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RESPONSE OF POLYCHAETE POPULATIONS TO DISTURBANCE: AN EVALUATION OF METHODS IN HARD SUBSTRATUM

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SUMMARY

The spatial dispersion of hard substratum polychaete populations was studied in two sub-areas of the North Aegean: Thermaikos Gulf, which is under the influence of organic pollution and Chalkidiki. The evaluation of the proposed methods to assess disturbance, originally developed on soft substratum communities, reveals that multivariate analyses based on relative abundance are better applicable in hard substratum, compared to univariate ones and to those based on size changes of the fauna.

KEYWORDS: biodiversity, hard substratum, sublittoral, organic pollution, Aegean Sea.

INTRODUCTION

The eutrophication of coastal marine habitats is a current issue in the Mediterranean, due to its negative effect on biodiversity [1]. The biomonitoring of these ecosystems is a subject of priority for the members of the European Union, in order to assess their ecological quality, as forced by many international directives and conventions (e.g. Rio 1992, Water Framework Directive 2000/60/EC). Approaches to this subject have been manifold and several analyses have been proposed, which can be briefly summarized in three categories: (1) univariate techniques, including diversity and biotic indices, (2) multivariate techniques and (3) graphical/ distributional representations. These analyses, originally developed from soft substratum studies, have been applied to other marine habitats, with varying degrees of success [2-6]. Still, they are usually based on an extensive list of species of the investigated area [6] and thus on species richness that is the important indicator of diversity across spatial scales and habitats [7, 8].

Polychaetes have been widely used as a key taxon in biomonitoring studies, since they constitute a species-rich group with a perceptible response to disturbance [3, 9-12]. Still, the data on its population structure and biodiversity patterns in hard substratum are scarce and thus the response of their populations to pollution remains unclear [13]. In this study an evaluation of the applicability of the analyses of ecological monitoring in hard bottom polychaete communities is undertaken. Thus, the population structure of polychaetes from an organically polluted algal-dominated community and from a clear one was analyzed with the aforementioned techniques, and these results were further compared *inter-se*.

MATERIALS AND METHODS

Study area

The study area includes two hydrological different subareas of the North Aegean Sea: (1) Thermaikos Gulf and (2) Chalkidiki peninsula (Fig. 1). Thermaikos Gulf is a shallow-water embayment in the NW Aegean, which receives discharges from large river systems (Axios, Loudias, Aliakmonas) and also sewage and industrial effluents from the city of Thessaloniki [14, 15]. It is considered to suffer from eutrophication, especially on its northern part, where conditions of severe organic pollution have been commonly reported [16]. In contrast, both Toroneos and Sigitikos gulfs are exposed, deep-water oligotrophic areas, receiving insignificant discharges from inland waters, whereas Strymonikos Gulf receives the inputs from Strymonas River [17]. The basic physical and chemical parameters of the water column show increased variability in Thermaikos, whereas on the other three gulfs they are rather stable, showing a seasonal pattern characteristic for such temperate regions [13]. Twelve coastal stations were sampled, six located at the NE side of Thermaikos, three at the southern part of Toroneos, two at the inner Sigitikos and one at Strymonikos (Fig. 1).

Sampling

Sampling was carried out in summer at a depth ranging from 3 to 15 m, depending on the bathymetric expansion of the rocks (July & August of 1997-1998 for Chalkidiki and August 2001-2003 for Thermaikos). Macrofaunal samples were collected with SCUBA diving, using a quadrate sampler covering a surface of 400 cm^2, by totally scraping off the rocks in order to collect both the motile and sessile

