УДК: 577.353.9: 574.24: 57.084

Bioindication of the ecological state (health) of coastal waters based on the use of automated bioelectronic systems

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Received September 04, 2020;
Revised September 25, 2020;
Accepted September 27, 2020

The paper presents the data obtained in the study of the health of the ecosystems in a number of recreational areas of the Black Sea and the Eastern Gulf of Finland. Prompt assessment of the ecosystems health was carried out by testing the health of adult bivalve mollusks inhabiting them, using the functional load method based on the analysis of their heart rate measured by a fiber-optic bioelectronic system. The goal of the study was to test the innovative bioindication technology developed earlier by the authors on the basis of the above-mentioned method in the regional programs for environmental monitoring of the ecosystems of coastal marine and freshwater recreational water areas (using the example of a number of water areas of Sevastopol, as well as several freshwater areas of the Kurortniy District of St. Petersburg. It has been established that the functional state of mollusks may serve as an indicator of excessive pollution of coastal waters by the objects that discharge insufficiently treated wastewater from their local treatment facilities, including household wastewater. The technology applied in this study for the assessment of functional state of local species of mollusks may be effectively used to solve the problems of early diagnostics of alterations in the health status of coastal aquatic ecosystems and contribute to ensuring the ecological safety of recreational water areas, serving as an infobase for the development of science-based environmental management decisions. Taking into account a high expressiveness and easy-to-use technology applied in the study (which does not require expertise for its practical implementation), this technology may be considered as the best available technology for biomonitoring the water quality of recreational waters in Russia.

KEYWORDS: assessment of the state of coastal water areas; biomarker studies; functional state of animals; bioindication; aquatic ecosystem health; heart rate of mollusks; biomonitoring; best available technologies

DOI: 10.17816/phf46438/2713-153X-2020-3-2-64-73

СОКРАЩЕНИЯ:

HR – heart rate;
IBSS – A.O. Kovalevsky Institute of Biology of the Southern Seas of the Russian Academy of Sciences;
BOD5 – biochemical oxygen demand on the fifth day;
COD – chemical oxygen demand;
SRCES RAS – Scientific Research Center for Ecological Safety of the Russian Academy of Sciences;
Trec – recovery time;
SPC RAS – Saint Petersburg Federal Research Center of the Russian Academy of Sciences

INTRODUCTION

In times of increasing anthropogenic pressure the value of many marine and freshwater coastal areas as recreational water bodies is constantly reducing. At the same time persistent water pollution inevitably leads to a decrease of adaptive capacities local aquatic organisms not related to saprabionts. Thus, in contrast to macrobenthos animals with a long life cycle inhabiting unpolluted water areas, animals from the polluted ones usually have a lower status of their functional state. Compared to similar organisms from conditionally clean waters, they require additional energy costs to maintain homeostasis, which inevitably adversely affects their health.

In a number of documents [1, 2] reference is made to the necessity of the development of new techniques and instrumental approaches for the development of some criteria for assessing the state of ecosystems based on the application of biological methods. Recent studies have demonstrated the effectiveness of the use of biomarkers in the indication of biological effects of the impact of pollutants or their mixtures on the ecosystem state [3–6].

In this regard a number of criteria and methods were proposed and substantiated based on biomarker indicators of macrobenthos invertebrates and fish, as well as on the determination of concentrations of contaminants in sediments and organs of aquatic organisms. A biomarker is understood as the response to biologically significant impacts of different nature [7–10]. The directions for the use of biomarker indicators were provided in such important international environmental documents as the ICES Working Group on Biological Effects of Contaminants (WGBEC) [2] and in the EU Directive [1].

The paradigm put forward by the British scientists turned out to be very successful: "Healthy animals, healthy ecosystem" [11]. According to this paradigm, biomarker studies conducted on individual organisms (randomly taken from the natural population) make it possible to extend conclusions to the state of the population as a whole and, thus, to indirectly assess the ecological state of the ecosystem inhabited by the animals under study - biological "targets" of integral toxic effects of pollutants [3, 8, 12].

