

Abstracts
of the International Conference
Scientific and Technological Developments
of Research and Monitoring
of Marine Biological Resources

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The biosynthesis of pharmacologically important quinone pigments in sea urchin cell culture

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Marine inhabitants demonstrate a significant potential for biodiscovery research as possible sources of valuable biologically active substances for the pharmaceutical industry. Sea urchins produce secondary metabolites, such as quinone pigments, namely echinochrome and spinochromes, having highly effective antioxidant, antibacterial, antifungal, and antitumor activities. Previously, we developed an *in vitro* technology for inducing pigment differentiation process in sea urchin embryonic cell cultures both with transfection of foreign genes and with the use of the specific components of the coelomic fluids from sea urchins. Here, two sea urchin species differing in the number of embryonic pigment cells are chosen: the sea urchin *Strongylocentrotus intermedius* and the sand dollar *Scaphechinus mirabilis* (Fig. 1). We showed earlier that echinochrome was produced only in the sand dollar cells, while spinochromes were produced in the cultivated cells of both sea urchin species. Pigmentation was more intense when the cells were cultured in sea urchin coelomic fluids rather than in seawater (Fig. 2A). The maximal number of pigment cells of *S. intermedius* was detected, when the cells were cultivated in the coelomic fluid of injured sea urchins. The same picture, but at

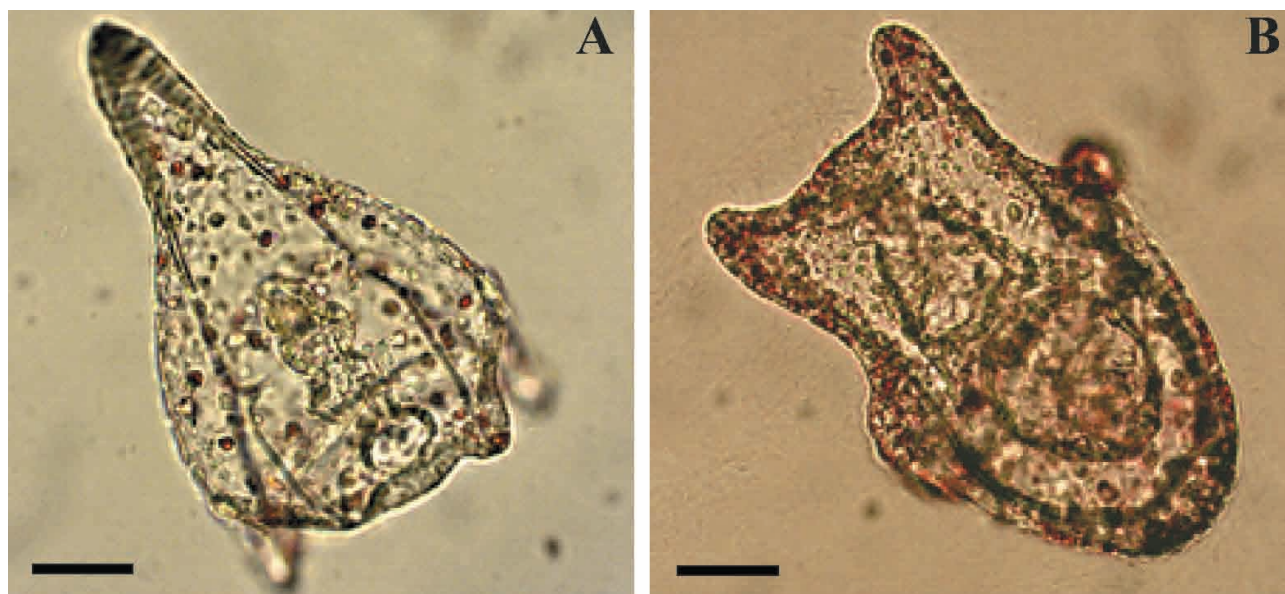


Fig. 1. Larvae at the pluteus stage of sea urchins *Strongylocentrotus intermedius* (A) and *Scaphechinus mirabilis* (B). Bars 20 μ n.

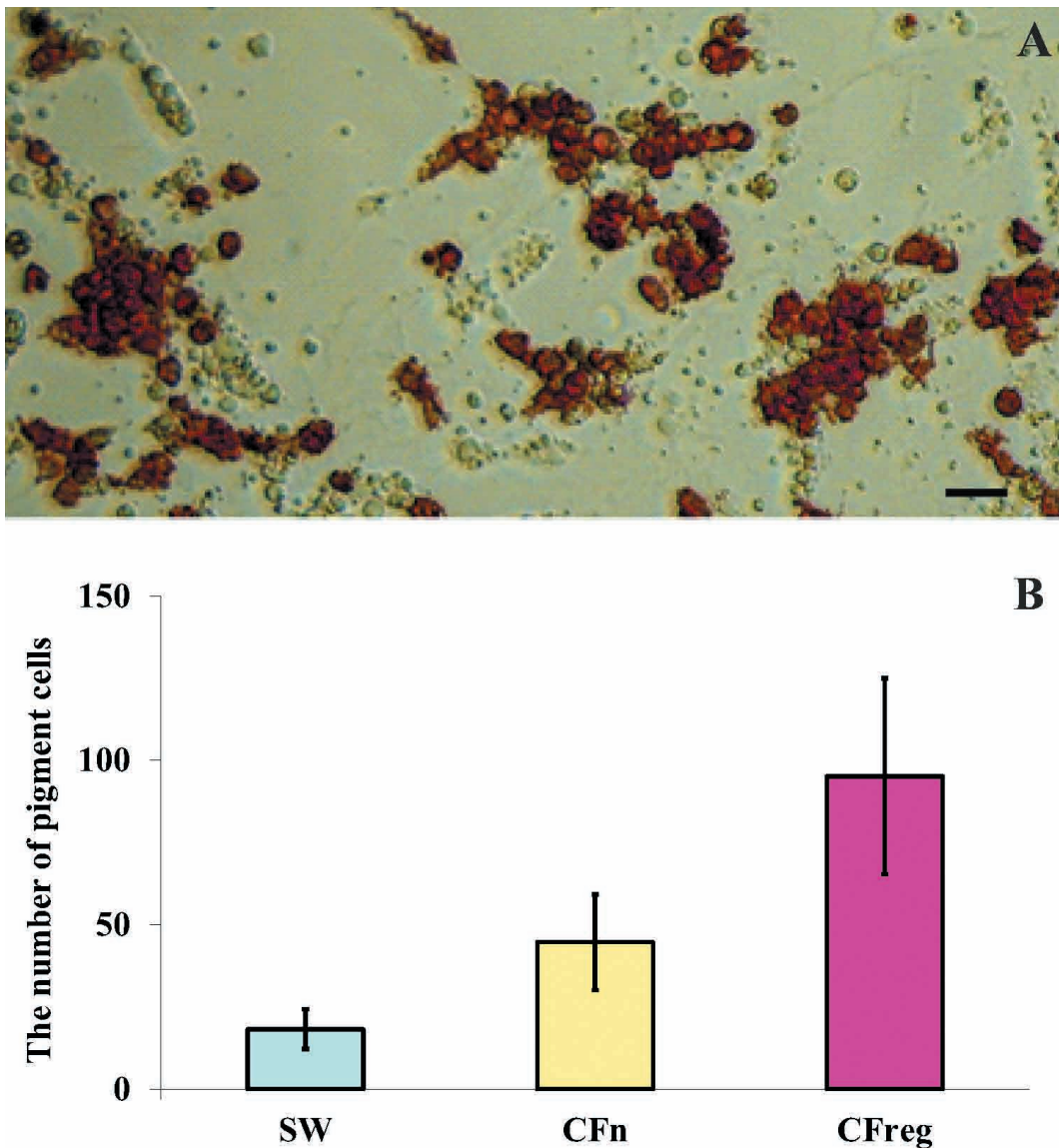


Fig. 2. Pigmentation in a blastula-derived culture from the sea urchin *Scaphechinus mirabilis*: Pigmented cells, cultivated in the coelomic fluid of normal sea urchins (A); the expression level of the pigmented genes in sand dollar cells cultivated in different culture media during four days (B). Bar 10 μ n.

much earlier timing culturing, was observed for cultivated sand dollar cells. The analysis of the gene expression associated with the induction of pigment differentiation suggests a significant difference between the expression levels of these genes (estimated by quantitative real-time PCR) in sand dollar cells cultivated in different culture media (Fig. 2B). This assay is a useful tool for assessing the production of naphthoquinone pigments during cultivation. Our data support the hypothesis that specific components of sea urchin coelomic fluids can act as inductive signals in pigment differentiation in the early stages of cultivation through the regulation of genes implicated in naphthoquinone synthesis. The study can help to reduce the impact on the adult sea urchin populations and is important for the development of new techniques in marine biotechnology and pharmacology, including the generation of cell cultures producing complex bioactive compounds with therapeutic potential.

Polyploid cells in haemolymph of scallops *Swiftopecten swifti* (Bernardi, 1858) in the Vostok Bay of the Sea of Japan (Russia)

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By means of light microscopy and flow cytometry, the nuclear morphology and DNA content were studied in haemocytes of scallops *Swiftopecten swifti* in parallel with some morphofunctional parameters of the cells (cell size, their aggregation activity and ability to form lamellopodia, cytoplasmic f-actin content and distribution). Adult scallops (the shell length 70–100 mm) were collected from a natural population of the Vostok Bay (the Eastern part of Peter the Great Bay, the Sea of Japan, 42°52' N, 132°43' E) in summer for three consecutive years – 2014 (21 individuals), 2015 (12 individuals) and 2016 (27 individuals).

In all three years, the haemocytes with pathologically altered nuclei were observed in the hemolymph: bi-, tri- and tetra-nuclear cells as well as the cells with one large pleomorphic nucleus (the bean-shaped, dumbbell-shaped, rod-shaped nuclei) corresponding to the mitotic block. The share of haemocytes with nuclear abnormalities grew significantly ($p \leq 0.0001$) over the three-year period, the mean values were 9.84 ± 2.51 , 31.61 ± 1.35 and $45.21 \pm 2.09\%$ in 2014, 2015 and 2016, respectively. Simultaneously, the mean values of DNA content in G_1 -cells (compared with the sperm haploid standard) gradually changed ($p \leq 0.0001$) during this time as follows: 2.23 ± 0.04 , 2.98 ± 0.21 and $4.12 \pm 0.14 c$ in 2014, 2015 and 2016, respectively. Thus, in 2014, most haemocytes were hyper-diploid, and, by 2016, the vast majority of the cells had become tetraploid with corresponding aneuploid “shift right”. Accordingly, the FSC (forward light scattering) values reflecting the cell sizes increased significantly ($p \leq 0.0001$) from 2014 (792.33 ± 22.80 cu) to 2016 (1265.89 ± 11.29 cu).

It should be noted that all DNA profiles in 2014 and 2016 had one dominant G_1/G_0 -peak (hyper- $2n2c$ and hyper- $4n4c$, respectively), and some cells were in G_2/M phase (hyper- $2n4c$ and hyper- $4n8c$, respectively). In 2015, most mollusks demonstrated one dominant hyper-diploid G_1/G_0 -peak (in some cases the DNA values reached $3c$), but there were some individuals in the sample with G_1/G_0 -haemocytes of two ploidy levels (about $2.5c$ and $5c$) in equal proportion. So, the mean DNA content value obtained in 2015 (about $3c$) is the result of summing of the values of both G_1/G_0 -peaks.

To assess the functional status of polyploid haemocytes, the samples of 2014 and 2016 years were compared for aggregations activity of the cells. Counting the cell conglomerates which had formed during the haemolymph withdrawing was performed by flow cytometry. The share of conglomerates from all events was $13.05 \pm 0.63\%$ in 2014, and this share dramatically decreased to $2.61 \pm 0.25\%$ in 2016. It corresponds to results of morphological analysis which showed a decrease in haemocytes ability to form lamellopodia in 2016 compared to 2014. The haemolymph samples collected in 2016 were also analyzed for f-actin content and distribution. In haemocytes of different shapes, the microfilaments had ring submembrane localization, but the greatest amount of f-actin

was detected in lamellopodia of amoeboid-shaped cells. By flow cytometry, the cells were clearly divided into two subpopulations, one of them showed the low f-actin content, another – the high content. The share of cells in the actin-rich subpopulation correlated with the number of lamellopodial haemocytes. The ratio of haemocytes number in two subpopulations was about 1:1 in most scallops, but there were two individuals with a significantly decreased number (~30 and ~15%) of the actin-rich lamellopodial cells.

In this way, the haemocytes polyploidization in *S. swifti* seems to be associated with a partial loss of functional capacity of the cells. It may indicate the malignant nature of this genomic instability and may reflect the development of hemic neoplasia in scallops of the Vostok Bay.

Phylogenetic methods for detecting natural selection in genomic sequences

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The goal of my talk is to highlight some of recently developed phylogenetic methods that provide a powerful toolkit for genetic diversity studies. Phylogenetic methods help to disentangle the molecular evolutionary history of homologous sequences and to relate changes at the molecular level to functional or phenotypic changes. Genomic sequences are shaped by a complex interplay of various biological factors, among which natural selection plays an important role. Phylogenetic Markov models of codon evolution are among the most successful methods for evaluating selection signature and have recently been implemented for phylogeny inference (Gil et al 2013; Anisimova 2015). Selection operates in many different ways, not only on the level of point mutations but also on genomic features. For example, tandem repeats are found abundantly in genomic sequences and are known to be involved in resistance and disease. Selection clearly operates also on tandem repeats. Therefore I will also review some of the recent methods we developed for analyzing tandem repeats (Anisimova et al 2015).

Evolutionary genetics in biodiversity conservation and monitoring

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Biodiversity represents a dynamic result of the past and ongoing evolutionary genetic processes. Consequently, the fundamental evolutionary genetic principles are critical to follow to prevent the fatal intrinsic changes in natural populations underlying a major loss of biodiversity. Indeed, there is a mounting evidence that anthropogenic perturbations (including harvesting and other business activities) have led to the detrimental genetic changes, which increase the extinction risks especially for overexploited populations. In this report, I highlight some important evolutionary genetic aspects relevant to biodiversity conservation and monitoring.

Intraspecific variability is the basic genetic resource ubiquitously represented in natural populations. The data on the king crab *Paralithodes camtschaticus* and the sea urchin *Strongylocentrotus intermedius* clarify why an intraspecific variability analysis is indispensable and should be integrated in biodiversity conservation and monitoring programs. The negative fitness consequences due to inbreeding and outbreeding depression are demonstrated with the examples from different groups of marine fishes and invertebrates.

Reliable species identification and phylogenetic reconstructions are crucial for biodiversity conservation and monitoring. The problem is challenging for marine biota characterized by a high abundance of morphological variants with uncertain taxonomic status. New approaches, based on the multi-locus analysis and complete mitochondrial genome sequencing, illustrate how the long-lasting taxonomical problems can be resolved in some complex groups of marine algae, lampreys, and fishes.

Human-mediated hybridization poses a major threat to aquatic biodiversity worldwide, leading to degradation of locally adapted genetic pools. An example of anthropogenic hybridization between salmonid fishes, such as taimen *Hucho taimen* and two lenok subspecies *Brachymystax lenok* and *B. lenok tsinlingensis*, is described. The phenomenon of reticulate evolution is exemplified by the data on historical hybridization between distantly related brown algae belonging to the orders *Ectocarpales* and *Laminariales*. Finally, the consequence of hybridization for the invasive species evolution is considered.

The data on microbiome content and structure of the brown alga *Saccharina japonica* and the sea urchin *S. intermedius* are presented to emphasize a potential role of symbiotic bacteria as a causative factor of morphological divergence and epigenetic speciation in the sea. The possible speciation reversal due to anthropogenic contaminations in populated areas of the Sea of Japan coast as well as the importance of microbiome conservation and monitoring are also discussed.

The evolutionary genetic methodology is a necessary prerequisite for successful practical realization of the long-term challenges in biodiversity conservation. Genetic and epigenetic monitoring offers an important and reliable approach to reveal diverse detrimental transformations and threat from anthropogenic impact to the genetic integrity of natural populations. The potential results are promising. However, taking in account that the human species represents the most powerful evolutionary force on Earth, the technical and scientific advances may not be enough efficient for protection of biodiversity without the complementary advances in human ethical and cultural attitudes toward Nature.

Leukocyte formula of scorpion fish (*Scorpaena porcus* L.) as a biomarker of antropogenic pollution in the Black Sea coastal waters

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It is known many species of flora and fauna in the coastal zone in conditions of anthropogenic pressure high intensity home to. These aquatic organisms have a certain range of adaptive opportunities to exist in extreme environmental conditions of hypoxia, anoxia, temperature fluctuations and salinity at high concentrations of pollutants due to the peculiarities of biology. Features of physiological and biochemical processes of these organisms during adaptation to environmental conditions can act as biomarkers. These species can be used as indicator species for assessing the status of the water environment. Typical representative of such species in the Black sea is scorpion fish *Scorpaena porcus* L. Scorpion fish is a sluggish predator with reduced metabolism and lives in littoral and sublittoral zone. Very important that scorpion fish always exists at the bottom, where many pollutants concentrate in the bottom layers and bottom sediments. The blood parameters are highly specific for each species and fluctuate in a fairly narrow range. It allows to use blood parameters as markers of physiological and pathological processes at the organismal and ecosystem level. So, White blood cell counts (WBC) is a universal signal indicator of the general nonspecific adaptive reactions, and the leukocyte number characterizes the physiological status of the individual not only at the organismal level, but can serve as an indicator of species functionality and for the broader systematic categories. The fish WBC study is an important element in monitoring of physiological state in fish practice and in comprehensive studies of populations under natural conditions. The aim of the study to determine scorpion fish WBC from areas with different pollution. Blood was sampled from the tail vein, and blood smears were dried and analyzed. The scorpion fish WBC changes in the bays with the different pollution were founded. The neutrophils and lymphocytes had most noticeable number changes. Increasing the level of contamination involved the lymphocyte number increasing and segmented neutrophil number decreasing ($p \leq 0.05$). Thus, lymphocytes and segmented neutrophils subtly react to changing environments and it can be used to judge the approximate level of ecological well-being and general status of the area. We offer simple, efficient and representative monitoring method of waters contamination by analyzing the number of lymphocytes and segmented neutrophils.

Contribution of symbiotic dinoflagellate (zooxanthellae) in lipid and fatty acid compositions of the soft coral *Capnella* sp.

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Coral reef ecosystem is very important for tropical shallow waters. One of the major threats for the coral reefs is increases of sea surface temperature due to a global warming effect. Most coral species are a symbiotic association between photosynthetic microalgae (zooxanthellae, dinoflagellates of the genus *Symbiodinium*) and the animal host. Under stresses, such as increases of sea surface temperature and ultraviolet radiation, algal endosymbionts are expelled and animal host die slowly. This phenomenon is known as coral bleaching. By photosynthesis, microalgae provide nutrients to coral colonies necessary for vital activity of a symbiotic organism. Lipids are the important part of these nutrients.

In this work, lipid and fatty acid (FA) compositions of the soft coral *Capnella* sp. were identified and compared with the data published for other coral species. Markers lipid classes and polyunsaturated FAs (PUFAs) of each side of symbiotic association of the coral were described. Glycolipids and monoalkyldiacylglycerols were characteristic for zooxanthellae and coral polyp tissues, respectively. The level of PUFA markers of zooxanthellae, namely 18:3n-6 and 18:4n-3, was determined. Two very-long-chain tetracosapolyenoic fatty acids (24:5n-6 and 24:6n-3) were the biomarkers of soft coral tissues. In several coral species from different taxonomic groups, we determined the chemical structure and distribution of wax esters, which is one of reserve lipid classes transferring in marine food webs.

On the whole, zooxanthellae make substantial contribution to the composition of different lipid classes and fatty acids of corals. The features of lipid class and PUFA compositions of the soft corals, which are defined by the presence of the symbionts, were highlight. The contribution of each side of the symbiotic association in total lipids of whole coral colonies can be estimated on the base of the levels of lipid and PUFA markers.

Improving the technology of reproduction Masu salmon

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Masu salmon – the most valuable in the food respect to Pacific salmon in the Primorsky territory. Of course the desire to increase their numbers through artificial reproduction. Local hatcheries were trying to artificially reproduce, but the great success was not achieved mainly because of its biological characteristics.

Masu salmon – salmon with a long freshwater period of life. About 50% of the males and a small number of Mature females in the river without going to sea. Males maturing in the second or third year, do not reach marketable size, and therefore their cultivation in hatcheries is becoming a waste of funds.

It is believed that early-ripening males – let's call them the accelerates are not able to adapt to seawater. To test this hypothesis, we conducted a series of experiments. Because akseleratov in fresh water grow faster than juvenile females and males, there is a possibility to separate them. When reaching the average weight of juveniles of about 5 g., there are instances 10–15, They will all accelerates. Larger males we were placed in separate pools. When recruited, a quarter of the population gradually began to increase the salinity of the water. Our results showed that under a certain mode of increasing salinity akseleratov able to adapt to seawater. Translated to sea water males on experimental station mariculture "Glazkovka" were transferred to sea cages, where they grew well. In the process of adaptation to sea water was carried out histological examination of the gonads showed a gradual "rejuvenation".

The study of scales of juvenile masu grown in natural conditions and cultivated at elevated temperatures showed a real opportunity to differentiate these salmon on the temperature of cultivation.

On the basis of the conducted research it is possible to create new technologies of reproduction Sims and other salmon with a prolonged period of life.

Current state of *Strongylocentrotus pallidus* resources in the western Bering Sea

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Pale or white sea urchin *Strongylocentrotus pallidus* is among the most common bottom invertebrate species in the Bering Sea. These sea urchins are found down to 330 m depth, and form relatively dense commercial aggregations below 100 m depth (Bazhin, Stepanov, 2012). There is no market for this species today, because of the lack of tradition to use its gonads for food. Nevertheless, taking into account an increasing variety of seafood products on the market in Russia and beyond, this sea urchin may be of potential interest for commercial fishery; therefore, reliable information on its resources and biology is needed. Major objective of this research was to analyze *S. pallidus* size structure and abundance in the western Bering Sea.

Material for this research was collected in bottom trawl survey at depths 19–310 m during May 23 – June 29, 2016. The following areas were investigated: Korf and Olutorskyi bays, Koryak coast, Karaginskyi Bay and the so-called southwestern region, a vast area to the south of Korf Bay and to the west of Olutorskyi Bay. A total of 200 trawl hauls were made, and total research area was 80507 km². All captured Pale sea urchins were counted and weighted, and shell diameter for most of them was measured to the nearest 1 mm.

The largest *S. pallidus* aggregation (based on area and number of positive hauls) was found in the northernmost part of the research area off the Koryak coast, and that aggregation occupied 27742 km². Quantitative parameters for the aggregation were also the highest: average biomass was 1184 kg/km² (Fig. 1) and average density 96788 individuals/km². The median biomass was 160 kg/km² and median density 11278 individuals/km². Total stock biomass was assessed at 43.4 thousand metric tons.

In other parts of the research area, parameters of Pale sea urchin abundance were an order of magnitude lower. In Olutorskyi Bay, sea urchin aggregation occupied 7984 km², average biomass was 322 kg/km² (median 19 kg/km²), density 24969 individuals/km² (median 655 individuals/km²), and stock biomass 2.2 thousand metric tons. In the southwestern region, the area occupied by the sea urchin aggregation was the lowest, of 1884 km², average biomass was 417 kg/km², density 17944 individuals/km², and stock biomass 1 thousand tons. In Karaginskyi Bay, the sea urchins occupied relatively large area, of 11361 km², however, parameters of abundance were low: average biomass was 44 kg/km² (median 30.6 kg/km²), density 1175 individuals/km² (median 628 individuals/km²), and stock biomass 0.45 thousand tons. The lowest sea urchin abundance was in Korf Bay, where average biomass was 18 kg/km², density 251 individuals/km², and stock biomass was assessed at only 0.04 thousand tons in the area of 2031 km².

In Korf Bay, Pale sea urchins had the largest average shell size, of 56 mm, and animals with shells 65–70 mm in diameter formed a modal group (44%) (Fig. 2). In Karaginskyi Bay, the average

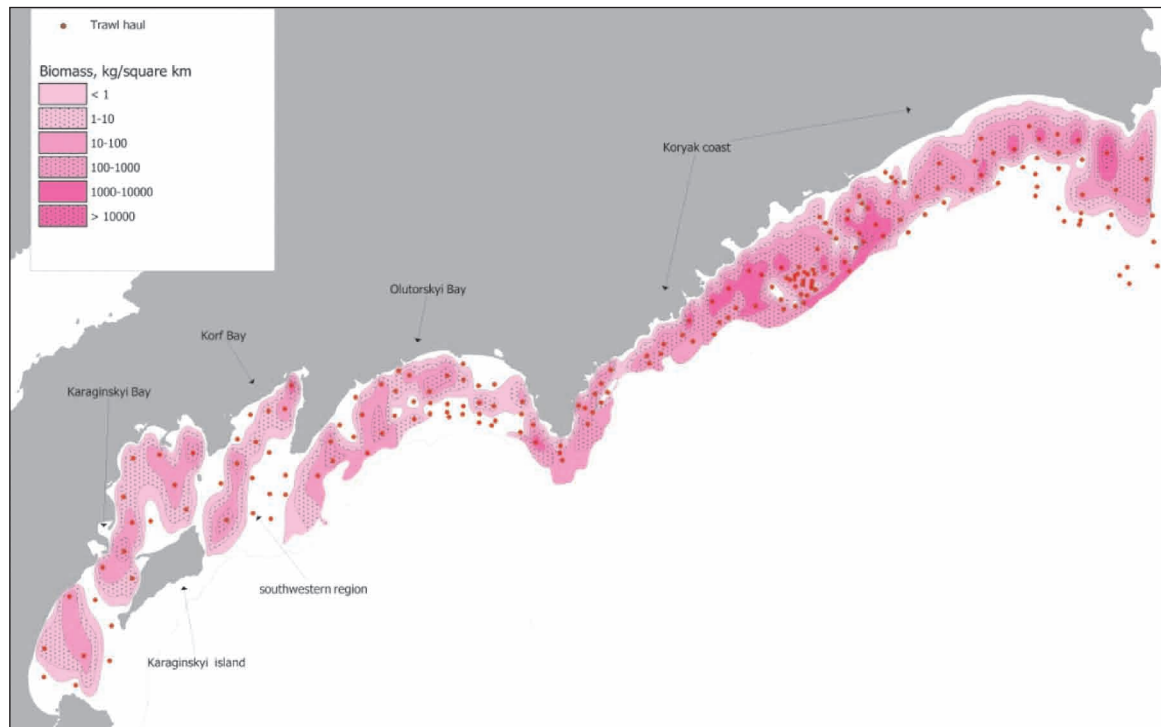


Fig. 1. Biomass spatial distribution (km²) of *S. pallidus* in the western Bering Sea.

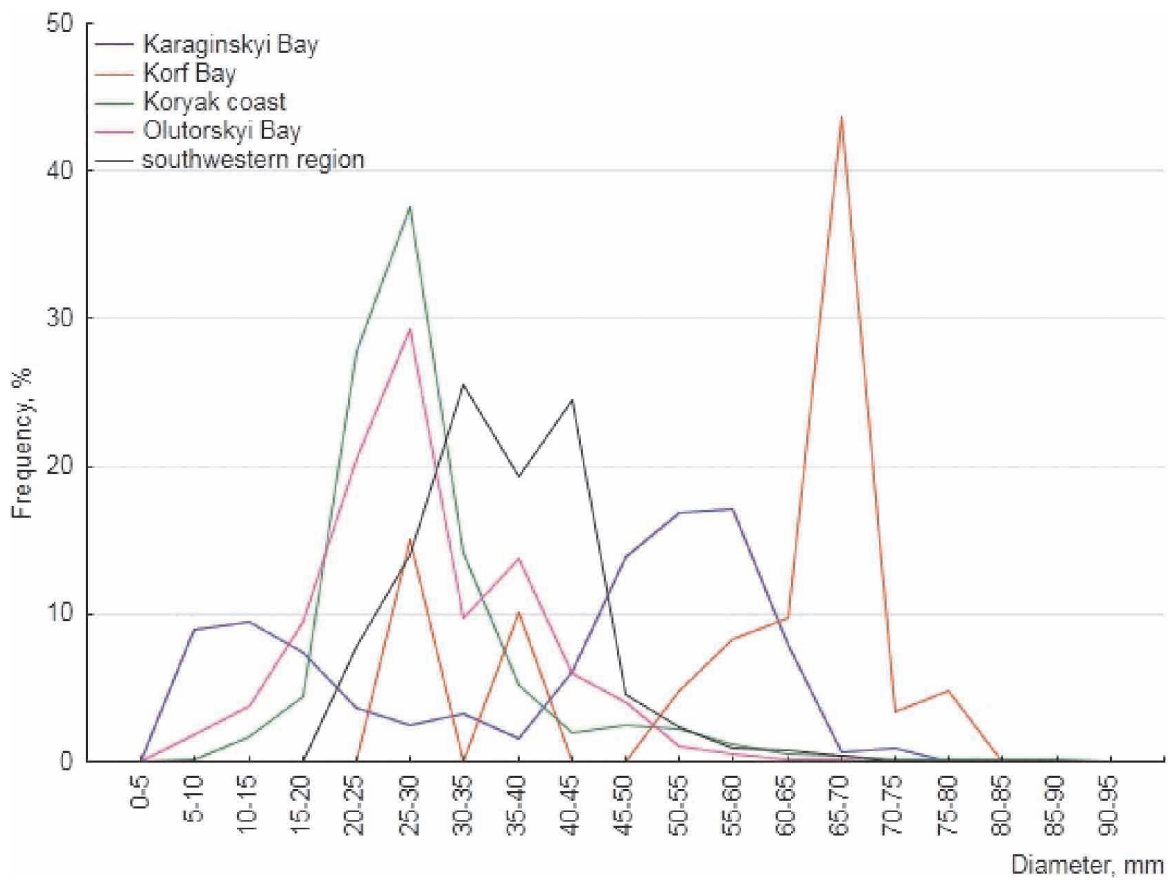


Fig. 2. Size composition of *S. pallidus* in the western Bering Sea.

shell size of the sea urchins was 39 mm, and modal groups were 5–20 mm (25%) and 45–60 mm (47%). In the southwestern region, the average shell size was 36 mm with a modal group 30–45 mm (68%). In Olutorskyi Bay, the average shell size was 28 mm with a modal group 15–30 mm (62%). Off the Koryak coast, the average shell size was 29 mm with a modal group 20–30 mm (66%). Relatively small size of sea urchins in Olutorskyi Bay and off the Koryak coast presumably reflects low growth rate due to less favorable forage conditions and hydrological features.

Summing up, total stock biomass of *S. pallidus* in the research area was estimated at 47 thousand metric tons, and total number 2689027 thousand individuals. The largest aggregations of Pale sea urchins were located off the Koryak coast, and the lowest abundance of these echinoderms was registered in Korf Bay.

Deep-sea nemerteans of the VEMA-Transit expedition

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Approximately 30 nemertean specimens were obtained from the samples collected with an epibenthic sledge during the VEMA-Transit expedition (2014–2015) to the Vema Fracture Zone in the equatorial Atlantic Ocean. This number is substantially smaller than in the previous deep-sea expeditions (SoJaBio, KuramBioI, KuramBioII, and SokhoBio). Members of only two nemertean groups were identified: tubulanid palaeonemerteans (three species) and eumonostiliferous hoplonemerteans (four species). A molecular phylogenetic analysis based on five genes (16S, 18S, 28S, COI, and H3) indicated that three tubulanid species formed a monophyletic group with the only known abyssal tubulanid (*Tubulanidae* gen. sp. IZ 45557) from the abyssal plain adjacent to the Kuril–Kamchatka Trench (depth 5225 m), but the monostiliferan species belonged to three different clades. Two new monostiliferous nemerteans, *Abyssonemertes kajiharai* gen. et sp. nov. and *Nemertovema hadalis* gen. et sp. nov., were described; the latter species is the deepest known nemertean collected in the Puerto Rico Trench from the depth of 8336–8339 m. *Abyssonemertes kajiharai* is distinguished from the other monostiliferous nemerteans by the unusual ventral arrangement of ovaries.

This research was supported by the Russian Foundation for Basic Research (grant No. 15-04-01933) and the Grant of Russian Science Foundation (grant No. 14-50-00034).