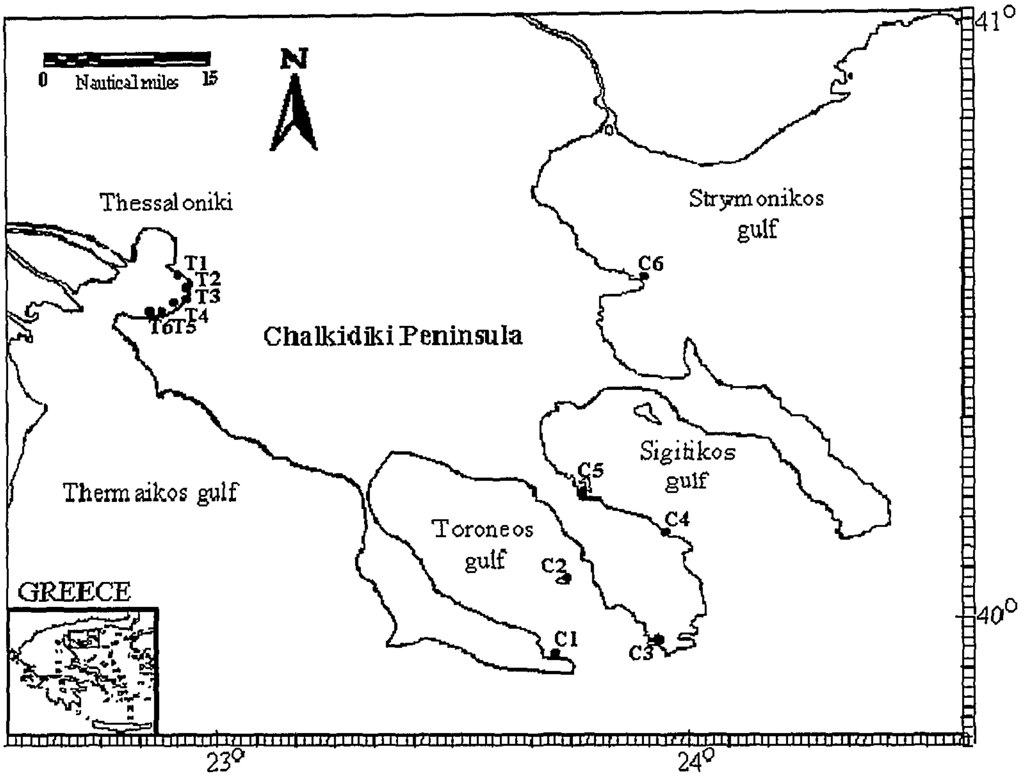

FIGURE 1
Map of the study area indicating sampling stations.

fauna [18-20]. Three to five replicates were collected at each station, thus a total of 90 samples were available (60 from Thermaikos Gulf and 30 from Chalkidiki peninsula). All samples were sieved (0.5 mm mesh size) and preserved in a 10% formalin solution. After sorting, all polychaetes were counted, identified down to species level and the dry weight of the specimens was measured. The feeding guilds for each polychaete species were classified according to Fauchald & Jumars (1979) and summarized in four categories (1) D = deposit feeders, (2) S = suspension feeders, (3) C = carnivores and H = herbivores.

Data analysis

The data matrix was first analyzed by standard biocoenotic methods (population density, mean dominance, frequency) [18, 20]. The first category of analyses, e.g. univariate techniques, included the calculation of (1) diversity indices (Margalef's richness d; Shannon-Wiener H; and Pielou's evenness J), (2) a functional/trophic relation based on the equation TR = D+S / C+H, which describes the proportion of the species utilizing the amount of suspended/deposited organic particles demanded for nutrition to the predatory or grazing species (TR is expected to increase proportionally to the organic matter), and (3) the biotic index of Borja, with the use of AZTI software [21, 22]. One-way ANOVA was used to assess the significance of the univariate results (Ho = no significant differences between the stations of the two sub-areas) [24].

The second category of analyses e.g. multivariate techniques, included the hierarchical cluster analysis and the multidimensional scaling ordination, both performed on log transformed numerical abundances data and the Bray-Curtis semimetric index, using PRIMER package [11]. The significance of the multivariate results was assessed with ANOSIM test (divergence of samples according to the geographical location of stations). SIMPER analysis was applied to identify the contribution of each species to the overall similarity within a group of samples and the dissimilarity among groups [11].

Finally, the third category of analyses, e.g. graphical/distributional representations, included the abundance-biomass comparison, ABC method [23]. For each station dominance curves for abundance and biomass were obtained as species were ranked in terms of importance on x-axis and in terms of percentage dominance of abundance or biomass on y-axis [11].

