO ₃ (µM)	NH ₄ (µM)	PO ₄ (µM)	Si (µM)	Chl- <i>a</i> (µg l ⁻¹)
Sep.08	29.63±0.47	39.50±0.31	0.32±0.36	0.96±0.84	0.06±0.07	2.40±1.77	0.56±0.89
Jan.09	17.79±0.22	39.40±0.05	0.33±0.08	0.36±0.51	0.04±0.01	0.91±0.08	0.18±0.10
Feb.09	16.35±0.86	38.61±0.81	4.57±4.29	0.85±0.72	0.05±0.03	2.60±1.63	0.60±0.95
Mar.09	16.94±0.33	38.26±0.88	2.86±0.86	1.80±0.62	0.08±0.02	3.17±1.04	0.58±0.24
Apr.09	20.21±0.49	37.00±1.75	1.03±1.10	0.34±0.27	0.05±0.02	3.11±3.10	0.74±0.36
Aug.09	30.21±0.58	38.80±0.56	0.34±0.53	0.87±0.61	0.06±0.03	2.63±2.40	0.66±0.64
Oct.09	25.70±0.29	38.86±0.50	0.26±0.22	0.50±0.36	0.05±0.02	3.90±0.84	0.41±0.29
Feb.10	15.96±0.96	37.71±1.25	6.70±3.88	0.63±0.55	0.04±0.02	6.20±3.12	1.14±0.97
Ap.2010	18.69±0.41	38.31±0.38	0.66±0.61	0.30±0.07	0.07±0.05	2.08±1.15	0.43±0.28
Jul.10	28.01±0.66	38.91±0.42	0.18±0.10	0.24±0.05	0.03±0.01	1.42±0.39	0.31±0.29
Nov.10	23.73±0.53	39.49±0.13	0.41±0.36	0.37±0.52	0.06±0.04	1.64±0.73	0.43±0.48
Feb.11	17.39±0.59	38.80±0.75	1.06±1.60	0.42±0.53	0.07±0.05	2.25±1.68	0.43±0.32
Offshore	T (°C)	Salinity (psu)	NO ₂ +NO ₃ (µM)	NH ₄ (µM)	PO ₄ (µM)	Si (µM)	Chl- <i>a</i> (µg l ⁻¹)
Sep.08	29.12±0.14	39.76±0.05	0.08±0.02	0.33±0.24	0.02±0.00	0.91±0.16	0.03±0.03
Jan.09	19.45	39.48	0.13	0.24	0.02	0.79	0.08
Feb.09	18.18±0.82	39.02±0.70	0.13±0.03	0.28±0.21	0.03±0.01	0.78±0.04	0.11±0.04
Mar.09	17.7	39.36	0.23	0.22	0.03	0.92	0.12
Apr.09	19.59±0.14	38.73±0.62	0.11±0.02	0.25±0.22	0.04±0.01	1.17±0.40	0.30±0.34
Aug.09	29.49±0.21	39.37±0.20	0.08±0.01	0.52±0.36	0.03±0.01	0.76±0.06	0.16±0.22
Oct.09	26.09±0.04	39.60±0.04	0.08±0.02	0.22±0.09	0.04±0.01	1.64±0.43	0.06±0.02
Feb.10	17.19±0.79	38.78±0.86	1.60±2.67	0.17±0.10	0.03±0.01	2.67±3.25	0.32±0.20
Ap.2010	18.56±0.20	39.04±0.17	0.11±0.02	0.22±0.07	0.03±0.00	1.00±0.07	0.07±0.05
Jul.10	27.33±0.30	39.22±0.06	0.13±0.02	0.17±0.04	0.03±0.00	1.11±0.11	0.07±0.03
Nov.10	24.22±0.21	39.56±0.02	0.15±0.06	0.11±0.05	0.03±0.01	0.98±0.10	0.10±0.05
Feb.11	18.06±0.19	39.29±0.02	0.36±0.24	0.14±0.08	0.03±0.01	1.06±0.29	0.12±0.04

Fig. 2: Surface distribution of chlorophyll-a in Mersin Bay during sampling times.

nutrients from terrestrial sources and from lower layer by vertical mixing (Table 1, Fig.2). The detailed description of spatial and temporal variations of environmental variables in the study area was given by Yücel *et al.* (2017) and Tuğrul *et al.* (2018).

