

## Ecological Indicators 52: 292-299

**Title:** Invasive ecosystem engineers and biotic indices: giving a wrong impression of water quality improvement?

**Authors:** Anastasija Zaiko<sup>a</sup>, Darius Daunys<sup>a,b</sup>

<sup>a</sup> Marine Science and Technology Center, Klaipeda University, H. Manto 84, LT 92294, Klaipeda, Lithuania

e-mail: [anastasija@corpi.ku.lt](mailto:anastasija@corpi.ku.lt)

<sup>b</sup> Ecology Department, Faculty of Natural Sciences and Mathematics, Klaipeda University, H. Manto 84, LT 92294, Klaipeda, Lithuania

e-mail: [darius@corpi.ku.lt](mailto:darius@corpi.ku.lt)

Corresponding author: Anastasija Zaiko, Marine Science and Technology Center, Klaipeda University, H. Manto 84, LT 92294, Klaipeda, Lithuania, Tel.: +370 46 398855, Fax.: +370 46 398845, e-mail: [anastasija@corpi.ku.lt](mailto:anastasija@corpi.ku.lt)

### Abstract:

Benthic component of an ecosystem is considered in ecological status assessment of the key European Directives. Most of the metrics proposed for the benthic quality assessment are biodiversity based. Their robustness and applicability are widely discussed in many recent studies. However an impact of invasive alien species on biotic indices and environmental quality assessments has been largely overlooked by researchers so far. In the current study we assessed Benthic Quality Index (BQI) in a coastal ecosystem, highly affected by the invasive zebra mussel *Dreissena polymorpha*. Zebra mussel is able of modifying benthic habitats and enhancing local biodiversity. In the analyzed ecosystem it affected benthic species richness, abundance and community structure. As a result the calculated BQI values were significantly higher in the presence of zebra mussel with evident outliers in samples with particularly high zebra mussel abundances. Therefore we found that BQI determined in our study was artificially elevated providing false signal of the ecological status improvement. Based on the results presented, we suggested data correction framework that has been tested on the current dataset and proved to be effective minimizing zebra mussel impact on BQI assessment. Our experience could be applied for other coastal ecosystems invaded by the zebra mussel or any other aquatic invasive species with resembling biological traits and bioinvasion impacts.

### Highlights:

- We test the effect of an invasive alien species on ecological quality assessment
- We calculate Benthic Quality Index for the coastal lagoon affected by zebra mussel
- Zebra mussel may modify benthic habitats enhancing local biodiversity
- This might bias BQI by showing false improvement of ecological status
- We suggest a framework how the this bias could be minimized

**Key words:** invasive species, bioassessment, BQI, *Dreissena polymorpha*, zebra mussel, Baltic Sea, Curonian Lagoon

## 1. Introduction

The demand for the universal biotic indicators aimed at ecological status assessment has increased with the development of the key EU Directives, focused on reduction of anthropogenic pressures, improvement of aquatic environment and preventing biodiversity loss (Borja et al. 2010, Borja et al. 2013, Tett et al. 2013). EU Water Framework Directive (WFD) and Marine Strategy Framework Directive (MSFD) consider a number of ecological quality parameters, both having benthic component involved (as “Macrofauna” in WFD and “Sea floor integrity” in MSFD). There is a number of biotic metrics proposed for the benthic ecological quality assessment, including (but not limited to) Infaunal Trophic Index (ITI) (Maurer et al. 1998); Benthic Index of Biotic Integrity (B-IBI) (Kerans and Karr 1994); Azti-Marine Biotic Index (AMBI) (Borja et al. 2000); Benthic Quality Index (BQI) (Rosenberg et al. 2004); Benthic Opportunistic Polychaetes and Amphipods Index (BOPA) (Dauvin and Ruellet 2007); Infaunal Quality Index (IQI) (Kennedy et al. 2011).

All of them are species richness based indices utilizing quantitative characteristics of benthic communities. Indices assume that bottom-dwelling fauna are sedentary enough to escape from deteriorating environmental conditions and therefore will relatively rapidly respond to human induced pressures (Pearson and Rosenberg 1978, Borja et al. 2000, Diaz et al. 2004, Villnas and Norkko 2011).

To be considered as appropriate for ecological status assessment an indicator should meet the following criteria: be scientifically based (Rice 2003, Rice and Rochet 2005, Mee et al. 2008, Niemeijer and de Groot 2008, Elliott 2011); ecosystem relevant and biologically important (Niemeijer and de Groot 2008, Elliott 2011); responsive, sensitive, specific and predictable (Rice 2003, Rice and Rochet 2005, Mee et al. 2008, Niemeijer and de Groot 2008, Elliott 2011, Kershner et al. 2011); accurate and practical in terms of measurability and cost effectiveness (Rice and Rochet 2005, Niemeijer and de Groot 2008, Kershner et al. 2011).

When evaluating the environmental status of marine waters the effects of chemical pollution, eutrophication, habitat destruction and overexploitation are being addressed (Olenin et al. 2011). Consequently, the suitability of indicators is being tested and validated predominantly in relation to those pressures. However an impact of invasive alien species (IAS) present in the considered ecosystem has been largely overlooked by researchers so far. IAS may induce multiple important alterations in the recipient ecosystem including changes in structure and distribution of native species assemblages, habitat properties, food web structure and biogeochemical processes (Elliott 2003, Reise et al. 2006, Olenin et al. 2007, Zaiko et al. 2011). Therefore, it is likely that impacts of other stressors may be surpassed and the correspondent ecosystem responses masked (Olenin et al. 2011).

In the current study we hypothesize, that presence and impact of an invasive ecosystem engineer may significantly influence quantitative metrics of biodiversity and therefore affect the overall ecological status assessment. In order to challenge this hypothesis, we assessed the performance of Benthic Quality Index (BQI) in a coastal ecosystem, highly affected by the zebra mussel *Dreissena polymorpha*.

BQI is a widely used multimetric indicator of benthic community condition and functionality (Rosenberg et al. 2004, Fleischer et al. 2007, Fleischer and Zettler 2009, Leonardson et al. 2009). Although designed for application in marine areas (Borja et al. 2003, Rosenberg et al. 2004), it has proved to be suitable for areas with strong salinity gradients given that tolerance levels of species are properly adjusted and assigned for the specific area (Zettler et al. 2007). BQI is reproducible and has been tested and validated in different marine ecosystems with

varying environmental conditions (e.g. Labrune et al. 2006, Fleischer et al. 2007, Zettler et al. 2007), therefore it was advised by the international expert groups (e.g. HELCOM CORESET) for distinguishing impacted habitats from undisturbed ones.