RESULTS

Univariate analyses

A total of 5,855 individuals were counted, belonging to 110 species of polychaeta. 25 species were dominant according to frequency and population density values (Table 1).

TABLE 1
Taxonomic list of the recorded species.

Polychaeta species
Abarenicola claparedei (Levinsen, 1883)
* •*Amphiglena mediterranea* (Leydig, 1851)
Amphitrite variabilis (Risso, 1826)
Amphitrite sp.
Ancistrosyllis cingulata (Korschelt, 1893)
Aonides oxycephala (Sars, 1862)
Aphelochaeta marioni (de Saint Joseph, 1894)
Aponuphis bilineata (Baird, 1870)
Aricidea fragilis Webster, 1879
Branchiomma bombyx (Dalyell, 1853)
* •*Capitella capitata* (Fabricius, 1780)
* •*Ceratonereis costae* (Grube, 1840)
Chaetozone setosa Malmgren, 1867
Chone filicaudata Southern, 1914
Chrysopetalum debile (Grube, 1855)
Cirriformia tentaculata (Montagu, 1808)
Dodecaceria concharum Oersted, 1843
Dorvillea rubrovittata (Grube, 1855)
Eteone picta Quatrefages, 1865
Euclymene oerstedii (Claparede, 1863)
Eulalia viridis (Linnaeus, 1767)
Eunice oerstedii Stimpson, 1853
Eunice torquata Quatrefages, 1865
**Eunice vittata* (Delle Chiaje, 1829)
Euphrosine foliosa Audouin & Milne Edwards, 1833
Eusyllis blomstrandi Malmgren, 1867
**Exogone naidina* Oersted, 1845
Ficopomatus enigmaticus (Fauvel, 1923)
* •*Glycera tesselata* Grube, 1863
Glycinde nordmanni (Malmgren, 1865)
Goniada maculata Oersted, 1843
Grubeosyllis limbata (Claparede, 1863)
**Haplosyllis spongicola* (Grube, 1855)
* •*Harmothoe areolata* (Grube, 1860)
Harmothoe spinifera (Ehlers, 1864)
Heteromastus filiformis (Claparede, 1864)
* •*Hydroides elegans* (Haswell, 1883)
* •*Hydroides pseudouncinata* Zibrowius, 1968
Janita fimbriata (Delle Chiaje, 1822)
Jasmineira candela (Grube, 1863)
•*Kefersteinia cirrata* (Keferstein, 1862)
Laetmonice hystrix (Savignyi, 1820)
•*Lanice conchilega* (Pallas, 1766)
Laonice cirrata (M. Sars, 1851)
Laonome salmacidis Claparede, 1870
Levinsenia gracilis (Tauber, 1879)
Lumbrineris latreilli Audouin & Milne Edwards, 1834
* •*Lysidice ninetta* Audouin & Milne Edwards, 1833
Magelona sp.
Maldane glebifex Grube, 1860
Marphysa fallax Marion & Bobretzky, 1875
Melina palmata Grube, 1870
Naineris laevigata (Grube, 1855)
Neanthes caudata (Delle Chiaje, 1828)
Nematonereis unicornis (Grube, 1840)
Nereiphylla pusilla (Claparede, 1870)
* •*Nereis rava* Ehlers, 1868
* •*Nereis zonata* Malmgren, 1867
Nicolea venustula (Montagu, 1818)
Notophyllum foliosum (M. Sars, 1835)
Odontosyllis ctenostoma Claparede, 1868
Palola siciliensis (Grube, 1840)
Paralacydonia paradoxa (Fauvel, 1913)
•*Perinereis cultrifera* (Grube, 1840)
Phalacrophorus pictus Greeff, 1879
•*Phyllodoce madeirensis* Langerhans, 1880
Pionosyllis lamelligera de Saint Joseph, 1886
Pista cristata (O.F. Muller, 1776)
Placostegus crystallinus Zibrowius, 1968
* •*Platynereis dumerilii* (Audouin & Milne Edwards, 1833)
Polycirrus aurantiacus Grube, 1860
Polydora caeca *(Oersted, 1843)*
Polygordiidae (archianelida)
**Polyopthalmus pictus* (Dujardin, 1839)
Pomatoceros triqueter (Linnaeus, 1767)
Protodorvillea kefersteini (McIntosh, 1869)
Protula sp.
Pterocirrus macroceros (Grube, 1860)
Pterosyllis formosa
Sabella fabricii Fauvel, 1927
Sabella pavonina Savignyi, 1820
Sabella spallanzanii (Viviani, 1805)
Sabellaria spinulosa Leuckart, 1849
Schistomeringos rudolphi (Delle Chiaje, 1828)
Sclerocheilus minutus Grube, 1863
Scolelepis bonnieri (Mesnil, 1896)
* •*Scoletoma funchalensis* (Kinberg, 1865)
* •*Serpula concharum* Langerhans, 1880
* •*Serpula vermicularis* Linnaeus, 1767
Spermosyllis torulosa Claparede, 1864
* •*Sphaerosyllis pirifera* Claparede, 1868
Spirobranchus polytrema (Philippi, 1844)
**Spirorbis* sp.
Syllidia armata Quatrefages, 1865
Syllis amica Quatrefages, 1865
•*Syllis armillaris* (O.F. Muller, 1776)
**Syllis cornuta* Rathke, 1843
Syllis gracilis Grube, 1840
* •*Syllis hyalina* Grube, 1863
Syllis krohnii Ehlers, 1864
* •*Syllis prolifera* Krohn, 1852
Syllis vittata (Grube, 1840)
Terebella lapidaria Linnaeus, 1767
Terebellides stroemi M. Sars, 1835
Theostoma oerstedi (Claparede, 1864)
Trypanosyllis coeliaca Claparede, 1868
**Trypanosyllis zebra* (Grube, 1840)
* •*Vermiliopsis infundibulum* (Philippi, 1844)
Vermiliopsis limbata (G.O. Costa, 1861)
Vermiliopsis sp

