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Investigation of the biota of Burgas Bay, Black Sea

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Abstract. *Burgas Bay is the largest one on the Bulgarian Black Sea coast. Industrial plants along the coast as well as ports for liquid and solid cargoes create conditions of anthropogenic pressure on hydrobionts in that area. Macrozoobenthos and phytoplankton samples were collected under a specific scheme in connection with the construction of the Burgas-Alexandroupolis oil pipeline, at stations located around the designated places for unloading tankers. During the study period (2009 – 2010), 88 phytoplankton species distributed in 13 classes were identified and over 50 species of benthic species distributed in four main groups: Polychaeta, Mollusca, Crustacea and the mixed group Diversa.*

Keywords: Black Sea, Burgas Bay, phytoplankton, macrozoobenthos, biodiversity, quantitative indicators

Abbreviations: ab – abundance, b - biomass

Introduction

The current state of marine life in Burgas Bay reflects the ecological status of the water and polluted coastal areas. The bay is subjected to high anthropogenic pressure and is defined as a "hot spot" on the Bulgarian Black Sea coast. Since the mid eighties biodiversity in the bay has reduced, eutrophication processes have deepened and organic compounds in the sediments have reached limit values. The exceedances reduce the natural potential for self-purification of water in the bay (EIA, 2011).

During previous studies in Burgas Bay (1990 – 2004), pronounced changes in planktocoenosis were observed – changes in seasonal dynamics of algae, reduction of their biomass, but also an increase in concentrations due to the development of small-sized phytoplankton cells. Large-sized diatoms reduce their number and algae community is often dominated by a small number of flagellates or heterotrophic species (Mavrodieva et al., 2005)

The first large study of macrozoobenthos in Burgas Bay over network of 87 stations was conducted by the Institute for Fish Resources – Varna (IFR-Varna). The summarized results are published by Li (1984) giving information on the quantitative and qualitative parameters of the benthic communities in the area.

The purpose of this publication is to present the results of the study on the basic structural characteristics and environmental status of phytoplankton and macrozoobenthos communities developing in the Bourgas Bay in the period 2009 – 2010.

Material and methods

In 2009 – 2010, macrozoobenthos and phytoplankton samples were collected in connection with the construction of Burgas-Alexandroupolis oil pipeline (Figure 1). Phytoplankton samples were collected from three horizons: surface (0-1m), medium (depth varies according to the total depth at a station) and bottom (one meter above the total depth for a station) using water samplers Niskin type (5l). The samples of water were put in 0.5 l plastic containers on board the ship. Fixing was performed by adding 5 ml formalin (37% formaldehyde solution stabilized with 10% methanol) in 500 ml water sample. After concentration by settling, the samples were stored in glass jars and analyzed on a light microscope NIKON E400 in a Sedgewick Rafter counting cell with volumes of 0.05 ml and 1.00 ml. The biomass of cells was calculated using a geometric method (Edler, 1979). This distribution of species in classes is according to electronic databases “WoRMS” (WoRMS, 2013) and “Algaebase” (Guiry and Guiry, 2013).

To study the concentration of chlorophyll-a, 500ml water samples were collected from surface and medium horizons as required by the Contracting Authority. The samples for chlorophyll were not fixed and were kept in the dark in a cooling bag or refrigerator, respectively, at temperatures below 10°C and analyzed in a laboratory using Turner Designs fluorometer.

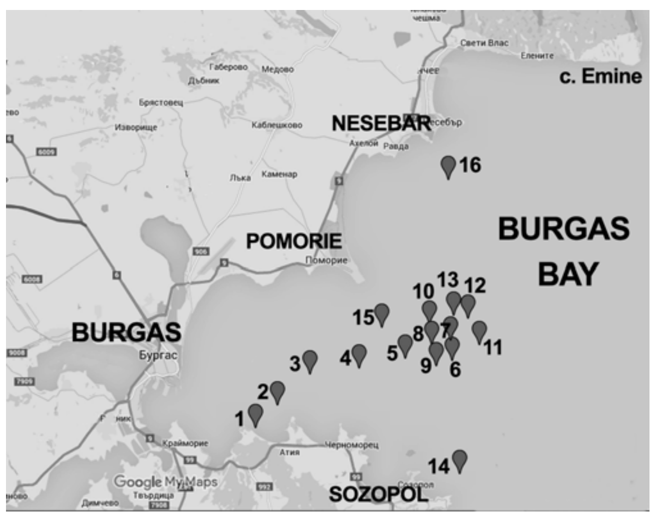

Figure 1. Location map of the sampling stations in Burgas Bay (Google Map)

