INTERNATIONAL SYMPOSIUM

MAPEEG-2015

PROGRAM &
ABSTRACTS

MAPEEG-2013
MAPEEG-2011

MODERN ACHIEVEMENTS IN POPULATION, EVOLUTIONARY AND ECOLOGICAL GENETICS

MAPEEG-2009
MAPEEG-2007
MAPEEG-2005

Conviner: Dr. Yuri Kartavtsev

MAPEEG-1998

VLADIVOSTOK & VOSTOK MBS

MAPEEG-1995

Modern Achievements in Population, Evolutionary, and Ecological Genetics : **International Symposium**, Vladivostok – Vostok Marine Biological Station, September 1–10, 2015 : Program & Abstracts. – Vladivostok, 2015. – 84 p. – Engl.
ISBN 978-5-7442-1563-7

HELD BY:
Far Eastern Branch of Russian Academy of Sciences,
A.V. Zhirmunsky Institute of Marine Biology FEB RAS,
Institute of Biology and Soil Science FEB RAS,
Far Eastern Federal University,
Administration of Nakhodka City District,
Vladivostok Public Foundation for Development of Genetics

SPONSORS:
Russian Foundation for Basic Research, Khimexpert Agency,
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Editors: Yuri Ph. Kartavtsev, Oleg N. Katugin

Современные достижения в популяционной, эволюционной и экологической генетике : Международный симпозиум, Владивосток – Морская биологическая станция «Восток», 1–10 сентября 2015 : Программа и тезисы докладов. – Владивосток, 2015. – 84 с. – Англ.
ISBN 978-5-7442-1563-7

ОРГАНИЗАТОРЫ:
Дальневосточное отделение РАН,
Институт биологии моря им. А.В. Жирмунского ДВО РАН,
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Администрация г. Находка,
Владивостокский общественный фонд развития генетики

ФИНАНСОВАЯ ПОДДЕРЖКА:
Российский фонд фундаментальных исследований,
ООО «Химэксперт», ООО «ГеноТек»,
ООО «СкайДжин»

Ответственные редакторы: Ю.Ф. Картавцев, О.Н. Катугин

PROGRAM

MAPEEG-2015 / MolSy-2015

Held by:
FAR EASTERN BRANCH OF RUSSIAN ACADEMY OF SCIENCES,
A.V. ZHIRMUNSKY INSTITUTE OF MARINE BIOLOGY FEB RAS,
INSTITUTE OF BIOLOGY AND SOIL SCIENCE FEB RAS,
FAR EASTERN FEDERAL UNIVERSITY,
ADMINISTRATION OF VLADIVOSTOK CITY,
ADMINISTRATION OF NAKHODKA CITY DISTRICT,
VLADIVOSTOK PUBLIC FOUNDATION FOR DEVELOPMENT OF GENETICS
Sponsors:
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KHIMEXPERT AGENCY, GENOTEK COMPANY, SKYGENE CO. RUSSIA

Organizing Committee:
Yu.Ph. Kartavtsev, S.D. (convener),
D.M. Atopkin, Ph.D., A.Y. Chichvarkhin, Ph.D., O.V. Chichvarkhina, I.V. Kartavtseva, S.D., O.N. Katugin, Ph.D., A.O. Katugina, S.V. Masalkova N.A., Redin A.D., Rozhkovan K.V., Sharina S.N., Shedko S.V., Sheremetieva I.N., Turanov, Ph.D., (members).

SUNDAY, AUGUST 31

Arrival, Hotel accommodation in Vladivostok

MONDAY, SEPTEMBER 1

MolSy Workshop/ School

8-30 **: Breakfast & Departure to the IMB + MBS Vostok**
11-30-12-30**: Registration & Accommodation**

12-30-13-30**: Lunch**

13-30-14-00**: Opening Remarks, MolSy-2015**

Andrey V. Adrianov, Director of the Institute of Marine Biology.
Maxim A. Melnikov, Ecology Department Head, Nakhodka City District Administration.
Yuri Ph. Kartavtsev, MAPEEG-2015 Convener.
Anton Yu. Chichvarkhin, MolSy-2015 Moderator.

14-00-19-00 **: Chair Person – S.D. Yuri Ph. Kartavtsev**

1. **Yuri Ph. Kartavtsev.** Practical training on nucleotide sequences: sequencing, edition, submission, alignment, tree building, analysis of

phylogeny and barcoding gap search (60').

2. **Anton Yu. Chichvarkhin**. Genomic insights into the marine invertebrates systematics and molecular Phylogenetics (60').

3. **Sergei V. Turanov**. DNA barcoding and molecular systematics of fishes (60').

4. ….(50').

5. .... (50').

#. General Discussion on Reports and First Day Activity (20').

19-30-20-30 **: Dinner**

SEPTEMBER 2-6

MolSy: Goals and Program (September 2-6, 2015)

Main goal of MolSy workshop is training for yang scientists of Russia in techniques for research in thr field of molecular systematics and phylogenetics. The aim is to start with collection of materials and finishing with usage of molecular genetic data in gene tree building, phylogeny inferences, and species identification through DNA barcoding.

The framework of the workshop aims to join in a team of an experienced researcher/teacher and few students (1-4 persons) to perform real small research in molecular systematics. In the interactive mode students will get knowledge training and outcome, a final product that could lately be transformed in a journal paper referring in WOS/Scopus science sources.

Participants of the workshop will get the following knowledge and skills:

- How to collect specimens and material in hydrobiology/ichtiology in general for DNA research? Participants will get opportunity to collect specimens, make conservation for current investigation.
- How correctly to keep, label, and make voucher records of specimens collected.
- How isolate DNA with different techniques.
- How label an appropriate section of a genome (primer design) for a research and using PCR reaction for gene amplification.
- How make DNA sequencing.
- How use GenBank and other database in a field of DNA barcoding or molecular phylogenetics.
- How to build gene (phylogenetic) trees with usage of DNA nucleotide sequences or amino acid sequences by modern techniques.

SUNDAY, SEPTEMBER 6

8-30-9-00 **: Breakfast & Departure from MolSy to the IMB**
12-30 : **Arrival and Hotel Accommodation All the Participants**

MONDAY, SEPTEMBER 7

9-30-10-00**: Registration**

10-00-10-15**: Opening MAPEEG-2015, Greetings**

I.S. Pushkarev, Head of Administration of Vladivostok City.
A.V. Adrianov, Director A.V. Zhirmunsky Institute of Marine Bology; Director School of Natural Sciences, Far Eastern Federal University.
Yu.Ph. Kartavtsev, Convener MAPEEG-2015.

Session 1. Evolutionary Genetics & Genomics
(Oral Presentations)

10-15-12-30**: Chair Person – Academician RAS, S.D. Vladimir N. Bolshakov**

1. **Kartavtsev Yuri Ph.** Gene introgression between species and gene tree reticulations: are these events challenged Neo-Darwinism and DNA barcoding**?** (30').
2. **Stegniy Vladimir N.** The chromosome's spatial organization, heterochromatin and speciation (30').

Coffee Break (10')

3. **Amstislavsky Sergey Ya., Kizilova E.A., Abramova T.O., Brusentsev E.Yu., Rozhkova I.N., Igonina T.N., Danilenko A.V., Naidenko S.V.** Cryobanking of Far East cat (*Prionailurus bengalensis euptilurus*): freezing and cryopreservation of semen (20').
4. **Antosyuk Olga, Marvin N.A.** The analysis of change of wing`s spatial structure of the Drosophila melanogaster as a method in evolutionary and ecological genetics (20').
5. **Sergey Turanov, Lee Y-H, Kartavtsev Yu.Ph.** Structural features and variation of mitochondrial control region among eelpouts (Cottoidei: Zoarcales) (20').

Lunch (30')

13-00-14-30**: Chair Person – S.D., RAS corresponding member Vladimir D. Bogdanov**

6. **Hanzawa Naoto.** Evolutionary aspects of Ctenophore and Cnidaria genomes and the jellyfishes (30').
7. **Nedoluzhko Artem V., Sokolov A.S., Sharko F.S., Boulygina E.S., Tsygankova S.V., Tikhonov A.N., Skryabin K.G., Prokhortchouk E.B.** Whole genome sequencing of the wooly mammoth tracks its evolution history (20').
8. **Bazhanov Dmitry P.**, **Li H., Li Ch., Li J., Bazhanova A.A., Yang. H.** Phylogenetic diversity of atrazine-degrading bacteria in Shandong province, P.R. China (20').
9. **Ragaeva Diana**, **Abramova T.O., Igonina T.N., Rozhkova I.N., Brusentsev E.Yu., Kalinichenko E.V., Amstislavsky S.Ya.** Effects of embryo transfer and in vitro culture on the development of hypertensive phenotype in rats genetically predisposed to hypertension (ISIAH strain) (20').

Coffee Break (10')

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

14-40-16-10**: Chair Person – S.D. Vladimir N. Stegniy**

10. **Sherbakov Dmitry Yu.**, **Kovalenkova M.V., Petunina J.V., Pudovkina T.A., Peretolchina T.E.** Paradoxes of evolutionary histories among members of Baikal species flocks (30').
11. **Romanova Elena**, **Aleoshin V.V., Mikhailov K.V., Logacheva M.D., Kamaltynov R.M., Sherbakov D.Yu.** Phylogeny of baikalian endemic amphipods based on sequences of their mitochondrial genomes (20').
12. **Chichvarkhin A.Yu., Chichvarkhina Olga V., Ekimova I.** Ressurection of *Placida babai* and other implementations of DNA barcoding approach in Opisthobranch systematics (Mollusca, Gastropoda) (20').
13. **Rozhkovan Konstantin V.**, **Shedko M.B.** Phylogenetic relationships of Paradiclybothrium pacificum and Diclybothrium armatum (Monogenoidea: Diclybothriidae) inferred from 18S rDNA sequence data (20').

Coffee Break (10')

16-20-18-20**: Chair Person – S.D. Dmitry Yu. Sherbakov**

14. **Kolbasova G.D., Zalevsky A.O., Gafurov A.R., Gusev P.O., Ezhova M.A., Zheludkevich A.A., Konovalova O.P., Kotlov N.U., Lanina N.O., Lapashina A.S., Medvedev D.O., Nosikova K.S., Nuzhdina E.O., Bazykin G.A., Neretina Tatyana V.** Sixteenth-eyed Lion mane – A new Cyanea species from the White Sea (20').
15. **Konovalova Olga, Bubnova E.** Study of Arctic marine fungi as example of necesserity of barcoding (20').
16. **Chichvarkhin A.Yu., Chichvarkhina O.V., Katugina A.O., Katugin Oleg N.** Molecular barcoding of Gonatid squeeds in Northwestern Pacific (20').

Coffee Break (10')

17. **Peretolchina Tatiana E., Sitnikova T.Ya., Sherbakov D.Yu.** Microevolutionary processes in endemic gastropods of genus *Baicalia* (20').
18. **Yuri Ph. Kartavtsev, Natalia M. Batishcheva, Nina G. Bogutskaya, Anna O. Katugina, Naoto Hanzawa.** Molecular systematics and DNA barcoding of Altai osmans, *Oreoleuciscus* (Pisces, Cyprinidae, Leuciscinae), and nearest relatives, as revaled by sequences of cytochrome *b* (*Cyt-b*), cytochrome oxidase *c* (*Co-1*), and complete mitochondrial genome (20').
19. **Kovalenkova Maria V., Petunina J.V., Sherbakov D.Yu**. Nuclear and mitochondrial polymorphism in baikalian *Gmelinoides fasciatus* (20').

#. Discussion on Reports (10').

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

TUESDAY, SEPTEMBER 8

8-30 - 9-00**: Breakfast**
10-00-12-00**: Receipt by the Mayor of Vladivostok City**
13-00-14-00**: Lunch**

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

14-00-15-00**: Chair Person – Ph.D. John Eimes**
20. **Shedko Sergei V.** Mitochondrial DNA sequence variation and demographic history of kaluga and Amur sturgeon (Acipenseridae): a comparison (20').
21. **Yurij Bukin**. Eestimates of convergence the results of population-

genetic analysis based on the DNA sequences sample (20').
22. **Katugina Anna O.**, **Kartavtsev Y. Ph.** Comparative analysis of 3 species of Far Eastern Dace of genus *Tribolodon* based on some morphological and genetic markers (20').

15-00-16-00**: Chair Person – S.D. Aleksander B. Mulik**

23. **Masalkova Natalia A.**, **Kartavtsev Y.Ph., Katolikova M.V., Sharina S.N., Chichvarkhina O.V.** Genetic and Morphometric Variability and Hybridization of Two Species of Mussels, *Mytilus trossulus* and *Mytilus galloprovincialis*, in the North-West of Sea of Japan (20').
24. **Ekimova Irina**, **Chichvarkhina O., Schepetov D., Chichvarkhin A., Martynov A.V..** DNA barcoding Dendronotid mollusks of Russian seas (20').
25. **Shumenko Polina G.**, **Tatonova Y.V., Besprozvannykh V.V.** Molecular characterization of *Metagonimus yokogawai* (Trematoda: Heterophyidae) in the Russian southern Far East (20').