* indicate dominant species, • indicate species contributing on about 50% of the average in-group similarity or among groups dissimilarity resulted from SIMPER analysis.

Richness values (d) ranged from 3.84 to 7.53, H values from 1.44 to 4.56 and J values from 0.26 to 0.90, indicating the evenly dispersion of numerical abundance among species in most sampling stations (Fig. 2).

One-way ANOVA showed that the values of H and J indices were significantly different among Thermaikos and Chalkidiki stations, only after the exclusion of spirorbids (F=10.34; $p<0.05$; F= 21.64 $p<0.05$, respectively), whereas d didn't showed any significant variation (F= 0.09; p=0.76).

The functional/trophic ratio (TR) showed higher values in Thermaikos stations (mean value 1.137±0.91), whereas in the stations of Chalkidiki was around 0.18±0.06 (Fig. 3). Still, these differences were not statistically confirmed (F= 0.02 p=0.88), as the high value of TR in C2 amplifies the variance in the group of Chalkidiki stations.

The increased value in C2 station is due to the massive settlement of spirorbids, which are typical filter feeder organisms, often classified in meiofauna [18]. The recalculation of TR, after the exclusion of spirorbids reinstates its values, which are now found significantly different among Thermaikos and Chalkidiki stations (F=6.46, $p<0.05$). Accordingly, the relative abundance of species utilizing the organic particles was increased in Thermaikos.

The biotic coefficient (BC) ranged from 1.38 to 2.9. Thus, the biotic index of Borja (BI) had a value of 2 in all cases and as a result all sampling stations were classified as slightly polluted. However, the percentage contribution of the five ecological categories of species showed a different pattern among Thermaikos (dominance of group III) and Chalkidiki (dominance of group II) stations (Fig. 4).

