

Summary

The rapid development of the economy, the intensive exploitation of environmental resources and the increasing anthropopressure associated with these phenomena force us to think about future generations. Direct threatening effects of human activity on the marine environment include pollution by undesirable chemicals, increased water temperature associated with climate change, eutrophication, invasions by alien species or overfishing due to overexploitation of populations relative to their self-reproducing capacity (HELCOM 2018; Jakubowska et al. 2002). Close observation of the surrounding environment as well as careful analysis of the impact of anthropogenic factors are important elements in protecting the world around us. The impact of human activity on the environment can be monitored using international guidelines, such as those contained in the Water Framework Directive (2000/60/EC), through the analysis of biological, hydrological or physico-chemical elements. However, for this purpose, it is the best to use indicators based on whole-community studies, as only a sufficiently rich grouping of species with diverse ecological requirements can provide complete information on the effects of factors related to anthropopressure (Pennesi and Danovaro 2017). Hence, the present study undertook to investigate and describe changes in microphytobenthic communities caused by selected factors associated with human activities.

Microphytobenthos in this study is broadly defined as a formation that includes plant microorganisms associated with the seabed or various types of substrate found in the water (Cahoon 2019). It is part of a microecosystem in which, as in any ecosystem, there are interactions between organisms and their environment (Bosserman 1983). It is functionally particularly important, especially for ecosystems of coastal zones, estuaries or shallow seas, as it contains organisms that are important producers and an important part of the trophic chain. Organisms forming microphytobenthic communities are considered reliable indicators of environmental change due to their rapid response to both abiotic and biotic factors (Potapova and Charles 2007). Their use in both monitoring and ecotoxicological studies has a rich and long history dating back to the 19th century (e.g., Dickman 1969; Blanck 1985; Schmitt-Jansen and Altenburger 2005; Schmitt-Jansen and Altenburger 2005; Roubex et al. 2011; Zhu et al. 2021). Factors of natural or artificial origin not only limit the growth and development of plant microorganisms, but also form the structure and functioning of entire communities. A major advantage of studies using microphytobenthic communities is the simplicity of obtaining data from the environment (Dahl and Blanck 1996; Blanck et al. 2009; Sylwestrzak et al. 2014b,

PAPERS 1, 2, 3). In addition, the literature on research conducted on microphytobenthos constitutes a valuable source of information for practical applications of this formation (e.g., Dahl and Blanck 1996; Underwood et al. 1999, Cohn and McGuire 2000; Underwood et al. 2004; De La Iglesia et al. 2013; Vannoni et al. 2022). In recent years, it has been a significant increase in interest on the use of microphytobenthic communities in ecotoxicological studies testing, among other things, of potentially toxic substances of anthropogenic origin (e.g., Blanck et al. 2009; Araújo et al. 2010; Åsa Arrhenius et al. 2014; Bácsi et al. 2016; Pennesi and Danovaro 2017; Du et al. 2021; Vannoni et al. 2022). For example, experiments have been conducted on marine microphytobenthic communities using antifouling substances and paints (Blanck et al. 2009; Arrhenius et al. 2014), medicines (Pérez et al. 2009), cosmetics (Mason et al. 1996), or herbicide products (Downing et al. 2004). Among other things, these scientific studies contributed to the planning of the dissertation research, in which the bioindication potential of entire communities was used. A completely novelty was the use of communities obtained directly from the environment, interesting results concerning the response of the communities to substances of anthropogenic origin.