Carrying out biomonitoring investigations for the diagnostics of the state of the surrounding aquatic environment it is recommended to use a number of living organisms as "targets" of the toxic effects of pollution, among which bivalve mollusks play an important role [2]. In this case, the study of the biomarker "response" of mollusks could be carried out at different levels: genetic, organismal, tissue, and others [6, 7, 11, 13, 14].

An obvious advantage of this approach is that the recorded effects may manifest themselves at the organismal level when exposed to even sub-lethal concentrations of water pollution [4, 15–18]. This makes it possible to detect changes in the health status of target species long before the onset of serious changes or even degradation of their populations, communities, and disturbances of the ecosystems they inhabit [7, 12, 13].

It is a matter of common observation, that a decrease in the body's ability for quick recovery after stress loads (within the tolerance of the species) characteristic for poor health may be detected by the method of functional loading long before the obvious manifestation of signs of a serious disease not only in humans [19], but also in the invertebrates [20-22]. In particular, the cardiovascular system indicators are currently used for assessing the adaptive capacity and health of the organism [19, 20, 22, 23, 24]. In a number of studies with higher crayfish and shell mollusks it was reported that [5, 6, 12, 15, 25], a study of higher crayfish and shell mollusks showed that the speed of recovery of their cardiac rhythm after standardized stress influence characterized the ability of organism to compensate for changes caused by external factors, which was an important sign of the tested animal's health. Indeed, the adaptive capabilities of the cardiorespiratory system reflect the intensity of physiological processes, and also, in many cases, make it possible to assess the functional state of the organism as a whole [9, 19, 20, 23]. Heart function of mollusks has lately become one of the most widely used indices in assessing effects of heavy metals [26–28], ammonia [29], detergent [12], and oil [30]. Previously, it had been found by the biomarkers of the locomotor activity of the valves of bivalve mollusks [31–34].

Within the framework of previously mentioned ecotoxicological biomarker studies, the authors found that both marine and freshwater shell mollusks taken from ecologically safe clean zones differ from the animals from polluted water areas as they demonstrate higher adaptive capacity [6, 10, 24, 25, 35]. It is expressed, in particular, in a significantly shorter HR recovery time to the background level after removing one or another type of short-term functional load. In this regard, a new physiological biomarker was proposed [15, 24, 35]: the time of adaptive recovery of HR to the background level after the removal of a short-term (within 0.5–2 hours) functional load [22]. At the same time, as further studies showed, it was most convenient to use a rapid (in 2–3 minutes) change in water salinity (within the tolerance of the species) per one hour as a standardized functional load. Based on the use of this method, an innovative technology was proposed for bioindication of ecosystems state in the water areas in which the animals under study live [6, 10, 36]. Later, the technique was tested in a number of domestic and international projects [24, 25, 37–42].

This paper presents the data obtained in the study of health characteristics of ecosystems in a number of recreational areas of the Black Sea and the Eastern Gulf of Finland. The main goal of the studies performed was to test the prospects of using the bioindication technology discussed above in regional programs for environmental monitoring of the state of ecosystems in coastal marine and freshwater recreational water areas (using the example of a number of water areas in Sevastopol, as well as in the Kurortniy District of St. Petersburg).

MATERIALS AND METHODS

Four Sevastopol water areas (in the area of Cape Khrustalniy, Kruglaya Bay, Kazachya Bay, Matyushenko Bay) with different levels of recreational load (Fig. 1, Table 1) were selected as objects of research on the Black Sea coast, and the subject of research was the Mediterranean mussel, Mytilus galloprovincialis (Lam.).

Mytilus galloprovincialis (Lam.) is a typical representative of the Black Sea malacofauna. The mussels were sampled in May (before the beginning of the tourist swimming season) and in October (at the end) of 2019.

In Fig. 1 the sampling sites are marked with red dots.