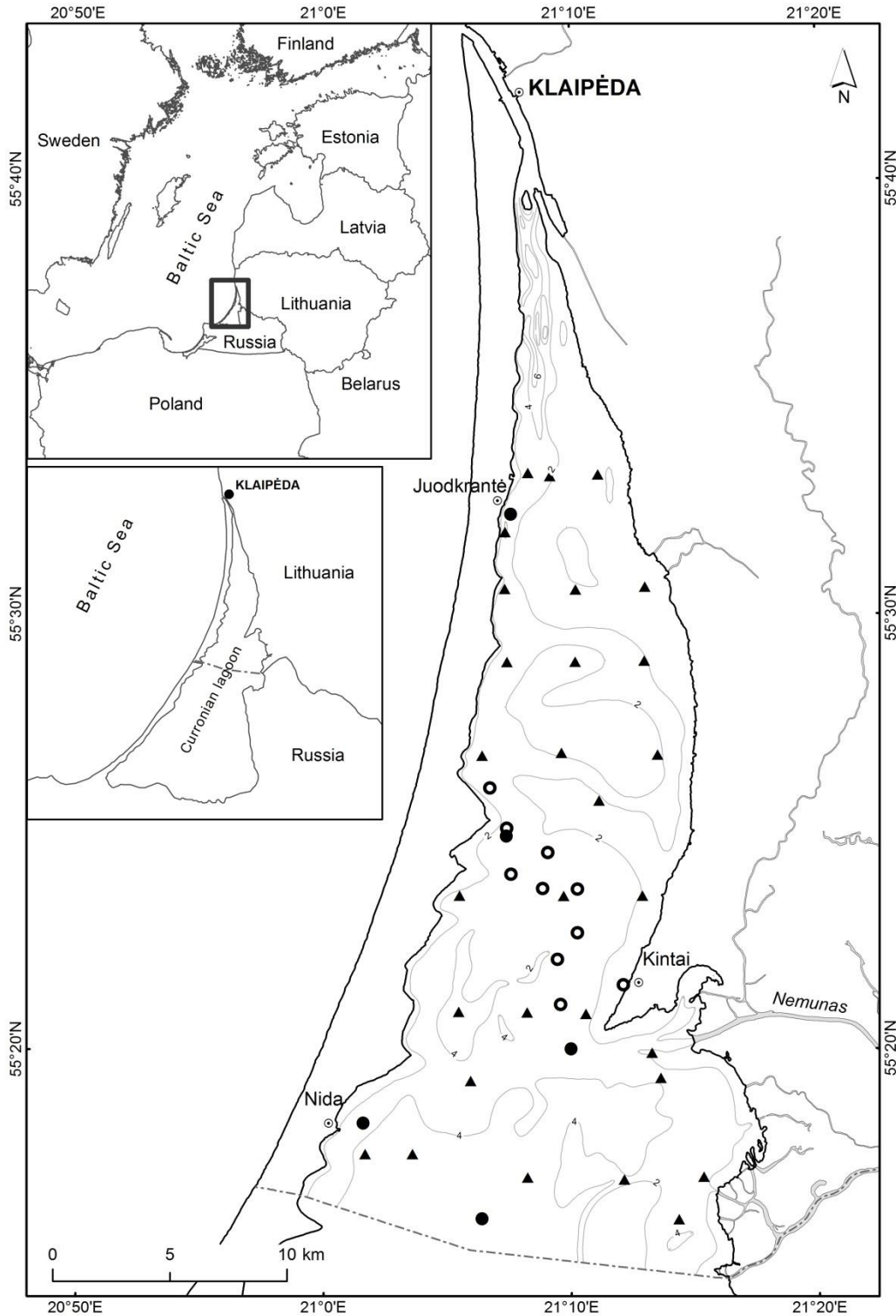
Zebra mussel is known as a powerful ecosystem engineer capable of modifying physical, morphological, biological and biogeochemical properties of the bottom habitats (Stewart et al. 1998, Karatayev et al. 2002, Minchin et al. 2002, Zaiko et al. 2009, Zaiko et al. 2010). As it was previously reported, zebra mussels are generally associated with increased benthic macrofauna abundance, species richness and decreased community evenness (Ricciardi et al. 1997, Zaiko et al. 2009, Atalah et al. 2010). Being one of the most abundant and widely distributed IAS in the oligohaline regions of the Baltic Sea (Zaiko et al. 2011, Fenske et al. 2013) zebra mussels produce dense colonies and beds of empty shells forming patches of high biodiversity and facilitating establishment of native and non-indigenous species (Zaiko et al. 2007, 2009). It has been shown recently that some eutrophication-related metrics (e.g. nutrient levels, chlorophyll concentrations, water clarity) might be affected and lose their explanatory value in ecosystems invaded by zebra mussel (Atalah et al. 2010; Zaiko et al. 2014). Therefore, in this study we test its effect on a benthic quality assessment and suggest a framework how the IAS-related bias could be minimized.

## **Material and methods**

### *Study area*

The Curonian Lagoon is a large (1.584 km<sup>2</sup>), shallow (average depth 3.8 m) coastal water body connected to the south-eastern Baltic Sea by the narrow (0.4–1.1 km) Klaipeda Strait (Fig. 1). The ecosystem is greatly dependent on the Nemunas river runoff (98% of the total freshwater discharge), draining substantial amount of nutrients from the basin (Zaromskis 1996). Ongoing eutrophication is one of the most important problems in the lagoon, affecting all ecosystem components including bottom habitats (Olenina and Olenin 2002, Olenin and Daunys 2004, Aleksandrov 2010).

The lagoon is oligohaline in its narrow northern part (with irregular rapid salinity fluctuations in the range of 0.5 to 5-6 PSU) and limnic in its central and southern parts (with a relatively closed water circulation and lower current velocities. Therefore these parts serve as the main depositional area of the lagoon (Olenina and Olenin 2002, Gasiunaite et al. 2008).



**Figure 1.** Sampling sites in the Curonian Lagoon. Filled circles indicate permanent monitoring stations, open circles - survey stations in 2006, triangles – survey stations in 1999.

*D. polymorpha* was probably introduced into the Curonian Lagoon in the early 1800s. The molluscs were presumably attached to timber rafts and reached the lagoon via the central

European invasion corridor (Olenin et al. 1999, Fenske et al. 2013). Currently, zebra mussels are highly abundant in the Curonian Lagoon, occupying the littoral zone down to 3–4 m depth and occurring on both hard substrates and soft bottoms. The habitats affected by zebra mussel comprise nearly ¼ of the lagoon bottom area with the largest zebra mussel community located in the central part (Zaiko et al. 2009). Soft bottom devoid of zebra mussels is dominated by oligochaetes, chironomids and another IAS *Marenzelleria neglecta* (Zettler and Daunys 2007).

#### *Data collection*

In this study, data on macrofauna abundances from the Curonian Lagoon were analyzed for BQI development and assignment of sensitivity values. We used a long-term (2000-2010) dataset of 12 sampling events at 5 permanent monitoring sites resulting in 113 benthic macrofauna samples (Fig. 1). To enhance the data resolution, additionally we included 30 macrofauna samples from 10 sampling sites surveyed in the course of comprehensive study of zebra mussel population in 2006 (Fig. 1). For the further validation of results, the developed framework was tested with a smaller dataset from the 1999 survey (32 benthic macrofauna samples). Index response to organic carbon content in sediments (as a proxy of eutrophication-related pressure) in the context of zebra mussel presence was verified on those data.

All the samples were collected using Van Veen grab with 0.1 m<sup>2</sup> sampling area, and analyzed following standard guidelines for bottom macrofauna sampling (HELCOM 1988). Due to the high small-scale bottom patchiness (Olenin and Daunys 2004, Zaiko et al. 2009), replicate grabs from one station and sampling event were not averaged for abundance and species number and considered as individual samples (as e.g. in Leonardsson et al. 2009).

To reduce the inconsistency in the taxonomic resolution of the dataset, part of the species were pooled into the higher taxonomic groups: e.g., Oligochaeta (excluding *Eiseniella tetraedra*), Chironomidae, Trichoptera, Turbellaria, Nematoda, Heteroptera, Nemertea, Gammaridae, Unionidae, *Pisidium*, *Valvata*, *Sphaerium*). Species with presence/absence data only (e.g. hydroids *Hydra vulgaris*, *Cordylophora caspia*) were not included into analysis.

#### *Benthic quality index calculation*

The macrofauna abundance data were used for the computation of the Benthic Quality Index (BQI) (Rosenberg et al. 2004). Since the original version of BQI is known to be sampling effort dependent (e.g. increase in sampling effort results in higher probability of obtaining rare species), the adjusted calculation was applied (Fleischer et al. 2007, Fleischer and Zettler 2009):

$$BQI_{ES} = \left( \sum_{i=1}^n \left( \frac{A_i}{A_{tot}} \times ES_{50,0.05i} \right) \right) \times \log(ES_{50} + 1) \times \left( 1 - \frac{5}{5 + A_{tot}} \right) \quad (1)$$

In Eq. (1) above,  $n$  denotes the observed species number.  $A_i$  stands for the abundance of the species  $i$  and  $A_{tot}$  is the sum of all individuals within this square meter. Finally,  $ES_{50-0.05}$  is the sensitivity/tolerance value for the species  $i$  and  $ES_{50}$  denotes the expected number of species for 50 individuals randomly taken from the square meter (Hurlbert Index).

The Primer software package (Clarke and Warwick 2001) was used for calculation of the Hurlbert Index ( $ES_{50}$ ). Species recorded in 10 samples or less (occurrence approx. less than 10 % in our case) were excluded from further sensitivity determination, but were considered when estimating  $n$  and  $A_{tot}$  in Eq. (1) following approach used in other studies (e.g.