The research underlying this dissertation is based on the on an analysis of the impact of factors associated with two types of anthropogenic pressure - pollution caused by the introduction of chemical substances into the environment and temperature increases caused by climate change. To analyse the response of microphytobenthic communities to chemical pollution, three substances from various chemical groups with different modes of action and varying degrees of recognition were selected. One of the substances used was copper (II) chloride (**PAPER 1**). Copper is functionally important for aquatic plant microorganisms and its action mechanism is relatively well recognised (Stauber and Florence 1987; Manimaran et al. 2012; Serwatka et al. 2015; Li et al. 2021). It is a component of many proteins and enzymes involved in metabolic reactions, hence it plays an important role in the metabolism of photosynthetic organisms (Morelli and Scarano 2004). However, in excess, it can interfere with physiological processes. At high concentrations and prolonged exposure, copper ions slow down photosynthesis (Fernandes and Henriques 1991; Guasch et al. 2002) generating oxidative stress by inducing the production of reactive oxygen forms (Morelli and Scarano 2004) and affecting metabolic processes also related to growth (Maksymiec and Krupa 2006) and biochemical composition, including carotenoids, proteins, lipids and carbohydrates (Neethu et al. 2021). Copper ions are used as one of the ingredients in antifouling paints because they negatively affect the condition of microalgae (Traon et al. 2021).

Another substance used was glyphosate in the form of Roundup® (**PAPER 2**). Glyphosate is an organic compound from the phosphonate group, which is a broad-spectrum biological active substance used in many herbicides. Herbicides include many excipients in addition to glyphosate as the active substance. Roundup®, due to its complex composition and rapid degradation rate, can be a source of carbon and nitrogen, and its low concentrations can stimulate microalgal cell growth (Malik et al. 1989; Wong 2000; Berman et al. 2020). A long-term study of pesticide content in surface waters has shown that the most common pesticides are bentazone and glyphosate (Stenström et al. 2021). Glyphosate as a pesticide is used on an increasing scale due to the massive development of agricultural production, the high yield of herbicide formulations containing this substance, their low production cost and the still liberal laws in many countries with highly developed agricultural economies (Brovini et al. 2021). Once in plant cells, this compound inhibits, for example, the production of the enzyme EPSP (5-enolpyruvylshikimate-3-phosphate) synthase, which slows down the organism's formation of aromatic amino acids important for growth and included in the composition of many plant pigments (Franz et al. 1997). Reduced or absent of photosynthetic pigments lead to damage to chloroplast structure and cell degradation (e.g., Sylwestrzak et al. 2015; Kim and Ponomarev 2021).

The last substance was 1-Butyl-3-methyl-imidazolium-chloride [BMIM]Cl, which chemically belongs to the group of ionic liquids (**PAPER 3**). Ionic liquids are substances that are gaining popularity due to their potentially desirable properties such as non-flammability, high thermal and electrochemical stability, low vapour pressure, good conductivity and excellent catalytic properties (Mai et al. 2014; Chen et al. 2020). The properties listed above correspond to the requirements for the products of so-called 'green chemistry'. However, after years of research, the 'green' nature of ionic liquids has been questioned, although the mechanism of the toxic effects of these compounds is still not well understood (Nikitenko et al. 2007; Freire et al. 2010; Kumar et al. 2011; de Jesus and Filho 2022; Maculewicz et al. 2022).

Among the factors related to climate change, the study investigated the impact of a short-term increase in water temperature on the organisms that form microphytobenthic communities (**PAPER 4**). As has been shown, it is not only the steadily increasing temperature that has a huge impact on the environment, but also the frequency and intensity of extreme climatic events such as heat waves (Vieira et al. 2013). Based on studies conducted during the last few years, it has been shown that in summer on the southern Baltic Sea, average coastal

water temperatures are around 19° C, but for short intervals these values rise above 23° C (Siegel et al. 2006; Bradtke et al. 2010; Rak and Wieczorek 2012; Stramska and Białogrodzka 2015). Results from earlier studies suggest that microphytobenthic communities can undergo major changes due to temperature increase. Results from work conducted on diatom-dominated microphytobenthic communities indicate that short-term increases in temperature stimulate photosynthesis, while long-term exposure to higher temperatures leads to a reduction in photosynthetic productivity and to changes in species composition as well as intense cyanobacterial growth (Hicks et al. 2011; Vieira et al. 2013; Cartaxana et al. 2015; Kazanjian et al. 2018).