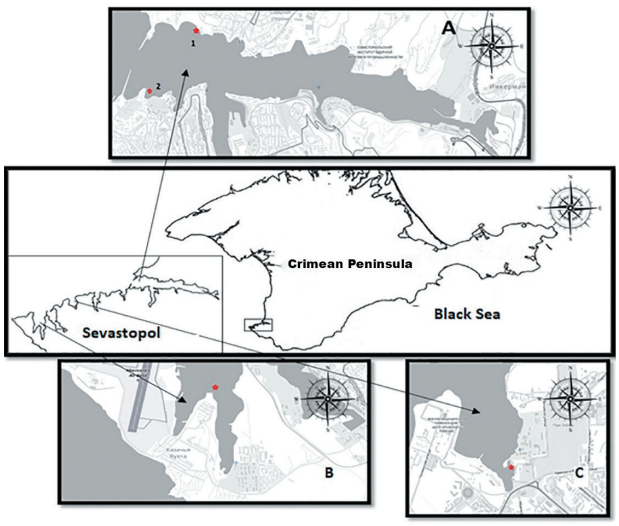

Fig. 1. Schematic map of the areas under study: A - b. Sevastopolskaya (1 – b. Matyushenko, 2 – Cape Khrustalniy); B – b. Kazachya, C – b. Kruglaya

Рис. 1. Карта-схема районов исследования: А – б. Севастопольская (1 – б. Матюшенко, 2 – мыс Хрустальный); В – б. Казачья, С – б. Круглая

The data of the hydrochemical characteristics of the seawater samples from the studied water areas, regularly measured by the experts from the A.O. Kovalevsky Institute of Biology of the Southern Seas of the Russian Academy of Sciences are as follows: salinity, dissolved oxygen content, biochemical oxygen demand on the fifth day, chemical oxygen demand, pH value, mineral and organic forms of phosphorus and nitrogen, - met the water quality standards imposed to fishery reservoirs [43]. However, it should be noted that the salinity of water in the Kruglaya Bay (15 ‰) was noticeably lower compared to the other three water areas, in which this indicator was practically the same and corresponded to the usual value for the upper horizon of the Black Sea - 18 ‰ (see Table 2). This, apparently, is associated with the inflow of wastewater from local treatment facilities of numerous public catering enterprises located in the coastal strip into the water area of the bay.

The experiments were carried out on bivalve molluscs sampled in the coastal zone at a depth of 0.5–2 m. The selection of organisms was collected by hand. In total, 250 individuals were taken from all studied sites.

Four water areas of the beaches of the Kurortniy District of St. Petersburg were selected as objects of research on the coast of the Eastern Gulf of Finland. The salinity of the waters in all of them is practically the same and does not exceed 1,7 ‰.

Table 1.

Табл. 1.

Brief description of the areas under study

Краткая характеристика районов исследования

The sampling areas (coordinates)	Visual inspection results	Characteristics of infrastructure facility/ level of recreational load
Khrustalniy Cape (44.617626,33.511528)	The sampling area is the surface of the metal supports. The water is transparent, the visibility is 1.5–2 meters. The bottom is clean, rocky, with small silting zones. Contamination with various plastic waste and pieces of rope was noted.	The beach is located in the city center; the coastline is equipped with sun loungers, awnings, a rescue point, with a large number of cafes and restaurants. In the immediate vicinity there are yachts and boats parking, a place for cruising boats, a ferry, small-size boats.
Kruglaya Bay (44.597430, 33.448286)	Sampling area - the surface of the concrete support of the berth. The water is turbid, visibility is no more than 30 cm. The mussels are covered with a layer of silt. There is no obvious oil pollution, but there is a smell of oil products.	The coastline is equipped with a beach (sun loungers, awnings, lifeguard), with a large number of cafes and restaurants. There is a yacht club in the immediate vicinity.
Kazachya Bay (44.579081, 33.409535)	Sampling area is the surface of the chain securing the buoy. The transparency of the water is about one meter. The bottom is sandy with separate islands of algae thickets. There are no visual signs of oil pollution, no dead organisms, plastic.	The coastline is not equipped for a beach. In the apex part of the right horn of the bay there are boat stops; a residential area of apartment buildings, a military unit and a military training ground are adjacent to the reservoir. There is also a military unit on the southern coast of the bay. There is a cottage village on the central promontory. The left apex of the bay is not equipped.
Matyushenko Bay (44.629388, 33.522752)	Place of sampling - concrete reinforcement surface. The water is clear, the visibility is about three meters. The bottom is clean, pebble, not silted.	The coastline is not equipped for a beach (there are no sun loungers, awnings, a rescue point), catering facilities are removed. The Mikhailovsky Battery Military History Museum adjoins the beach. In the immediate vicinity there is a cruise boat and a ferry.