Coffee Break (10')

16-10-17-20: Chair Person – **S.D. Aleksandr V. Borodin**

26. **O'Brien Stephen.** A Moving Landscape for Comparative Genomics in Mammals (30').
27. **Eimes John,** **Lee S.-I, Jablonski P., Didinger C., Townsend A., Nishiumi I., Satta Y.** Towards explaining the unusually complex MHC of the Passeriformes (20').
28. **Tatonova Yulia V.**, **Besprozvannykh V.V., Hung N.M., Ngo H.D.** Genetic diversity of Clonorchis sinensis (Trematoda: Opisthorchiidae) from Russia and Vietnam using IRAP (20').

Coffee Break (10')

17-30-18-50: Chair Person – **Ph.D. Naoto Hanzawa**

29. **Roslik Galina V.**, **Kartavtseva I.V.** Diversity of *Apodemus peninsulae* (Rodentia) B chromosomes DNA (20').
30. **Yalkovskaya Lidia E.**, **Sibiryakov P.A., Netunaeva O.N.** Chromosomal and mitochondrial DNA variability in the striped field mouse (*Apodemus agrarius* Pall., 1771, Muridae, Rodentia) in the Urals (20').
31. **Pavlenko Marina V.**, **Korablev V.P., Tsvirka M.V**., **Puzachenko Andrey Yu.** Genetic and morphological differentiation and systematics of

North China Zokor *Myospalax psilurus* (Rodentia, Spalacidae) (20').
32. **Kartavtseva Irina V., Rubtsov N.B., Karamysheva T.V.** Pericentromeric heterohromatine in *Apodemus* genus (Mammalia, Rodentia) (20').

#. Discussion on Reports (10').

19-00-19-30**: Dinner**

WEDNESDAY, SEPTEMBER 9

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

10-00-12-00**: Chair Person – Ph.D. Sergei V. Shedko**

33. **Smirnova Maria, Orlova S., Mugue N., Orlov A.** Population structure of Pacific cod in the Sea of Okhotsk (20').
34. **Skazina Maria A., Genelt-Yanovskiy E.A., Katolikova M.V., Nazarova S.A., Gagarina A.A., Strelkov P.P.** New data on hybridization in Scottish blue mussels enable comparison among known hybrid zones between *Mytilus edulis L.* and *M. trossulus* Gould (20').
35. **Grizanova Ekaterina, Dubovskiy I., Glupov V., Butt T.** Some resistance mechanisms of *Galleria mellonella* (Lepidoptera, Pyralidae) larvae selected for resistance to *Bacillus thuringiensis* in laboratory (20').
36. **Sibiryakov Petr A., Markova E.A.** Variability of mtDNA in *Microtus arvalis obscurus*: new data and prospects for future research (20').

Coffee Break (10')

11-30-13-10 **: Chair Person – Ph.D. Stephen O'Brian**.

37. **Kryukov Alexey, Spiridonova L., Mori S., Lobkov E., Redkin Y., Haring E.** Geographic isolation and phylogeography of the magpie *Pica pica* (20').
38. **Padyukova Asiya D., Lavryashina M.B., Skhalyakho R.A., Agdzhoyan A.T., Balanovskaya V.** Polymorphisms of Y-chromosome in populations of Tumen-Turin and Tobolo-Irtysh Tatars in Tumenskaya oblast (20').
39. **Homel Konstantin V., Kheidorova E.E., Nikiforof M.E., Shpak A.V.** Intraspecific genetic differentiation of *Haematopus ostralegus* Linnaeus,

1758 on the territory of Belarus (20').
40. **Atopkin Dmitri M., Beloded A.Yu., Ngo H.D.** Molecular systematics and phylogeny of the trematodes of mugilid fishes in the Far Eastern Region
41. **Pavlova Svetlana.** Chromosomal hybrid zones of the common shrew in Russia (20').

13-10-14-30**: Lunch**

14-30-16-10 **: Chair Person** – **S.D. Sergey Knyazev**

42. **Dudnikov Alexander Ju.** Intraspecies divergence of *Aegilops tauschii*: investigating an example of intermediate stage of adaptive speciation (20').
43. **Protopopova Marina V., Pavlichenko V.V., Chepinoga V.V.** Some aspects of genetic polymorphism in two relict plant species from Baikal Siberia (20').
44. **Kalinkina Valentina A., Mikhaylova Y. V.** The molecular phylogeny of the genus *Trifolium* L. section Lupinaster (Fabr.) Ser. (Fabaceae) (20').
45. **Volkov Ilya. KhimExpert Agency Lecture.** News on equipment and reagents distributed by KhimExpert Agency in Russia. NGS, the high-speed sequencing (30').
46. **GenoTek Co Lecture.** Application of next-generation sequencing technologies in population genetics research (30').

#. Discussion on Reports (10').

Session 4. Poster Presentations (30')

16-20-17-20 **: Chair Persons – S.D. Irina V. Kartavtseva & Ph.D. Konstantine V. Rozhkovan**

Section 1. Molecular Systematics, Barcoding, Phylogenetics and Theoretical Genetics

1. **Anahit Atoyants, Aghajanyan E.A., Avalyan R.E., Atoyants A.L., Hovhanisyan G.G., Aroutiounian R.M.** Detection of genotoxic and clastogenic effects of soils pollution in Armenia with application of Tradescantia (clone 02) test system.
2. **Kartavtsev Yu.Ph., Sharina S.N., Saitoh K., Imoto J., Hanzawa N., Redin A.D.** Molecular phylogeny of Russian Far Eastern flounders (Pleuronectiformes, Pleuronectidae) based on mitochondrial sequences for genes, *Co-1* and *Cyt-b*, and complete mitogenome.

3. **Voronova Anastasia N., Chelomina G.N., Besprozvannykh V.V.** Restitution and genetic differentiation of *Nanophyetus* populations.
4. **Spiridonova Lyudmila N., Valchuk O.P.** NUMTs as a source of new mtDNA haplotypes: Siberian rubythroat *Luscinia calliope* (Muscicapidae, Aves) case.
5. **Polityko Yulia K., Sibirykov P.A., Bogdanov V.D., Koporikov A.R., Borodin A.V.** The Arctic char (*Salvelinus alpinus*) from the Polar Urals and its position in the species' phylogeographic structure.
6. **Kalinichenko Ekaterina V., Ragaeva D.S., Rozhkova I.N., Igonina T.N., Danilenko A.V., Brusentsev E.Yu., Abramova T.O., Amstislavsky S.Ya.** Effects of embryo transfer and in vitro culture on characteristic behavior in genetically catatonic rats (GC strain).
7. **Solodovnik Darya A., Tatonova Y.V., Besprozvannykh V.V.** Molecular adaptation of *Clonorchis sinensis* (Trematoda: Opisthorchiidae) from rat's liver and intestine.
8. **Zhukova Alina.** The phylogeny of *Leucochloridium sp*. Trematodes.
9. **Prokhovova Elena E., Zhemchuzhnikova E.A.** Genetic variability between populations of *Planorbarius corneus* snails (Gastropoda, Pulmonata).
10. **Sirotinina Elena A., Sherbakov D.Yu.** Mechanism of mitochondrial genome simplification in evolution.
11. **Sokolov Alexey S., Nedoluzhko A.V., Sharko F.S., Boulygina E.S., Tsygankova S.V., Polilov A.A., Prokhortchouk E.B., Skryabin K.G.** De-novo assembly of mitochondrial genome of the smallest insect *Megaphragma amalphitanum* (Hymenoptera: Trichogrammatidae).
12. **Rastorguyev Sergey M., Nedoluzhko A.V., Gruzdeva N., Prohorchuk E.B.** MicroRNA differential expression of marine and freshwater forms of the three-spined stickleback (*Gasterosteus aculeatus*).

Section 2. Microevolution. Population Genetic Structure of Species. Ecological Genetics. Animal & Plant Genetics

13. **Krivenko D.A., Kulakova N.V., Verkhozina A.V., Kazanovsky S.G., Dymshakova O.S., Kotseruba Violetta V.** Karyological and molecular genetic differentiation *Chelidonium majus* L.: to the question of cryptic species in flowering plants.
14. **Kinoshita Gohta, Nunome M., Kryukov A.P., Kartavtseva I.V., Han S.-H., Yamada F., Suzuki H.** Reticulate evolution of the genus *Lepus* in northeastern Asia.
15. **Dymshakova Olga S., Krivenko D.A., Zhmud' Y.V.** Molecular genetic differentiation of species Astragalus section Cenantrum (Fabaceae).

16. **Polezhaeva Maria A., Tikhonova N.A., Pimenova E.A.** cpDNA diversity of some species of *Rhododendron* from Siberia and the Far East of Russia.
17. **Chaika Marina, Sheremetyeva I., Zhuravlev Yu.** On the variability in mtDNA of *Ursus thibetanus* on south of the Far East of Russia.
18. **Gorobeyko Ulyana V., Kartavtseva I.V.** Chromosomal variability of Far Eastern *Myotis petax*.
19. **Voronina O.L., Ryzhova N.N., Kunda M.S., Aksenova Ekaterina I., Zaytseva E.A., Adgamov R.R., Semenov A.N., Ermolaeva S.A., Gintsburg A.L.** Application of ecological genetics for surveillance of *Listeria monocytogenes*.
20. **Rakitin Sergey B., Bolshakov V.N., Borodin A.V.** Intraspecies genetic differentiation of red vole populations inhabiting the Urals based on mtDNA.
21. **Nazarova G.G., Knyazev Sergey P., Nikitin S.V.** A comparative study of genetic basis of birth weight in Water vole and domestic pig.
22. **Zhuykova Elena, Kiselyova I.** Molecular genetic characteristics of *Plantago media* l. subpopulations from the environs of kachkanarsky ore mining and processing enterprise.
23. **Ponomareva Ekaterina V., Volkov A.A., Ponomareva M.V., Shubina E.A.** Characteristics of European grayling (*Thymallus thymallus*) of Kuloy and Northern Dvina rivers (White Sea basin), based on mitochondrial DNA fragments analyses.
24. **Veiko Natalia, Lyapunova N.A., Porokhovnik E.N., Kostyuk S.V.** Copy number variation of the human ribosomal genes in health and disease.
25. **Vakurin Alexey, Pavlenko M.V.** In search of reproductive isolation between *Cricetulus barabensis* and *C. griseus*: hybridization and cytogenetic analysis.
26. **Pilganchuk Oksana A., Kositzyna A.I., Shpigalskaya N.Yu., Savenkov V.A.** Population genetic structure of sockeye salmon, *Oncorhynchus nerka* (Walbaum), Kamchatka Peninsula, Chukotka Peninsula and the Kuril Islands.
27. **Sheremetyeva Irina N.** The role of the isolates in shaping phylogeographical structure of *Microtus fortis*.
28. **Shatyr Yu., Bondarev A., Mulik Alexander.** Genetic background of variability in the phenotypic expression of nonspecific reactivity of an organism in the indigenous groups of various territories in the Volgograd region, Russia.
29. **Chertilina Olga.** Phylogenetic relationships of the mountain voles (*Articola*) inferred from cytochrome *b* sequences.
30. **Dymshakova O.S., Cherepanova Olga E.** Population genetic variability of *Calluna vulgaris* (L.) Hull.
31. **Frolov S.V.** Is Capelin one species? Karyotype analysis.

32. **Chernysheva Nadezhda U., Medvedev A.U., Romashko D.A., Stenkova A.M., Nedashkovskaya O.I., Isaeva M.P.** Draft genome sequence of *Vitellibacter vladivostokensis* harbors biodegrading enzyme genes.
33. **Kharchenko Victoria, Koksheeva I.** Transformation of architecture inflorescences in *Arabidopsis thaliana*.
34. **Triboy Tatiana I.**, **Nepokrytykh A., Sherbakov D.Yu.** Evolutionary history of endemic Trichoptera from Lake Baikal.
35. **KhimExpert Co.** Posters & exhibition by Main Sponsor.
36. **GenoTek Co.** Posters & exhibition by Main Sponsor.
37. **SkyGene Co.** Posters & exhibition by Regular Sponsor.

THURSDAY, SEPTEMBER 10

8-30 - 9-00 **: Breakfast**
9-00 - 19-00**: Free day, Excursions to Vostok Bay & Site Seeing around Vladivostok**
19-00-23-00**: Dinner, Closing Reception & Evening party**

FRIDAY, SEPTEMBER 11

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

ABSTRACTS

CRYOBANKING OF FAR EAST CAT (*PRIONAILURUS BENGALENSIS EUPTILURUS*): FREEZING AND CRYOPRESERVATION OF SEMEN

[1]Sergei Ya. Amstislavsky, [1]Elena A. Kizilova, [1]Tatyana O. Abramova, [1]Eugeny Yu. Brusentsev, [1]Irina N. Rozhkova, [1]Tatyana N. Igonina, [1]Alexey V., Danilenko, [2]Sergei V. Naidenko

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Moscow, 119334, (Russia)

Leopard Cat (*Prionailurus bengalensis euptilurus*) is existing in the wild in the Russian Far East and there is small farm population bred in captivity in Tschernogolovka (Russia). The aim of this study was to freeze and cryopreserve semen in this *Felidae* subspecies. Sperm was obtained from four captive bred tomcats on the Tchernogolovka biological station by the electroejaculation under general anaesthesia. Ejaculates were collected with P-T Electronics (Oregon, USA) electroejaculator. Semen was diluted and incubated for two hours at +4^{0}C with CaniPlus Chill (Minitube, Germany) supplemented with egg yolk and with glycerol (4%). The samples of semen were frozen in 0.25 mL straws (Cryo Bio System, France). Straws were placed horizontally 2 centimeters above the liquid nitrogen surface for 7 min before plunging into liquid nitrogen. Straws were transported to Novosibirsk in frozen state, thawed (10 sec 24^{o}C, then 30 sec 37^{o}C) and analyzed with two methods: (1) by double staining with SYBR Green and PI with the subsequent confocal microscopy and (2) with Hamilton-Thorne Ceros 12.1 computer-assisted sperm analyzer. The first method revealed that an average 42.3% of spermatozoa retained their viability after freezing and thawing. The second method confirmed that an average 32.5% of spermatozoa were motile after thawing. CaniPlus Chill was initially designed for dog semen chilling. In our preliminary experiments we demonstrated its applicability for domestic cat semen freezing. The present study represents successful attempt of cryopreservation in *Prionailurus bengalensis euptilurus* and demonstrates the applicability of the chosen cryopreservation protocol for *Felidae*.