Leonardsson et al. 2009). No samples were discarded from the analysis due to the low total abundance (less than 50 individuals) as advised by the other authors (Rosenberg et al. 2004, Puente and Diaz 2008, Fleischer and Zettler 2009). As originally proposed by Rosenberg et al. (2004), the sensitivity value of a species was set to the 5<sup>th</sup> percentile of the  $ES_{50}$  ( $ES_{50-0.05}$ ). This approach follows the assumption that the most tolerant species are likely to be associated with the lowest biodiversity, lower  $ES_{50}$  values and therefore attaining lower sensitivity estimates.  $ES_{50-0.05}$  was calculated as described by Leonardsson et al. (Leonardsson et al. 2009). Based on the estimated sensitivity values, the pre-selected species were classified by expert judgment as ‘*very tolerant*’, ‘*tolerant*’, ‘*sensitive*’ and ‘*very sensitive*’.

#### *Dataset correction*

In order to minimize the IAS effect on the BQI assessment outcome, following correction framework was applied on the original dataset. First, the species observed only in samples with zebra mussel were eliminated in order to reduce artificially elevated  $ES_{50}$  values in locations with zebra mussels. Then, the samples with particularly high zebra mussel abundances were excluded. Since habitats dominated by zebra mussel maintain benthic communities structurally different from those observed in areas with no or low numbers of zebra mussels (Thayer et al. 1997, Strayer et al. 1998, Zaiko et al. 2009, Minchin and Zaiko 2013), we set a threshold of zebra mussel abundance at approx. 1000 ind/m<sup>2</sup>, corresponding to a few average size clumps which are capable to modify the soft-bottom habitats to the stage when zebra mussel-specific communities form (Zaiko et al. 2009). Finally, abundance correction was applied for species demonstrating significant correlation with zebra mussel. Here we used a proportional correction, based on the coefficients determined in the regression model:

$$A_{i-cor} = \frac{A_i}{\beta \cdot A_{zm}} \quad (2)$$

In Eq. (2) above  $A_{i-cor}$  stands for the corrected abundance of the species  $i$ ,  $A_i$  – initial species abundance observed in the sample,  $\beta$  – slope (standardized) coefficient from the fitted linear regression model ( $y = \alpha + \beta x$ ),  $A_{zm}$  – zebra mussel abundance in the sample (note: the correction should be applied for samples with zebra mussel only; otherwise the initial values are left).

#### *Statistical analysis*

Logarithmic transformations were applied to macrofauna abundance and organic carbon content data in order to avoid distortion resulting from the outlying values, defined during the exploratory data analysis (visual assessment of box-plots and QQ-plots).

Non-parametric Mann–Whitney (Wilcoxon) W test was used to test differences (e.g. pairwise comparisons of BQI values in samples with and without zebra mussels) when datasets were unbalanced and did not meet normality assumptions. Linear regression model with a robust fitting algorithm was applied to ascertain the effect of the zebra mussel abundance on BQI values. In case of multiple pairwise comparisons or correlation analyses (e.g. for correlations between abundances of zebra mussel and other species) the Bonferroni correction for  $\alpha$  was applied.

Analysis of Covariance (ANCOVA) was used to verify the effects of organic carbon contents (% of sediment dry weight) on BQI values, with zebra mussel presence as a co-variate (two groups) and compare the regression slopes and intercepts between groups. Prior to that, the compliance with assumptions of homogeneity of group variances and independence of

predictor variables was tested. The results confirmed that assumptions are fulfilled ( $F=2.47$ ;  $p=0.09$  and  $F=3.79$ ;  $p=0.06$  respectively).

The analyses were implemented in the R v3 statistical computing environment (R-project 2014).

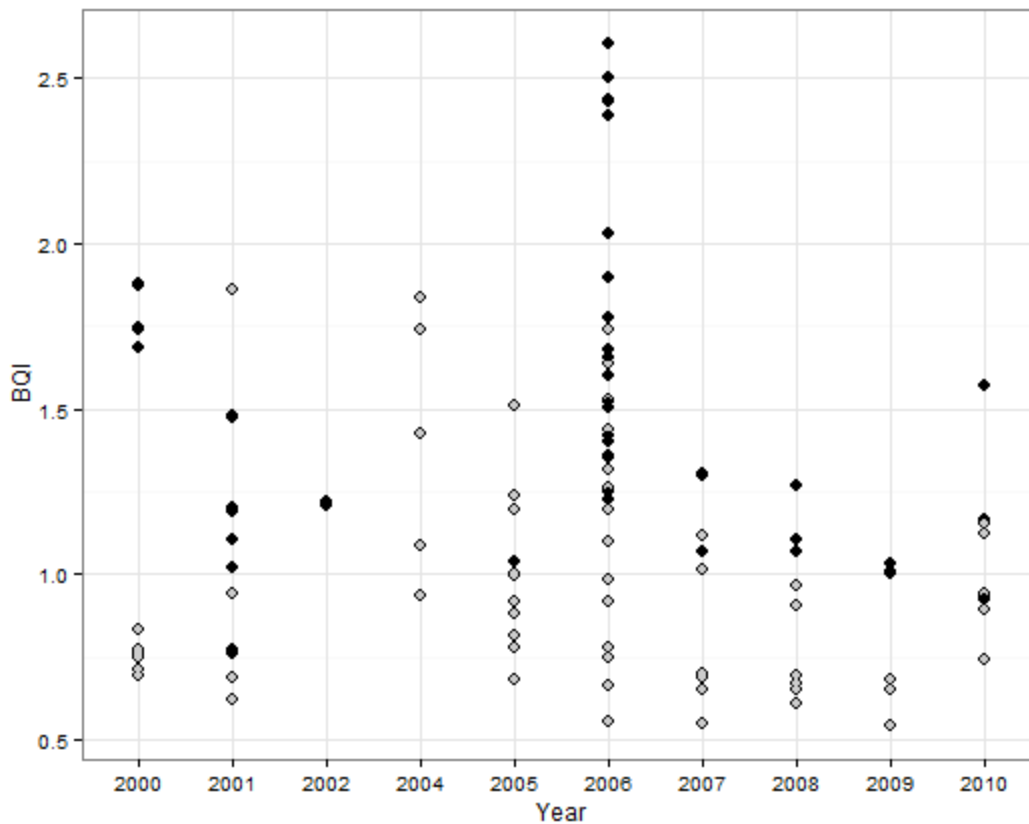
## 2. Results

When applying the rule of  $ES_{50}$  calculation for species occurring in  $\geq 10$  samples only, we were able to assign sensitivity values for 19 species/taxa (Table 1).

**Table 1. Sensitivity values of the pre-selected 19 taxa, from uncorrected data analysis and suggested sensitivity class (1 – very tolerant; 2 – tolerant; 3 –sensitive; 4 – very sensitive)**

Taxa	$ES_{50-0.05}$	Sensitivity class
Oligochaeta	1.4	1
Chironomidae	2.0	2
Gammaridae	2.2	2
Unionidae	2.2	2
<i>Valvata spp.</i>	2.4	2
Hydracarina	2.5	2
Ostracoda	2.8	2
<i>Glossiphonia complanata</i>	3.0	3
<i>Eiseniella tetraedra</i>	3.0	3
Trichoptera	3.1	3
Turbellaria	3.1	3
<i>Helobdella stagnalis</i>	3.2	3
<i>Pisidium sp.</i>	3.4	3
<i>Dreissena polymorpha</i>	3.5	3
<i>Erpobdella octoculata</i>	3.5	3
<i>Sphaerium spp.</i>	3.9	3
<i>Glossiphonia heteroclita</i>	4.0	4
<i>Viviparus viviparus</i>	4.1	4
<i>Bithynia spp.</i>	4.7	4

Four sensitivity classes were determined and assigned to the species using the nearest default non-decimal numbers for estimated  $ES_{50-0.05}$  values: very tolerant ( $ES_{50-0.05} < 2.0$ ); tolerant ( $2.0 \leq ES_{50-0.05} < 3.0$ ); sensitive ( $3.0 \leq ES_{50-0.05} < 4$ ) and very sensitive ( $4 \leq ES_{50-0.05}$ ). There were 15 other species or higher order taxa with the occurrence ranging from less than 1% to 8%: Nemertea, Nematoda, Hemiptera, Heteroptera, Ceratopogonidae, Corophiidae, Simuliidae, *Marenzelleria spp.*, *Asellus aquaticus*, *Caenis macrura*, *Glossiphonia concolor*, *Gordius aquaticus*, *Piscicola geometra*, *Potamopyrgus antipodarum*, *Radix auricularia*. These taxa were not included into sensitivity assessment.