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Sampling of macrozoobenthos was carried out by Van-Veen grab sampler with bite size 0.1 m^2. During the studied period were collected 19 benthic samples (with a replica at each station). The primary treatment included washing the samples on board through a series of sieves with mesh sizes of 1 mm and 5 mm, and fixation with 4% formaldehyde solution.

Washing of the samples, taxonomic identification (Morduhay-Boltovskoy, 1968, 1969, 1972; Marinov, 1977), defining of the quantitative characteristics (number and biomass) – wet weight dried on filter paper and weighing on an electronic analytical balance was done in a laboratory. The quantitative parameters were treated as equivalent to one square meter area.

Results and discussion

Phytoplankton

A total of 88 species distributed in 13 classes were observed during this study. With the highest species diversity was represented the group of *Dinophyceae* 47.73%, followed by *Bacillariophyceae* 19.32%. Of the remaining 10 classes with the best representation were class *Chlorophyceae* – 9.09% and *Prymnesiophyceae* – 4.55%. Diatom-peridinian complex comprised 67.05% of the registered species, which was a relatively low value for marine phytoplankton and was caused by the large influx of fresh water from the adjacent bay lakes, mainly during the spring season of the year.

In August 2009, the amount of chlorophyll-a in the Bay was relatively low, within the range from 1.05 $mg.m^{-3}$ to 0.12 $mg.m^{-3}$ (Figure 2). For comparison, in 2001 – 2006 the average concentration of chlorophyll-a was 0.231 $mg.m^{-3}$. Measurements of chlorophyll-a in Burgas Bay in 1987 – 1997 showed different seasonal structures with late winter-spring peak (~4-5 $mg.m^{-3}$ and > 8 $mg.m^{-3}$) and a second peak in the summer (Atanasova et al., 1995; Stoykov et al., 1994; Petrova-Karadjova, 1990; Velikova et al., 1999). In front of the Bulgarian coast were identified three peaks between 2-4 $mg.m^{-3}$ with significant changes over the years, while the lowest concentration of chlorophyll was registered in the summer, about 0.5 $mg.m^{-3}$ (norm <3.0 $mg.m^{-3}$) (Hiebaum and Karamfilov, 2005; Mavrodieva et al., 2005).

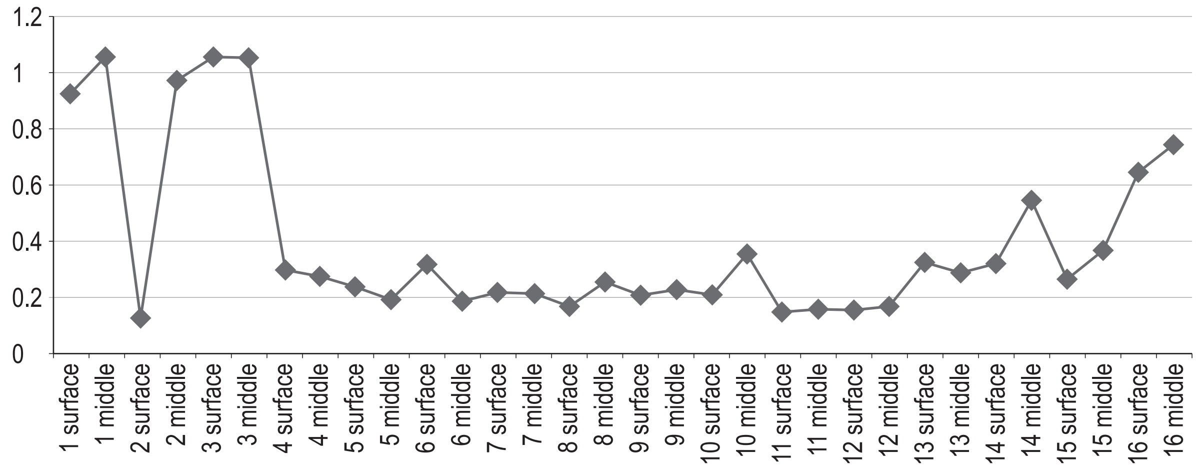

Figure 2. Chlorophyll-a ($mg.m^{-3}$) distribution at the surveyed stations, August 2009

