Article

Using meiobenthos for biomonitoring of ecological health in southern Caspian Sea shores, Mazandaran, Iran

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Received 14 May 2020; Accepted 31 May 2020; Published 1 September 2020

Abstract

Human activities, including industry, agriculture, mining, dredging, and dumping introduce large amounts of pollutants into marine areas, causing permanent and significant disturbance to and a major impact on ecosystems. For assaying ecological health of south of Caspian Sea shores (Mazandaran) biodiversity and distribution of meiobenthos was measured as a bioindicator. From 12 stations (ranging in depths from 5, 10, 20 and 50 meters), sediment samples were gathered for four seasons (2012). Temperature, salinity, dissolved oxygen and pH were measured during sampling with CTD (conductivity, temperature and depth). Percentage grain size and total organic matter and calcium carbonate were measured. The average water temperature ranged from 9.52 to 23.93 ℃, dissolved oxygen from 7.71 to 10.53 mg/L, salinity from 10.57±0.07 to 10.75±0.04 ppt, pH from 7.44±0.29 to 7.41±0.22, EC from 17.97±0.12 to 18.30±0.04 μs/cm^2, TDS from 8.92±0.04 to 9.14±0.02 mg/L, total organic matter from 5.83±1.43 to 6.25±0.97% and calcium carbonate fluctuated from 2.36±0.36 to 1.68±0.19%. From 4 groups of animals (Foraminifera, Crustacea, Worms and Mollusca), the results indicate that following Foraminifera, the worms had the maximum density in the present region. In particular, benthic foraminifera have been demonstrated to be particularly sensitive microorganisms and they have been successfully utilized for their value as bioindicators of environmental change in a wide range of marine environments. Account of Shannon index (below<1) showed that this area is under pressure. In Bandar Amirabad (Behshahr) station we observed lowest Shannon index and the west of study area had been better condition compare with east of study area. Account of Pielou index showed that the distribution in this area was not steady.

Keywords meiobenthos; Caspian Sea; biomonitoring; bioindicator; ecological health.

Proceedings of the International Academy of Ecology and Environmental Sciences
ISSN 2220-8860
URL: http://www.iaees.org/publications/journals/piaees/online-version.asp
RSS: http://www.iaees.org/publications/journals/piaees/rss.xml
E-mail: piaees@iaees.org
Editor-in-Chief: WenJun Zhang
Publisher: International Academy of Ecology and Environmental Sciences

1 Introduction

The term “meiobenthos” denotes microscopically small, and motile aquatic animals living mostly on soft substrates at all depths in the marine and freshwater realm (Giere, 2009). The meiobenthos are regarded as an important component in benthic ecosystems due to their small size, high abundance and fast turnover rates (Cai et al., 2012). The production of the meiobenthos is equal to or higher than that of the macrobenthos in some estuaries, shallow waters and deep seas. The meiobenthos thus may play an important role in the recycling of nutrients (Raghukumar et al., 2001; Giere, 2009). Generally, ecological and pollution studies at assemblage level have focused on macrofauna rather than meiobenthos, despite the fact that their assemblage structures may be determined by complex mechanisms. The importance of meiobenthos as indicators of environmental quality, however, is recognized (Bradshaw et al., 2006; Sutherland et al., 2007; Grego et al., 2009). Not only the meiobenthos as a whole, but also the individual species of the most abundant taxonomic groups, such as nematodes, harpacticoides and oligochaetes, are known to be important elements of benthic communities (Giere, 2009; Nazarhaghighi et al., 2014), since they can survive in highly saline and poorly oxygenated water. Overall, they are better adapted to conditions that are no longer favorable for the macrobenthos (Olafsson et al., 2000; Warwick et al., 2002). The meiobenthos may reach densities as high as 105–106 ind./m^2 and a diversity of up to 100 species may be attained at specific sites (Mokievsky et al., 2004; Mokievsky, 2009; Cai et al., 2012).

The Caspian Sea is the largest enclosed water body in the world and it is located on the border of Asia and Europe. Biodiversity of flora and fauna of the Caspian Sea are unique on the Earth. Approximate number of plant and animal species native to the Caspian Sea (Simonett, 2006), due to the presence of a very precious kind of fish called ‘sturgeon’, preserving the water quality in the Caspian Sea is of critical importance. Sturgeons of Caspian produce the expensive high quality caviar. The mid-1990s oil and gas brought an influx of foreign investment in energy development in the region. Oil and gas extraction, along with transportation and industrial production has been the source of soil, air and water pollution in the Caspian region (Nasrollahzadeh, 2010). There is no doubt that development of the oil and gas industry does have the significant impacts to the environment. The chemicals and pesticides are threats to the flora and fauna. Since 2000 due to the pollution thousands of seals died in the Caspian Sea., the pollution has weakened their immune systems. The Caspian is an ecosystem under stress. Existing pollution has damaged marine terrestrial communities. The over fishing of Sturgeon has caused a dramatic decline in fish stocks, the number of commercial fish has considerably been reduced. Some fish species have been included into the red book (Simonett, 2006). Weak environmental laws and regulation and the ability to enforce them is affecting efforts to protect the Caspian’s environment (Nasrollahzadeh, 2010).

Biodiversity of flora and fauna of the Caspian Sea are unique. A number of plant and animal species are native to the Caspian Sea (Simonett, 2006). The ecology of the Caspian Sea is threatened due to several issues such as petroleum extraction, river and sea pollution, water-level rise, biological damage, the decline in numbers of Caspian seals and lack of legal regime among the neighbors. Infrastructural developments have had serious impacts on the ecosystems around the Caspian Sea and have often imposed long term damages (Nasrollahzadeh, 2010).

