Long-Term Shifts of the Zooplankton Community in The Western Black Sea (Cape Galata Transect, Bulgarian Coast)

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ABSTRACT:-- The aim of the current study was to assess the state and long-term trends since 1967 till 2005 of zooplankton communities of the Western Black Sea (c. Galata transect - Bulgarian region) as a response to anthropogenic and environmental shifts. Zooplankton revealed important year to year fluctuations in density and it decreased as a function of distance from the coast to open sea. Higher abundance variations at the coastal stations in comparison with the offshore ones, suggested that zooplankton community and assemblages could be more influenced and disturbed by the anthropogenic and climatic effects. The standard CUSUM procedure and anomalies were used to detect changes with time. Two main periods of zooplankton community development, before and after 1973 were distinguished as indication of anomalies in certain periods and coincided with phases of the Black Sea ecosystem development. The temporal distribution pattern is similar for all stations and shows decreasing trend for the whole study period. Variability of mesozooplankton community attributes (abundance, taxonomic structure, annual dynamics) appeared vulnerable to external forcing, resulting in unsteady standing stock especially for the coastal waters where the influence of local drivers is significant.

Keywords:-- mesozooplankton community, alterations, standing stock, Bulgarian Black Sea coast

I. INTRODUCTION

Plankton fauna is a key link between primary producers and larger predators. Recently, zooplankton has been used as an indicator to observe global change because it seems to be strongly influenced by climatic features. Multi-year zooplankton time series provide useful tools for examining climate-ecosystem interactions [1]. The variability of zooplankton and its distribution is due to: abiotic (climatic or hydrological parameters), biotic (food limitation, predation, competition) and a combination of both.

The investigations on ecological significance and biological characteristics of zooplankton community of the Bulgarian Black Sea coast have been performed extensively since the beginning of the 20th century. The most studies emphasize the assessment of the state and trends of plankton fauna in relation to Black Sea environmental changes [2, 3, 4, 5, 6, 7, 8, 9, 10, 11]. The structural alterations were linked to direct drivers such as eutrophication, aliens, overexploitation, and climate changes [12]. The aim in the current study was to assess the state and long-term trends of zooplankton communities of the Western Black Sea (Bulgarian region) as a response to anthropogenic and environmental shifts.

It is here noteworthy that zooplankton sampling has mostly been seasonally conducted on the transect Galata. Even though this series was not considered to be the monthly series data, these biological series could be the best and the most useful series available as indicators of the system’s alteration over long-term time since from 1967 to 2005.

II. MATERIAL AND METHODS

Study area and sampling
Published and unpublished data were used to extract an overall trend in zooplankton dynamics in the Western Black Sea (Bulgarian part) in the period 1967-2005 (Table 1). Time-series data of the zooplankton were collected seasonally (four times per year) at Cape Galata transect regularly till 1990, then irregularly up to date with two years (1998, 1993) missed. Names’ stations were designated as the distance from the coastline: 3, 10, 20 and 30 miles in a period of 1967 to 1992 (Table 2). Since 1994 new sampling locations close to the former stations have been conducted by Institute of Oceanology-BAS, Varna: assigning them as st.301, st.302, st.304, st.306 (Fig. 1). The first two stations are determined as coastal, the third as shelf and st. 306 as open sea (Fig.1).
At each station, zooplankton was collected using a closing Juday net (150 µm mesh size, 36 cm mouth opening in diameter = 0.1 m² mouth surface area). The net was vertically hauled from a depth of two meters above the bottom to the sea surface or in discrete layers of 0-10 m, 10-25 m, 25-50 m, 50-100 m, and 100-150 m where the bottom depth allows. The samples were preserved in 4% formaldehyde buffered to pH 8-8.2 after the gelatinous species (*Aurelia aurita*, *Pleobrachia pileus*, *Mnemiopsis leidyi* and *Beroe ovata*) were removed, rinsed, measured and counted on board.