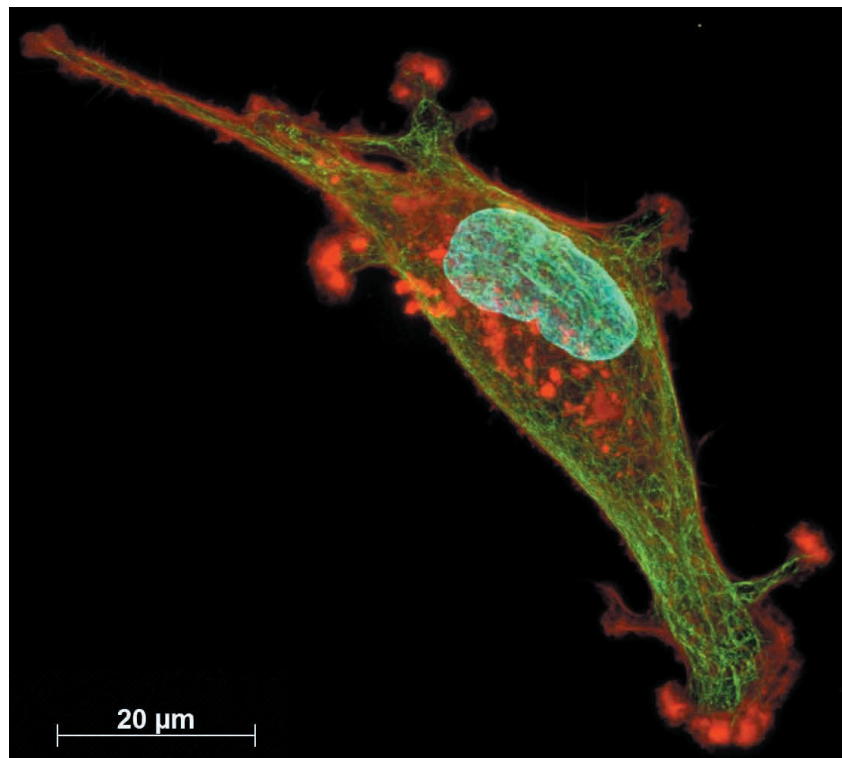
Cryobanking biological material of marine mammals: purpose, methods and successes

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The extinction of some mammal species is natural and inevitable evolutionary process, but now it happens more often than speciation mainly due to human actions: destroying mammal habitat, industrialization, anthropogenic pollution, competition with mammals, excessive hunt, and others. The conservation of every separate species is required to maintain the biodiversity, because eliminating any of them may lead to malfunction of whole ecosystems. Gamete cryobanking widens the interchange potential within and between the populations, increasing genetic diversity and slowing inbreeding. However, taking gametes from marine mammal is highly difficult and possible either from well-trained animal in dolphinarium or aquarium, or from a recently fallen animal. Cryopreservation of sperm usually is simple, but successful conservation efforts of oocytes are few. One of the ways to restore populations of rare marine mammals is reprogramming genome of somatic cells, for example skin fibroblasts, into induced pluripotent stem cells. If the artificial insemination of rare species is needed, these stem cells obtained from both female and male could be differentiated into functional oocytes and sperm cells for the following obtaining of new specimens. The less invasive approach of taking



The morphology and distribution of some cytoskeletal proteins in skin fibroblast derived from the walrus *Odobenus rosmarus*. The specimens were imaged using a confocal laser scanning microscope Zeiss LSM 780. Tubulin is stained (tubulin antibodies) in green, and actin is stained (TRITC-labelled phalloidin) in red. The nuclei (blue) are labeled with DAPI. Scale bar 20 μm.

“live” biological material from marine mammal is skin biopsy, making it the best source of cell isolation. Cells of rare species can help to determine multiple parameters of both a particular animal (sex, physiological peculiarities, chromosomal and DNA polymorphism) and its population (phylogenetic relationships, social structure). The foundation of cryobanks of cells cultures and tissues may become a basis for preservation of genetic diversity of rare and endangered marine mammals. But obtaining these cultures are labor- and time-consuming because of difficulties in finding and taking tissues of live animals under protection of international laws, therefore the successful attempts of cell culture isolation from marine mammals are not numerous. Correctly frozen sample contains all “data” for genomic, genetic, proteomic and other analysis, it could be stored for decades, transported worldwide and recovered anytime even from a small quantity of thawed cells without the necessity of taking tissue from an animal. Conservation of biological material from each specimen of marine mammal is crucial to have a constant access to it regardless of the animal fate (see Figure).

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Sea stars of the genus *Henricia* Gray, 1840 (Echinodermata: Asteroidea) from the northwestern Sea of Japan

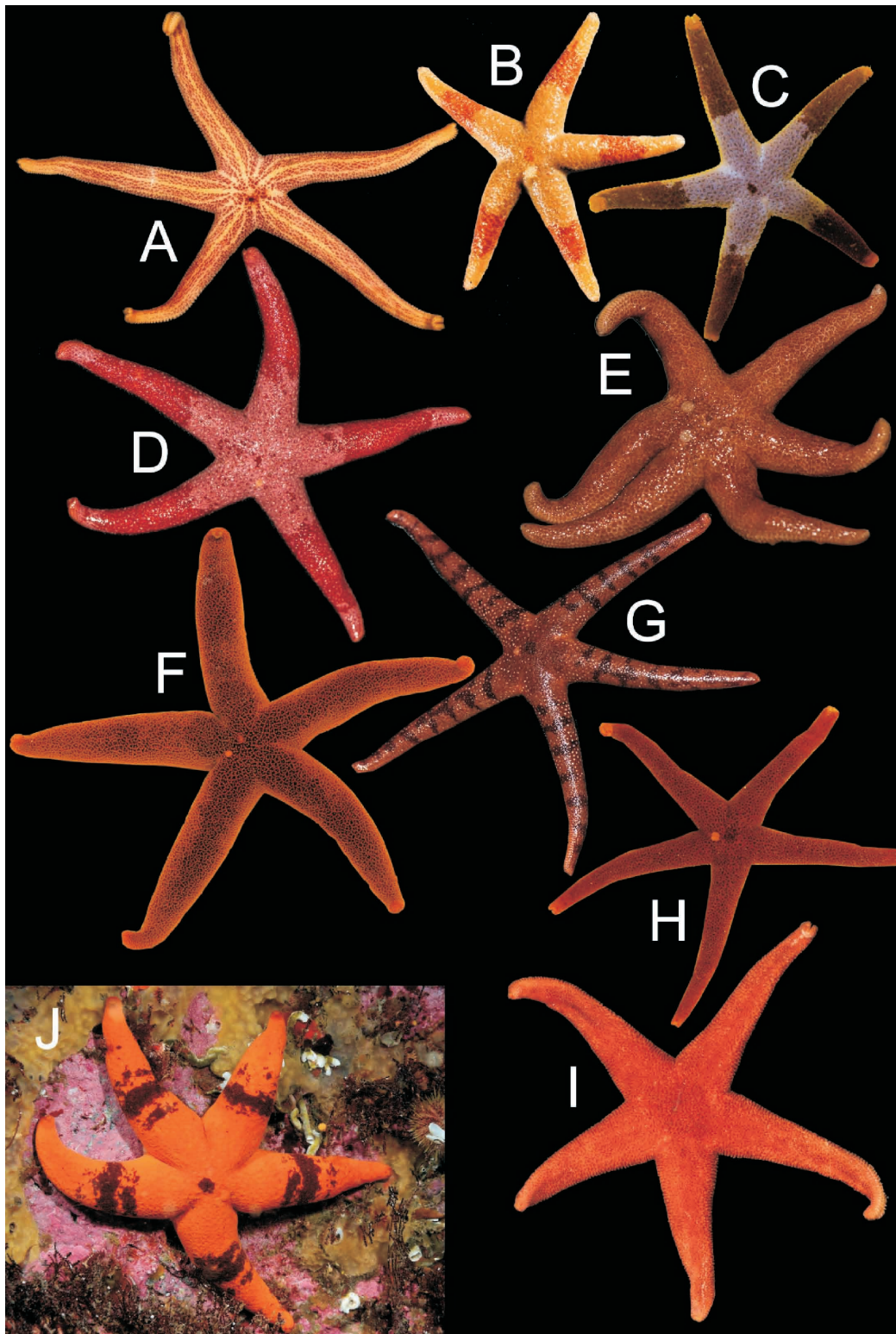
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Sea stars of the genus *Henricia* Gray, 1840 belonging to the family Echinasteridae (Asteroidea, Spinulosida) are a group of organisms with poorly developed systematics despite their wide distribution and abundance in world seas, especially in the northern Pacific (Verrill, 1909, 1914; Fisher, 1911, 1928, 1930; Hayashi, 1940; Shin, Rho, 1996; Djakonov, 1961; Clark et al., 2015). To date, the fauna of echinasterids of Russian Pacific seas totals about 30 nominal species belonging to the genera *Henricia* and *Aleutihenricia* (Smirnov, 2013; Chichvarkhin, 2017; Chichvarkhin et al., 2017). This number is based mainly on Djakonov's publications (1949, 1950, 1958, 1961) and taxonomical rearrangements by Clark and Jewett (2010) because of a lack of recent studies on echinasterid fauna and systematics in this region. Therefore, actual species abundance in Russian Pacific is still in need of study.

In this study, we have explored three coastal locations: Vostok Bay, Kievka Bay, and Rudnaya Bay. Also, museum collections were studied. Totally, we confirmed occurrence of the 11 species. One of them (*H. pachyderma*) was firstly reported for this area. Three species (*H. alexeyi*, *H. djakonovi*, and *Henricia* sp.) are new for science. *H. pacifica* is firstly reported for Russian continental shore. We also firstly report amphiboreal *H. oculata* from the Russian waters, which was confused with its junior synonym *H. aspera robusta*. We refer *H. reniessa asiatica* as *H. asiatica* because of its very large ray length and peculiar spine morphology. Also, a population of this relatively deep-water species is geographically isolated from *H. reniessa* and similar morphs known from the northern Japan, Sakhalin and Kurile islands. The only doubtful record in our list is *H. cf. leviuscula*: a single obtained specimen of this form resembles *H. hayashii* but its spines are crown-shaped as in *H. leviuscula* according to Djakonov (1961).

Most species recorded in this publication are not abundant. Although, *H. djakonovi* is the most abundant one across investigated area: about 80% of examined individuals belong to this species. The second abundant species (about 10% of collected specimens) is *H. pachyderma*. Also, *H. hayashii* was abundant in Rudnaya Bay, while it was very rare in Vostok Bay and absent in Kievka Bay. Abundance of the first two species can be explained by their tolerance to decreased salinity. This is why all species except *H. djakonovi* were not found in inner part of Vostok Bay, while this species was abundant in its inner part too. All studied species are associated with the sponges. But *H. djakonovi* can be also associated with the aggregations of the mussels. Their feeding remains unknown but a hypothesis by Rasmussen (1965) about phytoplankton feeding using water currents generated by the sponges and the bivalves seems plausible.



Sea stars *Henricia*: **A** – *Henricia* sp., Rudnaya Bay; **B** – *H. alexeyi*, Rudnaya Bay; **C** – *H. alexeyi*, Vostok Bay; **D** – *H. pacifica*, Vostok Bay; **E** – *H. pachyderma*, Vostok Bay; **F** – *H. granulifera*, Vostok Bay; **G** – *H. djakonovi*, Vostok Bay; **H** – *H. hayashii*, Vostok Bay; **I** – *H. densispina*, Vostok Bay; **J** – *H. oculata*, Commander Islands.

The following species are confirmed for studied region:

1. *Henricia alexeyi* Chichvarkhin et Chichvarkhina, 2017 (see Figure, B, C)
2. *Henricia pacifica* Hayashi, 1940 (see Figure, D)
3. *Henricia pachyderma* Hayashi, 1940 (see Figure, E)
4. *Henricia oculata* (Pennant, 1777) (see Figure, J)
5. *Henricia granulifera* Djakonov, 1958 (see Figure, F)
6. *Henricia djakonovi* Chichvarkhin, 2017 (see Figure, G)
7. *Henricia hayashii* Djakonov, 1961 (see Figure, H)
8. *Henricia asiatica* Djakonov, 1958
9. *Henricia densispina* (Sladen, 1878) (see Figure, I)
10. *Henricia* sp. (see Figure, A)
11. *Henricia* cf. *leviuscula* (Stimpson, 1857)

Population dynamics of a high latitude coral *Alveopora japonica* Eguchi (1968) in Jeju Island, off the south coast of Korea

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While coral reefs are facing severe challenges from a variety of natural and anthropogenic stresses, a coral species, *Alveopora japonica* Eguchi (1968), is experiencing a population explosion around Jeju Island, Republic of Korea (Fig. 1). The present study provides the first ecological data



Fig. 1. *Alveopora japonica* Eguchi (1968).

on this high-latitude coral species in Jeju Island. Colony abundance, size-frequency distributions, age and growth rates of two *A. japonica* populations located in the northwestern and southeastern coasts of Jeju Island were analyzed and compared to determine eventual spatial heterogeneity

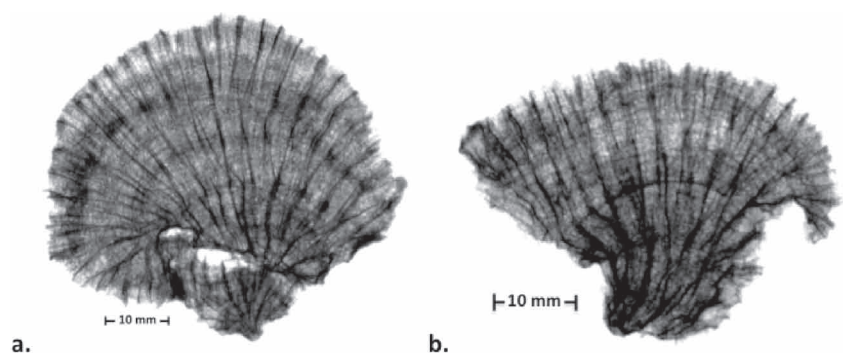


Fig. 2. X-radiograph of *Alveopora japonica* slabs from northern Jeju Island.

(Figs. 2, 3). The results showed that *A. japonica* is a small species characterized by a mean colony size of 30 cm², slow growth-rate (4.8 mm yr⁻¹), and a short lifespan of ~10 years. *A. japonica* revealed a dense population of 120 colonies/m² in both sites. Normally distributed colony size at both sites reflected a healthy status and indicated a local stability with

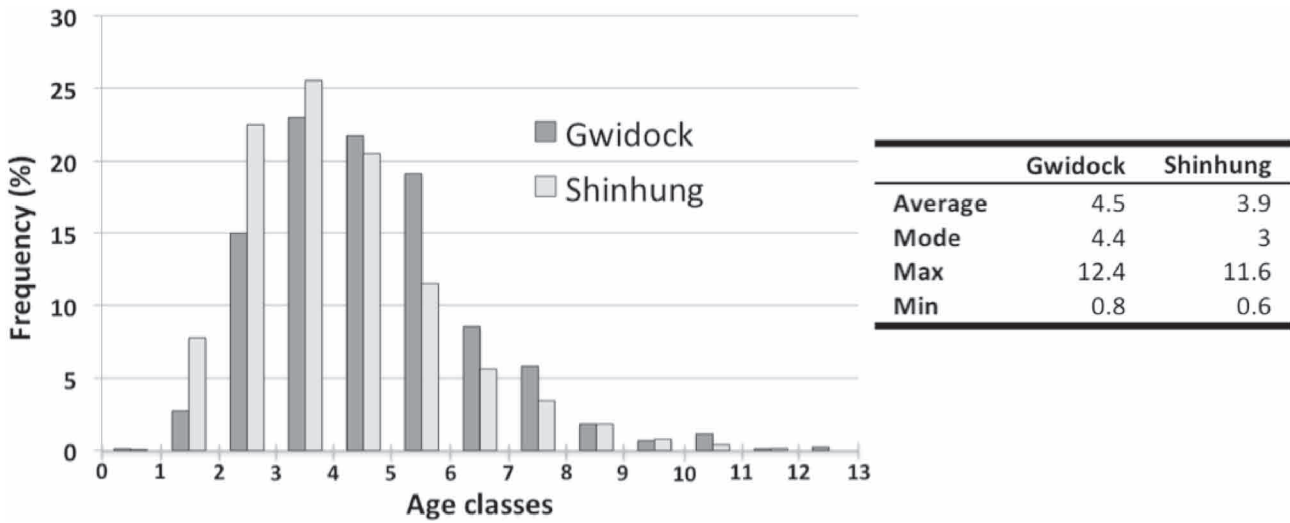


Fig. 3. Estimated age distribution of *Alveopora japonica* populations from the northern (Gwidock) and southern (Shinhung) Jeju Island. *A. japonica* populations were surveyed in 2011.

a stationary size structure allowing the population maintenance over time. However, the differences in distribution parameters of two populations reflected probable temporal variations in recruitment or/and environmental variations between two sites. These new insights about *A. japonica* population ecology provide important tools for understanding life-history processes of this coral species and its increasing abundance.

Influence of plasmalogens (1-*O*-alkyl-glycerols) on the body of elderly people

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Plasmalogens play an important role in the regulation of lipid metabolism. Plasmalogens is a special group of phospholipids. Precursors of plasmalogens are 1-*O*-alkyl-glycerol (AG) in the human body. In the world of medical practice, their use for the prevention of development and in the treatment of immunodeficiency and cancer diseases, inflammation of various etiologies, infectious, cardiovascular and neurodegenerative diseases. The lipids of the digestive gland of the commander squid *Berryteuthis magister* are a rich source of AG. On the basis of these lipids were derived biologically active additive to food "Lipidomarin".

The presented research was aimed at assessment of the influence of AG on the protective status and decrease in the risk of development of pathological processes in human body. Studies were conducted in a group of volunteers aged 60±3 years. A source of AG was a biologically active additive, the dietary supplement "Lipidomarin". "Lipidomarin" was given to the volunteers in a daily dose of 500 mg during 90 days. The physiological action of AG was assessed by the method of functional-topical diagnostics of a human body with the use of "RS MEGI-01", included in the Directory of the Ministry of Health of the Russian Federation in 2003, "Diagnostic and Health-Improving Technologies of Restorative Medicine", the spectral analyzer of rhythmic brain activity. The spectral analysis of total bioelectric activity of human brain has detected the early signs of disorders in neurotrophic vegetative regulation of the internal organs as well as alterations of this regulation under an effect of biologically active substances.

The evaluation criteria were: vegetative index (VI) – characterizes the balance of ergotropic and trophotropic central influences that control predominantly adrenergic and cholinergic actuators, the stress index (SI) – characterizes the dominance of the functional system, the index of individual health (IIH) – the stability of the biosystem at a significant increase Degree of tension, leading to a decrease in functional resources, the decentralization index (DI) – characterizes the degree of neurodystrophy (tissue exit from the influence of the central and autonomic nervous system)

The analysis of the obtained results showed that after taking the dietary supplements to food "Lipidomarin" the whole group of volunteers showed a general decrease in all indices. A significant decrease in DI was registered, a decrease of 6.7 times in comparison with the baseline before taking dietary supplements to food "Lipidomarin". Admission reduces the risk of neurodegenerative disorders in the elderly. The additive can be recommended as a potential enrichment ingredient for dietary preventive foods.

The study was supported by the Russian Scientific Fund (project No. 14-50-00034).

Optical methods of monitoring dynamics marine biological resources

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It is known that dynamics of number or biomass of phytoplankton is defined by the photosynthesis processes consisting in creation of organic substance from mineral compounds. The solar energy is played the main role in this dynamics. In this regard the main attention was paid to seasonal, annual and interannual variations in case of a study of dynamics primary producer of organic substance in different reservoirs. Thus practically no attention was paid to small-scale processes which can have significant effect on dynamics of the chain “phytoplankton-zooplanktons-fishes-marine animals”. To study the dynamics of this chain, a measuring complex is created, consisting of a laser measuring instrument of hydrosphere pressure variations, fluorimeter, and underwater video monitoring system. In addition the complex is equipped with power supply and telecommunication for information transfer from places of setting in coastal laboratory locations, a multi-bit analog-to-digital converter (ADC), system of exact time, the computer designed for primary processing and recording of the experimental data on the hard drives. Depending on the basic membrane parameters of the laser measuring instrument of hydrosphere pressure variations and the accuracy of measuring the change in the path difference in the interferometer arms is equal to 1 μm , accuracy measurements of hydrosphere pressure variations is in the range of 1 mPa to 1 μPa .

The fluorimeter C3 is fixed in a frame near the laser measuring instrument of hydrosphere pressure variations. The submersible fluorimeter C3, manufactured by the Turner Designs company is completed with two photosensitive sensors in vivo for measuring of a luminescence of phytoplankton chlorophyll in blue 460 nm and red 635 nm radiation spectra. The range of blue radiation allows fixing the concentration in the range of 0.025–500 mkg/l. The range of red radiation is more than 500 mkg/l. The device is also equipped with sensors of pressure and temperature. The durable case is made of acetyl Delrin that allows working in the hostile marine environment. The surfaces of the case near optical sensors are equipped with copper inserts to reduce biological contamination of photosensitive elements. The weight of the device is 1.64 kg, length is 23 cm, and diameter is 10 cm, the range of temperatures from -2°C to 50°C . Depth of immersion is up to 600 m. Maximum discretization of data acquisition is 1 Hz. The fluorimeter has to output digital data in ASCII format, or receipt of analog data in the presence of an appropriate adapter. All acquired information of measuring installations of a complex on cable lines arrives in the coastal laboratory building where after preliminary processing is recorded in the created experimental database.

The complex was installed at a depth of 8 m in the Vityaz Bay, Posyet Gulf of the Sea of Japan. In processing the synchronous experimental data of the laser measuring instrument of hydrosphere pressure variations and the fluorimeter in the range of wind sea waves it is found, that oscillations of fluorimeter indications correlate with change of intensity of wind disturbance, with daily and semi-diurnal wrong tide, with seiche of the Vityaz Bay.

Naphtaquinoid pigments in clypester sand dollars

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Sea urchins are very promising source of secondary metabolites, in particular naphthoquinones, used for pharmaceutical, food and other commercial purposes. The presence of naphthoquinoid pigments is one of the characteristic biochemical synapomorphies of sea urchins, caused by genes encoding the polyketide synthase enzymes, which are necessary for the biosynthesis of spinochromes. In regular globular sea urchins, for example in *Strongylocentrotus purpuratus*, naphthoquinoid pigments appear at the blastula stage. The pigment cells of epidermis and gametes for Clypesteroidae sand dollar *Scaphechinus mirabilis* were isolated with help an enzymatic method and naphtaquinone pigments of their alcohol extract were studied. It was found that naphtaquinone pigments present in the pigment cells that imbedded into their jelly coat of the oocytes but not in the cytoplasm of unfertilized eggs. Mass spectra laser desorption ionization showed that pigments of the jelly coat are composed of spinochromes E and D, whereas echinochrome A and spinochrome D were measured in the pigment cells isolated from epidermis of adult urchins. Spinochromes of the eggs deprived jelly coat were found. It is known that the sea urchin fertilization accompanied by oxidative burst associated with the production of hydrogen peroxide from molecular oxygen by quinone and naphthoquinone-dependent oxidase Udx1. Because of the fact lacking quinones in eggs cytoplasm, then biosynthesis of hydrogen peroxide to stimulate embryogenesis apparently used the spinochrome E water soluble and penetrating by diffusion into the cytoplasm. The dominant in epidermal cells echinochrome A is not soluble in water and, presumably, immobilized by hydrophobic anchor ethyl group in the surface cells of the epidermis of the animal.

Genetic analysis of gray whales during 2012–2015 near of Sakhalin Island

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Gray whales (*Eschrichtius robustus*) live in the northern Pacific and regularly migrate over seasons. There are two known gray whale schools: Sakhalin and Chukotka-Californian. The former winters and spawns near Korea and South Japan, feeding in summer in the Sea of Okhotsk on the northeastern Sakhalin shelf. This is a small group consisting of about 180 whales over one year old (WGWAP-16, 2015). The latter winters and spawns in the Gulf of California, feeding in summer in the Chukchi Sea, Bering Sea, and Beaufort Sea and, on rare occasions, entering the East Siberian Sea all the way to Nolde Bay and moving to the east to Cape Barrow. This is a larger school consisting of more than 20,000 animals; its size is being gradually restored. Based on certain biological features (specifically the population dynamics, with the Californian population increasing in size over the years since the 1947 whaling ban from a few hundred to more than twenty thousand, while the Sakhalin whale population was reduced to a few hundred), we can assume that these two populations are largely isolated. Nevertheless, only genetic data can provide a definitive answer to the question regarding the existence of population structure and the scale of migrations between gray whale schools. Several genetic methods are used to determine the population-genetic structure of species include: analysis of nuclear marker and mitochondrial DNA variability.

Based on the available capabilities (namely the small number of test specimens provided), we continued the genetic testing of gray whale samples via comparative analysis of nucleotides of combined mitochondrial DNA (mtDNA) gene fragments of the 1-st subunit of cytochrome oxidase I (*COI*), cytochrome b (*Cytb*), control region (*CR*), and a fragment of the NADH dehydrogenase subunit 2 (*ND2*). The skin tissues of gray whales sampled by the remote biopsy method in accordance with permission of the Russian Federal Supervisory Natural Resources Management Service (Rosprirodnadzor) along the coast of Sakhalin Island, Pil'tun Bay in August-September of 2012 (16 animals), 2013 (8 animals), 2014 (28 animals), and 2015 (9 animals). In total, 21 combined haplotypes were identified in the 2012–2015 samples (61 animals) for the combined subunits of *COI+CytB+CR+ND2*. An analysis of molecular variability computed from a comparison of mtDNA Control Region (*CR*) sequences did not show any differentiation between the samples of the Gray Whale from North Pacific (Sakhalin) in 2012–2015 ($p>0.05$). A comparison of the samples taken from the same region in different years, in two cases, identified reliably meaningful differences. The Exact test of pairwise differentiation between all specimens revealed reliably meaningful differences between the 2015 and 2012 samples, and also between the 2012 samples and the 2014 samples. Three animals (2015) were identified in the 2014 sample, some animals (2012–2014) were identified in all three samples. This

fact may indicate the affinity of the 2012–2015 samples as population. Based on the obtained data it could be supposed that in different years this area is selected as the feeding area by animals from different reproduction areas related to each other, or that these groups come here to feed in the same year, but at different times. Thus, our data indicate that the population of Sakhalin gray whales is not a genetically separate population. The thesis that the Sakhalin population is self-replicating is also countered by the relatively high genetic variability resulting from a large number of unique haplotypes which occur in each of the samples. Besides, new haplotypes appear in samples every year which also confirms their genetic heterogeneity.

Seasonal variation of microzooplankton in the Zuari estuary, West coast of India

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We report here the seasonal variation of microzooplankton (MZP) in the Zuari estuary located along the Central West coast of India in relation to certain physicochemical and biological parameters. MZP communities were studied for one year (January to December 2013) covering the pre-monsoon, monsoon and the postmonsoon seasons. A total of 26 species of 14 genera were identified, which include loricate ciliates (13 species of 8 genera), aloricate ciliates (3 species of 2 genera), heterotrophic dinoflagellates (10 species of 4 genera) and copepod nauplii. Among these, five species of loricate ciliates (*Dictyocysta* sp., *Leptotintinnus nordquisti*, *Stenosemella* sp., *Tintinnopsis beroidea* and *Tintinnopsis uryguayensis*) and three species of heterotrophic dinoflagellates (*Proto-peridinium breve*, *Proto-peridinium granii* and *Proto-peridinium latistriatum*) were dominant in this estuarine system through out the year. The MZP abundance was high during the premonsoon (520 to 1130 org/L) and low during postmonsoon (190 to 290 org/L) and monsoon (90 to 400 org/L). Statistical analysis (CCA) showed that, salinity and dissolved oxygen are the major environmental variables influencing the MZP assemblages in this estuarine system. Interestingly, heterotrophic dinoflagellates were found dominant (15 to 80% of MZP community) during monsoon season when estuarine system experiences intrusion of low oxygen water from the coastal region. Whereas, loricate ciliates were dominant during the premonsoon (28 to 62%) and postmonsoon (39 to 55%) associated with prevailing high saline waters in the region. Further, investigation on MZP grazing and species-specific salinity and low oxygen concentration preference and/or tolerance would help to understand their pivotal role in the food web dynamics in a tropical estuarine system.

Inclusion of 1-*O*-alkyl-*sn*-glycerol and docosahexaenoic acid in rat liver lipids

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Reduced levels of plasmalogens (PLs) in the nervous tissue in humans frequently depend on age; this is associated with a reduction in the peroxisomal enzymes activity at the initial stage of PLs biosynthesis. These alterations are especially characteristic for patients with neurodegenerative disease such as Alzheimer's disease. At present, one of the perspective methods of prevention of the negative effects resulting from impairment of plasmalogen biosynthesis is plasmalogen replacement therapy, by which the patient's diet is supplemented with plasmalogens themselves or their precursors, which include 1-*O*-alkyl-*sn*-glycerol (AG). Another critical metabolite included in the PLs composition is docosahexaenoic acid (DHA). DHA synthesis also takes place in the peroxisomes.

We have simulated a course of plasmalogen replacement therapy. Wistar rats were fed for 4 month diets that contained 150 mg/kg AG (chimyl alcohol 94%) and 500 mg/kg of polyunsaturated fatty acid (PUFA) ethyl esters (DHA 32%). In groups AG and AG+PUFA, an increase of the ratio 16:0-dimethylacetal/16:0-fatty acid methyl ester $\times 100$ (1.41 ± 0.11) was observed compared to controls (0.72 ± 0.05), that confirms the inclusion of chimyl alcohol in PLs biosynthesis in rat liver. In the group AG+PUFA and PUFA, the content of DHA in liver lipids increased to $5.35\pm 0.30\%$ and $5.77\pm 0.40\%$, respectively, compared to control $3.86\pm 0.50\%$. Thus, our results indicate that AG and ethyl esters of PUFA introduced in the diet can be incorporated into PLs and increased DHA level in liver.

We have developed a technological approach of complex processing of squid liver and isolation of AG and n-3 PUFA from liver oil of the squid *Berryteuthis magister*. The proposed procedure allows obtaining of purified AG ($\geq 99\%$), with chimyl alcohol as the main component (94%) in 57% yield. After AG separation, the lipid mixture was used to obtain PUFA concentrates (eicosapentaenoic acid (32%), DHA (32%)) and highly purified eicosapentaenoic acid and DHA ($\geq 99\%$).

The use of stationary underwater video surveillance systems for the evaluation of life indicators barnacle

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Stationary underwater surveillance systems are increasingly used around the world for long-term observation of coastal ecosystems. Three such surveillance systems are currently deployed by POI scientists as part of a network of instruments dedicated to scientific monitoring of Peter the Great Bay (Sea of Japan). Two of them were installed in 2013 and 2014 in the Alekseeva Bay (Popov Island, the Amur Bay). Another system was established in 2016 in the Vityaz Bay (Pos'eta Bay). The systems use sealed stationary IP-cameras for continuous observation of underwater scenes. At scheduled times information from the cameras (snapshots and short videos) is sent to the monitoring system database. In addition, live videos from the cameras are available online through "POI FEB RAS Underwater Video" web-service.

The surveillance systems include a set of programs for quantitative analysis of video data that complement visual observation. For example, with their help it is possible to obtain estimates of characteristics of underwater currents, surface waves, sea level fluctuations, color and turbidity. Another application of the programs is quantitative description of the state of the underwater biodiversity. One of the programs detects moving fishes and calculates their number in a camera's field view. Another program is useful in the study of sedentary biological objects performing visually noticeable periodic motions of body parts. The program allows to record a periodic process, determine its fundamental frequency, stability of this frequency in time, continuity of the process.

With a help of the surveillance systems a study of leg waving process of adult and young barnacles was carried out in summer and winter periods. By waving its legs a barnacle provides the flow of water with nutrients in its shell. The barnacles were located on an artificial reef in the field of view of one of the cameras. The program recorded the changes in the average brightness of the video frames in places where legs of the barnacles appeared and disappeared with the strokes. Several sessions of continuous observations, each lasting up to 8–10 hours were performed. In each session between 3 and 6 barnacles were observed at the same time. It was established that the fundamental frequency of leg waving process in the summer on average ranged from 0.7 to 1.1 Hz, in winter it fell almost 3 times to 0.3–0.4 Hz. In the summer, there was a significant difference in frequency stability and continuity of leg waving process for different individuals. Winter observations were characterized by very high frequency stability and almost complete absence of stops in the waving process. Juveniles barnacle were observed in the summer directly at the front glass of the underwater video system. Their main feature was a very high frequency of leg waving legs, 3–4 strokes per second.

An aquaculture safe for other inhabitants of sea

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Modern aquaculture technologies are based on collecting a lot of larvae and spores of certain species on collectors in the natural environment. These cultured organisms, after being transplanted, remain on land and die. This phenomenon has been given the term “ecological trap” (Dwernychuk, Boag, 1972). However, the dead larvae often have a higher cost than the cultivated species, and their mortality is caused by their smaller size and inconspicuous body color. These species include red king crab, Japanese sea cucumber, sea urchins, *Cucumaria japonica*, some species of sea squirts and sponges. Transplantation of thalli of Japanese kelp onto new ganging causes a month delay in their growth, and destruction of the community that formed in rhizoids by autumn reduces the rate of thallus growth, because the rhizoid community is in fact “nodule bacteria” that provide algae with nitrogen. However, our studies have shown that the use of realized, rather than fundamental, niche when selecting an area and depth for settling the artificial substrates, optimization of the density of kelp zoospores, as well as improvement of collectors’ design make it possible to grow the cultivated species without transplanting them to other facilities. In this case, many valuable associated species on artificial substrates can reach a marketable or viable size. The tests of collector – cages constructed in 1981 (Rus. Patent No. 826998; Fig. 1), placed on the bottom, showed that due to the use



Fig. 1. The collector-cage for gathering and breed larvae of molluscs to one-year-old age.

of covering net with a mesh size of 10 mm, they can be left in the sea until the spring of the following year, without any damage to scallop and accompanying species. In the case of equipping these collector – cages with a covering net with the mesh size of 15 mm, Japanese scallop can be cultured in them to a marketable size with no transplantations. Within three years, Japanese sea cucumber, sea squirts, and sponges, also growing in them, can reach the marketable size, while red king crab, sea urchins, and *C. japonica* can reach a viable age. Cultivation of scallop in these collector – cages with this of an overall covering net provides even more promising results. Applying an overall net cover to a facility for cultivation stimulates construction of a machine for hauling the facility out of the water, cleaning it from fouling, putting on a new net cover, and dipping it back into the sea. A mechanization of the entire cultivation process would largely reduce the cost of the products obtained and make it profitable in many countries (Rus. Patent No. 11178371; Fig. 2).