The meiobenthic is a remarkable and diverse component in sandy intertidal environments, having much greater diversity and abundance than the macrofauna in exposed beaches (Brown and McLachlan, 1990). The study of the meiobenthos in sandy beaches has great interest because of its accessibility and importance in coastal processes and especially because of its use as indicator of environmental quality. Kennedy and Jacoby (1999) reviewed the role of the meiobenthos in marine benthic systems and concluded that the state of meiobenthos communities may reflect the overall health of the system, and that changes in their populations could affect different trophic levels up setting other system components with recognized ecological value.

During the last years, these studies have become of great value for their use as bioindicator of pollution (Moreno et al., 2008; Cabria et al., 2015).

Meiobenthic of the Caspian Sea are poorly studied despite macrobenthos. Hence, the objective of the present study is to provide a detailed account of the distribution of meiobenthos collected from the southern coast of the Caspian Sea at depths ranging from 5 to 50 m.

The aims of the present work are (1) assay ecological health of south of Caspian Sea shores (Mazandaran Province) according to distribution and diversity meiobenthos (2) the characterization of the sandy beaches using selected abiotic factors, and (3) the comparison of based on species diversity statistical methods.

2 Study Area and Methodology

2.1 Study site

The study was carried out in spring, summer, autumn and winter 2012 in Mazandaran province, from Behshar (Bandar Amirabad) to Bengal solution immediately after its arrival on boat to distinguish living specimens, and then being mixed with 5% concentrated formalin solution (Moghaddasi et al., 2009; MOOPAM, 2010; Sadoogh et al., 2013; Zarghami et al., 2019a,b,c).

2.2 Sampling method

Samples were collected by boat, stations depths were measured with echo sounder and sampling coordinates were recorded with the Global Positioning System. At each station, a 0.1 m^2 Van-Veen grab sampler was used to collect bottom sediments. Three sets of samples were taken at each station by a 6.60 cm^2 area core sampler with 5cm depth and were stored in plastic boxes. For benthic studies, each sediment (33 cm^3 sample in volume) were treated with 1 g/L Rose Bengal solution immediately after its arrival on boat to distinguish living specimens, and then being mixed with 5% concentrated formalin solution (Moghaddasi et al., 2009; MOOPAM, 2010; Sadoogh et al., 2013; Zarghami et al., 2019a,b,c).

2.3 Meiobenthos analysis

For determining meiobenthic, in the laboratory, wet samples were washed through 63 μm mesh sieve to remove any excess stain and were then oven dried (75ºC, 8 hrs) (Schratzberger et al., 2002).

Foraminiferal, Ostracod and Mollusca tests were floated off using the heavy liquid CCl4 with the upper layer of the liquid consisting of floated meiobenthos tests, which were then filtered by paper and allowed to dry. A stereomicroscope was used to examine and identify tests with reference to several previous studies (Birshtain et al., 1968; Loeblich and Tappan, 1988). For identification of worms, in the laboratory, wet samples were washed through 63 μm mesh sieve to remove any excess stain and then fixed with ethanol (70%). Stereo-disecting microscope and compound microscope were used to examine and identify tests with reference to several previous studies (Birshtain et al., 1968; Hayward and Ryland, 1996).

Fig. 1 Situation of sampling stations in the southern Caspian Sea.

Table 1 Position of sampling stations.

Stations	Depth (m)	Longitude (°N)	Latitude (°E)
A1	5	36° 51' 31"	53° 16' 16. "
A2	10	36° 53' 10"	53° 16' 12"
A3	20	36° 56' 48"	53° 16' 09"
A4	50	37° 00' 52"	53° 16' 16"
B1	5	36° 43' 18"	52° 39' 33"
B2	10	36° 43' 58"	52° 39' 36"
B3	20	36° 45' 55"	52° 39' 28"
B4	50	36° 48' 41"	52° 39' 29"
C1	5	36° 40' 32"	51° 27' 43"
C2	10	36° 41' 04"	51° 27' 44"
C3	20	36° 41' 47"	51° 27' 42"
C4	50	36° 43' 47"	51° 27' 41"
D1	5	36° 56' 47"	50° 39' 20"
D2	10	36° 57' 18"	50° 39' 21"
D3	20	36° 58' 29"	50° 39' 26"
D4	50	37° 03' 17"	50° 39' 16"

Foraminiferal, Ostracod and Mollusca tests were floated off using the heavy liquid CCl_4 with the upper layer of the liquid consisting of floated meiobenthos tests, which were then filtered by paper and allowed to dry. A stereomicroscope was used to examine and identify tests with reference to several previous studies (Birshtain et al., 1968; Loeblich and Tappan, 1988). For identification of worms, in the laboratory, wet samples were washed through 63 µm mesh sieve to remove any excess stain and then fixed with ethanol (70%). Stereo-disecting microscope and compound microscope were used to examine and identify tests with reference to several previous studies (Birshtain et al., 1968; Hayward and Ryland, 1996).

2.4 Environmental factors

The benthic environmental factors including temperature, dissolved oxygen, salinity and pH were measured by CTD meter during the sampling time. Sediment grain size, Total Organic Matter (TOM) and calcium carbonate concentration ($CaCO_3$) were measured. For the grain-size analysis, 100 g of oven-dried sediments (70°C, 8 hrs) were mixed with 250 ml of tap water and 10 ml of sodium hexameta phosphate (6.2 g/L) to disaggregate the sediment. The sediments were then stirred mechanically (15 min), allowed to soak (8 hrs), stirred mechanically (15 min) and dried again (70°C, 24 hrs). Fifty grams of dried material were then transferred to the uppermost of a stacked series of graded sand sieves with 4, 2, 1, 0.5, 0.25, 0.125 and 0.063 mm mesh. The material that remained on the sieves were removed and weighed. Finally, the percentage of each particle was calculated (Moghaddasi et al., 2009; MOOPAM, 2010).