Table 2.

Water salinity (S, ‰) in different bays of the city Sevastopol

Табл. 2.

Величины солености (S) воды из разных бухт города Севастополя

	Cape Khrustalniy	Kruglaya Bay	Kazachya Bay	Matyushenko Bay
S, ‰	18	15	18	18

The Eastern Gulf of Finland is characterized by a shallow depth: on average - 38 m, with an area of 29,000 km2. The main environmental problems of this water area are associated with water pollution as a result of the discharge of untreated or insufficiently treated industrial, agricultural and domestic wastewater, as well as with a high degree of population of the coastal area (St. Petersburg and the Leningrad Region). This is fully typical for the Kurortniy District, located on a considerable length of the northern coast of the Eastern Gulf of Finland.

Despite the fact that currently the State Enterprise "Vodokanal" [water and wastewater treatment plant] of St. Petersburg purifies 98,5% of domestic wastewater, the coastal waters of the Gulf of Finland within the city continue to become polluted due to the unsatisfactory operation of treatment facilities and unauthorized discharges of untreated and insufficiently treated wastewater from the Leningrad Region. Thus, according to the data of the North-West Directorate for Hydrometeorology and Environmental Monitoring, the concentration of easily oxidizable organic substances exceeds the established standard in water samples taken in many large and small rivers of the Leningrad Region flowing into the Gulf of Finland.

The resort area of St. Petersburg is one of the most ecologically clean areas of the Northern capital. It stretches along the coast of the Gulf of Finland in a strip 6–8 km wide and 45 km long. The center of the district is the city of Sestroretsk; it also includes the city of Zelenogorsk and nine villages: Beloostrov, Solnechnoye, Pesochniy, Komarovo, Repino, Serovo, Ushkovo, Smolyachkovo and Molodezhnoye.

The district is a territory with a unique natural potential and recreational resources of both regional and federal significance. There are more than 40 permanently functioning sanatoriums, dispensaries, boarding houses, rest houses, ski and tourist complexes.

However, in recent years, the services of Rospotrebnadzor [Federal Service of Supervision of Consumer Protection and Welfare] increasingly impose a ban on swimming in coastal waters due to water pollution above permissible standards. This is due to the fact that the level of anthropogenic load on these water areas, apparently, exceeds their assimilation capacity.

The following territories were selected for the study:

1. Water area adjacent to the Dubki park (Sestroretsk).
2. Beach "Chudniy" in Repino settlement, which is a resting place for a large number of people. In the immediate vicinity of the bay there are 3 restaurants, a hotel and an administrative building.
3. Beach "Zolotoy" of Zelenogorsk town, also adapted for recreation of the population. Catering establishments are located in the immediate vicinity of the water area.
4. The area next to the Detsky beach in Ushkovo settlement. It is not so crowded. There are no retail outlets and catering establishments in the adjacent territory, and boarding houses and country houses of the village are located quite far from the coast - on a high sandy hill.

When assessing the ecological state of the selected recreational areas, it should be kept in mind that pharmaceuticals (for example, those listed in HELCOM as hazardous pharmaceuticals: ibuprofen, diclofenac, estradiol, etc., and others, and their metabolites), which may also have a significant effect on aquatic organisms studied in the water areas.

The bivalve mollusks Unio pictorum inhabiting all the studied waters of the Kurortniy District were chosen as animal bioindicators. The number of 3–6 year-old animal required for testing was collected by hand at a depth of 0,5 meters. Then they were placed in plastic containers and, within 1–2 hours, were delivered to the laboratory of the Scientific Research Center of Chemistry and Economics of the Russian Academy of Sciences, where they were placed in 10 L aquariums with continuously aerated natural water from their habitat.