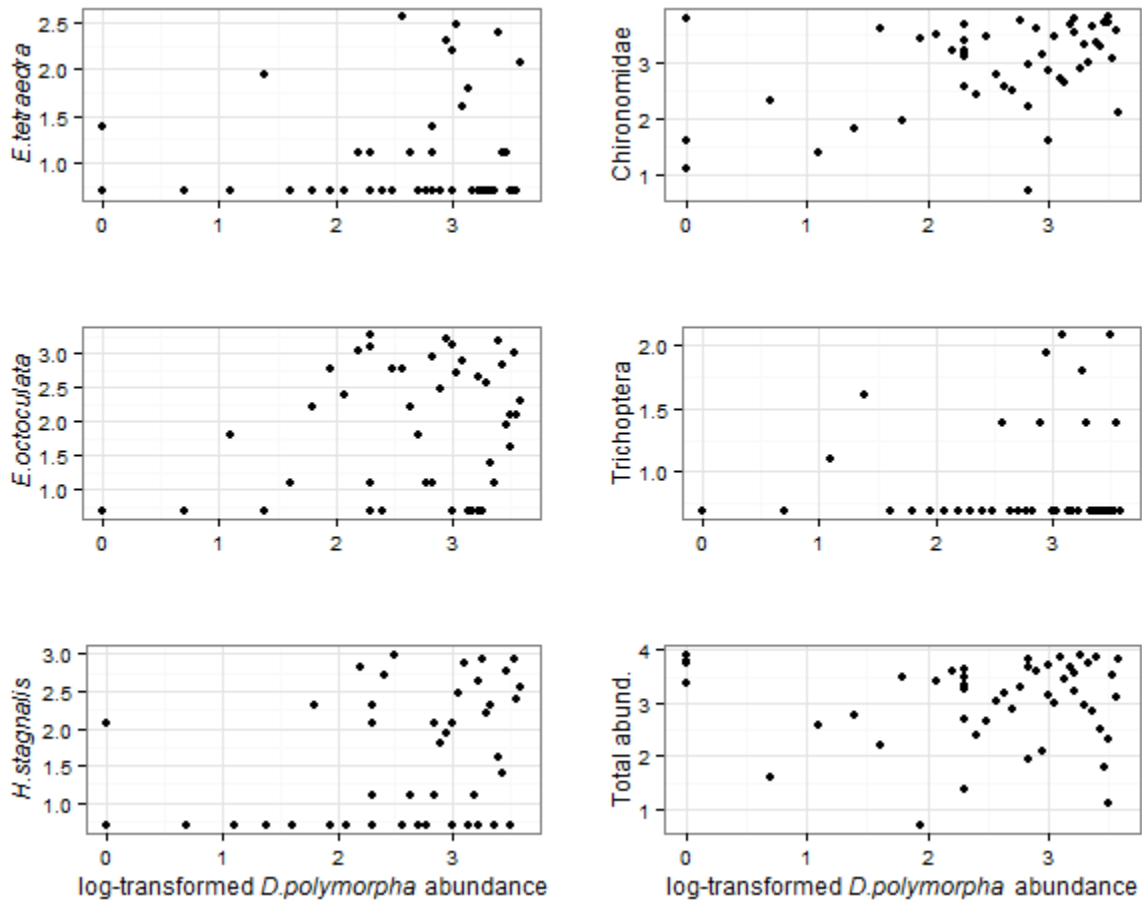
**Figure 2:** Temporal variability of BQI values in the analyzed samples. Gray dots represent samples without zebra mussels, black dots – those with zebra mussels.

The calculated BQI values varied between 0.54 and 2.61 with four apparent outliers in data from 2006 (Fig. 2). The highest ( $>2$ ) obtained BQI values coincided with more than tenfold elevated abundances of zebra mussels ( $1513 \pm 1862$  ind/m<sup>2</sup> versus  $94 \pm 207$  ind/m<sup>2</sup> average in the other samples), *Pisidium sp.* ( $1190 \pm 1425$  ind/m<sup>2</sup> versus  $42 \pm 107$  ind/m<sup>2</sup>), *Valvata spp.* ( $592 \pm 818$  ind/m<sup>2</sup> versus  $42 \pm 160$  ind/m<sup>2</sup>) and Ostracoda ( $4852 \pm 3699$  ind/m<sup>2</sup> versus  $155 \pm 709$  ind/m<sup>2</sup>).

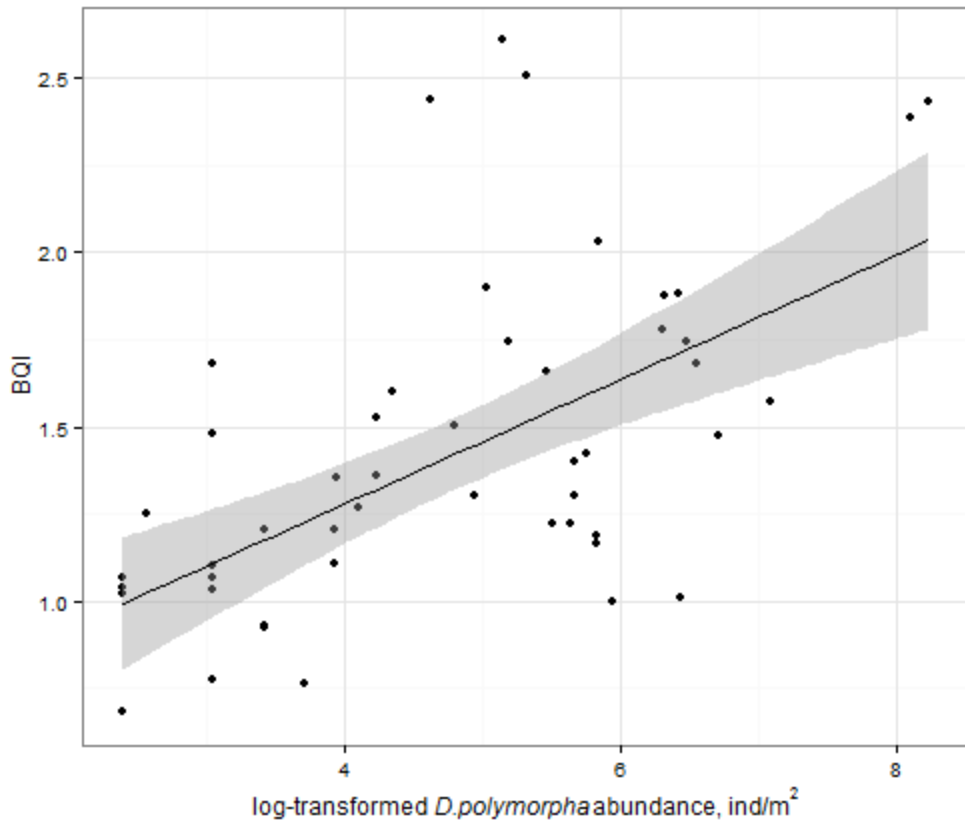
In general, BQI values in samples with zebra mussels were significantly greater ( $W=2548$ ,  $p<0.001$ ) comparing to those devoid of zebra mussels, with no apparent temporal trend (Fig. 2). Additionally, analysis of samples with presence of zebra mussels demonstrated an evident effect of *D. polymorpha* on the total macrofauna abundance (Fig. 3), significantly correlating with *Eiseniella tetraedra* ( $r=0.42$ ,  $p=0.002$ ), *Erpobdella octoculata* ( $r=0.62$ ,  $p<0.001$ ), *Helobdella stagnalis* ( $r=0.61$ ,  $p<0.001$ ), Chironomidae ( $r=0.51$ ,  $p<0.001$ ) and Trichoptera ( $r=0.53$ ,  $p<0.001$ ). On the other hand, only first three species showed significantly higher abundances (Mann-Whitney test,  $p<0.0001$ ) in the presence of the zebra mussels. Positive correlation was also found between the zebra mussel abundance and species richness ( $r=0.43$ ,  $p<0.001$ ). There were 3 species recorded from zebra mussel-free samples only: *Gordius aquaticus*, *Glossiphonia concolor* and non-indigenous gastropod *Potamopyrgus antipodarum*. However due to their low abundances and occurrence below 5%, these species had minor effect on the estimated BQI values. Seven taxa were observed exclusively in samples with zebra mussels: *Asellus aquaticus*, *Caenis macrura*, *Radix auricularia*,



Ceratopogonidae, Corophiidae, Simuliidae, Hemiptera. Consequently, calculated BQI values showed statistically significant correlation with zebra mussel abundance (Fig. 4).

