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ORIGINAL RESEARCH ARTICLE

Long-term monitoring of a coastal marine ecosystem after landfill remediation using oxidative stress biomarkers and heavy metal analysis in mussels

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Abstract: The aquatic area surrounding the Gornostay Landfill, which is located near the city of Vladivostok (Far East of Russia), has long served as a site for the placement and operation of a solid domestic waste landfill. The functioning of the landfill resulted in extensive and complex pollution of the coastal water area. Remediation and closure measures in 2010 were expected to improve the ecological condition of the bay. This study demonstrates the application of biochemical markers of oxidative stress as indicators of metabolic recovery processes in marine invertebrates, using bivalves as a model group. The results demonstrated a time-dependent decrease in the concentrations of potentially toxic elements in mussel tissues and a reduction in oxidative lipid degradation to control levels. These findings indicate an improvement in the quality of the marine environment in the years following the 2010 landfill remediation activities. Based on molecular indicators of oxidative stress and the content of heavy metals in the digestive gland and gills of *Crenomytilus grayanus* from the polluted aquatic area surrounding the Gornostay Landfill, the study concludes that a gradual restoration of the marine environment has occurred.

Keywords: Biomonitoring; Potentially toxic elements; Landfill; Marine environmental remediation; Mussels; Oxidative stress

1. Introduction

Biomonitoring, based on the accumulation of toxicants in the tissues of bivalves, is essential for the detection of bioavailable forms of chemicals in the environment. However, it does not allow for the assessment of the impact (toxicity) of accumulated pollutants on hydrobionts. At the same time, the evaluation of unfavorable environmental conditions through biological parameters at both the molecular and cellular levels facilitates the determination of the risk posed by sublethal concentrations of chemical compounds, in cases where populations and communities do not yet exhibit overt signs of environmental degradation.[1]

Understanding the role of reactive oxygen species (ROS) and free-radical processes in the mechanisms of toxicity of a wide range of pollutants has led to the active use of various indicators of oxidative stress in recent decades.[1-4] The extent of oxidative damage inflicted on biomolecules by ROS is directly correlated with the efficacy of antioxidant defense mechanisms,

which are represented by antioxidant enzymes and low-molecular-weight antioxidants.

Gray's mussel, *Crenomytilus grayanus,* is a widely distributed species of bivalve mollusks in the coastal zone of Primorsky Krai and a traditional fishing resource. The commercial stock of this mussel in Peter the Great Bay (Sea of Japan) is estimated at 32.6×10^3 tons.[5]

This species is considered a promising target for artificial reproduction. For more than three decades, it has been the subject of both laboratory and field research focused on the bioaccumulation of toxicants that are widespread in the environment, including potentially toxic elements (PTEs), organochlorine pesticides, and radionuclides.[6-11] Furthermore, the mechanisms underlying the impacts of contamination have been a major focus of study.

The present work focuses on the aquatic area surrounding the Gornostay Landfill, where the long-term placement and operation of a solid domestic waste landfill resulted in severe and complex pollution of the coastal water area with PTEs, especially lead (Pb) and copper (Cu), as well as pesticides, petroleum products, and phenols.[12-14] Remediation and closure of the landfill were carried out in 2010 to improve the ecological condition of the bay.

The presence of toxicants such as organochlorine compounds, petroleum hydrocarbons, phenols, and PTEs has been detected in the water and bottom sediments of the aquatic area surrounding the Gornostay Landfill. Consequently, an increase in ROS levels in *C. grayanus* tissues has been observed through various mechanisms,[15,16] inducing oxidative stress. The progression of oxidative stress is evaluated by the levels of antioxidant activity and the accumulation of lipid peroxidation products.[17-19]

The aim of the present study is to evaluate changes in marine environmental quality as a consequence of remediation activities in the aquatic area surrounding the Gornostay Landfill, which is subjected to anthropogenic pollution. This evaluation is based on the content of heavy metals and molecular indicators of oxidative stress in the tissues of *C. grayanus*.