After 1995 the reduction of phytoplankton biomass is a fact described for the entire Bulgarian coast due to economic collapse and reduction of anthropogenic pressure (Danubs, 2005; Mee et al., 2005; Petrova et al., 2006; Petrova and Gerdzhikov, 2008). In August 2009, blooms of phytoplankton species were not identified. Diatomea *Proboscia alata* dominated in number and biomass with a maximum of 0.67 x 10^6cells.l^{-1} and 3.36 $g.m^{-3}$ at station1 (surface).

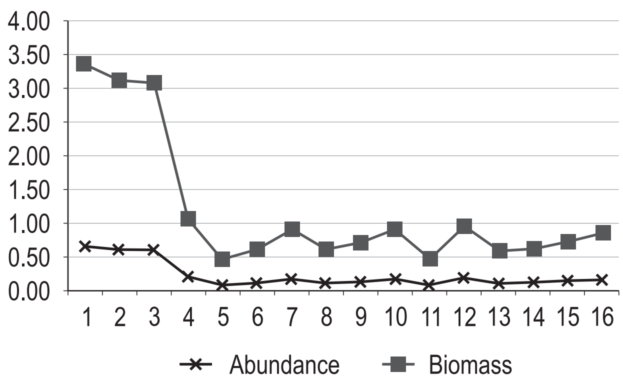

Figure 3. Maximum values of Proboscia alata; abundance (x10^6 cells.l^{-1}), biomass ($g.m^{-3}$), August 2009

The species occurred throughout the Bay, predominantly in the upper and middle horizons (Figure 3).

The abundance at the studied stations was formed by *Proboscia alata*, small *Cryptophyceae* 0.27 x10^6 cells.l^{-1} and 0.07 $g.m^{-3}$, *Merismopedia sp.* 0.175 x10^6 cells.l^{-1} and 0.003 $g.m^{-3}$, *Pseudo-nitzschia delicatissima* 0.029 x10^6 cells.l^{-1} and 0.006 $g.m^{-3}$, *Pronoctiluca pelagica* 0.027 x10^6 cells.l^{-1} and 0.058 $g.m^{-3}$, *Dactyliosolen fragilissimus* 0.026 x10^6 cells.l^{-1} and 0.081 $g.m^{-3}$, *Emiliania huxleyi* 0.022 x10^6 cells.l^{-1} and 0.006 $g.m^{-3}$ and *Gymnodinium sp.*(20 μм) 0.018 x10^6 cells.l^{-1} and 0.076 $g.m^{-3}$. The remaining 40 species occurred in numbers below 0.015 x10^6 cells.l^{-1}.

With the highest biomass differed stations 1, 2 and 3, mainly due to the development of the diatomea *Proboscia alata.* At these stations the average number of phytoplankton was 0.684 x10^6 cells.l^{-1} and the average biomass 2.69 $g.m^{-3}$. The index of Shannon - Weaver was low, an average of 1.22 (Figure 4).

At all other stations phytoplankton biomass was an average of 4-fold lower and the number 2-fold lower. At stations from N4 to N16 the average number was 0.303 x10^6 cells.l^{-1} with an average biomass of 0.73 $g.m^{-3}$. Shannon-Weaver index in that area was higher, 1.64 on average. The highest value of the index of Shannon-Weaver was registered at the farthest from shore station N 11 (2.03).