This investigation supported by the Russian Foundation for Basic Research (Project 15-04-03258).

THE ANALYSIS OF CHANGE OF WING'S SPATIAL STRUCTURE OF THE DROSOPHILA MELANOGASTER AS A METHOD IN EVOLUTIONARY AND ECOLOGICAL GENETICS

Olga N. Antosyuk

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Drosophila is convenient model object for the majority of researches in the field of evolutionary and ecological genetics. Changing of spatial structure of a wing of the drosophila as the sensitive parameter, which demonstrate to change of both the external, and internal environment was used. The morphometric analysis of a wing, which includes 18 linear and 6 squares parameters of a wing of the drosophila is used.

Using the morphometric analysis of a wing we can determine more sensitive and resistant strains to influence of various factors of a stress by 24 indicators, for example a methotrexate, an aminopterin or ionizing radiation. It was shown that strains can be resistant against action of one factor and are extremely sensitive to action of another factor.

The emergence of a mutation of scalloped of the strain, which received from crossing of individuals of Bar and vestigial which underwent the long directed selection by the morphometric analysis, successfully established. Hybrid individuals of vestigial contain heterozygote vestigial individuals and individuals whose phenotypes didn't differ from heterozygotes. Using morphometric analysis it is found, that nature of damages on a wing differed in both analyzed cases. Concordantly, it is established that the individuals allocated in this strain and that defined as scalloped phenotypes, had the changes in scalloped gene causing change of a product of a gene. Also found, that even when absence of damage is observed on a wing like "cutting", for example after influence mitomicin-C or etoposide, usage of morphometric analysis of a wings helps to determined changes of spatial structure of a wings after drugs impact. Different regions of a wing can change in various degree depending on what factor of a stress influences the change.

Thus, change of spatial structure of wings of Drosophila is a convenient method in evolutionary and ecological genetics. Wings imaginal discs allowed to estimate impact on somatic cages, and also indirectly the level of activity of transposons.

MOLECULAR SYSTEMATICS AND PHYLOGENY OF THE TREMATODES OF MUGILID FISHES IN THE FAR EASTERN REGION: TWO EXAMPLES FOR DIGENEAN INTRASPECIFIC GENETIC DIFFERENTIATION

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Present communication is about molecular genetic investigations of some trematode species that infected mugilid fishes of Russian Far East and Vietnam. We provide here molecular data on two species, *Lecithaster mugilis* Yamaguti, 1970 and *Skrjabinolecithum spasskii* Belous, 1954.

Nucleotide sequences of complete 18S rRNA gene and partial 28S rRNA gene were obtained for 9 specimens of *Lecithaster mugilis* from Vietnam (n=6) and Primorye Region (n=3). Comparative analysis of nucleotide sequences of *L. mugilis* from Vietnam and Primorye showed 40 (1.9%) and 28 (2.6%) variable sites for 18S rRNA and 28S rRNA genes, respectively. Genetic differentiation between *L. migilid* from Vietnam and Primorye with *L. gibbosus* was 5.4% and 5.5%, respectively for 18S rDNA gene sequence data and 10 and 10.3% for 28S rRNA gene sequence data. Phylogenetic relationship based on combined ribosomal gene sequence data showed two phylogenetic lineages of *L. mugilis*, which corresponded to specimens, collected in Primorye Region and Vietnam. Species *L. gibbosus* appears as sister species to *L. mugilis*.

Nucleotide sequences of the 28S rRNA gene and ITS1-5.8S-ITS2 rDNA fragment were obtained for 27 specimens of the trematode *Skrjabinolecithum spasskii* collected from mugilid fishes *Liza haematoheila* and *Mugil cephalus* from different rivers of the Primorye Region and Vietnam. These data were used for estimation of genetic differentiation and phylogenetic reconstructions of *S. spasskii*. Variation of ribosomal DNA fragments was revealed between specimens of *S. spasskii* from Russian Far East and Vietnam. There were 3 sequence variants (=genotypes) of *S. spasskii*. The first genotype occurred in specimens from *L. haematocheila* and *M. cephalus* collected from different rivers, including the type location for *S. spasskii* – Razdolnaya River of the Russian Primorye Region. The second genotype was revealed for specimens of *S. spasskii* from *M. cephalus* of the Cat Ba Island, Vietnam. The third genotype was found for 3 trematode specimens collected from *L. haematochelia* of the Kievka River, Primorye. Differentiation between genotypes I and III from Primorye Region was 0.4% and 0.65% by 28S rDNA and ITS rDNA, respectively. A highest identity between Vietnamese genotype II and genotype III was observed: 0.1% and 0.33% by 28S rDNA and ITS rDNA, respectively. Phylogenetic tree topologies, generated by combined 28S rDNA and ITS rDNA sequence data indicated intraspecific differentiation of *S. spasskii* into three well supported groups, representing 3 genotypes. A possible factors of genetic differentiation of *S. spasskii* are discussed.

PHYLOGENETIC DIVERSITY OF ATRAZINE-DEGRADING BACTERIA IN INDUSTRIAL AND AGRICULTURAL SOILS OF SHANDONG PROVINCE

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First registered in Switzerland and the US in 1958, atrazine has soon become one of the world's best-selling and heavily applied herbicides. As a result, atrazine and its stable metabolites nowadays are commonly detected contaminants of soils, surface and underground streams and basins. Even in low, ecologically relevant concentrations atrazine causes endocrine disturbance in many species of fish, amphibians and reptiles. Some bacteria can rapidly degrade atrazine through its dechlorination and further conversion to cyanuric acid readily mineralized in soil. The enhanced degradation substantially decreases the harmful consequences of atrazine application by reducing both its conversion to dealkylated metabolites and the leaching of atrazine and its metabolites to deep soil horizons. The diversity of atrazine degraders is poorly characterized because their isolation requires prior enrichment that eliminates many members of atrazine-degrading microbial communities. We isolated 116 atrazine degrading strains from geographically distant industrial and agricultural sites by direct plating of soil dilutions on the specially developed selective agar SM. ERIC-PCR genotyping of the isolates followed by 16S rRNA gene phylogenetic analysis gave evidence that the soils harbored communities of genetically diverse atrazine-degrading bacteria. Genetic structures of the communities in soils differentially exposed to atrazine did not overlap, indicating intensive selection of bacteria better adapted to local environments. However, the taxonomic diversity of atrazine degraders was limited to a variety of *Arthrobacter* spp. and few phylotypes within the genera *Nocardioides*, *Pseudomonas* and *Gulosibacter*, suggesting that the individual atrazine utilization is a trait of soil bacteria belonging to a limited number of lineages. In contrast to the abundance of the diverse *Arthrobacter* spp. in industrial soils, genetically similar *A. ureafaciens* bacteria were dominating atrazine degraders and the only found atrazine degrading representatives of the genus in the maize rhizosphere at all the agricultural sampling sites. The genetic structure of atrazine degrading communities of industrial soils appeared to be governed mainly by the rate of the contamination. More complex factors seem to influence the survival of atrazine degraders in agricultural soils which normally are exposed to low rates of atrazine once a year. The contribution of *A. ureafaciens* bacteria to the enhanced degradation of atrazine in agricultural soils and mechanisms causing their wide spreading and prevalence among atrazine degraders in the maize rhizosphere deserve further study.

This work was supported by the 1,000 Talents Plan, Science Foundation for Distinguished Young Scholars of Shandong Province (BS2014SW030) and Youth Foundation of Shandong Academy of Sciences.

ESTIMATES OF CONVERGENCE THE RESULTS OF POPULATION-GENETIC ANALYSIS BASED ON THE DNA SEQUENCES SAMPLE

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When we conduct population genetic research it is often raise the question of the necessary and sufficient data to obtain reliable results. Often to assess genetic diversity in populations using DNA sequences. On the basis of samples of DNA sequences calculated various population genetic criterions and parameters. For example such characteristics is the criterion of nucleotide diversity *Theta Pi* determining the genetic variability inside populations and the criterion *Fst*, that determine the genetic variability between populations or the degree of genetic isolation between the populations. All of the above criteria are calculated on the basis of genetic distances obtained by pairwise comparison of the DNA sequences of data samples. The main characteristic of the genetic variability in this case will be the average value of genetic distance between organisms population. The average genetic distance always will be defined with some statistical error since it is based on a limited sample of DNA sequences, taken from the general population.

The main task, which was implemented in the course of the work is the building of an algorithm implemented in the form of functions written in the programming language R, assessing the statistical error and confidence interval which is defined with average genetic distance between individuals in a population.

The algorithm of functions is based on bootstrap analysis and other theoretical methods. The functions can be easily integrated into scripts for handling large array of data, written as R language and other programming languages.

The results of testing algorithms coincide with the theoretical and experimental concepts published in other works.

POPULATION GENETIC VARIABILITY OF *CALLUNA VULGARIS* (L.) HULL

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Geographic variation and differentiation of the chloroplast DNA haplotypes, allozymes and morpho-anatomical leaf parameters were assessed in a number of certain European and Zauralye groups of *Calluna vulgaris* (L.) Hull populations, which have long been isolated from each other. Sharply pronounced genetic and phenotypic differences between the studied population groups were revealed. Genetic distances were many times higher than those between the relatively homogeneous European groups. The data obtained generally supported the hypothesis on the phenogenetic divergence of the Zauralye populations of *C. vulgaris*.

The reported study was supported by RFBR, research project No. 15-12-4-13.

DRAFT GENOME SEQUENCE OF *VITELLIBACTER VLADIVOSTOKENSIS* HARBORS BIODEGRADING ENZYME GENES

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Marine microorganisms are known to produce a number of bioactive primary and secondary metabolites, whose synthesis is determined by occupied ecological niche and symbiotic relationships. Recently, members of the phylum *Bacteroidetes* are considered as specific destructors of the both biological and synthetic macromolecules. They inhabit a marine environment with high levels of organic compounds and are often associated with algae and marine invertebrates. The family *Flavobacteriaceae* is one of the major branches of the phylum *Bacteroidetes* and has a significant role in ocean bioremediation processes.

Here we present the draft genome of the type strain *Vitellibacter vladivostokensis* KMM 3516^T (Nedashkovskaya et al., 2003), isolated from gut of the holothurian *Apostichopus japonicus* in 1997 (Troitsa Bay, Sea of Japan). The genome sequencing was conducted using the Roche 454 Titanium pyrosequencing shotgun technology on GS Junior instrument. Reads were assembled with Newbler v3.0 software. Automated annotation was performed using the RAST annotation server (Aziz et al., 2008) and manually by BLAST search of nonredundant protein sequences from the NCBI, Swiss-Prot, COG and KEGG databases. Additionally, genome was analyzed for identifying extracellular hydrolytic enzymes using CAZy and MEROPS databases, as well as SignalP v3.0 software to predict signal peptides.

The draft genome was assembled *de novo* in 25 contigs (N50=176,552 bp) with 24× coverage depth and evaluated to 3.3 Mbp size with GC% = 40.8. The RAST annotation server has identified 3,195 coding sequences and 37 RNAs genes. A total of 1,792 genes were assigned as encoding non-hypothetical proteins and 987 genes were categorized into COGs functional groups (including putative or hypothetical genes). The bacterium harbors 150 proteases and 49 glycoside hydrolases, 31 and 4 of which, respectively, are an exo-enzymes that may point out their ecological role of organic compounds decomposition, nutrient recycling and bioremediation processes in the ocean, as well as for potential application in biotechnology and industry.

PHYLOGENETIC RELATIONSHIPS OF THE MOUNTIAN VOLES (ALTICOLA) INFERRED FROM CYTOCHROME B SEQUENCES

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The analyzed 17 samples of five species of rocky voles: *Alticola semicanus*, *A. tuvinicus, A. olchonensis, A. strelzowi* and *A. barakschin*, collected in different areas: Northern Kazakhstan, South-Eastern Altai, Southwest Tuva, Republic Khakassia, the Baikal region and northern Mongolia.

On the basis of variability of a fragment of the cytochrome *b* gene (783 bp) phylogenetic tree reflecting the relationship of nine species of the genus *Alticola* was obtained. According to the reconstruction, the genus is divided into two large groups, first of which includes 5 species: *A. semicanus, A. tuvinicus, A. olchonensis, A. strelzowi* and *A. barakschin*. While the second consist of four, *A. montosa, A. argentatus, A. albicaudus, and A. lemminus*. There are two distinct clades inside the first group. The first branch consists of *A. barakschin*. Division of species in a second branch corresponds to differentiation obtained on the basis of morphological characteristics.