For this purpose, the activity of antioxidant enzymes, the level of reduced glutathione (GSH), and the index of integral antioxidant antiradical activity, as well as the content of lipid peroxidation products and heavy metal concentrations, were determined in the digestive gland and gills of mussels collected in the polluted aquatic area surrounding the Gornostay Landfill (Ussuriysky Bay) and relatively clean (Reineke Island) water areas of Peter the Great Bay (Sea of Japan). Mussels were collected in 2011, 2013, and 2024. The findings were compared with data from investigations carried out in 1999 on *C. grayanus* mussels from the same two water areas.[20,21]

Based on the quantitative characteristics of oxidative damage indicators and the state of the antioxidant system, it is possible to assess adaptation strategies to various environmental factors, including anthropogenic pollution. Consequently, the obtained results can serve as criteria for developing programs for environmental monitoring of coastal marine areas, thereby contributing to the preservation of marine ecosystem biodiversity.

2. Materials and methods

2.1. Materials

Aquatic areas surrounding the Gornostay Landfill (43.130910°N, 132.044114°E (WGS84)) and Reineke Island (42.933333°N, 131.741944°E (WGS84), chosen for mussel sampling, are approximately 44 km apart and show little to no variation in their primary hydrochemical and hydrological characteristics[22] (Figure 1).

The coastal waters of Reineke Island are considered clean. It is important to note that there are no industrial facilities on the island that could contribute to environmental pollution. The bottom sediments and water column in this area exhibit low levels of PTEs and pesticides.[8,14,23]

The municipal landfill for solid waste in Vladivostok was situated on the coast of the Ussuriysky Bay from 1967 to 2010, resulting in the contamination of nearby waters. Based on a study conducted by Shulkin,[23] the average concentration of PTEs in seabed sediment specimens taken from the the aquatic area surrounding the Gornostay Landfill and nearby waters, within a range of 50–100 m from the coast, were 3,739 μg/g dry wt for zinc (Zn), 2,659 μg/g dry wt for Cu, and 2,344 μg/g dry wt for Pb in the period from 1996 to 1999.

At the same time, the concentration of these PTEs in the sediments of the waters adjacent to Reineke Island were 21 μg/g dry wt for Zn, 3 μg/g dry wt for Cu, and 10 μg/g dry wt for Pb.[8] The overall concentration of organochlorine pesticides (including β-hexachlorocyclohexane, dichlorodiphenyltrichloroethane, and their derivatives) in seabed sediment specimens from the aquatic area surrounding the Gornostay Landfill was 5 times higher than in the waters adjacent to Reineke Island, reaching 1.67 ng/g dry wt. In 2008–2009, the levels of oil and phenol in the aquatic area surrounding the Gornostay Landfill were 2 and 5 times higher than the maximum