Transparency (m) in the waters of the Bay varied both over the

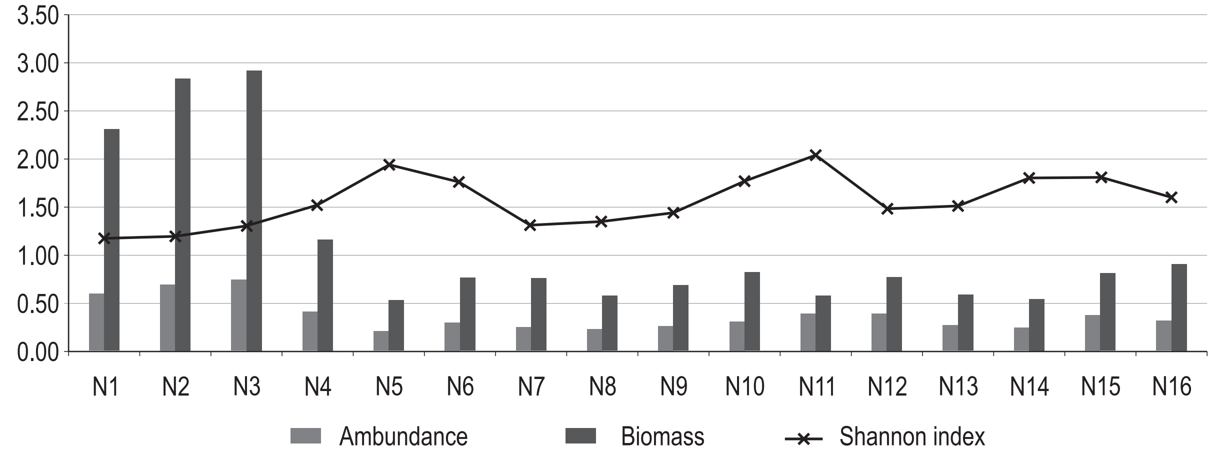

Figure 4. Comparative distribution of abundance (x10^6 cells.l^{-1}), biomass (g.m^{-3}) and index of Shannon – Weaver by stations, August 2009

years and throughout the seasons. In a multiannual perspective the lowest value was in the spring of 2005 (1.1 m) and the highest (11.10 m) - in August 2007 (Hiebaum and Karamfilov, 2005; Mavrodieva et al., 2005; Velikova et al., 1996). In August 2009 because of the greater abundance of phytoplankton in coastal waters (stations 1, 2, 3), the values of transparency were low (Figure 5). At those stations

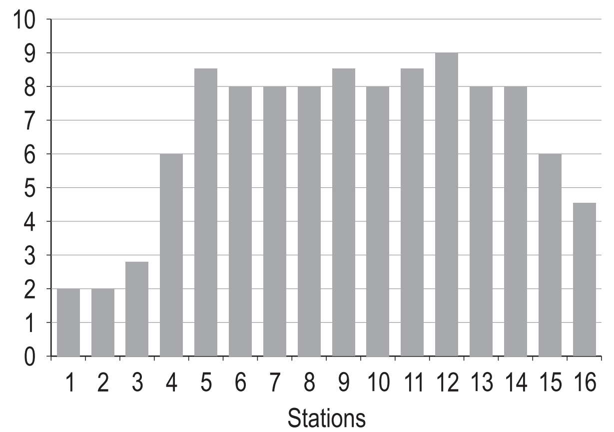

Figure 5. Transparency of the water body during the investigation (m), August 2009

was measured value of about 2 m which was below the regulatory requirement (> 2 m) and signalled poor state of seawater. By moving away from shore the values increased and this indicator was over 5 – 6m on average, which was good assessment for water quality (WFD, 2000).

In August 2009 diatoms (47%) and the group of others (47%) dominated in phytoplankton number in the Bay and in biomass dominated only diatoms (82%) (Figure 6).