All factors described above have a profound impact on the functioning of plant micro-organisms, hence their use in testing the response of microphytobenthic communities has adequate justification. It is worth noting that marine organisms associated with brackish environments have so far rarely been used in ecotoxicological tests. The vast majority of such tests are conducted on algal strains, e.g. tests recommended by the OECD (OECD 1984; OECD 2006) or the International Organization for Standardization (ISO 2012). Tests conducted according to standardised methods on selected monocultures are extremely valuable as they allow comparison of the degree of influence of different substances, but give information on the reaction of a few, selected organisms only. Furthermore, many of the species recommended for toxicological testing are maintained for a long time under artificial laboratory conditions to which they adapt, e.g.: *Chlorella vulgaris* Beijerinck (Beijerinck) is maintained in monoculture since 1889 (Beijerinck 1890), *Navicula pelliculosa* (Kützing) Hilse since 1955 (Lewin 1955), and *Selenastrum capricornutum* Printz since 1959 (Guiry 2013). Analysis of the impact of anthropogenic factors on natural microphytobenthic communities is a relatively new and innovative approach used in ecotoxicological testing. Although such studies have been conducted for more than 20 years and the literature is rich, a standard method for conducting these tests has still not been developed and the measurement techniques used during the studies vary (Cibic et al. 2008; Duong et al. 2008; Morin et al. 2017). Within the scope of the doctoral dissertation, a number of tools and methods for conducting ecotoxicological tests on microphytobenthic communities were checked, which undoubtedly represents an interesting contribution to the development of this field of science (Dahl and Blanck 1996; Perkins et al. 2001; Serôdio 2004; Araújo et al. 2010; Arrhenius et al. 2014).

The main objective of the dissertation was to characterise the response of the Baltic Sea microphytobenthic communities to factors described as of anthropogenic origin - substances

from different chemical groups, i.e., copper (II) chloride, glyphosate (in the form of Roundup®) and the ionic liquid [BMIM]Cl, as well as for a short-term rapid increase in water temperature.

The study assumes the following research hypothesis:

The introduction of contaminants into the environment in the form of copper (II) chloride, glyphosate (as Roundup®), the ionic liquid [BMIM]Cl and a short-term rapid temperature increase affect the composition and structure of the microphytobenthic communities of the Gulf of Gdańsk, and these changes can be estimated by observing the cells of microorganisms included in microphytobenthos communities.

Conducting the experiments forming the basis of the doctoral dissertation required a number of preliminary studies that allowed the development of the most appropriate methodology for assessing changes in the microphytobenthic communities under the influence of selected factors. The identification of the research material, i.e. the composition and structure of the Baltic microphytobenthos, took place within the scope of Z. Sylwestrzak's master's thesis entitled 'Succession of microphytobenthic communities in the Gulf of Gdańsk (experimental studies)', as well as on the basis of extensive literature of the subject. The experiments conducted in the initial stages of the doctoral dissertation were aimed at testing, among other things:

- the optimal exposure time in the environment of culture slides from which organisms forming microphytobenthic communities were obtained (Sylwestrzak, Zgrundo and Pniewski 2014);
- the influence of the culture medium (seawater plus standard f/2 medium) on the results of ecotoxicological tests conducted on microphytobenthic communities (Sylwestrzak and Pniewski 2014; Sylwestrzak and Zgrundo 2014).

Due to the fact that commonly used methods for assessing the condition of microorganisms, e.g. photosynthetic pigment concentration or photosynthetic activity (PAM fluorescence) (Brotas et al. 2007; Morelle et al. 2018) often did not provide unambiguous results, other less common indicators were tested during the initial study, such as:

- survival rate of representatives of individual microphytobenthos taxa, determined as the ratio of live to dead cells (Sylwestrzak and Zgrundo 2014),

- cell condition of microphytobenthos representatives in relation to the state of chloroplasts. The condition was determined on the basis of changes in the shape and structure of chloroplasts according to previously developed research methodology (Sylwestrzak 2014; Sylwestrzak et al. 2015; Sylwestrzak and Pniewski 2014). Examples and microscopic photographs of cells with correctly formed chloroplasts and abnormally shaped chloroplasts are shown on Fig. 1.