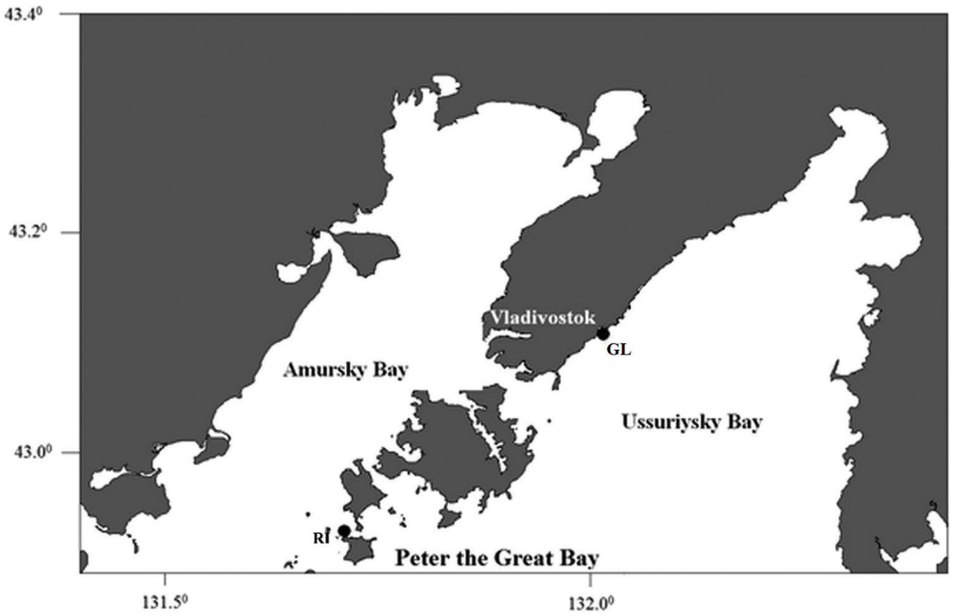


Figure 1. Locations of the sampling points in the Peter the Great Bay, which is part of the East Sea (also known as the Sea of Japan) in Russia: GL - the aquatic area surrounding the Gornostay Landfill and RI - Reineke Island.

allowable limits. The landfill was fully remediated by the end of 2010. Annual monitoring in the Peter the Great Bay, 500–800 m offshore, revealed a consistent downward trend in PTE content in the sediments of the aquatic area surrounding Gornostay Landfill by 2012.[24]

In 2011, 2013, and 2024, *C. grayanus* mussels were collected from two locations: Reineke Island, which served as the reference site, and aquatic area surrounding the Gornostay Landfill, which was considered polluted. To assess biochemical characteristics, three mussels (their gills and digestive glands) were combined to form one sample, with four samples collected for the population in total. The specimens were promptly frozen in liquid nitrogen and stored at −80°C. For metal content analysis, the tissues of six mussels were divided into three replicates, with two specimens per replicate. The tissues were then dried at 75°C and homogenized (1:10, w/v) at 0°C in 50 mM Trishydrochloride buffer (pH 8.0) with 0.1 mM phenylmethylsulphonylfluoride. The resulting homogenate was centrifuged for 20 min at 5000 g and then for an additional 40 min at 10,800 g at 4°C. The supernatant was used to assess the activity of antioxidant enzymes.

The physiological state of 20 bivalves was assessed through measurements of their condition indices (CI) and condition index based on shell length (CIL). The CI was determined as the proportion of soft-tissue mass to the total weight of the mussel, calculated as the ratio of wet soft-tissue mass (in grams) to the sum of shell, soft tissue, and pallial fluid (in grams), and multiplied by 100.[25] The CIL is based on the shell length, calculated as the soft-tissue mass (in grams) divided by the cube of shell length (in millimeters), multiplied by 10,000 (g/m).[26]

2.2. Biochemical analyses

The activity of antioxidant enzymes was evaluated at a temperature of 20°C. The activity of superoxide dismutase (SOD) (EC 1.15.1.1) was measured using the method described by Paoletti *et al*.[27] The activity of catalase (CAT) (EC 1.11.1.6) was determined by measuring the rate of hydrogen peroxide decomposition, and the activity of glutathione reductase (GR) (EC 1.6.4.2) was measured by monitoring the decrease in absorbance at 340 nm due to the oxidation of nicotinamide adenine dinucleotide phosphate.[28] The activity of glutathione peroxidase (GP) was assessed in a coupled enzyme system, in which the oxidized form of GSH was converted to its reduced form by GR.[29] The substrate used was cumene hydroperoxide, which served as a substrate for both the selenium (Se)-dependent (EC 1.11.1.9) and Se-independent (EC 2.5.1.18) activities. The concentration of reduced GSH was determined using the method described by Moron *et al*.[30] The concentration of 2-thiobarbituric acid (TBA) reactive substances was measured through a color reaction with TBA.[31] The amount of conjugated dienes (CD) was quantified using spectrophotometry.[32]

Lipofuscin (LF) was extracted from tissues following the protocol developed by Folch *et al*.[33] and quantified using the method proposed by Dillard and Tappel.[34] The concentration of protein was measured using the modified Lowry method.[35] The overall antioxidant activity of the tissue extracts was assessed by measuring the inhibition of the oxidation reaction of 2,2'-azinobis (3-ethyl-benzothiazoline-6-sulfonic acid) by peroxyl and alkoxyl radicals generated during the thermal decomposition of 2,2'-azobis (2-methyl-propionamidine) dihydrochloride.[36]

All biochemical measurements were performed using a Shimadzu UV-2550 (Germany) spectrophotometer and a Shimadzu RF-5301PC spectrofluorophotometer (Germany).