In both studies, artificial lighting around the clock was used in the laboratory room. During the experiments the water temperature was kept stable, depending on the season, within the range of 16–19 C°. The animals were not specially fed, although they could receive some food from natural water.

In the laboratory, miniature holders were glued to the shells of mollusks in the region of the projection of the heart for attaching fiber-optic sensors intended for continuous recording of heart rate and heart rate analysis in on-line mode. Following several hours of continuous registration, the functional state of the mollusks was tested according to the functional load method developed by the authors. The physiological state of the mussels Mytilus galloprovincialis (Lam.) was assessed using the original bioelectronic system developed by the authors [10, 44–47] and the method of functional loads: a rapid decrease in water salinity to 9 ‰ for one hour, followed by restoration of ambient salinity. In the case of freshwater mollusks Unio pictorum, a rapid increase in water salinity up to 6 ‰ for one hour was used as a functional load. The functional state of the mollusks was tested following their stabilization and HR maintaining for at least two hours at a given temperature.

Following the experiments the sensors were carefully removed from the shells all mollusks were returned to their habitat without any damage.

RESULTS AND DISCUSSION

The results (after appropriate statistical processing) of testing the functional state of mussels from all sampling sites in both the spring and autumn seasons of 2019 are shown in Fig. 2.

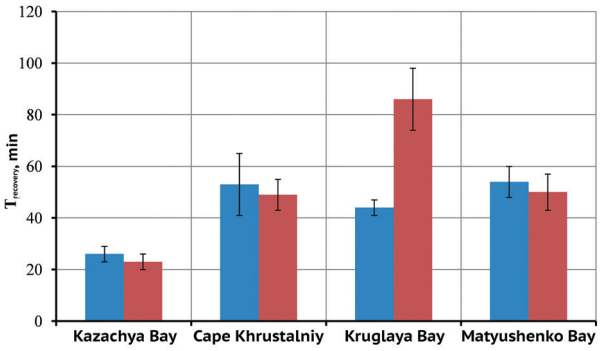

Fig. 2. Histogram of Trec values measured in May (blue bars) and October (red bars) in 2019 in the investigated waters of Sevastopol
Рис. 2. Гистограмма величин Твосст, измеренных в мае (голубые столбики) и в октябре (красные столбики) 2019 г. в исследованных акваториях Севастополя

The analysis of the status of water areas was carried out using the recommendations for ranking the ecological status of water areas set forth in [6, 36] on the basis of a large number of approbations of this method over ten years in freshwater, brackish and marine areas of different countries and continents [5, 25, 41, 42].

The results of studies of the ecological state of ecosystems in the waters of Sevastopol are presented in Table 3.

Data presented in Fig. 3 and in Table 3 shows that, regardless of the season, the status of ecosystems in the water areas of the Kazachya and Matyushenko Bays and near Cape Khrustalniy remains at a level not lower than "good". At the same time, the Kazachya Bay is the most ecologically safe throughout the year, therefore, it can be considered a reference, and mussels inhabiting it can be used as reference animals in cage studies with translocation of mussels from one site to another to exam Crimean coastal waters quality.

Table 3.

Results of testing of the mussel's functional state from different bays of Sevastopol and analysis of water quality before and after the resort season 2019

Табл. 3.

Итоговые результаты тестирования функционального состояния мидий различных акваторий Севастополя и анализа статуса этих акваторий до и после курортного сезона 2019 года

The area under study	T_{rec}, min in the period from 10 to 20 May 2019	Status of water areas in May 2019	T_{rec}, min in the period from 5 to 11 October 2019	Status of water areas in October 2019
Kazachya Bay	26±3	high	23±3	high
Cape Khrustalniy	53±12	good	49±6	good
Kruglaya Bay	44±3	high	86±12	satisfactory
Matyushenko Bay	54±6	good	50±7	good

Ecosystems in the area of Matyushenko Bay and Cape Khrustalniy have, apparently, a sufficiently high self-cleaning ability, therefore, throughout the year they remain in good ecological condition despite their rather intensive recreational use.