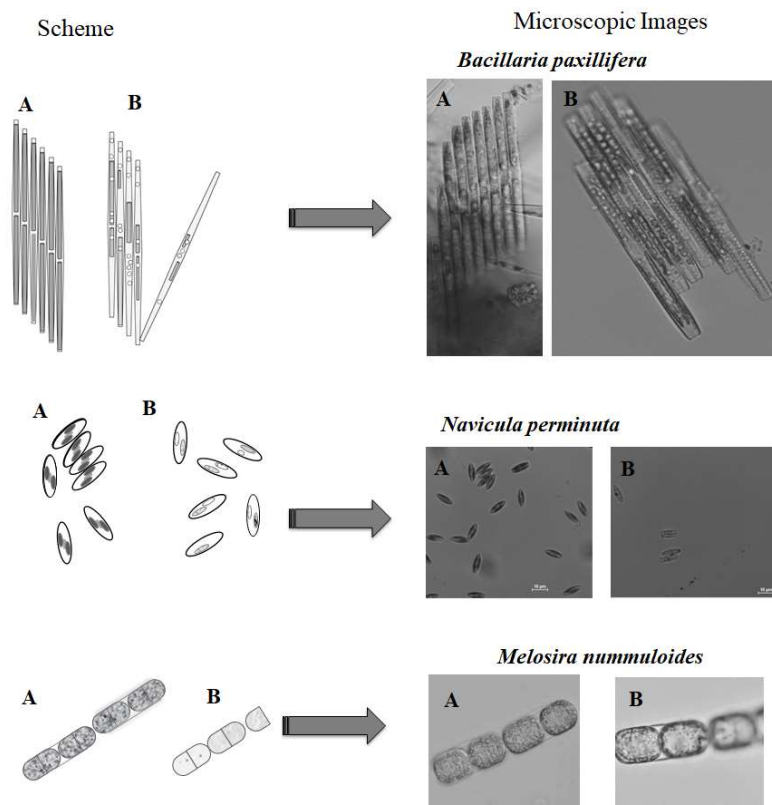


Fig. 1. Examples of cells with correctly formed chloroplasts (A) and abnormally shaped chloroplasts (B) for *Bacillaria paxillifera*, *Navicula perminuta*, *Melosira nummuloides* illustrated by schemes and photographs of the cells made using an optic microscope.

The material used in the targeted tests consisted of microphytobenthic communities obtained from culture slides exposed in the coastal zone of the Gulf of Gdańsk (southern Baltic Sea) for a period of 14 days in July and August 2015. During the exposure of the culture slides, the water temperature oscillated between 17 and 19 °C and the salinity varied between 7.9 and 8.4 PSU. The methodology of the fieldwork is described in detail in the studies comprising this

dissertation (**PAPERS 1, 2, 3 and 4**). After transporting the culture panels to the laboratory, the microphytobenthic communities were scraped from the culture slides using a scalpel, saturated with nitrogen gas to induce hypoxia and sonified. By using the above procedures, animal organisms were eliminated from the microbenthic solution and the test material was standardised (Rosenberg et al. 1991). Microphytobenthos were then placed in 250 ml flasks in 100 ml of filtered seawater collected *in situ*. Irrespective of the experiment, the average community abundance at the start of the tests was 41319 (\pm 1133), and 95% of the total community were diatoms. The initial concentrations of biogenic compounds in the seawater were: 9.4 mg·m⁻³ N-NH₄, 102 mg·m⁻³ N-NO₃, 36 mg·m⁻³ P-PO₄, 600 mg·m⁻³ Si-SiO₄. During the preliminary studies a biogenic nutrient analysis was carried out before and after the tests. Based on the data obtained, it was shown that during experiments conducted for 7 days, biogenic nutrients are not depleted and do not limit the growth of microalgae (Sylwestrzak 2016). To identify the response of the microphytobenthic communities to the selected chemicals (**PAPERS 1, 2, 3**) concentrations of compounds were used, established based on concentration values that are defined as threatening to the environment (based on standards, e.g. Dz.U.2011.257.1545 or the literature on the subject, i.e. Kulacki and Lamberti 2008; Liu et al. 2015; Skeff et al. 2015) and values whose real effects on plant microorganisms were observed during previous preliminary studies (Sylwestrzak 2012; Sylwestrzak et al. 2014; Sylwestrzak and Zgrundo 2014; Serwatka et al. 2015; Sylwestrzak et al. 2015). The following concentrations were used during the tests: for copper (II) chloride - $2 \cdot 10^{-5}$ g·dm⁻³ and $2 \cdot 10^{-3}$ g·dm⁻³, for glyphosate - 0.042 g·dm⁻³, 0.85 g·dm⁻³ and 8.5 g·dm⁻³, and for the ionic liquid [BMIM]Cl - $1.13 \cdot 10^{-3}$ g·dm⁻³ and $1.75 \cdot 10^{-2}$ g·dm⁻³. In the study presented in **PAPER 4**, the range of temperatures used was selected following measurements carried out in the southern Baltic in recent years (<http://www.satbaltyk.pl>) and based on publications describing the phenomenon of short-term temperature increases, which can have a strong influence on community composition and structure (Siegel et al. 2006; Bradtke et al. 2010; Rak and Wieczorek 2012; Stramska and Białogrodzka 2015).