The analysis supported that the Olkhon vole is a valid species that distinct not only by morphology, but also phylogenetically. At the same time *A. semicanus, A. tuvinicus* and *A. olchonensis* constitute a single group of closely related species, sister to *A. strelzowi*. Our data confirm as well the results of previous researches on illegality of allocation of *A. strelzowi* to a separate genus (or subgenus) *Platicranius*.

The analysis of variability mtDNA allowed revealing essential intraspecific differentiation in a species of *A. barakschin*. It is revealed that individuals from the territory of the Republic of Tuva differ significantly from that of the central and eastern Mongolia. Genetic distances on cytochrome *b* correspond to the level of subspecies, ranging from 1.8 to 2.1 %.

DNA BARCODING COMMERCIAL ARMHOOK SQUIDS OF THE FAMILY GONATIDAE (CEPHALOPODA, TEUTHIDA) IN NORTHWESTERN PACIFIC

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The family of armhook squids are part of the Cephalopoda class, which occupies an important position in most oceans both at an ecological level and at a commercial level. Nevertheless, some gonatid species are difficult to distinguish with traditional morphological identification in cases when specimens are heavily damaged during collection or when closely related taxa are existent. As a useful tool for rapid species assignment, DNA barcoding may offer significant potential for gonatid identification. Here, we used mitochondrial fragment of Cytochrome Oxidase I to assess whether 12 gonatids accounting for entire northwestern Pacific fauna could be identified by DNA barcoding technique. The pairwise intra- and interspecific K2P distances were assessed, and relationships among species were estimated by NJ analysis. All the species forming well-differentiated clades in NJ and Bayesian trees. The only specimen morphologically identified as *Gonatus berryi* exhibited high levels of intraspecific distances with the members of *B. berryi* clade, presumably suggesting the occurrence of a cryptic species. Previously proposed sister species *G. tinro* and *G. okutani* demonstrated intraspecific level of divergence (0.3%) and low clades confidence limits supporting the hypothesis of conspecific status of these taxa. High levels (>6.5%) of genetic differentiation within *Bathyteuthis borealis* suggested existence of two cryptic species. Analysis of intra- and intergeneric distances led to an overlap between their ranges hence not confirming applicability of COI sequences for taxonomical delimitations of gonatid taxa at generic level. This study confirms the usefulness of DNA barcoding for identifying species as well as discovering cryptic diversity of NW Pacific gonatids. It also lays a foundation for other ecological and biological studies of Gonatidae.

RESURRECTION OF *PLACIDA BABAI* EV. MARCUS, 1982 (MOLLUSCA: GASTROPODA), A SACCOGLOSSAN SEASLUG SPECIES FROM THE SEA OF JAPAN AND ADJACENT WATERS OF THE PACIFIC

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In 1955, Kikutaro Baba published review on opisthobranch mollusk fauna of Sagami Bay, Japan (Baba, 1955). In fact, Baba was asked to publish this work by Japanese Imperor Horohito, who personally investigated the fauna of Sagami because the Imperor’s status did not allow him to use His name as an author of a scientific paper. Among the other opisthobranchs, Baba recorded *Placida dendritica* (Alder et Hancock, 1843) and gave brief description of local population morphology. Later Marcus (1982) described new species, *Placida babai* using mentioned above Baba’s publication. Quite soon in 1985 Baba responded – in his brief but informative paper, he considered *P. babai* as *nomen nudum* but gave very detailed comparative morphological analysis with other *P. dendritica* forms including nominative form from North Sea. Although, he did not provide name for this species, calling it *P. sp*. (=*P. babai* Marcus, 1982). However, Marcus’s discpription meet all the requirements of ICZN, so this name is well valid. We suppuse, Baba was confused with the situation where his ‘modest’ name was given to a species collected by the Imperor, thus he refused this name and never offered a new one.

In this study, we collected *Placida* af. *dendritica* from Vostok Bay in Primorye and Nevelsk in Sakhalin specimens that were morphologically identical with *Placida babai* as described (Baba, 1955, 1985). We compared COI gene marker sequences from specimens obtained in White and North seas populations; specimens from Californian, and Japanese sequences from GenBank obtained earlier in whole mtDNA sequencing project. The sequences of the specimens from Genbank, and Sea of Japan were 100% identical. The *p*-distances between COI sequences Sea of Japan vs. North Sea and White Sea populations have score 0.133-0.136 and 0.140 respectively, while *p*-distance between White Sea and North Sea populations are just 0.039-0.043% distant. Thus, Western Pacific and Atlantic populations are represented by two clearly distinct species. This is supported by their morphological characters: *P. babai* possesses awl shaped teeth of radula vs. blunt chisel shaped in the other populations. The processes of digestive gland in *P. babai* are numerous, fine, filling the rhinophores completely, and partially penetrating the foot, while such processes in *P. dendritica* are thick, never approach the foot, and entering proximal part of the rhinophores only. The COI sequences from California possess intermediate morphology between *P. dendritica* and *P. babai* (*p*-distance is 7% and 12%, respectively) possessing morphology similar to *P. dendritica*. We are proposing to preserve *P. dendritica* name for both Atlantic and E. Pacific forms before detailed reassessment of their taxonomical status while resurrecting validity of *P. babai* Ev. Marcus, 1982 for the species occurring in the Sea of Japan and adjacent waters.

INTRASPECIES DIVERGENCE OF *AEGILOPS TAUSCHII*: INVESTIGATING AN EXAMPLE OF INTERMEDIATE STAGE OF ADAPTIVE SPECIATION

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Aegilops tauschii Coss. is a diploid goat-grass, a wild relative of common wheat. It is mostly self-pollinating species with occasional cross-pollination. *Ae. tauschii* occupies the vast area in Central Eurasia, from Turkey to Kirgizia. It is presented by many small, fairly well isolated local populations, belonging to the two subspecies, *tauschii* and *strangulata*, which considerably differ genetically and ecologically. Genetic exchange between ssp. *tauschii* and ssp. *strangulata* exists in nature; also, no evident genetic barrier found between them in experimental crosses. It seems that intermediate, "hybrid" genotypes are eliminated by natural selection as disadvantageous.

Polymorphism of non-coding sequences of chloroplast DNA (cpDNA) and polymorphism of a nuclear gene *Got2*, encoding the enzyme glutamatic-oxaloacetatic transaminase, was investigated in a set *Ae. tauschii* accessions, representing all the species area.

Cluster analysis of the polymorphism of DNA sequences of *Got2* revealed that at the beginning of its existence as a species, *Ae. tauschii* was presented by its subspecies *strangulata*. In the course of the species evolutionary history, several phylogenetic lineages of ssp. *strangulata* had originated before the origin of ssp. *tauschii*. These results are in line with the levels of polymorphism of cpDNA: in ssp. *stangulata*, cpDNA is much more variable than in ssp. *Tauschii.*

Gene-geographic study revealed that *Ae. tauschii* intraspecies divergence was an adaptive process. So, geographic occurrences of cpDNA genetic markers, characteristic for different lineages of *Ae. tauschii* ssp. *strangulata*, distinctly correlate with climatic conditions of an area. Geographic patterns of *Got2* polymorphism showed that previously, the ancient lineage of *Ae. tauschii* ssp. *tauschii* occupied the vast area but then it was almost totally forced out by the major lineage of ssp. *tauschii*, which originated relatively recently. To say in general, ssp. *strangulata* is mostly adapted to sea-side climate and occupy the area along the southern part of Caspian Sea. The origin of ssp. *tauschii* enabled the species to occupy the vast area with continental climate.

Chronological sequence of the major events of *Ae. tauschii* intraspecies divergence, as revealed by cluster analysis of *Got2* DNA sequences, is as follows: first, the origin of the most ancient, relict lineage, T-9(1)s, of ssp. *strangulata*; then, the origin of the major lineages of ssp. *strangulata*; and finally, the origin of ssp. *tauschii*. This sequence is in line with the allozyme polymorphism of another nuclear gene, *Acph1*: its allele *Acph1*(100) is characteristic for ssp. *tauschii*, while the relict lineage T-9(1)s and all the other lineages of ssp. *strangulata* have the allele *Acph1*(95). Cluster analysis of cpDNA non-coding sequences polymorphism displayed another chronological sequence: T-9(1)s - ssp. *tauschii* - the major lineages of ssp. *strangulata*. Apparently, chloroplast genome of *Ae. tauschii* had diversified considerably by the time of ssp. *tauschii* origin. Surprisingly, it was an ancient variant of chloroplast genome, which was involved in the origin of relatively young subspecies.

MOLECULAR GENETIC DIFFERENTIATION OF SPECIES ASTRAGALUS SECTION CENANTRUM (FABACEAE)

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Astragalus L. (Fabaceae), the largest angiosperm genus in the world, comprises about 3,000 species and about 200 sections. It represent a remarkable example of adaptive radiation. Eurasian section *Cenantrum* Bunge has 41 species and is one of the most difficult in the genus. Member of the section characterized by morphological polymorphism, whereby the views of their independence and volumes set ambiguously. Molecular genetic methods in recent years have increasingly attracted to address issues of plant systematics, taxonomy specifying the boundaries previously selected taxonomic groups based on classical methods of taxonomy.

The goal of this study establish the characteristics of the genetic diversity of populations and make evaluation of the degree of genetic relatedness of species in *Astragalus* of systematically challenging section *Cenantrum*; also the refinement based on the results of borders delimiting at the species and intraspecific levels is planned.

We studied 23 populations of *Astragalus*: *A. frigidus* (9 population), *A. mongholicus* (11), *A. sericeocanus* (2) and *A. saralensis* (1) based on the analysis of chloroplast (PCR-RFLP) and total (AFLP) DNA variability. These data confirm the division of the section into subsections studied morphologically. Thus, revealed high values of genetic differentiation between populations of *A. frigidus*, *A. saralensis* and the population group, *A. mongholicus-A. sericeocanus*, relating to subsections *Elliptica* and *Semilunaria*, respectively. Identification of genetic differences and similarities between *A. mongholicus* and *A. sericeocanus*, which indicate their specific independence, on the one hand, and close relation to the other, is made. Peripheral population *A. mongholicus* (Middle Ural) different from populations of the central part of the species range of molecular genetic data and is monomorphic. This is probably due to prolonged isolation of the population and genetic drift.

The reported study supported by RFBR, research projects No. 14-04-31023-mol_a.

POPULATION GENETIC VARIABILITY OF *CALLUNA VULGARIS* (L.) HULL.

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Geographic variation and differentiation of the chloroplast DNA haplotypes, allozymes and morpho-anatomical leaf parameters were assessed in a number of certain European and Zauralye groups of *Calluna vulgaris* (L.) Hull populations, which have long been isolated from each other. Sharply pronounced genetic and phenotypic differences between the studied population groups were revealed. Genetic distances were many times higher than those between the relatively homogeneous European groups. The data obtained generally supported the hypothesis on the phenogenetic divergence of the Zauralye populations of *C. vulgaris*.

The reported study supported by RFBR, research projects No. 15-12-4-13.

TOWARDS EXPLAINING THE UNUSUALLY COMPLEX MHC OF THE PASSERIFORMES

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A distinguishing characteristic of genes that code for the major histocompatibility complex (MHC) is that alleles often share more similarity between, rather than within species. There are two likely mechanisms that can explain this pattern: convergent evolution or trans-species polymorphism (TSP). Our previous study analyzed exon 2 of the MHC class IIB in three passerine bird species in the genus *Corvus*: jungle crows (*Corvus macrorhynchos japonensis*) American crows (*C. brachyrhynchos*) and carrion crows (*C. corone orientalis*). Carrion and American crows are recently diverged, but allopatric, sister species, whereas carrion and jungle crows are more distantly related but sympatric species, possibly sharing pathogens linked to MHC IIB polymorphisms. These patterns of evolutionary divergence and current geographic ranges enabled us to test for trans-species polymorphism and convergent evolution of the MHC IIB in crows. Phylogenetic reconstructions of MHC IIB sequences revealed several well supported interspecific clusters containing all three species, and there was no biased clustering of variants among the allopatric carrion crows and jungle crows. There were just four identical variants shared among any of the three species. The phylogenetic patterns we observed suggest that TSP, rather than convergence, explains the interspecific allelic similarity of MHC IIB genes in these species of crows. A new analysis of the same gene, which includes six additional corvid species (including new and old world jays), reveals three strongly supported MHC IIB lineages common to at least five species. By focusing on variant lineages that are likely under balancing selection, it is possible to simplify song bird MHC analysis—where as many as 20 variants may be present in a single individual. Current and future work includes expression assays of different MHC variants to assess if, like other bird species, one or a few loci are preferentially expressed. In addition, we are estimating T-cell repertoire in song birds to test, and potentially challenge, the “optimal MHC variant number hypothesis” that has become dogma in general immunology.

DNA BARCODING DENDRONOTID MOLLUSKS OF RUSSIAN SEAS

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The family Dendronotidae (Gastropoda: Nudibranchia) includes one monotypic genus *Dendronotus* Alder et Hancock, 1845. These molluscs are one of the most common marine epifaunal invertebrates in the northern hemisphere. They as many other opisthobranchs are emerged model group in various fields including taxonomic, phylogenetic, developmental, physiological and molecular studies. However, the diversity of *Dendronotus* in Russian Seas remains poorly explored mostly because of polymorphic colouration pattern in different species and variations of radula morphology during late ontogenesis. Till recent, only three *Dendronotus* species were registered in the Russian seas, amphiboreal *D. frondosus* (Ascanius, 1774) and *D. dalli* Bergh, 1879 and circumpolar *D. robustus* Verrill, 1870. DNA barcoding combined with molecular species delimitation methods and morphological studies is a useful approach for taxonomy studies of this genus.

