INTERNATIONAL SYMPOSIUM

MAPEEG-2019

PROGRAM & ABSTRACTS

MODERN ACHIEVEMENTS IN POPULATION, EVOLUTIONARY AND ECOLOGICAL GENETICS

MAPEEG TENTH ANNIVERSARY!

VLADIVOSTOK & VOSTOK MBS

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PROGRAM

MAPEEG-2019 Held by:
FAR EASTERN BRANCH OF RUSSIAN ACADEMY OF SCIENCES (FEB RAS),
A.V. ZHIRMUNSKY NATIONAL SCIENTIFIC CENTER OF MARINE BIOLOGY,
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Program Committee:
Yuri Ph. Kartavtsev, Dmitry Atopkin, Sergey Turanov, Tadeusz Malewski

SUNDAY, SEPTEMBER 8

Arrival, Hotel accommodation in Vladivostok

MONDAY, SEPTEMBER 9

8-30**: Departure to the NSCMB**

9-30 - 10-15**: Registration**

10-15 - 10-30**: Opening Remarks**

Vyacheslav S. Odintsov, Director of the NSCMB (Russia).
Victor V. Bogatov, Science Secretary of Far East Branch, RAS (Russia).
Gulab D. Khedkar, Paul Hebert Centre for DNA Barcoding and Biodiversity Studies (India).
Tadeusz Malewski, Lab Head at Museum and Institute of Zoology (Poland).
Yuri Ph. Kartavtsev, MAPEEG-2019 Chairman (Russia).

Session 1. Evolutionary Genetics & Genomics
(Oral Presentations)

10-30 - 12-00**: Chair Person – Ph.D. Eugene Gladyshev**

1. **Tadeusz Malewski, Kamil Oleński, Małgorzata Tokarska, Wiesław Bogdanowicz, Stanisław Kamiński**. Comparative genomics of the

posthitis-associated chromosome region in bovines (30').
2. **Kartavtseva I.V., Roslik G.V., Karamysheva T.V. Sheremetyeva I.N., Rubtsov N.B.** Features of additional chromosomes *Apodemus peninsulae* in Upper Amur using data of FISH analysis (20').
3. **Polityko Y.K., Redina O.E.** Renal functions and gene expression in kidney tissues of hypertensive ISIAH rats (20').
4. **Roslik G.V., Kartavtseva I.V.** Distribution of B chromosome morphotypes in *Apodemus peninsulae* (Rodentia) from the Upper Priamurie (20').

Lunch (60')

13-00 - 15-40**: Chair Person – Ph.D. Tadeusz Malewski**

5. **Gladyshev E.** Recombination-independent recognition of DNA homology for meiotic silencing in *Neurospora crassa* (30').
6. **Rutenko O.A., Turanov S.V., Kartavtsev Yu.Ph.** Characterization of the complete mitochondrial genome sequence of the *Opisthocentrus ocellatus* (Zoarcales: Opisthocentidae) and its phylogenetic position among eelpouts (20').
7. **Spiridonova L.N., Surmach S.G.** The OCA2 gene mutations determined the yellow eye color of the Japanese Scops-owl *Otus semitorques* Pennant, 1769 (Strigidae, Aves) (20').

Coffee Break (10')

Session 2. Molecular Systematics, Barcoding and Phylogenetics
(Oral Presentations)

8. **Khedkar G.D., Tiknaik A., Kalyankar A., Prakash B.** Taxonomic validation of species belonging to genus *Clarias* in India (20').
9. **Zolotova A.O., Kartavtsev Yuri Ph.** Morphological variability and genetic diversity of ITS1-5.8S-ITS2 rDNA fragment of three species of the genus *Tribolodon* (Osteichthyes: Cyprinidae) from Far East of Russia (20').
10. **Chichvarkhin A.Yu., Chicvarkhina O.V.** Species diversity of genus *Henricia* in the Sea of Japan (20').
11. **Voronina O.L., Kunda M.S., Aksenova E.I., Ryzhova N.N., Romanenko L.A., Novikova O.D., Gintsburg A.L.** Comparative analysis of transporters in marine and freshwater proteobacteria (20').

#. Discussion on Reports (10').

19-00 - 21-00**: Dinner & Welcome Party**

TUESDAY, SEPTEMBER 10

8-30 - 9-00**: Breakfast**
10-00 - 18-00**: Receipt by the Mayor of Vladivostok City and Tour around Vladivostok City with Visit to Primorsky Aquarium**
12-00 - 13-00**: Lunch**
18-00 - 19-00**: Free Evening**

WEDNESDAY, SEPTEMBER 11

8-30 - 9-00**: Breakfast**

Session 2. Molecular Systematics, Barcoding and Phylogenetics
(Oral Presentations, Continue)

10-00 - 11-50**: Chair Person – Ph.D. Gulab D. Khedkar**

12. **Kartavtsev Y.Ph.** DNA barcoding: from sea food mislabeling detection and biodiversity assessment to modern general biology paradigm validation (30').
13. **Sharina S.N., Chernyshev A.V., Zaslavskaya N.I.** Molecular-phylogenetic research of the *Lottia tenuisculpta* species complex (Patellogastropoda: Lottiidae) (20').
14. **Solodovnik D.A., Tatonova Yu.V., Nguyen H.M.** Clonal variability of *Clonorchis sinensis* in *Parafossarulus* snails (20').
15. **Kramina T.E., Samigullin T.H., Degtjareva G.V., Meschersky I.G., Lysova M.V., Sokoloff D.D.** Genus *Lotus* (Leguminosae): phylogeny and genetic structure of species (20').

#. Discussion on Reports (10').

Lunch (60')

12-50 - 15-00: **Chair Person – Ph.D. Dmitry M. Atopkin**

16. **Oleinik A.G., Skurikhina L.A., Kukhlevsky A.D., Bondar E.I.** New genetic data about the phylogeography of Bering Phylogenetic Group of Charr genus *Salvelinus*: Existence of a cryptic Pleistocene refugium in the Kamchatka River basin (20').
17. **Shumenko P.G., Tatonova Yu.V., Solodovnik D.A., Nguyen H.M.**

The genetic diversity comparison of the *Metagonimus* spp. from different parts of the Amur River basin (20').
18. **Tatonova Yu.V., Solodovnik D.A., Besprozvannykh V.V.** Phylogenetic relationships within Echinochasmidae Odhner, 1910 (Trematoda) (20').
19. **Marchenko Ju., Khaitov V.M., Katolikova M.V., Strelkov P.P.** Reliability of *Mytilus edulis* and *M. trossulus* identification by semi-diagnostic conchological character varies between oceanic and estuarine habitats across Kola Peninsula (White Sea, Barents Sea) (20').
20. **Lectures by SkyGene sponsor team** (In Russian & English). Presentation of two-three themes: Tatiana Timoshenko, Head of Marketing and Business Development. Federico Biella, EMEA Channel Account Manager of 10X Genomics (40').

#. Discussion on Reports (10').

19-00 - 20-00**: Dinner**

THURSDAY, SEPTEMBER 12

Session 2. Molecular Systematics, Barcoding and Phylogenetics
(Oral Presentations, Continue)

9-00 - 10-30**: Chair Person** – **Ph.D. Yuliya V. Tatonova**

21. **Turanov S.V.** The recent divergence of two pricklebacks species (Stichaeidae: *Stichaeus*) revealed by the data on complete mitochondrial genome (20').
22. **Izrailskaia A.V., Tatonova Yu.V., Besprozvannykh V.V.** Variability of ITS1 rDNA region in the Notocotylidae (Trematoda) species (20').
23. **Kartavtsev Yu. Ph., Sharina S.N., Chichvarkhin A.Yu., Chichvarkhina O.V., Masalkova N.A., Lutaenko K.A.** Molecular phylogeny of mussels (Mollusca, Mytilidae) based on three nuclear genes (28S RDNA, 18S RDNA and H3) (20').
24. **Pavlenko M.V., Kartavtseva I.V., Vakurin A.A.** Genetic differentiation and systematics of chromosomally polymorphic rodent, striped hamster *Cricetulus barabensis* sensu lato (Rodentia, Cricetidae): review of achievements in “molecular era” of taxonomy and a role of “old” cytogenetic methods (20').

Coffee Break (10')

Session 3. Microevolution. Population Genetic Structure of Species. Ecological Genetics

10-30 - 11-50**: Chair Person – Ph.D. Marina V. Pavlenko**

25. **Pankova V.V., Malyar V.V., Radashevsky V.I.** Molecular analysis and new records of the invasive polychaete *Boccardia proboscidea* (Annelida: Spionidae) (20').
26. **Pavlova S.V., Shchipanov N.A.** Microevolutionary processes in the common shrew: chromosomal races and hybrid zones (20').
27. **Dudnikov A.Yu.** *Aegilops tauschii* phylogeography and how does ecology form the shape of phylogenetic tree (20').
28. **Cherepanova O.E., Petrova I.V., Sannikov S.N., Sannikova N.S.** Genetic and phenotypic differentiation of *Calluna vulgaris* (l.) hull in Pritobolie and Europe (20').

11-50 - 13-00**: Lunch**

13-00 - 14-50**: Chair Person – Ph.D. Alexander Yu. Dudnikov**

29. **Leoke D.Yu., Trukhina A.V.** The common cuckoo *Cuculus canorus* as an object for research on the genetics basis of the evolution of the strategy of reproduction and migration (20').
30. **Frisman L.V., Brykova A.L.** Genetic Investigation of sable (*Martes zibellina*) of Middle Priamurye region (20').
31. **Rautian M.S., Beliavskaya A.Y., Kiselev A.D.** Triple symbiotic system *Paramecium bursaria*-symbiotic *Chlorella*-chlorella *viruses*: identification and correlated biodiversity (20').
32. **Atopkin D.M.** Comparative population genetic analysis of far eastern trematode species of the family Haploporidae Nicoll, 1914 – a parasites of mullets (20').
33. **Mudrik E., Goroshko O., Smirenski S., Kuznetsova N., Surmach S., Kashentseva T., Politov D.** Genetic differentiation of the vulnerable white-naped crane Antigone vipio (Gruidea, Aves) by mitochondrial DNA data (20').

Coffee Break (10')

Session 4. Poster Presentations (30')

14-50-15-20**: Chair Person – S.D. Irina V. Kartavtseva**

1. **Batishcheva N.M., Brykov V.A.** Characterization of polymorphic dinucleotide microsatellite loci in white-edged rockfish, *Sebastes taczanowskii*.
2. **Batishcheva N.M., Polyakova N.E., Brykov V.A.** Characterization of microsatellite markers for a new species of far eastern redfins of the genus *Tribolodon* (Pisces, Cyprinidae) and cross-species amplification in the related species.
3. **Skurikhina L.A., Oleinik A.G., Kukhlevsky A.D.** Genetic justification of a 3000-km long gap in the range of the smelt (*Hypomesus olidus*, Osmeridae).
4. **Maliar V.V., Sharina S.N.** Comparative phylogenetic analysis of two sister species of genus Lottia Gray, 1833 from the Far Eastern seas of Russia.
5. **Nikulin V.Yu.** New insight on the relationships in genus Rhodiola L. (Crassulaceae) based on the internal transcribed spacer (ITS) rDNA data.
6. **Nikulin A.Yu., Gontcharov A.A.** The study of biodiversity of green soil microalgae based on molecular data
7. **Egoraeva A.A., Yu.V. Tatonova., Prozorova L.A.** The taxonomic status of the Semisulcospiridae mollusks in the Russian Far East.
8. **Kalinina K.A., Tatonova Yu.V., Besprozvannykh V.V.** Phylogenetic relationships of Psilostomatidae (Trematoda) species based on genetic data.
9. **Katugin O.N., Chichvarkhin A.Yu., Chichvarkhina O.V., Zolotova A.O.** Genetic approach to the study of phylogeny of the gonatid squid.
10. **Katugina L.O., Tatonova Yu.V., Besprozvannykh V.V., Solodovnik D.A., Nguyen H.M.** Molecular data for *Erschoviorchis lintoni*, highly pathogenic trematode of fish-eating birds.
11. **Redin A.D., Kartavtsev Yu.Ph.** Molecular phylogeny of Russian Far Eastern flounders (Pleuronectiformes, Pleuronectidae) based on sequences for 16S rRNA genes.
12. **Dutova D.I., Shevchenko O.G., Ponomareva A.A., Turanov S.V., Shulgina M.A., Tevs K.O.** Using molecular genetic techniques for identification of *Skeletonema* (Bacillariophyta) species occurring in the Sea of Japan.
13. **Trukhina A.V., Leoke D.Yu., Nekrasova A.A., Smirnov A.F.** Instinctes as an object of research in the field of evolutionary and ecological genetics.
14. **Voronina O.L., Kunda M.S., Aksenova E.I., Ryzhova N.N., Logunov D.Yu., Gintsburg A.L.** Transport activity in the *Pseudomonas* nosocomial

isolates from different clonal complexes.
15. **Drozdov A.L., Tyurin S.N., Petrov N.B.** Phylogenetic position of brachiopod based on analysis of cytological and molecular genetic attributes.
16. **Zaitceva E.A., Toroschina A.V., Atopkin D.M.** Genetic characterization of Far Eastern hermit crabs of the genus *Pagurus* by mitochondrial 16S rRNA gene partial sequences.
17. **Efimova K.V., Kukhlevskiy A.D., Balanov A.A.** The first complete mitochondrial genome of a new *Lycodes* Reinhardt, 1831 (Perciformes: Zoarcidae) species.
18. **Vu T.K., Kartavtsev Y.Ph.** Discriminant, canonical and ANOVA analyses of otolith shape morphology and their utility for stock assesment and identification of two smelt species, *Hypomesus japonicus* and *H. nipponensis* (Pisces: Osmeridae) from the north-western Sea of Japan.
19. **Kamenskaya D.N., Pankova M.V.** Estimation of exon and intron nucleotide diversity in salmonids growth hormone gene.
20. **Gornikov D.V., Kartavtseva I.V., Roslik G.V.** Varyability variants of upper molars of the korean field mouse.
21. **Oleinik A.G., Skurikhina L.A., Kukhlevsky A.D.** Distribution, Composition and Relationships of Arctic Phylogenetic Group of Charr genus *Salvelinus* in the Northeast Asia and North America.
22. **Gorobeyko U.V., Kartavtseva I.V., Guskov V.Yu., Sheremetyeva I.N., Kazakov D.V.** The preliminary data about intraspecific genetic variability in Far Eastern Myotis petax based on mtDNA nucleotide sequence data.
23. **Guskov V.Yu.** Population diversity of Asian black bear at his north range.
24. **Shchepin O.N., Novozhilov Y.K., Prikhodko I.S., Schnittler M.** Large-scale DNA metabarcoding study provides new insights on myxomycete diversity and ecology
25. **Petrov N.B., Vladychenskaya I.P.** Intra- and interspecies genetic COI distances within freshwater mussel's genus *Unio* from Volga basin
26. **Minasbekyan L.A., Aydarkhanova G.S., Avagyan I.A.** Promising biomarker for assessing the effects of the environment on a population of plants.
27. **Semenchenko A.A., Seliverstov N.A., Rumyanceva A.Yu.** Molecular phylogeny for the non-biting midges of the diamesinae and prodiamesinae subfamilies (DIPTERA: CHIRONOMIDAE) of the Palearctic.

Mini Workshop/School

15-20-16-20**: Chair Person – S.D. Yuri Ph. Kartavtsev**

1. **Yuri Ph. Kartavtsev, Sergei V. Turanov and others on request.** News in MAPEEG, Barcoding World and close fields: Update on RUS-BOL Recent Activity, Metabarcoding, etc. (50').

#. General Discussion on Reports (10').

16-20-17-20**: Chair Person – Prof. Yuri Kartavtsev**

Yuri. Ph. Kartavtsev. Concluding remarks & VFDG meeting (60').

19-00-20-00**: Dinner**

FRIDAY, SEPTEMBER 13

8-30 - 9-00**: Breakfast**
9-00 - 19-00**: Free days, Excursions to Vostok MBS & Vladivostok City.**
19-00-23-00**: Dinner, Closing Reception & Evening party by the fire at Vostok MBS.**

SATURDAY, SEPTEMBER 14

8-30-9-00: **Breakfast**
16-00-16-15: **Departure to Vladivostok and Airport.**

ABSTRACTS

COMPARATIVE POPULATION GENETIC ANALYSIS OF FAR EASTERN TREMATODE SPECIES OF THE FAMILY HAPLOPORIDAE NICOLL, 1914 – A PARASITES OF MULLETS

Atopkin D.M.

Department of parasitology, Federal Scientific Center of the East Asia Terrestrial Biodiversity, Far Eastern Branch of the Russian Academy of Sciences, Vladivostok, 690022, Russia

The topic of phylogenetic interrelationships of trematodes of the family Haploporidae Nicoll, 1914 is an actively discussed problem at the present time. Recent studies using complex morphological and molecular approaches have resulted in the identification of several new species, genera and subfamilies within the family, along with leading to some taxonomical rearrangements. Most studies of haploporid trematodes have focused on high taxa, thus omitting the micro evolutionary processes which are an important factor during speciation. Population genetic studies are useful for evaluating these processes through the analysis of intraspecific polymorphism and differentiation using molecular data.