### Table 1. Sampling inventory and data sources

<table>
<thead>
<tr>
<th>Year</th>
<th>Frequency</th>
<th>Sources</th>
</tr>
</thead>
<tbody>
<tr>
<td>1967-1990</td>
<td>Seasonally (4 times per year)</td>
<td>[2, 13]</td>
</tr>
<tr>
<td>1991</td>
<td>Winter; spring</td>
<td>[2]</td>
</tr>
<tr>
<td>1992</td>
<td>Winter; summer</td>
<td>unpublished</td>
</tr>
<tr>
<td>1994</td>
<td>Spring</td>
<td>unpublished</td>
</tr>
<tr>
<td>1995-1996</td>
<td>Monthly</td>
<td>EROS project</td>
</tr>
<tr>
<td>1997-2001</td>
<td>Summer; Autumn</td>
<td>[3, 4, 5, 14, 15, 16]</td>
</tr>
<tr>
<td>2002-2005</td>
<td>Seasonally</td>
<td>[8, 9, 10, 11, 17]</td>
</tr>
</tbody>
</table>

### Table 2. Depth and location of stations

<table>
<thead>
<tr>
<th>Station</th>
<th>Latitude</th>
<th>Longitude</th>
<th>Depth</th>
</tr>
</thead>
<tbody>
<tr>
<td>301 (coastal)</td>
<td>43° 10'</td>
<td>28° 00'</td>
<td>22</td>
</tr>
<tr>
<td>302 (coastal)</td>
<td>43° 10'</td>
<td>28° 10'</td>
<td>27</td>
</tr>
<tr>
<td>304 (shelf)</td>
<td>43° 10'</td>
<td>28° 30'</td>
<td>75</td>
</tr>
<tr>
<td>306 (open sea)</td>
<td>43° 10'</td>
<td>28° 50'</td>
<td>360</td>
</tr>
</tbody>
</table>

Fig. 1. Map of the investigated cape Galata transect in front of the Bulgarian coast (Western Black Sea)
III. DATA ANALYSIS

Abundance data derived from samples collected in various cruises at the shelf and offshore (>200 m depth) were examined for shifts in spatial-temporal distribution. Dispersion of the mesozooplankton abundance was tested using “normtest” of the PASTECS. All data of abundance were not normally distributed at p<0.05, so the data were log10-transformed. Partly irregular series of abundance of the zooplankton in a period from 1967 to 2005 was regularized and interpolated at regular intervals (12 months per year) with a “Spline” function (PASTECS) [18]. The standard CUSUM procedure was used to detect changes with time in the mean of a variable at one location. Long-term trends were extracted with a simple linear regression and seasonal. Spearman analysis was performed on the time-series to be statistically tested for monotonic trend [19] at p<0.05. The tests were performed on both densities and environmental series.

Prior to the series analyses, Bray-Curtis dissimilarity matrix based on log10-transformed zooplankton abundance was tested by both one-way ANOSIM and one-way orthogonal nonparametric (Permutation-based) MANOVA (PERMANOVA) [20, 21] for differences of stations with the zooplankton community using a MatLab programme, FATHOM [22]. Canonical correlation analysis, CAP [23, 24], extremely informative, was applied to a dissimilarity matrix (Bray-Curtis) based on Log10(X+1) transformed abundance matrix of the zooplankton assemblages to test the difference among the stations. The resulting scores were then analyzed using generalized discriminant analysis (CAP), giving tests based on the canonical correlations [22]. Spatial and temporal patterns in community structure were examined by multivariate techniques using PRIMER software package (version 5.2.6, PRIMER-E Ltd.) [25]. Species abundance data were Log x+1-transformed to balance the contributions from the few very abundant species with the many rare species [25] (Clarke and Warwick, 2001). Bray–Curtis similarity was used to construct a similarity matrix which made-up the basis for a 2-D ordination plot using the non-metric multi-dimensional scaling (nMDS). Multidimensional Scaling was also applied to the community analyses as a linkage and conformation of ANOSIM analyses. Those analyses helped to make decision whether individual or a whole of the stations at once was subjected to the series analyses.