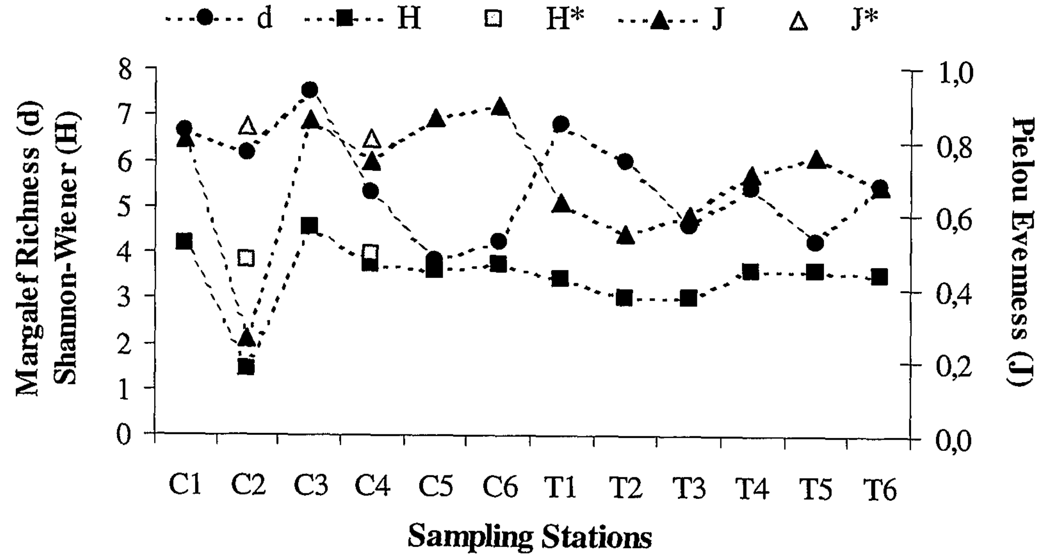

FIGURE 2
Spatial variation of diversity indices. Asterisk refers to the recalculated values after the exclusion of spirorbids.

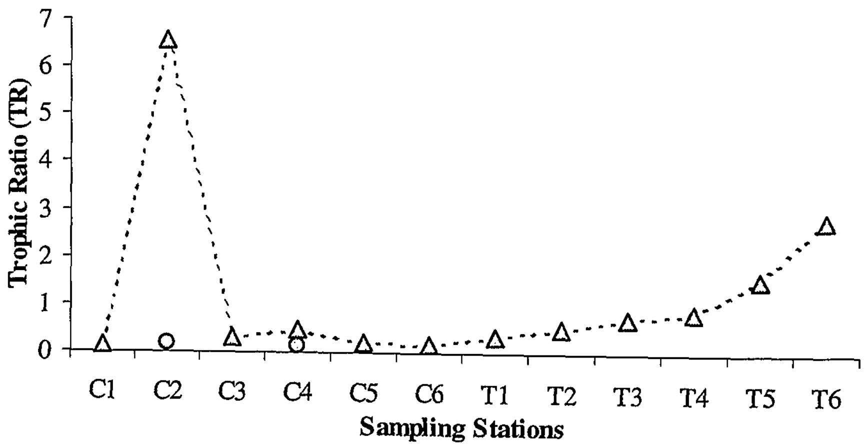

FIGURE 3
Value of the trophic ratio (TR) per sampling station. The circle refers to the recalculated value after the exclusion of spirorbids.