Intraspecific variation of *Parasaccocoelium mugili , Skrjabinolecithum spasskii* and *Carassotrema koreanum*, collected from mullet fish and cyprinid fish of the south of Russian Far East and Vietnam has previously been estimated on the basis of two molecular markers: ribosomal ITS1 and mitochondrial cytochrome oxidase I (COI) gene sequences. In the present study, molecular identification of these species from the Kievka River, Primorye and from Vietnam was performed by analysis of 28S rDNA sequences. Analysis of ITS1 rDNA sequences variation revealed two highly differentiated main groups for *P. mugili*, representing trematode specimens from the two regions. Genetic variation within each region was relatively low. Mitochondrial COI gene sequence data analysis revealed fixed nucleotide and amino acid substitutions, and supported the existence of two genetically different groups associated with geographical origin. Analysis of the COI gene fragments showed extremely high variation within Russian and Vietnamese *P. mugili* samples. Three main divergent groups were detected by ITS rDNA within S. spasskii: two groups (I and III) for Primorye and one (II) for Vietnam. Groups II and III were closely related to each other that to the group I. Frequency of both groups and its genetic variation in Primorye was almost identical to each other. Vietnamese group possess highest genetic diversity.

Carassotrema koreanum from Primorye and China were different by several sites of 28S rDNA, containing fixed substitutions. Ribosomal ITA and mitochondrial COI gene of this species within Primorye were almost identical.

Our results most probably represent a case of initial step of allopotric speciation for *P. mugili*, caused by living strategy of its definitive host at evolutionary scale. Mitochondrial DNA sequence data show that existence of gene flow between local populations of P. mugili in the Primorye Region caused by definitive hosts can be proposed. For *S. spasskii*, speciation process is more deeply in comparison with *P. mugili*. Second contacts of mullet fish in Primorye doesn't excluded.

This study was supported by Russian Science Foundation, project № 17-74-20074.

CHARACTERIZATION OF POLYMORPHIC DINUCLEOTIDE MICROSATELLITE LOCI IN WHITE-EDGED ROCKFISH, *SEBASTES TACZANOWSKII*

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Viviparous teleosts are trophically adapted to various modes of fetal-maternal relationships and have reproductive mechanisms, which are lacking in oviparous teleosts, including internal fertilization and embryogenesis in the ovary (Takemura et al., 1991). The white-edged rockfish, *Sebastes taczanowskii,* is a marine viviparous teleost inhabiting the Sea of Japan from the coasts of Korea to Sakhalin; Aniva and Terpeniya bays in the Sea of Okhotsk; waters of the northern coast of Hokkaido, as well as Pacific waters of Japan and the southern Kuril Islands (Kolpakov, 2006). In this study we select and characterize 19 polymorphic microsatellites that may be useful for parent-offspring comparisons in parentage studies and other population genetics assessments. Previously 19 di- microsatellite loci have been developed for other species and in the current research were tested for *S. taczanowskii*. Totally 10 microsatellite markers were successfully amplified and allele frequencies for 5 loci were determined with the sample of 48 individuals that collected from natural population. Polymorphism at these loci revealed from 9 to 16 alleles. The observed heterozygosity ranged from 0.64 to 1.00, and the expected heterozygosity ranged from 0.71 to 0.88. Null alleles were detected at one microsatellite locus. There was no evidence of scoring error, large allele dropout, or evidence of linkage disequilibrium at any locus. Two loci were significantly deviated from HWE ($P < 0.01$). The described polymorphic loci for *S. taczanowskii* could be suitable for further genetic analysis, including taxonomic inconsistencies, parentage/relatedness analysis and population genetics assessments.

CHARACTERIZATION OF MICROSATELLITE MARKERS FOR A NEW SPECIES OF FAR EASTERN REDFINS OF THE GENUS *TRIBOLODON* (PISCES, CYPRINIDAE) AND CROSS-SPECIES AMPLIFICATION IN THE RELATED SPECIES

Batishcheva N.M.[1], PolyakovaN.E[1], BrykovV.A.[1,2]
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Previously we have showed that *Tribolodon* sp. (southern form of *T. hakonensis*) has a mosaic of nuclear genes received from the Pacific redfin *T. brandtii* and big scaled redfin *T. hakonensis*. It was suggested that the new species could have formed as a result of homoploid hybridization between the true *T. hakonensis* (by original description) and *T. brandtii*, which probably made it possible for this species to occupy a new ecological niche (Polyakova et al., 2015). In the present study 21 polymorphic microsatellite loci were evaluated for PCR amplification (Koizumi et al., 2011) of genetic variability in samples of *T. hakonensis, T.* sp. (southern *T. hakonensis*) and *T. brandtii* to clarify the taxonomic status of *T.* sp. Among the 21 tested markers, 6 microsatellite loci were further used for genotyping in 5 wild populations. The numbers of alleles varied from 10 to 38. The observed and expected heterozygosities ranged from 0.037 to 0.884 and from 0.027 to 0.805, respectively. Null alleles were detected in two loci (*Thp6* and *Thp7*). These loci were successfully cross-amplified in three *Tribolodon* species. These microsatellite loci will be useful for clarifying the status of the southern form of *T. hakonensis*, for investigating the intra- and inter-specific genetic variation and population structure of this species.

GENETIC AND PHENOTYPIC DIFFERENTIATION OF *CALLUNA VULGARIS* (L.) HULL IN PRITOBOLIE AND EUROPE

Cherepanova O.E.*, Petrova I.V., Sannikov S.N., Sannikova N.S.
Botanical Garden of the Ural Branch of the Russian Academy of Sciences
620144, Ekaterinburg, March 8, 202 a, Russia

The investigation of intraspecific genetic and phenotypic divergence in natural plant populations from the marginal island and central part of the species range, along with the study of the role of this divergence in microevolution, is one of the key problems of general biology. The hypothesis of E. Mayr on homozygotization and gene pool depletion in small isolated populations has been confirmed only in laboratory experiments with the *Drosophola species*.

A group of marginal eastern isolated populations of heather (*Calluna vulgaris* (L.) Hull.) growing in Pritobolie, in the southwest of Western Siberia, represents an interesting object for ecogenogeographic analysis of the role of longterm isolation in the microevolutionary divergence of plants.

Geographic variation and differentiation of the chloroplast DNA haplotypes and morphological and anatomical leaf parameters were assessed in a number of eastern European groups of *C. vulgaris* populations and in the Pritobolien group of populations of this species in Western Siberia, which have been long isolated from the European populations. Sharply pronounced genetic and phenotypic distances and their gradients between the Pritobolien and European population groups were revealed. These distances were many times higher than those between the relatively homogeneous eastern European groups.

Data obtained generally supported the hypothesis on the phenogenetic divergence of the Pritobolien marginal populations of *C. vulgaris* from the European, probably at the subspecies level.

This work was supported by the Russian Foundation for Basic Research (grant no. 18-34-00506).

HENRICIA SEASTARS OF VOSTOK BAY

Chichvarkhin A., Chichvarkhina O.

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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. To date, the fauna of echinasterids of Russian Pacific seas totals about 30 nominal species.

The name for previously defined *pertusa*-group of the genus *Henricia* was designated as *Setihenricia* Chichvarkhin & Chichvarkhina, 2017. We are considering it as a subgenus, while *perforata*-group is considered as the subgenus *Henricia.*

We collected seven species in Vostok Bay. Partial sequence (598-609 b.p.) for *16S* rRNA mitochondrial gene was sequenced in 47 specimens. All of them were well delimited using neibour-joining and ABGD approaches with barcoding gap of 0.05 with *p*-distance. The two species, *H. asiatica* and *H. oculata* were included in the list basing on video footage and previous records.

Investigation on Vostok Bay revealed two new species:
H. alexeyi Chichvarkhin & Chichvarkhina, 2017, and *H. djakonovi* Chichvarkhin, 2017.

The following species are confirmed for Vostok Bay and its vicinity:

1. *Henricia alexeyi* Chichvarkhin & Chichvarkhina, 2017
2. *Henricia asiatica* Djakonov, 1958
3. *Henricia densispina* (Sladen, 1878)
4. *Henricia djakonovi* Chichvarkhin, 2017
5. *Henricia granulifera* Djakonov, 1958
6. *Henricia hayashii* Djakonov, 1961
7. *Henricia oculata* (Pennant, 1777)
8. *Henricia sp.*
9. *Henricia pacifica* Hayashi, 1940

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PHYLOGENETIC POSITION OF BRACHIOPOD BASED ON ANALYSIS OF CYTOLOGICAL AND MOLECULAR GENETIC ATTRIBUTES

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The phylum Brachiopoda is a relict group of marine invertebrates known from lower Cambrian Deposits. Their position in the animal system remains controversial, with morphological data often being inconsistent with the results of molecular phylogenetic analysis. According to classical concepts, brachiopods were divided into two classes: Articulata (Testicardines) with a hinge between the dorsal and ventral valves and Inarticulata (Ecardines), which have no hinge. We studied the structure of the sperm and the nucleotide sequences of the *18S* and *28S* rRNA genes in the articulate brachiopods *Coptothyris adamsi* and analyzed on this basis the macro-phylogeny and classification of modern brachiopods.

Phylogenetic trees of the combined sequences of *18S* and *28S* rRNA genes were built by Bayesian analysis using MrBayes version 3.2, as well as by the method of maximum likelihood using RAxML v. 7.4.2 with a raxmlGUI v. 1.3 graphical interface.

The spermatozoa of *C. adamsi* are similar to those of other articulate brachiopods. Two types of sperm structure can be distinguished within Brachiopoda: the first type is characteristic of the articulate brachiopods (Rhynchonelliformea), the second type, the inarticulate brachiopods (Linguliformea and Craniiformea). Rhynchonelliformea spermatozoa are similar to those of the deuterostome animals, in particular to the sperm of the Echinodermata, whereas Linguliformea and Craniiformea spermatozoa are similar to the typical sperm of the Trochozoa, such as annelids and mollusks. The two types of sperm structure suggest great differences between the inarticulate and articulate brachiopods. Perhaps this is the result of an ancient origin of brachiopods, whose ancestors were probably close to the group of animals that are ancestral to protostome and deuterostome animals.

Our tree of the combined sequences of the *18S* and *28S* rRNA genes, as well as the trees that are based on a great number of genes from the representative taxonomic sample show that brachiopods and phoronids form monophyletic groups that join together in grouping Brachiozoa, with phoronids occupying the basal position and brachiopods dividing into two groups corresponding to the articulate (Rhynchonelliformea) and inarticulate (Linguliformea and Craniiformea) species.

Thus our results of the molecular phylogenetic analysis and the structure of the spermatozoa support the division of Brachiopoda into two groups, viz., Inarticulata (with the classes Linguliformea and Craniiformea) and Articulata (with the class Rhynchonelliformea); this corresponds to the traditional notion on the systematic position of brachiopods.

PHYLOGEOGRAPHY OF *AEGILOPS TAUSCHII* AND HOW DOES ECOLOGY FORM THE SHAPE OF PHYLOGENETIC TREE

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Aegilops tauschii Coss. is a wild diploid mostly self-pollinating goat-grass (2n = 14, genome DD), a relative of common wheat, *T. aestivum* (2n = 42, genome AABBDD). In nature *Ae. tauschii* is presented by many local populations of subspecies (subsp.) *strangulata*, mostly occupying pre-Caspian area from Caucasia to Central Copet-Dag; and subsp. *tauschii*, occupying mostly continental area from Caucasia to Western Tian Shan and Himalayas.

Phylogeography of *Ae. tauschii* was investigated using DNA sequences of enzyme-encoding gene *Got2* (about 3,000 b.p.). Totally 114 *Ae. tauschii* specimens were involved in the study. It was found that on phylogenetic tree based on *Got2* DNA sequences subsp. *strangulata* presents five major lineages which share the subspecies area between each other, plus relict lineages. In contrast, subsp. *tauschii* presents the only one major lineage occupying all the subspecies area, plus some relict lineages. Phylogeographic patterns obtained through the study indicated that in the course of *Ae. tauschii* evolutionary history there were several "waves" of geographic expansion; and in the case of subsp. *tauschii* several times the "younger" and better adapted lineage almost completely forced out the previous "older" lineage from its area. In subsp. *strangulata* some of the major lineages have disjunctive areas as a result of competition between them, but anyway they "managed" to co-exist rather well.

Intraspecies ecological differentiation of *Ae. tauschii* was studied using WORLDCLIM database of computer system ArcGIS. For 307 *Ae. tauschii* local habitats, representing all the species area, 49 climatic parameters were pointed out (i.e. precipitation, minimum, mean and maximum temperatures for each month, and also the total annual level of precipitation). Principal components analysis of the data obtained revealed that despite the area of subsp. *strangulata* is much smaller than the area of subsp. *tauschii*; subspecies *strangulata* is relatively much more variable ecologically. It seems that rather wide ecological niche of subsp., *strangulata* gives an opportunity for several lineages of the subspecies to co-exist rather successfully, while in the case of relatively narrow ecological niche of subsp. *tauschii* the lineages other than the single major lineage had a chance to survive only as relicts.

USING MOLECULAR GENETIC TECHNIQUES FOR IDENTIFICATION OF SKELETONEMA(BACILLARIOPHYTA) SPECIES OCCURRING IN THE SEA OF JAPAN

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Species of the genus *Skeletonema* are among the most common marine planktonic diatoms of the temperate zone of the World Ocean. Despite the worldwide distribution of the genus, there are only fragmentary data on its ecology and quantitative characteristics of habitats, as well as limited information on the species diversity in the northeastern Pacific Ocean. Molecular genetic approaches have proven to be an auxiliary method for the identification of *Skeletonema* species, the investigation of population-genetic patterns that underlie the modes of reproduction and origin of variety of ecological forms, as well as in the study of phylogeography of the genus in general. For the first part of the present study, 13 strains of *Skeletonema* were obtained from planktonic and benthic samples of microalgae collected during the period from 2009 to 2017 from the northwestern Sea of Japan. We identified *Skeletonema* species based on light and electron microscopic observations and molecular genetic analysis of *28S* rDNA sequences. A detailed description, as well as new data on water temperature, salinity and species distribution, is provided for each species and their morphological features are discussed. The presence of several rimoportula processes on the terminal valve of *Skeletonema* species is shown for the first time. Our results support the usage of the topological criterion for the delimitation of the *S. dohrnii–marinoi*, as opposed to the distance-based criterion when dealing with *28S* rDNA fragments.

At the same time, there still no consensus view on the boundaries of *S. dohrnii-marinoi* species. An exceptional morphological plasticity as well as explicit methodological dependence of the delineation of genetically discrete groups makes most of researchers to consider these species as a complex. The information about reproductive isolation could be useful for such cases to indicate the presence of species boundaries. Hence, one of the indirect methods of rationalization of reproductive incompatibility among lineages (well-studied on green algae) is identifying compensatory substitutions in the second internal transcribed spacer (*ITS2*) of the ribosomal operon or rDNA. In the second part of the study, we used the complete sequences of ribosomal operon for three species of the genus *Skeletonema* (*S. dohrnii*, *S. marinoi* and *S. japonicum*), obtained from GenBank. We have conducted a prediction of the secondary structure and obtained information about the compensatory substitutions in *28S* rRNA (domains D1-D2) and *ITS2*, followed by transformation them into a phylogenetically informative character matrix. The consensus secondary structure of *28S* rRNA among investigated species of *Skeletonema* possesses one compensatory substitution which point at the species differences of *S. dohrnii* and *S. marinoi*, and, moreover, the secondary structure of *ITS2* does not form fours hairpin, where we have found indels. Also, *ITS2* comprises one compensatory substitution indicating potential reproductive isolation whose boundaries do not coincide with the taxonomic reference, resulting in paraphyly of *S. dohrnii*.

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THE FIRST COMPLETE MITOCHONDRIAL GENOME OF A NEW *LYCODES* REINHARDT, 1831 (PERCIFORMES: ZOARCIDAE) SPECIES

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The five specimens of *Lycodes* sp. were collected from the Gulf of Patience (southern part in the Sea of Okhotsk). All fishes were assigned to a new species based on morphology (here *Lycodes* sp.1 – sp.5). The complete mitogenome of the 5 individuals/fishes was determined by polymerase chain reaction (PCR) amplification and primer-walking sequencing. The complete mtDNA of *Lycodes* sp.1 – sp.5 (16.534 - 16.604 kbp) consisted of 13 protein-coding genes, 22 tRNAs, two rRNA genes, and a control region. We observed tRNA genes and heteroplasmy. Interesting cases of short overlap of 1 bp between two tRNA genes ($tRNA^{Thr}$ and $tRNA^{Pro}$) were encountered in one of the copies mtDNA of *Lycodes* sp. 4 and *Lycodes* sp. 5. Besides, the biggest intergenic spacers (69 bp and 36 bp) located between genes $tRNA^{Thr}$ and $tRNA^{Pro}$ were observed in one of the copies mtDNA of *Lycodes* sp. 2 and *Lycodes* sp. 4. Only one of the fishes (*Lycodes* sp. 1) had a homoplasmic mtDNA. Heteroplasmy was observed in intergenic spacer between the $tRNA^{Thr}$ and $tRNA^{Pro}$ genes, located close to the control region. The ratio of one variant to other variant of intra-individual copies of the mtDNA was approximately 2:1 for *Lycodes* sp. 2, but 1:1 for *Lycodes* sp. 3 and *Lycodes* sp. 4.

THE TAXONOMIC STATUS OF THE SEMISULCOSPIRIDAE MOLLUSKS IN THE RUSSIAN FAR EAST

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The mollusks of the Semisulcospiridae family are the first intermediate hosts of various trematoda species including *Nanophyetus*, *Metagonimus* and *Paragonimus*, whose definitive host is a human. The object of this study is the mollusks of the *Parajuga* genus. Their systematic position is not clear and molecular data are practically absent. These mollusks are usually identified by conchological features, but shell sculpture can be corroded. At the same time, mollusks of different species often inhabit common water reservoirs; therefore, it is difficult to determine the species by morphological characteristics. Formerly, the *Parajuga* genus was attributed to the *Juga* genus and the Pleurochidae family. To date, the mollusks of this genus belong to the family Semisulcospiridae. It should be stressed, that despite importance of this genus as the hosts of epidemiologically significant parasites, for the last ten years there is almost no molecular, anatomical and morphological data for these mollusks.