In case of Pacific oyster cultivation, technology involves several labor-intensive operations. To collect larvae from plankton in the sea, scallop shell valves, strung on a wire, are immersed into the water. After settlement of oyster larvae, separating spindles are put between the neighboring scallop valves; in autumn, excess juveniles are cleaned off from each valve. Settled juvenile oysters attach to the substrate with their entire bottom valve, and removing them in the process of harvesting poses a significant danger both to oyster and to collector's hands. Our plastic collector for collecting and on-growing of oyster to the marketable size (Rus. Patent № 886869; Fig. 3) resolves many problems associated with its cultivation and, at the same time, facilitates harvesting operations.



Fig. 2. The collector-cage for gathering and breed larvae of molluscs to three-year age.



Fig. 3. The collector for gathering and breed larvae of *Crassostrea gigas* to commercial size.

The presented materials allow us to conclude that aquaculture provides an opportunity to obtain products without damage to the environment, and the cultured organisms will have a higher taste value and useful properties, as they are grown in their natural habitat.

Distribution of benthic fauna on the Vulkanologov Massif – Komandor Basin slope: investigations with ROV “Comanche 18”

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During the 75th cruise of RV “Akademik M.A. Lavrentyev” (June 2016) landscape-ecological environment and the distribution of macro and mega benthos on the northern slope of the Vulkanologov Massif (south-west Bering Sea) have been explored using ROV “Comanche 18”. Seven dives were performed in the depth range from 4278 up to 349 m. From technical considerations routes of dives were directed upwards along the slope. Continuous visual observations (total duration 40 hrs 16 min) have been provided. For the estimation of size of objects and the area of the surveyed bottom the 10 cm laser scale was used. The observations were accompanied by photographic and video recording. A series of video transects were performed at a speed of approximately 0.5 knots. In total, 1372 photo images and 22 hrs 29 min videos were analyzed. During each dive samples of bottom fauna were collected using mechanical arm of ROV. After preliminary sorting and preparation aboard the ship the animals were preserved for different kinds of analyses. Obtained biological collection comprises 206 units of storage. With the help of deep-sea taxonomic experts, collected, photographed and video recorded animals were identified to the lowest taxonomic level possible. Totally more than 150 macro- and megafaunal species have been distinguished. Using video transects relative abundance of dominant (=landscape determining) taxa were calculated for different depth ranges. As a result general patterns of vertical distribution of bottom communities were established. At maximal explored depth the abyssal community is dominated by several holothurian species, among them *Kolga kamchatica* are most abundant. Starting from the depth 3600 m sedimented slope is occupied by the community dominated by *Scotoplanes kurilensis*. At the depths 3000–2800 m dense settlements of ophiurids (several dozens of specimens per m⁻²) were observed. Starting from the depth of 2650 m synallactid holothurians *Pelopotides solea* and red benthopelagic Trachymedusae (pres. fam. Rhopalonematidae) dominate the community. In the depth range 2290–1830 m mass development of the acorn worms Enteropneusta (fam. Torquaratoridae) were recorded (up to 10 specimens per m⁻²). Above 1700 m aggregations of hexactinellid sponges *Farrea occa* compose the basis of landscape. Live and dead sponges are inhabited by diverse fauna. Starting from the depth 700 m the community is dominated by soft corals (Alcyonaria and Corallimorpharia). The summit area (390–349 m) is occupied by dense settlements of zoantharians *Epizoanthus* sp. Based on vertical distribution of megafaunal taxa the depths of most prominent faunistic boundaries have been revealed.

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Biological investigations of the Piip Volcano with ROV “Comanche 18”

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During the 75th cruise of RV *Akademik M.A.Lavrentyev* (June 2016) landscape-ecological environment, hydrothermal evidences and mega and macro benthos on Piip Volcano (south-west Bering Sea) have been explored using ROV *Comanche 18*. In total, 17 dives were performed in the depth range from 1806 up to 349 m. Visual observations were accompanied by photographic and video recording and samples collecting. In the observed area distinct vertical zonal distribution of benthic fauna was revealed. In the depth range from 1700 up to 750 m aggregations of live and dead hexactinellid sponges *Farrea occa* forming a kind of reefs inhabited by diverse fauna compose the basis of landscape. Above 700m the community is dominated by soft corals *Anthomastus* sp. and *Corallimorphus pilatus*. The summit area (390–349m) is occupied by dense settlements of zoantharians *Epizoanthus* sp. and small pink sea anemones. Hydrothermal manifestations were generally observed above 450 m. On the slope of volcano extensive fields (up to several dozens of m²) covered by bacterial mats (mainly *Beggiatoa*) were recorded. Most hot vents (more 100°C) are concentrated in summit area. There are small (up to 1.5 m) anhydrite and carbonate structures, and cavities marked by shimmering water. In the zone of diffuse venting through loose deposits, settlements of bivalves *Calyptogena pacifica* (fam. Vesicomidae) were observed. This is northernmost known location of these symbiotrophic animals in Western Pacific. Adjacent bacterial mats were inhabited by numerous small provannid gastropods (gen., sp. n.) and lysianassid amphipods. Besides these groups, purple Corallimorpharia, ampeliscid amphipods, Calcyspongia, ampharetid polychaetes and Leptostraca (crustaceans, first recorded in this region) were obviously attracted to the vent zone. The role of chemosynthetic production in feeding of different animals is discussed. So far, Piip Volcano represents most shallow and most northern West-Pacific hydrothermal site inhabited by true obligate vent fauna.

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Development and creation of a remote-controlled underwater laser induced breakdown spectrometer for analysis of the chemical composition of sea water and bottom sediments

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The main purpose of the work is the development and creation of a underwater spectrometer for monitoring the elemental composition of seawater in field conditions. The design of the underwater laser induced breakdown spectrometer was made on the results of the experimental studies in the frame of Russian Science Foundation (agreement no. 14-50-00034 “Modern technologies and technical means of monitoring the state of marine ecosystems and marine biological resources”). This spectrometer is designed to in-situ measure the elemental composition of sea water and bottom sediments at depths of 1–100 meters. Laser induced breakdown spectrometer is mounted on remotely operated finding class underwater vehicle which will be created during this project.

The underwater laser induced breakdown spectrometer consists of a laser excitation source (DF251, Sol Instruments, Belarus), a spectrum recording system (optical part, Maya 2000 Pro spectrometer, Ocean Optics) and a microcomputer. These components are mounted on a separate chassis, which is inserted into a rugged case with a diameter of 35 cm and a length of 80 cm.

The laser source has the following parameters: DPSS Nd: YAG type, radiation wavelength 1064 nm, pulse repetition rate 1-10 Hz, single-pulse and two-pulse generation mode, single-pulse energy 100 mJ, two-pulse 2·50 mJ, pulse duration 10–12 ns. By the focusing of laser radiation on the bottom surface or in the water a laser plasma was generated. Plasma emission registered by the spectrum recording system and transmitted by the Ethernet to the operator computer in the real time condition. Registered spectral data is automatically processed in a Matlab software system and displayed on a computer screen.

Now a mechanical part of the spectrometer underwater vehicle, a rugged case with a laser source and a recording system have been created. Also, work is underway on aligning all systems and laboratory testing of laser plasma detection and excitation systems. It is planned to complete the entire system by the end of 2017.

The autonomous underwater robotic complex for marine bio-resources monitoring

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Increasing of anthropogenic and technological pressures on the marine environment leads to a long-term decline of biological diversity and general marine ecosystem degradation. The major part of data for an objective evaluation of marine bio-resources is received during monitoring of space-time variability of chemical, biological and physical environmental characteristics. In the presentation the autonomous robotic complex “CH-2” for marine bio-resources monitoring is presented. The complex was designed and manufactured in Institute of Marine Technology Problems FEB RAS.

The complex represents a combination of the autonomous underwater vehicle (AUV) and the surface repeater connected by the optical cable. The feature of the AUV is the build-in variable buoyancy system that allows controlling target depth without energy consumption gaining data from on-board sensors and cameras. The AUV is equipped with propulsion system composed of two horizontal and two vertical thrusters. Such allocation of thrusters allows maneuvering in the target region with active control of target depth, yaw and pitch. Also, the vehicle includes sensitive acoustic sensors for noise measurement of the marine ecosystem.

The surface repeater provides global navigation of the complex by the GPS receiver. In addition to the GPS system, the surface repeater includes the Wi-Fi adapter that allows controlling the vehicle and transmitting sensors data from one to escort small vessel or coast station in real-time mode. The surface repeater can be equipped with satellite communication system “Iridium” or “Gonets” if needed to provide connection to a remote station.

The original software of the complex and scalable data bus (CAN, RS-485 and Ethernet) allows quickly equipping new monitoring instruments: a manipulator for grabbing of marine organisms, a water and microbiota sampler, a CTD-sensor and others.

The presentation shows results of marine trials of the autonomous robotic complex “CH-2” took place in summer 2016 in Vityaz Bay, Primorsky Krai. The possibility of equipping the AUV with manipulation system is considered. The engineering solution of variable buoyancy system for noiseless depth control for recording noise of sea, and its inhabitants is presented.

The reproductive pathways of floating *Ulva prolifera* and the response mechanisms to stress

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The green tide has been developed in the Yellow Sea, China since the year 2008, of which free-floating *U. prolifera* is one of the causative species. The key research of the project aims to study the photosynthetic carbon fixation in the rapid accumulation and floating state of the thalli and the molecular mechanism of upper floating thalli to stress.

(1) Rapid reproductive means and mechanism of sporogenesis in floating *Ulva prolifera*. During fragments excised from intact thallus were cultured in laboratory, the cell in thallus fragments changed greatly. Those cells developed into sporangium after 48 h culture. Subsequently zoospores were released from those sporangia. The cells in fragments of floating *Ulva prolifera* were vegetative during 0–48 h culture. Meanwhile photochemical quantum yield of PS I Y(I) and effective PS II quantum yield Y(II) were relatively stable and the variation tendencies between them were similar. These results indicated that both CEF enhancement and the oxidation of the plastoquinone pool are essential for sporulation onset. The further research suggested carbon fixation in photosynthesis has a relationship with sporulation. The decline of LEF can decrease inorganic carbon fixation.

(2) The pigment composition of floating *U. prolifera* thalli and the carbon source for photosynthetic carbon fixation process. *Ulva prolifera* thalli in free-floating mats could be separated into three groups according to color: light green, intermediate green, and dark green. The effective photosystem II quantum yield (YII) of the light green thalli cells had the highest value. These thalli usually are exposed to the air for long periods of time, during which it is difficult for them to acquire dissolved inorganic carbon from the seawater. So, why the growth rate of free-floating *U. prolifera* is extremely high when green tides occur? Our results found that *U. prolifera* thalli can utilize CO₂ in the air directly. CO₂ assimilation was enhanced as CO₂ increased in the air, even when under moderate stress, such as dehydration and salt stress.

(3) The tolerant mechanism of *Ulva prolifera* to desiccation. We found that during desiccation, the PSI-driven cyclic electron flow in *U. prolifera* increased significantly. Although the activity of cyclic electron flow is lower than that of linear electron flow which dominates under optimal conditions, the cyclic electron flow might provide some protection of the photosynthetic apparatus and extra ATP under desiccated conditions. Based on these results, we propose that the physiological tolerance and stability of photochemical systems, especially PSI-driven cyclic electron flow, might be one of the most important factors that make *U. prolifera* well suited to withstand repeated cycles of desiccation and re-hydration during daily low and high tides.

Lipid molecular species and lipidom analysis as a new approach in the study of trophic and symbiotic relationships of marine invertebrates

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For more than forty years, fatty acids (FAs) have been applied as qualitative and quantitative markers to confirm trophic relationships in the marine environment and resolve dynamic processes of the marine ecosystems. Certain marker FAs of marine producers may be transferred to and can be recognized in their consumers. FAs rarely exist in free form and are usually esterified to a glycerol backbone of acyl lipids. Thus, FAs are transferred between marine trophic levels in the form of acyl lipids, but the structure, content, and distribution of the individual acyl lipids with marker FAs are still unknown. In a consumer organism, a part of dietary acyl lipids are decomposed to yield acylglycerols and FAs, which can be used for biosynthesis of new acyl lipids of the consumer. Each acyl lipid class is known to consist of a lot of lipid molecular species with different acyl groups, and the sum of molecular species of the organism forms his lipidom. Lipidomic analyses have been recently started for marine organisms. In our opinion, the distribution of marker FAs both in producer lipidom and in consumer lipidom is important for the explanation of the features of transfer, accumulation, and metabolism of FAs in marine food webs. In addition to a common analysis of FA profiles, a lipidomic analysis of molecular species profiles may provide us with new tools necessary to conduct marine ecological studies. A comparison of the producer and consumer lipidoms seems to be useful for understanding of lipid and FA metabolism in marine organisms. Probably, the same hypothesis is truth for the transferring of lipids between organisms in marine symbiotic associations.

To verify these hypotheses, the polar lipidoms of one mollusk species, several species of soft corals, and coral endosymbiotic microalgae (zooxanthellae) were determined for the first time and compared with the special attention to the molecular species with marker FAs. This approach allowed us to find the dietary source of marker FAs in the mollusk, explain the reason of abnormally high level of these acids in the consumer, unambiguously confirmed the transfer of PUFAs from zooxanthellae to the coral host, and determine the features of biosynthesis of phospholipids in mollusks and corals.

On the selection of the configuration of autonomous underwater robots for monitoring marine biological resources

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Autonomous underwater vehicles/robots (AUV) are increasingly used to monitor marine biological resources in coastal waters. The key to scaling up the use of AUVs is to increase the efficiency of their work; i.e., minimizing the cost per unit area of the examined region.

Traditional technologies relating to AUV applications require significant overhead costs which are associated with navigation infrastructure to support AUV missions; e.g., transect sequences. As an example, consider the task of monitoring populations of red king crabs in the Peter the Great Bay of the Sea of Japan. The dimensions of the study area indicate that the appropriate AUV path length is about 500 km. Because of the requirement to deploy bottom moored acoustic transponders of the Long Baseline (LBL) navigation system, there are 14–28 AUV sorties and the same number of LBL redeployments. The use of an Ultra-Short Baseline (USBL) navigation system eliminates the need for repeated transponders to be relocated on the bottom, but instead requires permanent surveillance of the support ship using a USBL transceiver (in our estimation the total time is six days or more to cover the entire study area).

An increase in AUV efficiency under these conditions is possible in several ways:

- 1) Minimizing the assistance of the time of use of a support ship by:
 - allowing the vessel to execute shore-based AUV missions or use a subsea docking station;
 - reducing the level of AUV supervision and permanent tracking, and increasing the level of autonomy;
 - using AUV navigation without LBL bottom acoustic transponders.
- 2) Allowing on-board processing and decision-making processes to reduce AUV mission duration, while keeping the mission's scope.
- 3) Increasing the mission's scope by extending AUV endurance.

The experience of IMTP FEB RAS engineers gained from both the development process and AUV field operations allows us to offer two versions of robotic systems for these purposes. The first is to use a type of AUV MT-2012 with dead reckoning navigation based on Inertial Measurement Units and Doppler Velocity Logs, which surfaces for GPS fixes (every 3–6 hours, depending on specified positioning accuracy). The same surfacing can be used for communication/data transfer between the AUV and surface/operator control station.

The second option involves the use of marine autonomous robotic complex (MARC), which consists of underwater and surface (USV) robots. Coordinated movements of AUV and USV simplify the navigation system and make it cheaper. Control and data management become easier by using a through-air communication channel.

Real time object recognition tools are available as an AUV option for contour tracking or finding the center of closely grouped crab clusters in our case.

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Water-soluble polysaccharides from brown algae *Desmarestia viridis*, *Dictyota divaricata*, *Dictyota dichotoma* and *Padina boryana*

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The search of safe antitumor drugs from natural sources for the treatment and prevention of cancer is very crucial. People used algae to treat and prevent various diseases for years. The most valuable biologically active substances from brown algae are polysaccharides: water-soluble fucoidans and laminarans, and alkali-soluble alginic acids.

The aim of this work is the isolation of water-soluble polysaccharides from brown algae *Desmarestia viridis*, *Dictyota dichotoma*, *Dictyota divaricata*, and *Padina boryana*; determination of their structural characteristics and anticancer activity.

Water-soluble polysaccharides were extracted with diluted solution of hydrochloric acid from defatted brown algae. Negatively charged fucoidans were separated from neutral laminarans by anion exchange chromatography. Further laminaran fractions were purified by hydrophobic chromatography.

The yields of laminarans from *D. viridis* and *D. dichotoma* were 0.43% and 1.36% of defatted algae weight, respectively. The analysis of the monosaccharide composition of laminarans from *D. viridis* and *D. dichotoma* (DvL and DdL) showed that they are glucans. The ¹³C NMR spectra of laminarans contained signals with chemical shifts of 103.7 (C1), 74.4 (C2), 85.5 (C3), 69.3 (C4), 76.8 (C5) and 62.0 (C6) ppm specific for 1,3-linked β-D-glucose residues, the signals with chemical shifts of 104.0 (C1), 74.7 (C2), 75.7 (C3), 70.8 (C4), 77.2 (C5), and 70.0 (C6) ppm characteristics of 1,6-linked β-D-glucose residues, as well as signals at 86.1 (C3) and 76.0 (C5) ppm specific for 1,3,6-linked residues of β-D-glucose. The ratio of bonds 1.3:1.6 was calculated from a comparison of the intensities of the signals of anomeric protons in ¹H NMR spectra and was 10:1 and 3:1 for DvL and DdL, respectively.

The yields of fucoidans from *D. viridis*, *D. dichotoma*, *D. divaricata* and *P. boryana* were 0.51, 1.02, 0.32 and 0.25% of defatted algae weight, respectively. According to the analysis of the monosaccharide composition, all obtained fucoidans were sulfated heteropolysaccharides. The fractions did not include proteins and contained polyphenols in small amounts (0.7–4.3% of the sample weight). The fucoidan, isolated from *D. viridis*, consisted of the fucose, galactose, xylose, and mannose residues and sulfate groups (12.5%). Heterogeneous fucoidan from *D. dichotoma* contained fucose, galactose, xylose, mannose, and glucose residues. This fraction contained a small amount of sulfate groups (2%), it is very unusual for fucoidans. Two fractions of fucoidans from

D. divaricata were represented by sulfated polysaccharides (11.2 and 18.3%), consisting of fucose, galactose (Fuc:Gal=2:1 and 1:1) and trace amounts of mannose, xylose and glucose residues. Fucoi-dan from *P. boryana* was sulfated (18.2%) galactofucan (Fuc:Gal=2:1), containing small amounts of mannose and glucose residues. The galactofucans from *D. divaricata* and *P. boryana* were choosing for the study of the biological activity. It was shown that they have anticancer effect against DLD-1 carcinoma cells *in vitro*.

Laminarans of brown algae and their sulfated derivatives: isolation, structure, and biological activity

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Brown algae have drawn worldwide attention due to their involvement in many industrial applications. Laminarans are reserved polysaccharides of brown algae and they consist of β -1,3;1,6-linked D-glucose residues. Laminarans have a broad spectrum of biological activities including antimicrobial, immunomodulating, and anticancer activities. Laminarans can be modified enzymatically or chemically to obtain polysaccharides with more potent biological action. Nowadays modification of laminarans such as sulfation has a great interest.

The aim of the present work is to isolate the laminarans from Far Eastern brown algae *Saccharina cichorioides*, *Saccharina japonica*, and *Fucus evanescens*, obtain their sulfated derivatives, determine the structural characteristics of polysaccharides and to investigate their anticancer activity *in vitro*.

Laminarans were isolated from brown algae *S. cichorioides*, *S. japonica*, and *F. evanescens* using combination of extraction methods and hydrophobic chromatography. It was shown that the main chain of laminarans from *S. cichorioides* and *S. japonica* consisted of β -(1 \rightarrow 3)-linked D-glucopyranose and branch to C6. The laminaran from *F. evanescens* consisted of not only β -(1 \rightarrow 3)-linked D-glucopyranose, but also includes single β -(1 \rightarrow 6)-linked D-glucose residues. The branches at C6 are presented as glucose or as gentiobiose. The sulfated laminarans with different degree of sulfation were obtained by the chlorosulfonic acid-pyridine method. The sulfated laminaran from brown seaweed *F. evanescens* was found to have the highest degree of sulfation (50%), the laminaran from *S. cichorioides* was sulfated on 42% and the sulfated laminaran from *S. japonica* contained 34% of sulfates.

In vitro cytotoxic and antiproliferative activity of investigated compounds against human colorectal adenocarcinoma HCT 116, malignant melanoma SK-MEL-5, and breast adenocarcinoma MDA-MB-231 cell lines was determined by MTS method. It was shown that all investigated polysaccharides were less cytotoxic against tested cells at the concentration range up to 800 μ g/mL after 24 h of treatment. However, they inhibited cancer cells proliferation at dose of 200 μ g/mL in different manner. The effect of the polysaccharides (at concentration 200 μ g/mL) on colony formation of human cancer cells was studied using soft agar clonogenic assay. The laminaran from *S. cichorioides* and its sulfated derivative suppressed colony formation of HCT 116 cells on 16 and 32%, respectively, and SK-MEL-5 cells on 17 and 27%, respectively, but not colony formation of MDA-MB-231 cells, compared with non-treated cells. The polysaccharides from *S. japonica* were observed to inhibited

colony formation of SK-MEL-5 on 29 and 34%, respectively, and MDA-MB-231 cells on 45 and 53%, respectively, but not HCT 116 cells. On the other hand, the treatment with laminaran from *F. evanescens* and its sulfated derivative had a profound inhibitory effect on colony formation of all tested cell lines. The sulfated laminaran from brown alga *F. evanescens* possessed a powerful inhibitory activity and almost completely prevented the formation and growth of colonies of these human cancer cells.

In conclusion, our findings indicate that laminarans from Far Eastern brown algae and their sulfated derivatives may be one of the therapeutic approaches for cancer treatment.

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Diversity of tanaidaceans in harbors of the South Korea, with three new species of the family Tanaididae Nobili, 1906 (Crustacea: Tanaidacea: Tanaidomorpha)

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Tanaidaceans are small peracarid crustaceans and distribute in all marine habitats and over the full range of depths. They are small benthic crustaceans with a benthic life cycle and low dispersion rates. Their life history and strategies are closely related to environmental factors, so they can be available as indicators of ecosystem health. Some of the taxa are dispersed by marine vertebrates, floating algae, anthropogenic transport such as shipping, and by prevailing sea-currents. In particular, the genus *Hexapleomera* has been known to be taxon commensal on turtles, or manatees and regarded to show cosmopolitan distribution. However, recent studies revealed that some species of this genus inhabit diverse environments such as on the hulls of yachts and benthic sediments, as well as marine turtles do not show a cosmopolitan distribution as population. To assess the species diversity of tanadiaceans in the harbor or pier, samplings were conducted in seven locations of the southern and western regions of the South Korea (Anheung, Yokjido, Geomundo, Ulsan and three locations of Jeju Island), using diverse sampling methods: light trap set, direct picking, epi-sledge net, and grab. As a result, two genera, only in the family Tanaididae Nobili, 1906, were found. Five new species (3 of *Hexapleomera* and 2 of *Zeuxo*) were confirmed based on the combined analysis including full descriptions and molecular analysis using mitochondrial cytochrome c oxidase subunit I (mtCOI) gene. Species of *Hexapleomera* from the Geomundo and Jeju Island showed the same morphology and genetic results. These locations are much far from each other, as compared to distances between other study areas. Our result suggests the possibility of shifting of tanaids by the Tsushima Warm Current (TWC) and/or shipping between these two areas.

**New record of the genus *Hexapleomera* Dudich, 1931
(Crustacea: Tanaidacea: Tanaididae)
from the South Korea,
based on morphological and molecular evidence**

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Harbors are well known gateways for species introductions in marine environments but little work has been done to uncover relationships between species diversity, harbor type, and geographic distance to reveal the secondary spread. As a part of a study to monitor introduction of alien invasive species by shipping in harbors, we sampled tanaidaceans from two harbors, using a light trap set overnight at the entrance. Two new species are described for *Hexapleomera* Dudichi, 1931 from the southeastern coast and Jeju Island of South Korea (North West Pacific). *Hexapleomera* sp. 1 is clearly differentiated from other species of the genus by a characteristic combination of antennule with six aesthetascs, uropod with five segments, a maxillule palp with four distal setae, epignath with short and blunt spiniform seta, and maxilla with a rugged shape of the distal margin. *Hexapleomera* sp. 2 can be distinguished from other species of the genus by antennule with four aesthetascs, maxillule palp with six distal setae, propodus of pereopods 2–3 bearing one short seulouse seta, and one slender simple seta on the ventral margin. To resolve the taxonomic complexity frequently occurring in *Hexapleomera* and to prove two the Korean species to be new to science, full descriptions of the matured females and males are provided, and these are supported by distinct genetic differences in mitochondrial cytochrome c oxidase subunit I (mtCOI) gene between *Hexapleomera* sp. 1 and *Hexapleomera* sp. 2 (ca. 16%), which is first gene data in the genus. In addition, close examinations on the appendages, known to have morphological variations between the individuals of one species and/or between different genders, are carried out based on two new species and the results are discussed.

**A new genus and species of sphyrapodid tanaidaceans
(Crustacea: Peracarida)
from the southern coast of the South Korea**

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Species diversity and distribution pattern of macrobenthos in the southern subtidal zone of the South Korea were investigated. Samplings were conducted using the epi-sledge net. Potential new genus and species belonging to the order Tanaidacea was found and described in this study. The genus is included in the subfamily Sphyrapodinae Guțu, 1980 by mandible without palp and has close affinity with the genus *Poligarida* Bamber and Marshall, 2013 by shape of the third pereonite with lateral apophyses, antennule male fringed with rows of aesthetascs on the outer flagellum, and an elongate article 2 of antenna, but is distinguished from that genus by having maxilluar palp. A new genus is unique in the subfamily Sphyrapodinae by a character combination of the proximal peduncle of male antennule forming a robust projection covered with denticles and a deep groove on the distal margin, uni-articulated maxillular palp, pereonites each with a ventral keel, and the third pereonite with lateral spines. To prevent the confusion and difficulties in identification by morphometric variations with size and little information in sexual dimorphism, detailed characteristics of new genus and species are fully described for mature adults of both sexes and compared to different sized individuals. A comprehensive comparison for main characteristics of genera within Sphyrapodinae is provided and discussed. To our knowledge, this report is the first record of sphyrapodinae in the NW Pacific.

The diel vertical migration of zooplankton in the hypoxia area observed by Video Plankton Recorder

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The vertical distribution of 4 zooplankton taxa (copepods, gelatinous zooplanktons, chaetognatha, euphausiacea) were studied during a 24 hour survey using a Video Plankton Recorder (VPR) stationed offshore of Zhejiang province in the East China Sea. Temperature, salinity, fluorescence and dissolved oxygen were investigated together with the vertical distribution of four taxa in order to discuss the reason for diel vertical migration (DVM). Copepods and chaetognatha performed typical DVM, but only a small part of the population appeared under 40 m. Gelatinous zooplanktons aggregated at the surface water layer shallower than 30 m. DVM of euphausiacea remained uncertain because of the small number of individuals investigated in the study. Our study confirmed that VPR could be used as a valuable tool to study zooplankton DVM. DVM of most zooplankton living in the coastal area of the East China Sea might be affected by multiple environmental elements such as feeding activities, predator presenting, stratification of water column and energy utilization. This is the first time VPR has been used in our country, and its use can improve our zooplankton observation.

Phenotypic plasticity during larval development of sea urchin, *Strongylocentrotus intermedius*

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Phenotypic plasticity, i.e. the ability of a single genotype to produce different phenotypes in response to variations in environmental conditions, is often considered as an important mechanism of organisms' adaptation to the changing environment. Numerous studies of terrestrial and aquatic organisms showed alterations in their morphology, physiology or behavior in response to particular environmental conditions in order to increase their fitness.

According to the experimental studies, pre-feeding larvae of many sea urchin species grow longer arms when develop in food-limited conditions as compared to their well-fed siblings of the same age. As a result, poor-fed larvae develop larger ciliary bands and are able to capture food particles from the water more efficiently and increase their feeding rate when food is scarce. To date, larvae of nearly twenty sea urchin species were tested for phenotypic plasticity during their development. However, the degree of the plastic growth of arms among closely related sea urchin species remains poorly investigated. Here, for the first time, we provide data on phenotypic plasticity in pre-feeding larvae of sea urchin, *Strongylocentrotus intermedius*, under different feeding regimes. Larvae at the blastula or early gastrula stage (24 hours post-fertilization) were partitioned into glass containers with three feeding regimes: no food (NF), low food (LF), and high food (HF). By the end of pre-feeding period, on day 4 of development, 45 two-armed plutei from each feeding regime were photographed under a Zeiss AxioImager Z.2 microscope, and the lengths of their postoral arms and body rods were measured using the ImageJ image processing software. The data were analyzed by one-way analysis of variance (one-way ANOVA) using the R system for statistical computing. Multiple comparisons between different feeding regimes were done using the Tukey's honest significance (Tukey's HSD) test. Our results indicate a statistically significant effect of feeding regime on postoral arm length (ANOVA: $F(2.132)=21.66$, $p<<0.001$) on day 4 of development. Tukey's HSD test showed a statistically significant difference in length of postoral arms between larvae from the HF feeding regime and those from the NF and LF feeding regimes ($p<0.001$). No difference in length of postoral arms between larvae from the NF and LF feeding regimes was found. In absolute values, postoral arm length in larvae from the HF feeding regime on day 4 of development was approximately 10% shorter than those in the other two experimental groups. Besides, a statistically significant effect of feeding regime on the body rod length was also observed (ANOVA: $F(2.132)=6.29$, $p=0.003$). Tukey's HSD test showed that the body rod length in larvae from the HF feeding regime was statistically significantly different from those in the NF and LF feeding regimes ($p<0.05$). A detailed analysis of the data showed that the body rod length in larvae from the HF feeding regime tends to be slightly smaller (approximately 3%) than that in larvae from the other experimental groups. Our results clearly indicate that pre-feeding larvae of *S. intermedius* display a phenotypic plasticity in response to variations in available amount of food in the environment.

Electron microscopy analysis of embryonal and larval development of *Mizuhopecten yessoensis* (Bivalvia: Pectinidae)

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Due to its high taste value and large size (the biggest among pectinids), *Mizuhopecten yessoensis* has become a very popular food product and is used as an object of aquaculture in a number of countries. In spite of the high economic value of this scallop, no ultramicroscopic studies of embryonic and larval stages of its development have been carried out to date. At a water temperature of 13°C, gastrulae of *M. yessoensis* are formed within 24 hours after fertilization. They are rounded, oval, or tapered in shape and have two invaginations: the primordium of shell gland is located on the dorsal side, and the blastopore is on the ventral side. In the shell gland primordium, there are cells that begin differentiating to secrete the larval shell subsequently. Embryos are composed of cells morphologically similar to each other. Their cytoplasm contains yolk granules of two types: with a lipoprotein and glycoprotein content. Within 51 hours of development, most embryos are represented by trochophores. The apical plate of these larvae is fringed by the locomotor organ, prototroch, formed by a double ring of cilia. There is long ciliary tuft of the apical flagellum, formed by a group of associated cilia, at the centre of apical plate. The cells of archenterone and the external wall of the larva body are still morphologically similar to each other at this stage. In the primary cavity of the body, a pool of undifferentiated mesenchymal cells is observed. Within 70 hours of development, most larvae become late trochophores. The growth of the prodissoconch I is active; prototroch gradually transforms into velum. Synthetic processes are activated in cells of the larval gut. The myogenic differentiation of mesodermal myoblasts begins. On day 8 of development, all larvae are represented by early veligers. Their body is covered with a semitranslucent shell. The apical plate of larvae is edged with locomotor organ, velum. The anterior adductor is still undeveloped at this stage. The larval velar retractor muscles have cross striation with a period of 1 µm. The larval intestine is well differentiated. It consists of esophagus, stomach, and short gut opening into the mantle cavity. The esophagus wall is formed by ciliated epithelium. The stomach wall, in spite of the early stage of development, already shows a clear division into regions corresponding to the gastric shield and style sac of adult animal. The digestive gland at this stage is a swelling of the stomach wall, without forming the lobes. It is formed by cells of two types: undifferentiated cells, division of which provides the growth of organ, and cells that perform the intracellular digestion processes. Thus, we have studied the structure of embryonic and early larval stages of *M. yessoensis* development, using SEM and TEM, for the first time.

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Isolation and characterization char's growth hormone genes and their application in biotechnology

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Growth hormone (GH), also known as somatotropin, plays an important role in regulation of somatic growth. In addition, it is involved in numerous physiological processes including maintenance of ion balance, lipid and protein metabolism, growth, reproduction, immunity response and affects behavior. A crucial physiological role of somatotropin suggests that the growth hormone gene sequence should be conserved. Indeed, most vertebrate taxa, possessing a single gene copy, show a conserved somatotropin sequence. But, in salmonids' genome the GH gene is represented by two unrelated paralogous genes *gh1* and *gh2*. Both genes exist throughout the time of divergence of species in this group. Therefore, salmonids are a suitable model system for investigating the origin, evolution, and functions of duplicated genes. We isolated two types of growth hormone genes from four *Salvelinus* species and determined their complete nucleotide sequence.