TOM in each sample was measured by calculating the loss of weight during combustion. An empty crucible was weighed and then half-filled with wet sediment and dried in an oven (70°C) until a constant weight was reached (about 24 hrs.). After removal from the oven, the sample was allowed to cool and reweighed (A). It was then placed in a Muffle furnace (550°C, 8 hrs), removed, cooled and reweighed again (B). The TOM content was determined by the loss of weight on ignition at this temperature: TOM % =100(A-B)/(A-C) (Moghaddasi et al., 2009; MOOPAM, 2010; Zarghami et al., 2014a, b; Zarghami et al., 2018). Calcium carbonate concentration was measured based on the reaction with dilute Hydrochloric Acid (HCl).

Twenty-five grams (W1) of dried sediment (7 – 8 hrs) were mixed with HCl (0.1 N) and stirred until no CO2 bubbles were discernible, and then allowed to soak (24 hrs). The upper liquid phase was discharged and the remaining sediments were filtered (with filter paper), dried (7 – 8 hrs) and reweighed again (W2). Calcium carbonate percentage was measured by the following formula: $CaCO_3\% = 100(W1- W2)/W1$ (Moghaddasi et al., 2009; Zarghami et al., 2014a, b; Zarghami et al., 2018, 2019a, b, c).

2.5 Data analysis

Principal component analysis (PCA) was used to investigate the relationship between seven variables collected during seasonal sampling cruises in 2012 (temperature, pH, dissolved oxygen, salinity, TOM %, $CaCO_3$ % and granulometry). Discriminant Analysis (DA) was used in different depths and stations. One Way ANOVA were performed to test for possible differences. Shannon-Wiener (H) diversity index and Peilou's evenness index were measured assaying species diversity and ecological assessment in this area (Marques et al., 2009).

3 Results and Discussion

3.1 Environmental factors

The results of environmental factors were shown in Tables 2, 3 and 4. The temperature of the water near the bottom was nearly similar at all stations (17.54 to 18.52°C).

The results of measuring dissolved oxygen concentration indicated enough oxygen in water near the bottom and the high average of dissolved oxygen concentration was in Ramsar transect. Salinity was also had low difference between stations (10.94 to 11.28) and increased with depth, pH was nearly equal at all stations (8.28 to 8.43).

The grain size analysis of the sediments showed that the structure of the sediment samples mostly consisted of; sand, silt and clay and seldom gravel. The grain size decreased with water depth. The silt and clay rate increase with depth at all stations (Fig. 2).

Table 2 Mean temperature, salinity, DO, pH, Total Organic Matter (TOM) and CaCO3 in different seasons at the southern Caspian Sea from Behshahr to Ramsar (±SD).

Season	Temperature (C^0)	Salinity (ppt)	DO (mg/l)	pH	TOM %	CaCO3 %
Spring	20.74 ±0.02	11.01 ±0.01	10.23 ±0.04	8.27 ±0.01	7.00 ±1.00	9.00 ±4.47
Summer	23.93 ±0.008	11.22 ±0.005	8.17 ±0.014	8.56 ±0.005	8.52 ±1.64	9.61 ±3.29
Autumn	17.34 ±0.007	11.14 ±0.01	8.10 ±0.007	8.11 ±0.051	8.08 ±1.03	9.19 ±2.22
Winter	9.52 ±0.009	11.39 ±0.02	10.53 ±0.01	8.41 ±0.01	8.23 ±1.60	9.72 ±3.92

Table 3 Mean temperature, salinity, DO, pH, Total Organic Matter (TOM) and $CaCO_3$ in different seasons at the southern Caspian Sea from Behshahr to Ramsar (±SD).

Depth	Temperature (^0C)	Salinity (ppt)	DO (mg/l)	pH	TOM %	CaCO3 %
5	20.83 ±0.011	11.08 ±0.019	8.71 ±0.034	8.28 ±0.016	3.41 ±0.66	3.33 ±0.653
10	20.71 ±0.023	11.2 ±0.008	8.72 ±0.015	8.29 ±0.019	6.43 ±1.14	7.09 ±1.968
20	18.8 ±0.019	11.25 ±0.009	8.6 ±0.03	8.35 ±0.03	7.86 ±0.881	13.4 ±5.873
50	11.27 ±0.013	11.21 ±0.04	9.20 ±0.007	8.48 ±0.01	14.56 ±2.77	12.76 ±4.572

Table 4 Mean temperature, salinity, DO, pH, Total Organic Matter (TOM) and $CaCO_3$ in different depths in the southern Caspian Sea from Behshahr to Ramsar (±SD).