The following species of the *Parajuga* genus were analyzed in this study: *P. amurensis*, *P. nodosa*, *P. heukelomiana* and three samples of an unspecified species (*Parajuga* sp.). Identification of the species was based on the shell structures. The nucleotide sequences of the 28S rRNA gene of nDNA and the *cox*1 mtDNA gene were used as the markers in genetic analysis.

Based on the 28S rRNA gene, 1% difference was found between *Semisulcospira* and *Parajuga*, whereas higher value of distances was between *Juga* and *Semisulcospira* (3%) and *Juga* and *Parajuga* (2.4%). Most probably, *Semisulcospira* and *Parajuga* belong to the same genus. But to clarify their status, it is necessary to include other members of the family in the analysis. Specimens within *Parajuga* have no differences with the exception of the unique sequence of *P. nodosa*, for which a nucleotide substitution was found at 115 bp position.

The analysis of the *cox*1 gene revealed that the sequences of *Koreoleptoxis amurensis*, *P. nodosa* and *Parajuga* sp. are nearly 100% identical. Therefore, all specimens belong to the same species. At the same time, *P. heukelomiana* and *P. amurensis* differ by 2% from *P. nodosa*, *Parajuga* sp. and *K. amurensis*. According to the analysis of amino acid sequences, all the detected substitutions were synonymous. Obtained variability is apparently intraspecific. Thus, all studied specimens of the *Parajuga* genus are one species, *Koreoleptoxis amurensis*.

GENETIC INVESTIGATION OF SABLE (*Martes zibellina*) OF MIDDLE PRIAMURYE REGION

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The purpose of the work was to analyze population structure of sable in the Middle Priamurye Region. Based on the microsatellite allelic diversity we investigated geographic variability for sable community in the region. The annual change in allelic composition was investigated as well.

Our study of geographical variability was based on biological material of five geographical areas of the Middle Priamurye. Samples of following areas were explored: 1) ”Magan’- northwestern slope of the Bureinsky Ridge, 2) “Sutyr” and 3) “Tyrma” in western macroslope of the Bureinsky Ridge in its middle part, 4) “Kamenuschka” in northern spur of the Lesser Khingan and 5) “Manoma” in western macroslope of Middle the Sikhote-Alin Ridge. Totally 148 specimens were collected in 2012-2013 hunting season. Genotyping on 3 microsatellite markers (Ma-2, Ma-3, Mer-041) was made. Twenty three alleles per locus ranging from 4 (Ma-3) to 12 (Mer-041) were found. Most of the alleles were found in all geographical samples. As usual their frequencies were variable when moving from one sample to another ones. The largest amount of alleles were represented in the Sikhote-Alin sample “Manoma’ where four unique alleles were found. Additional unique allele was found in the sample of north-western slope of the Bureinsky Ridge (“Magan”).

The Hardy-Weinberg equilibrium (HWE) test for each locus revealed that among 5 geographical samples, only one of them deviated from the HWE at Ma-2 locus and another one deviated from the HWE at Ma-3 locus. For other geographical samples and for all 148 specimens across five geographical areas, all loci followed the HWE expectations. Pairwise comparison of 5 geographical samples reflected a low level of their subdivision ($0{,}0004 \leq Fst \leq 0{,}0127$).

The change of allele composition from year to year was traced on biological material of four localities. These localities were 1) “Magan”, 2) “Sutyr”, 4) “Kamenuschka”, and 5) “Manoma”. The material was collected in 2011-2018 years. Two to four annual samples were collected in each of the geographical areas under consideration. About 24 specimens for geographical sample of each year were analyzed (total sample was 276 specimens). Genotyping on Ma-3 locus has shown that differences in time are insignificant, but sometimes exceed those between geographical samples of the region.

Data of pairwise comparison Fst and results of HWE equilibrium test of loci investigated suggest the existence in the Middle Priamurye region of a single sable populations with a fairly high migration exchange among its geographical subpopulations. The result of the study of year to year changes in the allelic composition on Ma-3 locus are consistent with this extraction. However allelic diversity indicates a certain uniqueness of the Sikhote–Alin subpopulation. Increasing the number of microsatellite loci and attracting other characteristics will allow us to understand this phenomenon.

RECOMBINATION-INDEPENDENT RECOGNITION OF DNA HOMOLOGY FOR MEIOTIC SILENCING IN NEUROSPORA CRASSA

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Homologous chromosome pairing represents a critical aspect of meiosis in nearly all sexually reproducing species. While meiotic pairing relies on the formation of double-strand DNA breaks in some organisms, in many others it can proceed in the apparent absence of DNA breakage and recombination. The mechanistic nature of such recombination-independent pairing represents a fundamental question in molecular biology. Using "meiotic silencing by unpaired DNA" (MSUD) in the fungus *Neurospora crassa* as a model system, we demonstrate the existence of a principally new solution to the problem of inter-chromosomal homology recognition during meiosis. Here we take advantage of the unique ability of MSUD to efficiently detect and silence (by RNA interference) any relatively short DNA fragment lacking a homologous allelic partner. We show that MSUD does not require the function of eukaryotic RecA proteins and the type II topoisomerase-like protein Spo11. We further show that MSUD recognizes weak interspersed homology in which units of sequence identity as short as 3 base-pairs (bp) are spaced apart with a periodicity of 11 bp, approximating double-helical DNA pitch and corresponding to an overall sequence identity of only 27%. Taken together, these results reveal the role of a recombination-independent homology-directed process in guiding the expression of small interfering RNAs and suggest that meiotic chromosomes can be evaluated for sequence homology at base-pair resolution by a mechanism that operates on intact DNA molecules.

VARYABILITY VARIANTS OF UPPER MOLARS OF THE KOREAN FIELD MOUSE

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The Korean field mouse *Apodemus peninsulae* Thomas, 1906 inhabits deciduous forests in the Asian Palaearctic. Nine subspecies can be distinguished based on differences in dimensional characteristics of the body and skull; however, that differentiation is ambiguous. It is known that the structure and morphology of teeth are genetically inherited traits (Jernvall et al., 2000). Morphology of occlusal surfaces of molars is sometimes used as a diagnostic feature in many species of *Apodemus* s. str. That feature is frequently used in paleontological studies.

A study of the fossil and modern material of island populations in Japan: *A. speciosus* Temminck, 1894 and *A. argenteus* Temminck, 1894, revealed variability in occlusal surface pattern of the upper molars; however, such variability was not described in *A. peninsulae* (Kawamura, 1989).

For the first time the structure of occlusal surface of 112 upper molars (M 1-3) was investigated from mice *A. peninsulae* caught during 7-11 August 2018 in the Ussuriysky Reserve. Classification of occlusal surface structures is based on Jacobs (1978). Different variants can be distinguioshed based on the fusion pattern of the tubercles (from t1 to t8) as well as on the appearance of additional tubercles, and the enteroconule.

Sixteen variants (4 of them were known earlier in *A. speciosus* and *A. argenteus* and 2 in *Sylvaemus* = *Apodemus witherbyi* Thomas, 1902) were described for M1, two for M2 and 11 for M3 (Kawamura, 1989, HosseinPour, 2009). Therefore, we have found a total of 23 new variants of M 1-3 in *A. peninsulae*.

As a result, more variants of the *A. peninsulae* occlusal teeth surface than in other *Apodemus* species s. lato were revealed, which may indicate higher genetic diversity in this species. Research of the upper molar variants may be useful in population studies of this species.

INTRASPECIFIC GENETIC VARIABILITY IN THE FAR EASTERN BAT *MYOTIS PETAX* BASED ON MTDNA NUCLEOTIDE SEQUENCE DATA

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Myotis petax Hollister, 1912 is a common widespread, but poorly studied East Asian bat species. It was recognized not so long ago as a separate species from *Myotis daubentonii* Kuhl, 1817.

Molecular genetic study allows to reveal the cryptic genetic variability in the Far Eastern populations of *Myotis petax* and to determine intraspecific structure, which is very important for estimating the state of the species natural population. In this study, the sequences of two mtDNA genes, cytochrome c oxidase subunit I (COI) and control region of d-loop (CR), were used to analyze intraspecific variability in *Myotis petax.*

COI sequences were obtained from 20 specimens of *M. petax* and CR sequences from 26 specimens. All bats were collected in the caves Spasskaya and Primorsky Velican (Primorsky Krai), in the vicinity of Komsomolsk-on-Amur (Khabarovsk Krai), in the vicinity of Zeya (Amur Oblast) and from the cave Dolganskaya Yama (Republic of Buryatia). The sequences from the Genbank were also used in the analysis.

All specimens of *M. petax* studied had only slight differences in COI sequences. On the other hand, they had significant differences in CR, including variations in nucleotide length due to different number of R1-repeats.

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POPULATION DIVERSITY OF ASIAN BLACK BEAR AT HIS NORTH RANGE

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Decline of biodiversity is one of the problems that accompany the development of mankind and scientific and technical progress. In turn, humanity has become a "catalyst" for the process of extinction of species or phylogenetic lines, increasing the speed of these processes hundreds of times. Such changes are perfectly noticeable in wide-adrenal species, which have vast areas of distribution and a large number of genetic variants. One such species is *Ursus thibetanus*, Cuvier 1823, known as the Asiatic black bear or moon bear. At the moment, genetic diversity is an important characteristic in the study of a given animal, especially species under threat of extinction, since diversity is one of the important parameters of the evolutionary development and preservation of a viable population.

In the course of this work, we considered the diversity and genetic structure of Asian black bear population from the north-east range. The original material of the Asian black bear from Russian Far East was taken as a basis for the work. Additionally, homologous sequences stored in the GenBank/NCBI database were used. As markers we used cytochrome *b* gene and control region sequences of mtDNA. A total of 250 sequences were used to analyze the control region and 87 for cytochrome *b*. The parameters of genetic diversity were calculated and phylogenetic reconstructions were constructed using the NJ and MP methods. Trees and networks for both mtDNA sites had a similar topology.

As a result of the work, we studied the samples from the south of the Far East of Russia and placed it in a general phylogenetic picture. Genetic differentiation from the population of the Japanese islands was very significant. Samples from the territory of the Korean Peninsula were included in the same clade with our animals and differed by only one nucleodite substitution. Chinese samples from Sichuan province were also included in the common clade with the Korean bears. The genetic diversity within the population of the south of the Far East was rather low. Animals from Southeast Asia, as expected, had their own clade.

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VARIABILITY OF ITS1 RDNA REGION IN THE NOTOCOTYLIDAE (TREMATODA) SPECIES

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Species of two trematoda genera, *Notocotylus* and *Catatropis*, are cosmopolitan and parasitize mainly in birds, less often in mammals (rodents). These genera are recognized by the following morphological features: three rows of papillae ar present in *Notocotylus* and median ridge and two lateral rows of papillae are present in *Catatropis*.

Phylogenetic tree previously constructed based on the 28S rRNA gene sequences did not support subdivision of these genera based on the above mentioned morphological features (Gonchar et al., 2019; Izrailskaia et al., 2019). One group included representatives of the genus *Notocotylus*; another group included representatives of the genus *Catatropis*, and the third group included representatives of both genera, as well as the newly described species, *Pseudocatatropis dvoryadkini* (Izrailskaia et al., 2019).

Further studies of Notocotylidae family using the ITS rDNA region involving one more species revealed significant differences between nucleotide sequences of *Notocotylus*, *Catatropis* and *Pseudocatatropis*. Differences in sequence length, presence of insertions and number of substitutions were observed.

PHYLOGENETIC RELATIONSHIPS AMONG PSILOSTOMATIDAE (TREMATODA) SPECIES BASED ON GENETIC DATA

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Trematodes parasitize practically in all classes of animals. The Psilostomatidae family is a cosmopolitan group of parasitic flatworms that can cause diseases in mammals and birds. Phylogenetic relationships within this family are not well investigated, and there are few available molecular data for this group.

In this study, we have analyzed phylogenetic relationships among species within the Psilostomatidae family based on nucleotide sequences for the 28S ribosomal RNA gene. Three species from Primorye, *Sphaeridiotrema monorchis*, *Sphaeridiotrema newmilleardamensis* and *Psilotrema acutirostris*, as well as GenBank data were used.

On the tree, two separate clusters can be distinguished for representatives of the *Sphaeridiotrema* and *Psilotrema* genera. It was found that, in the first cluster, *S. monorchis* from Primorye (Russia) differs from *S. monorchis* from Vietnam, despite the fact that both genus representatives were assigned to the same species using morphological features. However, based on genetic analysis, they belong to different species, which appeared as a result of geographical isolation. The genetic distance between these species is comparable to the interspecific distances between *S. monorchis* and *S. newmilleardamensis*. Both *S. monorchis* from Vietnam and Russia, as well as *S. newmilleardamensis* form a separate group within a cluster of the *Sphaeridiotrema* genus.

When analyzing the second cluster, it was found that *P. acutirostris* from the Arsenyevka River (Primorye, Russia) and *Psilotrema simillimum* do not differ from each other. Based on previously published data (Atopkin, 2005), there is only one nucleotide substitution between these species. It can be assumed that this is the same species. In contrast, there is an interspecific distance between *P. acutirostris/P. simillimum* and *P. acutirostris* from the Ilistaya River (Primorye, Russia). All these species form a separate group within a cluster of the *Psilotrema* genus.

Therefore, to obtain more reliable results and clarify the species composition and phylogenetic relationships in both genera, it is necessary to include additional morphological and molecular data.

PROMOTOR REGION IN SALMONIDAE GROWTH HORMONE GENE

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Salmonid fish are a unique group, originated through autotetraploidization with a subsequent divergence. Many genes in this taxonomic group were multiple, including growth hormone (GH) gene. In salmonid genome, the GH gene is represented by two unrelated paralogous genes, *gh1* and *gh2*. Both genes exist throughout the time of species divergence in this group. Generally, duplicated copies of genes accumulate changes at a higher frequency and may eventually lose or acquire new functions. However, high conservation of coding sequences (exons) of paralogous growth hormone genes in salmonid fish indicates that both genes are either functional or subfunctional. If a gene acquires new functions, its regulatory regions may also change. Comparison of promotor region of paralogous growth hormone genes *gh1* and *gh2* in salmonids suggested that both *gh1* and *gh2* promotor include the same set of regulatory sequences (TATA box, CRE, GRE and RAR/RXR elements) and binding sites for the pituitary transcription factor (Pit-1 (F1–F4)). However, promoter of *gh2* gene has a greater degree of polymorphism compared with *gh1* gene promoter in all investigated species of salmonids. The observed differences in the rates of accumulation of changes in growth hormone encoding paralogs could be explained by differences in the intensity of selection.

MOLECULAR MARKERS: FROM SEA FOOD MISLABELING DETECTION AND BIODIVERSITY ASSESMENT TO MODERN GENERAL BIOLOGY PARADIGM VERIFICATION

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The evidences of possible impact of gene introgression on species evolution, evolutionary fate of taxa, including reticulations in phylogenetic trees, and consistency of the latest molecular genetic data with the main modern paradigm, Neo-Darwinism, are considered in many of works (Barton, Hewitt, 1985; Campton, 1987; Avise, 2000; Gerber, 2001; Arnold, 2009; Arnold, Fogarty, 2009). In this assignment, the author will focus on animals, although many ideas suit other phyla too. The main issues of the report are as follows: (1) What methods are most appropriate for the hybrid detection and estimation of genetic introgression or gene flow? (2) What facts, obtained on gene introgression by nDNA and mtDNA markers, are the evidence for? (3) Is there in the literature any data on correspondence of molecular diversity in lineages or in taxa with Biological Species Concept (BSC). (4) How frequently are reticulations in gene trees observed, and what is a major signal from their topology?

(1) A combination of nDNA and mtDNA markers best suits the hybrid identification and estimates of genetic introgression or gene flow. (2) The available facts for both nDNA and mtDNA diversity seemingly make the introgression among many taxa of animals and plants obvious, although even in wide hybrid zones of *Mytilus* ex. group *edulis*, for example, introgression may be quite restricted or asymmetric, thus holding at least the "source" taxon (taxa) intact. (3) If we accept that sexually reproducing species in marine and terrestrial realms are introgressed, as it is still evident for many cases, then we should recognize that the orthodoxal BSC, in terms of complete lack of gene flow among species, is inadequate due to the fact that many zoological species are not biological species yet. However, sooner or later they definitely become biological species. This conclusion is supported by the genetic distance increasing with taxa rank and by the lowest diversity at intraspecies level as for single mtDNA genes, for complete mito-genome, and for nDNA data (Kartavtsev et al., 2016; Hedges et al., 2015). (4) The recent investigation of fish taxa divergence (Kartavtsev, 2017) using vast BOLD (www.boldsystem.org) database shows that gene trees for taxa up to the family level are basically monophyletic, and interspecies reticulation are rare for most of gene trees.

All the four above-listed outcomes have a great importance to the paradigms of General Biology, Evolutionary Genetics, and to iBOL (www.ibol.org) science policy, and to the practice of species identification in particular. Evidently, the most common successful delimiting of species based on barcoding technique is possible due to the prevailing species origin throughout geographic speciation mode that allows random accumulation of numerous mutations/substitutions after isolation of sister populations/taxa, which are detectable with molecular markers (barcodes). It seems that claims on the invalidity of the modern BSC paradigm (Arnold, Fogarty, 2009) due to the large-scale gene introgression and phylogeny reticulation are too premature. Contrary to that, the evidences available in the literature shows that molecular genetic data are concordant in general with the BSC and Neo-Darwinism.