Species composition and community structure

A total of 85 taxa (26 agglutinated and 59 hyaline loricae) representing 32 genera were recorded. The tintinnid species found in the study area are shown in Table 2 with abundance and frequency of occurrence of each species. The maximum species number (48) in the area was in January 2009 and November 2010 while the minimum species number (30) was in March 2009. During 2008, a total of 42 tintinnid species were identified. In 2009, the lowest and highest number of species was 30 and 48, respectively. The tintinnid species number in 2010 ranged from 42 in February to 48 in November. During one sampling period in 2011 (February), the species number was

38. The agglutinated genera *Tintinnopsis* was the most diverse and was represented by 12 species, followed by hyaline loricated *Eutintinnius* (9 species), *Proplectella* (7 species), and *Salpingella* (8 species) (Table 2). These species accounted for 42% of the total number of species. *Stenosemella ventricosa*, *Tintinnopsis beroidea*, and *Helicostomella subulata* were the most common species during the study, and they were present during all the sampling periods. In contrast, the species such as *Acanthostomella minutissima*, *Bursaopsis punctatostrata*, *Metacylis Jörgenseni*, *Undella hadai*, and *Xystonella lohmani* were observed for a single period at a single site. In terms of spatial distribution, *S. ventricosa*, *T. beroidea*, *H. subulata*, *F. azorica*, *Codonellopsis schabi*, and *Dadayiella ganymedes* were the most common species both at the coastal and offshore stations (Table 2). Other relevant taxa, *T. radix* and *T. campanula* were common at coastal stations and *E. undella*, *S. acuminata*, *P. ostensfeldi*, *P. fastigata*, and *S. steenstrupi* were common at offshore stations, but they were observed in low abundances. The tintinnid species number was relatively low near coast-

Table 2. The abundance and frequency occurrence of tintinnid species in coastal and offshore area.