Stations	Temperature (^0C)	Salinity (ppt)	DO (mg/l)	pH	TOM %	CaCO3 %
A	18.52 ±0.34	10.97 ±0.032	8.69 ±0.036	8.4 ±0.024	10.59 ±1.88	14.73 ±6.26
B	18.18 ±0.011	11.24 ±0.024	8.42 ±0.036	8.3 ±0.033	7.78 ±1.1	7.95 ±2.59
C	17.37 ±0.01	11.28 ±0.014	9.01 ±0.007	8.43 ±0.013	7.04 ±0.95	7.27 ±1.66
D	17.54 ±0.017	11.24 ±0.01	9.1 ±0.006	8.28 ±0.005	6.85 ±1.52	6.63 ±2.544

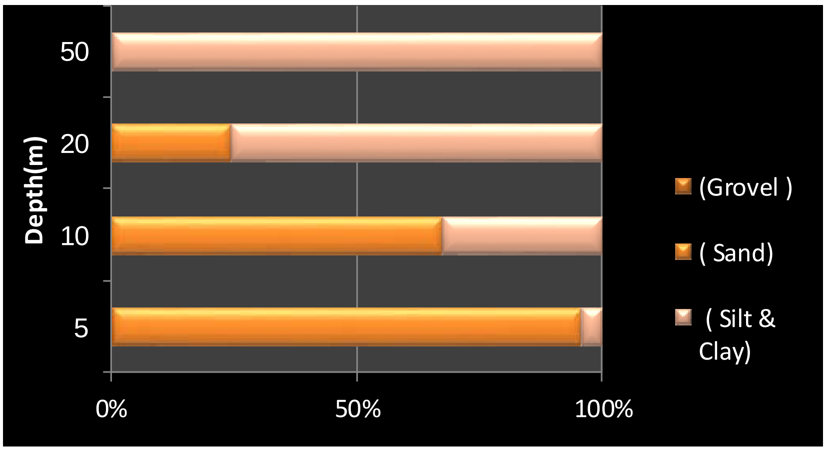

Fig. 2 Percentage of gravel, sand, silt and clay in different depths in the southern Caspian Sea from Behshahr to Ramsar.

Table 5 Shannon and Pielou indices for meiobenthos animals from different seasons at the southern Caspian Sea from Behshahr to Ramsar.

Index	Spring	Summer	Autumn	Winter
Shannon	0.5	0.57	0.85	0.9
Pielou	0.31	0.3	0.4	0.46

Table 6 Shannon and Peilou indices for meiobenthos animals from different depths at the southern Caspian Sea from Behshahr to Ramsar.

Depth (m)	Index	
	Shannon	Pielou
5	0.93	0.52
10	0.82	0.39
20	0.66	0.31
50	0.49	0.39

Table 7 Shannon and Pielou indices for meiobenthos animals from different stations at the southern Caspian Sea from Behshahr to Ramsar.

Stations	Index	
	Shannon	Pielou
Behshahr (A)	0.28	0.13
Babolsar (B)	0.88	0.43
Noshahr (C)	0.78	0.5
Ramsar (D)	0.96	0.55

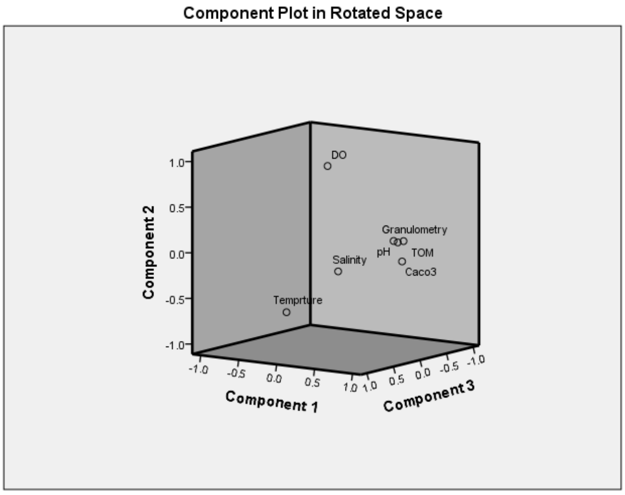

Fig. 3 PCA of environmental factors in the southern Caspian Sea.

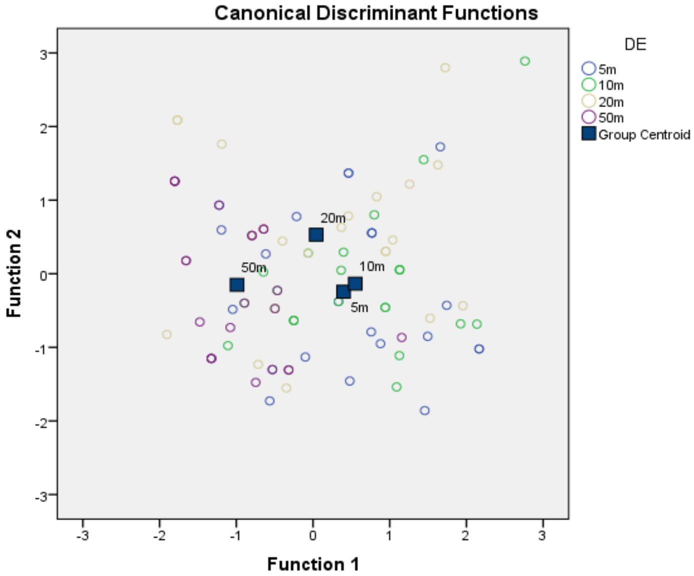

Fig. 4 DA analysis of meiobenthos animals at different depths in southern Caspian Sea.