On the contrary, the ecosystem of the Kruglaya Bay water area has, apparently, insufficient self-cleaning ability for the season with the peak intensity of the coastal catering establishments. Despite its fairly good ecological state in the autumn-winter period, with the beginning of the holiday season (and not under the influence of any local industrial enterprises) its ecological state deteriorates sharply. This certainly reduces the recreational attractiveness of the bay during the holiday season.

The results of ranking the waters of the Gulf of Finland in accordance with the established average Trec of samples from the group of mollusks were as follows (see Table 4).

Table 4.

Results of testing Unio pictorum mollusks from different areas of the Kurortniy District of Saint Petersburg using the functional load method and subsequent analysis of the status of these water areas in the summer period May-August 2019

Табл. 4.

Итоговые результаты тестирования моллюсков Unio pictorum, различных акваторий Курортного района Санкт-Петербурга методом функциональной нагрузки и последующего анализа статуса этих акваторий в летний период май–август 2019 года

Ecological status of the water area	T_{rec} values, min	Sestroretsk town T_{rec} values, min	Repino settlement T_{rec} values, min	Zelenogorsk T_{rec} values, min	Ushkovo settlement T_{rec} values, min
High	⩽50	45±11			
Good	50–70				70±8
Satisfactory	70–100				
Bad	100–200		120±10	180±14	
Very bad	>200				

The results obtained are in accordance with the results of sanitary and chemical studies of Rospotrebnadzor specialists in St. Petersburg (see Table 5 [48]), according to which (in particular, in terms of COD and BOD5 indicators), the characteristics of organic water pollution of the investigated beaches of Repino and Zelenogorsk do not comply with sanitary rules and norms. At the same time, the Dubkovskiy (Sestroretsk) and Detskiy (Ushkovo settlement) Beaches are water areas, the ecosystems of which are at a fairly high level.

The contrast between characteristics of the ecological state of the water areas of the beaches in Sestroretsk and Repino and in Zelenogorsk indicates that they are not affected by wastewater from urban areas of St. Petersburg. And their difference from the state of the ecosystem of the beach water area in Ushkovo reveals the main source of excess pollution - the insufficiently efficient operation of local treatment facilities of boarding houses and catering enterprises located close to the coast. We have the same picture as in the case of Round Bay in Sevastopol. At the same time, in our opinion, the increased value is due to pollution associated with the presence of high concentrations of detergents in the water, and BOD5 is due to the discharge of untreated wastewater from domestic sewage. The latter is also indicated by the data of Rospotrebnadzor, demonstrating the excess content of coliform bacteria in the water of the water areas of Repino and Zelenogorsk.

It should be noted that these types of surface water pollution typical for domestic wastewater have a negative impact not only on the quality of bathing water, but also on the health of organisms living here. We also observed this effect on the Mediterranean mussels of the Boka-Kotor Bay of the Adriatic Sea [41] and, as indicated above, in the mussels of the Kruglaya Bay in Sevastopol.

Thus, mollusks can be considered as good indicators of excess pollution of coastal waters with household wastewater, and the technology used in this work for the assessment of functional state of local species of mollusks may be used as an effective, most relevant method for the rapid detection of objects located near coastline that dispose of insufficiently purified wastewater from their local treatment facilities into this water area.

Table 5.

The values of the COD and BOD5 indicators on the beaches of the Kurortniy District of St. Petersburg in June 2019 (according to Rospotrebnadzor in St. Petersburg)

Табл. 5.

Величины показателей ХПК и БПК5 на пляжах Курортного района Санкт-Петербурга в июне 2019 года (по данным Роспотребнадзора по Санкт-Петербургу)

Water area of the Gulf of Finland	COD June 2019 mgO_2/dm^3	Standard mgO_2/dm^3	BOD_5 June 2019 mgO_2/dm^3	Standard BOD_5 mgO_2/dm^3
Beach "Dubkovsky", Sestroretsk	≤30	≤30	4,1	≤4
Beach "Chudniy", Repino settlement	81	≤30	23,1	≤4
Beach "Zolotoy", Zelenogorsk	71	≤30	18	≤4
Beach "Chudniy", Ushkovo settlement	39	≤30	6,4	≤4

CONCLUSIONS

Insufficiently purified wastewater from boarding houses and catering establishments located close to the coast affects not only the sanitary-chemical and microbiological characteristics of coastal waters that are unfavorable for people, but can also lead to disruption of the health of ecosystems of coastal waters and their accelerated degradation.