2.3. Metal analyses

The digestive gland and gill tissues were dried and weighed, then digested in Teflon bottles using 16 M nitric acid. The concentrations of iron (Fe), Zn, Cu, cadmium (Cd), and Pb were determined using a Shimadzu AA-6800 atomic absorption spectrophotometer (Germany). The quality of the analysis was verified by comparing the results with those obtained from a certified standard sample (ERM-CEZ78 mussel tissue). The precision of the analysis exceeded 90%. Mercury content was determined by the atomic absorption method using a RA915M mercury analyzer with the PIRO915 pyrolytic attachment.

2.4. Statistical analysis

An unpaired *t*-test was used for statistical analysis. A significance level of $p<0.05$ was adopted to determine statistical significance (STATISTICA, StatSoft, United States of America).

3. Results

Morphometric data showed that the age of the bivalve specimens harvested from the aquatic area surrounding the Gornostay Landfill and Reineke Island in 2011 and 2013 was approximately 8–16 years old, whereas in 2024, the age was estimated to be 18–30 years. The Gray's mussel is a long-lived mollusk;[37] therefore, the reproductive age of the animals used for analysis from the two water areas can be considered physiologically similar. Mussels from the polluted aquatic area surrounding the Gornostay Landfill exhibited smaller mean shell sizes across all years, measuring 9.66 ± 0.80 cm (2011), 10.36 ± 0.83 cm (2013), and 11.57 ± 0.70 cm (2024), compared to mussels inhabiting the clean waters near Reineke Island, which measured 11.03 ± 0.84 cm (2011), 11.63 ± 0.75 cm (2013), and 13.59 ± 0.87 cm (2024).

The CI did not differ significantly between mussels from the aquatic area surrounding the Gornostay Landfill and Reineke Island, amounting to 21.74 ± 2.44 and 20.96 ± 2.84, respectively. The CIL values were consistent with expected ranges, averaging 0.28 ± 0.06 for mussels from the aquatic area surrounding the Gornostay Landfill and 0.23 ± 0.04 for mussels from Reineke Island.

3.1. Toxic trace elements

Examination of the metal composition within the tissues of bivalve mollusks from Reineke Island (Table 1) revealed that the annual fluctuations in Fe and Zn content were not significant. However, fluctuations in Cu and Cd levels exhibited a consistent pattern over the observed period. It should be noted that Cu content in the digestive gland increased twofold, whereas Cd content in the gills decreased fivefold compared to the levels recorded in 1999. As demonstrated in the 2024 study of mussels from the Reineke Island area, Pb content in their tissues increased threefold compared to 1999 levels.

In 1999, it was found that the concentration of Cu in the digestive organs and gills of mussel specimens collected from the aquatic area surrounding the Gornostay Landfill exceeded those from Reineke Island by factors of 5 and 50, respectively (Table 1). Fe and Zn contents were also higher in both tissues compared to mussels from Reineke Island waters (Table 1). The results indicated that the concentrations of all metals in mussels collected in the aquatic area surrounding the Gornostay Landfill in 2011, 2013, and 2024 were lower than those recorded in 1999. However, an increase in Cd content by 1.7 times in the digestive gland was observed in 2024 compared to 1999.

In 2011, 2013, and 2024, following the remediation of the waste site, the Fe and Zn contents in the mussels from the aquatic area surrounding the Gornostay Landfill were found to be comparable to those in mussels from Reineke Island. During the final observation period, mussels from the polluted site showed a significant decrease in Cu and Pb concentrations in the gills (by 69 and 30 times, respectively) and digestive gland (by 5 and 10 times, respectively) compared to 1999 levels. In 2024, Cu content in the digestive gland of mussels from the aquatic area surrounding the Gornostay Landfill was 2.3 times lower than that of mussels from Reineke Island, whereas Cu content in the gills was comparable in both cases. Cd and Pb contents in 2024 in mussels

Table 1. Mean concentrations of toxic trace elements (±standard deviation, μg/g dry weight) determined in the tissues of mussels collected from polluted areas (GL) and pristine sites (RI)