IV. RESULTS

Year – to – year abundance variability

Long term variability was a significant source of variation explaining the density fluctuations of all species. Zooplankton showed important year to year fluctuations in density and it decreased as a function of distance from the coast (Fig. 2). Higher abundance variations at the coastal stations in comparison with the offshore once, suggested that zooplankton community and assemblages could be more influenced and disturbed by the anthropogenic and climatic effects as compared with open-water zooplankton community in terms of abundance and species composition.

Abundance data of the zooplankton were not normally distributed along the seasonal sampling period (normtest, PASTECS, p<0.05). Interannual variations in the zooplankton numbers were large, both in time and magnitude. Long-term time series analyses showed that abundances of the zooplankton shifted as a term of the anomalies at stations in certain periods (Fig. 3). As indication of anomalies (deviation from the relevant data),
starting with a positive phase at the beginning of the time-series 1967 till 1972, zooplankton abundance decreased steadily and it was consistently low during much of the 1970s and 1980s (Fig. 3, left panel). Figure of abundance at coastal stations (st 301 and 302) shows clear gradual decrease from 1973 till 1977 (with minimum in 1976) then a gradual increase till 1980, followed by a gradual decrease till 1983 and then no clear trend (Fig. 3, right panel). At shelf and offshore stations (st 304 and 306), the decline occurred after 1973 and it seems like there was a “delay of the decrease” between the two groups of stations. The major shift in the zooplankton has started with a decline of the zooplankton abundance since 1973 (Fig. 3). Minor positive anomalies were observed in the late period (middle 1990s). Thus, the major tendency distinguished two main periods, before and after 1973 as the second differentiated two phases of i) severe negative alterations (from 1973 to 1992) corresponding to negative anomalies and ii) the transition period 1994-2005 manifesting large variations of zooplankton quantity resulted in unsteady standing stock. Generally, the temporal distribution pattern is more or less similar for all stations and showed an overall decrease of values after 1972-73.

**Taxonomic structure, key groups and species (variability along the longitudinal gradient-stations)**

Plankton fauna is represented by species of phylum Myzozoa, Cnidaria, Rotifera, Annelida, Mollusca, Arthropoda, Chaetognatha, Chordata and Ctenophora. In general, key groups dominated in the taxonomic structure are copepods and clodocerans. The genera Acartia, Paracalanus, Oithona were mostly presented inshore, while Pseudocalanus, Calanus were dominant in offshore waters. Cladocerans (Evadne spinifera, Pseudevadne tergestina, Penilia avirostris and Pleopis polyphemoides) co-dominated in the summer and fall mainly at coastal stations. Co-dominant species to plankton fauna community structure during the whole year were also Parasagitta setosa and Oikopleura dioica. Benthic larvae (mainly Cirripedia, Polychaeta, Decapoda, Mollusca) contributed substantially to zooplankton community inshore. Usually, the estuaries and lagoons along the Western Black Sea were enriched by brackish and fresh water species as rotifers [26].
Fig. 3. Year-to-year anomalies from series of the total zooplankton abundance (left panel) during the Period 1967 to 2005.

Particular ‘Cumulative Sum’ curve (right panel). Monotonic decreased trend in the series with significant trended anomaly was found between the series of years for each of the stations (PASTECS, non-parametric Spearman, \( r_s = -0.1149615, p = 0.017350 \) for St 301; \( r_s = -0.1252591, p = 0.01941 \) for 302; \( r_s = -0.1542309, p = 0.003349 \) for St 304; and \( r_s = -0.1035267, p = 0.04288 \) for St 306).