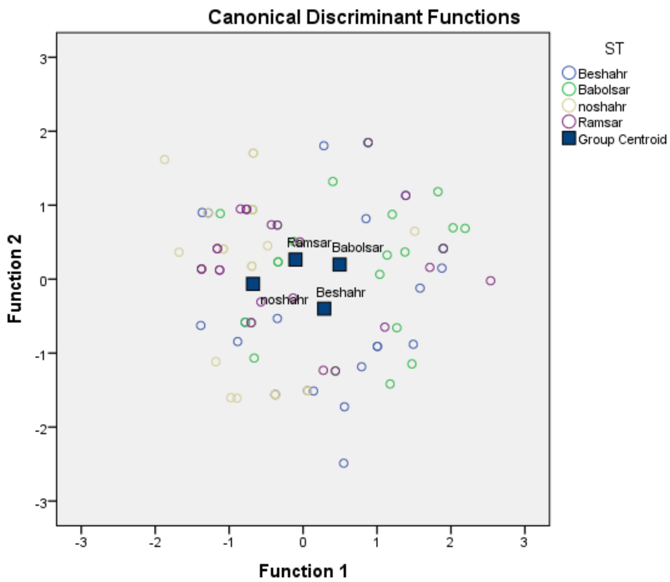

Fig. 5 DA analysis of meiobenthos at different stations in southern Caspian Sea.

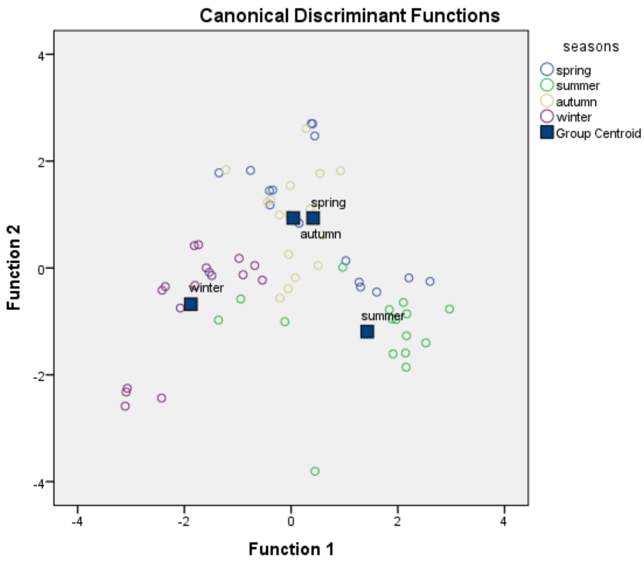

Fig. 6 DA analysis of meiobenthos at different seasons in southern Caspian Sea.

In the previous study, a highly significant negative correlation was obtained between meiobenthic abundance and salinity. This is different from the trend observed in the Changjiang River (Yangtze River) Estuary (Hua et al., 2005) and an offshore area of the Bohai Bay, China (Zhang et al., 2009). The results Jameyla et al. (2020) in Muduing Bay showed that Populations of the different species of gastropods may have been affected by the various environmental factors and disturbance regimes such as kinds of habitat, habitat degradation, and unregulated harvests, which vary from site to site. It was also observed that there were variations in the effects of selected factors on species richness and abundance. It was shown that pH affects the population of a species in one site while other sites by total dissolved oxygen. While physicochemical factors such as temperature, salinity, and total suspended solids (TSS) indicated no significant effects on the diversity and abundance of gastropods in this bay (Jameyla et al., 2020).

However, in the present investigation the meiobenthic abundance may not have been significantly related to salinity and salinity gradient was not found. It is known that the smaller the organisms, the more fine environmental heterogeneity they could perceive. Furthermore, the short generation time of small-sized organisms causes high temporal variability of their communities (Burkovsky et al., 1994; Azovsky, 2000, 2002; Azovsky et al., 2004). The overall higher densities in the Mediterranean are explained by the coarser and more oxygenated sands, higher meiobenthic in coarser sediments has been reported frequently (Giere, 2009). Benthic communities in brackish water have lower densities and fewer species than either pure marine or pure freshwater communities (Gheiskiere et al., 2005).

The distribution and dynamics of the communities of meiobenthos in the ecosystems are strongly influenced by fluctuations of the physicochemical factors. According to the results of One Way ANOVA the density of meiobenthos was significantly different with the stations, seasons and depths. Among seven parameters evaluated, the result of Pearson correlation showed that the density of meiobenthos, TOM and depth were negatively correlated. The present results indicate that all these factors affect the abundance and diversity of meiobenthic organisms in the area of the study (with low density and high diversity) characterized by coarse grain sediments and low TOMs. On the contrary, Ashraf et al. (2011) found that meiobenthos community of the Suez Canal and the Mediterranean Sea showed high meiobenthic density and low diversity in fine grain size and high Tom's sediments.

Grain size and the degree of sorting of the sand grains determine the available space for interstitial meiobenthos and thus their abundance. Meiobenthos assemblages are largely determined by spatial gradients in factors such as grain size, depth or organic matter contents; therefore large variations in meiobenthos abundance depending on these factors might be expected (Giere, 2009; Deudero and Vincx, 2000).

Even though water depth has been proposed as an environmental factor that modifies meiobenthos assemblages (Deudero and Vincx, 2000), as well as grain size (Coull and Bell, 1979), there is a lack of knowledge on the relationship between these two factors. Water depth affects hydrodynamism, the deeper the profundity is, and the lesser is the hydrodynamism at the bottom and therefore smaller grain size scan be found. This fact, translates into higher sedimentation and organic accumulation rates (Parenzan, 1979; Guerra-García and García-Gómez, 2005). The results of Cabria et al. (2015) in the sandy beaches of the western Mediterranean showed that the combinations of environmental factors that best explain the observed meiobenthos distribution have similar correlation values in both BIO-ENV analyses. The present results showed that depth was the main factor influencing meiobenthic assemblages in this region of the Caspian Sea. Additionally, we provide quantitative and qualitative data for future assessment of shallow communities of meiobenthos under natural or human-induced perturbations (Zarghami et al., 2019c).