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MOLECULAR PHYLOGENY OF MUSSELS (MOLLUSCA, MYTILIDAE) BASED ON THREE NUCLEAR GENES (28S RDNA, 18S RDNA AND H3)

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On the basis of nucleotide sequences of three nuclear genes and using molecular phylogenetic and evolutionary genetic approaches, the phylogeny of the main representatives of one of the largest taxa of bivalve mollusks, the family Mytilidae, was studied, and its system and taxonomy were refined. A phylogenetic system for the family Mytilidae and closely relative taxa of the order Mytilida, which currently has no consensus among specialists on the basis of traditional characters, is presented. Using nucleotide sequences of the *28S* rDNA, *18S* rDNA, and histone *H3* genes, this consensus was established by the study of Mytilidae. Some concerns of mussel systematics were resolved; in particular, the monophyly of the family Mytilidae Rafinesque, 1815 was established with the strongest support for the subfamily Mytilinae Rafinesque, 1815. The data obtained disproved Distel's et al. (2000) conclusion on polyphyly of the family Mytilidae Rafinesque, 1815. Isolation of the taxa in the rank of the Modiolinae G. Termier & H. Termier, 1950 and Bathymodiolinae Kenk & Wilson, 1985 subfamilies in the family Mytilidae and also the family Septiferidae Scarlato et Starobogatov, 1979 was confirmed, although the rank of the later taxon is not universally recognized and it remains to be clarified in an additional study.

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MORPHOLOGY OF ADDITIONAL CHROMOSOMES IN *APODEMUS PENINSULAE* FROM THE UPPER PRIAMURIE REGION (BASED ON FISH ANALYSIS)

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Korean field mouse, *Apodemus peninsulae* (Thomas, 1906) is widely distributed throughout the Asian mixed forests. Karyotype of this species contains 48 acrocentric A chromosomes of the basic set and additional B chromosomes varying in number (0–30) and morphology (from metacentric to acrocentric). B chromosomes in mice from the Russian Far East were shown to exhibit low variability in DNA content while, the DNA composition of B chromosomes in species from Siberia and Transbaikalia were highly variable (Rubtsov et al., 2015). Amur region (Upper Priamurie) is located between Transbaikalia and the Far East of Russia. The DNA of B chromosomes of mice from this region are still not investigated.

We assumed that the DNA content of B chromosomes in animals from the Upper Priamurie are similar to the B chromosomes in mice from the Middle, Lower Priamurie and Prymorskii Krai.

Morphotypes of these B chromosomes were similar the B chromosomes described in the Far Eastern, Transbaikalian and Siberian populations (Roslik , Kartavtseva, 2019 see this conference).

We compared the DNA content of B chromosomes in mice from the Upper Priamurie region with the B chromosomes in mice from the other regions. The analysis of additional chromosomes in *A. peninsulae* inform the Upper Priamurie using FISH revealed several variants of chromosomal regions enriched by new DNA repeats, as well as variants of chromosomal regions enriched by repetitive DNA typical for the Far Eastern, Transbaikalian and Siberian populations.

Therefore, we can assume that the Upper Priamurie, as well as Transbaykalia are the areas, where mice from Siberia and the Far East intermingle. Earlier, we suggested that only Transbaikalia is a territory, where mice with both types of B chromosomes occur (Kartavtseva, Roslik 2006).

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GENETIC APPROACH TO THE STUDY OF PHYLOGENY OF THE GONATID SQUID

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The gonatid squid (Gonatidae, Oegopsida, Teuthida) are widely distributed in cold and temperate waters in the Northern and Southern hemispheres; however, most species are known from the boreal North Pacific, where this family presumably originated and diverged. The gonatid squid are represented by different morphological and ecological forms, including benthopelagic deep-sea species, as well as meso- and epipelagic species, some of which are known to conduct diel vertical migrations. These squid also exhibit strikingly different reproductive strategies, one of which involves egg-brooding. Genetic approaches using allozymes and mitochondrial DNA were used to infer phylogenetic relationships among gonatid species. Two major phylogenetic lineages were recovered by both approaches: one lineage includes species with seven rows in the radula and do not exhibit egg-brooding behavior, and another includes species with five rows in the radula and females of which are known to brood their eggs between the arms and floating in water column until the eggs are hatched.

MOLECULAR DATA FOR *ERSCHOVIORCHIS* REPRESENTATIVE, HIGHLY PATOGENIC TREMATODE OF FISH-EATING BIRDS

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This study presents the trematodes of a new species, *Erschoviorchis anuiensis* sp. nov. Adult worms are parasitic in the pancreas and liver of fish-eating birds. Infected organs are increased in size, also tissue destruction occurs and numerous capsules are formed where the parasites are located. Such changes indicate a high pathogenicity of this trematode, which presence is usually fatal to the hosts.

In the *Erschoviorchis* genus, the only one species was currently known, which has been described for the first time in North America (Linton, 1928 cited in Skrjabin, 1950). Later, this parasite was studied in more detail and transferred to the *Amphimerus* genus by Gower (1939). On the basis of morphological data, its taxonomic status and position in the trematode system as a new species of *Erschoviorchis lintoni* were identified by Skrjabin (1945) and confirmed by Scholz (2008). But genetic data for this species has not yet been obtained.

A representative of the *Erschoviorchis* genus was described in the Russian Far East (Oshmarin, 1963). It was assigned to *E. lintoni*. However, the morphological data on the detected worms were absent, and information on their morphology was limited to a brief description provided by Linton (1928). It can be assumed that the individuals found could have been mistakenly attributed to the species *E. lintoni*, since for the first time this species was described in North America. Between the continents (Asia and North America) there are restrictions of the bird migration (definitive hosts of the studied trematodes). Therefore, a new species, *Erschoviorchis anuiensis* sp. nov, can be established, at least, based on a geographic isolation. This hypothesis is also confirmed on the basis of the detected differences in morphological characters between species.

Despite the lack of genetic data for the *Erschoviorchis* genus, to establish the phylogenetic position of the species within the family Opisthorchiidae, we obtained molecular data for nuclear (*28S* rRNA, *ITS2*) and mitochondrial (*cox*1) DNA markers. Based on the phylogenetic tree topology, it is clear that a representative of the *Erschoviorchis* genus forms a sister group with species of the *Opisthorchis*, *Clonorchis* and *Metorchis* genera. At the same time, it is not *Amphimerus* representative, which confirms the fallacy of the Gower's theory (1939).

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TAXONOMIC VALIDATION OF SPECIES BELONGING TO GENEUS CLARIAS IN INDIA

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Catfishes (order Siluriformes) the most diversified group of fish comprising 3,093 species belong to 478 genera and 36 families. Of which, freshwater clariids are one of the catfish families within the order Siluriformes. The dilemma about fish representing to family clariidae is still not clear and varying numbers frequently appear in the literature that ranges species number between 89-113 belonging to 15-16 genera. Similarly, Genus Clarias (Scopoli 1777) represents "Old World" family of clariid catfish ranging 48-56 species and supposed to be the largest genus. Although, some species are reported in Syria, southern Turkey and throughout Southeast Asia, their diversity is the highest in Africa. Moreover, of the 16 genera, three are endemic to Asia, 12 endemic to Africa, and one, Clarias Scopoli, 1777, is present on both continents represented by about 56 species. Some authors are of opinion that there are 21 valid species of Clarias in Asia. Of the 21 Asian species, perhaps none is well studied than *Clarias batrachus* among others. In parts, Ng (2004) trying to establish the identity of *C. batrachus* for the description of a new *Clarias* and experienced issues with the identity and nomenclature of *C. batrachus*, demanding the designation of a neotype. Recently Ng & Kottelat (2008) suggested that the *C. batrachus* found in South-East Asia, Java and India are actually comprised of four subspecies as (i) *C. batrachus*, (ii) *C. aff. batrachus* ''Indochina'', (iii) *C. aff. batrachus* ''Sundaland'' and (iv) *Clarias magur*.

The dilemma of neotype designation, subspecies, synonyms and misidentification reported in the published literature is basically supported either based on morphological forms or historical descriptions by taxonomists and no genetic consideration is been given in most of the cases. Due in part, our study evaluated the taxonomic status of key Indian species of clariid catfish by using mitochondrial DNA sequence data from COI, partial cytochrome *b* gene and control region. Species differentiation based on rigorous taxonomical procedures supported by molecular markers is the basis of this study in evaluating taxonomic status of Clarias species in India. Our study banked with four putative new species and resolved the ambiguities between *C. batrachus* and *C. magur*.

GENUS *LOTUS* (LEGUMINOSAE): PHYLOGENY AND GENETIC STRUCTURE OF SPECIES

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Lotus includes ca. 130 species of herbs, semishrubs and shrubs native to the Old World. Previous nrITS-based phylogenies were incongruent with all taxonomic systems of the genus. Members of the former genus *Dorycnium* were placed near species of *L.* sect. *Lotus*. The aim of the present study was to conduct a global phylogenetic study of *Lotus* using both plastid and nuclear markers with special attention to relationships between *Lotus* and *Dorycnium*. Widely sampled genus *Lotus* (incl. *Dorycnium* and *Tetrago-nolobus*): 167 accessions ITS 1-2 nrDNA, 74 accessions 5'ETS nrDNA, 65 accessions plastid set (*psb*A-*trn*H, *rps*16 intron, *trn*L intron, *trn*L-F); outgroup including species of *Cytisopsis*, *Hammatolobium* and *Tripodion*. Eight nuclear SSR loci were studied in the *L. corniculatus* complex. Bayesian (MrBayes 3.2.2) and ML (MEGA X) phylogenetic analyses, S-DIVA analysis. Genetic structure of *L. corniculatus* complex (Structure, TCS and Arlequin programs).

I. Sequences variability (total length/variable sites/parsimony informative sites): ITS: 680/280/212; ETS: 589/254/212; plastid set: 2378/305/153. Among plastid markers, *trn*L-F region plays important role in tracking the *Lotus* evolution, especially due to the presence of long indels. *Lotus schimperi* has 50 bp insertion in *trn*L-F IGS, a group *L. creticus* - L. *pseudocreticus* has 91 bp deletion in *trn*L intron, and *Lotus corniculatus* complex is characterized by 73 bp repeats covering parts of *trn*F and *trn*L-F IGS.

II. Taxonomy and Biogeography: Southern lineage of *Lotus* includes the majority of sections and is supported by most markers. Northern lineage includes *L.* sections *Lotus*, *Dorycnium* and *Bonjeanea* (the last two were members of the former gen. *Dorycnium* sensu Lassen) and supported by plastid data only. Phylogenetic relationships within Northern lineage are the most controversial and its evolution is essentially homoplastic. Sections in traditional view are not supported by most markers, except for Bayesian analysis of ETS, which confirms *L.* sect. *Lotus* and gen. *Dorycnium* sensu Lassen. S-DIVA analysis of the ETS data set suggests that plants of Europe or N. Asia (area A) were ancestral for two Northern lineages and plants of Africa or S. Asia (area B) - for Southern lineage of *Lotus*. A group corresponding to gen. *Dorycnium* sensu Lassen may have ancestral forms in area A. Sect. *Bonjeanea* contains three unrelated species, which combine characters of *Lotus* and *Dorycnium*. *L.rectus* is a widely distributed Mediterranean species, and *L.strictus* occurs from East Mediterranean to West China. *L.hirsutus* demonstrates a tendency towards subdivision into the west and east lineages and close connections with *L.* sect. *Dorycnium*, which is inconsistent with data on morphology. Sect. *Dorycnium* is not monophyletic based on all markers. *L. dorycnium* complex is widely distributed and geographically differentiated in Mediterranean. It is supported by ITS marker, but reveals introgression with *L. hirsutus* by plastid data. *L.graecus* from East Mediterranean and Black Sea region is a monophyletic, geographically non-structured species, with two closely related Turkish endemics. Sect. *Lotus* apparently has ancestral forms in area A. It is polyphyletic by ITS and plastid data, but its core group, *L. corniculatus* complex, is well supported by all markers. *L. conimbricensis* and a cryptic species *L. carpetanus* form sister group of the complex. We suppose the European origin of *L. corniculatus* complex and its geographic expansion parallel to the changes in cp haplotypes from A group in western Europe to D group in central Asia. Within the complex, a correlation between population genetic structure and species reproductive strategy was found.

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THE COMMON CUCKOO *Cuculus canorus* AS AN OBJECT FOR RESEARCH ON THE GENETICS BASIS OF THE EVOLUTION OF THE STRATEGY OF REPRODUCTION AND MIGRATION.

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The Common Cuckoo *Cuculus canorus* is a widespread species among birds of Eurasia. It belongs to family Cuculidae (140 species), whose species demonstrate a transition from nest-building and care for offspring to obligate nesting parasitism. Transition looks like this: nest- building and raising offspring → collective nesting (several pairs build a joint nest and hatch eggs together) → tossing in of eggs into the nests of their own species under adverse conditions → tossing in of eggs in nests of other species under adverse conditions → seizing of other nests and care of offspring in them → tossing in of eggs into nests of other species that hatch them and feed the chicks, but after the nestlings are released from the nest, the cuckoo feeds them → obligate breeding parasitism.

The Common Cuckoo is one of the most perfect nesting parasites. The objects of parasitism are small birds of the order of Passeriformes (about 300 species). In addition, each female parasitizes only on the type of birds to which its adoptive parents belonged. Having thrown an egg into another's nest, the cuckoo no longer cares about it, and the host birds hatch and feed its chick. It should be noted that the color of the eggshell of the cuckoo eggs coincides with the color of the eggshell of the host bird. The type of egg shell color is regulated by a gene that is transmitted along the line of females.

The Common Cuckoo is a distant migrant. Wintering sites are located in sub-Saharan Africa and more recently they were also believed to be in India (eastern populations). Having become independent, young cuckoos migrate one by one to wintering grounds. How they find wintering territories is not exactly known. Studies conducted in the past few years by scientists from Russia, Denmark and China using satellite transmitters have allowed not only to establish the route for the migration of cuckoos from European populations, but also to show that Eastern Common Cuckoos also migrate to sub-Saharan Africa, south of the Sahara, and not to India. At the same time, cuckoos from European and Asian populations winter in different parts of Africa.

The genetic basis for the migration of the common cuckoo and the breeding strategies of the representatives of the family Cuculidae have not yet been studied. Thus, the Common Cuckoo is a good subject for research in the field of evolutionary genetics and genetics of behavior.

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COMPARATIVE GENOMICS OF THE *POSTHITIS*-ASSOCIATED CHROMOSOME REGION IN BOVINES

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About 5-6% of male European bison (*Bison bonasus*) are affected by a severe disease called *posthitis* (inflammation of the foreskin). A genome–wide association study showed that most SNPs associated with this disease are located in a 1.1 Mb region (UMD 3.1.1, whole genome shotgun sequence AC_000172.1:45238956-48143907) of the cattle (*Bos taurus*) chromosome 15. This chromosome region is similar to the American bison (subsp. *Bison bison bison*) genomic scaffold (NW_011495061). Surprisingly, regions of high identity are interrupted by long gaps, which may indicate the presence of quite long regions in the genome specific only to bison.

Alongside the *B. b. bison* genome, the SRA section of GenBank has deposited raw sequence reads from eight individuals of *B. bonasus*, i.e. Planta, Platen, BBO 3569, BBO 3574, MdL2, MdLC, Cc1 and Cc2. Although this quantity of deposited sequences would seem to sustain an expectation of approximately 10-fold genome coverage, the number of mapped reads to the *B. taurus* reference sequence in the regions of SNP location is in fact of much lower value. Because sequencing reads are not distributed evenly over the target areas, higher coverage of this region is needed if the above issue is to be addressed.

Among the 18 SNPs located in the AC_000172.1: 45238956-48143907 region, eight are in 5'-flanking sequences, seven in introns, one in a 3'-flanking sequence and two in exons. This shows that most SNPs associated with *posthitis* are located in regulatory elements of genes. SNPs located in the 5'-flanking sequences can affect gene expression by altering the ability of transcription factors to bind to the sequence and activate or repress transcription, while others can be located in enhancers or in not yet described gene regulatory elements.

COMPARATIVE PHYLOGENETIC ANALYSIS OF TWO SISTER SPECIES OF THE GENUS *LOTTIA* GRAY, 1833 FROM THE FAR EASTERN SEAS OF RUSSIA

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There are about 20 species of limpets in the Patellogastropoda (Mollusca: Gastropoda) in Far Eastern seas of Russia, nine of which are within the most numerous and taxonomically complex genus *Lottia* Gray, 1833 (Chernyshev, Chernova, 2005). However, these results were based entirely on the analysis of morphological characters. At the same time, taxa that belong to the genus *Lottia* have been insufficiently studied using molecular genetic markers. Previous studies were based on relatively small number of taxa and individuals, which makes it impossible to fully describe the biodiversity of the genus *Lottia* and each species separately.

Phylogenetic analysis was based on nucleotide sequences of mitochondrial (*12S rRNA*, *16S rRNA*, protein-coding *COI*) and nuclear (*28S rRNA*, protein-coding *Histone 3*) genes in *L. persona* (Rathke, 1833) and *L. versicolor* (Moskalev in Golikov et Scarlato, 1967) species, which occur in the Russian Far East. Intraspecific genetic variability in *L. versicolor* is quite high for both nuclear and mitochondrial markers (for *Histone 3* gene the *p*-distance inside the clade is 0.5%, for gene *16S rRNA* – 7%), while boreal taxon *L. persona*, on the contrary, showed relatively low intraspecific variability over a significantly wide range (from the Kuril Islands to the southern regions of Primorsky Krai). The *p*-distance between individuals within the *L. persona* clade is 0.1% for *Histone 3* gene, 0.13% for *16S rRNA*, and 0.2% for *12S rRNA*. As a result, these two sister species showed different levels of genetic variation, which requires further research.