In May 2010, we examined three stations located in the "small" Burgas Bay (N1, N2 and N3). With bloom concentrations dominated diatomea *Pseudo-nitzschia delicatissima* 2.36 x10^6 cells.l^{-1}, 58.28% ab (B.zalN1, 8m) and 1.72 x10^6 cells.l^{-1}, 65.00% ab (B.zalN3, 0m). Among subdominants evolved representatives of *small Flagellates* 1.31x10^6 cells.l^{-1} and 30.24% ab (B.zalN3, 15m). With the highest biomass evolved the peridinea *Prorocentrum micans* 21.88 x10^6 cells.l^{-1}, 0.37 g.m^{-3} and 38.78% b (B.zalN1, 0m), *Ps.nit. delicatissima*, *small Flagellates*, *Gymnodinium sp.*, *Neoceratium furca*, *Protoperidinium divergens*, *Glenodinium paululum* and *Heterocapsa triquetra*.

The average quantitative parameters were: abundance 2.91 x10^6 cells.l^{-1}, biomass 1.69 g.m^{-3}, Shannon-Weaver index with good values 2.22 (Figure 7). From an environmental perspective, the water of Burgas Bay can be divided into two parts: internal Bay with

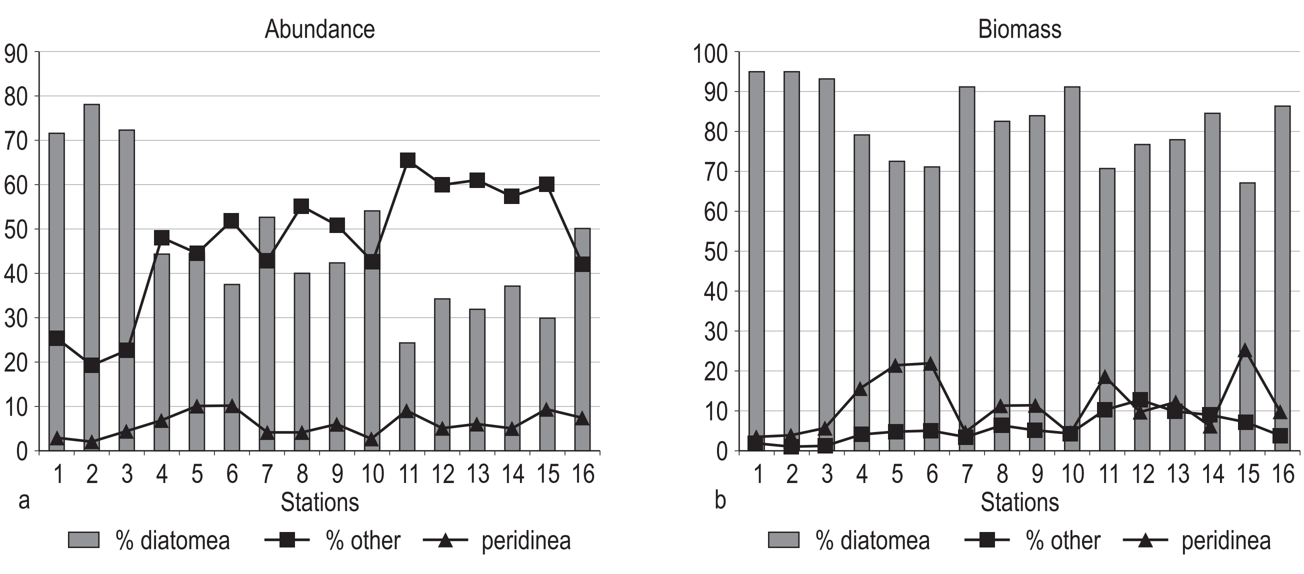

Figure 6. Percentage distribution of main taxonomic group by abundance (a) and by biomass (b), August 2009