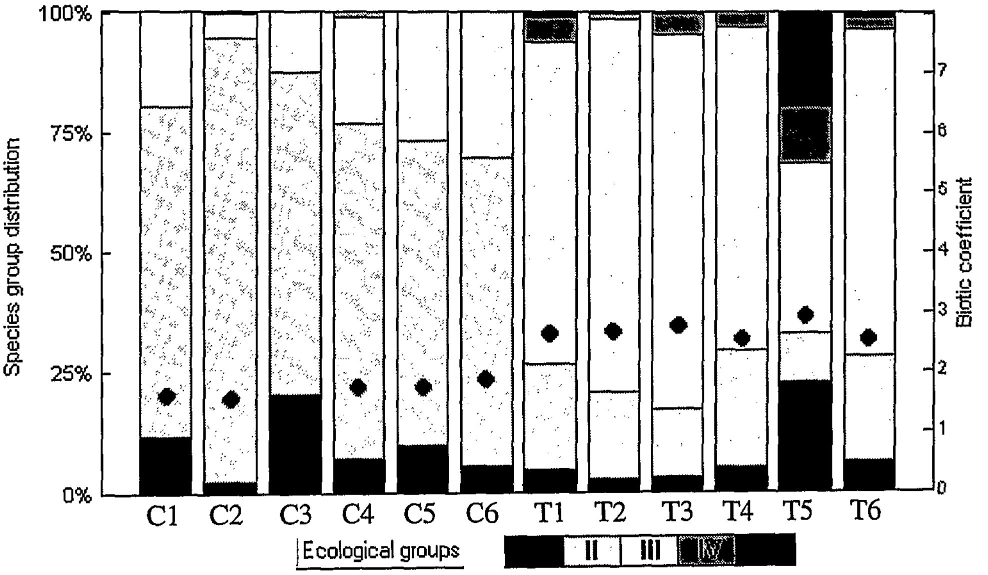

FIGURE 4
Biotic coefficient value (BC) and percentage of the ecological groups per sampling station.

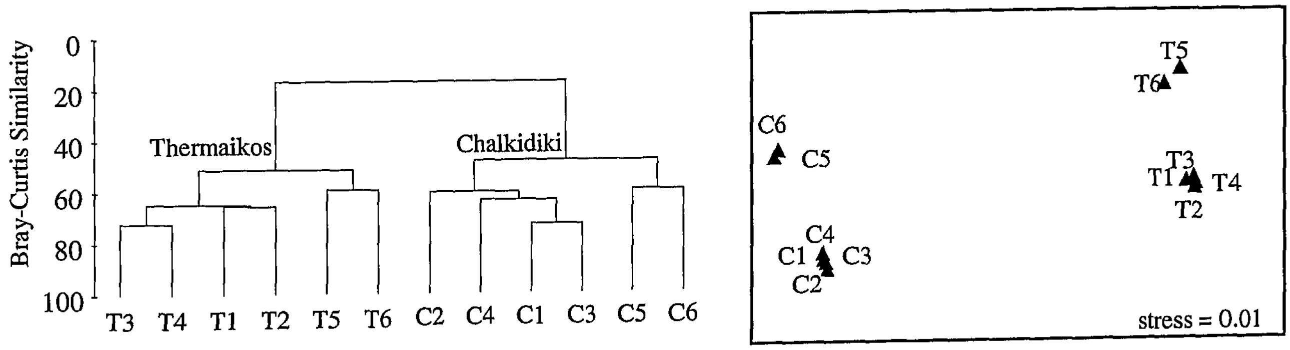

FIGURE 5
Non-metric multidimensional scaling, based on Bray-Curtis similarity index, calculated from root transformed numerical abundance data.

Multivariate analyses

Hierarchical cluster analysis and non-metric MDS show the separation of the samples in two groups according to the sampling site, e.g. Thermaikos or Chalkidiki (Fig. 5). Within each group a second minor division is apparent, according to the community type. In Thermaikos group the facies of *Mytilus galloprovincialis* (T5, T6) is distinguished from the photophilic algae community (T1, T2, T3, T4). In Chalkidiki group the facies of the filamentous algae (C1, C2, C3, C4) is distinguished from the facies of encrusting algae. The discrimination of the samples from Thermaikos and Chalkidiki stations is confirmed by one-way ANOSIM (global $R = 0.1$; $p < 0.1$). The SIMPER analysis identified 15 species as most contributing to in-group similarity and 22 species to among-groups dissimilarity (Table 1).

Graphical/distributional representations

The ABC curves obtained for Thermaikos and Chalkidiki stations are presented in Figure 6. In most stations of Thermaikos, the two curves coincide or cross each other, except of T4 and T5, in which the biomass curve was above that of abundance. Regarding the stations from Chalkidiki, the biomass curve was above that of abundance, producing a k-dominance pattern. For C2, however, the pattern was reverse, and for C4 the two curves coincide.