Plankton abundance and species composition are characterized by a very high degree of spatial and temporal variability especially in coastal and shelf waters. Taxonomically, both coastal stations demonstrated similar pattern with prevalence of copepods and cladoceras in late 60s-early 70s. After 1976 the abundance of the dinoflagellate *Noctiluca scintillans* increased substantially especially in coastal and shelf stations (301, 302, 304). Also, the most specific feature of the mesozooplankton composition in the coastal waters was the important presence of meroplankton group especially during the last decade (Fig. 4). Shelf station (304) was characterized as transition area which consisted peculiarities of inshore and offshore waters. The deepest station showed different taxonomic pattern with copepods predomination (40-80%) and *N. scintillans* co-domination all over the years (Fig. 4).
One-way PERMANOVA showed significant difference in the zooplankton distribution among the stations during the years 1967-2005 (F - 13.8264, p<0.001). The canonical plot is extremely informative as it shows that real differences existed in the zooplankton assemblages at the four different stations (Fig. 5). Squared canonical correlations were 0.7441 and 0.1573 and explained 94.69% of total variation. All the test statistics resulted in the highly significant results with 9999 permutations (p=0.000), indicating strong evidence against the null hypothesis where there is no difference in the zooplankton assemblages among the four sites. In general, zooplankton assemblages along the gradient were oriented in association with the distance from the coast. Two main groups occurred as shallow water stations (St. 301 and St. 302) - coastal, and shelf (St. 304) and offshore stations (St. 306) were clearly clustered in all sampling months (Fig. 5). The ordination of samples seems to be related to the stations depth and their distance from the coastline. All two canonical axes together explained 94.69% of the variability, but the first axis contributed 83.14%. Thus the distance from the coast and the station explained the major part in the total variation associated with the zooplankton community. Presumably year as a function of the date series data was associated with the second CCA species axis (Fig. 17). Together, these two axes explained the spatiotemporal variation in the relation of the species between the stations and the long-term series dates.

![Graph showing interannual variation of zooplankton community structure](image-url)

**Fig. 4.** Interannual variation: Zooplankton community structure represented by abundance of key Taxa in percent of total zooplankton (st. 301, 302, 304 and st. 306).

**Fig. 5.** Canonical correlation analysis (CAP), applied to a dissimilarity matrix (Bray-Curtis) based on Log10(X+1) transformed zooplankton abundance matrix at stations 301 (1), 302 (2), 304 (3), and 306 (4).
To show similarities/dissimilarities among years Multidimensional Scaling (MDS) was applied. According to the samples ordination, three periods were discriminated corresponding with Black Sea ecosystem evolution (Fig. 6). The first period (1996-1973) is associated with phase of early eutrophication, the second period (1974-1993) is related with coincidence events of intensive eutrophication, invasion of M. leidyi, overfishing and loss of biological diversity, the last one (1994-2005) corresponds to B. ovata introduction, M. leidy/B. ovata interaction, post-eutrophication process, changes in phytoplankton community.

V. DISCUSSIONS

Zooplankton density showed significant oscillations through the years. The abundance of the zooplankton decreased as a function of distance from the coast. Especially in coastal and shelf waters, plankton abundance and species composition are characterized by a very high degree of spatial-temporal variability suggesting importance of local drivers. Coastal zones are transitional areas in which processes are controlled by complex interactions and fluxes of material between the land, marine water and atmospheric systems. As a result, coastal zones are among the most dynamic, rapidly changing and most vulnerable environments [27, 28].

Figure 6. MDS, applied to a dissimilarity matrix (Bray-Curtis) based on Log10(X+1) transformed zooplankton abundance matrix to discriminate the years. St.301 - circle, st. 302- square, st304 – Triangle, and st.306- diamond

The combination of a large drainage area and a high population density with the long residence time of water within the system (~ 2000 years) [29] clearly makes the Black Sea ecosystem very sensitive to land-use practices [30]. The pelagic Black Sea ecosystem has been exposed to significant changes during the last years of twenty century including habitat loss [12], collapse of predatory fish stocks [31], invasion of ctenophore Mnemiopsis leidyi [32, 33, 34, 10], and massive phytoplankton blooms resulting in hypoxia and loss of benthic communities [35, 15]. It is widely accepted that these chronically changes are at least partially attributable to intense eutrophication, particularly in coastal waters [36, 12, 37, 38, 39]. Thus, according to the Black Sea ecosystem evolution several phases were depicted – up to the early 70-ties (preutrophication), eutrophication period with two phases of early and intensive eutrophication (from the 70-ties to the early 90-ties) and a transition period after 2000. The eutrophication phase included also “era” of M. leidy invasion and wide-spread, a further diminishing of fish stocks, increasing of frequency of phytoplankton blooms [40]. The period after mid 90s coincides with new ctenophore species introduction - Beroe ovata and subsequently attributed to Beroe/Mnemiospis (predator/prey) interaction. Long-term time series analysis (anomaly and CumSum) discriminate two main phases of zooplankton community development (before 1973 and after that), abundances of the zooplankton shifted as a term of the anomalies at all shallow and deep stations in certain periods. After the pristine period (1950s early 1960s) the first response to eutrophication was clear increase in zooplankton abundance. The eutrophic Black Sea ecosystem has produced more zooplankton biomass than it used to in its pre-eutrophication phase. But, many of the dominant mesozooplankton species supporting fish populations were
replaced by smaller, less-valuable species [41]. Minor positive anomalies were recently observed, better presented in offshore stations.