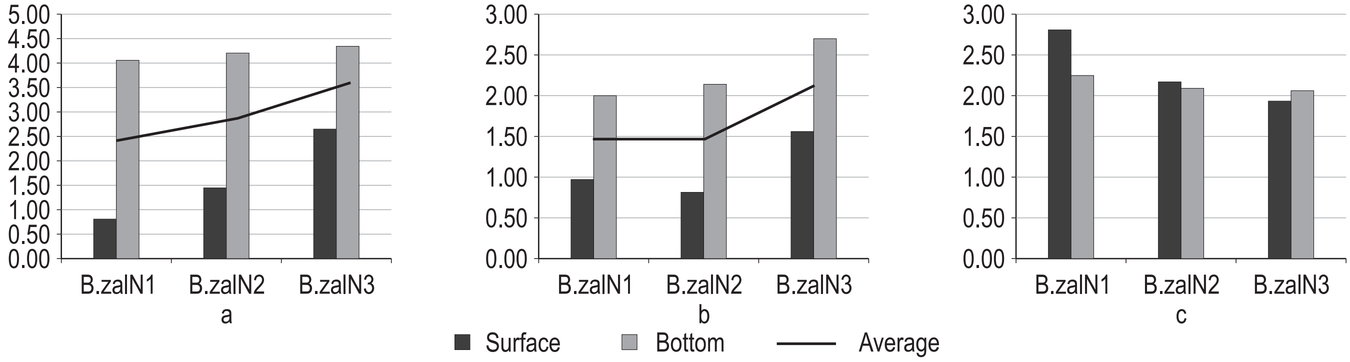

Figure 7. Distribution of phytoplankton abundance (x10^6 cells.l^{-1}) (a), biomass (g.m^{-3}) (b) and Shannon-Weaver index (c), Bourgas Bay, May 2010.

stronger anthropogenic impact and external Bay, where the ecological state is better.

Relatively high quantitative values of phytoplankton development were characteristic of spring 2010 (May) (Table 1). High phytoplankton values were also registered in May 2008 (*Pseudo-nitzschia delicatissima* 7,5 x10^6 cells.l^{-1} ; *Emiliania huxleyi* 2.9 x10^6 cells.l^{-1}) (Gerdzhikov et al., 2008).

Table 1. Average monthly values of phytoplankton abundance (x10^6 cells.m^{-3}), biomass (mg.m^{-3}), Shannon-Weaver index and transparency (m) in Burgas Bay (2009–2010)

Year	Month	Abundance (x10^6 cells.l^{-1})	Biomass (g.m^{-3})	Shannon	Transparency (m)
2009	August	0.375	1.099	1.56	6.61
2010	May	2.914	1.690	2.22	-
	Average	1.645	1.395	1.89	6.61
	Stdev	1.795	0.418	0.47	-

Zoobenthos

In 2009 – 2010 over 50 macrozoobenthos groups were established and divided into four main groups: Polychaeta, Mollusca, Crustacea and Diversa. The percentage distribution of the different groups is presented in Figure 8. With the highest percentage was represented the group of polychaetes (36%) in 2010, while traditionally with the lowest one was the mixed group Diversa (8%) in the same year. The average number of macrozoobenthos in the surveyed area was estimated at 280 ind. m^{-2}. The highest values were registered at st.9 and st.13 with 560 ind. m^{-2} and the lowest one at st.4 with 75 ind. m^{-2}.

In numbers dominant in both periods of the study was the group of polychaetes (192 ind.m^{-2}), followed by mollusks (53 ind. m^{-2}) and crustaceans (28 ind. m^{-2}). The average values of abundance by taxonomic groups and years are visualized in Figure 9.

The total average biomass of the macrozoobenthos was

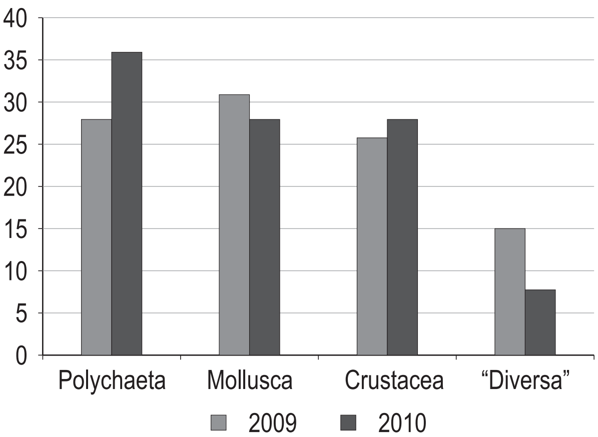

Figure 8. Percentage of the macrozoobenthos groups in species composition in Bourgas Bay