Each variant of the experiments described in **PAPERS 1, 2, 3 and 4** was performed three times, and methods of observing the response of microphytobenthic communities to a factor of anthropogenic origin used in **PAPERS 1, 2 and 3** were different from those used in **PAPER 4**. The material spent to analyse the effect of factors considered to be chemical pollutants was microphytobenthic communities preserved with Lugol's fluid, and observations were made for samples collected after three and seven days (**PAPERS 1 and 2, 3**). Qualitative

and quantitative community analysis and chloroplast condition analysis was performed in 50 fields of view in Utermöhl Sedimentation Chambers (2 ml) under a Nikon Eclipse TS100 Inverted Microscope at magnifications of x200, x400. The condition of the cells was determined based on the state of the chloroplasts, grouping them into two categories: cells with normal-shaped chloroplasts (i.e., matching the shapes presented in the publications as typical and normal for the taxa in question) and cells with abnormal-shaped chloroplasts (showing deviations from those presented in the literature). The results of the experiments were presented as the number of all organisms and representatives of each taxon in relation to the number observed in the control solution, according to the principles adopted in the OECD guidelines used to assess the toxicity effects of chemical compounds on plant microorganisms (OECD 2006).

An analysis of the effect of a short-term temperature increase (**PAPER 4**) was carried out on material collected as in **PAPERS 1, 2 and 3**, but using a special laboratory treatment to obtain pure diatom material, which involves the removal of the remaining organisms forming the microphytobenthos. An analysis of the effect of a short-term temperature increase (**PAPER 4**) was carried out on material collected as in **PAPERS 1, 2 and 3**, but using a special laboratory treatment to obtain pure diatom material, which involves the removal of the remaining organisms forming the microphytobenthos. The observation was performed on permanent slides made with Naphrax resin of refractive index ~ 1.7 , using a Nikon 80i Fluorescence Microscope equipped with a DS-U2 camera at x1000 magnification. In this case, up to 300 diatom clades were identified and counted. Other parameters such as photosynthetic activity (determined by changes in chlorophyll fluorescence measured using a Fluorescence Monitoring System (FMS1; Hansatech, Norfolk, UK)), and photosynthetic pigment concentration (qualitative and quantitative analysis of pigments was measured using high-performance liquid chromatography (HPLC) using the Waters system equipped with two Waters 515 pumps and the Waters 2998 Photodiode Array Detector). were also analysed during the test connected with short-term temperature increase. The detailed methodology of the analyses performed is presented in the publication. For the necessary thematic consistency of this dissertation, the results related to the response of diatoms to temperature increase are included hereafter only in relation to community composition and structure.