Species	Code	Coastal		Offshore	
		Abundance (ind. l ⁻¹)	Frequency	Abundance (ind. l ⁻¹)	Frequency
<i>Acanthostomella minitissima</i> Kofoid et Campbell	Ami	0	0	0-2	1
<i>Acanthostomella conicoides</i> Kofoid et Campbell	Aco	0	0	0-3	3
<i>Acanthostomella norvegica</i> (Daday) Jörgensen	Ano	0-6	19	0-15	15
<i>Amphorides amphora</i> (Claparede et Lachmann) Strand	Aam	0-7	57	0-4	63
<i>Amphorides quadrilineata</i> (Claparede et Lachmann) Strand	Aqu	0-4	16	0-1	13
<i>Amphorellopsis tetragona</i> (Kofoid et Campbell) Kofoid et Campbell	Ate	0-62	6	0-1	3
<i>Amplectella tricollaria</i> (Laackmann) Balech	Atr	0-1	1	0-2	2
<i>Ascampbelliella armilla</i> (Kofoid et Campbell) Corliss	Aar	0-5	11	0-20	38
<i>Bursaopsis punctatostriata</i> (Daday) Kofoid et Campbell	Bpu	0	0	0-1	1
<i>Codonella aspera</i> Kofoid et Campbell	Cas	0-8	5	0-18	23
<i>Codonella amphorella</i> Biedermann	Cam	0-3	3	0-4	20
<i>Codonella apicata</i> Kofoid et Campbell	Cap	0-2	1	0-1	1
<i>Codonella perforata</i> Entz Sr.	Cpe	0	0	0-1	2
<i>Codonellopsis schabi</i> (Brandt) Kofoid et Campbell	Csc	0-20	69	0-9	59
<i>Coxiella laciniosa</i> (Brandt) Brandt	Cla	0	0	0-2	3
<i>Cyttarocyclus cassis</i> (Haeckel) Fol	Cca	0	0	0-1	3
<i>Cyttarocyclus eucryphalus</i> (Haeckel) Kofoid et Campbell	Ceu	0	0	0-2	3
<i>Dadayiella ganymedes</i> (Entz) Kofoid et Campbell	Dga	0-26	59	0-22	92
<i>Dictyocysta elegans</i> Ehrenberg	Del	0-1	3	0-4	14
<i>Dictyocysta lepida</i> (Kofoid et Campbell) Balech	Dle	0-1	3	0-2	3
<i>Dictyocysta mitra</i> Haeckel	Dmi	0-2	6	0-5	29
<i>Epiplocyclus undella</i> (Ostenfeld et Schmidt) Jörgensen	Eun	0-5	8	0-12	57
<i>Eutintinnus apertus</i> Kofoid et Campbell	Eap	0-4	17	0-3	13
<i>Eutintinnus elegans</i> (Jörgensen) Kofoid et Campbell	Eel	0-4	4	0-4	15
<i>Eutintinnus fraknoi</i> (Daday) Kofoid et Campbell	Efa	0-4	22	0-5	41
<i>Eutintinnus lusus-undae</i> (Entz) Kofoid et Campbell	Elu	0-8	47	0-10	70
<i>Eutintinnus macilentus</i> (Jörgensen) Kofoid et Campbell	Ema	0-3	8	0-5	32
<i>Eutintinnus medius</i> Kofoid et Campbell	Eme	0	0	0-2	2
<i>Eutintinnus pinguis</i> (Kofoid et Campbell) Kofoid et Campbell	Epi	0	0	0-4	6
<i>Eutintinnus stramentus</i> (Kofoid et Campbell) Kofoid et Campbell	Est	0-7	21	0-9	48
<i>Eutintinnus tubulosus</i> (Ostenfeld) Kofoid et Campbell	Etu	0-2	4	0-5	22
<i>Favella azorica</i> (Cleve) Jörgensen	Faz	0-341	52	0-5	28
<i>Favella campanula</i> (Schmidt) Jörgensen	Fca	0-39	11	0	0
<i>Favella ehrenbergi</i> (Claparede et Lachmann) Jörgensen	Feh	0-37	19	0-2	3
<i>Favella serrata</i> (Möbius) Jörgensen	Fse	0	0	0-2	1
<i>Favella</i> sp.	Fsp	0	0	0-2	2
<i>Helicostomella subulata</i> (Ehrenberg) Jörgensen	Hsu	0-304	67	0-8	45
<i>Metacyclis jørgenseni</i> (Cleve) Kofoid et Campbell	Mjö	0	0	0-1	1
<i>Ormosella bresslaui</i> Kofoid et Campbell	Obr	0-2	1	0-52	22
<i>Parundella lohmanni</i> Jörgensen	Plo	0-2	1	0-2	2
<i>Petalotricha ampulla</i> (Fol) Kent	Pam	0-4	8	0-4	21