A comparison of the complete paralogous GH genes *gh1* and *gh2* of salmonids has shown that the conserved regions are associated with exons, and the variable regions correspond to intron sequences. It should be noted that not all intron sequences are variable; conserved regions can also be found. The presumable regulatory elements, localized in some introns (Pit-1 motifs, CRE, ERE), are also conserved. Also, we identified the potentially functionally important regions in promotor. There are a TATA box, which is typical of most eukaryotic genes and other regulatory elements, such as A/T – rich regions (Pit-1 sites), cAMP response elements (CRE), a glucocorticoid response elements (GRE) and RARE/RXRE elements, which are responsible for interaction with other transcription factors.

As is known, gene constructs containing the GH gene have been widely used for transformation of various fish species. We are going to use the obtained construction for transformation both model and economic fish. It is noteworthy that the use of transgene elements homologous to both the structural gene and promoter region sequences are more effective than heterologous ones for growth hormone transgenesis. In this structure as the GH promoter and a structural part of a gene derived from the same char species and the structural part of the gene comprises exons and introns, which provides stability of transformed species. Gene transfer provides a powerful tool for the study of fish development and the determination of the role of different genes.

Antioxidant activity and hepatoprotective potential of shellfish protein hydrolysates

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In recent years, fish protein hydrolysates (PH) have attracted much attention of food biotechnologists due to the availability of large quantities of raw material for the process, and presence of high protein content with good amino acid balance and bioactive peptides (antioxidant, immunomodulatory and antimicrobial peptides). Enzymatic hydrolysis of proteins is often employed to improve the functionalities of food proteins. Hydrolysis can be used to improve or modify the physicochemical, functional properties of native proteins.

Shellfish tissue (*Mercenaria stimpsoni*, *Corbicula japonica*, *Anadara broughtoni*) formed the material of the study. Frozen whole shellfish minced muscle, including viscera, was subjected to protein recovery using the extraction of fish muscle protein.

In the present study, shellfish frame proteins were separately hydrolyzed by alcalase 0.6 L, neutrase 0.5 L, protamex 1.5 MG, flavourzyme 500 MG. Enzyme activity was high in range of 5.5–8.0, but showed considerable loss of activity at pH 10.0. The optimal pH for hydrolysis of shellfish protein extract was 7.0. The type of enzymes used and the conditions of the hydrolysis had a marked effect on the yield of the final product. The degree of hydrolysates (DH) of *M. stimpsoni*, *C. japonica* and *A. broughtoni* was observed to be 15.4, 12.6 and 22.2% for protamex-treated hydrolysate, respectively, the other proteolytic enzymes showed DH values lower than 10%.

The antioxidant activity of a substance can be identified by assessing scavenging activities on free radicals generated in oxidative systems. Peptides from shellfish PH were evaluated for their antioxidant activities using scavenging 1,1-diphenyl-2-picrylhydrazyl (DPPH) activity assays. The DPPH radical scavenging activity of all samples was evaluated and their EC₅₀ values are presented. The DPPH radical scavenging activity enhanced with the increase of DH for shellfish PH from *M. stimpsoni*, *C. japonica* and *A. broughtoni*. Among the three hydrolysates, the highest antioxidative activity was observed in the *Corbicula* protein hydrolysate and its EC₅₀ values was 11.8 mg/ml.

To confirm the antioxidant capacity as well as the hypothesis of hepatoprotective activity on shellfish PH, HepG2 cells were co-treated with tert-butyl hydroperoxide (t-BHP) and different concentrations of PH for 24 h. The viability of HepG2 cells treated by 0.4 mM t-BHP alone decreased to 66.4±4.8% of normal control group. Shellfish PH prevented t-BHP-induced cell death ($p \leq 0.01$) and the cytotoxicity-inhibitory activity was dependent on the concentration of PH. The highest protection (82.3±4.4) was achieved by *Corbicula* PH. These results demonstrated that the antioxidant activities of PH were positively correlated with the improvement of the cell viability.

Biodiversity learning and use: DNA barcoding as a tool for progress at regional (RUS-BOL) and global (iBOL) levels

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The world-wide, iBOL (www.ibol.org), as well as the regional Russian, RUS-BOL (<http://www.wimb.dvo.ru/misc/barcoding/index.htm>), biodiversity programs are in focus of this report. Based on molecular markers (MM) of genes, delimitation of species identities was performed for over 500 members of several taxa of fish and shellfish from the Far East seas of Russia. This work has essentially improved the understanding of the systematics of these taxa and identification of individuals, including many objects of fishery, thus having both a fundamental and an applied value. According to the general iBOL standards, and firstly, in Russia, the complex approach for biodiversity description (CABD) is realized. The CABD includes 5 basic items: (1) accompanying all type (or paratype) specimens with color digital photographs, (2) holding all type (or paratype) specimens with voucher documentation, (3) maintenance of cryo-preserved or alcohol-preserved collections of tissues, (4) creation and management of own database (DB) by iBOL standards, and (5) integrating our own data into a world DB, starting with BOLD (www.boldsystems.org), GenBank (www.ncbi.nlm.nih.gov), and others.

Several approaches, based on MM, are applied as a work bench for DNA barcoding: 1. Gene tree building by MM data (usually nucleotide sequences); 2. Estimation of sustainability of gene trees based on several techniques (ML, MP, BI, NJ), and searching for the consensus gene and species trees; 3. Searching for specimens in DB, their classification according to the identity mode, and their identification (delimitation) by means of coalescent approaches, such as PTP (Poisson Tree Processes), GMYC (Generalized Mixed Yule Coalescent), etc.; 4. Searching for gaps in MM of DBs, based on non tree-like techniques, e.g. ABGD (Automatic Barcoding Gap Discovery), etc.; 5. An empirical estimation of identification precision of all specimens in BOLD DB by so-called BIN (barcoding index number) descriptor which is associated with the voucher lists of specimens from museums (museums' collections).

Currently, 6 research works may be listed as the NSCMB achievements in DNA barcoding: 1. Molecular systematics research, DNA barcoding of Altai Osman, *Oreoleuciscus* (Pisces, Cyprinidae, Leuciscinae), and nearest relatives, inferred from sequences of cytochrome *b* (*Cyt-b*), cytochrome oxidase *c* (*Co-1*), and complete mitochondrial genome (Kartavtsev et al., 2016; WoS IF=1.76); 2. Molecular-phylogenetic reconstruction and taxonomic investigation of eelpouts (Cottoidei: Zoarcales), based on *Co-1* and *Cyt-b* mitochondrial genes (Turanov et al., 2016; WoS IF=1.76); 3. DNA barcoding for squids of the family Gonatidae (Cephalopoda: Teuthida) from the boreal North Pacific

(Katugin et al., 2015; WoS IF=1.76); 4. The colonial ascidian *Didemnum vexillum* Kott, 2002 is an alien species in Peter the Great Bay (Sea of Japan) (Zviagintsev et al., 2016); 5. *Janolus fuscus* O'Donoghue, 1924 (Gastropoda, Proctonotidae), a species of the nudibranch family that is new to the marine fauna of Russia (Chichvarkhin et al., 2016; WoS IF=0.61).

In brief, iBOL Current Progress comprises (i) 482,051 barcode clusters for animals (BINs), (ii) a total of 6,016,392 of all sequences, and (iii) 5,219,015 barcode sequences. Over 10 Campaigns are scheduled worldwide: The Formicidae Barcode of Life campaign, the All Birds Barcoding Initiative, the Fish Barcode of Life Initiative (FISH-BOL), etc. RUS-BOL has its own contribution to the world initiative as its regional node. The BOLD search has found 30,634 published records, forming 6,820 BINs, with specimens from 1 country, deposited in 232 institutions. Of these records, 12,421 have species names and represent 3,448 species.

The analysis of trophic relationships in bottom ecosystems of the Far Eastern seas using marker fatty acids

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Marker fatty acids (FA) are widely used in food web research. We conducted a large-scale study of FA composition of more than 200 species of marine benthic invertebrates. Among them, FA compositions of foraminifera, sponges, mollusks (bivalve and scaphopods), and echinoderms were of special interest. Foraminifera, together with sponges, were sources of many unusual polyunsaturated FA, but, unlike sponges, they contained “normal” even chain FA with methylene interrupted double bonds. FA compositions of demosponges with and without bacterial symbionts were very different. The C₁₄–C₂₀ FA composition of the latter group of sponges was close to those of glass sponges. Scaphopods that are specialized to feed on foraminifera have a high concentration of arachidonic acid, typical of many benthic foraminifera. A widely diverse fatty acid composition, which reflects the differences in feeding modes, was found in lipids of bivalve mollusk. Data on FA composition of marine invertebrates were used to analyze the trophic structure of marine ecosystems. These studies in the Sea of Japan were carried out in the seagrass ecosystem, in the ecosystem of subtidal sands, in marine rocky intertidal habitat, and in the bathyal and abyssal ecosystems. Especially interesting was the trophic structure of the abyssal zone in the Sea of Japan, where the key invertebrate species are carnivorous. In the Sea of Okhotsk, the studies were conducted on the continental shelf, as well as in the bathyal and abyssal zones. It was found that many invertebrates in the bathyal zone of the Sea of Okhotsk utilize more refractory organic matter than invertebrates which inhabit the continental shelf. The abyssal zone of the Sea of Okhotsk is characterized by a high biomass of surface and subsurface deposit feeders. In food webs of the Kurile-Kamchatka Trench, rapidly sedimented algal detritus plays a more important role than in food webs of the surrounding abyssal ecosystems.

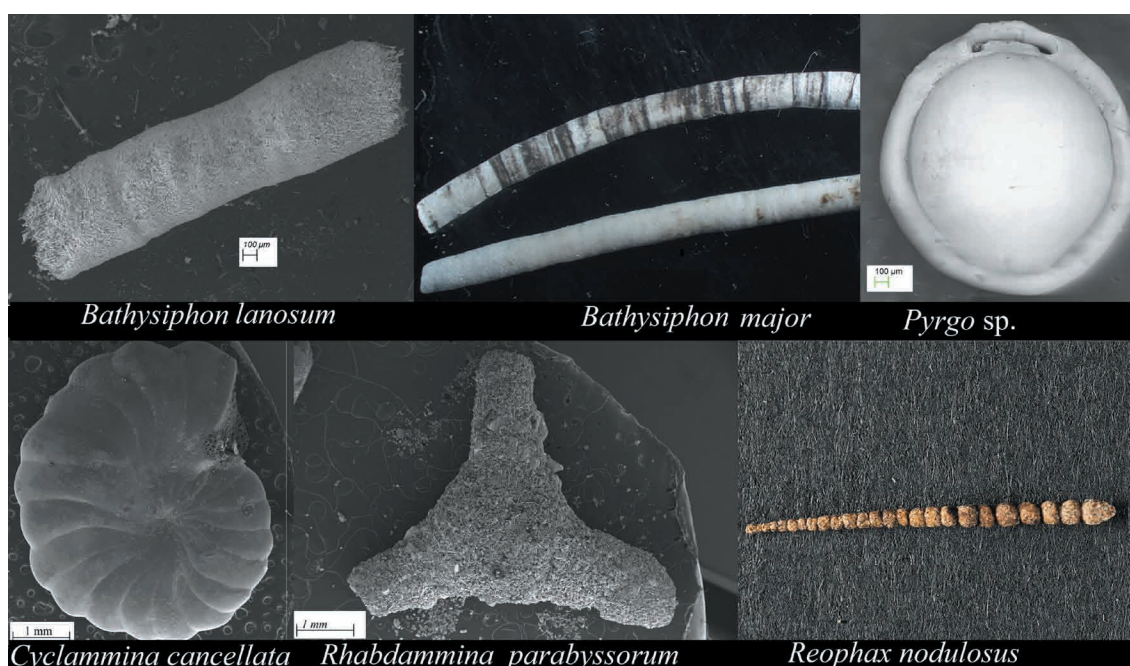
This work was supported by the Russian Foundation for Basic Research (RFBR), project No. 16-04-01477.

Fatty acids of deep-sea Foraminifera

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Foraminifera are an important component of marine benthic communities, especially in abyssal ecosystems. They are able to consume a wide range of food sources, which affect their fatty acid composition. We analyzed the fatty acid composition of 12 species of deep-sea benthic foraminifera (photos of these foraminifera are presented in Figure). It allowed us to determine the food sources of these species, and establish the role of foraminifera as sources of polyunsaturated fatty acids in deep-sea ecosystems. Among the species analyzed, we found the carnivorous foraminifera of the genus *Pilulina*, which feed on bottom copepods and abyssal foraminifera of the genus *Hippocrepinella*, consuming fresh diatoms. Foraminifera are able to synthesize a variety of polyunsaturated fatty acids, from C_{18} to long-chain fatty acids C_{26} . Most deep-sea foraminifera contain high concentrations of 20:4n-6, often significantly exceeding the concentrations of 20:5n-3. The diversity of polyunsaturated fatty acids in foraminifera exceeds the diversity of polyunsaturated fatty acids in other



Foraminifera from Kuril Basin (the Sea of Okhotsk).

systematic groups, including sponges and thraustochytrids. Analysis of the fatty acid composition of the common deep-sea species confirmed the assumption that foraminifera are an important component of deep-sea food webs and strongly affect the fatty acid composition of benthic invertebrates.

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Sea anemones and biological activities of their extracts (the insular shelf and slope of the Kurile and Commander Islands)

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The present work is based on samplings along the insular shelf and slope (above 100 m and up to 500–600 m depth) of the Kurile and Commander Islands in 2008–2015, during cruises of the R/V “Akademik Oparin” (Fig. 1). According to literature and own data, based on a modern



Fig. 1. Schematic map of the sea anemone samplings along the insular shelf and slope of the Kurile and Commander Islands.

higher-level taxonomic classification of Hexacorallia, the sea anemone fauna of the insular shelf (subtidal zone) and slope (bathyal zone) of studied region includes about 30 species belonging to Actiniaria (Anenthemonae and Enthemonae suborders) and Corallimorpharia orders. In the collected material, at least 13 species of sea anemones are taxonomically identified. Among Enthemonae, species of *Cribrinopsis* and *Urticina*, *Actinostola callosa* (Verrill, 1882), *Stomphia coccinea* (Muller, 1776), and *Liponema brevicorne* (McMurrich, 1893) mainly prevail. Other groups of sea anemones presumably represent by only 1–2 species in 100–600 m depth (*Actinernus* cf. *robustus* (Hertwig, 1882) of Anenthemonae as well as *Corallimorphus* cf. *pilatus* Fautin, White et Pearson, 2002 of order Corallimorpharia often occur) (Fig. 2).

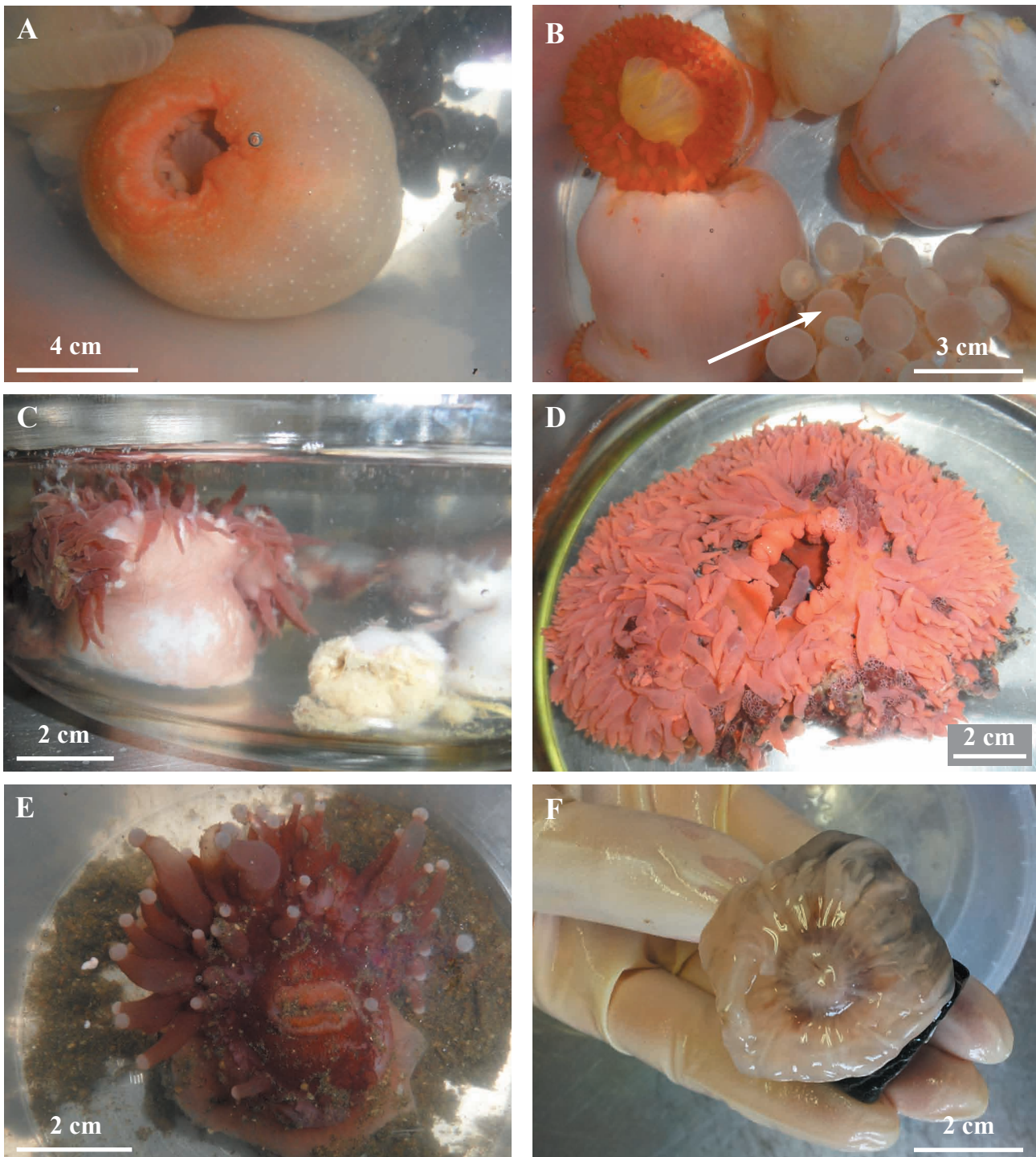


Fig. 2. Some sea anemones inhabiting the insular shelf and slope of the Kurile and Commander Islands. **A** – *Cribrinopsis* sp. (Simushir Island, 46°55.08' N, 152°06.8' E, from 113 m depth), **B** – *Actinostola callosa* (Chirpoy Island, 46°23.8' N, 150°46.5' E, from 145 m depth; “giant larvae” are indicated by arrow), **C** – *Actinernus* cf. *robustus* (Urup Island, 46°00.7' N, 149°52.4' E, from 210 m depth), **D** – *Liponema brevicorne* (Simushir Island, 46°56.6' N, 152°16.3' E, from 447 m depth), **E** – *Corallimorphus* cf. *pilatus* (Urup Island, 45°57.9' N, 149°24.4' E, from 560 m depth), **F** – *Corallimorphus* sp. (Bering Island, 54°34.0' N, 166°51.7' E, from 250 m depth).

Cribrinopsis spp. and *Urticina* sp. (Actiniidae) are typically subtidal species, but they can be found in the upper bathyal zone (Iturup, Simushir, and Rasshua islands, Kurile Islands, 103–370 m depth). *S. coccinea*, *A. callosa*, *Actinostola* sp., and *Actinostola* cf. *faeulentata* (McMurrich, 1893) (Actinostolidae) dwell in both the subtidal zone and bathyal zone (Iturup, Urup, Chirpoy, and Simushir islands, Kurile Islands, 118–223 m depth; Bering Island, Commander Islands, 207–320 m depth). In the northern Pacific, *A. callosa* and *A. faeulentata* live up to 2 km depth. *A. robustus* (Actinernidae) is typically bathyal species, and it can go down above 3 km depth (*A.* cf. *robustus* is found at Urup Island, Kurile Islands, in 210–490 m depth). *C. pilatus* (Corallimorphidae) is also the bathyal species living up to 2 km depth (*C.* cf. *pilatus* is found at Iturup and Urup islands, Kurile Islands, in 290–560 m depth). In general, species of *Corallimorphus* are typically bathyal species, but they can go down to the upper abyssal (above 4 km depth) and also go up to the lower part of the shelf. *Corallimorphus* spp. occurred at Chirpoy Island, Kurile Islands, in 455 m depth and Bering Island, Commander Islands, in 250–289 m depth. *L. brevicorne* (Liponematidae) inhabits both the bathyal zone and upper abyssal zone (above 4 km depth); sometimes this species can be found in the shelf up to the upper subtidal zone (46 m depth, Bering Sea). It is found at Simushir and Onkotan islands, Kurile Islands, in 113–447 m depth and Bering Island, Commander Islands, in 153 m depth.

Most species (species of *Cribrinopsis*, *Urticina*, *Actinernus*, and *Corallimorphus*) attach themselves to the bedrock outcrops, boulders, gravel, shell debris, and other hard surfaces often with sandy or silted overlay. *S. coccinea* also prefers bouldery-gravelly substrata (sometimes occurs in silted seabed), but it can detach from substratum and swim. *A. callosa* can be found in both sandy or silted bottom and hard surfaces, but it is not always attached to the substratum like *S. coccinea*. In the Sea of Okhotsk insular shelf of Urup Island, *A. callosa* inhabits volcanic sands. *L. brevicorne* lives unattached on bottom of loose substrata (sand, silt, fine gravel, and shell debris) sometimes with admixture of small boulders.

The aqueous and ethanol extracts of sea anemone collected in the studied region were tested to explore their hemolytic, cytotoxic, antimicrobial, and antifungal activities, as well as inhibitory activity for trypsin, α -galactosidase and its mutant form – C493N. The water extracts of tentacles of sea anemones *A. callosa* (Commander Islands) and *L. brevicorne* (Kurile Islands) and also water-ethanol extract of *L. brevicorne* (Commander Islands) exhibited high hemolytic activity. On rat splenocytes, the ethanol extract of *A.* cf. *faeulentata* (Commander Islands) exhibited high toxic activity. The aqueous extracts of tentacles of *S. coccinea*, *Actinostola* sp., and *Corallimorphus* sp. of the Kurile Islands were effective against bacteria *Bacillus subtilis*, *Staphylococcus aureus*, and pathogenic yeast *Candida albicans*. However, the ethanol extract of *Corallimorphus* sp. showed antibacterial activity only for *B. subtilis*. The aqueous extract of *L. brevicorne* (Kurile Islands) restrained growth of the *B. subtilis* colonies, and extracts of *Cribrinopsis* sp. (Kurile Islands) and *A. callosa* (Commander Islands) were effective only against *S. aureus*. Interestingly, the extract of *L. brevicorne* of the Kurile Islands showed a pronounced antimicrobial activity against *C. albicans*, but, at the same time, it did not contain active cytolytic substances. Any investigated specimens had no demonstrable a trypsin-inhibitory activity. However, the ethanol extracts of *A.* cf. *faeulentata* of the Commander Islands and *L. brevicorne* of the Kurile Islands inhibited the activity of enzyme α -galactosidase, and extract of *A.* cf. *faeulentata* also inhibited the activity of mutant forms C493N of the enzyme.

Marine biological resources of India: conservation and management

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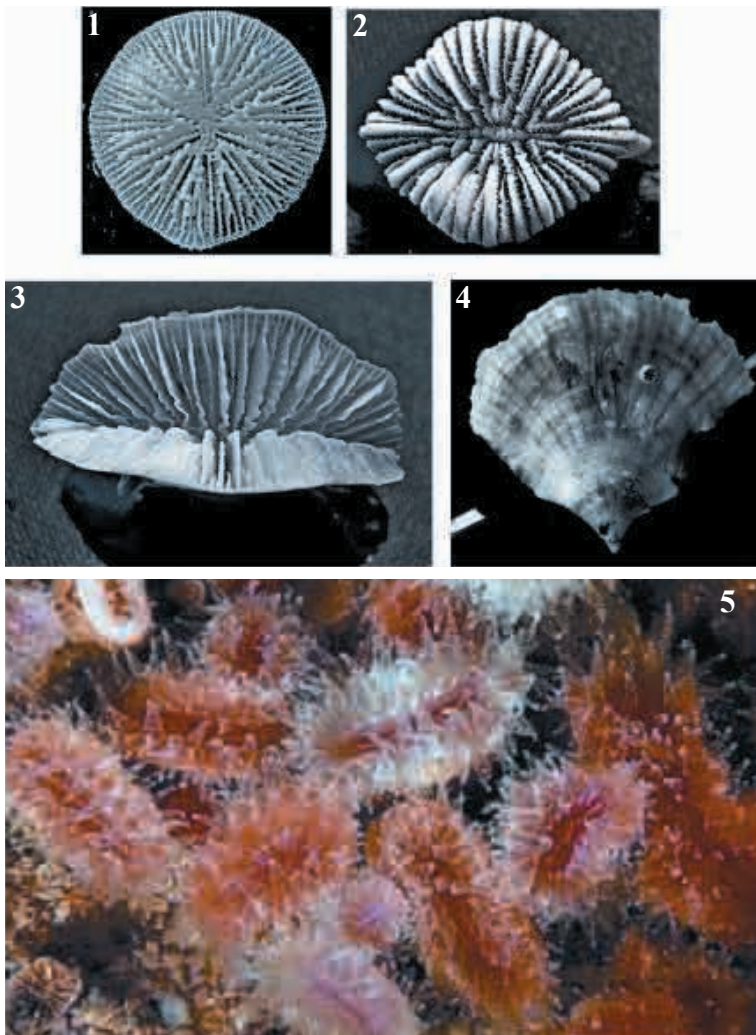
India is one of the 12 mega biodiversity countries of the world. With a coastline of 8129 km bounded by the Arabian Sea to the West and the Bay of Bengal to the East and Exclusive Economic Zone (EEZ) of 2.02 million km², India harbours 15% of the global biodiversity. Of the 32 animal phyla, 15 are represented in India's marine ecosystem with more than 15,000 species. The recent blue revolution and the blue growth initiative recognizes the importance of sea and oceans as drivers of the blue economy having potential for growth and innovation with a focus on marine bioresources, sea farming, marine biotechnology, industry and fishing harbours. The newly launched Sagar Mala Scheme envisions fishermen's welfare through a revolution in coastal development by creating industrial and tourism hubs. A new multi institutional national program (Phase II) on Drugs from the Sea has been initiated for the discovery of a variety of unique and biologically potent natural products. Government of India has established several marine protected areas for the conservation of fauna and flora because of increased fishing activities, pollution, climate change and aquaculture practices which are threatening the coastal and marine genetic resources. The Government of India has brought into force a number of laws (the Biological Diversity Act – 2002, and Biodiversity Rules 2004, the Indian Wildlife Protection Act 1972, the Marine Fisheries Regulation Acts of Indian Maritime States) and the recent National Marine Policy, 2016. The presentation summarizes the current state of knowledge and future strategies for the conservation and management of marine bioresources including the application of modern and innovative technologies, ecosystem based approach and national and international regulations.

New information about the solitary deep-water corals of the Vietnamese shelf

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Described solitary corals collected of the shelf Vietnam when deep-water drilling on the R/V “Akademik Oparin” in 2007 and 2016. In general were identified 54 species, 16 genera attributed to the 9 families. Tree species were not previously known to science, 14 species not previously known for Vietnam. Most massive settlements have formed two species *Flabellum pavonium* (21 species in samples) and *Tropidocyathus longiseptum* sp.nov (23 species in samples). Most taxonomically rich coral genera were *Flabellum* and *Truncatoflabellum* (see Figure).



The high density of corals noted at stations with depths 90 and 180 m, the greatest variety at a depth of 120–136 and 285–390 m. Deep water corals in Vietnam (54 species) are integral with the coral fauna of the Indo-Polynesian Province. These data complement the general characteristics of the knowledge of the composition, morphology and chorology world fauna solitary ahermatypic coral. Rich in species diversity, as a single deep-water corals (for Vietnam 54 species) and marked by high diversity of previously hermatypic scleractinian (376 species) waters of Vietnam (Latypov, 2016) confirms the earlier opinion (Latypov, 2011, 2014) that coast and shelf of Vietnam constitute a single unit in the coral fauna of Indonesian-Malaysian Centre of its origin and belong to the Indo-Polynesian province of Indo-Pacific region.

1 – *Stephanophyllia neglecta*, 2 – *Tropidocyathus lesson*, 3–4 – *Flabellum politum*, 5 – *Flabellum in situ*.

Estimates of species richness of bivalve mollusks in various parts of the Sea of Japan

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Biodiversity of the bivalve mollusks of the Sea of Japan is of high interest as Japan and neighboring areas (China and South China Sea) are the most richest areas in terms of species richness. The Sea of Japan lies in two different geographical and climatic zones – temperate in the north, and subtropical in the south, thus, making the fauna a combination of species of various origins, boreal, boreal-arctic and tropical. A complicated geological history and oceanography of the Sea of Japan influence biodiversity patterns of molluscan fauna. Biogeography and biodiversity of bivalve mollusks in the Sea of Japan are relatively well-known up to date except for North Korea. There are comprehensive catalogues of bivalves for both eastern (Higo et al., 1999) and western (Lutaenko, Noseworthy, 2012) parts of the sea but there were no attempts to compile a check-list for the entire basin.

Kantor and Sysoev (2005) estimated that the Russian part of the sea is inhabited by 169 species of bivalves. According to literature data, species richness of bivalve faunas in large bays and other marine localities of the Sea of Japan varies from 88 (Vostok Bay, Russia) to 211 (Sado Island, Japan) (see Table). In order to preliminarily estimate total species richness of bivalves in the Sea of Japan,

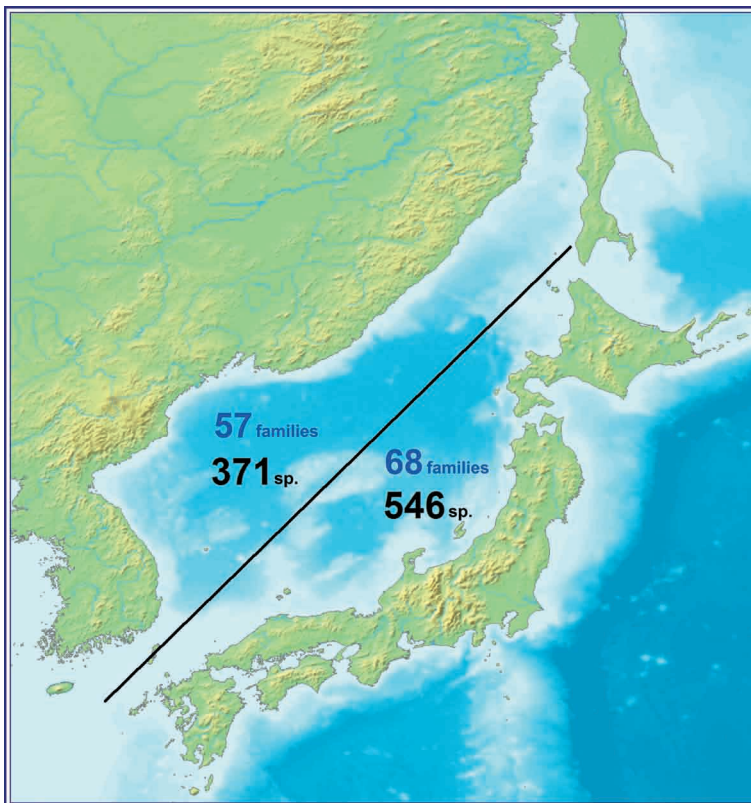
Species richness of bivalve molluscan faunas in various localities in the Sea of Japan

Locality	Species richness	Reference
Wakasa Bay (Honshu)	207	Ito (1990)
Tsukumo Bay (Noto Peninsula, Honshu)	104	Habe (1973)
Ichikawa Prefecture (includes Noto Peninsula)	119	Ito et al. (1986)
Sado Island	211	Kuroda (1957); Honma, Kitami (1978, 1979, 1995)
Mano Bay (Sado Island)	103	Ito (1978)
Yeongil Bay (Korea)	98	Lutaenko et al. (2003, 2006)
Possjet Bay (Russia)	97	Golikov, Scarlato (1967); Scarlato (1981)
Vostok Bay (Russia)	88	Evseev (1981)
Amursky Bay (Russia)	119	Lutaenko (2002, 2003)
Ussuriysky Bay (Russia)	124	Lutaenko (2005, 2006)
Western Tatarsky Strait (Russia)	99	Dulenina (2013)

we have compiled a list of bivalves separately for the western and eastern parts of the sea based largely on above-mentioned catalogues. A list for Sakhalin is not prepared yet while two separate lists for western and eastern Sea of Japan have been processed but they are not fully merged: there are some taxonomic problems and various treatments of species validity and synonymy among Russian, Korean and Japanese malacologists and we have to critically analyze some species. The western part of the Sea of Japan is inhabited by 371 species and subspecies of bivalve mollusks. Among them, 316 species are known for South Korea, 50 species for North Korea, and, in Russia, 163 species for southern Primorye (Peter the Great Bay), and 130 species for middle/northern Primorye, the area of the continental coast of the sea from Cape Povorotny to Tatarsky Strait. The South Korean fauna is quite rich, and is comparable with that of the East China Sea, which has 337 species (Xu, Zhang, 2011). The species richness of bivalves along the western shore of the Sea of Japan shows a clear latitudinal gradient from north to south which is a fundamental pattern of ecology and biogeography: the increase in biological diversity from polar to equatorial regions. In the eastern part of the sea (excluding Sakhalin shelf), 546 species and subspecies of bivalves are known based on tentative calculations. There is difference in family richness between eastern and western parts: 57 families are found in western part against 68 families in eastern part of the Sea of Japan (see Figure). At least, ten families of bivalves are not known in the western part but present along the Japanese coast: Tindariidae, Nucinellidae, Cucullaeidae, Malleidae, Sportellidae, Tridacnidae, Cardiliidae, Pholadomyidae, Parilimyidae, Verticordiidae. Preliminary observations show that 1, eastern sector of the Sea of Japan is more species-rich (175 species difference); 2, the degree of penetration of tropical-subtropical

and subtropical species to the north in eastern part is higher due to warming effect of Tsushima Current; 3, eastern sector contains more warm-water-species.