One of the main elements analysed during the experiments was the total abundance of microphytobenthic communities. In the case of copper (II) chloride (**PAPER 1**) and glyphosate in the form of the Roundup® preparation (**PAPER 2**), the total size of communities in the

tested solutions remained at a similar level throughout the study period. Only the community structure, i.e., the number of representatives of individual microalgal taxa, changed. It was observed that taxa sensitive to the test substance were replaced by tolerant or resistant taxa, leading to a change in community structure. A different response was observed for communities treated with [BMIM]Cl ionic liquid, where most of the taxa composing the community under study responded with a reduction in cell number (**PAPER 3**). In the case of a short-term temperature increase, the total community size was practically constant, with only the percentages of individual taxa changing (**PAPER 4**).

Among the dominant species, present in the initial communities and also present in all experiments, mainly diatoms were distinguished, such as: *Bacillaria paxillifera* (O.F.Müller) T.Marsson, *Diatoma moniliformis* (Kützing) D.M.Williams, *Diatoma vulgare* Bory, *Melosira nummuloides* C.Agardh, *Navicula perminuta* Grunow, *Tabularia fasciculata* (C.Agardh) D.M.Williams and Round. It was observed that the most numerous cyanobacterium was the taxon *Merismopedia* sp. Meyen. Irrespective of the degree of dominance in the initial community, individual taxa showed a different type of response to the presence of an anthropogenic factor. Thus, among other things, a group of organisms was distinguished that were stimulated to grow in the presence of the tested factors. An example of a species that was stimulated by all the factors tested was *N. perminuta*. Indeed, when exposed to ionic liquid, it showed a double increase in cell number under both copper chloride and elevated temperature (**PAPERS 1 and 4**) and eight times increase in number in the presence of glyphosate (**PAPER 2**) as well as a ten times increase in the presence of [BMIM]Cl ionic liquid number compared to the control solution (**PAPER 3**). Some taxa were stimulated by the presence of chemicals only during the initial phase of testing. For example, the cell number of *Achnanthes brevipes* C.Agardh under a concentration of copper (II) chloride of $2 \cdot 10^{-5} \text{ g} \cdot \text{dm}^{-3}$ increased by 35% on the third day, and on the seventh day already 40% less cells were observed than in the control solution. An unusually large, up to four times, increase in number under copper (II) chloride was observed in *Grammatophora marina* (Lyngbye) Kützing at a concentration of $2 \cdot 10^{-3} \text{ g} \cdot \text{dm}^{-3}$ CuCl_2 on the third day (**PAPER 1**). Some taxa reacted to the presence of chemicals in the last phase of the tests. For example, in *Navicula ramosissima* (C.Agardh) Cleve, seven times more cells were observed on the seventh day of testing than in the control solution (**PAPER 3**). Glyphosate at a concentration of $8.5 \text{ g} \cdot \text{dm}^{-3}$ increased the cell number of *T. fasciculata* by 40% compared to the control solution, on the seventh day. The number of cyanobacteria representatives, i.e., *Merismopedia* sp. and *Spirulina* sp. Turpin ex Gomont, increased several

times during the tests (416% and 1750%, respectively). Due to the presence of glyphosate, it is likely that the culture medium was enriched in phosphorus nutrients (Delpy et al. 2022), leading to a complete remodelling and domination of the microphytobenthic community by cyanobacteria (**PAPER 2**).

Among the taxa identified as neutral to agents of anthropogenic origin, in case of copper (II) chloride, were, for example: *Brebissonia lanceolata* (C.Agardh) R.K.Mahoney and Reimer, *Cocconeis pediculus* Ehrenberg, *Fallacia* sp. Stickle and D.G.Mann and *Rhoicosphenia abbreviata* (C.Agardh) Lange-Bertalot (**PAPER 1**). In contrast, glyphosate at the concentrations tested did not affect changes in number in, among others: *Halamphora coffeaeformis* (C.Agardh) Levkov, *T. fasciculata* (**PAPER 2**). Species described as neutral to the applied factors almost weren't observed, in the case of the ionic liquid and short-term temperature increase (**PAPER 3 and 4**).