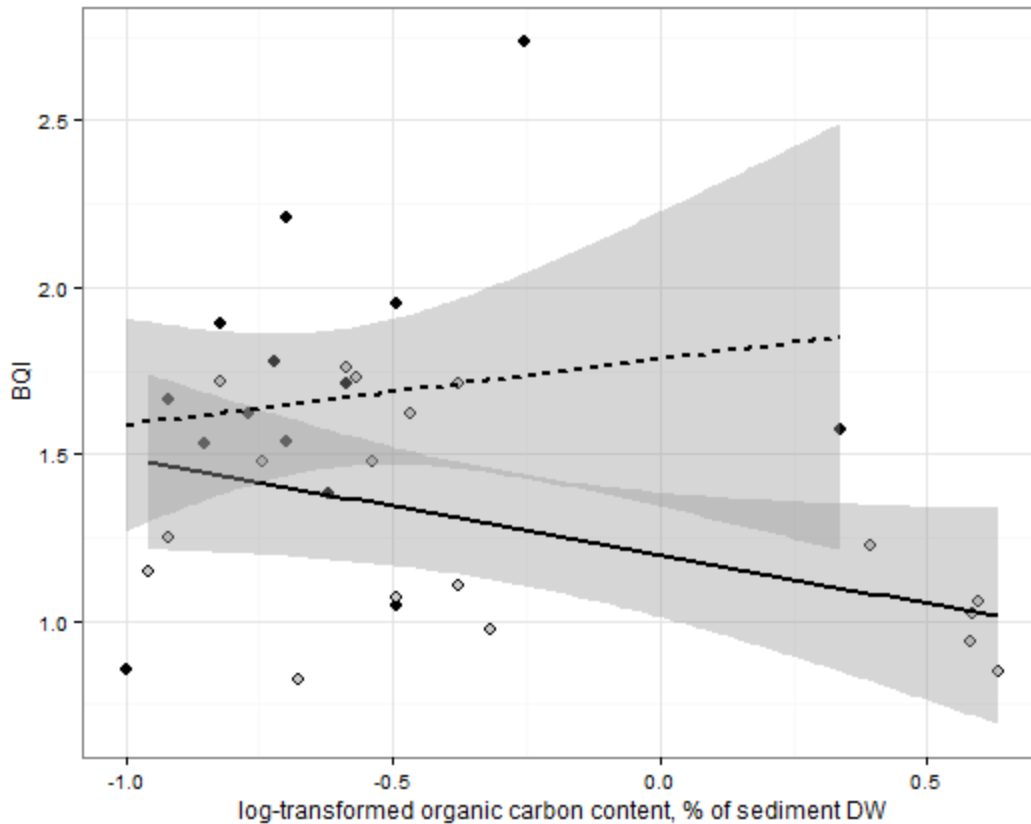


**Figure 3.** Abundances (log-transformed) of *E. tetraedra*, *E. octoculata*, *H. stagnalis*, Chironomidae, Trichoptera and total abundance (ind/m<sup>2</sup>) versus log-transformed *D. polymorpha* abundance (ind/m<sup>2</sup>).



**Figure 4.** BQI values for samples with zebra mussels *versus* *D. polymorpha* abundance (log-transformed) with fitted linear model trendline ( $R^2=0.33$ ,  $r=0.58$ ,  $p<0.001$ ) and standard error represented by shaded area.

When verifying the results on 1999 data (applying the pre-assigned sensitivity values), ANCOVA revealed statistically significant effect of zebra mussel presence ( $F=5.67$ ;  $p=0.02$ ) and marginal effect of organic carbon ( $F=3.63$ ,  $p=0.07$ ) on the BQI values. Moreover, there was a shift from negative to positive regression in the samples with zebra mussels (Fig. 5).



**Figure 5.** BQI values calculated for 1999 dataset based on the pre-assigned species sensitivity values (Table 1). Gray dots – samples without zebra mussel: solid regression line ( $t=12$ ;  $p<0.001$ ;  $BQI= 1.19-0.29x[\log(C\_org)]$ ); black dots – samples with zebra mussel: dashed regression line ( $t=3$ ;  $p= 0.006$ ;  $BQI=1.89+0.36[ \log(C\_org)]$ ); multiple  $R^2=0.32$ .

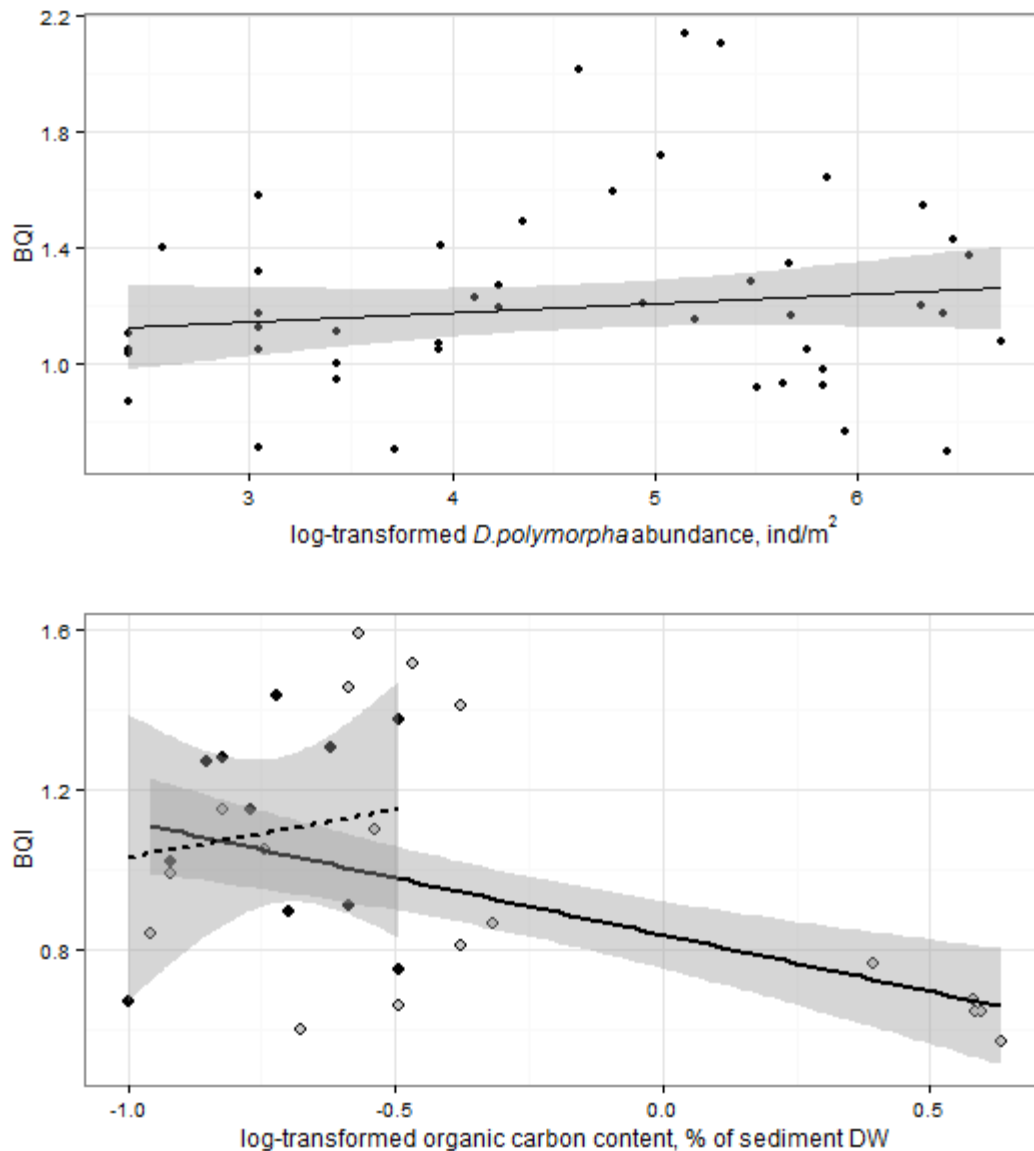
After applying the dataset corrections, new species sensitivity values and their ranking were obtained (Table 2).