We suggest that about 600 species of bivalves inhabit the entire sea. This is first estimate of total bivalve species richness for the Sea of Japan. This is sufficiently higher than in neighboring seas: 87 and 175 species are known for Bohai and Yellow seas, respectively (Xu, Zhang, 2011), about 288 species of bivalves were found in all Russian Far Eastern seas (Lutaenko, 2013), and 337 species are identified for subtropical East China Sea. Bivalve mollusks show a low level of endemism in the Sea of Japan, and only two species living in the western part of the sea can be regarded as endemics. However, thorough analysis of the full list of bivalves of the entire sea may reveal more endemics in the eastern part.



Family and species richness of bivalves in eastern and western Sea of Japan.

The main deep sea habitat types of the Far Eastern Seas of Russia

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“Deep sea” is defined here as sea floor area below 150 m, where sunlight penetration is too low to support photosynthetic production. The geological, physical and geochemical settings of the deep-sea floor form a number of different habitats with unique characteristics that support very specific benthic communities. Approximately 9000 species of free-living invertebrates inhabit the Far Eastern Seas of Russia and Kuril-Kamchatka Trench and great many of them have been found in the deep sea environment. In the present report the following habitats are discussed.

Coral gardens. Northern Pacific deep sea coral communities consist of predominantly by gorgonians, alcyonaceans and sea pens, which form so called “coral gardens” (Fig. 1). The bottom surface covered by them is quite prominent and more than 40 species of different corals have been found in these communities. Coral gardens are situated along both sides of Kuril Islands.

Sponge fields. Polyspecific settlements of sponges are common in different sites of the Kuril Islands. At least a dozen of sponge fields have been already found in this region, but the real substrate coverage by them is extremely difficult to evaluate but undoubtedly it is huge. Similar to deep-sea coral gardens, the presence of large sponges creates a three-dimensional structure of the seafloor, increasing habitat complexity and attracting an invertebrate and fish fauna at least twice as rich as that on surrounding soft bottom substrates (Fig. 2).

Seamounts are topographically-isolated peaks with relatively steep (14 to 35°, sometimes more) slopes, rising more than 1000 m above the surrounding seabed with their summits do not reach the ocean surface and which do not situated on the continental slope, usually with a rocky substratum that is quite distinct from the sedimented deep-ocean floor. There are more than 110 seamounts and volcanoes nearby the Kuril Islands and off the Commander Islands, but only two of them have been studied in some details (Fig. 3).

Submarine canyons are noticeable and topographically complex features. They are valleys carved into the continental margin where they form deep incisions on the edge of a continental shelf and slope in a global scale and act as conduits for transport of sediment from the continent to the deep-ocean floor. As other habitats mentioned above, canyons are heterogeneous habitats. They are widespread features on most of the ocean’s continental and island margins. Submarine canyons of the Russian Far East Seas are very poor studied and mostly this attributes to the biota. One can find information only on Great Gamov Canyon.

Hydrothermal vents. In the region under consideration we can say with some confidence only about a few known manifestations of hydrothermal activity – on the Kuril Islands underwater slopes and on the Kashevarov Bank in the Okhotsk Sea.

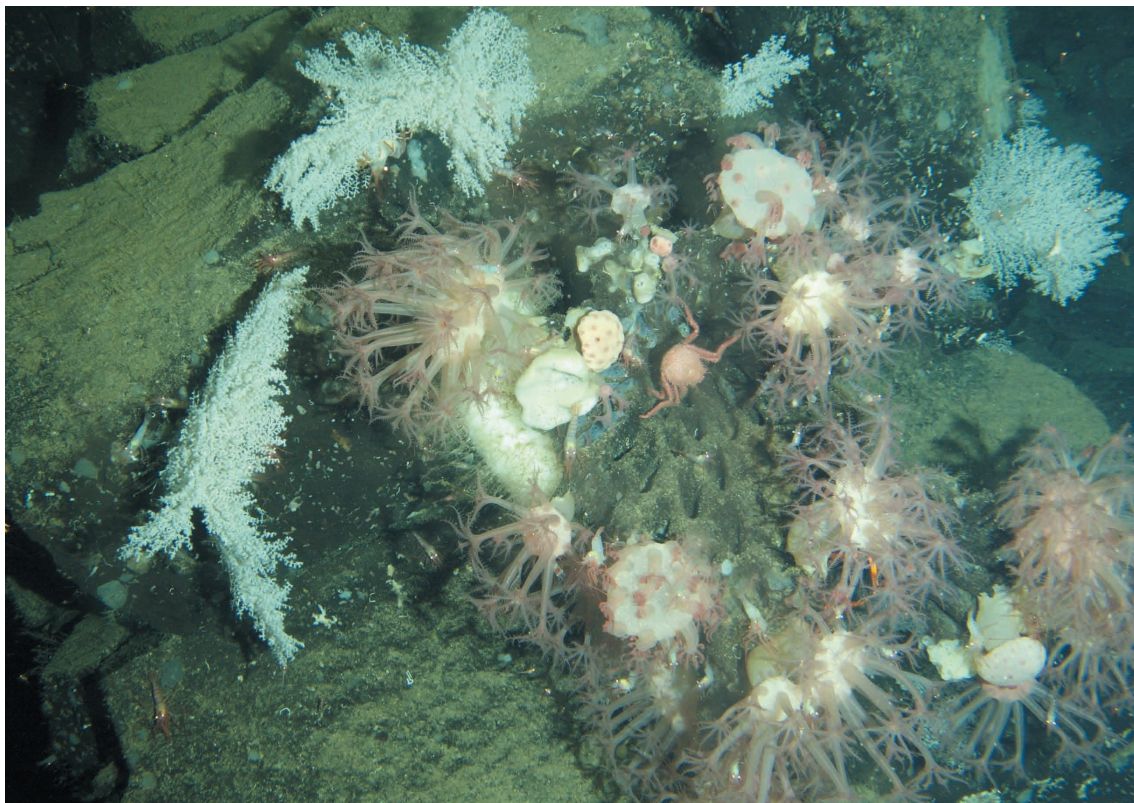


Fig. 1. Coral gardens.

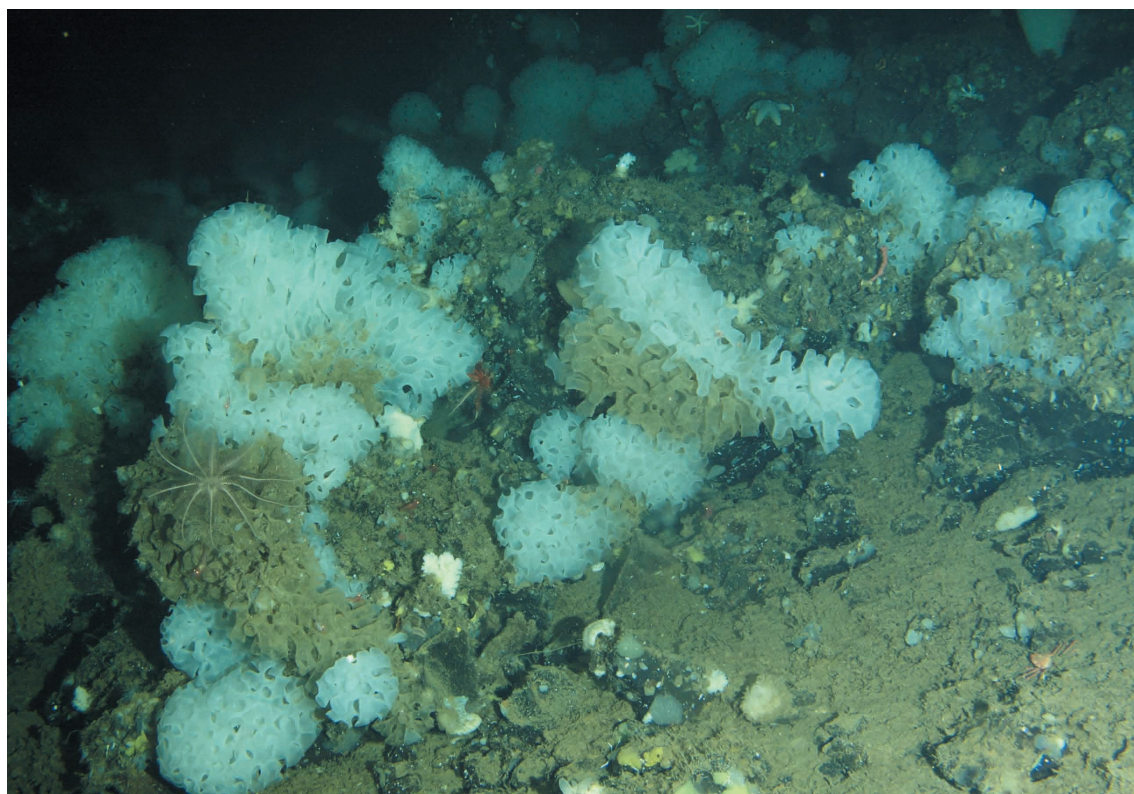


Fig. 2. Sponge fields.

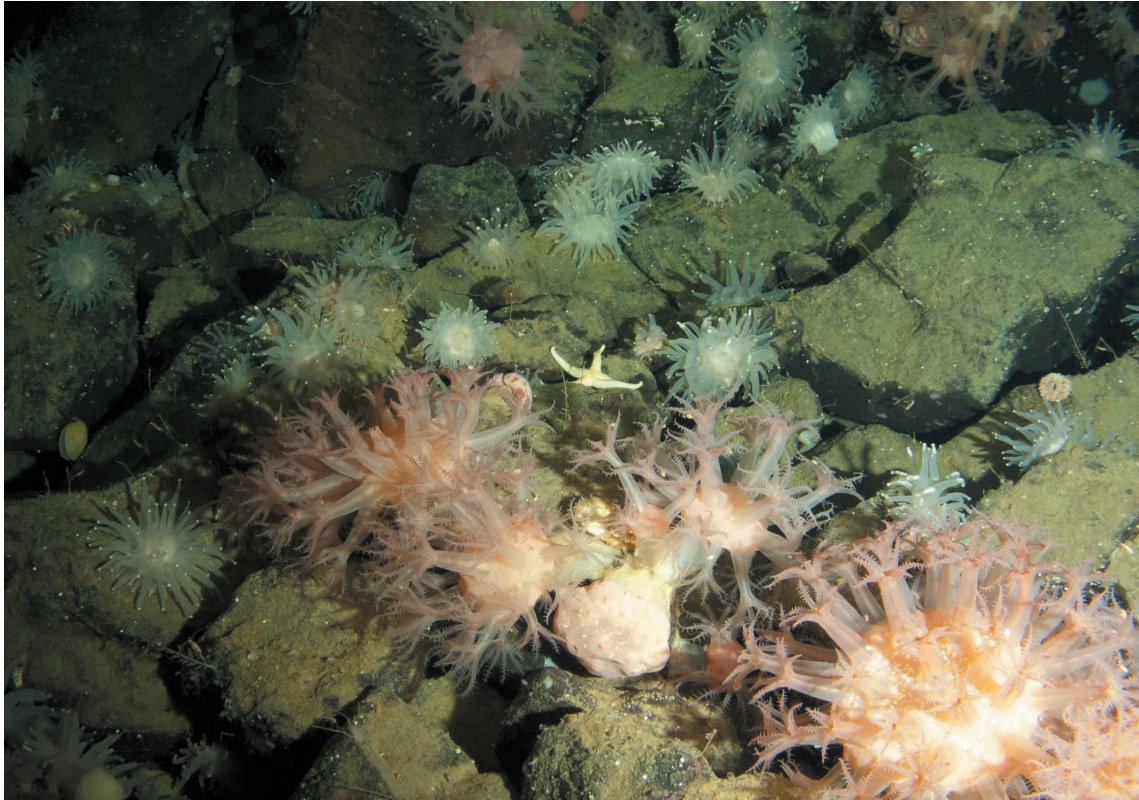


Fig. 3. Seamounts.

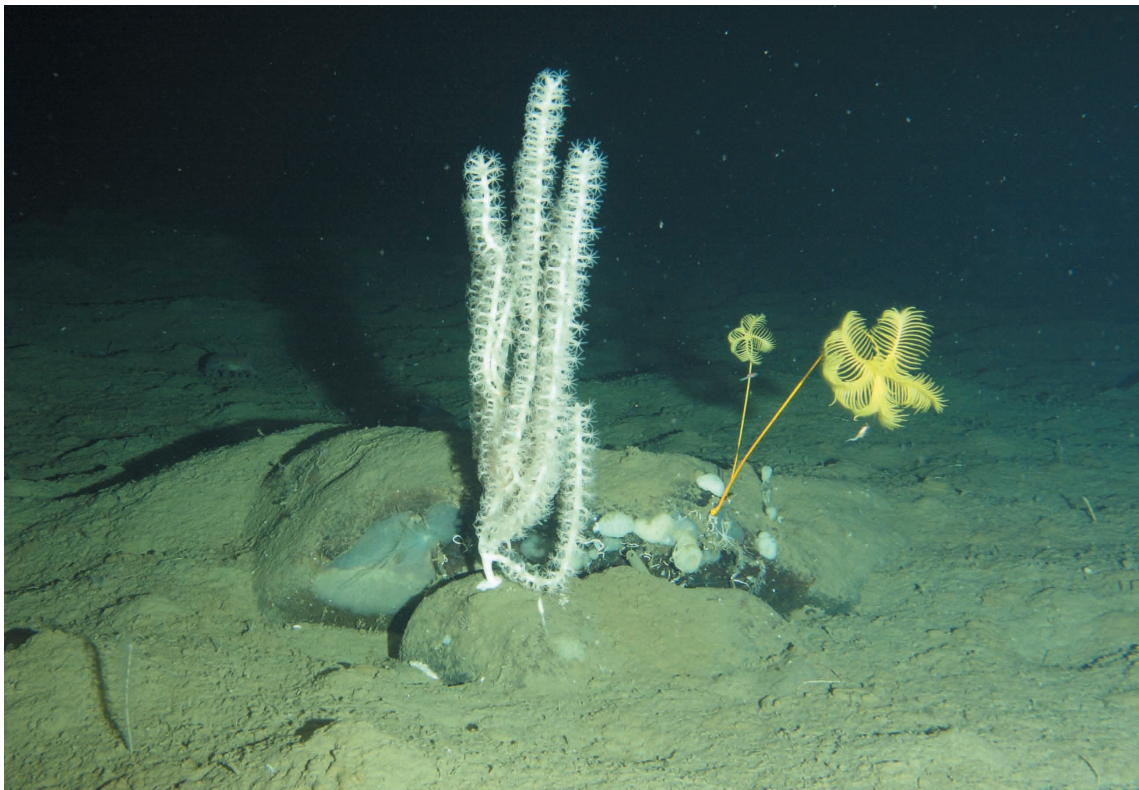


Fig. 4. Abyssal plains.

Cold methane seeps. Unfortunately, the biological study of the seep fauna in Russian waters until current century is virtually not conducted. It is possible to find a fragmentary information contained in the articles of marine geologists, geophysicists and of other non-biological specialists and much less – marine biologists. Just two seep areas have been investigated recently by biologists in the Okhotsk Sea – near Paramushir Island and in the Deryugin Basin.

Abyssal plains. These areas of the deep ocean floor usually situated between the foot of a continental slopes and a mid-ocean ridges. Vast abyssal plains, flat or sometimes hilly, look like a kind of “underwater desserts”, but in fact they are quite inhabited areas (Fig. 4).

Trenches. Deep ocean trench is any long, narrow, steep-sided depression in the ocean bottom (“scars” on the planet surface) in which maximum oceanic depths occur. This is an area where old ocean crust is subducted beneath the margins of continental plates. The Kuril-Kamchatka Trench has been investigated only twice, but samplings demonstrate a rich bottom fauna of the trench.

Comparison of polar lipidoms of the soft corals from tropical and cold-water regions

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Each acyl lipid class consists of lipid molecular species with different acyl groups, and the sum of all molecular species forms lipidom of the organism. Lipidomics, integrated with genomics, proteomics, and metabolomics, will contribute toward understanding how lipids function in biological systems. Lipidomic analyses were recently started for marine organisms. Initial data on the lipidom were obtained for some species of several groups of marine organisms, such as microalgae, seaweeds, sharks, mollusks, squids, and sea anemones. Corals are very important for both tropical and cold-water marine ecosystems. The information about coral lipidoms is very limited. The polar lipidoms of several soft coral species (*Simularia macropodia*, *Capnella* sp., and *Xenia* sp. from Vietnam shallow waters and the cold-water soft coral *Gercemia rubiformis* from the Sea of Okhotsk) were determined for the first time by HPLC followed high resolution tandem mass spectrometry. The chemical structure and the content of all phospholipid molecular species of the tropical symbiotic species and the cold-water species without endosymbiotic dinoflagellates were compared. Ethanolamine glycerophospholipids (PE), choline glycerophospholipids (PC), serine glycerophospholipids (PS), inositol glycerophospholipids (PI), and ceramide aminoethylphosphonate (CAEP) were the major polar lipid classes. 18:1e/20:4 PE, 18:0e/20:4 PC, 18:0e/24:5 PS, and 18:0/24:5 PI were the major molecular species of the tropical corals, whereas 16:1e/20:4 PE, 16:0e/20:4 PC, 20:1/24:5 PS, and 16:0/24:5 PI predominated in phospholipids of the cold-water species. PE, PC, and PS mainly consisted of alkyl acyl and alkenyl acyl forms, but diacyl forms predominated in PI. Two very-long-chain tetracosapolyenoic acids (TPA, 24:5n-6 and 24:6n-3) were the principal fatty acids in PS and PI, whereas 20:4n-6 was more abundant in PE and PC. Selective incorporation of TPA in the molecules of PS and PI are supposed to be a specific feature of the biosynthesis of phospholipids in alcyonarians. Some molecular species of PS and PI with TPA may be used as molecular lipid markers during the study of symbiotic and trophic relationships of soft corals.

Cells number dynamic and photosynthetic apparatus function under copper pollution

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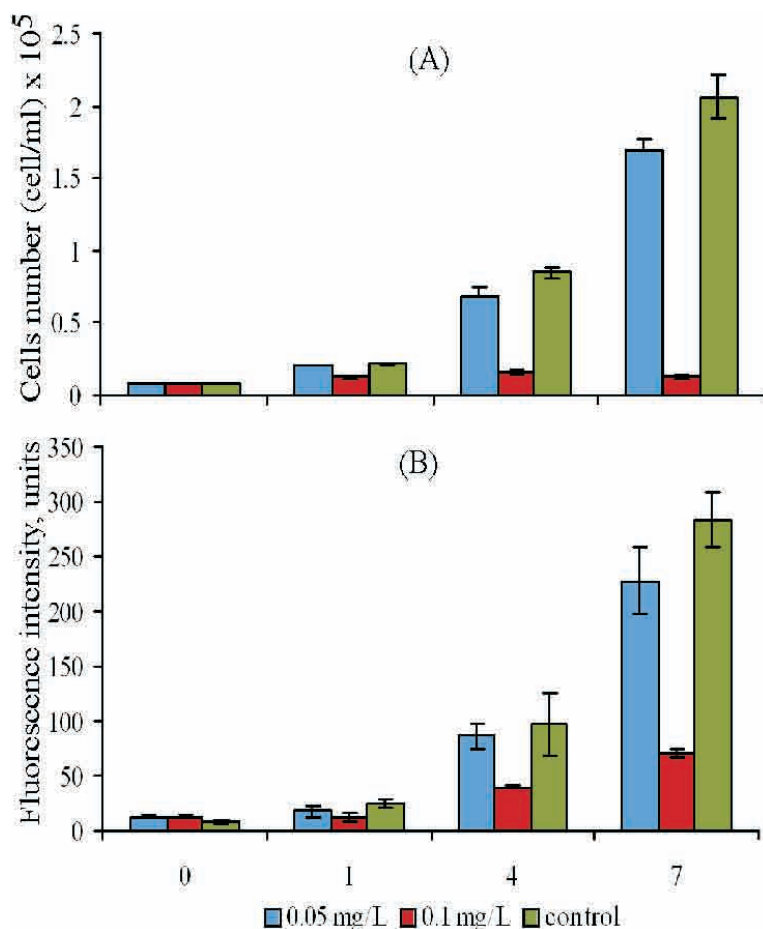
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Copper is one of the heavy metals that are most commonly applied in many industries and in agriculture. When entering a body of water, copper accumulates in bottom sediments, affecting the benthic organisms. Benthic microalgae are an important component of aquatic ecosystems

(Yuruela, 2005; Nagajyoti et al., 2010). Photosynthetic apparatus is the main source of energy for cells. A conventional technique, usually applied to diagnose its condition, is measurement of fluorescence chlorophyll *a* parameter (Voznesenskiy et al., 2016). The goal of this work was to study the effect of copper on the cells number dynamic and the laser-induced fluorescence (LIF) of chlorophyll *a* in the benthic microalgae *Attheya ussurensis* (Bacillariophyta).

The culture of the benthic diatom *Attheya ussurensis* Stonik, Orlova, Crawford (Stonik et al., 2006) was object of investigations. The laboratory-based culture was grown in the medium *f* (Guillard, Ryther, 1962), using 100 mL conical Erlenmeyer flasks, at a temperature of 20±2°C, a salinity of 32‰, a photosynthetic photon flux density (PPFD) of 70 μmol/m²s under fluorescent lamps, and with the light-dark period of 14:10 h. The medium was prepared in sterile seawater. Cells were counted in a Nageotte counting chamber 0.044 mL. LIF was excited



Parameters of the microalga *Attheya ussurensis* exposed to copper before subculturing: (A) number of cells; (B) laser-induced fluorescence level.

using continuous laser radiation at a wavelength of 442 nm and a power of 38 mW. The duration of sample's exposure to the laser radiation, was 2 s in all cases. The LIF spectra of phytoplankton were measured on a Shamrock 303i spectrometer (Andor Technology, USA).

The number of *A. ussurensis* cells increased gradually at a concentration of 0.05 mg/L Cu²⁺, but was slightly smaller than in the control during the entire experiment. A passage to the medium with 0.1 mg/L Cu²⁺ resulted in inhibition of growth: the number of cells remained unchanged throughout the exposure period (see Figure, A). The dynamics of LIF at 0.05 mg/L Cu²⁺ correlated with those for the number of cells. After passaging to 0.1 mg/L Cu²⁺, the fluorescence intensity was more lower than in control, but increased more intensively as compared with cells number and reached a quarter of the control intensity by the end of the experiment (see Figure, B).

Thus, copper at concentrations of 0.05 and 0.1 mg/L had an inhibitory effect on the population growth of *A. ussurensis* cells. Its negative influence on the state of alga's photosynthetic apparatus. The obtained results confirm the necessity of monitoring of bottom sediments for copper content.

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Preservation method of quality of sea urchin roe during storage

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Sea urchins are major components of marine communities and highly fecund, producing millions of gametes each year. Sea urchins are benthic marine echinoderms distributed across all of the world's oceans. Genus of *Strongylocentrotus* is important fishery catch in the Sea of Japan.

It is known that bioactive substances of sea urchin roe exhibit antioxidant, antitumor, hypolipidemic, anti-inflammatory, geroprotective, antimicrobial and other healing properties. And sea urchin roe regulates the hormonal system of the body, normalizes the functions of the mammary glands, contributes to restitution after injuries, illnesses and surgeries, increases the organism resistance to unfavorable ecological conditions. Therefore, some of the technologies of biologically of active food supplements and medicinal products have been developed based on sea urchin roe.

Sea urchin roe is consumed as fresh roe or processed product. In Japan, fresh roe are eaten both raw and cooked. There are various methods of conservation. The product can be consumed as non-heated (preserve) or heated (steamed, baked). Snack flavoured with sea urchin roe are also available. In Russia, the rational processing technology, providing quality and stability of sea urchin roe during storage, are underdeveloped. And food products with functional ingredients are not available to all population groups.

Oxidation and hydrolysis reactions are reasons of food spoilage of roe and roe products during storage. And methods to extend the viability of sea urchin roe have not received much attention.

The purpose of this study is to develop the ensuring food safety method of sea urchin roe and roe products during storage.

Our studies have indicated that the protein content of sea urchin roe of *S. intermedius* is $13.9 \pm 1.1\%$, *S. nudus* is $13.8 \pm 0.7\%$ The relative biological value compared to casein (protein standard) was $128.0 \pm 9.0\%$. The free amino acid content of the roe of *S. intermedius* is $3.2 \pm 1.1\%$, *S. nudus* is $2.9 \pm 0.8\%$.

The fat content of the roe of *S. intermedius* is $7.3 \pm 1.2\%$, *S. nudus* is $6.3 \pm 1.5\%$. Polyunsaturated fatty acids of total fatty acids are 35.3–42.1%. The ratio of omega-6 (32.2–34.3%) and omega-3 (30.9–33.4%) of fatty acids is 1:1, indicating the high value of sea urchin roe lipids. The phospholipids is 25.5–27.1% of total lipids roe. Research shows strong evidence that the phospholipids can help lower cholesterol and blood pressure.

The mineral composition showed that sea urchin roe is a source of magnesium, sulfur, iodine, selenium and other substances.

During the past few years, some interest has been shown in commercial production of frozen sea urchin roe. The freezing process and subsequent frozen storage can produce undesirable flavor and texture changes in sea urchin roe.

Studies have indicated that the water blanching of sea urchin roe for 20–30 seconds leads to inactivation of hydrolytic enzymes. The connective tissue protein coagulation of the roe surface film and the roe adjacent layer to the film occurs during the short-term heat-treatment. After freezing, sea urchin roe is stored without changing the quality and the chemical composition at minus 18°C for 4 months, at minus 25°C for 6 months.

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Search for new bacterial producers of tetrodotoxin (perspective analgesic agent) for pharmaceutical industry

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Marine organisms are a rich source of biologically active compounds with antibiotic, cytotoxic, and antitumor properties. Neurotoxins extracted from marine organisms are widely used in neuroscience, pharmacology and medicine. Tetrodotoxin (TTX) is one of the most studied low-molecular-weight toxins – a guanidine neurotoxin that selectively targets voltage-gated sodium channels in the nerve and muscle tissues. In the 1960s, TTX acquired wide popularity as an instrument of experimental neurobiology, although recently wider interest emerged in using this toxin in therapy for treatment of addiction, epilepsy, as local and general anesthetic and analgesic, and for other medical purposes. The widespread occurrence of TTX across phylogenetically diverse groups of organisms has led to a hypothesis about the bacterial origin of the toxin. Despite of numerous TTX-producing bacterial strains isolated from various TTX-bearing animals industrial toxin production for pharmaceutical manufacture on the base of bacterial cultures is yet to be developed. The main problems are lack of optimal conditions for bacterial cultivation and TTX synthesis, and rapid loss of the ability to produce the toxin by bacterial strains.

Many ribbon worms secrete different neurotoxins and use them for hunting and protection from predators. Some nemertean species have been shown to contain extremely high concentrations of TTX. The aim of the present study was to investigate taxonomic structure and properties of associative cultivable microflora of TTX-bearing and non TTX-bearing nemerteans and search for new TTX-producing bacteria.

The taxonomic composition of bacteria associated with the two TTX-bearing (*Hubrechtella juliae*, *Lineus alborostratus*) and two non-TTX-bearing (*QuasitetraSTEMMA stimpsoni*, *Malacobdella grossa*) species of ribbon worms was studied. Bacterial isolates were identified using 16S rRNA gene sequencing and phenotypic characteristics. 38 bacterial strains from 8 genera: *Pseudoalteromonas*, *Shewanella*, *Ruegeria*, *Pseudomonas*, *Defluviicoccus*, *Vibrio*, *Alteromonas* and *Bacillus*, were isolated. γ -Proteobacteria dominated among the associated microflora of nemerteans (76.3% of the total number of isolates). The sensitivity analysis of the strains to antibiotics of various classes revealed multiple resistance to three or more antibiotics in all of the studied isolates. The 15 bacterial strains isolated in the present study exhibited antimicrobial activities against at least one of five indicator microorganisms, most of which were correspondent to *Pseudoalteromonas* genus. Screening of the TTX-producing bacteria with confocal laser scanning microscopy and polyclonal antibodies against TTX revealed TTX-producing bacterial strain *Pseudoalteromonas* sp. among symbiotic microflora of *Hubrechtella juliae*. The correlation between the presence of TTX-positive microflora and the toxicity of nemerteans was determined.

A method of assessing the health of marine bivalves and the state of their habitat

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The assessment of the marine environment plays an important role in environmental safety. The classic method of the assessing physiological state of the aquatic organisms is a histological analysis of the gills, digestive gland, and kidneys, because it is the main accumulating and excretory organs. Despite the high effectiveness, this method is very lengthy. In this respect, a promising and highly sensitive biomarker of the hydrobionts state and their habitat is the assessment of the immune status, based on the hemolymph vital assay and rapid analytical procedures.

Sexually mature *Modiolus kurilensis* bivalves with a shell length of 75–95 mm were collected from wild populations in non-impacted water areas (Vostok Bay) and impacted area of Amursky Bay (Sportivnaya Gavan Bay) of the Sea of Japan. To identify the histomorphological alterations and lesions, sections of paraffin-embedded tissue fragments of kidneys and digestive glands were obtained using a rotary microtome, and were then stained with hematoxylin and eosin. Twenty histological changes of the investigated organs related to the presence of parasites, changes in cells, tubule and intertubular spaces have been analyzed, on the basis of which formulas are developed. Afterwards, the individual quantitative histopathological condition indexes were estimated for each individual based on the weighted indices (alterations in the organs) approach proposed by Bernet and colleagues (1999) for fish and adapted by Costa and colleagues (2013) for bivalves. To assess the immune status, the parameters of the cell-mediated (phagocytic activity (PA), phagocytic index (PI), reactive oxygen species (ROS), total and differential hemocyte count (THC and DHC) with granularity assay (SSC)) and humoral immunity (hemagglutination (HA), hemolytic activity (HL), protein concentration (PC)) were measured. To test the significance of the hypothesis of either the absence or the presence of differences between the studied samples we used the paired t-test and the Mann-Whitney test. To identify correlations of other parameters, a Spearman R correlation was performed ($p < 0.05$).

The histopathological condition indexes, i.e., combining all reaction patterns, revealed that, for both organs, the bivalves collected Sportivnaya gavan attained the highest values, significantly different from Vostok Bay. In comparison with molluscs from the Vostok Bay *M. kurilensis* from the impacted water area had considerably ($p < 0.05$) lower values of average PA, ROS, HL, but THC was considerably higher ($p < 0.05$). Correlation analysis revealed a significant ($p < 0.05$) inverse moderate correlation between the total histopathological index (for two organs) and PA, percent and SSC of granulocyte, and weak correlation with HL; but direct moderate correlation with PC of

bivalves from Vostok Bay transformed to inverse strong correlation of *M. kurilensis* in Sportivnaya gavan. However, the greatest contribution to the correlation, as a rule, was made by the pathology of the digestive gland.

Thus, the suggested approach in this work, based on the evaluation of immune parameters is more effective alternative to labor-consuming methods of health diagnosis of the marine hydrobionts and has promising potential for biomonitoring application. The revealed immunological parameters characteristic of physiological alterations can be used as objective features in the diagnosis of homeostasis disorders preceding the development of histopathological processes.

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Fauna of soft sediment and inactive hydrothermal sulfide fields in the Russian Exploration Area on the Mid-Atlantic Ridge

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In November 2012 the Russian Federation signed a contract with ISA to prospect and explore mineral resources on the MAR over 15 years. The Russian Exploration Area lies between 12°48.36' N and 20°54.36' N and includes 100 blocks, approximately 10 kilometres by 10 kilometres in size. The environmental impact assessment requires information on the status of reference ecosystem in the exploration area. Hydrothermal ecosystems of the northern Mid-Atlantic Ridge are among the best studied habitats in the deep-sea. However the periphery of active deep-sea hydrothermal vents and fauna of inactive hydrothermal sulphide fields remain poorly known. We provide here first results on the megafauna associated with soft sediment and inactive hydrothermal deposits on the Mid-Atlantic Ridge based on geological video records and limited material collected on several cruises of the RV *Professor Logachev* in the Russian Exploration Area, including new data obtained in February–March 2015 in 37th cruise of RV *Professor Logachev*. Significant effort of the last survey was aimed at studies of benthic non-vent soft sediment fauna at depths from 2270 m to 3900 m. Samples of this fauna at seven stations revealed ~780 specimens and at least 136 species. Very limited biological data was obtained for the hard substrate fauna. The sea conditions were not favourable for TV profiling. Nevertheless, extensive fields of empty bivalve shells were observed on the TV profile in the vicinity of two new ore fields Pobeda-1 (17°08.7' N, 46°23.44' W) and Pobeda-2 (17°07.45' N, 46°24.5' W) discovered in the course of the cruise (Beltenev et al., 2015). All species discovered alive on the ore deposits in our study were of small size (<3 cm) that agrees with results of Dobretsova et al. (2013) for the MAR segment 12°58'–13°31' N.