Site	Toxic trace element	Digestive gland			Gills			
		August 1999[a]	July 2011	July 2013	November 2024	August 1999[a]	July 2011	July 2013	November 2024
RI	Fe	123±27	119.7±14.7	194.8±27.8	162.3±37.52	225±25	116.1±7.4	159.3±45.4	155.58±4.72
GL		620±211*	153.1±17.6	144.51±19.42	190.43±3.85	377±169	129.6±25	169.9±101	161.1±48.76
RI	Zn	108±50	97.1±13.6	137.4±13.3	146.03±9.11	109±7	95.3±11.6	118.5±25.5	73.25±0.04
GL		229±74	90.4±15.1	100.83±12.88*	156.8±14.69	272±94*	87.0±1.6	75±10	77.54±2.57
RI	Cu	12.0±1.1	24.9±0.4	19.4±0.7	27.61±4.35*	9.1±0.9	13.4±10.7	7.6±2.9	7.76±1.43
GL		61.0±5.8*	89.3±10.9*	24.14±2.96	12.0±1.77	448±179*	36.1±7.7*	15.7±2.1*	6.52±0.66
RI	Cd	3.8±2.3	4.6±0.9	11.0±6.2	8.42±2.34	20.2±2.25	7.9±5.0	16.7±4.9	3.81±0.57
GL		9.73±2.47*	2.7±0.8	4.19±2.28	16.75±1.12*	14.9±3.5	1.2±0.3	0.7±0.1*	6.18±1.41*
RI	Pb	n.d.	n.d.	1.7±0.6	3.12±0.40	n.d.	n.d.	1.19±0.8	2.91±0.23
GL		169±50	28.9±12.9	26.31±4.71*	16.3±0.74*	233±53	80.3±9.3	77.3±24.9*	7.75±2.10*
RI	Hg	-	-	-	0.058±0.034	-	-	-	0.02±0.004
GL		-	-	-	0.142±0.033*	-	-	-	0.02±0.004

Notes: aFrom Ref.[21] *indicates significant differences between the levels of metals in the tissues of mussels from the reference and polluted sites (n=3, $P<0.05$).
Abbreviation: n.d.: Not determined, GL: the aquatic area surrounding the Gornostay Landfill, RI: Reineke Island.

from the polluted site were higher in the digestive gland by 2 and 5 times, and in the gills by 1.5 and 2.6 times, respectively. For the 1st time, mercury content was determined in mussels from the aquatic area surrounding the Gornostay Landfill. The analysis revealed that mercury content in the gills did not differ from that of mussels from Reineke Island waters; however, in the digestive gland, it was found to be 2.4 times higher.

3.2. Lipid oxidative damage products

The results indicated that in 1999, there was a marked increase in the concentration of all lipid peroxidation metabolites in the digestive gland and gills of bivalves collected from polluted site compared to those from Reineke Island (Figure 2). Following landfill remediation, there was no significant discrepancy in the concentrations of lipid peroxidation metabolites in the gill tissues of mussels. The malondialdehyde (MDA) content in the digestive gland exhibited a marked increase (80%) 1 year after remediation (in 2011), whereas LF content in mussels from the aquatic area surrounding the Gornostay Landfill was significantly lower than that in mussels from the control site, Reineke Island, in both 2011 and 2013. Fourteen years after remediation, the concentrations of MDA in the tissues of mussels collected from the aquatic area surrounding the Gornostay Landfill were not significantly different from those found in the water area around Reineke Island.

3.3 Antioxidant system

Among the antioxidants investigated in 2011, the activity of SOD in the digestive glands and gills of mussels collected from the aquatic area surrounding the Gornostay Landfill exhibited a remarkable increase of approximately 1.8- and 1.3-fold, respectively, relative to those from the pristine water area (Figure 3). The level of glutathion-S-transferase (GST) activity was found to be twofold higher in the gills of mussels from from polluted site. In addition, a 1.5-fold increase in the level of integral antiradical activity (IAA) was observed in mussels from the aquatic area surrounding the Gornostay Landfill. The activities of other antioxidant enzymes (CAT, GP, and GR) did not differ significantly (Figure 3). In 2013 and 2024, the activity of SOD in the digestive gland and gills of mussels from the aquatic area surrounding the Gornostay Landfill was found to be 1.3- and 1.9-fold higher, respectively, compared to mussels from Reineke Island. In 2024, the IAA level in the mussel tissues from the two water areas was found to be equivalent.