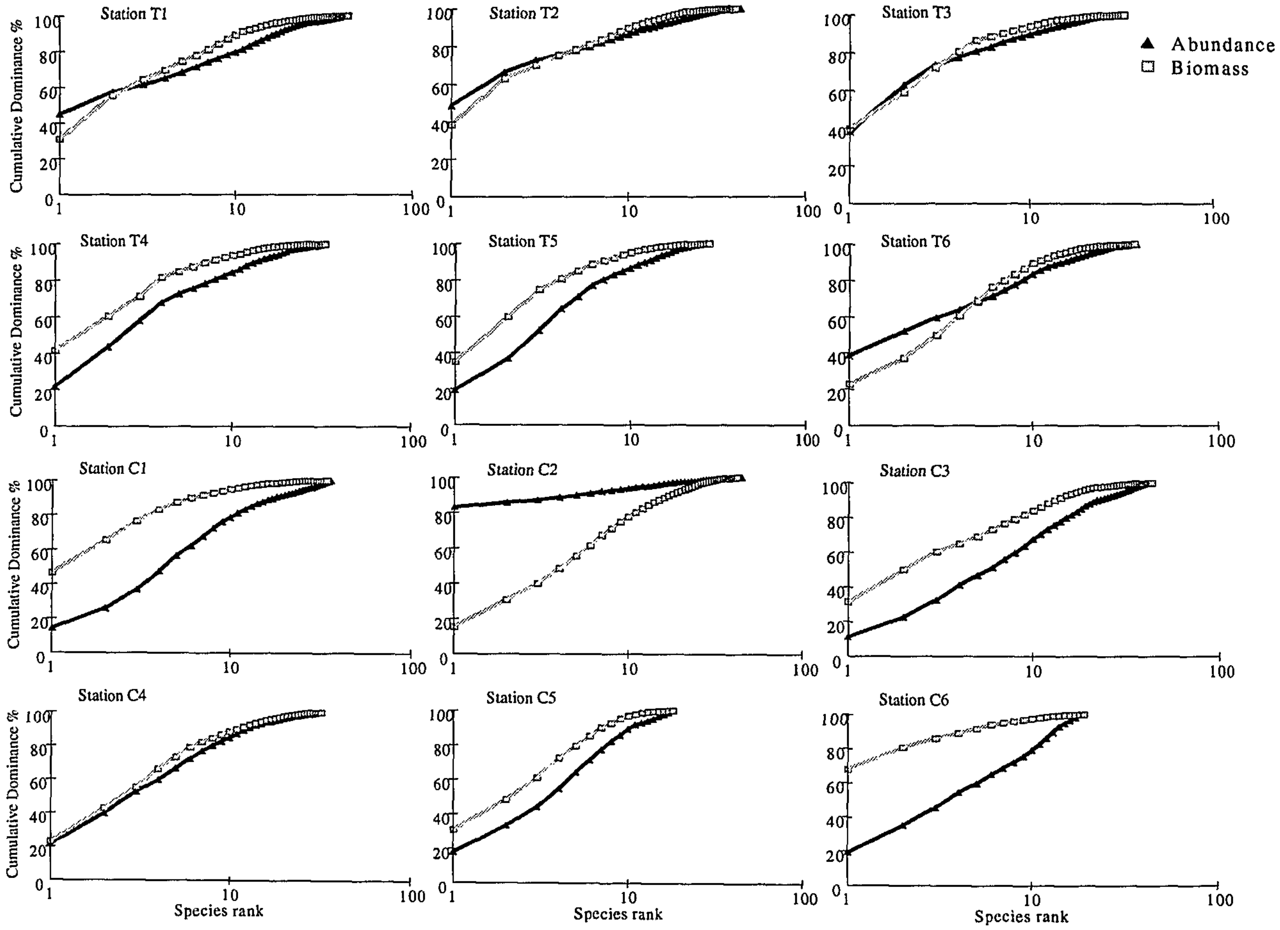

FIGURE 6
ABC curves for the stations of Thermaikos (T1-T6) and Chalkidiki (C1-C6).

DISCUSSION

Algal-dominated communities prevail the sublittoral rocky bottoms throughout the Mediterranean [25]. These communities are characterized by increased biodiversity and complexity. They host a large number of zoobenthic species with various ecological fit, among which polychaetes usually comprise over one third of both the species richness and the numerical abundance [5, 13, 20, 26]. In this study a total of 110 species of Polychaeta were recorded, all previously reported in the Aegean Sea [27, 28].