continued

Table 2 continued

Species	Code	Coastal		Offshore	
		Abundance (ind. l ⁻¹)	Frequency	Abundance (ind. l ⁻¹)	Frequency
<i>Petalotricha serrata</i> Kofoid et Campbell	Pse	0-24	10	0-1	2
<i>Poroceus apiculatus</i> Cleve	Pap	0-1	1	0-2	3
<i>Proplectella angustior</i> (Jørgensen) Kofoid et Campbell	Pan	0	0	0-3	12
<i>Proplectella fastigata</i> (Jørgensen) Kofoid et Campbell	Pfa	0-7	27	0-9	65
<i>Proplectella claparedei</i> (Entz) Kofoid et Campbell	Pcl	0-2	4	0-3	11
<i>Proplectella ostenfoldi</i> Kofoid et Campbell	Pos	0-4	12	0-14	68
<i>Proplectella ovata</i> (Jørgensen) Kofoid et Campbell	Pov	0-3	4	0-13	7
<i>Proplectella parva</i> Kofoid et Campbell	Ppa	0-2	1	0-1	1
<i>Poroceus apiculatus</i> Cleve	Pap	0-1	1	0-2	3
<i>Protorhabdonella curta</i> (Cleve) Jørgensen	Pcu	0-62	26	0-7	18
<i>Protorhabdonella simplex</i> (Cleve) Kofoid et Campbell	Psi	0-20	57	0-10	40
<i>Rhabdonella elegans</i> Jørgensen	Rel	0-3	4	0-5	9
<i>Rhabdonella spiralis</i> (Fol) Brandt	Rsp	0-5	11	0-3	20
<i>Salpingella acuminata</i> (Claparede et Lachmann) Jørgensen	Sac	0-6	28	0-18	75
<i>Salpingella attenuata</i> Jørgensen	Sat	0-8	8	0-15	23
<i>Salpingella cuneolata</i> Kofoid et Campbell	Scu	0-3	7	0-12	39
<i>Salpingella curta</i> Kofoid et Campbell	Scr	0-3	1	0-1	1
<i>Salpingella decurtata</i> Jørgensen	Sde	0-7	12	0-9	39
<i>Salpingella gracilis</i> Kofoid et Campbell	Sgr	0-2	1	0-2	5
<i>Salpingella glockentögeri</i> (Brandt) Kofoid et Campbell	Sgl	0-15	6	0-24	21
<i>Steenstrupiella steenstrupii</i> (Claparede et Lachmann) Kofoid et Campbell	Sst	0-13	77	0-24	104
<i>Stenosemella nivalis</i> (Meunier) Kofoid et Campbell	Sni	0-136	25	0-16	3
<i>Stenosemella ventricosa</i> (Claparede et Lachmann) Jørgensen	Sve	0-682	207	0-243	96
<i>Tintinnopsis baltica</i> Brandt	Tba	0-8	2	0	0
<i>Tintinnopsis beroidea</i> Stein	Tbe	0-319	170	0-123	80
<i>Tintinnopsis brandti</i> (Nordqvist)	Tbr	0-93	22	0-1	1
<i>Tintinnopsis campanula</i> (Ehrenberg) Daday	Tca	0-34	76	0-7	12
<i>Tintinnopsis cylindrica</i> Daday	Tcy	0-90	20	0	0
<i>Tintinnopsis compressa</i> (Daday) Laackmann	Tco	0-575	81	0-27	15
<i>Tintinnopsis everta</i> Kofoid et Campbell	Tev	0-1	1	0-2	2
<i>Tintinnopsis levigata</i> Kofoid et Campbell	Tle	0-287	22	0-2	2
<i>Tintinnopsis nana</i> Lohmann	Tna	0-1	1	0-1	1
<i>Tintinnopsis radix</i> (Imhof) Brandt	Tra	0-66	93	0-8	22
<i>Tintinnopsis tocaninensis</i> Kofoid et Campbell	Tto	0-120	60	0-2	2
<i>Tintinnopsis</i> sp.	Tsp	0-4	2	0-1	1
<i>Undella clevei</i> Jørgensen	Ucl	0-8	17	0-17	19
<i>Undella hyalina</i> Daday	Uhy	0-2	2	0-3	7
<i>Undella hadai</i> Balech	Uha	0	0	0-2	1
<i>Undellopsis marsupialis</i> (Brandt) Kofoid et Campbell	Uma	0	0	0-2	3
<i>Xystonella lohmanni</i> (Brandt) Kofoid et Campbell	Xlo	0	0	0-2	1
<i>Xystonella longicauda</i> Brandt (Lackmann)	Xyl	0-4	7	0-6	23
<i>Xystonella treforti</i> (Daday) Laackmann	Xtr	0-2	3	0-3	15
<i>Xystonellopsis cymatica</i> (Brandt) Jørgensen	Xcy	0	0	0-3	2
<i>Xystonellopsis paradoxa</i> (Cleve) Jørgensen	Xpa	0	0	0-2	2

al stations while it increased towards offshore stations (Table 3). The highest number of species was found at offshore stations (stations 46 and 48). The maximum species number was at station 48 (24 species), followed by stations 40, 43, 47, and 22 (17, 14, 14, and 15 species, respectively) (Table 3).