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MYTILUS EDULIS AND *M.TROSSULUS* IDENTIFICATION USING CONCHOLOGICAL CHARACTERS IN OCEANIC AND ESTUARINE HABITATS, THE KOLA PENINSULA (WHITE AND BARENTS SEAS)

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Mussels *Mytilus edulis* (ME) and *M. trossulus* (MT) are species that demand identification using multilocus genotypes. ME and MT co-occur and hybridize along the coasts of Kola Peninsula. Previous studies have revealed morphological differences between these species in the White Sea: the majority of MT bear an uninterrupted dark strip of the prismatic layer under the ligament on the inner side of the shell (T-morphotype) while the majority of ME lack this character (E-morphotype). Hybrids are not numerous (about 18% frequency of occurrence in a mixed sample), and morphologically are similar to that parent species, whose genes dominate in their genotypes. This morphological character was first introduced for the purpose of discrimination between MT and *M. galloprovincialis* in the Russian Pacific Ocean coast. However, discriminative power of the character has been poorly tested in other geographical regions. The goal of our study was to find out whether morphological differences between ME and MT persist in populations along the Barents Sea coast.

ME and MT were sampled in the White Sea (Kandalaksha Bay, 24 samples from estuarine habitats, salinity up to 25 ppt) and the Barents Sea (Murman coast, 12 samples from estuarine habitats, salinity as in the Whites Sea; 14 samples from oceanic habitats, salinity from 25 to 35 ppt). Mussels were genotyped by four allozyme loci. Contribution of MT and ME genes into individual genotypes was estimated in terms of individual scores obtained by means of a model-based Bayesian clustering method implemented in the program Structure 2.3.4. To assess associations among the genetic (Structure scores), morphological (T, E-morphotypes) and ecological (oceanic vs. estuarine habitats) variables we used a generalized linear mixed-effect models (GLMMs). Samples were considered as random factors in the analyses. The R programming language with lme4-package was used. We found out that, in the low salinity habitats (White Sea and estuarine Barents Sea samples) differences in morphotype frequencies between ME and MT were 67% and 63%, respectively. In oceanic habitats, differences were much lower: 21%, on the average, due to the increased frequency of T-morphotypes among ME. We hypothesize that recorded morphological differences between ME from estuarine and oceanic habitats could be due to the ability of a species to adjust the strength of the shell by active production of the nacreous layer under conditions of high acidity in estuaries. Unlike ME, MT does not show such plastic responses. Reliability of ME and MT identification using specified conchological character is satisfactory high in the estuarine, and low in oceanic habitats across the Kola Peninsula. The probability of correct identification (when it is conceivable that a mussel with T-morphotype is MT, and a mussel with E-morphotype is ME) depends on the taxonomic structure of a sample. E.g., in pure population of ME, rare mussels with T-morphotypes will be 100% ME. For the convenience of mussel researches we present the “T-calculator”: statistical functions relating the frequency of T-morphotype in population to frequency of MT genes and the accuracy of ME and MT identification using conchological character. The calculator has been developed for populations from places with salinity below 25 ppt.

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PROMISING BIOMARKER FOR ASSESSING THE EFFECTS OF THE ENVIRONMENT ON A POPULATION OF PLANTS

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In the last years, special attention has been given to population studies of pasture plants, since environmental pollution through animal food can be transmitted to humans. Therefore, it is essential to regularly monitor pasture plants, both for relocating animals to more environmentally friendly meadows, and for environmental protection measures aimed at improving damaged pastures. The great number of biotic and abiotic factors affects plant under natural growing conditions growth. DNA sequence does not carry complete information necessary to determine the phenotype of an organism. DNA methylation controls genomic integration, regulates genome expression and cell differentiation, as well as plant response to biotic and abiotic stresses. Epigenetic regulation involves various reversible chemical modifications occurring in both the DNA itself and the proteins interacting with it, which as a result affects the chromatin structure and function without, however, altering the sequence of nucleic residues in the DNA. Epigenetics holds the promise to explain at least a part of the influences the environment has on phenotype, *Bromus inermis, Medicago sativa, Onobrychis arenaria,* and *Agropyron pectinoforme* are widely distributed pasture plants. Currently we have studied the DNA methylation of esparcet (*Onobrichis arenaria*) and crested wheatgrass (*Agropyron cristatum*) from the Kazachstan grassland and discuss the environmental factors, which can lead to changes in epigenetic features. The obtained data on epigenetic changes of the studied plants will improve methods of pasture monitoring, taking into account ecological and climate conditions in various regions and agricultural sector of the countries' economies.

GENETIC DIFFERENTIATION OF THE VULNERABLE WHITE-NAPED CRANE *ANTIGONE VIPIO* (GRUIDEA, AVES) BASED ON MITOCHONDRIAL DNA DATA

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White-naped crane *Antigone vipio* (Pallas 1811) is a vulnerable species, and has a narrow breeding range divided into the western and eastern parts with corresponding two patterns of flyways. Western population of the species breeds in the northeastern Mongolia and Transbaikalian region of Russia and migrates south to China for wintering. Birds from the eastern population breed in the Far Eastern region of Russia and northeastern China and fly across Korean Peninsula to Japan. Some cranes from this population overwinter in Korean Demilitarized Zone. We are unaware of any data on population genetic structure of the White-naped crane. For the first time we tried to reveal spatial genetic differentiation between two geographically distant breeding groupings of this species using mitochondrial DNA Control Region (CR) and ND2 gene sequence data. Biological material for DNA analysis (blood samples, plucked and molted feathers) was taken from wild White-naped cranes and captive birds taken from the wild in Russian part of the species range.

We sequenced 642 bp of CR and 694 bp of ND2 of seven and eleven individuals from the western and eastern populations, respectively. ND2 was turned out to be completely monomorphic within the analyzed sample while the CR was highly polymorphic with 21 polymorphic sites. In total, we found 12 CR haplotypes in the studied sample. In general, the haplotype diversity of the species was high ($h=0.960\pm0.026$), but total nucleotide diversity ($\pi=0.0049\pm0.0007$) and average number of nucleotide differences ($k=3.1895$) were low. Four haplotypes were found only in the western population, six haplotypes were specific for the eastern one, and two haplotypes were shared among these samples. Both populations were characterized by similar levels of haplotype and nucleotide diversity and absence of fixed differences between them. Genetic differentiation between samples from two parts of the fragmented species range was weak: $F_{ST}=0.01111$ and $G_{ST}=0.00245$. In general, low differentiation is common for the migratory bird species including cranes. The tests for deviation from selective neutrality of Tajima ($D=-1.284$) and Fu and Li ($D=-1.843$) were negative and non-significant. Data reflect no evidence of recent bottle-neck effect in total population of the White-naped crane in the past. Median-joining network of CR haplotypes did not reveal highly diverged lineages. All haplotypes from both populations, shared and private, originated from a single haplotype 3 found in the eastern grouping. This study based on molecular data indicates shallow genetic differentiations between western and eastern White-naped crane breeding groups that may reflect their incomplete isolation caused putatively by occasional sharing of the two existing flyways by birds from both populations.

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THE STUDY OF BIODIVERSITY OF GREEN SOIL MICROALGAE BASED ON MOLECULAR DATA

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The main challenge in studying of the biodiversity of green soil microalgae is their reliable identification. A significant part of these organisms (especially in the class Trebouxiophyceae) is poor for phenotypic traits: simple, unicellular, immobile cells (coccoid morphology), making it difficult to identify them using light microscopy. In the past decade, a DNA-based approach has revealed the latent genetic diversity of microalgae and made it possible to describe a large number of new taxa of various levels (Cho et al., 2017). In this scope, we analyzed the ITS region and the fragment of the 18S gene of nuclear ribosomal DNA (nrDNA), widely used for barcoding algae and protozoa (Pawlowski et al., 2012).

For 12 strains of soil green microalgae collected from two localities (Kamchatka Peninsula and Egypt; hereafter, the letter “K” or “E” in the codes), length of the nucleotide sequences varied from 618 bp to 1373 bp for ITS and 610–627 bp for 18S nrDNA fragment. We then determined the percentage of their identity (% identity by ITS / 18S, respectively) with those already available in the GenBank database by BLAST. Thus, it was established that each strain belongs to one of the classes:

- Chlorophyceae Wille, 1884, order Chaetophorales (863E – 83/100% – *Stigeoclonium sp.*) and order Chlamydomonadales (817K – 85/99% – *Chlamydomonas sp.*; 868K – 96/99% – *C.* cf. *debaryana*; 845E – 92/95% – *Chlorococcum sp.*; 851E – 86/98% – *Lobomonas sp.*);
- Trebouxiophyceae Friedl, 1995, order Chlorellales (852E – 99/99% – *Oocystis solitaria*; 855E – 98/99% – *Marvania geminata*; 846E – 98/99% – *Nannochloris sp.*;) and 827K – 89/99% – *Coccomyxa sp.*;
- Ulvophyceae K.R. Mattox et K.D. Stewart, 1978, order Ulotrichales (842E – 98/99% – *Tupiella speciosa*; 861E – 88/99% – *Vischerioclonium sp.*); order Ulvales (865E – 92/97% – *Desmochloris sp.*).

In 8 out of 12 cases (<95%) low sequence similarity of ITS nrDNA does not have a reliable definition of species in the strains. However, the 18S region was more conservative and thus allows to define strains at genus level. We believe 863E, 817K, 845E, 851E, 827K, 861E and 865E may be a new species. Phylogenetic analyzes should be carried out to determine the appropriate phylogenetic position inside the classes of green algae; comparison of their morphological features and properties of life cycles with already described species from the corresponding genera will help to test this hypothesis.

The research was carried out with the grant FE №18-4-011.

NEW INSIGHT ON THE RELATIONSHIPS IN GENUS *RHODIOLA* L. (CRASSULACEAE) BASED ON THE INTERNAL TRANSCRIBED SPACER (ITS) rDNA DATA

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Genus *Rhodiola* L. (Crassulaceae) includes 60–90 species mainly distributed in high-attitude and cold regions of the Northern Hemisphere (Mayuzumi, Ohba 2004). Phylogenetic relationships in this morphologically polymorphic genus are poorly understood. We obtained 17 sequences of the ITS rDNA region of *Rhodiola* spp. and took all available sequences from the GenBank database (altogether 634 seq.; 61 species) to make a wide sampling of this genus.

Total 300 ribotypes were detected in the data set. Length of the ITS region (ITS1+5,8S+ITS2) varied from 546 to 579 bp and the average was 571.3±5.7 bp. The percentage of bases guanine+cytosine was in the range 52-56% (mean 54.2±0.9%). The length of ITS1 was 194.4±2.3 bp. Almost all sequences of the 5.8S exon in the data set were 161 bp long. The mean length of ITS2 was 216,1±3,7 bp.

We reconstruct consensus secondary structure models of ITS1 and ITS2 rDNA for *Rhodiola* species to analyze structural features of these regions. Models are generally consistent with those in other plant groups (Coleman, 2015) and were very similar to related Crassulacean genera (*Sedum* L. and *Orostachys* Fisch.; Nikulin et al., 2015; Nikulin et al., 2016).

For most of the *Rhodiola* species (33 of 61), we found common ribotypes, i.e. one ribotype was encountered in sequences of 2 or more species. This may indicate either a close relationship between species within one ribotype, or incorrect identification of samples.

Phylogenetic analyzes (ML and BI) revealed a tree with 5 clusters. The branching pattern between this clusters largely unresolved. Terminal branches characterized moderate (85–94%) to high support (95–100%). Only species with a small number (1–5) of sequences formed species clusters, while most species (>5 seq.) were mixed with other species.

Thus, *Rhodiola* genus is characterized by high ribotypic diversity, not always correlated with the traditional concept of the species, and complex intrageneric structure. Further, for a detailed analysis of the relationship between closely related species/ribotypes, we are planning to use more sensitive molecular markers (IGS rDNA, etc.).

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DISTRIBUTION, COMPOSITION AND RELATIONSHIPS OF ARCTIC PHYLOGENETIC GROUP OF CHARR GENUS *SALVELINUS* (SALMONIDAE) IN THE NORTHEASTERN ASIA AND NORTH AMERICA

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Charrs of genus *Salvelinus* is widely distributed in the North Pacific and Arctic seas and can be regarded as species that can clarify biogeography and evolutionary history of Arctic fauna. Phylogenetic analysis of charrs genus *Salvelinus* shows that *S. andriashevi, S. krogiusae*, *Salvelinus* sp. 4 (Lake Nachikinskoe), *S. boganidae* and *S. elgyticus* belong to the Arctic lineage of Taranetz charr *S. taranetzi* according to Oleinik et al. (2015). This phylogenetic group also includes the Arctic charr *S. a. erythrinus* from the Arctic coast of Canada (NWT), and isolated lake populations of the North Slope of Alaska. One of the unsolved problems is the relationships between *S. taranetzi* from Asia and *S. a. erythrinus* from North America (NA) within the Arctic phylogroup.

Data on the variability of sequences of the three mtDNA fragments (*Cytb, COI* genes, and *CR)*, as well as genealogy of mtDNA haplotypes suggest a phylogenetic similarity between populations of *S. taranetzi* from Chukotka and Arctic charr from NWT. The level of divergence mtDNA within the Arctic lineage was relatively low. At the same time, all representatives display similar divergence estimates (0.01±0.002) with *S. alpinus alpinus* (Atlantic and Sibirian lineages) and *S. alpinus aquassa* (Acadian lineage). The ratio of within- to between population divergence of the mtDNA nucleotide sequences was analogous to that previously reported for *S. taranetzi* (Oleinik et al. 2015). Phylogenetic and genealogical analyses confirm that the Arctic phylogroup of *S. taranetzi* and the phylogroup of *S. alpinus* (Eurasian and Acadian groups) have reached the state of reciprocal monophyly, without retaining the ancestral polymorphism. The genealogical analysis shows the existence of significantly diverged mitochondrial lineages each of which coalesces to their own ancestral mtDNA haplotypes. In general, we have managed to solve the issue of relationships between *S. taranetzi* (with closely related taxa) from Asia and *S. a. erythrinus* from North America by proving their phylogenetic proximity. Heterogeneity tests have shown significant differentiation among allopatric populations of *S. taranetzi* and *S. a. erythrinus*. Hierarchical analysis of molecular variance (490-bp fragment of the control region both for our and Genbank sequences were used in the analysis) revealed the following population structure: (i) high (87.5% of the variance) structuring between, and weak (12.5%) diversity within Asian populations of *S. taranetzi*; and (ii) moderate (34%) structuring between, and high diversity (66%) within North American populations. Overall, an estimated 45% of total variance was distributed between the Asian and NA population groups in the Arctic lineage. Consequently, the Arctic phylogroup in the studied part of the range is characterized by a pronounced structuring, the greatest contribution to which is made by the differences between the two phylogenetic subgroups. The region of Anadyr–Kolyma Basin, which determines the heterogeneity of the entire phylogroup, stands out on a wide geographical scale.

The genetic similarity of geographical samples of *S. taranetzi* and *S. a. erythrinus* (with homing and the lack of anadromous life history stage in the lacustrine Asian charrs) is probably associated with the postglacial colonization from common sources, rather than with the gene flow in modern populations. By assuming a common origin of *S. taranetzi* and *S. a. erythrinus*, we have shown a certain level of divergence between them owing to the probable fragmentation of the common ancestor's range and the subsequent evolution under conditions of isolation by distance. Our data indicate the survival of ancestral populations and the recolonization from different glacial refugia.

NEW GENETIC DATA ABOUT THE PHYLOGEOGRAPHY OF BERINGIAN PHYLOGENETIC GROUP OF CHARR GENUS SALVELINUS: EXISTENCE OF A CRYPTIC PLEISTOCENE REFUGIUM IN THE KAMCHATKA RIVER BASIN

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Territory of the Northeast Asia can be considered as the region of the endemism development of charrs genus *Salvelinus*, since lots of narrow ranged species are presented here. First description of the majority of endemics is based solely on the morphological features that are an essential problem for construction of modern genus phylogeny. The Kamchatka River basin (Kamchatka Peninsula, Russia) is populated by numerous anadromous and resident forms of charrs including the endemic stone charr *S. malma kuznetzovi* that inhabits the rivers and tributaries along with the Northern Dolly Varden *S. malma malma* and white charr *S. albus*. Nucleotide sequences of the entire gene sequence of cytochrome *b* and cytochrome *c* oxidase-1, control region and locus of the lactate dehydrogenase gene (*LDH-C1*) were used for analysis of charr differentiation and phylogenetic relationships. Our results suggest that *S. malma kuznetzovi*, *S. albus*, and *S. malma malma* represent a monophyletic (Beringian) group originating from a common ancestor. The ratio of within- to between-population divergence of mtDNA nucleotide sequences was analogous to that previously reported for *S. malma malma* populations (Oleinik et al., 2014).

The new data allowed us to clarify the phylogeography of Northern Dolly Varden and discuss the origin of stone charr. Two divergent mtDNA lineages have been identified in the Kamchatka River basin, with stone charr and white charr being the main carriers of the haplotypes of a new identified group in this region. Evidence has been obtained for the common origin of stone charr from the Kamchatka River basin and *S. malma malma* from the Chukchi Peninsula and Alaska. Haplotypes of the ancestral stone charr population, representing an independent radiation center, diverged from *S. malma malma* of the Kamchatka River. Presence of highly diverged haplotypes in the Kamchatka River basin is a primary feature of phylogenetic groups existing in stable conditions without sharp fluctuations in abundance for long periods of time or groups that diverged through genetic drift within different refugia. The range of *S. malma kuznetzovi* is limited to the Kamchatka River basin, so we infer the existence of a small cryptic refugium of Northern Dolly Varden. Such a scenario is supported by the complicated historical demography of *S. m. malma* reflecting the periodicity of Pleistocene glaciations, as well as by modern views on the paleogeology of the region. It is known that Pleistocene glaciations on the Pacific coast of the Bering Sea were of a mountainous rather than integumentary nature, as were large-scale glaciations of the Middle and Late Pleistocene in Kamchatka Peninsula. In this time the Kamchatka River valley was not influenced by glaciers and river retained a continuous runoff flow. Concurrently, various types of geomorphological activity in the region repeatedly created conditions for isolation of charrs.