**Table 2. Sensitivity values and re-assigned sensitivity classes for the pre-selected 19 taxa after dataset corrections (1 – very tolerant; 2 – tolerant; 3 –sensitive; 4 – very sensitive). An asterisk denotes species with shifted down sensitivity class comparing to the uncorrected analysis.**

Taxa	<i>ES</i> <sub>50-0.05</sub>	Sensitivity class
Oligochaeta	1.4	1
Chironomidae*	1.9	1
<i>Dreissena polymorpha</i> *	2.0	2
<i>Glossiphonia complanata</i> *	2.1	2
Gammaridae	2.1	2
Unionidae	2.1	2
<i>Valvata spp.</i>	2.4	2
Hydracarina	2.5	2

<i>Eiseniella tetraedra</i> *	2.8	2
Ostracoda	2.8	2
<i>Erpobdella octoculata</i> *	2.9	2
<i>Pisidium</i> spp.	3.0	3
<i>Helobdella stagnalis</i>	3.1	3
Trichoptera	3.1	3
Turbellaria	3.1	3
<i>Glossiphonia heteroclita</i> *	3.4	3
<i>Sphaerium</i> spp.	3.5	3
<i>Viviparus viviparus</i>	4.1	4
<i>Bithynia</i> spp.	4.7	4

The BQI values calculated with applied data corrections varied within a narrower range (from 0.54 to 2.13) without any significant correlation with zebra mussel abundances (Fig. 6). When the algorithm was tested on 1999 dataset, an insignificant positive trend of BQI values with enhanced organic carbon content still could be detected in the presence of zebra mussel. However impact of organic carbon content remained as the only important factor (F=6.72, p=0.01) explaining 21% of variance in BQI data.



**Figure 6.** Corrected BQI values for samples with zebra mussels *versus* zebra mussel abundance (log-transformed) with fitted linear model trendline ( $R^2=0.02$ ,  $r=0.14$ ,  $p=0.32$ ) (top) and corrected BQI values for 1999 dataset (bottom). Gray dots – samples without zebra mussel: solid regression line ( $t=11$ ;  $p<0.001$ ;  $BQI=0.88-0.31[\log(C\_org)]$ ); black dots – samples with zebra mussel: dashed regression line ( $t=1$ ;  $p=0.35$ ;  $BQI=0.39+0.24[\log(C\_org)]$ ); multiple  $R^2=0.21$ .

### 3. Discussion

There are at least three scenarios how zebra mussel might compromise the results of benthic quality assessment, if blindly incorporated into the data analysis: (I) by altering species richness; (II) by altering species abundance; (III) by restructuring the whole community in sites highly modified by zebra mussel colonies. Particularly, in the case with BQI calculation (see Eq. 1), all of its components can be potentially affected: number of observed species, abundance of a species, total abundance,  $ES_{50}$  and  $ES_{50-0.05}$  values.

As it was reported from earlier studies, many benthic invertebrates tend to aggregate in habitats modified by zebra mussels (Karatayev et al. 2002, Reed et al. 2004, Zaiko et al. 2009). In soft-bottom environments, zebra mussels provide substrata and shelter for the epifaunal (e.g. *A. aquaticus*, *C. macrura*, *E. octoculata*, *H. stagnalis*, *Radix auricularia*) and infaunal invertebrates (e.g. Chironomidae, Ceratopogonidae, *E. tetraedra*) (Bially and MacIsaac 2000, Minchin and Zaiko 2013). Both detritivorous and carnivorous species benefit from the structural complexity and resources availability (produced biodeposits, sheltered prey items) enhanced by zebra mussel colonies (Zaiko et al. 2009). This explains the fact that a few taxa in our study have demonstrated high level of association with zebra mussel presence and/or abundance.

Except for species significantly correlating with zebra mussel abundance (Fig. 3), there were at least 7 taxa observed exclusively in the presence of zebra mussel. Consequently, the enhanced species richness, total abundances and  $ES_{50}$  values might be affected by zebra mussel presence as well.

On the other hand, there were three species (*G. aquaticus*, *G. concolor* and *P. antipodarum*) observed exclusively devoid of zebra mussels. These species were reported from the west-coast monitoring stations with the prevalence of fine silty mud in sediments (Trimonis et al. 2003), where conditions were highly unfavorable for the zebra mussel population establishment (Fenske et al. 2013). However due to their low abundances and occurrence below 5%, these three species had minor effect on the estimated BQI values.

Based on the species ranking according to their sensitivity (original dataset), *D. polymorpha* hit the third quartile suggesting it as a rather sensitive species. However its high tolerance to variable environmental conditions and different levels of anthropogenic pressure is known from multiple observational and experimental studies worldwide (Claudi and Mackie 1993, Shkorbatov et al. 1994, Fenske et al. 2013). Therefore, we suspect that sensitivity values determined for some other species in this study could be also an artefact of their association with presence or high abundance of zebra mussels. Although for the part of the considered species (e.g. Oligochaeta, Chironomidae, *Bithinia*, *Erpobdella*, *Glossiphonia*, *Sphaerium*) sensitivity scores were consistent with the results reported by other studies in the region (Osowiecki et al. 2008, Kotta et al. 2012, HELCOM 2013), others could be artificially elevated or *vice versa* demoted due to their preference for modified habitats or other particular inter-specific relationships with zebra mussel.

Referring to the results presented, the simplest solution is to eliminate samples with zebra mussel (or any other IAS with strong impact on habitats and communities) from the benthic quality assessment. However, in many invaded ecosystems this would imply exclusion of significant part of the data from the analysis. For instance, in the analyzed dataset from the Curonian Lagoon samples with zebra mussels comprised nearly 50% of the monitoring data and were obtained from 13 locations (out of 15 sampled). Exclusion of these samples would significantly reduce the representativeness of the assessment and robustness of the conclusions.

On the other hand, due to the patchy and non-persistent distribution of zebra mussels (Olenin and Daunys 2004, Zaiko et al. 2009, Zaiko et al. 2014), it is difficult to estimate precisely the probability of finding the species in a particular location. It means that *a posteriori* exclusion of samples from the analysis should be applied, thus affecting the overall cost-effectiveness of the monitoring program. Therefore in this study we have demonstrated a framework of the

dataset correction that proved to be effective enough minimizing the zebra mussel effect on the BQI assessment outcome.

The applied corrections have resulted in a rather logical shift in sensitivity class of 6 species (Table 2). This time zebra mussel pooled within the tolerant species group that matched our general expectations based on expert knowledge. Five other species (*Chironomidae*, *Glossiphonia complanata*, *G.heteroclita*, *Eiseniella tetraedra* and *Erpobdella octoculata*) have been also assigned lower sensitivity class compared to the uncorrected data analysis. The BQI values calculated on the corrected dataset have demonstrated better responsiveness to the considered pressure (eutrophication, expressed by the organic carbon content) with minimized undesired “noise” caused by the presence of invasive ecosystem engineer (Fig. 6). Thus the reliability and overall robustness of the environmental status assessment was improved.