We studied 78 specimens of the genus *Dendronotus* collected the White, the Barents, the Kara, the Bering seas, in Avachinsky Bay (Kamchatka) and in the northwestern part of the Sea of Japan. External and internal morphology was studied using stereomicroscope and scanning electron microscope. Molecular analysis included an obtaining of partial sequences of mitochondrial genes cytochrome *c* oxidase subunit I and 16S rRNA and nuclear genes histone 3, 18S rRNA and 28S rRNA. Molecular species delimitation was performed with ABGD, GMYC and K/θ analyses. Eight species of the genus *Dendronotus* inhabiting shallow waters of Russian seas were described: *D. frondosus*, *D. lacteus*, *D. dalli*, *D. robustus* and four new species: *D. dudkai*, *D. kalikal*, *D. kamchaticus* and *D. niveus*. *D. frondosus* is common species in the shallow Barents and White Seas. However in the northern part of Pacific Ocean *D. frondosus* has been found only in Amur Bay of the Sea of Japan. In Vostok and Rudnaya bays of the Sea of Japan a cryptic species *D. dudkai* has been described. It is sibling species to *D. frondosus* and differs from it only by features of jaws and reproductive system and by means of molecular analysis. In the Bering Sea and on the Pacific coast of Kamchatka *D. frondosus* is replaced by two sympatric cryptic species *D. kamchaticus* and *D. kalikal* revealed not only by molecular analysis, but detailed studies of morphology, including the features of the radula and reproductive system. New data obtained have allowed excluding the Pacific species *D. dalli* from the lists of fauna of the White and the Barents Seas. Earlier this species has repeatedly pointed to these waters. At the same time, there were discovered a new cryptic species, *D. niveus* that well differs from *D. dalli* and other species of the genus by means of molecular analysis. At the first time *D. lacteus* was registered for the fauna of Russia. According to our data its distribution therefore includes north-east Atlantic, the Barents Sea and also the east coast of Kamchatka peninsula.

Results of molecular analysis show that specimens of *D. lacteus* from Russian Seas are very close genetically to specimens from the North Sea. *D. robustus* tends to separate into shallow water (4–10 m) and deep sea (100–200 m) populations. Molecular data showed some differentiation between them. Some transitional forms, probably hybrids, at intermediate depths (50–100 m) were also discovered. Therefore, to resolve the question of status of *D. robustus* as a single species an additional morphological and molecular study is necessary.

This work supported by Grant of Russian Scientific Fund, Project No. 14-50-00034.

IS CAPELIN ONE SPECIES? A KARYOTYPE ANALYSIS

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Capelin, *Mallotus villosus* is a circumpolar species and among Osmeridae representatives, it alone is only marine species. Pacific capelin was described by T. Pennant in 1784 as a real species, *Salmo catervarius*. Later it was showed, that it is a subspecies of Atlantic capelin, *Mallotus villosus catervarius*, though it its morphometric discrepancy and difference in its ecology of breeding were shown. Although, capelin of North-East of Russia in some morphological characters is closer to Atlantic, but in others to Pacific capelin (Chereshnev et al., 2002).

Thus, there is a great interest on the study of capelins in Atlantic, Arctic and Pacific regions by genetic methods. However, it was not so simple to resolve a problem. According to molecular-genetic data capelin of north-east Atlantic and Pacific-and-Arctic Region are closer to one another, than each of them to capelin of Kara Sea, which geographically intermediate between Atlantic and Pacific capelins (Mecklenburg et al., 2011).

The karyotype of capelin of Barents Sea (north-east Atlantic) that studied (Ghigliotti et al., 2008) has: $2n = 54$, $NF = 80$. But karyotype of Pacific capelin from the Sea of Okhotsk (north of the Sakhalin) studied by the author has $2n = 34$, $NF = 56$.

Such sharp differences in karyotypes of Atlantic and Pacific capelins do not leave doubts on the species status for each, Atlantic and Pacific capelins and rise the necessity of identification of their status jointly by molecular and karyological methods using the geographically intermediate capelin form of the Kara Sea. I am asking, what is Kara Sea capelin?

CHROMOSOMAL VARIABILITY OF FAR EASTERN *MYOTIS PETAX*

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Eastern water bat, *Myotis petax* Hollister, 1912, is a common and abundant vespertilionid species with wide distribution range. Eastern water bat is find from the Altai, Transbaikalia and southern part of Russian Far East to South Korea and Japan. Until recently, *M. petax* was included into *M. daubentoni* Kuhl, 1817 as a subspecies. However, a validation of the species distinctiveness of *M. petax*, is derived from molecular and morphological data.

The differences in the amount and location of heterochromatic material, position and number of the nucleolus organizer regions on chromosomes revealed by application of different banding techniques are the important and species-specific features of *Myotis* species. Previously the conventional staining of *M. petax* (=*M. daubentoni*) was studied to some individuals from the populations of South Korea and Primorsky Krai. The diploid chromosome number (2n=44) and the fundamental number of autosomal arms (NFa=50) of *Myotis petax* is not different from other species of genus *Myotis* Kaup, 1829.

The location and number of nucleolus organizer regions in the *M. petax* karyotype defined at the first time in our research. The distributional pattern of nucleolus organizer regions of *M. petax* is significantly different from those of *M. daubentoni* that also demonstrated a species distinctiveness of *M. petax*.

The study of distributional pattern of heterochromatic segments in the karyotypes of *Myotis petax* individuals from Amur Oblast and Primorsky Krai also conducted at the first time. The interspecific variation in extent and position of heterochromatic material on chromosomes is find.

INTRASPECIFIC GENETIC DIFFERENTIATION OF *HAEMATOPUS OSTRALEGUS* LINNAEUS, 1758 ON THE TERRITORY OF BELARUS

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Based on the analysis of polymorphism of mitochondrial gene of cytochrome oxidase subunit 1 (*COX 1*), the intraspecific differentiation of Oystercatcher' (*Haematopus ostralegus* Linnaeus, 1758) populations on the territory of Belarus was studied. The study is based on the hypothesis of M.E. Nikiforov about the existence of two subspecies: *H. o. ostralegus* and *H. o. longipes* in the north-west and south-east of Belarus respectively. For the hypothesis testing, the DNA samples extracted from muscle tissues of 4 individuals from the south-east (Homiel region, Turaŭ), 2 individuals from the north-west of Belarus (Braslaŭ region), as well as of 1 sample from Kirov (Russia) and of 1 sample from Sakhalin (Russia), were examined. In addition, DNA sequences from GenBank (Norway, Sweden, Netherlands and Russia) were involved in the analysis. All analyzed samples correspond the chorologic structure of Oystercatcher' subspecies on the territory of Palearctic.

The length of the analyzed fragment is 575 bp. Sequence contains seven variable sites, representing 1.2% of the total. The share of parsimony-informative sites is 0.2% (1 site). The average nucleotide diversity is 0,002%, haplotype diversity is 28.6% bp. The results of phylogeographic analysis using the program MEGA 6.0 (Maximum likelihood method, Hasegawa-Kishino-Yano model; IB≥50%, 500 replications) confirmed the presence of subspecies *H. o. longipes* on the territory of Belarus: sequences from the south-east of Belarus were united in one cluster (IB = 59%) with the sequences from the main area of this subspecies in Russia (Kirov). Individuals from the north-west of the Belarus and from Russia (Kandalaksha Bay, Astrakhan, Krasnoufimsk), Norway, Sweden and the Netherlands (main area of *H. o. ostralegus*) were included in another cluster (IB = 36%).

EFFECTS OF EMBRYO TRANSFER AND IN VITRO CULTURE ON CHARACTERISTIC BEHAVIOR IN GENETICALLY CATATONIC RATS (GC STRAIN)

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Assisted reproductive technologies (ART) are used for infertility treatment in humans. In particular, granulocyte–macrophage colony stimulating factor (GM-CSF) is applied to achieve higher implantation rates. There are some clinical data indicating that the frequency of some autistic disorders may be increased in people born as the result of ART, but these data are controversial. The aim of this study was to investigate long-term effects of *in vitro* culture at the preimplantation stages of embryo development combined with the embryo transfer on the behavior of the adult offspring in rats with genetic catalepsy (GC strain). GC rat preimplantation embryos were collected at two-cell stage and cultured *in vitro* for 48 h in rat one-cell culture medium (R1ECM) supplemented with GM-CSF up to morula stage, then transferred to recipient female rats. At the age of 5 mo. GC rats born after transfer of cultured embryos were compared with naturally born GC and Wistar rats of the same age with the open field test and the elevated plus-maze test. The results of the open field test indicate that GC rats of both groups possess less locomotion and less rearing activity as compared to Wistar rats. GC rats born after *in vitro* culture with GM-CSF and the subsequent embryo transfer (GC GM-CSF group) demonstrated lower exploration activity as compared to GC rats born naturally (GC group). Elevated plus maze test revealed increased anxiety in GC GM-CSF rats: they spent less time in open arms of the maze as compared to GC group. There were also differences in the rates of head-dipping and some other behavioral characteristics between these groups. Our data suggest that some combinations of ART applied to GC rats at preimplantation stages of development, affect their adult behavior.

Research supported by the Russian Foundation for Basic Research (Project No.13–04–00685).

THE MOLECULAR PHYLOGENY OF THE GENUS TRIFOLIUM L. SECTION LUPINASTER (FABR.) SER. (FABACEAE)

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Section Lupinaster is difficult of morphology and not fully investigated in terms of the phylogenetic relationships inside the group. For molecular phylogeny study we used 35 samples, which represent of genus *Trifolium*, section *Lupinaster* (for some species we used several samples from different regions). The genomic DNA was isolated using the method of (Doyle, Doyle, 1987) with modifications (Rodionov et al., 2005). The ITS region was amplified using the primers ITS-1P (Righway et al., 2003), ITS-2, ITS-3, ITS-4 (White et al., 1990). In preliminary study we decided to focus on unexplored species (*Lupinaster albus*, *T. pacificum*, *T. neurofillum,* hybrid *T. lupinaster x T. baicalense*) and obtained ITS for 7 samples. Sequence data were aligned by Clustal implementation in MEGA5 software (Tamura et al., 2011). We added in the alignment *Trifolium* ITS sequences from GenBank (http://www.ncbi.nlm.nih.gov/genbank/) for comparison. *Lotus* was used outgroup. Phylogenetic tree was constructed by NJ method with 1,000 bootstrap replicates.

Species we study diverged on two phylogenetic clades. Clade with a good bootstrap support consisted of section *Involucrarium* species (*T. nanum* and *T. neurophillum).* Another clade consisted of complex *T. lupinaster s. l.* species. On the NJ tree *Lupinaster albus* appeared non monophyletic and group together with *Trifolium* species. *Complex T. lupinaster s. l.* divided on subclades with low bootstrap support. We found some genetic differentiation inside this complex, but result was not congruent with species recognized morphologically. *T. pacificum* and *Lupinaster albus* seems to be polyphelitic. Phylogenetic study of *T. lupinaster s. l.* complex is difficult due to polyploidy. There are known a range of chromosomes numbers (2n= 16, 32, 40 и 48) has been reported (Bobrov, 1945; Pavlovs, 1989). This could be explained by hybrid origin or series of autopolyploidy events.

GENE INTROGRESSION BETWEEN SPECIES AND GENE TREE RETICULATIONS: ARE THESE EVENTS CHALLENGED NEO-DARWINISM AND DNA BARCODING?

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This assignment discuss 3 points: 1. On methods for the hybrid detection, 2. On facts obtained on gene introgression by mtDNA and nDNA markers, and 3. On the correspondence of data on diversity with Biological Species Concept (BSC). DNA data on evolutionary divergence of animals are summarized and concluded that they concordant with the BSC. Also evident that introgression exists, although even in a wide zone of *Mytilus* spp., for example, it may be quite restricted or be asymmetric, so holding intact at least the “source” taxa. If accept that the sexually reproducing animal species are introgressed, then we should recognize that the orthodox BSC, in terms of complete gene flow absence among species, is inadequate in a sense that many zoological species are not biological species yet. However, in future they definitely become biological species. Such a conclusion supported by genetic distance increase with taxa rank, lowest diversity at intraspecies level and absence of interspecies reticulation for bulk of gene trees. It seems that claims on a crush of the modern BSC paradigm and Neo-Darwinism in general due to wide-scale gene introgression and phylogeny reticulation are too premature, especially keeping in mind the long history of many hybrid zones. These outcomes have direct impact on the science policy of iBOL (www.ibol.org\), a world-wide DNA barcoding initiative and species discrimination in particular. Species identification by DNA barcode is successful for bulk of attempts because prevalence of geographic mode of speciation and long time for the formation of new taxa in isolation; hence it allows accumulation of random mutations and many different nucleotide substitutions that are detectable by molecular markers.

The author gratefully acknowledge financial support for research received from the Russian Science Found: grant #14-50-00034 in a part connected with the section of molecular phylogenetics and tree reticulation, and as well as from Russian Foundation for Basic Research grants #14-04-00758 and #15-29-02456 in a part connected to gene introgression between species.