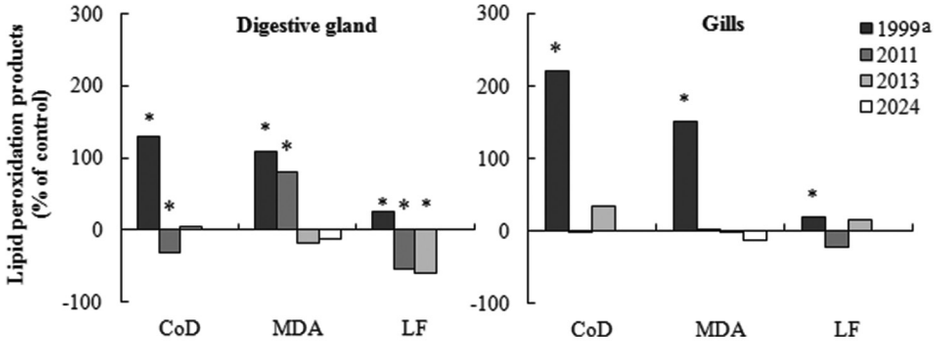


Figure 2. Levels of conjugated dienes, malondialdehyde, and lipofuscin in tissues of *Crenomytilus grayanus* mussels harvested from the aquatic area surrounding the Gornostay Landfill compared to those from Reineke Island, serving as a control group. The results are expressed as percentages of the control values.
Notes: [a]From Ref.[21] *denotes statistically significant differences ($n = 4$, $p<0.05$).

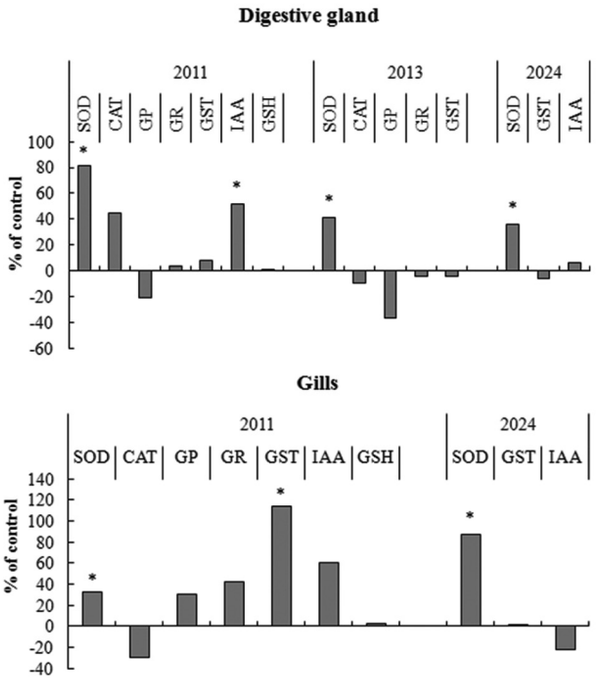


Figure 3. Antioxidant defense parameters in *Crenomytilus grayanus* mussel tissues collected from the polluted site, expressed as a percentage of the control (mussels from Reineke Island)
Note: *indicates statistical differences($n = 4$, $p<0.05$).
Abbreviations: CAT: Catalase; GP: Glutathione peroxidase; GR: Glutathione reductase; GSH: Glutathione; GST: Glutathion-S-transferase; IAA: Integral antiradical activity; SOD: Superoxide dismutase.

4. Discussion

The CI is widely utilized in environmental monitoring to evaluate the physiological condition of mussels. It has been demonstrated that exposure to pollutants has a detrimental effect on the growth of organisms due to the high energy costs associated with detoxification processes.[38] The results demonstrated that mussels from the contaminated aquatic area surrounding the Gornostay Landfill exhibited a CI that was indistinguishable from that of mussels inhabiting the uncontaminated waters of Reineke Island. This finding suggests that environmental pollution does not induce a state of physiological disturbance.