Abundance and distribution of tintinnids

Tintinnid abundance ranged from 0 (October 2009) to 820 ind l⁻¹ (September 2008), with the highest values at coastal area (Fig. 3 A, B; Fig. 4). The seasonal differences in tintinnid abundance were statistically significant ($p < 0.01$). In addition to first peak in September, a second maximum was observed in August 2009 as 749 ind l⁻¹ (Table 3). The first maximum in September 2008 was dominated by the enhanced *T. beroidea*, *H. subulata*, and *F. azorica*, and the second maximum was dominated by *T. compressa*, *H. subulata*, and *F. azorica*. The lowest abundance was recorded in October 2009 and the abundance values were also very low in March and April 2009, and in April 2010 at the coastal stations (Fig. 3A). In the offshore region, the lowest tintinnid abundance was found in February 2009 and October 2009. *S. ventricosa*, which was observed during all the sampling periods, was the most common and abundant species during the study, accounting for 25% of the total tintinnid abundance. Abundance of this species reached 682 ind l⁻¹ in February 2009 (Table 2) at station 33. Species of the genus *Tintinnopsis* (*T. beroidea* and *T. compressa*) were also dominant as 319 ind l⁻¹ in September 2008 at station 33 and 575 ind l⁻¹ in August 2009 at station 18, and they accounted for 28% of the total tintinnids. Other important taxa, in order of their decreasing significance, were *F. azorica*,

H. subulata, and *T. radix* sharing about 7%, 5%, and 2%, respectively, of the total tintinnid abundance. The difference in tintinnid abundance between coastal and offshore stations was statistically significant ($p < 0.01$). In this context, when coastal and offshore areas were evaluated separately in terms of abundance, the highest values were found at coastal stations (stations 2, 18, 22, and 33). Total tintinnid abundance ranged from 3 ind l⁻¹ to 820 ind l⁻¹ at the coastal stations, and from 0 ind l⁻¹ to 267 ind l⁻¹ at the offshore stations (Fig. 4). In the coastal zone (<50 m), the highest abundances of tintinnids were found in the surface waters. Depth-dependent variations of tintinnid abundance in the offshore waters are depicted in Figure 5, showing that tintinnids abundance was relatively high in upper layer in August 2009 and February 2010, and in the lower depth in April and November 2010.

Values of Shannon Wiener diversity varied from 0 (due to the presence of one species) during April 2009 (at st. 4) and October 2009 (at stations 10 and 40) to 2.91 during November 2010 (at st. 48). High tintinnid diversity in the offshore area was accompanied by a high number of species and low abundance values (Fig. 3B, Fig. 6B). Diversity values as low as 0.12 were observed due to a bloom of *S. ventricosa* in February 2009. In terms of the spatial distribution of diversity values, clear fluctuations were observed among the coastal and offshore stations (Fig. 6A, B). Tintinnid species diversity generally increased from the coastal to the offshore waters during the sampling periods.

The abundance of tintinnids in the Mersin Bay was positively correlated with all surface nutrient concentrations ($p < 0.01$, $p < 0.05$), temperature ($p < 0.05$), and chlorophyll-*a* ($p < 0.01$). When the correlations were separately calculated for surface water of coastal and offshore stations, the results of the coastal stations were almost

Table 3. Range of abundance and species number of tintinnids for coastal and offshore areas during sampling times.

	Coastal area	Offshore area		
	Range (ind.l ⁻¹)	Range (sp.number)	Range (ind.l ⁻¹)	Range (sp.number)
September 2008	26-820	3-13	21-72	5-17
January 2009	22-42	10-14	61-103	14-24
February 2009	11-697	3-10	1-24	3-9
March 2009	14-51	3-9	17-62	5-20
April 2009	4-58	1-5	11-46	2-14
August 2009	28-749	4-11	22-106	3-18
October 2009	3-102	1-5	0-22	1-10
February 2010	52-154	3-12	26-267	3-11
April 2010	14-45	3-10	20-104	6-21
July 2010	18-124	2-10	13-147	3-13
November 2010	17-423	5-12	14-48	6-19
February 2011	11-378	4-15	19-56	6-18

Fig. 3: Temporal changes in total abundance of tintinnids in coastal (A) and offshore areas (B) (Bottom and top whiskers show the lowest and highest values, respectively. The median is shown by a line the box represents the middle 50% of values).