MOLECULAR SYSTEMATICS AND DNA BARCODING OF ALTAI OSMANS, *OREOLEUCISCUS* (PISCES, CYPRINIDAE, LEUCISCINAE), AND NEAREST RELATIVES, AS REVALED BY SEQUENCES OF CYTOCHROME *B* (*CYT-B*), CYTOCHROME OXIDASE *C* (*CO-1*), AND COMPLETE MITOCHONDRIAL GENOME

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Mitochondrial DNA (mtDNA) at the protein-coding *Cyt-b* gene along with attraction of data from GenBank for *Co-1* gene fragments and complete mitochondrial genome (mitogenome) of Altai osmans and nearest relatives of Leuciscinae fish species have been compared. Phylogenetic trees were built by 4 techniques: Bayesian (BA), maximum likelihood (ML), maximum parsimony (MP), and neighbour-joining (NJ). Resolution of *Cyt-b* trees for species of two genera (*Oreoleuciscus* and *Phoxinus*) was quite distinct in all approaches. For *Tribolodon*, the single gene trees were not well resolved; however, mitogenome tree was resolved. Species identification on per individual basis (DNA barcoding) was high for both *Cyt-b* and *Co-1* genes. Trees built using data for 13 protein mitochondrial genes revealed a complicated phylogenetic pattern within the subfamily Leuciscinae.

Scores of the average *p*-distances at three taxonomic levels were considerably different: (1) 1.16±0.96, (2) 8.21±1.01, and (3) 16.41±0.85 for *Cyt-b* and (1) 1.04±0.78, (2) 8.30±0.92, and (3) 10.74±0.79 for 13 protein genes of mitogenome. Mitogenome distance data were summarized for the taxonomic hierarchy for the first time in literature. A concordant increase in distance score, as the rank of taxa grows, with the minimum score at the intraspecies level, both for a single gene and the whole mitogenome, substantiates the concept that speciation in the subfamily Leuciscinae in most cases follows a geographic mode. The sharp clustering of Altai osmans, *Oreoleuciscus potanini*, and *O. humilis* species at *Cyt-b* and *Co-1* gene trees with small overall genetic distances, obtained for both genes, allows us to consider these taxa as separate but genetically sister species.

This work was supported in part by Russian Science Fund #**14-50-00034** (biodiversity) and the Russian Foundation for Basic Research grant **#15-29-02456** (phylogeny).

MOLECULAR PHYLOGENY OF RUSSIAN FAR EASTERN FLOUNDERS (PLEURONECTIFORMES, PLEURONECTIDAE) BASED ON MITOCHONDRIAL SEQUENCES FOR GENES, *CO-1* AND *CYT-B*, AND COMPLETE MITOGENOME

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To increase knowledge about the systematics of Pleuronectidae the primary sequence of nucleotides at subunit 1 cytochrome oxidase *c* (*Co-1*) and cytochrome *b* (*Cyt-b*) genes were determined. In total 17 newly collected species and some species from GenBank were analyzed in this research. Phylogenetic relationships among representatives of flounders were based on four types of trees: neighbor joining, maximum parsimony, Bayesian and maximum likelihood. These trees showed similar topology. Two separate clusters on the trees support subfamily Hippoglossoidinae and Hippoglossinae subdivision and monophyletic status of these taxa. The subfamily Pleuronectinae also can be considered monophyletic, if the tribe Microstomini is excluded from it and genus *Lepidopsetta* is moved in the tribe Pleuronectini. The phylogenetic status of *Hippoglossoides elassodon* and *H. robustus* is uncertain and need to be resolved in further investigation.

Mitogenome of 38-45 complete sequences from NCBI GenBank were analyzed. After alignment two sets of nucleotide sequences were formed and investigated independently, one set included only structural genes (15,068 bp) while the second set comprised by the whole mitogenome (15,120 bp). Both data sets gave congruent phylogenetic signal basically agreed with conventional views on the taxonomic system for the order Pleuronectiformes. In particular, the node which includes the representatives of suborder Pleuronectoidei and superfamily Pleuronectoidea is highly supported.

The incongruities between morphological and molecular issues that also were obtained suggest the need for reassessing the systematic value of some morphological characters and phylogeny both at family and order levels.

This work was supported in part by Russian Science Fund #**14-50-00034** (biodiversity) and the Russian Foundation for Basic Research grant **#15-29-02456** (phylogeny).

PERICENTROMERIC HETEROHROMATINE IN APODEMUS GENERA (MAMMALIA, RODENTIA)

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The wood and field mice of *Apodemus species* (*A. agrarius, A. peninsulae, A. specious, A. argenteus*) are widespread throughout their Far East Asian distribution area.

Chromosome numbers vary from 46 to 48, or from 48 to 56. Species have differentiation by number and locality of heterochromatin and nuclear organization on chromosomes. The B-chromsomes in amount from 1 to 30 in *A. peninsulae* and from 1 to 2 in *A. argenteus* were described. Cross-hybridization of DNA probes generated from pericentric C-positive blocks of chromosomes of *Silvaemus* species with chromosomes of *Apodemus* species and DNA probes from pericentric C-positive blocks of chromosomes of *Apodemus* species with chromosomes of *Apodemus* and *Silvaemus* species showed diffuse fluorescent signal along whole length of chromosomes except pericentric regions. The B-chromosoms of *A. peninsulae* from Siberia have pericentromeric heterochromatin of *A. peninsulae*, while B-chromosom's arms contain pericentromeric heterochromatin of *Silvaemus flavicollis.*

These results indicate that level of DNA homology in pericentric chromosome regions of *Apodemus* and *Silvaemus* species decrease, while the level of divergence rises and species become less related to each other.

COMPARATIVE GENETIC ANALYSIS OF THREE SPECIES OF FAR EASTERN DACE OF THE GENUS *TRIBOLODON* (CYPRINIDAE, CYPRINIFORMES) BASED ON SOME MORPHOLOGYCAL AND GENETICAL TRAITS

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Dace from waters of the Russian Far East is an object of recreational and commercial fisheries [1]. Expanding our knowledge on population biology and genetic composition of the Far Eastern dace species will be helpful for better understanding life strategies of these anadromous fish, and may also be useful for sustainable use of fish resources and for conservation of genetically distinct and unique fish stocks. Species of the genus *Tribolodon* belong to subfamily Leuciscinae, family Cyprinidae. The difference between southern form and northern form has been find using electrophoretic analysis of protein cording locus Prot-2, Cyt B and D-loop sequences [2, 3]. However, the above-cited authors did not suggest taxonomic status for these genetically different groups of the Far Eastern dace using both morphological characters genetic traits. The aim of the present study is to compare three nominal species of the genus *Tribolodon* using sequence data of mitochondrial DNA *Co-1* gene and some morphological traits.

According to *Co-1* gene fragment nucleotide sequence signal, southern and northern forms of *T. hakonensis* form different clusters with high bootstrap level. The divergence level between samples *T. hakonensis* from the two locations of the south of Primorye and Sakhalin were 4.1 - 5.1%. For comparison the number of vertebrae as morphological trait was chosen. It showed the significant differences of *T. sahalinensis* from *T. hakonensis* and *T. brandtii*. The maximum number of vertebrae for *T.sahalinensis* were less than the minimum number of vertebrae for *T. hakonensis* and *T. brandtii*.

Our results well agree with former findings on biochemical genetics and molecular genetics of the Far Eastern dace [2 - 4] and present the same level of divergence for *Co-1*. This genetic divergence for species of *Tribolodon* genus is not big (maximum: 8.30 ± 0.17), it is lower than level of divergence for the most of animals (11.06 ± 0.53; [5]). X-rays of 3 species of *Tribolodon* genus demonstrate the existence of interspecific and intraspecific variation of the number of vertebrae. These results well correspond to the general rule for the non anadromous Cyprinidae. The obtained data do not give grounds to create a new taxonomic rank for one of *T. hakonensis* population groupings from the different regions.

This work was supported in part by Russian Science Fund #**14-50-00034** (biodiversity) and the Russian Foundation for Basic Research grant #**15-29-02456** (molecular phylogeny).

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TRANSFORMATION OF ARCHITECTURE INFLORESCENCES IN *ARABIDOPSIS THALIANA*

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Inflorescence structure now associated with a number of transcription factors, in particular *TFL* has a mutant expression for causing transformation of indeterminate inflorescences and determinate. Terminal flower has key in the classification of inflorescences. However, in plants mutant line *tfl 1-2 Arabidopsis thaliana* inflorescence structure does not match any of the type's defined inflorescences. Study focuses on clarifying the genesis of the terminal flower in *A. thaliana*. The study focuses on the identification of the genesis of the terminal flower in *A. thaliana* and the mechanism of mutual transfer of indeterminate inflorescences in determinate. Here has been analyzed variability of the structure of shoots and inflorescences of *A. thaliana* at the baseline *er1* and mutant lines *tfl1-2* during morphogenesis. It is established that the mutant line *tfl1-2 A. thaliana* reaction norm narrows and this leads to the transformation of the structure of the inflorescence. Determinate and indeterminate inflorescences have differences in spatial and temporal terms. Plants mutant line *tfl1-2* transition to flowering at an earlier stage of morphogenesis of shoots, and the morphogenesis inflorescences have terminated earlier than in the baseline *er1*. In mutant line *tfl 1-2*, flowering begins with the escape underdevelopment structure and morphogenesis of the inflorescence terminates at its underdevelopment. The real significance of *TFL* as a transcription factor is not so much in the formation of terminal flowers as in the initiation date of transition to flowering and implementation of the program of ontogeny as a whole. Thus, flowering (puberty) begins with underdevelopment of shoots and inflorescences finish itself development with genetically determined. Thus, neoteny has genetically determined. The results that were obtained can also be regarded as evidence for the theory of recapitulation.

RETICULATE EVOLUTION OF THE GENUS *LEPUS* IN NORTHEASTERN ASIA

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Introgressive hybridization is not rare phenomenon among closely rerated and neighboring species, which can confuse the estimate of real phylogenetic relationships, but inferring important histories of demographic and intra-species interactions. The genus *Lepus* is an appropriate model because approximately 30 species are recognized from variable habitats across North America, Asia, Europe, and Africa, and more than 10 species are involved in introgressive hybridizations. However, the most of studies have been done in European vs. North American species, and phylogenetic relationships and extent of introgression of northeastern Asian species have not been surveyed. We examined phylogenetic relationships among four hare species (*L. timidus*, *L. mandshuricus*, *L. coreanus*, and *L. brachyurus*) from Russia, Korea and Japan using sequences of eight nuclear gene loci (*Mgf*, *Tg*, *Tshb*, *Sptbn1*, *Mc1r*, *Asip*, *Phka2*, and *Sry*) and *Cytb* of mtDNA, with database sequences of other populations and neighboring species (*L. capensis* and *L. sinensis*) in China. Phylogenetic analyses showed multiple introgression of nuclear and mitochondrial genes among continental hare species, showing variety in genomic loci, species pairs, locality, and geographic patterns of the distribution of the introgressed lineage. An extreme case of introgression is observed in the *L. mandshuricus* complex (*L. mandshuricus* and *L. coreanus*), which has lost the original mtDNA lineage due to replacement by introgressed lineages from the northern species *L. timidus* and the southern species *L. sinensis*. In contrast, no evidence of introgression across genomes to insular populations of *L. timidus* in Sakhalin and Hokkaido exists, or of the participation of *L. brachyurus* in Japan in the reticulate evolution. Climatic changes during the Pleistocene promote distribution shifts and the reticulate evolution of hares in continental northeastern Asia, but long-term isolation on the marginal islands after colonization.

SIXTEENTH-EYED LION MANE – A NEW CYANEA SPECIES FROM THE WHITE SEA

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The genus *Cyanea* includes some of the most conspicuous and majestic representatives of megaplankton. Throughout the last century its taxonomy has been revised repeatedly due to high morphological plasticity of *Cyanea* species. Until recently all *Cyanea* from European Arctic have been identified as *C. capillata*. Here, we describe *C. tzetlinii* sp. Nov, a new *Cyanea* species sympatric to *C. capillata*. Morphologically *Cyanea tzetlinii* sp. nov. is easily distinguishable from all described *Cyanea* species by an eye-spot-bearing bulb in the basal part of each of the eight rhopalia upon the subumbrellar side of the bell, as well as by a unique combination of other morphological traits (the shape of bell margin, structure of gastrovascular system, color and nematocysts clusters on the exumbrellar surface). All the collected specimen of *C. tzetlinii* sp. nov. were somewhat smaller than *C. capillata* from the White Sea collected in the same time in the same place. This well-recognizable morphological characteristic is supported at the molecular level by a substantial genetic distance in mitochondrial (CO1: 9.6-10.6%, 16S RNA: 3.1-3.5%) as well as nuclear (ITS: 5.0%, 18S RNA: 0.1%) loci, making it the sister species to *Cyanea capillata*. In the ecological aspect we can note that mature *C. tzetlinii* sp. nov. form accumulations a bit later, than *C. capillata*. Taking into account the young geological age of the White Sea and a substantial genetic divergence between *C. tzetlinii* sp. nov. and the nearest sister species, we suppose that *C. tzetlinii* sp. nov. has been advected to the White Sea from elsewhere and may also inhabits other Arctic seas.