In general, in the context of long-term historical data the average abundance of crustaceans maintained lower levels in the 1980s, contrary to the period 1967-1980. Multidecadal oscillations might be coupled with modified environmental conditions, especially in the late 1980s-1990s in consequence of striking change in the climate regime and Mnemiopsis pressure [42].

Applied community analysis discriminated tree main groups of zooplankton associations, associations of shallow water (including stations 301, 302), shelf and open sea, (306). In general, zooplankton assemblages were as function of the distance along the gradient coast-shelf-open sea.

Taxonomically, both coastal stations demonstrated similar pattern with prevalence of copepods and cladocerans in late 60s-early 70s and mass development of N. scintillans during the 1980s. This species became dominant with frequent and massive blooms in the periods of early and intensive eutrophication. Sharp drop of Noctiluca abundance after 1997 could be associated with SST increasing since this species shows an inverse relationship with temperature. Being a boreal cold-water organism, N. scintillans had more favorable reproduction capability in the years with cooler late-spring (May–June) temperatures after more severe winters [43]. The deepest station showed different taxonomic pattern with copepods predomination and N. scintillans co-domination. Although it has been found regularly inshore, large aggregates occurred also offshore in result of upwelling events during episodes of bloom [4].

Through the years, community structure significantly shifted with reorganization mainly of dominant zooplankton species. The most important aspect of zooplankton community after the 1970s was the change in diversity. The alterations in phytoplankton taxonomic composition, together with mass development of mixotrophic algae trigger the expansion phytophagous and detritophagous zooplankters [44]. A. clausii, P. parvus, O. similis were a constant component of plankton fauna but large copepods Pontella mediterranea and Anomalocera patersoni were almost absent during 1980s-1990s. Recently these species were recorded more often during the summer. Similar trend was evident for warm temperate copepods C. ponticus and the cladocerans Penilia avirostris, resulting in the great similarity of the samples collected in late sixties and in 2001-2005.

VI. CONCLUSION

Variability of mesozooplankton community attributes (abundance, taxonomic structure, annual dynamics) appeared vulnerable to external forcing, resulting in unsteady standing stock especially for the pelagic ecosystem in coastal waters where the influence of local drivers is significant.

The zooplankton abundance decreased as a function of distance from the coastline toward the sea. Higher variations of the coastal zooplankton abundance in comparison with the offshore, suggested that zooplankton community and assemblages could be more influenced and disturbed by the local anthropogenic and climatic effects compared with open-water zooplankton community. Taxonomic structure and community analysis supported discrimination of drawn spatial-temporal distribution patterns of near-shore versus offshore waters.

Two main periods of zooplankton community development, before and after 1973 were distinguished as indication of anomalies in certain periods and coincided with phases of the Black Sea ecosystem development. The second period could differentiate two phases of i) severe negative alterations (from 1973 to 1993) corresponding with negative anomalies and ii) the period 1994-2005 manifesting large variation in zooplankton quantity leaded to more vulnerable and unsteady zooplankton community state. Generally, the temporal distribution pattern is similar for all stations and shows decreasing trend for the whole study period. Based on the community composition and structure analysis, the two latter periods are clearly differentiated and some characteristics of the community of the 2001-2005 “seem similar” to those in the 1970s. This evolution appears to be related to the temperature evolution, characterized by the alteration of warm with cold periods and vice versa.

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