3.2 Meiobenthos analysis

From the 4 animal groups (Foraminifera, Crustacea, Worms and Mollusca), 40 were recognized of which 38 species belonging to 29 genera of 25 families were identified.

About 34 species were alive (Table 8 and 9) and six species were dead or with empty shells (Table 10). Foraminifera were the dominant group of meiobenthos (70.3 %) (Fig. 7).

The results of experimental studies that have considered the overall effects of macrobenthos originating from processes such as predation, bioturbation and competition for food also indicate effects on meiobenthos (Ólafsson, 2003). The results of Zarghami et al. (2018) showed that the divergent seasonal variations of the meiofauna and macrofauna may be linked to their different life strategies, and that possible biological interactions between meiofauna and macrofauna may also play a significant role in structuring these associations.

In the present study the maximum density of meiobenthos, was observed in depth of 20m. These fluctuations in the meiobenthos were, to a large extent, be affected by variation in density of Foraminifera, which constituted more than 70% of the total meiobenthic (Fig. 7). Our results showed that the seasonal variability was highest for the univariate indices such as the Shannon–Wiener and Pielou index. These indices decreased with increasing depth and showed maximum in winter (Tables 5, 6 and 7). Thus in the Mediterranean protected area, Burullus Lake in Egypt exhibited maximum diversity at a depth of 5m (Mitwally and Abada, 2008) and the results showed that the diversity of meiobenthos was negatively related to grain size.). Foraminifera was the only group present in every core sample and dominated the fauna. The cosmopolitan Foraminifera *Ammonia beccarii caspica* was the dominant species at all sampling stations and was observed at all seasons (Fig. 15). This species was reported in earlier work in the Caspian Sea (Birshtain et al., 1968; Zarghami et al., 2019a, b). *Ammonia beccarii* is a common cosmopolitan species dwelling in littoral and neritic environments. It has been extensively studied in various aspects, such as, geographic distribution, ecology, biology, life-cycles, morphology, structure and environmental applications from all over the world (Debenay et al., 2009; Sadoogh, 2013; Zarghami et al., 2014; Zarghami et al., 2018; Zarghami et al., 2019a,b). Results of previous study showed on very dynamic sandy shores, waves and tidal currents can suspend fractions of sediment, therefore, disturb the infauna (Murray et al., 2002). The present results indicate that following Foraminifera, the worms had the maximum density in the present region (Fig. 7). Among these, benthic foraminifera and nematodes are highly suitable and sensitive biological organisms through which our comprehension of marine and transitional marine environments can be further explored. In particular, benthic foraminifera have been demonstrated to be particularly sensitive microorganisms and they have been successfully utilized for their value as bioindicators of environmental change in a wide range of marine environments. Since the complex interplay of different biological, chemical, ecological and physical element a general limitation of ecological investigations of nematodes is related to their difficult taxonomic identification. The taxonomic approach is certainly the best and more sensitive way for the evaluation of the nematode species response to the pollution effects. However, the results from the diversity index should be treated with caution because of the influence of some natural environmental parameters. At the present time, the maximum density of nematode was 92.14% of the total worms (Fig. 11). Highest density of nematode was observed in autumn. Nematodes usually dominate all marine meiobenthos samples in abundance and biomass occurring in each substrate and sediment in all climatic zones, where they are of considerable ecological importance (Ashraf et al., 2011). Regarding abundance, nematodes showed the highest densities at ‘5m, where the hydrodynamic conditions are expected to be more stressful (Zarghami, 2019b). In fact, nematodes are more tolerant to stressful conditions than most other groups (Deudero and Vincx, 2000). TOM values increased with depth. Then higher abundances at ‘5 m’ could be due to pollution events, since it is known that nematodes have a higher persistence in gradients with increasing pollution (Raffaeli and Manson, 1981).

Table 8 Species of meiobenthos, identified from the southern Caspian Sea (Behshahr to Ramsar) at different stations.

Group of Meiobenthos	Species	Stations			
		Behshahr (A)	Babolsar (B)	Noshahr (C)	Ramsar (D)
Foraminifera	*Ammonia beccarii*	*	*	*	*
	Ammonia tepida	*	*	*	*
	Ammonia parkinsoniana	*	*	*	*
	Elphidium littorale	*	*	*	*
	Criboelphidium sp.	*	*	*	*
	Elphidium excavatum	*	*	*	*
	Ammobaculites agglutinans	*		*	*
	Ammotium sp.		*	*	
	Miliammina fusca	*			
	Miliammina sp.	*	*	*	*
	Cornuspira sp.		*		
Crustacea	*Amnicythere longa*		*		
	Amnicythere bacuana	*		*	
	Amnicythere reticulata		*		
	Amnicythere striatocostata	*	*	*	
	Loxoconcha lepida	*	*	*	*
	Loxoconcha rhomboidea		*		*
	Xestoleberis depressa	*	*		*
	Cyprideis littoralis	*	*	*	*
	Darwinula stevensoni	*	*	*	*
	Polyphimidae		*		*
	Copepoda				*
	Mysidae	*	*	*	*
Mollusca	*Didacna protracta*	*	*	*	*
	Hypanis caspia	*	*	*	*
	Abra ovata	*	*	*	*
	Anisus kolesnikovi	*	*	*	

	Abeskunus sphaerion	*	*		*
	Ulskia ulskii	*			
Worms	*Paranais litoralis*	*	*	*	*
	S. gynobranchiata	*	*	*	*
	Nereis diversicolor		*		
	Annulovortex sp.	*	*	*	*
	Nematoda	*	*	*	*
	Total	26	29	23	24

Table 9 Species of meiobenthos identified from the southern Caspian Sea (Behshahr to Ramsar) at different seasons.