NUCLEAR AND MITOCHONDRIAL POLYMORPHISM IN BAIKALIAN AMPHIPOD *GMELINOIDES FASCIATUS*

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Gmelinoides fasciatus is a Baicalian amphipod and successful ecologically plastic invader. Previous study of *G. fasciatus* with mtCO1 showed existence of four clearly distinguished populations in Baikal. Appeared that one of them had large distance to others. We used the intron sequences of the gene coding for the ATP-synthase beta-subunit to investigate population structure. Intronic marker was shown to be very convenient for population level studies and is applicable for phylogenetic analyses only in case of closely related taxa. This may be illustrated by the example of the intron studied, which is extremely diverged between *G. fasciatus* and the representatives of 3 related Baikalian amphipod genera (*Eulimnogammarus*, *Pallaseopsis* and *Palicarinus*). This also suggests that evolution of this sequence is close to the neutral pattern, although it is currently hardly possible to prove in case of a non-coding sequence. It was shown that this marker is quite divergent: it's diversity (0.0236%±0.001) is comparable with the one of the mitochondrial marker CO1 (0.0409%±0.001). Interestingly, phylogenies inferred from intronic sequences for this species differ from phylogenies based on mitochondrial sequences if obtained for the same set of organisms. There are two populations and their division is not the same in comparison with previously defined population boundaries. This may be due to different modes of evolution or because of different modes of genetic segregation of nuclear markers. In any case combining these data with already published mtDNA sequences data may allow one to obtain species history.

KARYOLOGICAL AND MOLECULAR GENETIC DIFFERENTIATION *Chelidonium majus* L. sensu lato (Papaveraceae): TO THE QUESTION OF CRYPTIC SPECIES IN FLOWERING PLANTS

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Chelidonium majus L. sensu lato (Papaveraceae Juss.) is morphologically and karyologically polymorphic species. Aim of the study is to identify cryptic species through detailed taxonomic boundaries of the *Ch. majus* and differentiate the populations based on genetic and karyological data.

We analyzed the genomic and chloroplast fragments of 36 specimens of *Ch. majus*: internal transcribed spacers, ITS1 – 5.8S – ITS2 and spacers within the chloroplast region trnL – trnF. All genome fragments were amplified in PCR and sequenced. Obtained sequences were searched for close homologues with help of Basic Local Alignment Search Tool (NCBI/BLAST) and analyzed in BioEdit 7.0.9.0 program. The genetic similarity of ITS was 98.2 – 100 % while chloroplast fragment was more conservative, 99.6 – 100%. Only 10 of 36 sequences had single substitutions in chloroplast fragment. Both fragments except genes for 5.8S rRNA and trn were combined to construct alignment for phylogenetic analysis. The Bayesian phylogenetic tr ee was reconstructed on the basis 1,088 bp nucleotide fragments. Were also counted of the chromosome number ($2n$) in samples of *Ch. majus* included in genetic analyze. The chromosome counts were carried out using pressed temporary preparations of dividing cells (at metaphase) from the apical portion of roots by standard method.

There were three well separated clades (*A*, *B* and *C*) on the phylogenetic tree. Clades *A* and *B* had a common ancestor and separate *Ch. majus* from two areas West Siberia – Central Asia (clade *A*) and Europe – Caucasus (clade *B*) with chromosome number $2n = 12$. The clade *C* which was most distinct from others and includes specimens from Far East with $2n = 10$ and south part of East Siberia with $2n = 11$ and rarely detected samples with $2n = 12$. We assume that the clade *A* and *B* are independent species in which $2n = 12$. Also, we find that the clade *C* combines 2 species: one species with $2n = 10$, and a hybrid with $2n = 11$ or 12.

Cryptic species represent a significant gap in the study of biodiversity and taxonomy questions. Identification and study of such species is very important to disclose these issues to the knowledge of evolution and speciation, for the protection of rare and economically valuable species.

The reported study was partially supported by RFBR, research projects No. 14-44-04105-a_sibir, 14-47-04125-a_sibir and 15-04-05372-a.

GEOGRAPHIC ISOLATION AND PHYLOGEOGRAPHY OF THE MAGPIE *PICA PICA*

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The Eurasian magpie *P. pica* is a convenient model for phylogeographic and microevolution studies. It has high population number, wide Holarctic range and several peripheral isolates. Complete Control Region of mtDNA, 1,328 bp, was sequenced for 111 samples of 9 subspecies: *P. p. fennorum, P. p. pica, P. p. bactriana, P. p. leucoptera, P. p. camtschatica, P. p. jankowskii, P. p. serica, P. p. mauritanica,* and *P. p. hudsonia*. The phylogenetic network clearly illustrates the geographic distribution of haplotypes. One major group represents all European and Siberian subspecies, without any major haplotype and geographic affinity. The haplotype of Kamchatka differs from that clade by 19 and more substitutions. Its homogeneity reflects unambiguous founder effect or bottleneck of Kamchatka isolated population. The clade of *P.p.serica* and *P.p.jankowskii* representing samples from South Russian Far East, Japan and Korea, is separated from the western group by at least 66 substitutions. In total, this group is characterized by high nucleotide and haplotype diversity and besides it is subdivided into two subgroups. Yet, there is no clear geographic pattern. It is notable that even much diverged haplotypes were found at the same locality. The population of Kyushu Island introduced ca 300 years ago from Korea is very homogeneous due to founder effect. The other Japanese population of Hokkaido appeared first time 30 years ago, but is rather variable. It supposedly was originated from Primorsky krai population which is very heterogeneous itself. Divergence by mtDNA haplogroups which was described above corresponds well to differences in the magpie phenotypes and bioacoustic features, so mitochondrial and nuclear data prove to be in a good accordance. Taxonomic revision should be conducted in purpose to unite several subspecies. In majority of cases presented, genetic distinctness of the haplogroups may be explained by geographic isolation with further accumulation of mutations.

GENETIC AND MORPHOMETRIC VARIABILITY AND HYBRIDIZATION OF TWO SPECIES OF MUSSELS, MYTILUS TROSSULUS AND MYTILUS GALLOPROVINCIALIS, IN THE NORTH-WEST OF SEA OF JAPAN

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Pacific mussel, *Mytilus trossulus* and Mediterranean introduced species *M. galloprovincialis,* studied in the west-pacific part of the Japan Sea, basically in the Peter the Great Bay using genetic markers and traits of morphology. The genotyping of individuals from 8 settlements made at 8 polymorphic enzyme loci and 2 nuclear DNA markers, *Me-5* and *ITS-1,2*. Frequency of parent species and their hybrids in samples are analysed. Enzyme and nuclear markers exhibited concordant genetic variability. In the studied samples, the prevalence of local species *M. trossulus* is obtained. The fraction of introduced species *M.galloprovincialis* in the whole material is relatively low. Exclusion is settlement in the Possiet Bay, near the Zarubino town. Here in the zone of active international ferry transportation the frequency of this species reaches up to 42±2%. In this area There is also the greatest number of hybrids in the area. Considering sampling errors it apparently follows that the proportion of hybrids determined by allozyme loci and two different DNA markers do not differ significantly, however, semi-diagnostic biochemical genetic loci give somewhat higher portion of hybrids. Main outcome from obtained data is made that the invasion of *M. galloprovincialis* in west-pacific part of Japan Sea is continue; it is also stated that in the Possiet Bay even permanent settlements of this mussel are already available which never recorded here before.

Eleven morphology traits and their indices used to classify individuals and estimate frequencies of two species and presence of hybrids in five samples taken in 2012-2013. Discriminant analysis revealed 78% precision of classification. Only one F_1 hybrid and no invasive species-specimens assumed on these data.

This work was supported in part by Russian Science Fund #**14-50-00034** (biodiversity aspects) and the Russian Foundation for Basic Research grants # **13-04-06081** and **#15-29-02456** (hybrid zone investigation).

A COMPARATIVE STUDY ON THE GENETIC BASIS OF BIRTH WEIGHT IN WATER VOLE AND DOMESTIC PIG

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The impact of genetic factors on birth weight of water vole (*Arvicola amphibius)* was evaluated to compare the obtained results with those previously reported for domestic pigs (*Sus scrofa*). It was found that the strength of the joint effect of genotype of the parents on the birth weight of offspring is significantly higher in voles (0.68) than in pigs (0.47-0.48). This indicates either a larger number of loci controlling such trait in voles, or a larger variability of alleles in these loci. The strength of the maternal effect in voles and pigs did not differ (0.15 and 0.13–0.14). It may be due to the association of birth weight with common maternal factors acting in multifoetal mammalian species. The strength of the effect of dominance in voles is 0.25, in pigs 0.14– 0.16, whereas the powers of influence of intra locus and inter locus interactions are 0.02 and 0.02-0.05, respectively. In voles, uncertainty of phenotypic realization of genotype in a group of fathers is higher than that in pigs (0.39 vs. 0.15–0.17); however, in the groups of mothers these values are almost equal (0.27 and 0.29-0.30). Therefore, uncertainty of phenotypic realization of genotype among voles is much higher in males than in females, and vice versa in pigs. Perhaps this is due to the fact that high uncertainty of phenotypic realization of genotype can increase the competitiveness of males in wild nature (water voles), but is harmful during artificial selection, when the value of male prepotency (domestic pigs) comes around.

A STUDY OF POLYMORPHISM OF Y-CHROMOSOME IN POPULATIONS OF TYUMEN-TURIN TOBOL-IRTYSH TATARS OF TYUMEN REGION

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Siberian Tatars are the indigenous population of the Western Siberia with a vast territory of settlement "from the Urals to the Yenisei." Tobol-Irtysh Tatars are the largest ethno-territorial group of Siberian Tatars. Places of accommodation of this group Tatars are in the administrative borders of Tyumen and Omsk regions of Russia. As a part of the Tobol-Irtysh Tatars, there exist isolated Turin Tyumen-Tobolsk, Yaskolbinsk, Tarski and Kurdaksk-Sargatsk subgroups. These subgroups, in turn, include a number of smaller units. In Tyumen-Turin Tatars subgroup, Tyumen, Yalutorovsk, and Turin Tatars can be distinguished. Ethnogenesis of Siberian Tatars is a complex, long process of interaction between different ethnic composition components that is reflected in a special "genetic portrait" of each group.

In this study, the Tobol-Irtysh Tatars are presented by Yalutorovsk and Tyumen subgroups (Bukhara Tatars) of Tyumen-Turin group (165 people). Tyumen-Turin Tatars and Bashkirs participated (or group later became members of the Bashkirs) in the formation of "Yalutorovsk"."Bukhara" is a ethno-class group within Tyumen, Isker-Tobolsk and Tara Tobol-Irtysh Tatars.We know that, historically, descendants from Samarkand, Urgench, Bukhara and other cities of Central Asia took part in its formation.

The gene pool of the groups was studied using 17 microsatellite (STR) markers of the Y-chromosome. According to the results of genotyping in Tyumen-Turin Tatars Tyumen region 14 Y-chromosome haplogroups were revealed, including Haplogroup 2 (C3, O3) belonging to Eastern Eurasian, and 3 (N1b, N1c and Q) to the north-Eurasian. West Eurasian haplogroups branch is represented by 8 - E1b1b1, G2a, J1, J2, J2b, I, R1a, R1b1b2. Haplogroup R2 may have South Asian origin. Gene pool of Ialutorovsk Tatars has reasonably extensive range of 14 Y-chromosome haplogroups and a lack of a positive haplogroup - none of the haplogroups found is predominant and "significant" for their gene pool. The most common haplogroups are J2-M172, R1a-M198 and N1c-LLY22g.

A study of polymorphism of Y-chromosome in population of Tyumensk Tatars showed that the main contribution to the formation of their gene pool is made by the G2a-P15 haplogroup. It is noteworthy that this haplogroup is similar to a number of peoples of the Caucasus: 66% of the gene pool of the Ossetians, ranges from 24% or more at the Adygei-Abkhazian peoples, and in the Turkic-speaking Karachai and Balkars found with a frequency of 31-33%. The haplogroup G is presented in a contemporary population of the Middle East. Another three haplogroups constituting the gene pool of the Tyumen subgroup characteristic of the indigenous population of the region. These are R1a-M198, N1c-LLY 22, N1b-p43 haplogroups. In general, the range of the Y-chromosome haplogroups in population of Tyumen-Turin Tatars demonstrated their genetic similarity to the Turkic peoples of Siberia. In particular haplogroup R1a-M198, N1b-p43, N1c-LLY22g are typical of the indigenous population of the South Siberia (Khakassia, Shor, Altai). However, the ratio of the haplogroups differs. This diversity may be caused by special aspects in ethnogenesis: unequal contribution of tribes and peoples who lived previously in these areas.

This work was supported by grant RFBR number 14-06-00272a.

GENETIC AND MORPHOLOGICAL DIFFERENTIATION AND SYSTEMATICS OF THE NORTH CHINA ZOKOR *MYOSPALAX PSILURUS* (RODENTIA SPALACIDAE)

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Both genetic and morphometric methods have been applied for studying of differentiation in the North China zokor, *Myospalax psilurus*, which includes zokors from the Russian Far East (Primorsky Krai), North China, Eastern Mongolia and Russian Transbaikalia (Zabaykalsky Krai). There are two subspecies of North China zokor which were described earlier: *M. p. psilurus* and *M. p. epsilanus*. The taxonomic statuses of these forms are debated. All zokors under study had diploid number of chromosomes 2n=64, but significant differences in the karyotype structure have been revealed between populations from Transbaikalia (9-12M-SM, 10-13 ST, 8-9A: "*epsilanus*") and Far East region (9M-SM, 13-14ST, 8-9A: "*psilurus*"). These populations are clearly differed by spectra of blood serum proteins (transferrins): all samples from Primorsky Krai were found to be monomorphic by TF-B, while the samples from Zabaykalsky Krai possessed TF-C only (Pavlenko, Korablev, 2003). Marginal populations from Zabaykalsky Krai and Primorsky Krai have significant differences according to RAPD-PCR analysis. Genetic distance between them is high and it may be compared with the distance between another zokor species, *M. aspalax* and *M. armandii* (Tsvirka et al., 2011). These differences can be considered as corresponding to interspecies level. All samples were shown to be divided into two clades according to the geographical distribution of samples. A similar pattern of differentiation is detected by sequencing of mitochondrial markers: hypervariable region D-loop and cytochrome *b* (Tsvirka et al., 2009, 2014).