Fig. 4: The abundance distribution of tintinnids in surface water during sampling times.

Fig. 5: Vertical distribution of tintinnid abundance (ind. l⁻¹) at offshore stations during sampling times.

similar to the correlations of all the stations, but tintinnid abundance didn't correlate with nitrate+nitrite in the coastal waters. However, the correlations with environmental variables were not important in the offshore where these parameters display relatively low spatio-temporal variations as compared to the coastal water mean values (Table 1). According to results of CCA, the effect of all environmental variables on tintinnid species distribution was statistically significant and environmental variables used in the model explained 13% of the total variance in species composition (Table 4). The CCA ordination diagram shows that the first two CCA axes explained a

total of 69% of the variability (Figure 7). In CCA ordination, temperature showed a significant contribution to the variance in the species distribution. The distribution of abundant species such as *S. ventricosa*, *T. compressa*, *T. beroidea*, and *F. azorica* was associated with temperature. Of these, *T. compressa*, *T. beroidea*, and *F. azorica* increased at high temperature, whereas *S. ventricosa* increased at low temperature. The CCA diagram also showed that the distribution of tintinnid species was related to the nutrient concentrations (Fig. 7).

Fig. 6: Temporal changes in species number and species diversity of tintinnids in coastal (A) and offshore areas (B).

Table 4. Details of model properties and the first two axes of constrained analysis of principle coordinates (CAP).

Model properties	Inertia		Proportion
Total	8.639		1
Constrained	1.136		0.1315
Unconstrained	7.503		0.8685
Details CCA axes	CCA1	CCA2	Total
Eigen value	0.51	0.28	0.79
Proportion (Explained)	0.45	0.24	0.69
Biplot scores of environmental variables	CCA1	CCA2	Sig.
Depth	0.40	-0.54	***
Temperature	-0.97	-0.18	***
Salinity	-0.14	-0.79	***
Nitrate+nitrite	0.19	0.66	***
Silicate	-0.34	0.75	***
Phosphate	-0.20	0.54	**
Ammonium	-0.36	0.38	**
Chl <i>a</i>	-0.32	0.58	*

*** Significant at 0.001 level, ** Significant at 0.01 level, * Significant at 0.05 level.

Fig. 7: Distribution of tintinnid species in relation to environmental variables in the CCA ordination (A: ammonium; Chl: chlorophyll-*a*; D: Depth; N: Nitrate+Nitrite; P: phosphate S: salinity; Si: silicate; T: temperature; codes of tintinnid species is shown in Table 2. (Biplot scores of blue vectors are shown in the above and right axis).

Discussion

In the present study, the species composition and abundance of tintinnids showed clear spatial and temporal variations. While the number of species in Mersin Bay was lower than that reported for the Lebanese coastal waters by Abboud Abi Saab (2008), it was higher than those reported for the Ionian Sea (Sitran *et al.*, 2007), the Tyrrhenian Sea (Sitran *et al.*, 2009), the Gulf of Naples (Modigh & Castaldo 2002), Adriatic Sea (Krsinic & Grbec, 2006; Bojanic *et al.*, 2012; Monti *et al.*, 2012), the Tunisian coast (Hannachi *et al.*, 2008; Rekik *et al.*, 2015) and the Egyptian coast (Heneash *et al.*, 2015) (Table 5). Abboud Abi Saab (2008) reported that while the abundance of tintinnids displayed little variation from the east to the west, the number of species and genera increased. In the present study, number of species increased in offshore waters. Biodiversity of the Eastern Mediterranean Sea is affected by newly introduced species from the Red Sea and the number of new species has increased further in recent years (Kovalev, 2006). Lakkis (2012) reported that human activities and climatic changes in recent years have contributed to tropicalization, which has increased the number of alien species. Kovalev (2006) reported that the tintinnid species were dispersed into the Mediterranean through the Suez Canal. Moreover, Abboud Abi Saab (2008) reported that approximately 60% of the tintinnid species found in the coastal waters of Lebanon were also present in the Red Sea. Since the salty Eastern Mediterranean Sea and the Red Sea have similar physico-chemical properties (highly saline, warm and low algal production) the species passing through the Red Sea can easily move into and thrive in the Mediterranean Sea