Despite a low level of genetic divergence stone charr is of particular interest for studying problems of microevolution of charrs. Results suggest that further studies of charr populations in the Kamchatka River basin may reveal greater diversity than was previously estimated. Genetically distinct charr of Beringian group would not be surprising, given the environmental diversity of the Kamchatka region. Charr coexist with many species of Pacific salmon (*Oncorhynchus* spp.: chum salmon, coho salmon, chinook salmon, sockeye salmon, pink salmon) in this watershed, so geographic spread and Beringian group distribution may differ from those in other areas of the Holarctic. Additional research and integration of information on charr populations would allow comparative analysis that may identify determinative factors of evolution in *S. malma malma*.

MOLECULAR ANALYSIS AND NEW RECORDS OF THE INVASIVE POLYCHAETE *BOCCARDIA PROBOSCIDEA* (ANNELIDA: SPIONIDAE)

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The spionid polychaete *Boccardia proboscidea* Hartman, 1940 is a tube-dweller and shell/stone-borer widely occurring in temperate waters across the World and considered an invasive species in many areas. It was originally described from California, USA, and later reported from Pacific Canada, Asian Pacific, Australia, New Zealand, Argentina, South Africa, and northern Europe. Genetic similarity was reported only for worms from South Africa and the Pacific coast of North America.

We reviewed previous and analysed new records of this species using molecular approach with the aim to examine whether that species either has a cosmopolitan distribution or, in some locations, represents a complex of morphologically similar species, and further suggest possible ways of the species introduction.

Sequence data of three gene fragments (836 bp in total) of mitochondrial *16S* rDNA, nuclear *28S* rDNA, and *Histone 3* suggested that individuals from the Pacific coasts of Canada and USA, Argentina, Australia, South Africa, the United Kingdom, and Mediterranean France were genetically very similar (maximal average *p*-distance value, 0.49%, was between *16S* rDNA sequences). We consider these individuals to be conspecific and report the earliest records of *B. proboscidea* from the UK and for the first time from Mediterranean France, in the Gulf of Lion. Our analysis supports the conspecificity of individuals living in different habitats: in sandy and silty tubes in soft sediments and boring into the shells of various mollusks and into different kinds of stone. High *16S* haplotype diversity in *B. proboscidea* from the Pacific USA suggests that the species native distribution is in the northern Pacific, and subsequent introductions of this species to other parts of the World occurred through human activities.

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GENETIC DIFFERENTIATION AND SYSTEMATICS OF CHROMOSOMALLY POLYMORPHIC RODENT, THE STRIPED HAMSTER *CRICETULUS BARABENSIS* SENSU LATO (RODENTIA, CRICETIDAE): REVIEW OF ACHIEVEMENTS IN "MOLECULAR ERA" OF TAXONOMY AND A ROLE OF "OLD" CYTOGENETIC METHODS

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The superspecies *Cricetulus barabensis* sensu lato is represented by allopatric chromosome forms, taxonomic status of which is ambiguous. The superspecies include karyoforms "griseus" (2*n* = 22), "pseudogriseus" (2*n* = 24) and "barabensis" (2*n* = 20). They are distributed widely across the steppes in the eastern and central Palearctic. Morphologically, these forms are poorly differentiated (Lebedev, Lisovskii, 2008). Experimental hybridization between them under laboratory conditions is quite successful (Poplavskayaet al., 2012a, 2012 b). Several potential zones of natural hybridization were revealed in Mongolia and Russian Transbaikalia (Poplavskaya et al., 2012a, 2012 b; Korablev et al., 2013). Investigation of diversity of six microsatellite loci and partial *RAG1* exon of "barabensis" and "pseudogriseus" karyoforms supports subdivision into two well-differentiated clusters corresponding to the two karyoforms. It was shown that rare hybridization events between the two chromosomal races occur in nature in central Mongolia (Poplavskaya et al., 2017). Phylogenetic analysis of cytochrome b gene sequences based on about of 500 specimens from more than 100 localities revealed five well supported lineages divergent at 2–4%. Two of them correspond to "griseus" (2*n* = 22) and "pseudogriseus" (2*n* = 24) karyoforms and are placed as sister taxa. The "barabensis" (2*n* = 20) karyoforms is represented by three other branches and appears non monophyletic. All mtDNA lineages are distributed allopatrically or parapatrically; no indications of gene flow between populations of different chromosomal races were found. Common ancestor of the recent lineages hypothetically belong to the 2*n* = 20 karyoforms and originated in the eastern part of the modern range (Poplavskaya et al., 2019). In all cases, study of the karyotypes with the routine methods of analysis was preceded to molecular genetic analysis. First step was to outline the borders of distribution of these forms and to analyze the karyotypes of animals from potentially contacting populations of "barabensis" and "pseudogriseus". For most of molecular studied specimens, karyotypes were detected previously. The analysis of karyotypes of animals from 35 locations in the southeastern Transbaikalia (the region of potential hybridization) was performed in the Laboratory of evolutionary zoology and genetics FSC of Biodiversity FEB RAS. Two allopatric chromosomal forms presumably of the species rank (*Cricetulus barabensis* sensu stricto (2n = 20), and *C. pseudogriseus* (2n = 24)) inhabit the territory between the Ingoda, Shilka and Argun rivers. In the southern areas of this region, the border between these forms passes along the Onon River valley; it may be considered as a valid geographical barrier that separates the nearest known locations of two different chromosomal forms. Our results do not support the idea that the Shilka River is a zoogeographical barrier for different karyoforms (Korablev et al., 2013; Pavlenko et al., 2016). The pattern of distribution areas for contacting populations is more complicated than it was suggested earlier, based on morphological data. Molecular data confirm the absence of hybridization between these karyoforms in the southeastern Transbaikalia (Poplavskaya et al., 2019).

MICROEVOLUTIONARY PROCESSES IN THE COMMON SHREW: CHROMOSOMAL RACES AND HYBRID ZONES

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The common shrew, *Sorex araneus* (Mammalia, Lipotyphla) is one of the very good models to study the role of chromosomal rearrangements including Robertsonian (Rb) fusions and whole-arm reciprocal translocations (WARTs) in speciation. This species is subdivided into more than 75 parapatric chromosomal races distinguished by Rb and WART. Up to data about 35 chromosomal hybrid zones (an area where interracial hybrids occur) between different races are known and 17 of them located in Russia. Hybrids between races are often complex heterozygotes, forming complex configurations at meiosis, and most likely such hybrids could suffer infertility. Thus, common shrew chromosomal races could be reproductively isolated. However, there are no evidences of strong limitation of gene flow between races within hybrid zones. Based on analysis of karyotypic variability and distribution patterns of the 26 chromosomal races in Russia we found two centres of high karyotypic diversity: western (Baltic region) and eastern (near Lake Baikal). We supposed that those places were some sort of “refugee” from where further recolonisation processes started. We also found that ecological factors or geographical barriers did not play a significant role to limit race ranges or to “trap” hybrid zones. Apparently, the main factor to maintain current distribution of races is density-dependent processes. We will discuss all these questions in the presentation.

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INTRA- AND INTERSPECIES GENETIC COI DISTANCES WITHIN FRESHWATER MUSSELS OF THE GENUS *UNIO* FROM THE VOLGA BASIN

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Currently, only three species are known in the genus of freshwater mussels *Unio* in European Russia, but no taxonomic validation by molecular data for these species yet exists. We sequenced partial COI genes for 23 individuals of *Unio* freshwater mussels, collected in the Volga basin from 3 regions located within an area of 550 km in latitude and 1200 km in longitude (Uglich, Korozechna river; Ugori, Unza river; Buzuluk, Borovka river). Sequences obtained in this study were aligned with those taken from GenBank and analyzed using MrBayes and the MEGA7 programs. At phylogenetic trees, all sequences including obtained in this study fall into three major groups corresponding to three *Unio* species regardless of the methods of analysis. Sequences of geographically distant populations from West, Central and East Europe form homogenous species clusters supported by 100% bootstrap values on ML and MP trees and by 1.0 PP on Mrbayes tree. The sequences were distributed as follows: 10 sequences fall into the *U. pictorum* group (5 sequences from Uglich, 3 from Ugori, 2 from Buzuluk); 5 sequences fall into the *U. crassus* group (all from Ugori); and 8 sequences fall into the *U. tumidus* group (6 from Buzuluk, 1 from Uglich, 1 from Ugori).

The levels of intra- and intergroup polymorphism for the COI gene estimated from genetic distances in *Unio* ranged from 0.001–0.004 and 0.036–0.152. Therefore, the values of intra– and interspecies genetic distances for the genus *Unio* did not overlap and are close to those obtained for other groups of mollusks.

Therefore, the genus *Unio* is represented in the Volga basin by three species. Our study did not support the existence of hidden species in the genus *Unio*.

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RENAL FUNCTIONS AND KIDNEY GENE EXPRESSION IN HYPERTENSIVE STRESS-SENSITIVE ISIAH RATS

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Hypertension is one of the most common diseases, characterized by a persistent increase in blood pressure (BP). In general, hypertension is a multifactorial disease. Since genes and environment make practically equal contribution to variability in blood pressure, genetic studies on regulatory processes underlying the hypertension development are conducted mostly on inbred rat strains in order to control environmental stability.

Kidneys provide long-term regulation of blood pressure due to their ability to maintain water-salt balance of the body. It is known, that the salt loading or reduced blood flow in kidneys can lead to arterial hypertension development. The aim of this study was to evaluate how effectively hypertensive ISIAH rats with inherited stress-induced arterial hypertension may cope with the salt loading and to find the genes which may participate in kidney response to salt loading as compared to control normotensive Wistar Albino Glaxo (WAG) rats. Groups of intact and experimental (with chronic salt loading) ISIAH and WAG male rats were used. Each group included 6 animals at the age of 3 months. Animals were kept separately in metabolic cages (Techniplast, Italy). Experimental rats received water containing 1.5% NaCl for 7 days (chronic salt loading). The control groups of rats drank the tap water. Diuresis and the urine components were measured daily. Sodium was measured by the photometry of flame. Glomerular filtration rate (GFR) was calculated from serum and urine creatinine concentrations and minute volume of diuresis. Different levels of gene transcription in the isolated renal cortex and medulla were observed using whole genome RNA-Seq. The functional annotation of differentially expressed genes was performed in the Gene Ontology database. The diuresis, GFR and natriuresis did not differ in the groups of control ISIAH and WAG rats. At the saline load, a significant increase in natriuresis and glomerular filtration rate was observed in the ISIAH rats compared to WAG without changes in diuresis. However, when the salt loaded and the control ISIAH rats were compared, a significant increase in daily natriuresis and diuresis was found in the loaded ISIAH rats, but no changes in the GFR were observed. In contrast, no significant differences in all studied parameters were found in the loaded and control WAG rats. The analysis of transcriptional activity of genes in kidneys of ISIAH and WAG rats revealed several groups of genes described by the Gene Ontology terms, which might be associated with genetically determined specificity of natriuresis and GFR processes in the hypertensive kidneys. Groups of genes differentially expressed in the renal cortex of ISIAH and WAG rats were described in terms of regulation of ion transport, ion transport, ion homeostasis, regulation of response to stress, regulation of body fluid levels. Groups of genes differentially expressed in renal medulla of ISIAH and WAG rats were described in terms of regulation of ion transport, ion transport, ion homeostasis, regulation of sodium ion transport, sodium ion transport, regulation of response to stress, response to stress, response to osmotic stress, response to salt stress, response to water deprivation. The experiment showed an increased natriuresis in ISIAH salt loaded rats as compared to the control normotensive WAG rats, which may be due to the specificity of expression of many genes in the kidneys of ISIAH rat. Therefore, the hypertension in the ISIAH rats may be considered as sodium independent.

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TRIPLE SYMBIOTIC SYSTEM *PARAMECIUM BURSARIA*-SYMBIOTIC *CHLORELLA*-CHLORELLA *VIRUSES*: IDENTIFICATION AND CORRELATED BIODIVERSITY

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BACKGROUND. *P. bursaria* belongs to one of the best studied Ciliate's genus, *Paramecium*. This species diverged basically from other paramecia. They possess' symbiotic Chlorella in the cytoplasm. These chlorella were attributed to three Chlorella species: *Ch. vulgaris, Ch. variabilis* and *Micractinum reisseri*, according to their phylogeny inferred from *ITS1-5,8S-ITS2* sequence and their sensitivity to specific viruses (PBCV). Morphospecies *P. bursaria* consist of 5 syngens, reproductively isolated Intraspecies groups. Previously we have shown that syngens were represented by different branches on phylogenetic trees.

The purpose of our investigation was to study phylogeny of *P. bursaria* and their symbiotic Chlorella using the same set of strains collected in different geographic regions.

RESULTS. As molecular marker for Chlorella we have used partial sequence of gene RuBisCo. The tree consists of three separate branches. Diversity in each branch is negligible. Our data support strong correspondence between *P. bursaria* syngens and branch of Chlorella, while no correlation between Chlorella type and its geographic location.

CONCLUSIONS. According to our results we have concluded that successful symbiosis between *P. bursaria* ancestor and Chlorella has been established at least three times. The last time it had been happened before syngens R1 and R2 have diverged. We propose the strategy of evolution and geographic dispersion of this symbiotic system.

MOLECULAR PHYLOGENY OF RUSSIAN FAR EASTERN FLOUNDERS (PLEURONECTIFORMES, PLEURONECTIDAE) BASED ON *16S* rRNA GENE SEQUENCES

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To increase knowledge about the systematics of Pleuronectidae the primary sequence of nucleotides at *16S* rRNA genes were determined. In total 14 newly collected species and some species from GenBank were analyzed in this research. Phylogenetic relationships among representatives of flounders were based on three types of trees: neighbor joining, maximum parsimony and maximum likelihood. The largest subfamily in the family is Pleuronectinae. The subfamily Pleuronectinae is represented by two tribes, this is Microstomini and Pleuronectini. They do not form a monophyletic cluster. Thus, systematics at the subfamily level needs further refinement. Two separate clusters on the trees support subfamily Hippoglossoidinae and Hippoglossinae subdivision and monophyletic status of these taxa. The phylogenetic status of *Hippoglossoides elassodon* and *H. robustus* is uncertain and need to be resolved in further investigation. The genus Lepidopsetta (*L. mochigarei*) was included in the tribe Pleuronectini, whereas in the research of Cooper and Chaplau (1998) this genus was considered exclusively as part of the tribe Microstomini. Thus, the genus *Lepidopsetta* may be considered as part of the tribe Pleuronectini.

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DISTRIBUTION OF B CHROMOSOME MORPHOTYPES IN *APODEMUS PENINSULAE* (RODENTIA) FROM THE UPPER PRIAMURIE

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Karyotypes of *Apodemus peninsulae* Thomas, 1906 includes supernumerary (B) chromosomes in addition to chromosomes of the basic set. Number, size and morphology of B chromosomes are different in specimens and populations. We formerly described small and medium metacentric B chromosomes (Bs) as most prevalent morphotypes in territories of the Middle (n = 34), Lower (n = 67) Priamurie and Prymorskii Krai (n = 319). And large meta-, submeta-, subtelocentric; medium and small submeta-, subtelocentric and mini B chromosomes were classified as rare morphotypes (Roslik et al., 2016; Roslik, Kartavtseva, 2017). Descriptions of B chromosome morphotypes were previously performed only for single specimens (n = 5) of *A. peninsulae* in a large extended territory of the Upper Priamurie (Kartavtseva et al., 2000; Kartavtseva et al., 2015).

The aim of our work was to study the karyotypes of mice from the Upper Priamurie and to obtain a complete picture of the B chromosomes morphotypical diversity this species in the Far East of Russia.

Karyotypes of 45 specimens from 11 new populations of the Upper Priamurie (Amur Region: Bol'shaya Omutnaya River, Urusha vil., 3 localities from Zeya city, Argi River, Magdagachi vil., Zheltoyarovo vil., Malinovka vil.) and adjacent territories (Transbaikalia: Amazar vil.; Khabarovsk Region: imeni Polini Osipenko vil.) have been studied. Animals were captured during 2012-2016. Besides we analyzed karyotypes of five early studied mice from two populations of Amur Region: Belogorie vil. and Bomnak Reserve (Kartavtseva et al., 2000; Kartavtseva et al., 2015). B chromosomes were revealed in all specimens. We described 157 Bs in total.

Small metacentric Bs were prevailent (41%) in animals of the Upper Priamurie as well as in the Middle, Lower Priamurie and Prymorskii Krai. Further to, there are findings a B chromosome morphotypes changes in the karyotypes of *A. peninsulae* from this region, compared with the ones of animals from previously studied populations of the Russian Far East. So, almost a quarter of the findings of B chromosomes were micro Bs (24%), which are typical for karyotypes this species from the Siberia. These micro B chromosomes appear in the karyotypes of *A. peninsulae* populations, together with the middle metacentrics (18%) and mini (10%) B chromosomes. Large meta- (3%), middle submeta- (1%) and small submetacentric (3%) Bs are allocated as very rare. The diversity of B chromosome morphotypes in studied animals are slightly higher than those in the Middle and Lower Priamurie, but lower than in Primorskii Krai.

Thus, our study allowed us to describe the B chromosome morphotypes in *A.peninsulae* of the Upper Priamurie and to draw a more complete picture of the B chromosome morphotypical diversity both in this region and in the Far East as a whole. A comparative analysis of the *A. peninsulae* karyotypes in this region suggests that they have B chromosome morphotypes, which are characteristic both of far eastern and siberian populations of the species. Our data are conformed to the hypothesis of repeated settlement of various regions by mice from different refugiums.