Several anthropogenically sensitive taxa were identified in the communities tested. Organisms that responded with a reduction in number to the presence of both copper (II) chloride and ionic liquid included *B. paxillifera* and *T. fasciculata* (**PAPERS 1 and 3**). The diatom *M. nummuloides* was a species showing particular sensitivity to the chemicals used, e.g., its count under glyphosate was reduced to only 15% of that in the control sample (**PAPER 2**). Similar reactions were observed for *A. brevipes* and *Cylindrotheca closterium* (Ehrenberg) Reimann, whose cells were not observed in glyphosate and in ionic liquid, on the seventh day of testing. Negative effects of the presence of copper (II) chloride in the culture medium were also observed for representatives of cyanobacteria, e.g., in *Spirulina* sp. a reduction in counts by approximately 40% compared to the control solution was observed (**PAPER 1**). Among the species sensitive to the short-term temperature increase was *G. marina*, whose counts decreased by 43% on the last day of testing compared to the control sample (**PAPER 4**).

The state of chloroplasts, as determined by changes in shape and structure, is an additional indicator that complements information about the cellular condition of the organisms forming the communities used in the experiments. The results obtained indicate that the cell number of individual taxa can remain at a similar level or increase over short periods of time despite significant impairment of chloroplast function caused by the action of, for example, copper (II) chloride. Changes in chloroplast shape and structure in the presence of copper (II) chloride and ionic liquid were observed in the dominant diatom species *T. fasciculata* (**PAPERS 1 and 3**), while under the influence of glyphosate, changes in chloroplast shape and

structure were observed in *B. paxillifera* (**PAPER 2**). By observing chloroplasts, it is possible to gain a better understanding of the response of the taxa forming the communities to the chemical agents used, but it cannot be applied as a stand-alone indicator of community changes.

The research carried out as part of the doctoral dissertation allowed to observe changes in the composition and structure of marine microphytobenthic communities occurring under the influence of factors related to human activity, such as the introduction of chemicals into the environment or short-term increases in temperature, and thus the research hypothesis was confirmed. Microphytobenthos communities are more resistant than single strains of microalgae, a decrease in the number of anthropogenic factors used in single strains was observed at the early stages of research (Sylwestrzak et al. 2015). The decline of sensitive taxa and their replacement by resistant ones indicates the fact that communities as a functional whole are highly resistant to disturbances of anthropogenic origin. Studies based on a variety of microphytobenthos-forming organisms allow us to learn about the richness of responses both at the cellular level (e.g. by assessing the condition of chloroplasts) and at the population level (analysis of community composition and structure). The diatom *N. perminuta* has shown, on the one hand, a particular resistance to a relatively broad spectrum of chemical pollutants and, on the other hand, has flexibly adapted to short-term temperature increases. However, the vast majority of organisms forming the microphytobenthic communities were sensitive to the tested substances of anthropogenic origin. It should also be noted that increased concentrations of the chemicals tested, or increased temperatures favoured the mass growth of only individual taxa (e.g., *G. marina*, *N. perminuta*) or groups of taxa (e.g. cyanobacteria in the case of glyphosate).

The photosynthetic organisms that make up the microphytobenthos are extremely important elements of aquatic ecosystems due, among other things, to their role as primary producers. Understanding and quantifying the response of entire communities is extremely valuable, as it allows us to reliably estimate the changes that may occur under the influence of factors of anthropogenic origin, for each taxon included in their composition. A valuable element of the doctoral dissertation was the testing of different chemical agents based on a single methodology, which enabled different types of responses to be clearly traced in relation to microorganisms' communities and populations. The Baltic microphytobenthos includes many cosmopolitan taxa and species, which makes it possible to assume that similar responses to the tested agents, presented in the papers forming the basis of the dissertation, will be shown by communities existing in brackish waters of other regions of the world. The use of

microphytobenthic communities provides more reliable information on the likely changes that will occur in the environment as a result of human activities than tests conducted on single strains of microalgae. The results obtained in the course of the doctoral dissertation provide new insights into the functioning of microphytobenthic communities under conditions of severe stress caused by factors of anthropogenic origin, as well as enriching knowledge on the possibility of using microorganism communities in ecotoxicology.