(Por, 1978; Lakkis, 1998). It is, therefore, possible that some species found in the present study could have been dispersed from the Red Sea to the eastern Mediterranean Sea. However, it is not possible to make a conclusive evaluation as to the number of species and their origins as there have been no detailed studies on this subject.

The number of species was highest in November 2010, followed by January 2009 and July 2010 in offshore waters. Bojanic *et al.* (2012) and Sitran *et al.* (2009) reported that the diversity of tintinnids reached the highest level in autumn. In contrast, Abboud Abi Saab (2002; 2008) stated that the number decreased with an increase in temperature and that the highest number of species was observed in December. However, in the present study, high levels were observed at different times of the year, and this reveals that the number of species does not follow a regular temporal change. Dolan & Pierce (2013) categorized the species biogeographically as cosmopolitan, coastal, and warm water species and reported that those with an agglutinated lorica were generally dominant on the coast, while those with a hyaline lorica were dominant offshore. In the present study, the number of species with a hyaline lorica was found to be higher than those with an agglutinated lorica. In terms of the biogeographic distribution, the identified species are neritic, cosmopolitan, and oceanic. Of the neritic genera, *Stenosemella*, *Tintinnopsis*, *Favella*, and *Helicostomella* were found in high numbers in the present study. Among the species belonging to these genera, *S. ventricosa*, *T. beroidea*, *T. compressa*, *H. subulata*, and *F. azorica* were of high abundance. These species have been reported in high numbers in other marine environments (Dorgham *et*

Table 5. Number of tintinnid species found in different regions of Mediterranean waters.

Region	Species number	Reference
Lebanese coast (Eastern Mediterranean)	117	Abboud Abi Saab (2008)
Egypt coast (Eastern Mediterranean)	29	Heneash <i>et al.</i> (2015)
Gulf of Naples (Eastern Mediterranean)	57	Modigh & Castaldo (2002)
Adriatic Sea (Eastern Mediterranean)	70	Krsinic & Grbec (2006)
Adriatic Sea (Eastern Mediterranean)	51	Bojanic <i>et al.</i> (2012)
Northern Adriatic Sea (Eastern Mediterranean)	47	Monti <i>et al.</i> (2012)
Gulf of Gabes, Tunisia (Eastern Mediterranean)	61	Hannachi <i>et al.</i> (2008)
Tunisia coast (Eastern Mediterranean)	39	Rekik <i>et al.</i> (2015)
Ionian Sea (Central Mediterranean)	79	Sitran <i>et al.</i> (2007)
Tyrrhenian Sea (Western Mediterranean)	67	Sitran <i>et al.</i> (2009)
Present study	85	

al., 2013; Abboud Abi Saab, 2008; Dolan & Pierce, 2013; Rakshit *et al.*, 2016; Bojanic, 2001; Bojanic *et al.*, 2012).