Metal concentrations in mussel tissues are indicative of their bioavailability in the environment; therefore, the temporary reduction in metal concentrations in the investigated marine hydrobionts (mussels) observed in 2011, 2013, and 2024 was attributed to environmental factors such as reduced metal input due to rainfall runoff, as well as biological processes such as metabolism and depuration. The decrease in metal content observed in both bottom sediments[39] and seawater of the polluted site[40] is indicative of landfill remediation on the coast of Ussuriysky Bay in 2010. The remediation process was found to have a substantial impact on the levels of Pb, Cd, and nickel compounds present in the former landfill site. This decline was evident in the reduced concentrations of these metals in the tissues of the oyster *Magallana gigas*, as well as in the silty fraction of the bottom sediments. A reduction of heavy metal concentrations in the brown algae *Sargassum miyabei* and *Spirochaeta pallidum* as a result of waste site remediation was also described by Kozhenkova *et al*.[41]

In 2011, the level of Cu in the mollusk's digestive organ remained stable compared to 1999, whereas the level of lead in both organs had decreased significantly by 2011 and remained unchanged until 2013. Fourteen years after the remediation, the Cu content in the

digestive gland and gills of the mussels attained levels comparable to those found in mussels inhabiting the waters of Reineke Island. Concurrently, the Cu content in the digestive gland of mussels from Reineke Island exhibited higher levels compared to those found in the aquatic area surrounding the Gornostay Landfill in 2024. The concentrations of Cd and Pb in the tissues of mussels collected from the polluted site, however, exceed the acceptable limits.

The sediments of the aquatic area surrounding the Gornostay Landfill are composed mainly of fine and medium-sized sands (more than 50% of the total sediment) and large siltstones, while the proportion of pelites is among the lowest. The sediments are subjected to agitation by periodic wave action, which results in the suspension of finer fractions enriched in PTEs, forming a hydraulically lighter fraction.[42] It is probable that periodic hydrodynamic leaching of PTEs from heavily contaminated sediments in polluted site is the primary source of bioavailable forms of Cd and Pb at present. The intricate pattern of Pb excretion from mussel tissues may also be associated with its reabsorption from shell material under unfavorable environmental conditions.[43]

In this study, the suitability of mussels from polluted site for use as food was evaluated. Based on the data obtained by Kavun and Shulkin,[21] the total content of PTEs in all tissues was calculated. Knowing the approximate percentage of the total content of PTEs in mussel tissues, it is possible to extrapolate data on metal content in individual organs (digestive gland and gills) to the content in whole tissues (estimated total content). Comparing the estimated total content values obtained with quality standards, it can be concluded that mussels from the aquatic area surrounding the Gornostay Landfill contain excessive levels of Cd and Pb. The consumption of such mussels is therefore unacceptable and poses a potential risk to human health.

The products of lipid peroxidation (CD, MDA, and LF) are extensively employed as indicators of oxidative stress in aquatic organisms.[44-46] A comparative analysis of the state of the marine environment before and after the remediation of the solid waste dump revealed that the processes of uncontrolled oxidation of free radicals in the digestive glands and gills of mussels from polluted site had undergone a significant reduction. All three indicators (CD, MDA, and LF) correspond to those of mussels from a clean site.

The antioxidant defense system is considered to be one of the most significant systems in maintaining the integrity of the organism and the chemical constancy of its internal environment.[47] It has been demonstrated that in organisms exposed to pollution, an increase in the activity of antioxidant enzymes results in a reduction in sensitivity to oxidative damage. Thus, SOD catalyzes the disproportionation of the superoxide radical ($O_2^{\bullet -}$), which is constantly formed as an intermediate product in reactions involving oxygen. The activation of SOD in mussels from polluted site suggests an increase in the generation of $O_2^{\bullet -}$, which is capable of reducing Fe^{3+} and Cu^{2+}, thereby catalyzing the formation of the most harmful oxygen-based molecules—the hydroxyl radical (HO•). In addition, $O_2^{\bullet -}$ has the capacity to directly initiate the process of fatty acid peroxidation in biological membranes.[47] The antioxidant role of GST is the destruction of fatty acid peroxides and the detoxification of electrophilic compounds, including aldehydes and epoxides, which are formed during lipid peroxidation.[48] It is probable that the increased activity of GST observed in the gills in 2011 contributed to the prevention of MDA formation, which is also involved in the formation of LF.