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Application of metagenome analysis to time series monitoring of plankton in the Okhotsk Sea

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Sequencing technologies, such as Roche 454 or illumina MiSeq, have made it possible to obtain millions of sequence reads in a single experiment, and massively parallel sequencing (MPS) is a currently revolutionizing survey of eukaryotic diversity including Harmful Algal Bloom causing (HAB) species, because this technology enables us to detect several hundreds of operational taxonomic units (OTUs) of eukaryotes from seawater samples and facilitates the detection of low-abundance populations in complex eukaryote communities.

The eastern part of Hokkaido facing the Okhotsk Sea is where the coldest water (-2°C) flows in from Sakhalin in the winter. Therefore, the perturbations in the marine ecosystem has been considered as an environmental sensor, and Mombetsu city is especially recognized as the most important monitoring base for observing plankton biodiversity in the coldest waters of Japan. Therefore, we have conducted plankton biodiversity monitoring using MPS-based technology one a week for nearly five years at the Okhotsk tower. In this study, we analyzed the biodiversity in seawater samples taken from Apr 2012 to Jun 2014 ($n=112$). We also conducted HAB detection using this technology.

The variability of plankton biodiversity among samples was evaluated by applying similarity indices (the Jaccard index), and non-metric multidimensional scaling (NMDS) analysis was performed. Samples taken at the same month but in different years were more closely plotted and also the samples of closest months were located in the closest positions, showing the consecutive transition of species composition and the clear seasonal pattern in Okhotsk Sea. The relative abundance of identified OTUs at the supergroup levels at the four groups was compared. The relative abundance of Alveolata or Stramenopiles was highest, and the third highest groups were Opisthokonta, or Viridiplantae, which varied depending on the samples, and they accounted for 77.1–86.4% of the total. These patterns are common in coastal waters. The relative abundance of diatoms was 37.6% at Jan–Mar group and the highest among the groups, reflecting the conspicuous diatom spring blooms, which are represented by *Thalassiosira eccentrica*, *T. antarctica*, *Fragilariopsis cylindrus*, *Odontella aurita*, *Porosira pseudodenticulata*, and/or *P. glacialis*. Spring blooms by diatoms are also common in the Seto Inland Sea, Japan, but the dominant species are *Chaetoceros* spp., *Eucampia zodiacus*, *Coscinodiscus wailesii* and/or *Skeletonema* spp., showing the different compositions. The lowest

water temperature during diatom spring blooms in Okhotsk Sea is nearly -2°C , but $8-9^{\circ}\text{C}$ in the Seto Inland Sea. Diatom species isolated from western Japan cannot grow below 5°C under laboratory conditions (Nagai, unpublished data). We therefore are going to compare the eukaryotic biodiversity in Okhotsk Sea with other localities in more detail. We may also need to compare gene expression patterns by transcriptome analysis to reveal the differences.

In Okhotsk sea samples, we detected the genus *Alexandrium*, *Azadinium*, *Chattonella*, *Cochlodinium*, *Karenia*, *Karlodunium*, *Prorocentrum*, *Pseudochattnella*, *Takayama* including toxic and fish killing species. Our data indicated that these HAB species were carried by the Tsushima warm current in summer seasons, suggesting the possibility of settlements in northern Japan. The detection limit of MPS-based monitoring of *A. tamarense* was 50 copies of rRNA genes, indicating that the technology is sufficiently sensitive for practical use in the monitoring of HAB species.

Scientific and technological developments of monitoring HABs and biotoxins on the Russian east coast

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A growing number of harmful algal blooms have motivated the expansion of active monitoring programs in coastal areas of the Asia-Pacific region to detect the presence of toxin producing microalgae, and evaluate the toxicity of seafood. One of these monitoring programs was established in 2007 in Primorsky Krai, Russian Federation, by Center for Monitoring of HABs and Biotoxins of NSCMB Far Eastern Branch of the Russian Academy of Sciences (recently IMB FEB RAS) www.imb.dvo.ru/misc/toxicalgae/index.ttm. The initial purposes for Center creation were: monitoring of potentially harmful microalgae, which cause HABs and produce toxic for marine animals and human substances; revealing of toxin biodeposits (sea water, benthos, epibioz); monitoring of resting cysts as a potential initiators of HABs; developing of analytical methods, which help to predict HABs; participation in developing of analytical methods for toxin identification and application of these methods for providing healthy environment; educate school and university students as well as authorities about the problem of HABs and phyco-

toxins (see Figure).

The results of Monitoring of HABs and Biotoxins in coastal waters of Russia during the period 2007–2016 were summarized. A total number of 43 potentially toxic and bloom-forming microalgae have been identified on the Russian east coast. Several species of potentially toxic haptophytes, dinoflagellates, diatoms, raphidophytes and cyanobacteria are reported for the first time in the study area. The revealed changes and trends in the composition and distribution of microalgae communities are as follows: long-term changes in microalgae communities on the Russian East coast are observed;



Young scientists of the NSCMB FEB RAS undergo training in using Automated Biobanking Systems Liconic STC Compact ULT.

there is a decreasing dynamic of diatom component of microalgae communities, which substituted by the non-diatom component; new boom forming species were detected for the study area and new toxin producing species were revealed. The species richness increased; there are species-specific changes in the abundance and seasonal dynamic of HAB taxa like *Pseudo-nitzschia* spp., *Karenia* spp., *Dinophysis* spp.); a list of species causing blooms in the bay was expanded (dinoflagellates *Ostreopsis* spp. revealed in 2007 and *Prorocentrum foraminosum*. in 2014, diatoms *Skeletonema* spp. revealed in 2013, haptophytes *Pseudohaptolina sorokina* in 2016); the biomass of the non-diatom component of the phytoplankton increased. For the last two decades, some species of harmful algae (e.g., toxic dinoflagellates and haptophytes benefiting from land runoff and water column stratification, warm water benthic dinoflagellates and cyanobacteria responding to increased water temperatures) became more common. It is supposed that climate change combined with human activities, primarily through nutrient runoff, is an important factor of the increase in HABs activity in the study area. A growing number of harmful algal blooms have motivated the expansion of active monitoring programs in coastal areas of the Asia-Pacific region to detect the presence of toxin producing microalgae, and evaluate the toxicity of seafood.

Diatoms of the genus *Pseudo-nitzschia* are producers of domoic acid, which causes amnesic shellfish poisoning (ASP). Seven *Pseudo-nitzschia* species were found. Species of the genus *Dinophysis* and *Prorocentrum* that are capable to produce diarrhetic toxins causing diarrhetic shellfish poisoning (DSP) were observed in the study area. Species of the genus *Alexandrium* are known as producers of saxitoxins, causing the paralytic shellfish poisoning (PSP). This study revealed potentially toxic species *Protoceratium reticulatum*, *Gonyaulax spinifera* and *Lingulodinium polyedra* which are able to produce yessotoxins. Raphidophytes, cyanobacteria and diatoms, which are known as fish killers, were found in the study area too. Data on toxin concentration both in the samples of mussels and microalgae cultures isolated from Peter the Great Bay were received by Enzyme Linked Immunosorbent Assay (ELISA) and LC/MS-MS methods. One poisoning syndrome, diarrhetic shellfish poisoning has not been reported in this region, but its expansion to other subarctic waters has been observed. We show here the first measures of diarrhetic shellfish toxins (DST) on the east coast of Russia. Mussels contained DST concentrations above the recommended regulatory level of 16 µg/100 g shellfish at almost all sites sampled in the Primorsky Krai region, with maximum concentrations of 56.3 µg/100 g. The primary DST isoform was dinophysistoxin-1 (DTX-1; 94–100%) but minor amounts of okadaic acid (OA; 0–6%) also was detected. Other lipophilic toxins measured in shellfish were yessotoxin (YTX), pectenotoxin-2 (PTX-2) and azaspiracid-2 (AZA-2); the last two toxins observed in Russian water for the first time. Phytoplankton samples dominated by *Dinophysis acuminata* contained DST, indicating that it was likely responsible for shellfish toxicity. Particulate seawater samples with *Dinophysis* contained 2.1–10.2 ng DTX-1/L and 6.5–101.8 ng PTX-2/L, corresponding to cellular concentrations of 3.1 to 5.6 pg DTX-1/cell and 9.6 to 46.7 pg PTX-2/cell. These findings provide a foundation for a Federal Monitoring Program for harmful algal blooms (HAB) and phycotoxin control in seafood in Russia, and suggest that appropriate methods for both rapid screening and regulation of shellfish toxins be established to ensure safe harvest on the Russian East coast. The absence of federal monitoring program for HABs and phycotoxins is a huge disadvantage, which leads to underestimation of actual scale of negative influence of HABs for ecosystems; economic loses for aquaculture and level of people's health harm. The absence of obligatory screening for phycotoxins prevents from effective analysis of possible reasons of massive fish mortalities in the region. Currently, the Russian Federal Sanitary

Control Service has no guidance for PTXs, AZAs and YTXs. As well as there is no recommendations for microalgae monitoring in the regions with the most fish farms. Those are just some drawbacks in legislation that we face now.

These results have importance for local community and for global science community. The significance of these results for scientists all over the world is in understanding of microalgae migration patterns, analyzing influence of various environmental conditions on toxicity of microalgae and taking into account Far Eastern data for global predictions and estimations. On the local level, this information is used to raise people's awareness about severity of HABs problem and prevent possible intoxications. Each summer there is information is mass media spread by Center workers about possibility of poisoning by eating uncertified seafood, which helps people to make avoid intoxication.

Educational role of Center plays huge role in preventing the illness of people caused by phycotoxins. Center workers participate in science festivals with lectures and seminars for school student of different age, provide consultations for sanitary officers and representatives of people's health protection organizations and participate in local conferences where scientists, administrators and business representatives meet in order to collaborate for solving of problems. Multiple analytical reports were prepared by Center about HABs problem, showing the absolute need for developing of phycotoxin food safety monitoring programs, which should help not only protect people from possible intoxication, but also improve competitiveness of Russian seafood products on the international market, which is very beneficial for development of fish farms and regional economic situation.

In general, there will still be a lot of new challenges in the HAB research and management, but new technology and qualified personnel to minimize effects and protect public health and marine environmental and resources will be more in demand than ever.

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Complex monitoring of the state of sea water basins by optical methods

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Many remote methods for detecting different pollutants rely on identifying the contrast between clean and contaminated sea surfaces. The phenomenon of “smoothing out” the water surface due to changes in the coefficient of surface tension produced by organic films, and due to the suppression of small-scale wind induced ripples by the films, underlies the physical processes making it possible to record organic films on the sea surface in the visible wavelength range. The smoothing-out effect of oil is also observed during active radar sensing of the water surface in the radio wavelength range from 1.5 to 70 cm. These “smoothed out” regions are often called slicks, which are easily discernible in the visible, infrared, and millimeter wavelength ranges. Systems of video surveillance of slicks in the optical wavelength range are an efficient tool for the real-time monitoring of coastal water basins.

A simplified formalism is introduced for slick–sea surface contrast formation which takes into account the upward sea radiation and qualitatively explains in situ measurement results. The results of a detection of slicks on the coastal water surface are also described, which, together with the simplified formalism suggested, provide optimal experimental conditions for slick–sea surface contrast measurements; i.e., the contrast of the P component of reflected radiation is measured at viewing angles close to the horizon.

The panoramic system of video surveillance of the sea surface, developed in the Far East Branch of the Russian Academy of Sciences, has operated at a marine expedition station of the Pacific Oceanological Institute since 2006. This video surveillance system was used as the first level in the complex monitoring system of pollution of the sea surface by organic films in coastal regions. It detects slicks, determines their coordinates, and tracks the dynamics of variations; also, it helps to study the surface circulation of waters. Analysis of a time sequence of transformed to the plane panoramic images obtained using the system allows to estimate a velocity field of vortex structure, phase velocity of surface manifestations of internal waves, intensity and dynamics of surface films of oil pollution.

The second level of real-time monitoring, whose purposes include the identification of organic species in organic films, can be performed by a remote method with the help of mobile fluorescent lidar. This lidar can certainly record fluorescence spectra of organic substances from a range of up to 150 m from the installation site (on shore or on ship); this spectrum can then be used to identify organic species (such as determining the petroleum product type) provided that the database of

reference spectra is available. For ecological monitoring of the water basin of Amur Bay near Vladivostok, we performed lidar sensing of the sea surface at a range of about 150 m from where lidar was installed. The measurements provided fluorescence spectra of model spills of motor oil, kerosene, diesel fuel, fuel oil, and automobile brake fluid. The experiments demonstrated that the spectra could be confidently recorded when the signal-to-noise ratio was higher than 100 for the spill types studied.

The complex approach to implementation of multilevel monitoring system of sea water basins will make it possible to rapidly detect the region and degree of pollution of water basins. The systems for analyzing the sea waters in situ will make it possible to determine the environmental effect immediately during study. This will help to make timely decisions regarding maintenance and restoration of the ecology in coastal water basins.

Microalga as a natural bio-indicators of aquatic pollutants

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The important objects for environmental monitoring of water are natural bio-indicators – living objects, such as microalgae, capable of responding to changes in the condition their habitat. A promising method of measuring the microalgae concentration and their living conditions is the analysis of fluorescence of pigments; primarily it is laser-induced fluorescence (LIF) of chlorophyll-a. The report discusses perspectives of applying of microalgae reaction to detect to the presence in water of some metal ions.

Most widespread consequence of the industrial discharges is the excess of MPC of metal ions, which often leads to ecological disasters. The most commonly in water there are metal ions Cu^{2+} , Zn^{2+} , Fe^{2+} , Al^{3+} , Mg^{2+} , both natural and technogenic origin, and as a rule technogenic very toxic ions Cd^{2+} , Hg^{2+} , Pb^{2+} , Ni^{2+} .

Experiments were carried out with cultures of microalgae from different phylums: *Nannochloris maculata* (Chlorophyta), *Porphyridium purpureum* (Rhodophyta), *Chaetoceros muelleri* (Diatomeae), *Tisochrysis lutea* (Haptophyta), which were provided by the National Scientific Center of Marine Biology FEB RAS.

It was shown that to create sensors of the acute toxicity best suited *Nannochloris maculate* (Cu^{2+} , Zn^{2+}) and *Porphyridium purpureum* (Al^{3+} , Ni^{2+} , Pb^{2+}), due to their rapid reaction to metal ions. To create sensors of chronic toxicity (which appears with the time) all studied microalgae are suitable: *Nannochloris maculate* (Fe^{2+} , Zn^{2+}), *Porphyridium purpureum* (Fe^{2+} , Zn^{2+} , Ni^{2+} , Pb^{2+}), *Chaetoceros muelleri* (Cd^{2+} , Hg^{2+}), *Tisochrysis lutea* (Cu^{2+} , Al^{3+}).

The obtained results have shown promising use LIF of microalgae for the analysis of changes in environmental conditions of their habitat.

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The fluorescent bioindicators based on immobilized microalgae for monitoring systems of ecological state of water areas

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Application of living organisms as bioindicators is one promising areas of development measuring systems for the analysis of the ecological state of aquatic environments. Currently, as a promising biological indicator actively studied of phytoplankton microalgae, which react to changes in the environment by changing the parameters of the laser-induced fluorescence of its pigments. The main problems of such monitoring of water areas are the variability in species composition and concentration of microalgae in the investigated volume.

The report discusses the possibility of creating of volumetric biosensors based on the technology immobilize of pure cultures of microalgae at a predetermined concentration in a porous matrix of silicon oxide, which is permeable for the pollutant molecules but prevents the penetration of others microalgae from the environment.

We explored three variants of biosensors based on immobilized algae. The first variant, the matrix in the form of a parallelepiped, which allows to carry out fluorescence measurement perpendicular to direction of the exciting radiation. The second variant, the matrix in the form of a film (thickness of 1–2 mm) on a glass substrate for the measurement of fluorescence by external of optical fiber probe without it introducing into the volume of the matrix. The third variant, matrix of an arbitrary shape, inside which, during the formation, introduced optical-fiber probes, which enables to create biosensors without strict requirements for cleanness of their surface.

We consider the advantages and disadvantages of options proposed of biosensor, in terms of permeability of the matrix in the aqueous medium, methods of excitation and measurement of fluorescence and practical application in environmental monitoring of water areas.

This work was supported by a grant from RSF (agreement No. 14-50-00034) in the part of studies of immobilization of microalgae, FEB RAS grant program “Far East” No. 0262-2015-0089, in the part of experiments with different cultures of microalgae.

New products are sources of amino acids for the formation of collagen

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At present, the combination of vegetable and meat raw materials is common in the food industry. This allows for simultaneous intake different nutrients in the body. Sea holothuria – *Cucumaria japonica* is used in the production of food products, as it allows to receive various combined products. *C. japonica* is a source of collagen and triterpene glycosides. Her muscles contain 65–70% of collagen. Collagen contains many amino acids of proline and hydroxyproline, which provide collagen synthesis. These products belong to the group of functional food. They are very useful for the body, especially for people of older age groups. As these products are useful for people with reduced functions of skin regeneration and osteoarticular system.

The purpose of this work is to justify and develop the technology of combined canned foods with using *C. japonica* and chicken gizzards.

The composition of the new canned include *C. japonica*, chicken gizzards, browned onions and carrots, spices.

Finished canned food is sterile and is a combination of functional products. Canned food can be consumed in a warm way as a second course. Or like a cold snack. The product has a pleasant appearance, taste and smell, a soft and juicy consistency. Canned food without foreign smell and taste.

In 100 grams of canned food, the amount of collagen-forming amino acids allows us to meet the daily requirement of a person in them by 24.7%. These canned foods can be belonging to products of a functional purpose, since they contain amino acids involved in the synthesis of collagen in the human body. Consuming them will help restore and maintain the functions of connective tissue in the human body.

This product is characterized by a high content of amino acids proline and hydroxyproline, involved in the synthesis of collagen. During the sterilization of cucumber and chicken stomachs, the uptake of the collagen protein increases. As there is a decay of its molecules under the influence of high temperature.

As a result of the research, was developed a recipe and technology of new canned food based on *C. japonica*, chicken gizzards and vegetable raw materials. The content of collagen-forming amino acids in 100 grams of combined canned food allows satisfying the daily human need for them by 24.7%. Canned food as a functional product is recommended for nutrition to the elderly and people with reduced functions of skin regeneration and osteoarticular system.

Megabenthic communities in deep-sea Central Arctic

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A photographic survey was carried out during the expedition ARK-XXVII/3 of the research icebreaker RV *Polarstern* in summer 2012 (2 August – 29 September) to the Eastern Central Arctic basins when the minimum of sea ice cover in the Arctic Ocean was registered. Seafloor was photographed using a towed camera platform, an Ocean Floor Observation System (OFOS). Nine transects were carried out: four of them were located at Nansen Basin (3571–4066 m), five – at Amundsen Basin (4041–4384 m). Additional seven samples for identification and ground-truthing were taken by Agassiz trawl. For the first time in the history of the study Arctic Ocean we determined the composition and structure of megabenthic communities in different areas and evaluated their controlling factors in the conditions of climate changes and melting of sea ice. Megafaunal distribution was uneven with high variation of abundance at different stations. Nevertheless, two groups of stations were distinguished. One group located closer to ice margin under first year ice was characterized by relatively high densities and biomass of actinarians *Bathypheilia margaritacea* (mean 0.18–0.95 ind m⁻²; 0.16–0.85 g ww m⁻²) and high density of serpulids *Hyalopomatus claparedii* (mean 0.14–0.55 ind m⁻²) and relatively high indicators of productivity (including concentrations of chlorophyll a, phaeopigment, total organic carbon, prokaryotes abundance and others). Another group located closer to North Pole under multi year ice with 100% ice cover tended to have lower indicators of productivity and relatively high density and biomass of holothurians *Elpidia heckeri* (mean 0.92–1.53 ind m⁻²; 0.30–0.43 g ww m⁻²) and *Kolga hyalina* (mean 0.004–1.68 ind m⁻²; 0.01–3.48 g ww m⁻²). Macellicephalin polychaetes were abundant at some stations without detected correlation with any factors. The influence of diatom ice algal deposits on the sea floor founded during the cruise for the first time in the abyssal zone of the Arctic Ocean on the megabenthic communities was assessed. However, we couldn't find any direct relation of megafaunal abundance and biomass with coverage of algal aggregations at the seafloor or with their degree of freshness. But in small scale large mobile megafauna (holothurians and ophiuroids) accumulated around ice algae deposits and fed them.

The work was partly supported by RSF (grant No. 14-50-00095).

The structure and distribution of marine benthic communities in relation to body size in the northeastern Sakhalin shelf in the Piltun and Chayvo Area (the Sea of Okhotsk)

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Few studies consider meiofauna and macrofauna at the same time, even though both form parts of wider ecological networks, and fewer consider interactions between sample size, body size and spatial clustering. While there have been many surveys of the macrobenthos in the northeastern Sakhalin shelf (The Sea of Okhotsk) during the past few decades, particularly in nearshore (Fadeev, 2009), there have been no truly comprehensive quantitative survey, including different size groups. Benthic macroinvertebrates may influence occurrence, composition and distribution of the meio-benthos directly through predation and indirectly through alterations in interstitial physico-chemical characteristics and processes rates (e.g. transport of organic matter, mineralization rates, distribution and exchange of solutes). This study examines the degree to which infaunal assemblage structure varies with mesh size, sample size and sample dispersion within two different areas of apparently homogeneous sediment, a muddy sand and a coarse medium sand, in the northeastern Sakhalin shelf in the Piltun and Chayvo Area (the Sea of Okhotsk) based on materials of field studies in 2013–2015. The investigated sea area was 365.6 km². Benthic assemblages along the Piltun and Chayvo Area were examined to better understand the structure of the community. We have found very abundant and diverse meiofaunal and macrofaunal assemblages. Although the benthic community distribution pattern was mainly structured by environmental factors, sediment grain-size and organic compounds are key factors in structuring macro- and meiofaunal assemblages. The area was resampled in September–November 2013, September–November 2014, and June–November 2015 to determine if the identified effects were persisting. Considerable inter-year fluctuations of species abundances as well as species richness, diversity, trophic and size structure were recorded. Patterns observed for quantitative distribution of meiofauna were compared with those from larger-sized benthic fauna. The main “top-down” community control is maintained by macroinvertebrates, especially polychaetes and amphipods, which not only have the direct predatory effect but might be also mechanically disturbing the meiofaunal assemblages. As benthic infauna, can profoundly influence nutrients cycling and since food is an important limiting factor for many benthic populations the densities of macrobenthic species may be inducing changes in the investigated sea area via food web interactions. This study will be useful in understanding the abiotic factors that influence the benthic community and will facilitate future assessments of natural and man-made disturbances.

Bloom-forming phytoplankton species, new to marine waters of Russia

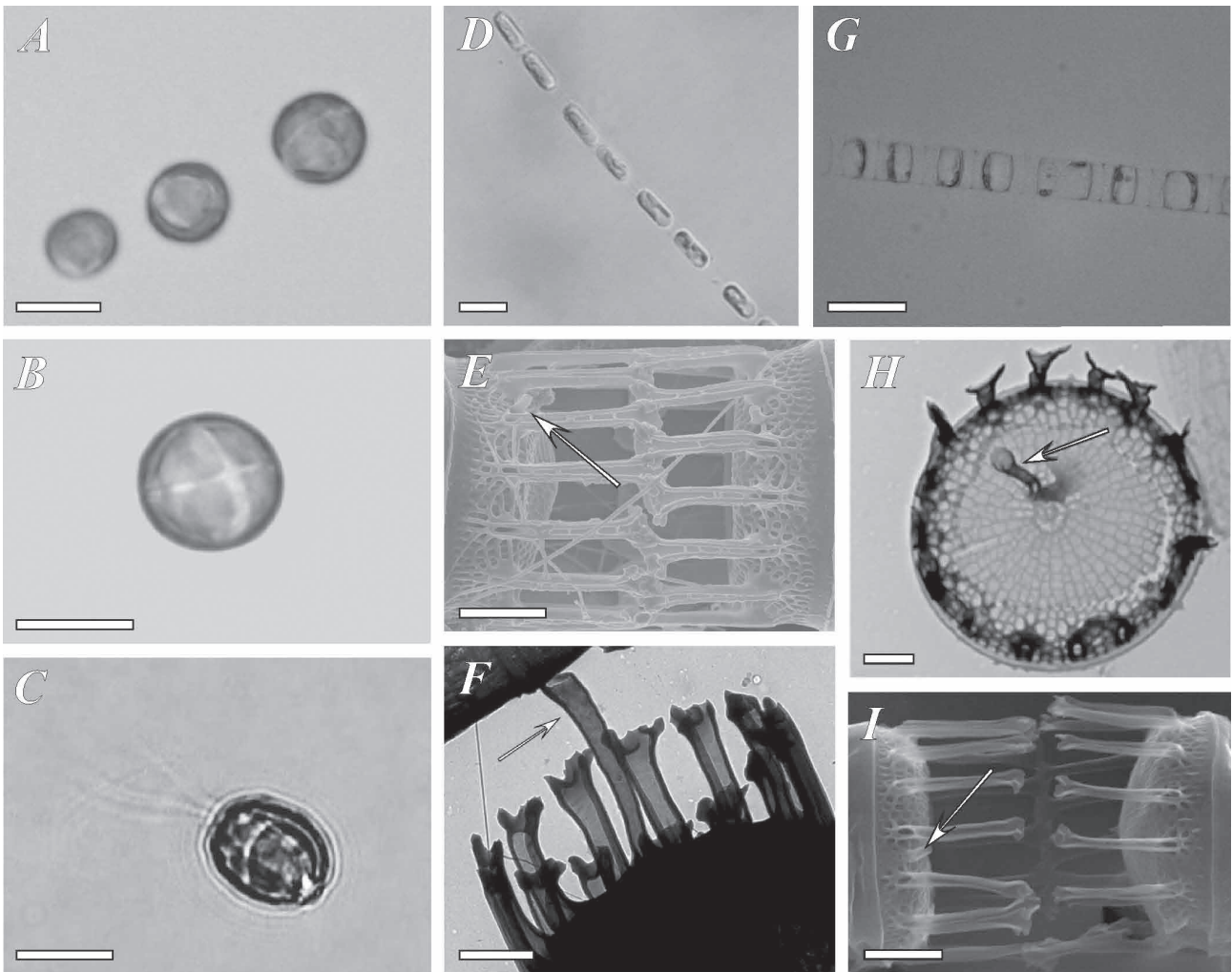
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As a result of monitoring of microalgae (2013–2015) in coastal waters of Paris Bay (Russky Island, Peter the Great Bay, Sea of Japan), a “bloom” of the haptophyte alga *Pseudohaptolina sorokinii* was recorded in the spring season of 2013 (see Figure, A–C). This is the second find of this species. The genus *Pseudohaptolina* was described relatively recently as consisting of one species, *P. arctica* Edvardsen et Eikrem, 2011. The second member of the genus, *P. sorokinii* Stonik, Efimova et Orlova, 2016, was described in 2016, when its mass proliferation occurred in Amur Bay, Sea of Japan. A “bloom” of *P. sorokinii* in Paris Bay was recorded on March 12, 2013: at the horizon of 0.5 m, the concentration of cells of this species reached 13.7 million cells/L (at a water temperature of -1.2°C and a salinity of 32.5‰). An analysis of horizontal distribution of the species showed that at the depth of 5 m the concentration of cells was insignificant, 300 cells/L; at the depth of 2 m, *P. sorokinii* was absent. During the following survey on March 26, 2013 (water temperature -0.3°C ; salinity in the near-surface horizon 13.8‰), the *P. sorokinii* concentration at the 0.5 m depth was 9.9 million cells/L; at the depth of 2 m, 100.6 thousand cells/L; and at the depth of 5 m, 27 thousand cells/L. The subsequent sampling on April 09, 2013 showed that even single cells of *P. sorokinii* were absent in the plankton of Paris Bay.

A comparison of the characteristic environmental parameters for development of a typical member of the genus, *P. arctica* (water temperature $0-3^{\circ}\text{C}$; salinity 33.5 ‰; vegetating under ice in Baffin Bay, Canada), with the data obtained for *P. sorokinii* in Paris Bay (water temperature from -1.2 to -0.3°C ; salinity 13.8–32.5‰) showed that both species develop in cold waters. Nevertheless, *P. sorokinii* is distinguished by a greater adaptive plasticity to changing salinity, as compared to *P. arctica*.

Two *Skeletonema* species new to marine waters of Russia, *S. japonicum* (see Figure, D–F) and *S. dohrnii* (see Figure, G–I), were identified using the electron microscopy methods. Members of *Skeletonema* are extremely widespread in plankton of the coastal zone of temperate seas and cause “blooms” of water. According to their distribution in the World Ocean, species of the genus are referred to as cosmopolitans, but the distribution pattern of most of them remains unclear. “Blooms” of *Skeletonema* spp. in the plankton of Paris Bay were observed in the summer and autumn of 2013–2015. A mass proliferation of *S. dohrnii* was observed in June, at a sea surface temperature of $11-17^{\circ}\text{C}$ and a salinity of 26.2–32.1‰; proliferation of *S. japonicum* was recorded in November, at a temperature of $3-9^{\circ}\text{C}$ and a salinity of 32.8–33.5‰.



Bloom-forming phytoplankton species. *Chrysochromulina* sp.: (A) general view of cells; (B) cell with four large chloroplasts; (C) cell with two flagella and haptonema. *Skeletonema japonicum*: (D) fragment of colony; (E) adjacent cells, fultoportula processes (8–12) much flared distally, short rimoportula (arrow); (F) terminal valve with long, tube-shaped rimoportula (arrow). *Skeletonema dohrnii*: (G) fragment of colony; (H) terminal valve with rimoportula (arrow); (I) adjacent valves, fultoportula processes (12–16) slightly flared distally. (A–D, G) LM; (E, I) SEM; (F, H) TEM. Scale bars: (F, H, I) 1 μm , (A–E) 10 μm , and (G) 20 μm .

The long-term year-round monitoring of phytoplankton, conducted at one observation point in the water area under study, provides a complete view of the floristic composition, an opportunity to register the emergence of new species, and observe the long-term trends in species composition.

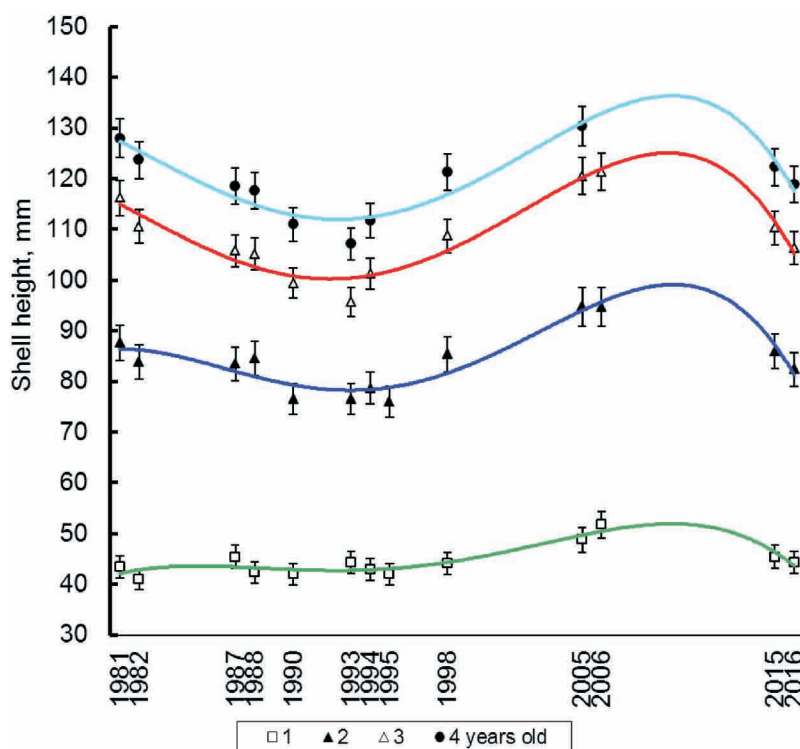
The work was financially supported by the Russian Foundation for Basic Research, grant No. 15-04-04838.

Monitoring of community of the Japanese scallop with its shell endobionts at the northwestern coasts of the Sea of Japan

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It is known, that there is a gradual enrichment of bottom sediments by organic matter in marine coastal areas throughout the world. The changes in quality of an environment entail the changes in a state of the organisms inhabiting coastal areas, especially in vicinity of cities and mouths of large rivers. The northwestern part of the Sea of Japan is subject to a long-term and continuously increasing effect of a complex of organic substances, which come there with the riverine drain of large Razdolnaya and Tuman rivers and with industrial and domestic sewages from Vladivostok city and other shore settlements. These pollutants gradually accumulate in the bottom sediments. Moreover, the settled organic material stimulates mineralization of this material, which entail a change in the benthos due to decrease of the water oxygen concentration. The purpose of the present study was to determine changes in the benthic community of the Japanese scallop *Mizuhopecten yessoensis* with endobionts



Dynamics of the mean shell heights of different aged scallops *Mizuhopecten yessoensis* inhabited north-eastern Amurskii Bay.

that erode its shell at the sites approached to main suppliers of organic pollution in the northwestern part of the Sea of Japan for last decades. The dynamics of the scallop growth rates, as integral vital sign, were studied. Also, degrees of the scallop shell bioerosion performed mainly by endolithic polychaete worm *Polydora brevipalpa* were compared.