Species	Spring	Summer	Autumn	Winter
Ammonia beccarii	*	*	*	*
Ammonia tepida	*	*	*	*
Ammonia parkinsoniana	*	*	*	*
Elphidium littorale	*	*	*	*
Criboelphidium sp.	*	*	*	*
Elphidium excavatum		*	*	*
Ammobaculites agglutinans		*	*	
Ammotium sp.		*	*	
Miliammina fusca		*		
Miliammina sp.	*	*	*	*
Cornuspira sp.			*	
Amnicythere longa	*			
Amnicythere bacuana		*		
Amnicythere reticulata	*			
Amnicythere striatocostata	*	*	*	
Loxoconcha lepida			*	*
Loxoconcha rhomboidea	*			
Xestoleberis depressa	*	*		
Cyprideis littoralis	*	*	*	
Darwinula stevensoni	*	*	*	*
Polyphimidae	*			
Copepoda			*	
Mysidae	*	*	*	*
Didacna protracta	*	*	*	
Hypanis caspia	*	*	*	
Abra ovata	*	*	*	
Anisus kolesnikovi	*			
Abeskunus sphaerion	*	*	*	
Ulskia ulskii	*			
Paranais litoralis	*	*		*
S. gynobranchiata	*	*	*	*
Nereis diversicolor				
Turbellaria	*	*		*
Nematoda	*	*	*	*
Total	25	24	22	14

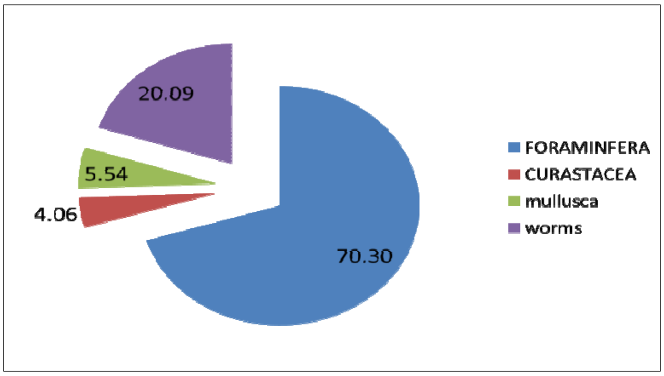

Fig. 7 Percentage of meiobenthos groups at the southern Caspian Sea from Behshahr to Ramsar.

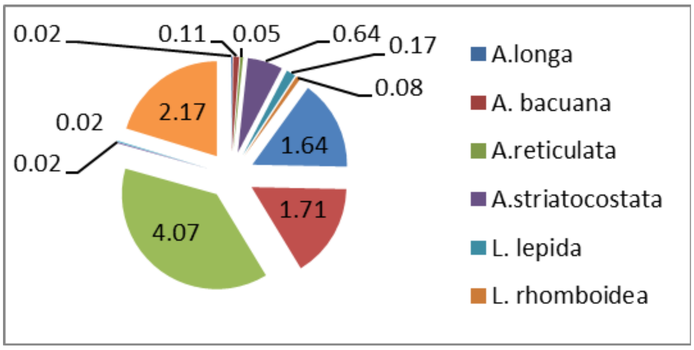

Fig. 8 Percentage of crustacea at the southern Caspian Sea from Behshahr to Ramsar.

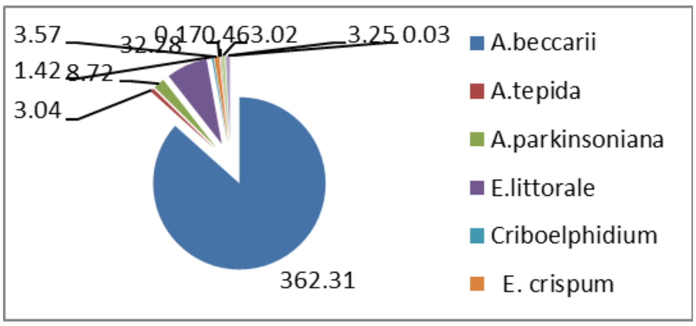

Fig. 9 Percentage of foraminifera at the southern Caspian Sea from Behshahr to Ramsar.

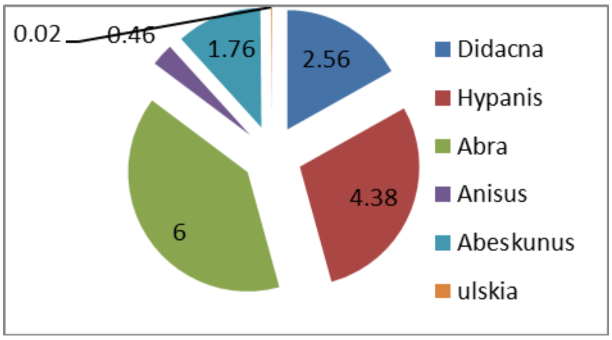

Fig. 10 Percentage of mullusca at the southern Caspian Sea from Behshahr to Ramsar.