CHARACTERIZATION OF THE COMLETE MITOCHONDRIAL GENOME SEQUENCE OF THE OPISTHOCENTRUS OCELLATUS (ZOARCALES: OPISTHOCENTRIDAE) AND ITS PHYLOGENETIC POSITION AMONG EELPOUTS

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The ocellated blenny, *Opisthocentrus ocellatus,* is one of the common species of the recently established family Opisthocentridae (Teleostei: Perciformes: Zoarcales). However, this view is still controversial, the scientific literature and particularly the GenBank Taxonomy Browser still list *O.ocellatus* in pricklebacks (Stichaeidae) family. In this study, we sequenced the complete mitochondrial genome of ocellated blenny and investigated the gene content and organization compared with other species. We also reconstructed phylogenetic tree for the purpose of analyzing the evolutionary relationships within Zoarcales infraorder.

The mitogenome of *O. ocellatus* comprised 16,525 bp in length, including 13 protein-coding genes, 22 tRNA genes, 2 rRNA genes and two noncoding regions (CR or D-loop and origin of L-strand replication or OL) which is in accordance with the mitogenomes of the typical ray-fined fish (Kartavtsev et al., 2007). Among these genes, 12 protein-coding genes (PCG) are encoded on the H-strand and the remaining 9 genes including one PCG encoded on the L-strand (ND6, Gln, Ala, Asn, Cys, Try, Ser, Glu, and Pro). The overall base composition of H-strand is 26.6% A, 27.5% T, 18.1% G, and 27.4% C, total nucleotide composition is 54.4% A+T rich, additionally, the AT skew (-2.1364) and its GC skew (−0,2064) is negative, indicating a higher content of Ts and Cs. Commonly methionine (ATG) is the start codon for most PCGs, except for COI, ND2, ND3 which utilizes GTG, ATA and ATC. Incomplete stop codons were revealed in ND2, ND3, ND4, COII, ATPase 6 and Cyt-b genes. The incomplete stop codon is usually found in metazoan mitogenomes, which is presumably completed via post-transcriptional polyadenylation. The total number of codons in PCGs is 292, where AGA and AGG codons are not represented. Leucine (Leu), alanine (Ala), and threonine (Thr) are the most common amino acids in *O. ocellatus* mitogenome PCGs. The AT skew of PCGs is negative (-0.0392) except for *ATP8* (1.8868).

By phylogenetic analysis, the mitogenome sequences could be used to resolve the higher-level relationships of Zoarcales infraoder. The results indicated that Opisthocenthridae are most closely related to the family of Pholidae then to Stichaeidae, as well as other families within eelpouts. Our findings provided important data for further studies on the population genetics and evolutionary biology of Opisthocentridae and additional taxonomic work is needed in the future.

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MOLECULAR PHYLOGENY FOR THE NON-BITING MIDGES OF THE DIAMESINAE AND PRODIAMESINAE SUBFAMILIES (DIPTERA: CHIRONOMIDAE) OF THE PALEARCTIC

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Amphibiotic insects play a greater role in the trophic relationships of freshwater and near-water biocenoses. Phylogeny of Diamesinae and Prodiamesinae is currently not enough studied. Ome authors (Brundin, 1966; Serra-Tosio, 1973; Rossaro, 1995) proposed detailed phylogenetic relationships of Diamesinae and Prodiamesinae based on morphological features. The phylogeny of Diamesinae constructed using multivariate analysis showed primitiveness of the tribe Boreoheptagyiini, and further divergence of two sister clades, Protanypini and Diamesini (Rossaro, 1995). Modern molecular multilocus approach used only for chironomids collected in southern hemisphere (Cranston et al., 2012). Using several mitochondrial and nuclear loci, we reconstructed the phylogeny of two subfamilies of non-biting midges. A number of *Boreoheptagyia* species were paraphyletic relatively to the genus *Shilovia*, which show necessity for a revision of the tribe Boreoheptagyiini. The high isolation of the tribe Protanypodini (Diamesinae) shows the need to change taxonomic status to independent subfamily Protanypodinae. Phylogeny of other genera in the subfamily Diamesinae corresponds to the morphological data.

Brundin, L. Transantarctic relationships and their significance, as evidenced by chironomid midges. With a monograph of the subfamilies Podonominae and Aphroteniinae and the Austral Heptagyiae. // Kunglika Svenska Vetenskapakadamiens Handlingar. 1966. V. 11. N1. P. 1-474.
Cranston P.S., Hardy N.B., Morse G.E. A dated molecular phylogeny for the Chironomidae (Diptera) // Systematic Entomology. 2012. V. 37. P. 172–188.
Rossaro B. The distribution of Palaearctic Diamesinae (Insecta, Diptera, Chironomidae) // Spixiana, Zeitschrift für Zoologie. 1995. V. 18. P. 177 - 186.
Serra-Tosio B. Ecologie et biogeographie des Diamesini d'Europe (Diptera, Chironomidae) // Trav. Lab. Hydrobiol. Piscicult Grenoble. 1973. V. 63. P. 5-175

MOLECULAR-PHYLOGENETIC RESEARCH OF THE *LOTTIA TENUISCULPTA* SPECIES COMPLEX (PATELLOGASTROPODA: LOTTIIDAE)

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Genus *Lottia* is the largest within family Lottiidae. Species within the genus differ mainly by the shape, color, and sculpture of the shell. These traits are highly variable and often lead to misidentification. Other features (the structure of the radula, jaws) can be very conservative within the genus. Distribution data and biology of closely related species from the Lottiidae are incomplete, and the previous studies suggest that there are a large number of sibling species.

In the current research, the species *L. tenuisculpta* was analyzed as one of quite taxonomically complex species. On the phylogenetic trees based on *16S* rDNA and *12S* rDNA, all individuals of this species were divided into 2 subgroups with high support and relatively large distances between them, *L. tenuisculpta* (1) and *L. tenuisculpta* (2). Both subgroups include specimens from the Amur Bay, Sakhalin Island and the Kuril Islands, subgroup *L. tenuisculpta* (1) also includes individuals from Kievka Bay. The *p*-distances within clades of *L. tenuisculpta* (1) and *L. tenuisculpta* (2) are 0.08% and 0.2%, respectively; while between clades they are 1.1%, which corresponds to the interspecific level of variability. One specimen from the GeneBank (NCBI), defined as *L. tenuisculpta* (AB106499), showed a separate position in relation to the other *L. tenuisculpta* specimens. The distances between the samples *L. tenuisculpta* from the GeneBank and *L. tenuisculpta* (1), *L. tenuisculpta* (2) are 12% and 13%, respectively. So, based on the obtained data, we can suggest that individuals of *L. tenuisculpta* (1), *L. tenuisculpta* (2) and *L. tenuisculpta* AB106499 are species complexes.

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LARGE-SCALE DNA METABARCODING STUDY PROVIDES NEW INSIGHTS ON DIVERSITY AND ECOLOGY OF MYXOMYCETES

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Myxomycetes constitute the most species-rich group within the Amoebozoa. Due to the presence of fruiting bodies in their life cycle they can be relatively easily detected in the field, compared to the other members of Amoebozoa. This allowed accumulate a large body of data on diversity and distribution of myxomycetes during the last 250 y. In contrast to data derived from fruiting bodies, we know much less about the populations of their trophic stages. DNA metabarcoding studies that use 'universal eukaryotic' primers generally fail to detect them due to highly divergent sequences of ribosomal genes in this group. Only four studies based on primers specific for myxomycetes were published to the moment. We report the results of a DNA metabarcoding study targeting myxomycete populations on large geographical scale, including several European mountain ranges and lowlands as well as mountains on Kamchatka peninsula. This data set allowed for the first time to compare communities of trophic stages of myxomycetes among different geographical regions across environmental gradients, bringing new insights on their diversity, ecology and distribution.

THE GENETIC DIVERSITY COMPARISON OF *METAGONIMUS SUIFUNENSIS* FROM DIFFERENT PARTS OF THE AMUR RIVER BASIN

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In 2017, based on the sequences of the *ITS2* region and the *28S* gene of nuclear ribosomal DNA, a new species *Metagonimus suifunensis* was described in the Russian Far East (Shumenko et al., 2017). The status of this species was confirmed using sequences of the *cox1* gene of mitochondrial DNA (Tatonova et al., 2018).

In this study, using the complete sequences of the *cox1* gene we compared genetic diversity level of *M. suifunensis* from different localities of the Amur River basin. We used the samples from six localities in Primorsky Region (Komarovka, Komissarovka, Sorochevka, Odarka, Ilistaya and Arsenyevka rivers), as well as samples from the Odyr and Anuj rivers in Khabarovsk Region and the Bira River in Jewish Autonomous Region. The complete nucleotide sequences of the *cox1* gene (1539 bp) were analyzed for these samples of *M. suifunensis*.

Previously, we have found that samples from Primorsky Region have a low level of variability, both in nuclear and mitochondrial marker. This study showed that samples from the Middle and Lower Amur have a higher level of variability than those from the Ussuri River basin. The sequences of *M. suifunensis* from the Primorsky Region showed 10 variable sites, while 15 sequences from the Khabarovsk Region showed 20 variable sites. This phenomenon of low variability presence in the whole *M. suifunensis* area may be due to the "founder effect". Probably, the parasite expansion was carried out in the investigated region from the Amur River basin to the southern territories (the Ussuri River basin including Khanka Lake and the Razdolnaya River). In our opinion, the distribution center of *M. suifunensis* is the Khabarovsk Region. At the same time, peripheral populations are located in the Primorsky Region.

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GENETIC JUSTIFICATION OF A 3000-KM LONG GAP IN THE RANGE OF THE SMELT *HYPOMESUS OLIDUS* (OSMERIDAE)

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Study of circumpolar species, to which the smelt *Hypomesus olidus* (Pallas, 1814) belongs, can elucidate not only the specific features of their evolution, but also could provide a deeper insight into the role of glaciations and hydrographic changes in forming the modern genetic structure of northern marine and estuarine fish species and their dispersal. It is known that the range of *H. olidus* has two extensive gaps in their distribution area: one about 3000 km long in its North Asian part from the Alazeya River to water bodies of the Polar Ural region (Klyukanov, 1975) and the other 1100 km long in the North American part (Degraaf, 1986). Recently, a small population of *H. olidus* was discovered in Varandey Bay (Barents Sea) at a considerable distance from the western distribution boundary of the species (Novoselov et al., 2019). Its origin is of much interest. The molecular genetic study of the combined mtDNA gene sequences (1–588 bp *CoI* and 589–1668 bp *Cytb*) of 119 specimens of *H. olidus* from nine populations in the Russian part of the range and three additional sequences derived from the GenBank showed that, like other osmerid fishes, *H. olidus* is characterized by high levels of mtDNA molecular diversity. The average values of haplotype and nucleotide diversity of *H. olidus* across its distribution range were similar (0.9004 ± 0.0225 and 0.0107 ± 0.0059, respectively). Haplotype diversity estimates of the sample from Varandey Bay ($h = 0.7320 \pm 0.0732$) were somewhat lower than the average for the range, the values of nucleotide diversity were significantly lower ($\pi = 0.0024 \pm 0.0019$). As indicated by AMOVA, *H. olidus* is characterized by a high geographical structuring of populations ($P < 0.001$). Evaluation of mtDNA polymorphism subdivision by the F_{ST} test confirmed differentiation between samples and the lack of contemporary gene flows between samples from remote geographic regions. These data indicate long isolation of these geographic populations of *H. olidus*. At the same time, the genealogical analysis of conjugate geographical distribution and the mismatch distribution of mtDNA haplotypes indicate a phylogenetic proximity and common demographic history of smelt from the Bering and Barents seas. The genetic-statistical evaluation of the demographic processes of *H. olidus* suggests demographic expansion and spatial expansion from the Bering Sea into the Barents Sea. Demographic expansion for the Barents and Bering seas dates to the same time period, 34000 - 59370 years ago ($P = 0.05$) and, according to our data, precedes spatial expansion. Statistical estimates of the historical demographic processes in *H. olidus* also suggest that some populations may have survived in glacial refugia of the Polar Ural region; their presence is confirmed by recent paleogeographic studies. Our results and literature data suggest that during the last ice age (15-22 thousand years ago), *H. olidus* could have survived not only in southern refugia, but also in the Berengian refugium and in Polar Ural refugia. Until *H. olidus* populations are found in the area from Baidaratskaya Bay (Kara Sea) to the Alazeya River, the vast gap in the modern range of the species, in fact, confirms its survival of adverse conditions in near-glacial refugia at the junction between Asia and Europe.

CLONAL VARIABILITY OF *CLONORCHIS SINENSIS* IN *PARAFOSSARULUS* SNAILS

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Clonorchis sinensis is the representative of trematodes with passive infection of the first intermediate host (snail). In the view of some researchers, snails can swallow several eggs with invasive miracidiae, which then will be developed to mother sporocysts. In contrast, there is an opinion that only one mother sporocyst is developed in snail's body. To try to go into the matter, we used the genetic marker, *CsRn1*retrotransposon, detected in *C. sinensis* by Bae et al. (2003).

It turned out that there was the specific pattern of retrotransposon loci within each line of cercariae for *Clonorchis* population in Russia (the line is all cercariae emitted from one snail). It is interesting that a variability was absent in retrotransposon pattern within all one-line cercariae. Moreover, the variability was also absent between embryo cells from one mother sporocyst. However, for *C. sinensis* population from Vietnam a clonal variability was revealed, despite the fact that a specific pattern remains unique for each one-line cercariae from different snails. In addition, cercariae lines without variability were obtained.

We suppose that only one miracidium develops in each snail. *Parafossarulus* snails have small digestive gland, in contrast to some other mollusks, for example, *Parajuga*, which can be infected by with cercariae at once four species of trematodes (own data). The presence of the clonal variability can be the result of asynchronous development of daughter generations in mother sporocyst. It means that both mother and its daughter sporocysts produce next generations periodically and different times, to minimize negative impact on the host (snail). At the same time, development features of separate parasite stages in snail's body and impact of retrotransposons on their adaptive facilities are still unknown. Additional data can be helpful to contribute to a better understanding of the issues.

THE *OCA2* GENE MUTATIONS DETERMINED THE YELLOW EYE COLOR OF THE JAPANESE SCOPS-OWL *Otus semitorques* Pennant, 1769 (Aves: Strigidae)

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Eye color, along with vocalization, is one of the main taxon-specific markers in the systematics of Asian scops owls (superspecies complex *Otus bakkamoena-letia-semitorques-lempiji*..). The Far Eastern form of Japanese scops owl is considered to contain 3 subspecies: nominative *O. s. semitorques* with fiery to dark red eyes (Northen Japan and Kuril Isl.), *O. s. pryeri* with dark yellow eyes (extremely Southern Japan) and continental race *O. s. ussuriensis* with intermediate yellow to orange-yellow eyes (Amurland). In fact, most of the birds registered within the range of *O. s. ussuriensis* during breeding season have bright orange-red eyes just like the nominative subspecies. Only some specimens have yellow or yellow-orange eyes. The narrow geographic localization of yellow-eyed individuals (extreme southwest Primorye) and only seasonal finds (winter time) suggest that we are either dealing with different forms, one of which is residential (yellow-eyed), the other - mainly migratory; or that the eye color is a function of the physiological condition or age of specific individuals and, therefore, cannot be a strict taxon-specific trait.

The aim of this study is to solve the problem of determining the yellow eye color of the mainland form of *O. semitorques* (race *ussuriensis)* using nuclear markers and in particular the *OCA2* gene fragment encoding the integral transmembrane *P*-protein and playing an important role in determining eye color. We analyzed 6 yellow-eyed and 4 red-eyed specimens of *O. s. ussuriensis,* collected in the south of Primorsky region. For comparison we took other representatives of yellow-eyed owls: a closely related species *O. sunia*, along with other yellow-eyed owls like *Ketupa blakistoni*, *Nyctea scandiaca* and *Aegolius funereus*, and also one species with orange eyes, *Bubo bubo*. For the outer group we used *Spizaetus nipalensis* with yellow eyes, *Buteo buteo* with yellow or brown eyes and *Falco peregrinus* with brown eyes (Genbank, NW_004929449 chromosome I fragment). There are no molecular studies related to the determination of eye color in birds and in this respect this paper is innovative. For the first time we studied the polymorphism of the *OCA2* gene fragment (about 1.2 kb) in the red-eyed (normal eye color) and yellow-eyed (rare eye color) forms *O. s. ussuriensis*. All red-eyed individuals had both of the wild-type alleles with single synonymous mutation. Yellow-eyed birds carried one allele of the wild type, the second was mutant with AATT duplication in the intron I. Despite of the insertion in the yellow-eyed birds, a fragment of the *OCA2* gene of the red-eyed and yellow-eyed forms of *O. s. ussuriensis* has the same sequence; which differs from the one in the *O. sunia* with yellow eyes. It was unexpectedly found that *B. bubo* with orange eyes had a sequence identical to red-eyed *O. s. ussuriensis*. The genetic differentiation between *O. semitorques*/*B. bubo* was only 0.04%, while for *O. semitorques*/yellow-eyed *O. sunia* it was 2.3%, which exceed the corresponding values between other yellow-eyed owls *N. scandiaca*/*K. blakistoni* (1.5%). Moreover, we detected a positive correlation between the yellow eye color and the homologous inserts in other species of studied birds. The *OCA2* gene alleles of the red-eyed and yellow-eyed specimens of *O. s. ussuriensis* were combined in one cluster on the reconstruction based on the maximum likelihood method (ML). A congeneric species *O. sunia*, with true

yellow eyes distanced significantly from the cluster of species characterized by red (*O. semitorques*) and orange (*B. bubo*) eyes on the phylogenetic reconstruction, which formed one clade. Two yellow-eyed but taxonomically distant species, *N. scandiaca* and *K. blakistoni*, were combined in one cluster. Yellow-eyed *A. funereus* is the most different from the other owls in terms of this trait. Such clustering demonstrates the non-identity of the OCA2 gene sequences in true yellow-eyed species and individual yellow-eyed birds in the population of *O. s. ussuriensis*, thereby indicating the different nature of their yellow-eye phenomenon.