Although data correction framework presented here showed good results in our BQI calculation exercise, the environmental context and ecosystem peculiarities should be considered before applying this approach. Our experience could be applied for other coastal ecosystems invaded by the zebra mussel or any other IAS with similar bioinvasion impacts, after a proper validation and ecosystem-specific adjustments (e.g. for sensitivity values and correlations).

#### **4. Conclusions**

The results of the BQI assessment exercise presented here indicate that several important characteristics of the indicator (including its responsiveness, sensitiveness, predictability, accuracy) could be compromised due to the impact of IAS present in an ecosystem. Zebra mussel ability of modifying benthic habitats and forming local patches of elevated biological diversity may bias the results of benthic quality assessment by showing false improvement of ecological status. If not considered in the course of the assessment, any species richness-based index may reflect IAS impact rather than anthropogenic pressure effect. Proper adjustments of ecological status assessment are desirable for the ecosystems strongly affected by IAS.

#### **Acknowledgements**

The current study was supported by DEVOTES (DEVELOPMENT OF innovative TOOLS for understanding marine biodiversity and assessing Good Environmental Status) project funded by the European Union under the 7th Framework Programme, ‘The Ocean of Tomorrow’ Theme (Grant Agreement No. 308392), [www.devotes-project.eu](http://www.devotes-project.eu) and by the BIO-C3 (Biodiversity changed investigating causes, consequences and management implications) within the BONUS, the joint Baltic Sea Research and Development programme, funded jointly by the EU 7th Framework Programme and Research Council of Lithuania (Grant Agreement No. BONUS-1/2014). We thank Lithuanian Environmental Protection Agency, Marine Research Department for the monitoring datasets provided and Ingrida Bagdanavičiūtė for the generated map of the study area. We also thank two anonymous reviewers for the valuable comments on the manuscript.

## References

- Aleksandrov, S. V. 2010. Biological production and eutrophication of Baltic Sea estuarine ecosystems: The Curonian and Vistula Lagoons. *Marine Pollution Bulletin* 61:205-210.
- Atalah, J., M. Kelly-Quinn, K. Irvine, and T. P. Crowe. 2010. Impacts of invasion by *Dreissena polymorpha* (Pallas, 1771) on the performance of macroinvertebrate assessment tools for eutrophication pressure in lakes. *Hydrobiologia* 654:237-251.
- Bially, A., H.J. MacIsaac. 2000. Fouling mussels (*Dreissena* spp.) colonize soft sediments in Lake Erie and facilitate benthic invertebrates. *Freshwater Biology* 43:85-97.
- Borja, A., M. Elliott, J. H. Andersen, A. C. Cardoso, J. Carstensen, J. G. Ferreira, A.-S. Heiskanen, J. C. Marques, J. M. Neto, H. Teixeira, L. Uusitalo, M. C. Uyarra, and N. Zampoukas. 2013. Good Environmental Status of marine ecosystems: What is it and how do we know when we have attained it? *Marine Pollution Bulletin* 76:16-27.
- Borja, A., M. Elliott, J. Carstensen, A.-S. Heiskanen, and W. van de Bund. 2010. Marine management - towards an integrated implementation of the European Marine Strategy Framework and the Water Framework Directives. *Marine Pollution Bulletin* 60:2175-2186.
- Borja, A., J. Franco, and V. Perez. 2000. A Marine Biotic Index to Establish the Ecological Quality of Soft-Bottom Benthos Within European Estuarine and Coastal Environments. *Marine Pollution Bulletin* 40:1100-1114.
- Borja, A., I. Muxika and J. Franco. 2003. The application of a Marine Biotic Index to different impact source affecting soft-bottom benthic communities along European coasts. *Marine Pollution Bulletin* 46: 835–845
- Clarke, K. R., and R. M. Warwick. 2001. Change in marine communities: an approach to statistical analysis and interpretation, 2nd edition edition. PRIMER-E, Plymouth, UK.
- Claudi, R., and G. L. Mackie. 1993. Practical manual for zebra mussel monitoring and control. CRS Press.
- Dauvin, J.-C., and T. Ruellet. 2007. Polychaete/amphipod ratio revisited. *Marine Pollution Bulletin* 55:215-224.
- Diaz, R. J., M. Solan, and R. M. Valente. 2004. A review of approaches for classifying benthic habitats and evaluating habitat quality. *Journal of Environmental Management* 73:165-181.
- Elliott, M. 2003. Biological pollutants and biological pollution - an increasing cause for concern. *Marine Pollution Bulletin* 46:275-280.
- Elliott, M. 2011. Marine science and management means tackling exogenic unmanaged pressures and endogenic managed pressures – a numbered guide. *Marine Pollution Bulletin* 62:651-655.
- Fenske, C., A. Zaiko, A. Wozniczka, S. Dahlke, and M. I. Orlova. 2013. Variation in length–frequency distributions of zebra mussels (*Dreissena polymorpha*) within and between three Baltic Sea subregions Szczecin Lagoon, Curonian Lagoon, and Gulf of Finland. Pages 725-740 in T. F. Nalepa and D. W. Schlosser, editors. *Quagga and zebra mussels: Biology, Impacts and Control*. CRC Press Taylor & Francis Group, Boca Raton, London, New York.
- Fleischer, D., A. Gremare, C. Labrune, H. Rumohr, E. V. Berghe, and M. L. Zettler. 2007. Performance comparison of two biotic indices measuring the ecological status of water bodies in the Southern Baltic and Gulf of Lions. *Marine Pollution Bulletin* 54:1598-1606.



- Fleischer, D., and M. L. Zettler. 2009. An adjustment of benthic ecological quality assessment to effects of salinity. *Marine Pollution Bulletin* 58:351-357.
- Gasiunaite, Z. R., D. Daunys, S. Olenin, and A. Razinkovas. 2008. The Curonian Lagoon. Pages 197-215 in U. Schiewer, editor. *Ecology of Baltic coastal waters*. Springer-Verlag, Berlin, Heidelberg.
- HELCOM. 1988. Guidelines for the Baltic monitoring programme for the third stage.
- HELCOM. 2013. HELCOM Core Indicator Report: State of the soft-bottom macrofauna communities. [www.helcom.fi](http://www.helcom.fi).
- Labrune, C., J.M. Amouroux, R. Sarda, E. Dutrieux, S. Thorin, R. Rosenberg, A. Gremare. 2006. Characterization of the ecological quality of the coastal Gulf of Lions (NW Mediterranean). A comparative approach based on three biotic indices. *Marine Pollution Bulletin* 52: 34-47.
- Karatayev, A. Y., L. E. Burlakova, and D. K. Padilla. 2002. Impacts of zebra mussels on aquatic communities and their role as ecosystem engineers. Pages 433-446 in E. Leppakoski, S. Gollasch, and S. Olenin, editors. *Invasive aquatic species of Europe - distribution, impact and management*. Kluwer, Dordrecht.
- Kennedy, R., A. Wallace, and B. F. Keegan. 2011. Long-term trends in benthic habitat quality as determined by Multivariate AMBI and Infaunal Quality Index in relation to natural variability A case study in Kinsale Harbour, south coast of; Ireland. *Marine Pollution Bulletin* 62:1427-1436.
- Kerans, B. L., and J. R. Karr. 1994. A Benthic Index of Biotic Integrity (B-IBI) for Rivers of the Tennessee Valley. *Ecological Applications* 4:768-785.
- Kershner, J., J. F. Samhouri, C. A. James, and P. S. Levin. 2011. Selecting indicator portfolios for marine species and food webs: a puget sound case study. *PLoS One* 6: e25248.
- Kotta, J., V. Lauringson, A. Kaasik, and I. Kotta. 2012. Defining the coastal water quality in Estonia based on benthic invertebrate communities. *Estonian Journal of Ecology* 61:86-105.
- Leonardsson, K., M. Blomqvist, and R. Rosenberg. 2009. Theoretical and practical aspects on benthic quality assessment according to the EU-Water Framework Directive "Examples from Swedish waters. *Marine Pollution Bulletin* 58:1286-1296.
- Maurer, D., H. Nguyen, G. Robertson, and T. Gerlinger. 1998. The infaunal trophic index (ITI): its suitability for marine environmental monitoring. *Ecological Applications* 9:699-713.
- Mee, L. D., R. L. Jefferson, D. A. Laffoley, and M. Elliott. 2008. How good is good? Human values and Europe's proposed Marine Strategy Directive. *Marine Pollution Bulletin* 56:187-204.
- Minchin, D., F. Lucy, and M. Sullivan. 2002. Zebra mussel: impacts and spread. Pages 135-146 in E. Leppakoski, S. Gollasch, and S. Olenin, editors. *Invasive aquatic species of Europe - distribution, impact and management*. Kluwer, Dordrecht.
- Minchin, D., and A. Zaiko. 2013. Variability of the zebra mussel (*Dreissena polymorpha*) impacts in the Shannon River system. Pages 587-597 in T. F. Nalepa and D. W. Schlosser, editors. *Quagga and zebra mussels: Biology, Impacts and Control*. CRC Press Taylor & Francis Group, Boca Raton, London, New York.
- Niemeijer, D., and R. S. de Groot. 2008. A conceptual framework for selecting environmental indicator sets. *Ecological Indicators* 8:14-25.