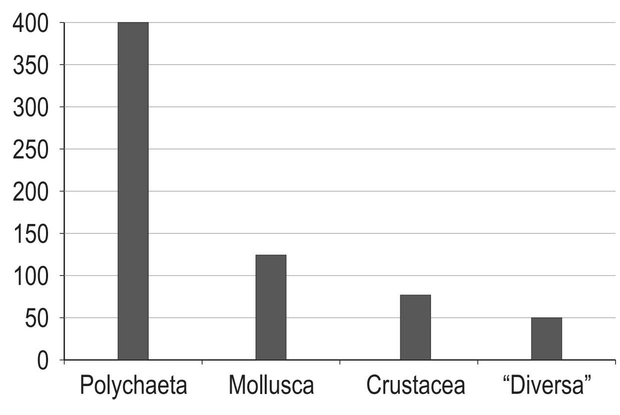

Figure 9. Average abundance of the macrozoobenthos groups in Burgas Bay in 2009 and 2010

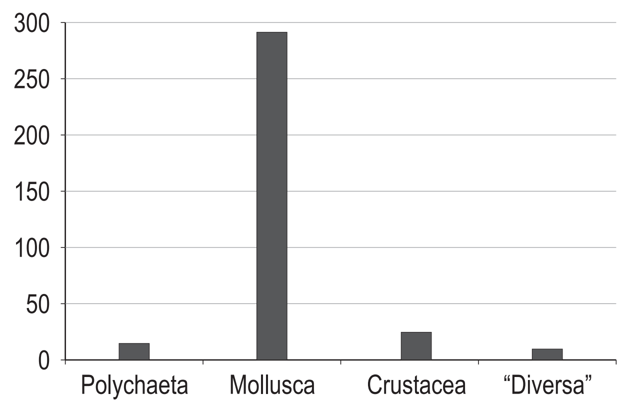

Figure 10. Average biomass of the macrozoobenthos groups in Burgas Bay in 2009 – 2010

estimated at 157 g.m^{-2} in 2009 – 2010. In both years of the study, biomass was formed by the weight of mollusc species (142.52 ind. m^{-2}). Standout species were *Mytilus galloprovincialis, Anadara inaequvalvis* and *Gastrana fragilis*, typical of the specific conditions of the surveyed water area (Figure 10).

Conclusion

The presence of 88 phytoplankton species was registered. In 2010, diatomea *Pseudo-nitzschia delicatissima* and the representatives of *small Flagellates* developed with bloom concentrations. In 2009, across the Bay dominated diatomea *Proboscia alata*.. Development of small cryptophytes was observed as their number increased in the open sea stations in the Bay. Diatomea *Pseudo-nitzschia delicatissima* dominated in 2010. The low indices of species diversity signalled for "instability" of the coastal phytocoenosis in the Bay. At the open sea stations biodiversity, respectively stability of the phytoplankton community was higher. In 2010 we observed an improvement in the ecological status. The index of Shannon-Weaver is with good values 2.22.

In Burgas Bay were identified over 50 species of macrozoobenthos (47 in 2009 and 25 in 2010) divided into four main groups: Polychaeta, Mollusca, Crustacea and mixed group Diversa. Rare or endangered species were not registered in the composition of zoobenthos. The relatively low values of quantitative indicators of zoobenthos were due to the location of most of the stations in the area of the fairway which reduced the risk of environmental issues when carrying out construction work. The comparison of the data with previous periods showed similar trends in species zoobenthos composition and higher values (between 400 – 500 ind/m^{-2} and 200 – 1000 gr/m^{-2}) for past years due to the different areas of investigations within Burgas Bay.

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CONTENTS

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The title needs to be as concise and informative about the nature of research. It should be written with small letter /bold, 14/ without any abbreviations.

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Oldenbroek JK, 1999. Genebanks and the conservation of farm animal genetic resources, Second edition. DLO Institute for Animal Science and Health, Netherlands.

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Hristova D, 2013. Investigation on genetic diversity in local sheep breeds using DNA markers. Thesis for PhD, Trakia University, Stara Zagora, Bulgaria, (Bg).

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Animal welfare

Studies performed on experimental animals should be carried out according to internationally recognized guidelines for animal welfare. That should be clearly described in the respective section "Material and methods".

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