It is not caused by a specific gene in *O. s. ussuriensis*, such as in *O. sunia*, but by the mutation of one of the alleles of the *OCA2* gene of the red-eyed form (AATT duplication), that is, it is an anomaly and therefore cannot be considered as a taxon-specific trait: both phenotypes definitely belong to the same genetic race. Said mutation probably may have affected the secondary structure and the normal functioning of the *P*-protein and limited the transport of melanin to melanosome leading to lighter (yellow) eye color.

PHYLOGENETIC RELATIONSHIPS WITHIN ECHINOCHASMIDAE ODHNER, 1910 (TREMATODA)

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Echinochasmidae Odhner 1910 includes trematoda species, adult worms of which are parasites of mammals, birds and reptiles. In 2016, Tkach et al. distinguished this new family and indicated main taxonomic problems within it to resolve. However, these authors have mentioned that there is no enough data to finalize the issues. In our study, the nucleotide sequences of both nuclear and mitochondrial DNA were obtained for the new Echinochasmidae representatives. These data are helpful for resolve problems of the validity and composition of genera in the family. In addition, it was found that this group of trematodes has features in secondary structure of *ITS2* rDNA region, which may also be an additional argument in clarifying phylogenetic relationships within the family.

Besides the question of the generic identification of the species, there is also the problem of the presence of cryptic species with recent ancestry in the family. For example, *Echinochasmus suifunensis* and *Echinochasmus milvi* have well resolvable differences in the morphology of all developmental stages. Biological features of their cercariae are also different. Despite this, the nucleotide sequences of the nuclear DNA (*ITS1-5.8S-ITS2-28S* rDNA region) were almost 100% identical for these species. At the same time, based on data of mitochondrial DNA (the *cox1* gene), it was obtained that these species have fixed nucleotide and amino acid substitutions.

Thereby, this study supplies new molecular data for this poorly studied group of parasites, and provides also the basis for more deep and comprehensive analysis of new issues in taxonomy of the Echinochasmidae family.

INSTINCTES AS AN OBJECT OF RESEARCH IN THE FIELD OF EVOLUTIONARY AND ECOLOGICAL GENETICS

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Instincts are species-specific and individual stereotypes of behavior that are organized on the basis of complex reflexes according to a genetic program. The most complex reflexions are formed by a sequence of complex reflexes, the completion of each of which is the beginning of the next. P.V. Simonov singled out three different classes of the most complicated unconditioned reflexes: 1) vital unconditional, 2) role (zoosocial), 3) unconditional reflexes of self-development. The second class of the most complicated unconditioned reflexes can be realized only by interacting with other individuals of the same species. These reflexes underlie sexual, parental, territorial behavior, and the formation of a group hierarchy.

Instinct is an evolutionarily worked out innate adaptive form of behavior inherent in a certain species of animal, representing a combination of complex reactions that occur in response to irritation. Konrad Lorenz believes that instincts should be understood as specific, strictly fixed actions (movements), the same in the same situations in all the presented species. Lorenz called instincts as "a complex of fixed actions" or stereotypical behavior. According to his ideas, under the action of a number of external and internal factors (hormones, temperature, light, etc.) in the corresponding nerve centers, an accumulation of "energy of action" occur specific to a certain drive (hunger, thirst, sexual need, etc.). The growth in this activity above a certain level leads to the appearance of the search phase of a behavioral act, which is characterized by a wide variation in performance, both in a given individual and in different representatives of the same species. It consists in the active search for irritants, under the action of which it is possible to satisfy the impulse that has arisen in an animal. When stimuli are found, the final act is carried out as a fixed complex of species-specific stereotyped movements, combined in one individual in each case, and in all individuals of a given species. This complex of movements is characterized by a high degree of genotypic conditioning.

Either way, instinctive reactions are innate. In the implementation of such complexes of instinctive fixed actions, the starting function is performed by external stimuli, which in their entirety create a starting situation. Each key-stimulus launches a corresponding set of programmed actions. Key-stimuli are environmental signs to which animals can react, regardless of their individual experience, with an innate behavioral act.

The study was conducted with the financial support of the Russian Foundation for Basic Research (Project 17-04-01321a).

THE RECENT DIVERGENCE OF TWO PRICKLEBACKS SPECIES (STICHAEIDAE: *STICHAEUS*) REVEALED BY THE DATA ON COMPLETE MITOCHONDRIAL GENOME

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The species *Stichaeus grigorjewi* and *S. nozawae*, typical representatives of the pricklebacks fishes, are found to have sharp discrepancies between morphological and molecular genetic characteristics. The nucleotide divergence between them, as revealed by common mitochondrial and nuclear molecular genetic markers, does not exceed the average values of intraspecific variability for most of eelpouts as well as other known fish. Along with the recent divergence, this may indicate the sophisticated features of the mitochondrial genome evolution in these taxa. To clarify the issue here we report on the sequencing, annotation and analysis of the complete mitochondrial genome sequences of these two species together with other eelpouts. The results of the analysis confirm the recent origin of *S. grigorjewi* and *S. nozawae*. In addition, within CR domain we found an insertion of 11 bp long marking the investigated representatives of *Stichaeus* genus, which may later shed light on the causes of the paraphyletic origin of the genus.

The study was partially supported by the grant of the President of the Russian Federation (M K-305.2019.4), as well as the FEB RAS program (project 18-4-040), and Russian Federal Government Finding for World-level Genome Centers.

TRANSPORT ACTIVITY IN THE *PSEUDOMONAS* NOSOCOMIAL ISOLATES FROM DIFFERENT CLONAL COMPLEXES

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Transport activity is very important for survival of bacterial pathogen in host. Acquisition and exclusion of metabolites and drags is regulated by transporters. Comparison of all transporters' pool in representatives of 3 clonal complexes (CC) of *P. aeruginosa* and one CC of *P. putida* was the goal of our investigation.

Two *P. aeruginosa* clinical isolates PA23 and PA169 of ST235 (2006 and 2010 year of isolation), one isolate PAKB6 of ST549 (2014), PA1840 of ST2592 (2015) and one *P. putida* clinical isolate PPKH115 of ST12 (2018) were sequenced by 454 Roche and/ or Illumina technologies and assembled by CLC Genomic Workbench v.12. RAST and other special servers were used for genomes annotation.

Four groups of transporters were analyzed in all genomes: porins, ABC, Tripartite ATP-Independent Periplasmic (TRAP) including TRAP associated extracytoplasmic immunogenic (TAXI), and Tripartite Tricarboxylate (TTT).

PA23 as the oldest representative of the nosocomial infection had the most number of porins, and PPKH115 had 20% less porins. PA ST235 and PPKH115 had alginate export system in contrast to PAKB6 and PA1840.

PPKH115 and PA ST235 had only one TTT, but PAKB6 and PA1840 revealed 2 complete and 2 incomplete TTT. The number of TRAP were more in PA genomes, than in PPKH115 one. The TRAP pool of PAKB6 and PA1840 included TAXI transporters, which were absent in PA ST235 and PPKH115.

The most difference in the transporter number was revealed in ABC group. The quantity of ABC transporters in PPKH115, PAKB6 and PA1840 was 6.5, 5.4 and 5.9 time higher than in PA ST235. Prolonged persistence of ST235 in the hospital led to a reduction in metabolic activity and to the loss of transporters of sugars, polyols, amino acids, oligo- and dipeptides, vitamins, most cations, efflux systems of antimicrobial peptides and antibiotics.

Only transporters of nickel and molybdenum as important for human pathogenic bacteria were detected in PA ST235 genomes. It should be noted that the number of nickel transporters was maximal in PPKH115 and amounted to 11.

Polyamines (spermidine, putrescine and etc.) have important role in protecting cells from physiological stress. The number of polyamines transporters was almost equal in all analyzed Pseudomonas genomes.

It is interesting that *P. aeruginosa* isolates distinguished the presence of gliding motility-associated ABC transporters, but the number of these transporters was two times more in PA ST235. These data suggested that this type of motility is important for pathogenic clone formation.

Thus, the study revealed a sharp reduction in ABC transporters among representatives of long-circulating *Pseudomonas* clonal complexes, but preservation and multiplication of transporters necessary for survival in the host.

COMPARATIVE ANALYSIS OF TRANSPORTERS IN MARINE AND FRESHWATER PROTEOBACTERIA

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Marinomonas primoryensis KMM3633 (MPKMM3633) was isolated Romanenko et al. (2003) from a coastal sea-ice sample, obtained from a sea-ice column at a depth of 0.8 m in Amursky Bay of the Sea of Japan, near Vladivostok in March 2001. Phenotypic characteristics of bacteria were analyzed for the new species registration in 2003. Whole genome sequencing (WGS) is very informative method for the prediction of metabolic and structural properties. Since Tripartite ATP-Independent Periplasmic (TRAP) including TRAP associated extracytoplasmic immunogenic (TAXI), and Tripartite Tricarboxylate (TTT) transporters were supposed to uptake to pathogenicity, the first goal of this research was revealing of these types of transporters in MPKMM3633. Porins and ABC transporters are necessary for environmental and host adaptation of bacteria, so the second task was to describe these pools of transporters too. At last we compared *M. primoryensis* transporters with WGS data of earlier sequenced in our laboratory clinical strains of other Gammaproteobacteria: *Pseudomonas aeruginosa, P. putida* and Betaproteobacteria: *Burkholderia cenocepacia*.

WGS of MPKMM3633 was performed using Illumina technology; data were assembled by CLC Genomic Workbench v.12. RAST and other special servers were used for genomes annotation. *P. aeruginosa* ST235, 549, 2592, *P. putida* ST12 and *B. cenocepacia* ST709 genomes were used in comparative analysis.

Bioinformatic analysis of genomic data revealed huge prevalence of TRAP transporters in MPKMM3633 in comparison with other Proteobacteria in sampling. 18 TRAP were C4-dicarboxylate specific and 7 TRAP had mannitol/chloroaromatic compound as substrate. 2 TRAP operon remnants presented only by solute receptor were revealed. 3 TAXI and 3 TTT transporters were found. In contrast other Gammaproteobacteria had 1-3 whole TRAP transporters, one whole TTT. TAXI was revealed only in ST549 and ST2592 genomes. *B. cenocepacia* had only 2 TRAP transporters.

ABC transporters in MPKMM3633 were almost equal (110) to these one in *P. aeruginosa* ST549 and 2592, but 3 times less than ABC transporters revealed in *B. cenocepacia*. It should be noted a lot of sugar and polyols ABC transporters; and transporter for osmoprotectant ectoine/hydroxyectoine in *M. primoryensis*.

But the most significant differences were found in the group of porins. If *Pseudomonas* had 22-27 porins (64-67% oprD), *B. cenocepacia* – 51 porins (92% ompC), then MPKMM3633 had only one porin of Porin_4 family.

Thus, investigation of marine bacteria genome demonstrated considerable differences in quantity and substrate specificity of transporters of different classes form freshwater Proteobacteria in our sampling.

DISCRIMINANT, CANONICAL AND ANOVA ANALYSES OF OTOLITH SHAPE MORPHOLOGY AND THEIR UTILITY FOR STOCK ASSESMENT AND IDENTIFICATION OF TWO SMELT SPECIES, *HYPOMESUS JAPONICUS* AND *H. NIPPONENSIS* (PISCES: OSMERIDAE) FROM THE NORTH-WESTERN SEA OF JAPAN

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There are many appropriate methods for studying a fish stock structure and species diversity. In the present paper, the analysis of otolith shape has been applied to identification of smelt stocks and two species, as was successfully done for many other fishes previously. In this study, four approaches (Shape indices, Elliptical Fourier descriptor, Elliptical Wavelet, and Regression functions) of otolith analysis have been used to determine differentiation of local populations, as well as two species, of the genus *Hypomesus*, which are most abundant and has a complicated taxonomic history in the north-western Sea of Japan. The main goal is to clarify the intra- and interspecies classification of smelt individuals (*H. japonicus* and *H. nipponensis*) based on the otolith shape analysis. The relationship between the basic otolith dimension parameters (area, perimeter, otolith width, otolith length, maximum feret, and minimum feret) and body measurements (standard length, head length, and head height) of two smelts (*H. japonicus* and *H. nipponensis*) is also in a focus of the research.

The Pearson correlation coefficient (r_p) and the coefficient of determination (R^2) ($P < 0.001$) showed for both species that the relationship between standard body length and otolith measurements is greater than the relationship between head length and otolith measurements. This study established a difference between otoliths of two species, *H. japonicus* and *H. nipponensis*. The obtained difference in otolith shape can be useful for taxonomy of these two species. Within the species *H. japonicus*, the individuals sampled from northern sample in Olga Bay (OB) are most similar morphologically to those from the coastal waters off the village of Terney. In contrast, the individuals of this species from the coastal waters off Russky Island (RI), including another southern sample and those from northern samples, manifest the greatest differences, thus providing evidence on the existence of at least two local stocks of the species in waters of Primorsky Krai.

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GENETIC CHARACTERIZATION OF FAR EASTERN HERMIT CRABS OF THE GENUS *PAGURUS* BY MITOCHONDRIAL 16S RRNA GENE PARTIAL SEQUENCES

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The family Paguridae is highly diverse, with species widely distributed through all oceans. In recent years, new genera have been added to almost all the families of Paguroidea, but most additions have been to the Paguridae, which is currently comprised of 74 genera. The genus *Pagurus* exhibits a high degree of species proliferation with 173 species currently assigned to it worldwide. Some of these species are closely allied to one another and can be assigned to any one of 11 informal species groups defined on the basis of morphological similarity. In some cases, morphology suggests very close relationships between members within these groups, and this raises the question as to their status as separate species. Molecular data are useful to resolve taxonomical questions at the species level in hermit crabs. Taxonomical re-examination of two South American hermit crab species of the "*comptus*" group (Group III, by Lemaitre and Cruz Castaño, 2004) was carried out successfully using partial sequences of the *16S* ribosomal RNA gene, rRNA (Mantelatto et al., 2009). Olguin and Montelatto (2013) presented the first molecular evidence validating some morphological groups. Three statistically stable clades were obtained using different phylogenetic algorithms that corresponded to three groups: "*provenzanoi*", "*comptus*" and "*exilis*". Nevertheless, there is a lack of molecular data for hermit crabs from the Japan Sea, which are not included in the species group list of Lemaitre and Cruz Castaño (2004). The common species for shallow waters of Japan and Russian Far East, *P. brachiomastus*, was described from a single specimen from Japan. The holotype of this species was lost so far and the neotype was designated. According to Komai (2000a), two additional morphologically distinct species (*P. proximus* and *P. simulans*), which were previously considered to be *P. brachiomastus*, inhabit the Japan Sea. Detailed morphological descriptions of these species have been previously provided by Komai, 2000a. Another species that is widely distributed along the Russian coast of the Japan Sea, *P. pectinatus*, also has ambiguous taxonomy. Detailed re-description, performed by Komai (2000b), suggested a taxonomical validity of this species. Another species, *P. middendorfii*, a common hermit crab of the shallow water of the Japan Sea, is considered a member of the "*comptus*" group. There are currently no reports about the distribution of *Pagurus* species of the "*comptus*" group in the Russian Far East. Membership of *P. middendorfii* in the "*comptus*" group therefore may be questioned. Our present study provides results on phylogenetic relationships of additional four far eastern *Pagurus* species: *P. minutus, P. parvispina, P. trigonocheirus* and *P. gracilipes*. Each of these species was placed into "*bernhardus*" group. Of these, *P. trigonocheirus* and *P. parvispina* were closely related to each other, *P. minutus* was a sister to [*P. trigonocheirus*/*P. parvispina*] *P. pubescens* subclade and *P. gracilipes* was as sister to *P. brachiomastus*/*P. proximus* subclade. In a whole, on *16S* rRNA based phylogenetic tree there were five clusters, corresponding to five morphological groups: "*bernhardus*", "*provenzanoi*", "*comptus*", "*forbesii*" and "*exilis*". Phylogenetic position of the "*forbesii*" group provided in the present study at the first time.

MORPHOLOGICAL VARIABILITY AND GENETIC DIVERSITY OF *ITS1-5.8S-ITS2* rDNA FRAGMENT OF THREE SPECIES OF THE GENUS *TRIBOLODON* (OSTEICHTHYES: CYPRINIDAE) FROM FAR EAST OF RUSSIA

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The *ITS1-5.8S-ITS2* rDNA fragment was used to assess whether species or different populations of the genus *Tribolodon* from the Primorskii region (Far East of Russia) could be identified using the secondary structure of *ITS2*. The molecular structure and diversity of the *ITS1-5.8S-ITS2* rDNA fragments, including the two intragenic spacers *ITS1* and *ITS2* were analyzed. For the first time inter- and intraspecific differences are shown for redfin of the genus *Tribolodon* using secondary structure of *ITS2*. The secondary structure of *ITS2* is significantly different for different species of the genus *Tribolodon*. Comparative morphological analysis of the same samples showed interspecific differences in the number of scales above and below the lateral lines of the body. Applying of number of scales in the lateral line of the body the similarity for individuals of different species (*T. hakonensis* and *T. brandtii*) from the Primorskii region and the similarity for individuals of different species (*T. hakonensis* and *T. brandtii*) from the Sakhalin island was found.

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