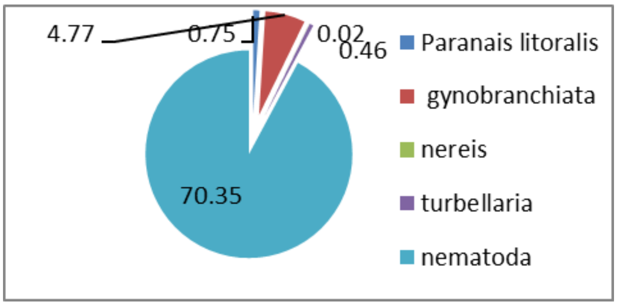

Fig. 11 Percentage of worms at the southern Caspian Sea from Behshahr to Ramsar.

3.3 Data analysis

Among seven parameters evaluated the results of PCA showed that granulometry, TOM % and $CaCO_3$ % had an important role (Fig. 3). The results of Pearson correlation showed that had been negative correlation between density of meiobenthic, TOM and depth. However, according to the results of One Way ANOVA the density of meiobenthos were significantly different with the stations, seasons and depths (Tables 10, 11 and 12).

Maximum Shannon–Wiener and Pielou index for meiobenthos were observed in winter. The values of Shannon and Pielou indexes were showed in Tables 5, 6 and 7. We observed highest Shannon-Wiener index and high Pielou index in winter. Then Shannon-Wiener index was high despite that we had maximum richness in spring.

Table 10 Density of meiobenthos in 10 cm^2 of sediment in different depths in the southern Caspian Sea from Behshahr to Ramsar (±SD).

Depth (m) / Density	5	10	20	50
Meiobenthos	606.65±309.46^a	524.08±325.19^a	705.93±418.54^a	279.79±150.46^b

Table 11 Density of meiobrnthos in 10 cm^2 of sediment in different sesons in the southern Caspian Sea from Behshahr to Ramsar (±SD).

Season / Density	Spring	Summer	Autumn	Winter
Meiobenthos	362.6±232.81^a	541.81±347.61^a	592.01±331.95^a	820.1±360.04^b

Table 12 Density of meiobenthos in 10 cm^2 of sediment in different stations in the southern Caspian Sea from Behshahr to Ramsar (±SD).

Station / Density	Behshar (A)	Babolsar (B)	Noshahr (C)	Ramsar (D)
Meiobenthos	475.6±291.12^a	654.82±406.75^a	587.64±270.84^a	398.39±234.94^b

The result of PCA showed that granulomere, TOM% and $CaCO_3$% had an important role on the distribution of meiobenthos. DA showed the depth of 10m is separated from the depth of 50 m. According to granolumetry (Fig. 9) the structure of sediment is different in two depths then density of meiobenthic is different. Stations Babolsar is separated from Noshhar (Fig. 5). Mean grain size did not vary significantly over the seasons. The substrate type was varied among the four depths (Fig. 2). The common substrate type consisted of coarse sand, find sand, silt and clay. The highest number of individuals and diversity was observed in depth of 5m; where substrate structure consisted of fine sand.

Therefore, it can be assumed that substrate is one of the major factors that influenced the distribution of meiobenthos. The result of PCA showed that granulometry had an important role (Fig. 3). The benthic communities at the study site exhibited a marked seasonal variability with maximum density and diversity were observed in winter (Table 12).

In a previous study the meiobenthos, both the number of taxa and abundances were significantly higher during the winter and autumn (Meurer and Netto, 2007). The results obtained by Maurer and Netto (2007) in shallow sub-littoral Laguna estuarine (South Brazil) showed an increase of reproductive activities of macrobenthic species during spring and summer, as indicated by the highest densities of temporary meiobenthos, coincided with the lower peak of the meiobenthos densities.

According to the European Marine Strategy Framework Directive seafloor integrity should be at a level ensuring the safeguarding of the structure and function of ecosystems. Consequently, monitoring the quality of the environment appears to be essential for devising effective protection strategies and appropriate forms of management of marine systems (Balsamo et al., 2012).

According the research of Yusal et al. (2019) the stations situated nearby the hotels and restaurants along the edge of Losari Beach had a high abundance of meiofauna because the activities taking place in these buildings supplied both organic and inorganic materials, i.e., the primary food of meiofauna, to the surrounding waters. As our result showed Meanwhile, the stations located close to the Soekarno-Hatta International Port, Paotere Harbor, and the mouth of Jeneberang River had a low abundance of meiofauna on account of the direct physical disturbances from the diverse anthropogenic activities to the habitat of meiofauna. Our results showed that and showed most Shaanon diversity index in tourism area Ramsar and Babolsar stations and lowest of this index in Amir Abad port. As our result showed in Behshahr (Bandar Amiriabad) because port Shannon-Wiener index was low thus in polluted area we observed lowest diversity and frequency of meiobenthos.

4 Conclusion

In summary assaying ecological health of south of Caspian Sea shores (Mazandaran) the biodiversity and distribution of meiobenthos as an bioindicator was measured the results showed four group of meiofauna (Foraminifera, Crustacea, Worms and Mollusca) was observed in research area. Data analyzing showed that Pearson correlation revealed a negative correlation between density of meiobenthos, TOM and depth. Shannon diversity index in shallow waters, was higher than in deeper water. Account of Shannon index (below<1) showed that this area is under pressure. Most Shannon diversity index in tourism area Ramsar and Babolsar stations and in Bandar Amirabad (Behshahr) station we observed lowest Shannon index and the west of study area had been better condition compare with east of study area. Account of Pielou index showed that the distribution in this area was not steady. In this respect, further similar investigation into other parts of the Southern Caspian Sea coastline is strongly recommended.

Acknowledgment

The authors would like to acknowledge the support which was received from Professor John Murray (University of Southampton) and Professor Simon Haslett (University of Wales) in confirming the identification of Foraminifera and their valuable suggestions.

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