The univariate analyses showed the same overall trends. Thus, the diversity indices had decreased values in Thermaikos, in contrast with the trophic ratio, which showed increased values. These two methods confirmed the diversification of the stations from the two sub-areas of the North Aegean. They also identified some pollution effects (in the sense of the decreased biodiversity and the predominance of suspensivores/ deposit feeders) to the set of Thermaikos stations. However, the methods showed two severe disadvantages. The first one is related to the very strong influence of the results to the relative proportion of some species. This was obvious from the C2 results. This station was characterized as polluted by the application of both trophic ratio and diversity indices. However, this was due to the increased abundance of spirorbids, which were attached to the thalli of the brown alga *Padina pavonica* [13]. Spirorbids are typical opportunistic filter feeding organisms that settle immediately after release from parental tubes [29]. Their presence to the assemblage should not be considered as stable, as they probably perish when the thalli of *Padina pavonica* decay after mid September [13]. Moreover, are often classified to the meiofaunistic component of the community, due to their very small size [18]. Thus, the exclusion of such species is recommended to better describe the community patterns [13, 30]. The second disadvantage is the relativity of the numerical values of indices, since the differences between Thermaikos and Chalkidiki stations were statistically confirmed only after the exclusion of spirorbids.

The biotic index of Borja failed to distinct the various stations, as all sites were classified as slightly polluted.

Thus, the BI values are obviously underestimated from Thermaikos and overestimated for Chalkidiki stations. This failure may have resulted for a number of reasons. First of all, many polychaete species were not yet assigned to ecological groups, and thus excluded from the analysis [21, 22]. Secondly, none of the species favored by the increased organic load showed a peak to their population density. The only apparent difference among Thermaikos and Chalkidiki stations refers to the relative proportion of two ecological groups. Thus, species indifferent to enrichment (group II) dominated Chalkidiki stations and species tolerant to increased organic load (group III) dominated Thermaikos. The species pool differs among hard and soft substratum communities, the later hosting exclusively species belonging to groups IV and V (e.g. certain cirratulids). Thus, a different arrange of the fauna to ecological groups adapted to rocky bottoms could improve the BI results allowing the eduction of a solid conclusion.

The abundance biomass comparison describes the relative proportions of r- to k- strategies. The taxon of Polychaeta is strongly recommended for this analysis, as the distributions of abundance and biomass among species is shifting with respect to organic pollution. The increment of organic load will generate the substitution of large by small sized species [23, 11]. However, this method has been scarcely performed on rocky bottoms [6]. Our results generally confirmed the predictions of the method. Still some misjudgments came up. In most of the stations of Chalkidiki large bodied polychaetes dominated, indicating stable community trends. The extremely high number of spirorbids, species indifferent to organic enrichment [21] with very small body size and thus negligible biomass can explain the exceptions of C2 and C4 results. Regarding Thermaikos stations, four of them (T1, T2, T3, T6) are classified as moderately perturbed, while T4 and T5 were unperturbed. In T4 the species *Eunice vittata*, *Lanice conchilega* and *Sabella spallanzanii* contribute most to the biomass, whereas *Marphysa fallax* and *Ceratonereis costae* in T5. All these polychaetes are either sensitive or indifferent to organic pollution [21, 28]. Thus, the results of ABC method in hard substratum communities should be viewed with caution, by referring to the species responsible for the configurations.

The last methodological approaches, e.g. the multivariate analyses produced the same overall patterns. According to these results, the polychaete populations are differently structured according to the investigated subareas. The organically polluted algal-dominated communities were undoubtedly distinguished from the clear ones, on the basis of the relative abundance of the species pool. Moreover, the various stations sampled were separated with respect to the prevalent facies, which indicates the increased sensitivity of the methods applied.

Concluding, the effect of the organic pollution, well documented in inner Thermaikos Gulf [3, 4, 5, 12, 16], was detectable on the current biotic pattern. The results support the usefulness of the multivariate techniques in ecological monitoring in hard substratum, since they clearly separated the unpolluted from the organically polluted sites. The other analyses performed were found less effective, mostly due to their dependence on the attributes of some species (abundance, biomass, sensitivity or feeding guild). Consequently, information on the biology of the species pool and their biotic interaction is required for the interpretation of the results produced by such methods.

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