At the coasts of Vladivostok, which is also under influence of Razdolnaya River run off, three main stages were revealed in the community development. During the first stage lasting until the beginning of 90th, there was the gradual depression of the community. The scallop growth rates decreased (see Figure). The shell bioerosion had increased considerably in

1982–1998. It was a period of an intensification of anthropogenic pollution of the near coastal water. Further, to the beginning of 2000th, the state of the community became better. The scallop growth rates had increased; the shell bioerosion had decreased to 2006. It was a period of a reduction of the pollution of the water and bottom sediments due to industry production decrease in the perestroika. In the third stage, which goes on to present day, the scallop growth rates declined again (see Figure). However, the degree of the scallop shell bioerosion increased. It was related to intensification of the water eutrophication due to a sharp increase in the number of automobiles. The stream of the nitrogen dioxide in an atmosphere increased. With rains this oxide tumbles down in the sea strengthening the water eutrophication.

At the site located further from Vladivostok and Razdolnaya River mouth, in open Reyneke Striate, the scallop shell infestation was lower than at the Vladivostok coasts, however, for two decades the significant increase of the degree of scallop shell bioerosion was also observed. In contrast, the scallop growth rates went down. At the coasts of Furugelm Island located not far from Tumen River mouth, the scallop growth rates went down, but the degree of scallop shell bioerosion statistically significantly increased for three decades. The increase of the degree of scallop shell bioerosion may be explained by the gradual increase of organic content in the bottom sediments. Organic matter is advantageous for development of bacteria and phytoplankton, which improves food potential for polychaete worm that erodes the scallop shells.

Thus, in coastal areas the process of biogeocoenosis reorganization permanently exists, and at presence of anthropogenesis influence it intensifies. Changes in the communities in some sites are statistically significant even for one–three decades. The anticipated ecological consequences are the most drastic for benthic organisms, including the scallop.

The work was supported in part by the FEB RAS (grant No. 15-I-6-059).

Extraction and screening of bioactive compounds from crab *Ocypode platytarsis*

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Marine organisms represent an excellent source for bioactive compounds. The field of marine natural products has been expanding in response to the growing number of structurally novel and bio medically promising natural products being isolated from the marine source. In general, marine crabs have been recognized as the most important and untapped resource for novel bioactive compounds. In the present investigation, the extracts were prepared from an intertidal crab *Ocypode platytarsis* which has been taken to evaluate pharmacological properties such as antibacterial, antioxidant, anti-inflammatory and immunomodulatory. The purified compounds from the extracts showed maximum zone of inhibition against *Klebsiella pneumonia* and *Salmonella typhi*. Moreover, the time kill curve studies and β -Gal assays revealed that the compounds were involved in β -Gal release by inhibited the cell wall and possess an excellent activity against *Klebsiella pneumonia*. The compound was also considered to be excellent potential antioxidant property against DPPH, FRAP and H_2O_2 . In anti-inflammatory activity, the compound down regulated the expression of NO and ALP production indicate that the positive evidence of dose dependent anti-inflammatory activity on LPS-induced RAW macrophages. It was also found that the ability of the compound significantly inhibited the proinflammatory cytokines production such as TNF- α , IL-1 β and IL-6 without inducing cytotoxicity. Such interesting of compound responsible for antibacterial, antioxidant, antiinflammatory and immunomodulatory activity could be claimed after characterization that 9-octadecenoic acid, methyl ester. Being a commonly available crab with least consumer preference, *O. platytarsis* is one of the potential sources of anti-bacterial, anti-oxidant, anti-inflammatory and immunomodulatory chemical agents and therefore a potential role is envisaged as a health-promoting food resource in human diets.

GIS employment in investigation of Pacific oyster beds in the northern part of Amursky Bay (Sea of Japan)

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The most abundant settlements of Pacific oyster *Crassostrea gigas* (Bivalvia, Ostreidae) are located in the northern part of Amursky Bay (Peter the Great Bay, Sea of Japan). This species is able to form large dense assemblages (oyster beds, banks and reefs) in shallow warm-water estuarine areas. The oyster beds work as biological filters standing on the way of river outflow, their maximum development is observed on silty grounds within 2–5 m depth range. These structures may reach up to 5 m above the bottom. Therefore, they are easily detected on satellite images exported from public Web mapping services (Google Maps, Yandex Maps, Bing Maps, etc.). On highly accurate navigation maps, big oyster banks can also be easily recognized.

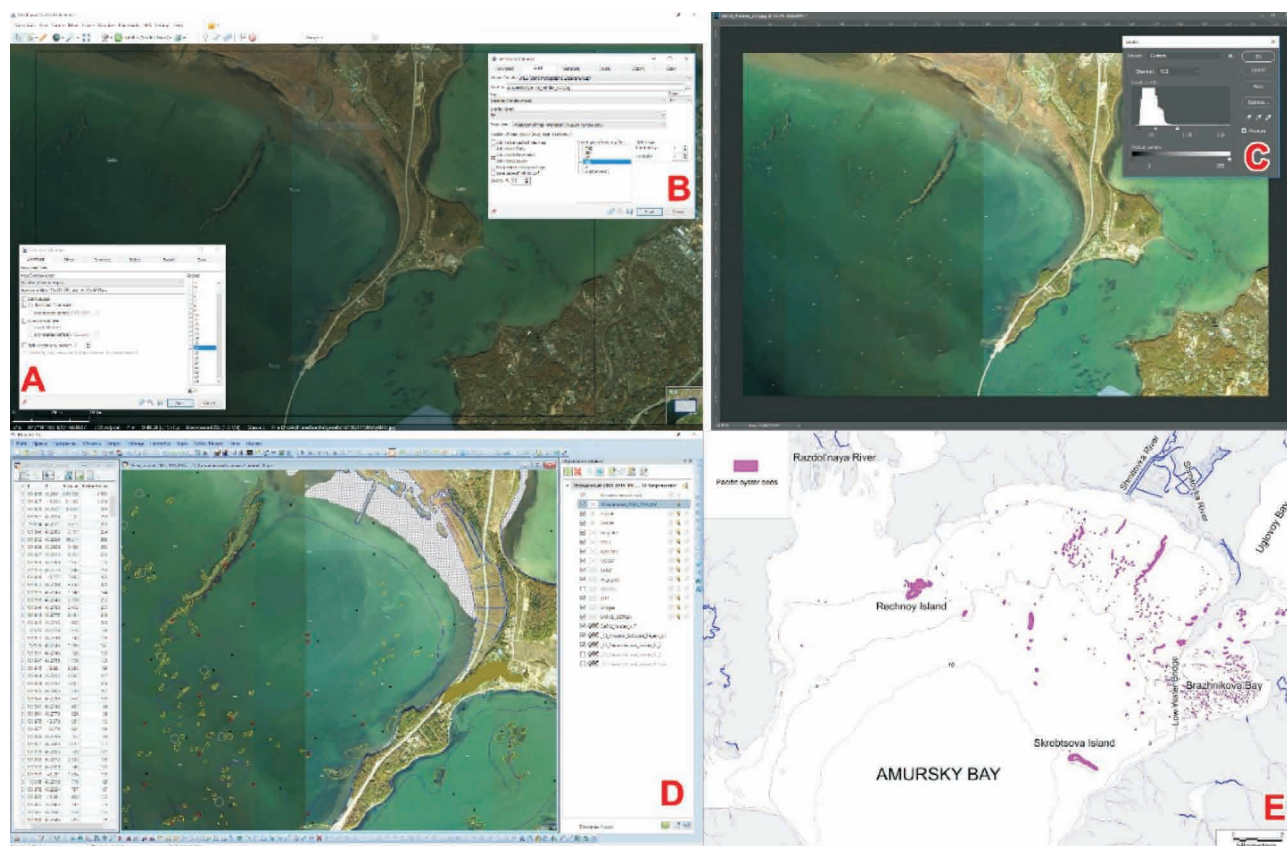
The main purpose of this work is collecting, processing and analyzing of the spatial data used for oyster beds investigation with the utilization of Geographic Information System (GIS).

At the first stage we downloaded images (tiles) of explored area with necessary zooming from one of the web-mapping sources in program cache of the SAS.Planet software (see Figure, A). Then we merged them into single graphic file (“.jpeg” format) and linked it to georeferencing file (“.tab” MapInfo format) (see Figure, B). Obtained image was processed in the raster graphics editor for better visibility of underwater structures (see Figure, C). Consequently, we acquired several integral satellite images that can augment each other.

Next step included merging of the acquired images with the vector map layers (coastal contour, depths, isobaths, types of bottom deposits, sandbanks, etc.) by means of the Desktop GIS Map-Info Professional (see Figure, D). Based on the analysis of spatial data set, 692 oyster beds were delineated in the area from the mouth of Razdol'naya River to Uglovoy Bay and Skrebtsov Island (see Figure, E). Obtained data was tested by SCUBA diving survey, conducted in the summer of 2015.

Total estimate of oyster beds area in the northern part of Amur Bay is 2.6 km². Single bed surface area ranges from 85 to 339,723 m² (average – 3773.2±692.4 m²), with perimeters ranging from 39 to 4563 m (average – 195.9±11.9 m). The biggest aggregation is located around Rechnoy Island near the mouth of Razdol'naya River at the spacious shoal with depths less than 2.5 m. The second largest oyster reef (area – 213.526 m², length – 2.3 km) is situated close to the mouth of Shmitovka River at 0.4–4 m depths. The third largest bed (211.994 m²) is located around Skrebtsova Island at the depths less than 6 m.

Four largest oyster aggregations only (0.4%) have individual areas exceeding 100,000 m², combined together they sum up to 33.7% of total oyster beds' area. 4% of oyster beds total number have surface areas ranging from 10,000 to 100,000 m², these beds occupy 22.7% of total acreage. Most numerous banks (52.3%) with individual areas from 1000 to 10,000 m² amount to 37% of total acreage. The greatest number and density of small-sized oval-shaped beds are registered in eastern



Step by step satellite image processing.

part of the examined bay section (from Low Water Bridge to Brazhnikova Bay) at the depths less than 3.5 m. Oyster reefs from the central and northern parts have bigger individual areas and more elongated shape. The 658 beds (95.1%) are located in shallow waters (up to 5 m). At the deeper locations, we found several dead oyster beds covered with silty deposits. Pacific oyster is a suspension feeder, thus most of mollusk assemblages are aligned transversely to coastline for better food supply from the coastal currents.

The obtained data will be used as a basis for the assessment of oyster beds' current state, estimation of sustainable fishing and for the further ecological monitoring.

Antioxidant and hepatoprotective potential of the polyphenol enriched extract from *Sargassum pallidum*, brown algae, against CCl₄ induced liver damage in rats

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The liver is one of the most vital organs in the body. It protects the organism from foreign substances by detoxifying and then their elimination. Due to these functions, liver ailments continue to be among the main threats to public health. The metabolism of various chemical agents such as alcohol, drugs or carbon tetrachloride (CCl₄) by the liver cause severe damage to the liver cells over time. Increased generation of the free radicals in the process of oxidative transformation of xenobiotics and decreasing the antioxidant defense mechanisms cause oxidative stress development of which enhances the severity of hepatic damage.

All of these negative processes could be reduced by the administration of the antioxidants. One of the best nominees to this role could be natural plant polyphenols, which can display antioxidant activity at low concentrations and thus be effective hepatoprotectors. Marine algal polyphenols – phlorotannins, which are known only to occur in brown algae have been shown to possess diverse biological activities, including antioxidant and hepatoprotection. In the course of searching for hepatoprotective agents from natural sources, we evaluated the polyphenols of marine brown alga, *Sargassum pallidum* for hepatoprotective activity. We received polyphenol enriched extract from *S. pallidum* (SPE) with total polyphenols content of 45% on a dry residue basis. Antioxidant and hepatoprotective potential of the obtained preparation was assessed using the model of CCl₄ induced liver damage in Wistar rats (180–200 g). Rats were injected in the dorsal neck fold with 2 ml/kg of a 50% solution of CCl₄ in olive oil for 4 days. SPE was administered intragastrically in a dose of 100 mg of total PP/kg of body mass. Wistar rats were divided into four groups of ten animals. Group 1 was normal control, group 2 – CCl₄ group; group 3 – CCl₄+withdrawal for 7 days; group 4 – CCl₄ +SPE for seven days. CCl₄ administration caused the typical picture of toxic hepatitis. In the group of rats induced CCl₄ alone the liver Mass Index was increased by 50% (p<0.001). Amount of total lipids in the liver was 3 times higher control level. This explained by an increase of on average 18–23% (p<0.01) in the contents tryacylglycerols (TAG), cholesterol (Chol), and free fatty acids (FFA) due to enhanced peripheral lipolysis in fat tissue – stress response to xenobiotic. We observed double increase in the TBARS level (p<0.001). Followed by, the level of antioxidant enzymes glutathione peroxidase (GPx), glutathione reductase (GRx), Superoxide dismutase (SOD) and reduced glutathione pool (GSH) was also significantly diminished (by 25–48%, p<0.01). This indicated a high level of free radical generation due to reducing dehalogenation of CCl₄ by CYP_{2E1},

which are actively, included in the phospholipid fatty acid chains, disorganizing the structure of cellular membranes. Within 7 days after CCl₄ withdrawal, most of the observed biochemical indices in the liver of rats (group 3) did not return to the control value, indicating a continuing toxic stress and insufficiency of the body defense to counteract the toxic pathology. Furthermore, the level of the antioxidant enzymes GPx, GRx, SOD and GSH pool was expressed by further decrease. The neutral lipids pattern remains misbalanced, which indicates further progress of fatty infiltration in the liver. The administration of the SPE during CCl₄ withdrawal (group 4) exhibited significant hepatoprotective activity by reducing the CCL₄ caused changes and led to the recovery of almost all studied biochemical parameters to the control value. We observed a restoration of the liver Mass Index and total lipids amount and neutral lipids pattern balance, which means improvement of fatty liver. TBARS level returned to normal and as well as the level of antioxidant enzymes GPx, GRx, SOD and GSH pool which indicates the reduction of the free radical activity level.

The results obtained by this study suggest that a polyphenol enriched extract from *S. pallidum*, containing phlorotannins, is a promising source for effective hepatoprotectors.

Alkyl glycerol ethers affect lipid metabolism and hematological parameters of blood in old-aged rats

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A beneficial effect of alkyl glycerol ethers (AGE) and omega-3 polyunsaturated fatty acids (n-3 PUFAs) on functions of many regulatory systems in organism of mammals, including humans, is widely known. Earlier studies in this direction have been conducted with the use of liver oil of deep-sea sharks. However, we find it interesting to assess the effect of AGE obtained from another marine object – the gonatid squid *Berryteuthis magister* – on lipid metabolism and homeostasis of old rats.

The experiments were carried out in 3 groups of old male rats (weight 440±35 g, age 30 months). The rats received biologically active components with a standard diet for 3 months: (I) docosahexaenoic acid (DHA), 100 µl/day; (II) AGE, 70 mg/day; and (III) DHA+AGE, (100 µl+70 mg/day). The comparison groups were: (IV) intact adult rats (weight 295±25 g, aged 6 months), (V) intact old rats. The lipid spectrum of blood serum was evaluated for total cholesterol, triglycerides, high-density lipoprotein cholesterol, low-density lipoprotein cholesterol, very low-density lipoprotein cholesterol, and the atherogenic index. The hematological parameters of the peripheral blood included counts of the number of erythrocytes, platelets, leukocytes, and lymphocytes; determination of hemoglobin, the average hemoglobin content per red blood cell, the color index, and the coagulation time.

The preparations for the studies were obtained in NSCMB FEB RAS: 45% DHA (as ethyl ester) concentrate and AGE preparation, 99% purity (biologically active food additive “Lipidomarin”, Certificate of state registration No. RU.77.99.11.003.E.008800.06.12), both produced from lipids of the gonatid squid *B. magister*.

The study allowed us to identify both individual and total pharmacological effects of AGE and DHA, which ensure their effectiveness at senile disorders of the homeostasis systems. While the course of DHA administration contributed to a moderate correction of hematological parameters, the response of blood cells to administration of alkyl glycerol ethers was manifested by a significant increase in their functional activity, increased synthesis of hemoglobin, and a higher number of immunocompetent cells. The greatest positive effect on the parameters of peripheral blood in conditions of senile immune deficiency, anemia, and hypercoagulation was achieved by the combined use of DHA and AGE. Therefore, in this case, we can talk about the synergistic effects of AGE and n-3 PUFA.

Immune correcting, antihypoxic, and antithrombotic activities of compounds of alkyl-glycerol structure in combination with lipid-lowering, cytoprotective effects of polyunsaturated fatty acids improves rheological properties of blood, the state of the immune system and the lipid metabolism at aging processes. The obtained results can be used in the development of targeted drugs for humans.

First data on foraminiferans from colonies of the bivalve *Calyptogena pacifica* (family Vesicomidae) on the Piip submarine volcano (Bering Sea)

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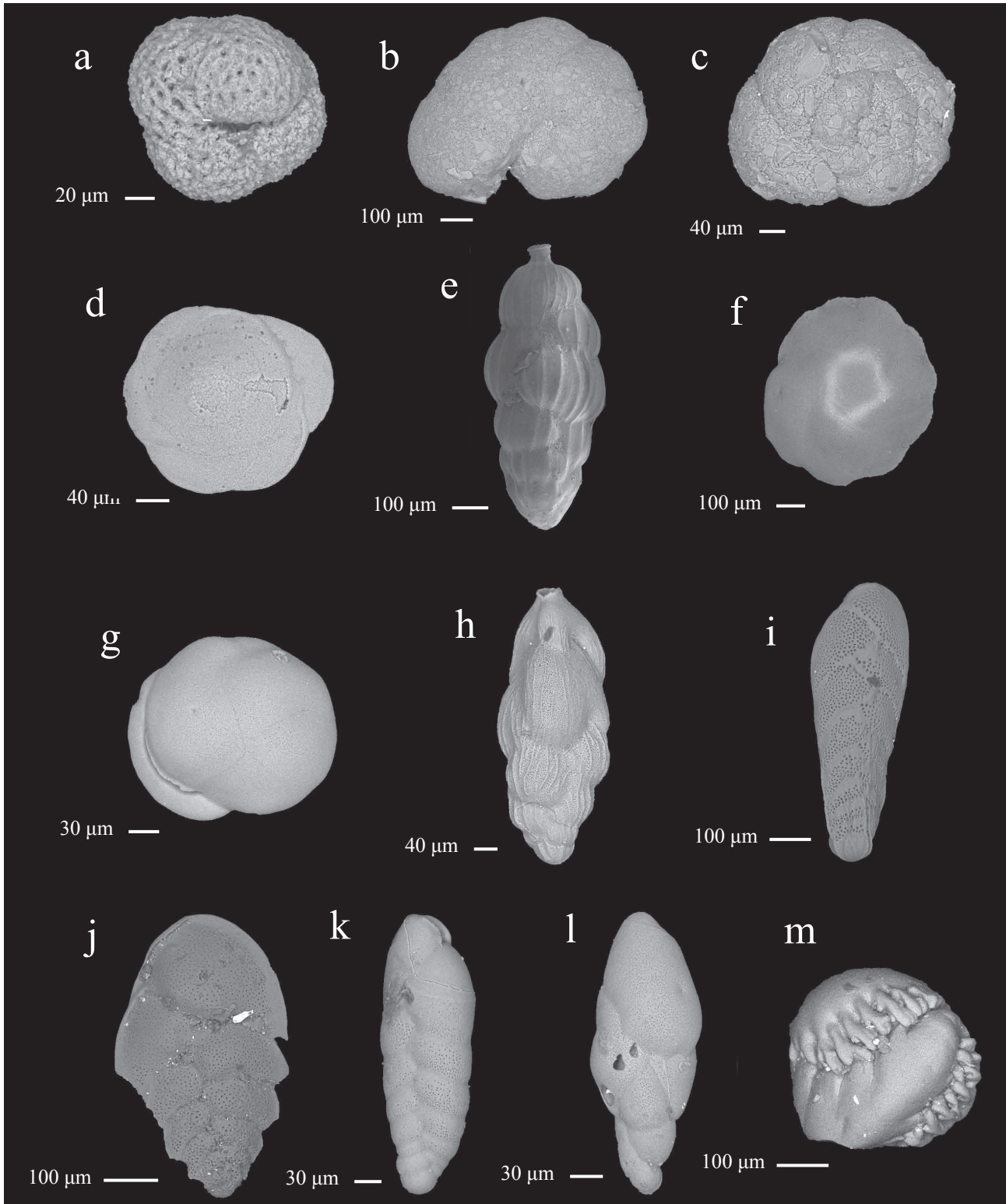
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Zones of deep-sea hydrothermal vents and cold seeps (including the studied areas of shallow-water volcanic activity in marine coastal ecosystems) are widely distributed in the World Ocean. In the Bering Sea, the only known volcanically active area is the Piip submarine volcano. We studied hydrothermal vents both on the southern and on the northern summits of Piip. The material for the study was the qualitative sediment samples collected during the 75th research cruise aboard R/V “Akademik M.A. Lavrentyev” in June 2016, using a scoop net attached to the manipulator of a remotely operated underwater vehicle (ROV) “Comanche 18”. At both summits of the volcano, there are large patches of bacterial mats that mark the sites of vents. The use of a ROV made it possible to determine the location of a *Calyptogena* colony in this area at a depth of 470 m (coordinates 55.3822° N, 167.2611° E). There clams were found buried in the sediment and exposing only ends of their shells. The composition of the *Calyptogena* community includes small animals that find shelter and food in aggregations of this clam. The unicellular benthos is dominated by foraminifera with hard shell. A total of 38 species with a predominance of secreted forms were identified, including three species of dead planktonic foraminifera: *Globigerina bulloides* d’Orbigny, 1926, *G. pachyderma* (Ehrenberg, 1861), and *Orbulina universa* d’Orbigny, 1839. Foraminiferas with organic shell were represented by single individuals of *Placopsilinella* sp. Among hard-shelled foraminifera, the most abundant were agglutinated (*Labrospira jeffreysi* (Williamson), 1858 and *Trochammina* sp.) and secreted species (*Buccella inusitata* Andersen, 1952 and *Uvigerina parvocostata* Saidova, 1961, *Cassidulina translucens* Cushman and Hughes, 1925, *Cassidulina delicata* Cushman, 1927); live cells (Rose Bengal stained) accounted for 12 to 16%. Such species as *Angulogerina angulosa* Williamson, 1858, *Bolivina spissa* Cushman, 1926, *Bolivina alata* (Seguenza, 1862), *Bolivina pacifica* Cushman and McCulloch, 1942, *Nonionella digitata* Norvang, 1945, *Chilostomellina fimbriata* Cushman, 1926, and *Stainforthia loeblichii* Feyling-Hanssen, 1954 were small in number but had a large proportion (an average of 40%) of live cells (see Figure). The rest of the species were found as single live cells and dead shells. A comparison of this complex with the data on the fauna of the region (1984; coordinates 55.0237° N, 167.180° E; depth 480–600 m; unpublished data) revealed



The hard-shelled foraminifera: **a** – *Globigerina pachyderma*, **b** – *Labrospira jeffreysi*, **c** – *Trochammina* sp., **d** – *Buccella inusitata*, **e** – *Uvigerina parvocostata*, **f** – *Cassidulina translucens*, **g** – *Cassidulina delicata*, **h** – *Angulogerina angulosa*, **i** – *Bolivina spissa*, **j** – *Bolivina alata*, **k** – *Bolivina pacifica*, **l** – *Stainforthia loeblichii*, **m** – *Chilostomellina fimbriata*.

only three common species. The investigated foraminifera complex is distinguished from the fauna of cold methane seeps of the Deryugin Basin (Sea of Okhotsk), in which the described complexes were small in number and had a poor species composition, by the predominance of agglutinated foraminifera and a low proportion of live cells (about 5%). Some differences from the sites of H₂S and hydrocarbon seeps in the Deryugin Basin have also been found: the abundance of all foraminifera was significantly higher, with the predominance of agglutinated species; live cells accounted for 13% of the total number of common species. Nevertheless, the composition of the bathyal fauna of the *Calyptogenia* colony on the Piip volcano has many forms common with the foraminiferal fauna of deep-sea regions across the oceans.

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Possible role of marine plants in recovery of coral reefs after severe physical disturbances

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The coral reef ecosystem is a collection of diverse species that interact with each other and with the physical environment. The latitudinal distribution of coral reef ecosystems in the oceans is determined by the seawater temperature, which influences the reproduction and growth of hermatypic corals – the main component of the ecosystem. Benthic algae are one of the most important components of reef ecosystems. Their role in healthy coral reefs is defined as: primary production of organic matter and its turnover, the construction of reefs, nitrogen fixation, the initial link of food chains, environment for marine animals, and protection of reefs against deleterious effects from surf.

Severe physical disturbances such as typhoons/hurricanes/cyclones and tsunamis cause extensive damages to coral colonies. Coral bleaching is often caused by unusually high sea temperatures combined with periods of slack wind, calm seas, high solar radiation, leads to reduced photosynthesis, a tissue growth, calcification and subsequently to the death of corals. The mass mortality of corals under the influence of severe physical disturbances leads to disruption of the homeostasis of the ecosystem, and at their frequent repetition to the destruction of coral reefs. The coral reef restoration can last for decades. Recovery of coral reef hampers or contributes to a variety of abiotic and biotic factors. Many abiotic and biotic factors hinder or contribute to the restoration of coral reefs. In the report we discuss the possible role of benthic algae in the process of restoring coral reefs damaged by severe physical disturbances and for the first time expressed the hypothesis of their positive role in this process. We suggest that this is mainly achieved through the colonization of newly formed substrates by marine algae, with the following characteristics:

1. Maintenance of high ecosystem productivity through settlement of highly productive morpho-functional algal forms.
2. Protection of coral reef basis and newly formed carbonate substrata (dead coral colonies) from erosion and continuation of carbonate reef base building.
3. Colonization of vacant substrates by algae enhances the biodiversity of an entire reef assemblage.

4. Symbiotic relations between algae and corals also promote homeostasis and coral reef recovery in damaged reef systems through transport of assimilates from endolithic symbiotic algae to coral tissue, which intensify during a bleaching episode or through coral digestion of own zooxanthellae that also intensify under extreme conditions.
5. Release of secondary chemicals by encrusting calcareous algae (or their bacterial biofilm) promoting planula settlement and growth on their surfaces.
6. Planulae and young colonies attached to calcareous algae at the base of algal turf are protected from predatory/grazing organisms and from desiccation and bleaching in the intertidal zone. Coral growth is enhanced by the accumulation of zooplankton and other organisms in algal turf.

Seasonal changes in meiofauna communities in Tien Yen Estuary (South China Sea, Vietnam)

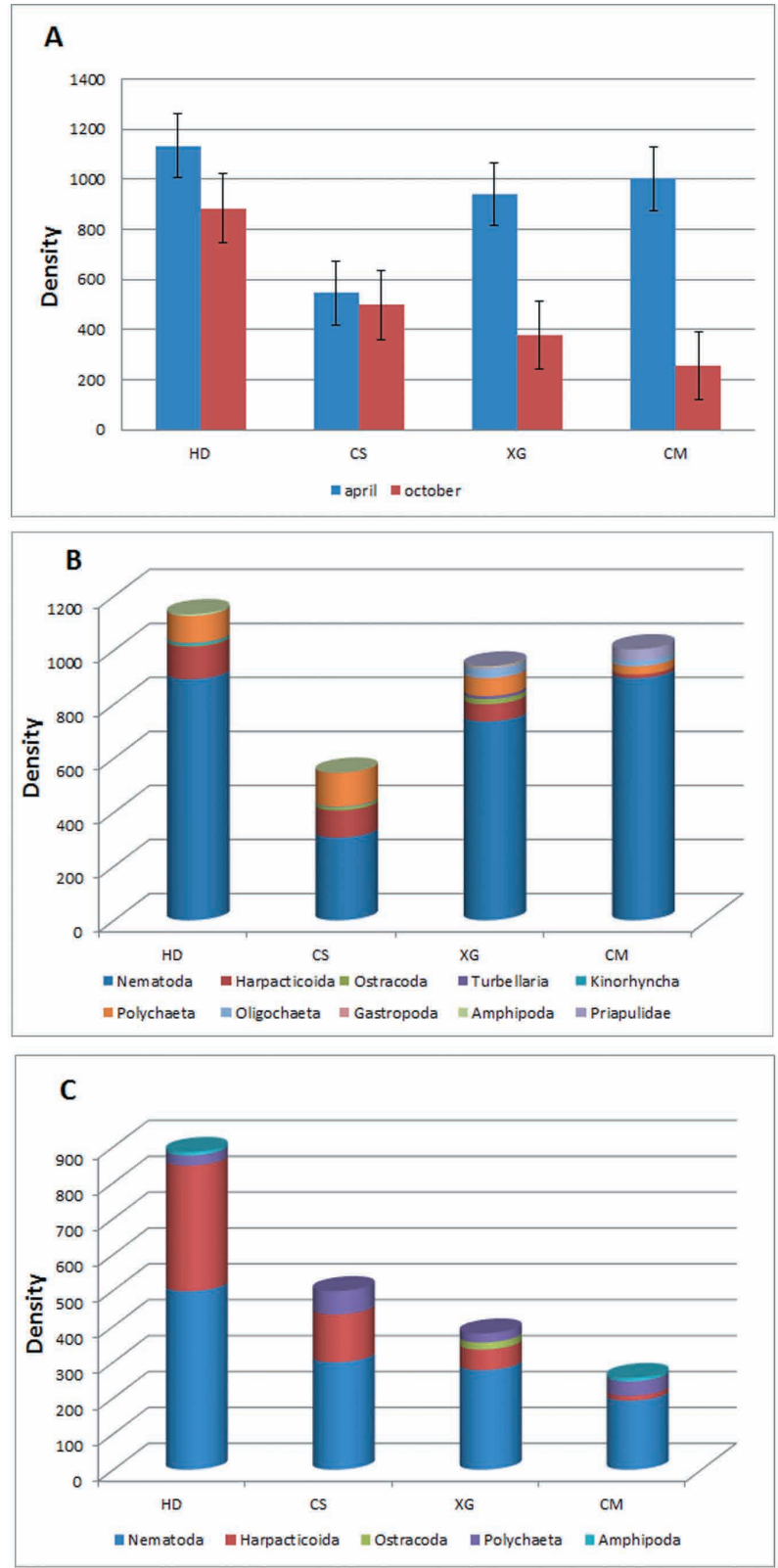
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Marine seagrasses are widely distributed in the coastal tropical and temperate areas. Due to high production capacity and diversity of fauna they create prominent habitats in coastal ecosystems. Seagrasses fulfill considerable role in primary production and organic matter cycling, in nutrients massing and deposits stabilization. The purpose of this study is to survey the seasonal changes in density, species biodiversity and trophic groups of free-living marine nematode communities inhabiting seagrass meadows with different seagrasses ratio in tropical deltaic system within Tien Yen District of Quang Ninh Province, Vietnam. The Tien Yen Estuary is located in the Tien Yen Bay and connects the river with the East Sea, north east of Viet Nam. The most important ecosystems in the estuary include mangroves, seagrass and intertidal mud flats. Sampling was conducted in April and October, 2015 on intertidal mud flats in Ha Dong (HD), Con Mat (CM), Xom Giao (XG) and Cua Song (CS) where seagrasses abundantly distributed. Water depths varied from 0.5 to 0.8 m. Chemical properties of the bottom layer determined with Water Quality Checker model WQC-22A. Granulometric composition of the sediments was determined by the separation of the sediment samples of natural humidity by two fractions: below 0.1 mm and above 0.1 mm. Even this rough classification allowed us to determine two basic types of the sediments: silted sands (stations HD and CS) and heterogenous silts with the small admixture of sand (stations CM and XG). In the meadows of the Tien Yen Estuary, low density and poor taxonomic diversity of meiobenthos were noted. Total meiofauna densities in April 2015 ranged from 547 ± 208 to 1148 ± 270 ind/10 cm². Highest density and diversity levels were reported for the Ha Dong site, at tide pools and puddles with high diversity of seagrasses (6 species: *Halophila ovalis*, *H. beccarii*, *Halodule pinifolia*, *H. uninervis*, *Zostera japonica*, *Ruppia maritime*) (see Figure, A). The taxonomic composition consisted of 6 groups: Nematoda, Harpacticoida, Ostracoda, Kinorhyncha Polychaeta and Amphipoda, nematodes were the dominant group accounting for 76.8% of the total meiobenthos density. The lowest density was marked at station Cua Song in estuary (3 species of seagrasses: *Z. japonica*, *H. ovalis*, *H. pinifolia*). The taxonomic composition of meiobenthos consisted of 5 groups (Nematoda, Harpacticoida, Ostracoda, Kinorhyncha and Polychaeta) (see Figure, B), nematodes dominated – 49.2%. In October there was a decrease in salinity and a change in the composition of bottom sediments, and as a consequence - a decrease in the density and taxonomic composition of meiobenthos (see Figure, A). Highest density and diversity levels were noted for the Ha Dong site (885 ± 127 ind/10 cm²) (see Figure, C). The taxonomic composition consisted of 4 groups: Nematoda, Harpacticoida, Polychaeta and Amphipoda, nematodes were the dominant

group accounting for 56.2%. The lowest density (256 ± 112 ind/10 cm².) was marked at station CM (1 species of seagrasses: *H. ovalis*). The taxonomic composition consisted of 4 groups Nematoda, Harpacticoida, Polychaeta and Amphipoda, nematodes were the dominant – 75.6%. Seagrass meadows sometimes create hydrodynamic “silent” benthic pools. However, in Vietnam’s monsoon climate featuring well-defined wet and dry seasons, littoral organisms are greatly affected by continental discharge that leads to ground structure changes. Precipitation in form of powerful rainfalls shows detrimental influence on meiobenthic community during the low tide, drastically decreasing the salinity of water and surface ground layer. Apparently, these factors affect seasonal changes in density, species diversity and nematode trophic groups. Thus, our studies confirm that the main factor responsible for seasonal changes of nematode community in intertidal zone of Tien Yen estuary is the precipitation (heavy rainfalls).