The technology presented in the research work does not require the involvement of highly qualified specialists for its practical implementation. Taking into account a rather high rapidity and ease of use, it may be considered as the best available technology for biomonitoring the water quality of recreational waters developed in Russia. It may be effectively used in solving problems of early diagnostics of the state of coastal aquatic ecosystems and preventing threats to the ecological safety of ecosystems in recreational waters.

The technology implemented in these studies complements modern methods of bioindication of the quality of surface waters quality as a habitat of aquatic organisms, and can be considered as an information basis for developing science-based management decisions to ensure the ecological safety of recreational water areas. After some refinement and appropriate approbation it may be recommended for use in regional programs for ecological monitoring of ecosystems of both coastal marine and freshwater areas.

This publication was prepared with the financial support of SRCES RAS - SPC RAS, project No. 0241-2019-0018, as well as with partial financial support from the RFBR grant No. 18-44-920010 "Evaluation of the recreational menu of the waters of the Sevastopol region by bioindication methods" and Cross-border cooperation programs "Russia-Estonia" for the period 2014-2020, project ER90 HAZLESS. The content of this publication is the sole responsibility of SRCES RAS - SPC RAS and in no way reflects the position of the countries participating in the Program and the European Union.

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The authors declare that they have no conflicts of interest.

Биоиндикация экологического состояния (здоровья) прибрежных акваторий на основе использования автоматизированных биоэлектронных систем

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Поступила в редакцию 04.09. 2020 г.
После доработки 25.09. 2020 г.
Принята к публикации 27.09. 2020 г.

В работе приводятся данные, полученные при исследовании здоровья экосистем ряда рекреационных акваторий Черного моря и восточной части Финского залива. Оперативная оценка здоровья экосистем проводилась путем тестирования здоровья обитающих в них взрослых особей двустворчатых моллюсков методом функциональной нагрузки на основе анализа их кардиоритма, измеряемого с помощью волоконно-оптической биоэлектронной системы. Основная цель работы состояла в апробации разработанной ранее авторами, на основе этого метода, инновационной технологии биоиндикации в региональных программах экологического мониторинга состояния экосистем прибрежных морских и пресноводных рекреационных акваторий (на примере ряда акваторий г. Севастополя, а также нескольких пресноводных акваторий Курортного района Санкт-Петербурга, расположенных вдоль северного побережья восточной части Финского залива). Установлено, что функциональное состояние моллюсков может служить индикатором сверхнормативных загрязнений прибрежных вод объектами, сбрасывающими недостаточно очищенные стоки своих локальных очистных сооружений, в том числе хозяйственно-бытовыми стоками. Использованная в работе технология оценки функционального состояния местных видов моллюсков может эффективно применяться для решения задач ранней диагностики состояния здоровья прибрежных водных экосистем и способствовать обеспечению экологической безопасности рекреационных акваторий, служа в качестве информационной основы для выработки научно обоснованных природоохранных управленческих решений. С учетом достаточно высокой экспрессности и простоты применения использованной в работе технологии, не требующей привлечения специалистов высокой квалификации для ее практической реализации, данную технологию, по мнению авторов, можно рассматривать, как разработанную в России наилучшую доступную технологию биомониторинга качества вод рекреационных акваторий. Она может эффективно использоваться для решения задач ранней диагностики состояния прибрежных водных экосистем и служить информационной основой для выработки научно обоснованных природоохранных управленческих решений.

КЛЮЧЕВЫЕ СЛОВА: оценка состояния прибрежных акваторий; биомаркерные исследования; функциональное состояние животных; биоиндикация; здоровье водных экосистем; частота сердечных сокращений моллюсков; биомониторинг; наилучшие доступные технологии