The observed increase in SOD and GST activity in the organs of mussels from polluted site (2011, 2013, and 2024), evidently reflects the compensatory reaction of the organism aimed to neutralizing the increased production of $O_2^{\bullet -}$ and detoxifying lipid peroxidation products. Higher activity of antioxidant enzymes has been documented in various species of marine invertebrates from polluted habitats compared to animals from unpolluted areas.[26,38,49] The upregulation of antioxidant enzymes may indicate a state of mild oxidative stress, in which the enzymes are not deactivated by ROS. Alternatively, it may be physiologically feasible for the organism to increase the expression of these enzymes.[50,51]

According to our data, the activity levels of CAT, GP, and GR did not differ significantly in mussels from both biotopes (2011 and 2013). Furthermore, no disparities were identified in the concentration of low molecular weight antioxidants (GSH) within the tissues of these mussels. However, assessing the overall physiological state of the organism based solely on individual components of the antioxidant defense system is challenging. Consequently, the total IAA was determined, which characterizes the total antiradical efficiency of the antioxidant defense of the cell in relation to peroxyl and alkoxyl radicals(ROO•, RO_2•). The higher IAA index in the digestive gland of mussels from polluted site in 2011 indicated an increased ability of mussels to neutralize toxic radicals. In 2024, the IAA level in the tissues of mussels from both water areas was found to be comparable.

The results of studies in 1999, 2011, 2013, and 2024 showed a decrease in the concentration of PTEs in mussel tissues and a reduction in lipid oxidation to reference values as a result of landfill remediation on the shore of polluted site. These results indicate that biochemical indicators used to measure the degree of oxidative damage can also serve as indicators for assessing the recovery of metabolic processes in aquatic organisms after environmental rehabilitation. The release of PTEs into aquatic ecosystems often leads to their accumulation in bottom sediments. Routine environmental monitoring programs employ the measurement of metal concentrations in sediment samples as a means of assessing potential impacts on biological diversity and ecological health. The findings indicated a marginal reduction in metal concentrations in sediments, coupled with a near-complete elimination of PTEs from mussel tissue, suggesting that a portion of the metals present in sediments exists in a relatively inactive state, rendering them inaccessible to aquatic organisms. Furthermore, it appears that the introduction of dissolved and particulate metal forms through terrestrial runoff has been diminished following remediation efforts. Thus, the content of PTEs in tissues serves as a critical indicator of potential stress on biological systems and the environment, as these parameters directly influence the bioavailability and subsequent biological effects. The findings of this study also reveal a strong correlation between the concentrations of toxic substances in tissues (PTEs) and their associated effects (lipid peroxidation products and activation of the antioxidant defense system).

5. Conclusion

It is evident from the results obtained that a comprehensive analysis of the antioxidant potential of tissues (including the activity of antioxidant enzymes and the integral antiradical index) is a useful approach in regional monitoring programs, both in relation to increasing anthropogenic impact and to the implementation of environmental protection measures. Undoubtedly, the efficacy of this approach is enhanced by the concurrent use of chemical analyses of tissues to detect the presence of toxicants.

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Conflict of interest

The authors declare no conflicts of interest.

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Ethics approval and consent to participate

All procedures in this study, including the methods for utilizing invertebrates, were approved by the Bioethics Commission of the V.I. Il'ichev Pacific Oceanological Institute of the Far Eastern Branch of the Russian Academy of Sciences (protocol no. 34, approved April 15, 2021) in Vladivostok, Russia.

Availability of data

Data will be made available upon reasonable request to the corresponding author.

Further disclosure

The paper has been uploaded as a preprint to ResearchGate (doi:10.2139/ssrn.5209249).

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