In this study, while the abundance of tintinnids was found to be higher at coastal stations, the number of species was found to be higher at the offshore stations. Accordingly, diversity index values were also higher at the offshore stations (Fig. 6). At coastal stations, species with an agglutinated lorica such as *T. beroidea*, *S. ventricosa*, and *T. compressa* were observed to be widespread and with high abundances. However, species with a hyaline lorica such as *H. subulata* and *F. azorica* were observed to be abundant at coastal stations. *H. subulata*, *T. beroidea*, and *S. ventricosa* reached high abundance levels because of their tolerance to environmental stress (Dorham *et al.*, 2013; Rekik *et al.*, 2015). The diet of tintinnids consists of small-sized phytoplankton such as nanoplankton and picoplankton. The reason they are more abundant in coastal waters having much higher phytoplankton abundance than in the offshore waters (Tuğrul *et al.*, 2018; Uysal & Köksalan, 2006). The significant positive correlation found between chlorophyll-*a* and tintinnid abundance at coastal stations supports this suggestion. Wang *et al.*, (2014) reported that the high abundance of tintinnid locally could be related to high concentrations of nutrients and coastal pollution, whereas Feng *et al.* (2015) found that tintinnid abundance was higher in extremely polluted environments. While the coastal waters of the Bay of Mersin were reported to be eutrophic due to terrestrial inputs, offshore waters were reported to have oligotrophic characteristics (Kaptan *et al.*, 2013; Tuğrul *et al.*, 2018). In the coastal zone of the bay, where large amounts of nutrients are discharged, phytoplankton biomass reaches (chlorophyll-*a*) high levels. The abundance of tintinnids was, therefore, found to be high at the coastal stations. The variations of tintinnids with depth were not as high as those between coastal and offshore stations. At the coastal stations (0-50 m), the highest abundance was found in the near surface waters (0-10 m depth range). Tintinnid abundance at most of the deep stations was low and relatively higher values were occurred during February and July 2010 (Fig. 5).

The abundance of tintinnids was higher in Septem-

ber 2008, August 2009 and February 2009 (Figure 3, 4). Dolan & Pierce (2013) reported that the species with a hyaline lorica were dominant in summer and those with an agglutinated lorica were dominant in winter. In the present study, the species showing an increase in numerical abundance in September 2008 were *T. beroidea* with an agglutinated lorica and *H. subulata* and *F. azorica* with a hyaline lorica, while in August 2009, they were *T. compressa* and *F. azorica*. The increase in February 2009 was due to *S. ventricosa* with an agglutinated lorica. The increase in the number of species with a hyaline lorica such as *F. azorica* and *H. subulata* in summer conditions and those with an agglutinated lorica such as *S. ventricosa* in winter conditions are similar to the results found by Dolan & Pierce (2013). However, while the increase in the number of the species *Tintinnopsis* in summer conditions is an exception in most of the studies conducted, it has been reported that the *Tintinnopsis* species could be found throughout the whole year (Abboud Abi Saab *et al.*, 2012; Dolan & Pierce, 2013).

Heneash *et al.* (2015) reported that the distribution and abundance of tintinnids are influenced by biological factors such as predation and food supply and physicochemical factors such as temperature, salinity, and the amount of oxygen. Thompson *et al.* (2001) reported that the abundance of tintinnids is influenced by abiotic factors rather than the food supply. In contrast, Wang *et al.* (2014) reported that the food supply is a significant factor influencing tintinnid communities and that factors such as predation and the abundance of competitors also affected the tintinnid abundance. In the present study, the significant correlation between chlorophyll-*a* and tintinnid abundance at coastal stations reveals that food availability affects tintinnid abundance as well as abiotic parameters. The significant positive correlation among the nutrients and tintinnid abundance can be explained in terms of the indirect influence of nutrients on tintinnids because nutrients affect phytoplankton, comprising the diet of tintinnids, and thus the amount of chlorophyll-*a*. Tintinnid abundance was also found to be related to water temperature in the coastal zone. This may be because the increase in abundance occurred at coastal stations and

generally during hot periods. However, the interaction of the environmental parameters should also be taken into account to explain the variations in tintinnid communities where there are differences between coastal waters and offshore.

The present results reveal that the diversity of tintinnids is high in the region, and some species can display significantly increased values. Species composition and abundance showed significant differences spatially, with high abundances found mostly at coastal waters where nutrient concentrations are high. The Eastern Mediterranean is a region where signs of climatic changes have been observed (Lelieveld *et al.*, 2012). The investigation of the diversity of species sensitive to environmental conditions, such as tintinnids, in this basin is of utmost importance. Moreover, the monitoring of tintinnids in the highly productive semi-enclosed bays under human pressures in recent decades, such as the Bay of Mersin, is also important to understand how tintinnids (in terms of abundance and species diversity) respond to environmental perturbations. In this respect, from the results of the present study, it can be concluded that tintinnids can provide important scientific information for both the water quality and trophic status of the marine environments.

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