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The quantitative characteristics and structure of meiobenthos communities in April and October 2015. **A** – total meiofauna densities (ind/cm²) in April and October; **B** – density (ind/cm²) and taxonomic composition of meiobenthos in April; **C** – density (ind/cm²) and taxonomic composition of meiobenthos in October.

The monitoring technology of organochlorine contamination in marine ecosystems

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Organochlorine pollutants are harmful and toxic substances affecting biota. Organochlorine pesticides, such as isomers of HCHs, DDT and its metabolites, are used in southern Hemisphere and may occur in polar regions due to atmospheric transfer. In this study, Pacific salmon, seabirds and marine mammals are used as bioindicators for assessment of the ecological status of the Sea of Okhotsk and the Bering Sea.

Fish inhabit almost the all aquatic environment of the planet. Most fish species are good accumulators OCPs. The many species of fish correspond to required criteria of bioindicators for the assessment of aquatic ecosystems. Therefore, fish are considered as one of the best organisms for the study of bioaccumulation characteristics and effects of pollutants on the ecosystem. Seabirds are useful bioindicators for organochlorine pollutants monitoring because they often are at the top of the food pyramid. Non-migrating birds can reflect the background contamination of their habitat. If there are no local pollution sources, birds reflect the global pollution resulting from the trans-boundary transfer of pollutants. Marine mammals can be considered key species for monitoring of persistent organic pollutants (POPs) in the marine environment all over the world. They can be used as bioindicators of global pollution and also as biomonitors of modern trends in anthropogenic pollution of the biosphere.

Isomers of HCH and DDT and its metabolites were detected in organs of marine mammals (*Eschrichtius robustus*, *Odobenus rosmarus divergens*) from the Bering Sea. Maximum concentration was obtained in walrus liver (90263 ng/g lipid). Seabirds (*Larus schistisagus*, *Aethia cristatella*, *Aethia pusilla*, *Fulmarus glacialis*, *Oceanodroma furcata*) were collected in the Okhotsk Sea. Total maximal pesticide concentration was determined in pen with skin of the Crested Auklet (16095 ng/g lipid), minimal concentration – in pen of the Northern Fulmar (29 ng/g lipid). Pacific salmon (*Oncorhynchus gorbusha*, *Oncorhynchus keta*, *Oncorhynchus nerka*, *Oncorhynchus tshawycha*) are the most abundant on epipelagial of North Pacific. In pink salmon, total pesticides concentration in body burden was about 20 ng/g, in chum salmon – 15 ng/g. High pesticide concentration was detected in sockeye liver (225 ng/g wet weight) and gonads (390 ng/g). Salmon are considered to be an important vector in the transport of pollutants in the North Pacific.

The use of bioindicators organisms facilitates the monitoring of the aquatic environment and indicates the degree of ecosystems contamination. The presence of considerable concentrations of pesticides in marine organisms from the Sea of Okhotsk and the Bering Sea, which areas are very far from the regions of industrial activities and pesticides application, is not a surprise, but demonstrate and confirm general global pesticides background existing in the world today.

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DNA barcoding and species diversity of fishes from the Lake Khanka

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Khanka is the largest lake on the far-eastern territory of Russia comprising the valuable part of the national resources in the field of freshwater fishery. Lake Khanka has a low value of endemism due to the recent origin and connection with large river systems of Amur and Ussuri. Currently the lake is undergoing the increasing impact of the human activity often being the cause of invasions including intentional introduction of some fish species for stabilization of relationships among communities and exhaustive utilization of the natural productivity of this lake. This makes challenging the development and implementation of the powerful techniques for documentation of species diversity of the Lake Khanka.

Previous attempts to describe the fish species diversity of Lake Khanka have been made based on the classic non-unified methods. Here for the first time we present the complex approach to documentation of fish species diversity.

We collected 65 fish specimens from 17 species representing 4 families within 3 orders of ray-finned fishes and analyzed their taxonomy and species diversity using both classic methods and DNA barcoding techniques and protocols. Specimens have been genotyped based of *Co-1* mitochondrial gene marker. Mean values of *K2P*-corrected intraspecific genetic distances were 0.1% (0–0.7), distances between different species within the same genera were 4.07% (0.15–6.6) and distances within families between different genera varied from 3.8 to 23.4 with 14.1% as a mean value. Phylogenetic analysis revealed monophyletic origin of all species clusters with large support (98–100% of bootstrap values), thus, supporting reciprocal complement of morphological and genetic species boundaries and confirming the great performance of the *Co-1* DNA barcoding in field of documentation of fish species diversity in the Lake Khanka.

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The usage of marine organisms for ecological monitoring of the Koz'mino Bay influenced by Transneft Kozmino Port, LLC

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The project for complex monitoring of the Koz'mino Bay influenced by the oil-landing terminal of "Transneft Kozmino Port, LLC" was established in 2010. Marine organisms as natural habitants were proposed for the purpose.

The monitoring aim is to achieve reliable data on ecological state of the aquatory. Further, man-made component of ecosystem transformation (oil-landing terminal activity) is to be established, reduced and prevented.

Complex monitoring of the Koz'mino Bay is a system of regular observations of natural ecosystems and artificial colonies of marine organisms (*Patinopecten yeyessoensis*, *Mytilus trossulus*, *Laminaria japonica*). They are grown on a special maricultural polygona belonging to "Transneft Kozmino Port, LLC". It is situated close to the oil-landing terminal (in 250 m). The monitoring is performed annually and includes 4 directions.

- Control of heavy metal concentration in tissues of marine organisms, estimation of their quantity, biomass, size and weight characteristics, survivability, reproduction timing and amount of span.
- Research on benthic community's state (with the privilege of mass macrobenthos species as indicators of environmental quality).
- Qualitative and quantitative composition of plankton communities in the Koz'mino Bay, with special interest in larvae which are sensitive to water pollution.
- Analysis of oil carbohydrates in marine water.

The achieved results reveal that oil carbohydrates concentration didn't increase. It is lower than MPC for fishery aquatories (0.05 mg/dm³). The state of marine organisms, their reproduction state and survival levels are common for the Southern Primorye. We didn't find any accumulation of heavy metals or carbohydrates in marine organism tissues, so that they are safe to eat. Both benthic and plankton communities are characterized by high biodiversity, found out to be stable, successfully reproducing and passing through all stages of the life cycles. The community structure is characteristic to the Nakhodka Bay. All the data demonstrate that ecological state of the Koz'mino Bay is stable despite the influence of oil-landing terminal.

Marine organisms – a treasure trove of novel, valuable, and uncommon lipids

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Marine organisms are a rich source of bioactive lipids with many of them absent or present at only low levels in terrestrial organisms. Here we report some of compounds we either discovered or reliably quantified for the first time.

Nisinic acid, an Omega-3 tetracosahexaenoic acid, is a precursor (= a hidden form) of docosahexaenoic acid, DHA. DHA is currently a popular component of dietary supplements for maintaining brain and overall health. We have found tetracosapolyenoic acids are characteristic for species relating to subclass Octocorallia (Alcyonaria, Gorgonaria, Helioporidae and Pennatularia). Gorgonian corals and sea feathers, a usual by-catch in fisheries operation both in the northern and southern seas and oceans might contain high levels of nisinic acid. Another example is detection family of C26 PUFA, including fatty acid 26:7(n-3) in Ophiura.

Phospholipids of edible brown algae attracted our attention due to an unusual pattern of signals observed in ³¹P-NMR experiments. A number of phospholipid candidates have been synthesised, and the structure of the main component was confirmed as that of phosphatidyl-O-[N-(2-hydroxyethyl) glycine]. An uncommon arsenic-containing phospholipid was found and quantified in both fresh algae and kelp dietary supplement, where its content was especially high, over 60 mg/kg.

Another interesting group is phosphonolipids, commonly found in shellfish, where, according to our studies, they might account for up to ¼ of phospholipids. Unlike most phospholipids, phosphonolipids have P-C bond, which is resistant to enzymatic hydrolysis, and thus may interfere with sphingolipids metabolism (which plays important role in controlling cell proliferation and apoptosis). The presence of phosphonolipids in oysters, mussels and other edible molluscs warrants the studies of their biological activities

While working with different species of red algae *Gracilaria* in both North Pacific and South Pacific we have observed distinctive eicosanoid profiles: for example, *G. vermiculophylla*, an edible alga from the Sea of Japan that, if mishandled, might produce a lethal (if ingested) set of prostaglandins, while its South Pacific relative, *G. chilensis*, under the same conditions produces no prostaglandins, but a set of leukotrienes, including unusual, 5,8-dihydroxyeicosatetraenoic acid.

The above mentioned lipids are either unavailable commercially, or are supplied as expensive synthetic compounds. As a result, only a limited number of reports highlighting their (potentially beneficial) properties have been published. At the same time, these compounds might be obtained with a lesser effort from marine organisms: for example, a prominent researcher in algal bioactives,

Dr William Gerwick (Scripps Institution of Oceanography, Center for Marine Biotechnology and Biomedicine, San Diego, United States), reported that algae represent a very substantial resource of this valuable class of specialty chemicals, and that “upon recognition of the structural identity of this natural product, we realized that (in our research) we had employed approximately \$100,000 worth of material (30 mg) at current market prices!”.

Economically important red seaweeds along china's coast: resources and utilization

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Red algal resources are abundant along the China Sea coast, many of which are economically important. In this paper, traditional uses of 60 red algal seaweeds ever recorded for various purposes were summarized and categorized. There are 34 edible species, 21 agarophytes, 13 carrageenophytes, nine species used as coating additive and seven species used for medical or textile purposes. These species are subordinated to seven orders and 15 families. A few species have been practiced to be potential candidates in the integrated multi-trophic aquaculture system (IMTA). Natural habitats and geographical distributions of these resources are illustrated based on marine algal flora of China seas. Seaweeds aquaculture supplies the source of sustainable biomass; *Pyropia* and *Gracilaria* are the current dominating cultivated species in China, while *Eucheuma* farming is facing challenge to the sustainability, and a general view of other red seaweeds farming, as well as the status of seaweed-based food and hydrocolloids industry are summarized. Since coastal aquaculture may cause adverse impacts on the coastal marine environment, on-going IMTA seems to offer a better solution and deep sea aquaculture will probably be developed in the future. Scientific approaches have revealed lots of valuable bioactive compounds stocked in red seaweeds which may have pharmaceutical applications, and the rich carbon source fixed through photosynthesis by red seaweeds will supply biomass for future bioethanol production. In general, the utilization of red seaweeds in China has been converting from traditional scattered uses to modern industrialized uses, and a high-efficient utilization will represent the future prospects for the red seaweeds.

Trophic structure of dominant species at a cold seep in the South China Sea

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Cold seeps occur in geologically active and passive continental margins and are rich in hydrocarbons, which are discharged into the surrounding seawater and sediment during the leakage process of fluids from subsurface reservoirs by pressure gradients. Compared with coastal zones and the common deep-sea environment, this extreme condition nurtures a special ecosystem. Biodiversity and biomass are extremely rich in hydrocarbon ecosystems. Abundant communities of animals (including tubeworms, polychaetes, crabs, mussels, clams, gastropods and fishes) are dependent on chemoautotrophs that derive energy from hydrocarbon sources. Nutritional relations among invertebrates from the cold seep at the summit of the Formosa Ridge southern peak of the South China Sea were studied via carbon and nitrogen stable isotope approach. A number of specimens of different species (the mussel *Bathymodiolus platifrons*; the gastropods *Bathymacmaea* sp., *Provanna glabra* and *Phymorhynchus buccinoides*; the squat lobster *Shinkaia crosnieri*; the shrimp *Alvinocaris longirostris*; the commensal scale worm *Branchipolynoe pettibonae* and the crab *Paralomis* sp.), seawater particulate organic matter (POM) and sedimentary organic matter (SOM) were analysed. The percentage of food source contributing to dominant species were calculated using the Stable Isotope Analysis in R package (SIAR in R software). $\delta^{13}\text{C}$ and $\delta^{15}\text{N}$ values ($-63.8071 \pm 1.14106\%$, $-0.0542 \pm 0.59481\%$, respectively) in *B. platifrons* were most negative among all invertebrates, which reflected that the carbon source was methane. To *B. platifrons*, 97.02% of trophic source was from endosymbiotic chemosynthetic bacteria. To *Shinkaia crosnieri* and *Alvinocaris longirostris*, scale worms, mussels and bacteria were primary sources. A suggested food web model for the cold seep ecosystem at the summit of the Formosa Ridge southern peak was elaborated according to the above associations and $\delta^{13}\text{C}$ and $\delta^{15}\text{N}$ values. Chemolithotrophic bacteria form the basis of the food web. Mussels and little gastropods are primary consumers. Shrimps, squat lobsters, commensal scale worms and large gastropods are secondary consumers. The top predators are bathyal species which make incursions into the ecological system.

Application of chlorophyll fluorescence technique for monitoring of oil pollution impact on macrofitobenthos in shallow coastal areas

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Due to the growing consumption of petroleum products, oil pollution is increasingly frequent in aquatic ecosystems. Oil and its derivatives have been found to be toxic to a wide array of marine organisms, including the representatives of macrofitobenthos, one of the major primary producers in the marine environment. Unfortunately, studies of petrochemical impacts on marine macrophytes are rare and largely confined to observations of spill events or laboratory physiological studies. This work was devoted to the application of chlorophyll fluorescence technique (Diving-PAM fluorimetry) as a non-invasive early “diagnostic” tool to be used in monitoring of coastal macrofitobenthos health under oil pollution pressure. We used field survey to examine special variation in the bulk values of minimal and maximal chlorophyll fluorescence (F_t and F_m' , respectively) in the light, as well as the effective quantum yield (ϕ_{PSII}) and the relative electron transport rate ($rETR$) in the dominant species of macrophytes at differentially oil-polluted upper-subtidal areas of the Peter the Great Bay (Sea of Japan, Russia). The results clearly showed that ϕ_{PSII} and $rETR$ were the most sensitive measures of the photosynthetic processes affected by oil-fuel. Among macrophytes investigated, four species (seagrass *Zostera marina*; brown macroalgae *Costaria costata*, *Sargassum miyabei*; red macroalgae *Neorhodomella larix*) were found to be sensitive bioindicators of oil pollution. Statistical treatment of the data revealed that the reference values for the ϕ_{PSII} of the above mentioned species should be in the interval (0.52–0.61; 0.58–0.65; 0.55–0.63 and 0.62–0.73, respectively) for normal (untreated) macrophyte populations. The values range for the $rETR$ of untreated pollution-sensitive species should be in the interval of 75–86; 40–60; 75–80 and 45–55, respectively. Macrophytes at sites with seawater oil concentrations above 0.12 mg/L are characterized by a 1.5–2-fold decrease in both the ϕ_{PSII} and $rETR$ biomarkers compared with non-polluted areas. In all, the use of the chlorophyll fluorescence parameters ϕ_{PSII} and $rETR$ proved to be sensitive indicators of early oil-fuel toxicity in marine macrophytes from shallow coastal areas. This technique is suggested for effective monitoring the onset and development of stress in upper-subtidal macrophyte populations when exposed to oil pollution pressure.

Growth and energy budget of the red king crab, *Paralithodes camtschaticus* larvae

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The red king crab, *Paralithodes camtschaticus* is an important crustacean fishery resource in the east sea of Korea. We studied the energy budget of red king crab larva. Carapace length of larval development stage is as follows; zoea I: 1.43 mm, zoea II: 1.59 mm, zoea III: 1.76 mm, zoea IV: 1.95 mm.

Larvae of *P. camtschaticus* were reared at three different temperature regimes (Natural sea water temperature, 8 and 10°C) to measure intermolt period, molt increment and survival. Mean intermolt period of larval stage were 43.1 days, 43.2 days and 32.3 days in NT, 8°C and 10°C, respectively. Intermolt period was declined with increasing temperature, whereas increased with size and instar number.

The survival rate at larval stage at NT and 10°C were higher than those NT. The survival rate at zoea I to zoea IV was 67.5% at NT and 70% at 8°C and 60% at 10°C.

The energy budget of the red king crab, *P. camtschaticus* larvae reared in the laboratory from zoea I to zoea IV was studied. Energy used by the larvae reared at three different temperatures (NT, 8 and 10°C) were calculated from data on feeding, growth, molting and metabolism. The energy used by growth during the larval stages at NT, 8 and 10°C were 14.52J, 15.09J and 14.62J, respectively. These results suggest that growth and metabolism of red king crab larvae may be driven by water temperature.

Lipids and fatty acids of nudibranch mollusks: potential sources of bioactive compounds

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The molecular diversity of chemical compounds found in marine animals offers good chance for discovery of novel bioactive compounds of unique structures and diverse biological activities. Mollusks, as well as the invertebrates in general, constitute a source of lipid bioactive compounds offering a variety of activities. Nudibranch mollusks that are not protected by a shell and produced chemicals for various ecological uses, including defense against predators, attract great interest to their lipid composition. Lipid analysis of eight nudibranch species has demonstrated for the first time that nudibranchs exhibit a wide diversity of lipids and differed greatly from that of other marine gastropods. This study revealed dominant phospholipids. Their concentration varied within a range from 74% in *Chromodoris geometrica* to 82% in *Glossodoris cincta*; this was much more than was found in other mollusks and invertebrates in total. Triacylglycerols, monoalkyldiacylglycerols and free fatty acids, which are the storage compounds of the cells, were minor components. The level of the neutral storage lipids is known to be species specific and depends mainly on the life history strategy and food availability. The phospholipid composition of the studied species was similar, with the dominance of phosphatidylcholine (up to 63% in *Chromodoris tinctoria*) and further, in descending order: phosphatidylethanolamine, phosphatidylserine, ceramide-aminoethylphosphonate, phosphatidylinositol and diphosphatidylglycerol. Among polar lipids 1-alkenyl-2-acyl glycerophospholipids (plasmalogens) were found in the mollusks. The highest percentage of phosphatidylethanolamine plasmalogens was found in *Risbecia tryoni*, accounting for 65%, and the phosphatidylserine plasmalogens contribution reached 61% in *Platydoris* sp.

The nudibranchs exhibited some unique features in their fatty acid composition. The fatty acid compositions of the nudibranchs differed greatly from those of other marine gastropods and exhibited a wide diversity: very long chain fatty acids (VLCFA), a series of non-methylene-interrupted fatty acids including novel 21:2 Δ 7.13 and an abundance of various odd and branched fatty acids typical of bacteria. In the nudibranchs, a significant amount of VLCFA specific to sponges, so-called demospongiic acids, was found. These nudibranchs are carnivorous and specialized feeders on sponges. Utilization of this food probably is responsible for the high level of the demospongiic acids in these mollusks. It is suspected that the majority of sea slugs feed on certain sponge species, which are known to be distinguished in their fatty acid composition. Indeed, a series of VLCFA with double bonds at Δ 5.9 positions in the chain was identified in the tropical nudibranchs. Concentrations of these components differed among the species. The specific distribution of the VLCFA suggests that these nudibranchs may feed on different sponge species.

Thus, the unique fatty acid composition of the nudibranchs is determined by food supply, inherent biosynthetic activities and intracellular symbiotic microorganisms. Symbiotic bacteria revealed in some species of nudibranchs participate presumably in production of some compounds serving for chemical defense of the mollusks. The current study has shown that these mollusks may be important resource of a wide range of bioactive compounds.

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The qualitative and quantitative composition of autumn phytoplankton in Peter the Great Bay, the northwestern Sea of Japan during 2012–2015

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The species composition and quantitative distribution of the phytoplankton were studied in Peter the Great Bay, Russian waters of the East/Japan Sea. Amursky and Ussuriisky bays are the second order bays in Peter the Great Bay. Long-term studies of the species composition of phytoplankton carried out during the autumn period from 2012 to 2016 revealed a total of 110 species of planktonic microalgae from five divisions. An annotated checklist of species is presented. The highest concentrations of phytoplankton ($1.8 \cdot 10^6$ cells/L) were recorded in November 2012 in Patrokl Bay and the lowest ($2.3 \cdot 10^4$ cells/L) in November 2015 in Lazurnaya Bay. Diatoms *Aulacoseira granulata*, *Skeletonema costatum*, *Thalassionema frauenfeldii*, *Rhizosolenia pungens*, *Pseudo-nitzschia calliantha*, *P. multistriata*, and species belonging to *Pseudo-nitzschia pseudodelicatissima/cuspidata* complex, and dinoflagellates *Torodinium robustum* and *Gymnodinium* sp. were predominant (30–85%). Potentially toxic diatoms belonging to the genus *Pseudo-nitzschia* were the most common bloom-forming microalgae. Morphological analysis based on transmission electron microscopy revealed *P. multistriata* and *P. calliantha* were the most numerous species. *Pseudo-nitzschia* species reached high cell densities (1.2 – $1.5 \cdot 10^6$ cells/L) in November 2012 in Patrokl, Lazurnaya and Sobol bays, when *P. multistriata*, *P. calliantha*, and species belonging to *Pseudo-nitzschia pseudodelicatissima/cuspidata* complex predominated. Furthermore potentially toxic *Pseudo-nitzschia* species exceeded their the threshold value ($1 \cdot 10^5$ cells/L) accepted in Europe as permissible limit for harvesting cultured mollusks (Andersen, 1996) on six occasions at stations in Ussuriisky Bay ($3.1 \cdot 10^5$ cells/L in October 2013 in Sukhoputnaya Bay; $2.1 \cdot 10^5$ cells/L in November 2013 in Lazurnaya Bay; $1.3 \cdot 10^5$ cells/L in November 2013 in Paris Bay; $4 \cdot 10^5$ cells/L in October 2015 in Sukhoputnaya Bay; $1.7 \cdot 10^5$ cells/L in October 2015 in Patrokl Bay; $2.2 \cdot 10^5$ cells/L in October 2016 in open waters of Ussuriisky Bay). Maps showing the distribution of *Pseudo-nitzschia* species and their density distributions in the studied areas are provided. Zones with increased risk of amnesic shellfish poisoning were found in Ussuriisky Bay.

This study was supported by the Russian Foundation for Basic Research (project No. 15-04-05643a).

**Sequence divergence in the genus *Tribolodon*
(Cypriniformes: Cyprinidae)
based on mtDNA and nDNA markers and its applications
to the systematics and genetics of speciation of redfin**

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The genus *Tribolodon* (Sauvage, 1883) is a member of the most common fish family, Cyprinidae, and, possibly, one of the largest families of vertebrates. To clarify relationship of species of the genus *Tribolodon* in the Russian part of their distribution ranges, 2 mitochondrial markers (*Co-1* and *Cyt-b*), a nuclear marker (*Rho*), and a gene marker (rRNA) of internal transcribed spacer (*ITS-1 – 5.8S – ITS-2*) were used. Depending on the marker, the different numbers of the species groups were detected by the ABGD method. These results were compared with the analysis of phylograms and data showed generally the support for known species clusters and regional intraspecies groups. A complex analysis of sequences from three redfin species within the area of the study, based on four marker genes and methods of molecular phylogenetics, ordination of genetic distances (ABGD software), recombinant analysis (RDP software), and population genetic approaches (MEGA-6 and DNA-SP5 software) is performed. Presented data revealed clusters of three commonly recognized species, regional intraspecific groups or individuals of local populations, and few hybrid individuals. The capture of hybrids in Kievka and Vostok bays supports the theoretical possibility of existence of a hybrid zone for *Tribolodon* in southern Primorsky Krai. However, the frequency of hybrid occurrence remains poorly studied. The findings of the present study support the existence of three formerly established species of the genus *Tribolodon* in southern Primorsky Krai and Sakhalin Island: *T. hakonensis*, *T. brandtii*, and *T. sachalinensis*. DNA barcoding technique proved to be efficient with the use of two mtDNA markers: *Co-1* and *Cyt-b*. Some insertions and substitutions within the *ITS-1 – 5.8S – ITS-2* have been found. This nuclear rRNA gene marker has a relatively high variability that allowed us to identify each of the three redfin species of the genus *Tribolodon*. *ITS-1 – 5.8S – ITS-2* marker has been sequenced for *Tribolodon* species for the first time, and it can be used in further studies for species identification and phylogenetic reconstructions of redfins, which is a challenging task. The results obtained on inter- and intraspecies differentiation do not confirm a sufficient level of divergence for defining any new taxa of a species rank in the genus *Tribolodon*.

Comparison of fungal complexes of Japanese scallop *Mizuhopecten yessoensis* from different areas in the Peter the Great Bay of the Sea of Japan

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Mycological investigation of the Japanese scallop *Mizuhopecten yessoensis* (Jay, 1856) (Bivalvia) from different areas of the Peter the Great Bay (Sea of Japan) was conducted. Subjects of mycological studies were the specimens of Japanese scallop *Mizuhopecten yessoensis* collected in various regions of the Peter the Great Bay. The following sites were selected for mollusk collection: Amursky Bay aquatic area exposed to industrial and household wastewaters of the city of Vladivostok; aquatic area around the Rikorda Island, where the bivalve mollusk breeding farm is located; and the relatively pure waters of the Vostok Bay.

In total, 72 species of filamentous fungi from 30 genera of ascomycetes, anamorphic fungi, and zygomycetes were isolated from internal organs of Japanese scallop.

The complex of filamentous fungi associated with the Japanese scallop from the Amursky Bay includes 35 species of fungi from 16 genera; 33 of the species belong to anamorphic micro-mycetes from 14 genera: *Acremonium* (3), *Aspergillus* (10), *Beauveria* (1), *Chrysosporium* (2), *Cochliobolus* (1), *Geomyces* (1), *Gliomastix* (1), *Phialophorophoma* (1), *Scopulariopsis* (1), and *Trichoderma* (1); also, there is a single species of ascomycetes (*Chaetomium globosum*) and a single zygomycete (*Pilaira anomala*) (Fig. 1).



Fig. 1. Taxonomic composition of the complexes of filamentous fungi associated with the Japanese scallop from various regions of the Peter the Great Bay (Sea of Japan).

The complex of filamentous fungi associated with the Japanese scallop from the coastal waters of the Rikorda Island comprises 39 species of filamentous fungi from 19 genera; among them, 30 species belong to anamorphic fungi from 15 genera: *Acremonium* (3), *Alternaria* (2), *Aphanocladium* (1), *Aspergillus* (5), *Botrytis* (1), *Cladosporium* (1), *Fusarium* (2), *Geomyces* (1), *Gliomastix* (1), *Hyalocylindrophora* (1), *Isaria* (1), *Penicillium* (8), *Phialophorophoma* (1), *Sarocladium* (1), and *Scopulariopsis* (1); 4 species belong to ascomycetes from 3 genera: *Eurotium* (1),

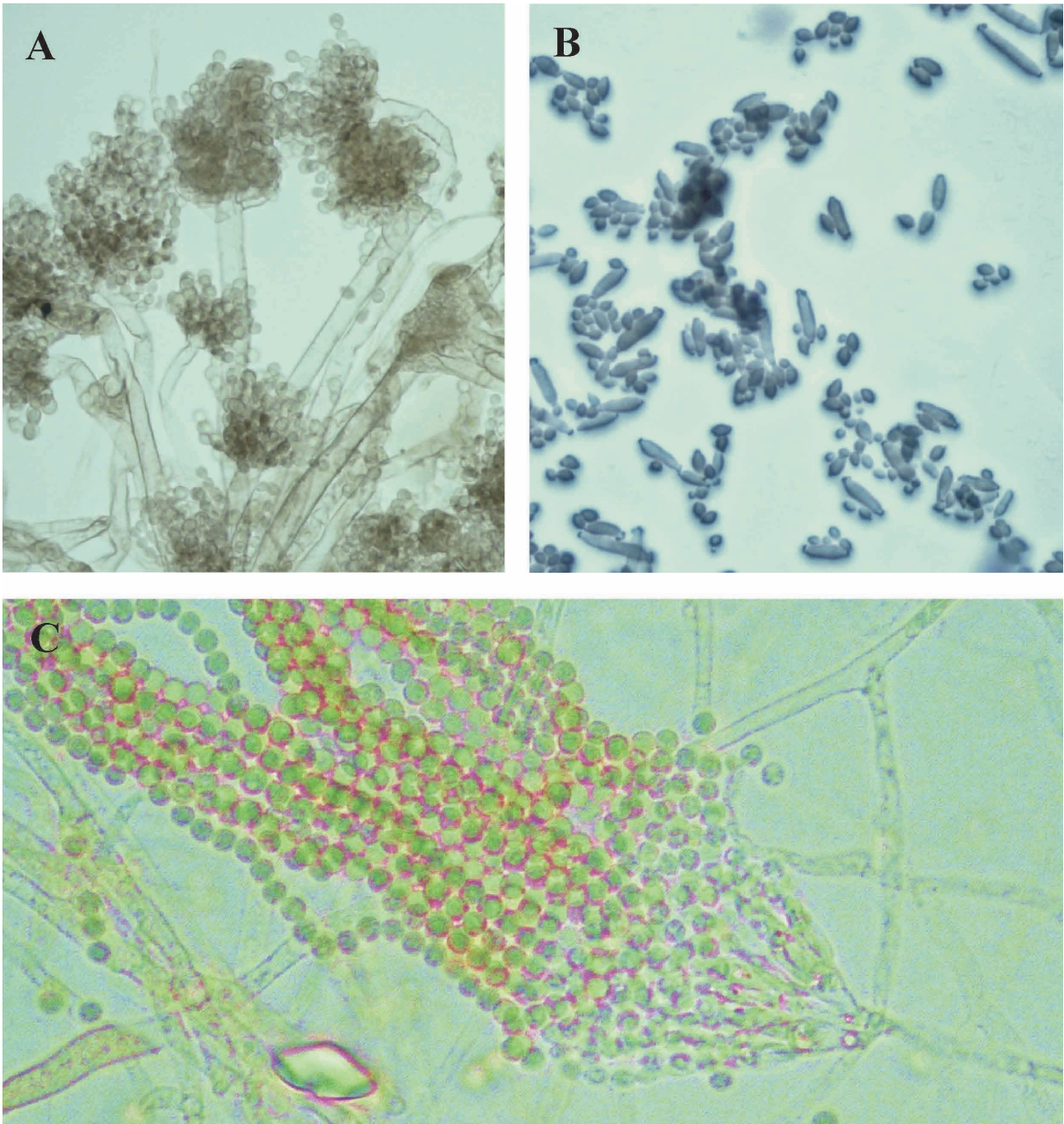


Fig. 2. Filamentous fungi: *Aspergillus melleus* Yukawa (A); *Cladosporium cladosporioides* (Frezen.) G.A. de Vries (B); *Penicillium simplicissimum* (Oudem.) Thom (C).

Purpureocillium(1), and *Talomyces* (2); 4 species belong to zygomycetes from the *Mucor* genus; and one species belongs to Mycelia Sterilia (Fig.1).

The Vostok Bay was chosen as a control aquatoria that is subjected to anthropogenic influence the least. The complex of fungi associated with Japanese scallop from this region comprised 14 species from 8 genera; among them, 14 species belonged to anamorphic fungi from 7 genera: *Alternaria* (2), *Aureobasidium* (1), *Aspergillus* (1), *Cladosporium* (3), *Geomyces* (1), *Penicillium* (4), *Trichoderma* (1); and a single zygomycete *Rhizopus stolonifer* (Fig. 1).

A low degree of the similarity in species composition of complexes of the filamentous fungi in bivalves from marine habitats with different indicators of environmental quality and degree of anthropogenic load was revealed.

Species richness of the conditionally pathogenic and toxigenic filamentous fungi, primarily the fungi of the genera *Aspergillus*, *Penicillium*, *Cladosporium* (Fig. 2), and *Chaetomium*, in internal organs of the bivalve mollusks increases in contaminated coastal waters.

Expedition of the National Scientific Center of Marine Biology FEB RAS to the Chukchi Sea

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These are the primary results of the expedition of the National Scientific Center of Marine Biology, Far Eastern Branch, Russian Academy of Sciences (NSCMB FEB RAS) to the Chukchi Sea aboard the research vessel “Academic Oparin” in September–October 2016. The main goals of the expedition were to conduct combined investigations of the Chukchi Sea ecosystems and to search for climate change footprints. To this end, biological materials were collected by several research instruments, including an underwater remotely operated vehicle (ROV) as the newest method of ocean observation that allows damage-free monitoring of marine ecosystems. Also, investigations were carried out in the same stations as those of the expedition of the icebreaker “Krasin” in 1935, in order to compare the results. More exact data were obtained by us about some benthic invertebrates dominating in the Chukchi Sea. The biggest population of tube-dwelling polychaetes with a high population density not recorded before was registered in the southern part of the Chukchi Sea. The stable isotope ($\delta^{13}\text{C}$ and $\delta^{15}\text{N}$) values for two crab species *Chionoecetes opilio* and *Hyas coarctatus* show that the benthic population of the south Chukchi Sea is influenced by Anadyr water, while fauna of the Chukotka nearshore and Wrangel Island areas depends on the Siberian Coastal Current and the East-Siberian Sea. It has been found that Herald Island’s nearshore flora has a Pacific Ocean origin. Fourteen algal species have been found for the first time in the Russian sector of the Chukchi Sea. Changes of some dominating fish species have been recorded in several parts of the Chukchi Sea. Rich and diverse collections of marine animals of the Chukchi Sea were made and placed to Museum of NSCMB FEB RAS for future species identification. Rich video materials can be used for describing different biocenoses to support future environmental investigations in the Arctic. It shows that traditional methods of benthos investigation such as bottom trawling and grabbing do not give a real picture of richness and diversity of bottom life. The use of ROV can be recommended for all environmental investigations in the ocean.

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