- Olenin, S., and D. Daunys. 2004. Coastal typology based on benthic biotope and community data: the Lithuanian case study. Pages 65-83 in G. Schernewski and M. Wielgat, editors. Baltic Sea Typology. Coastline Reports.
- Olenin, S., M. Elliott, I. Bysveen, P. F. Culverhouse, D. Daunys, G. B. J. Dubelaar, S. Gollasch, P. Gouletquer, A. Jelmert, Y. Kantor, K. Bringsvor Mezeth, D. Minchin, A. Occhipinti-Ambrogi, I. Olenina, and J. Vandekerckhove. 2011. Recommendations on methods for the detection and control of biological pollution in marine coastal waters. *Marine Pollution Bulletin* 62:2598-2604.
- Olenin, S., D. Minchin, and D. Daunys. 2007. Assessment of biopollution in aquatic ecosystems. *Marine Pollution Bulletin* 55:379-394.
- Olenin, S., M. I. Orlova, and D. Minchin. 1999. *Dreissena polymorpha* (Pallas, 1771). Pages 37-42 in S. Gollasch, D. Minchin, H. Rosenthal, and M. Voigt, editors. Case histories on introduced species: their general biology, distribution, range expansion and impact. Logos-Verlag, Berlin.
- Olenina, I., and S. Olenin. 2002. Environmental Problems of the South-Eastern Baltic Coast and the Curonian Lagoon. Pages 149-156 in G. Schernewski and U. Schiewer, editors. *Baltic Coastal Ecosystems*. Springer, Berlin Heidelberg.
- Osowiecki, A., E. Lysiak-Pastuszak, and Z. Piatkowska. 2008. Testing biotic indices for marine zoobenthos quality assessment in the Polish sector of the Baltic Sea. *Journal of Marine Systems* 74:S124-S132.
- Pearson, T. H., and R. Rosenberg. 1978. Macrobenthic succession in relation to organic enrichment and pollution of the marine environment. *Oceanography and Marine Biology. An Annual Review*. 16:229-311.
- Puente, A., and R.J. Diaz. 2008. Is it possible to assess the ecological status of highly stressed natural estuarine environments using macroinvertebrates indices? *Marine Pollution Bulletin* 56:1880-1889.
- Reed, T., S. J. Wielgus, A. K. Barnes, J. J. Schiefelbein, and A. L. Fettes. 2004. Refugia and local controls: benthic invertebrate dynamics in lower Green Bay, Lake Michigan following zebra mussel invasion. *Journal of Great Lakes Research* 30:390-396.
- Reise, K., S. Olenin, and D. W. Thieltges. 2006. Are aliens threatening European aquatic coastal ecosystems? *Helgoland Marine Research* 60:77-83.
- Ricciardi, A., F. G. Whoriskey, and J. B. Rasmussen. 1997. The role of the zebra mussel (*Dreissena polymorpha*) in structuring macroinvertebrate communities on hard substrata. *Canadian Journal of Fisheries and Aquatic Sciences* 54:2596-2608.
- Rice, J. 2003. Environmental health indicators. *Ocean & Coastal Management* 46:235-259.
- Rice, J. C., and M.-J. Rochet. 2005. A framework for selecting a suite of indicators for fisheries management. *ICES Journal of Marine Science* 62:516-527.
- Rosenberg, R., M. Blomquist, H. C. Nilsson, H. Cederwall, and A. Dimming. 2004. Marine quality assessment by use of benthic species-abundance distributions: a proposed new protocol within the European Union Water Framework Directive. *Marine Pollution Bulletin* 49:12.
- R-project. 2014. R: A language and environment for statistical computing. in. R Foundation for Statistical Computing, Vienna, Austria, [ISBN 3-900051-07-0], <http://www.R-project.org>.

- Shkorbatov, G. L., A. F. Karpevich, and A. P. I. 1994. Ecological physiology. Pages 67-108 in J. I. Starobogatov, editor. Freshwater zebra mussel *Dreissena polymorpha* (Pall.) (Bivalvia, Dreissenidae): systematics, ecology, practical Meaning. Nauka Press, Moscow.
- Stewart, T. W., J. G. Miner, and R. L. Lowe. 1998. Quantifying mechanisms for zebra mussel effects on benthic macroinvertebrates: organic matter production and shell-generated habitat. *Journal of North American Benthological Society* 17:81-94.
- Strayer, D. L., L. C. Smith, and D. C. Hunter. 1998. Effects of the zebra mussel (*Dreissena polymorpha*) invasion on the macrobenthos of the freshwater tidal Hudson River. *Canadian Journal of Zoology* 76:419-425.
- Tett, P., R. Gowen, S. Painting, M. Elliott, R. Forster, D. Mills, E. Bresnan, E. Capuzzo, T. Fernandes, J. Foden, R. Geider, L. Gilpin, M. Huxham, A. McQuatters-Gollop, S. Malcolm, S. Saux-Picart, T. Platt, M. Racault, S. Sathyendranath, J. van der Molen, and M. Wilkinson. 2013. Framework for understanding marine ecosystem health. *Marine Ecology Progress Series* 494:1-27.
- Thayer, S. A., R. C. Haas, R. D. Hunter, and R. H. Hushler. 1997. Zebra mussel (*Dreissena polymorpha*) effects on sediment, other zoobenthos, and the diet and growth of adult yellow perch (*Perca flavescens*) in pond enclosures. *Canadian Journal of Fisheries and Aquatic Sciences* 54:1903-1915.
- Trimonis, E., S. Gulbinskas, and M. Kuzavinis. 2003. The Curonian Lagoon bottom sediments in the Lithuanian water area. *Baltica* 16:13-20.
- Villnas, A., and A. Norkko. 2011. Benthic diversity gradients and shifting baselines: implications for assessing environmental status. *Ecological Applications* 21:2172-2186.
- Zaiko, A., D. Daunys, and S. Olenin. 2009. Habitat engineering by the invasive zebra mussel *Dreissena polymorpha* (Pallas) in a boreal coastal lagoon: impact on biodiversity. *Helgoland Marine Research* 63:85-94.
- Zaiko, A., M. Lehtiniemi, A. Narscius, and S. Olenin. 2011. Assessment of bioinvasion impacts on a regional scale: a comparative approach. *Biological Invasions* 13:1739-1765.
- Zaiko, A., D. Minchin, and S. Olenin. 2014. "The day after tomorrow": anatomy of an 'r' strategist aquatic invasion. *Aquatic Invasions* 9:145-155.
- Zaiko, A., S. Olenin, D. Daunys, and T. F. Nalepa. 2007. Vulnerability of benthic habitats to the aquatic invasive species. *Biological Invasions* 9:703-714.
- Zaiko, A., R. Paskauskas, and A. Krevs. 2010. Biogeochemical alteration of benthic environment by zebra mussel *Dreissena polymorpha* (Pallas). *Oceanologia* 52:649-667.
- Zaromskis, R. 1996. Oceans, seas, estuaries. Debesija, Vilnius.
- Zettler, M. L., and D. Daunys. 2007. Long-term macrozoobenthos changes in a shallow boreal lagoon: comparison of a recent biodiversity inventory with historical data. *Limnologica* 37:170-185.