The main morphological differences within the North China zokor were found in hard palate features: *foramina incisive* size and construction, length of maxillary bone (Puzachenko et al., 2009; 2014). According to the available data, zokors from Primorsky Krai are identical to the zokors from the southern part of the species range and are considered as typical *M. p. psilurus*. The zokors from the Great Khingan region must be considered as *M. p. epsilanus.* The zokors from Zabaykalsky Krai and Eastern Mongolia are definitely close to *M. p. epsilanus* by the morphometric data but have some specific features in *foramina incisiva* construction, the length of the M1 and M2 and the total length of the upper tooth-row.

Having considered all available data, we support the previously proposed hypothesis (Pavlenko, Korablev, 2003) that peripheral populations of North China Zokor from Transbaikalia and South of Russian Far East belongs to separate species *M. psilurus* Milne-Edwards 1874 and *M. epsilanus* Thomas 1912 or two semispecies in *statu nascendi* (Pavlenko et al., 2014). For further revision, integrated genetic and morphological research of zokors from the Chinese part of the species range should be done.

CHROMOSOMAL HYBRID ZONES IN THE COMMON SHREW IN RUSSIA

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The common shrew, *Sorex araneus* (Eulipotyphla, Mammalia) is one of the very good model species to study the role of chromosomes and whole-arm chromosomal rearrangements including Robertsonian (Rb) fusions and whole-arm reciprocal translocations (WARTs) in speciation. This species is subdivided into more than 70 parapatric chromosomal races distinguished by Robertsonian and WART chromosomal rearrangements. When races come into contact, interracial hybrids are produced with different sets of rearranged chromosomes in their karyotypes. At the moment, about 30 chromosomal hybrid zones (an area where interracial hybrids occur) between different races are known and 17 of them are located in Russia. Hybrids between races are often complex heterozygotes, forming medium-to-long chain or ring configurations at meiosis, due to differing sets of metacentrics in the hybridising races. Such hybrids could be expected to have meiotic abnormalities and consequently low fertility. Therefore, studies on meiosis and fertility in such hybrids provide keys in understanding the evolution of reproductive isolation between chromosomal races. Here, I report about results of cytogenetic and meiotic studies, which were conducted by our team on hybrids from several hybrid zones in central European Russia.

The study was supported by RFBR (15-04-04759) and President Grant for Russian Distinguished Young Scientists MK-4496.2015.4.

MICROEVOLUTION PROCESSES IN ENDEMIC MOLLUSCS OF THE GENUS *BAICALIA*

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Genus *Baicalia* Martens, 1876 belongs to endemic family Baicaliidae, which is a classic species group in Lake Baikal (Zubakov et al., 2003). According to the latest taxonomy it consists of five species and two subspecies: *Baicalia carinata*, *Baicalia turriformis*, *Baicalia carinatocostata* W. Dyb., 1875, *Baicalia dybowskiana*, *Baicalia rugosa* Lindh., 1909 and *B. turriformis inornata*, *B. dybowskiana carinatoides* (Sitnikova et al., 2004). Ecological properties of *Baicalia* species do not differ dramatically. There are no differences in food preferences and water depth range occurrence. The species of *Baicalia* mainly differ in reproductive behavior and substrate preferences. *Baicalia dybowskiana* and *B. carinata rugosa* attach eggs to sandstones, *B. turriformis* attaches them onto hard surface of rocks or stones, *B. carinatocostata* attaches eggs to sand particles and *B. carinata*, known for its peculiar behavior, attaches eggs to shells of snails of the same species. Reproductive behavior of *B. turriformis inornata* (Kozhov, 1962) and *B. dybowskiana carinatoides* (Kozhov, 1962) is still unknown. Ecological and biological peculiarities, and distribution patterns of these gastropods in Lake Baikal make them interesting for studies on speciation scenarios.

We studied the evolutionary history of endemic *Baicalia* gastropods using information about nucleotide sequences of two mitochondrial (cytochrome *c* oxydase subunit I (COI), large ribosome subunit (16S)) and two nuclear (internal transcribed spacer 1 (ITS1), intron of ATP synthase subunit alpha (ATPSa)) molecular markers.

Phylogenetic analysis based on both mitochondrial and nuclear molecular markers showed that *B. dybowskiana carinatoides* and *B. turriformis inornata* formed the same clade with *B. dybowskiana*. Also, we have found dramatic discrepancy between mitochondrial and nuclear phylogenies, which may be explained by secondary asymmetric break of reproductive isolation between *B. dybowskiana* and *B. turriformis* on the one side, and females of *B. carinata* on the other side. In both cases mitochondrial genomes of two relatively rare species (*B. turriformis* and *B. dybowskiana*) were swept away by mitochondrial genomes of *B. carinata*, the most numerous, flexible and opportunistic species. Therefore, we suppose that mitochondrial introgression events took place in evolutionary history the genus *Baicalia*.

POPULATION GENETIC STRUCTURE OF SOCKEYE SALMON *ONCORHYNCHUS NERKA* (WALBAUM) IN KAMCHATKA PENINSULA, CHUKOTKA PENINSULA AND THE KURIL ISLANDS

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Sockeye salmon *Oncorhynchus nerka* (Walbaum) is one of the most important commercial fish species in the Russian Far East. Throughout the species distribution range, populations of this species form a complex multi-level genetic structure. Allelic variations at seven microsatellite loci (*Ots*107, *Oki1*a, *Oki1*b, *One*104, *One*109, *Oki*6 and *OtsG*68) were examined in twenty-eight samples taken in water bodies of Kamchatka Peninsula, Chukotka and the Kuril Islands. The average differentiation between populations θ_{st} at seven loci was 5.34%, and is statistically significant. Hierarchical analysis (AMOVA) showed that variation due to regional differences was 5.02%. In the studied populations, differentiation exists mainly at the regional level, as well as at the level of small geographically or historically closely related areas, and in major spawning rivers and lake basins. We determined that there exist genetically distinctive populations of *O. nerka* in the study area. Values of pairwise genetic differentiation F_{st} showed that samples from Sarannoe Lake (Commander Islands), Krasivoe Lake (Kuril Islands), and Avacha River (Kamchatka Peninsula) differ from all other populations and are characterized by maximum values of this parameter. Results from this study can be used for stock identification of *O. nerka* in future.

GENETIC DIVERSITY OF CLOSELY RELATED RHODODENDRON SPECIES FROM SIBERIA AND THE FAR EAST OF RUSSIA

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The genus *Rhododendron* is the most numerous in the family Ericaceae; it includes more than 1000 species, 21 of which grow in Russia. Closely related species of rhododendrons with similar morphology and uncertain taxonomic status deserve special interest of researchers. Among these species, widely distributed representatives of the subgenus *Rhododendron*, section *Rhododendron*, subsection *Rhodorastra: Rhododendron ledebourii* Pojark and *Rhododendron dauricum* L. occur in Siberia, and *Rhododendron sichotense* Pojark and *Rhododendron mucronulatum* Turcz. occur in the Far East. The taxonomic status of *R. ledebouri* and *R. sihotense* remain controversial and in some systems these species are only considered as ecotypes of *R. dauricum*. In our study the genetic variation in these four rhododendron species was examined. After application of ten universal primer pairs for chloroplast regions four pairs were selected for further analysis. Fragments Tab CD, Tab AB, CS and K1K2 reveal polymorphism between and within species. The size of amplified fragments varied from about 550 to 1000 base pairs. Ten samples from different parts of each species range were analyzed (totally 160 individuals) per fragment. Two fragments (Tab CD and CS) contain mutations (nucleotide substitutions and deletions) specific for each species. In Tab AB and K1K2 fragments polymorphism within species was reveal. Data from the sequence analysis show variation and differentiation that confirm the independence of all the examined species. Genetic data were compared with the literature data for morphological features and the history of the formation of the species ranges.

THE ARCTIC CHAR (*SALVELINUS ALPINUS*) FROM THE POLAR URALS AND ITS POSITION IN THE SPECIES PHYLOGEOGRAPHIC STRUCTURE

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First data on the mitochondrial DNA (mtDNA) variability in the Arctic char (*Salvelinus alpinus*) from the Polar Urals are presented. MtDNA sequences (507 bp of the control region) were obtained from 18 individuals from two localities: Bolshoe Shuchye Lake and Bolshaya Hadata River. We describe four unique haplotypes of the mtDNA control region, and compare the original data (18 sequences) with the data reported in previous mtDNA studies (92 sequences) (Brunner et. al., 2001; Alekseyev et al., 2009).

Our results show that the previously described Atlantic and Siberian mitochondrial groups (Brunner et. al., 2001) are not separated from each other. Moreover, they do not represent different sub-groups in the Eurasian group as it was proposed by Alekseyev et al. (2009). We show that there is one Atlantic-Siberian supergroup, which has high genetic diversity. In the Brunner's work (2001) Atlantic and Siberian groups were separated from each other because the data were collected from the distant regions (from the western and eastern borders of the Atlantic-Siberian supergroup range). We show that the differences between these groups decrease after adding the data from some new regions such as the high Arctic Archipelago of Svalbard, Taimyr Peninsula (Alekseyev et al., 2009) and the Polar Urals (our data). Our results show that the major part of the Northern Eurasia and possibly the seas of the Arctic Ocean, are inhabited by one Atlantic-Siberian mitochondrial supergroup of the Arctic char.

CHARACTERISTICS OF EUROPEAN GRAYLING (*THYMALLUS THYMALLUS*) OF KULOY AND NORTHERN DVINA RIVERS (WHITE SEA BASIN), BASED ON MITOCHONDRIAL DNA FRAGMENTS ANALYSES

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Ichthyological studies (as well as fisheries studies) in European North of Russia have traditionally concentrated on the most valuable commercial species, namely Atlantic salmon (*Salmo salar* L.) and Brown trout (*S. trutta* L.). European grayling (*Thymallus thymallus*) populations inhabiting large territories remain underexplored. Grayling populations in the Asian part of Russia were morphologically and genetically studied extensively; however, genetic studies on European grayling in Russia are still based on small quantity of samples. Recently, mtDNA haplotypes characteristic for Siberian grayling (*Th. arcticus*) were found in two specimens of Nothern Dvina River. It was then suggested that hybridization zone between European and Siberian grayling exists more to the west than it has been supposed before. But the question of hybridization zone border is still not resolved. This study includes 5 European grayling (*Th. thymallus*) samples representing Northern Dvina and Kuloy rivers basins, 2 Siberian grayling (*Th. arcticus*) samples representing river lakes of the Ob' river basin, 1 grayling (*Thymallus* sp.) sample of the upper Yenisey River. MtDNA analysis is conducted based on 3 phragments ATP6, CR, COI. ATP6 gene and control region (CR, D-loop) of mtDNA were chosen along with COI as a more conservative fragment which is conventionally used in DNA barcode project. CR and ATP6 sequences for fish from tributaries of the Northern Dvina and the Kuloy rivers were obtained. In grayling samples representing European part of Russia, only European grayling haplotypes were found. Only one haplotype was found among CR sequences and three haplotypes were found within ATP6 gene. For the first time genetic data were obtained for the Kuloy river grayling population. Though it could seem more probable to find Siberian grayling haplotype in rivers, which, like Kuloy, are situated closer to the Urals Mountains, haplotypes close to Siberian grayling were not found in the examined specimens.

GENETIC VARIABILITY BETWEEN POPULATIONS OF *PLANORBARIUS CORNEUS* SNAILS (GASTROPODA, PULMONATA)

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Since *Planorbarius corneus* L., 1758 is a polymorphic species, morphological criteria are insufficient for its specific taxonomical identification. Two controversial viewpoints on *P. corneus* taxonomical status prevail in the literature. According to the first opinion *P. corneus* is a highly polymorphic species, according the second one, the snails form a superspecies (supra-species) complex *Planorbarius* with several subgroups. In this study, genetic variability in three populations of *P. corneus* was analyzed using internal transcribed spacers of rDNA (ITS1 and ITS2) and random amplified polymorphic dimorphism analysis.

Chromosome DNA in three populations of *P. corneus* snails from Leningradskaya Oblast and Kaliningradskaya Oblast (Russia) were investigated using specific primers for rDNA and random primers. The full sequences of ITS1 and ITS2 were obtained and analyzed. No intrapopulation differences in *P.corneus* ITS rDNA sequences were detected. Several single nucleotide polymorphisms were revealed between ITS1 and ITS2 in mollusks from different populations. The revealed minor polymorphisms among ITS in mollusks from different populations do not allow to define taxonomically distinct groups within the species *P. corneus*. Population variability was analyzed based on of RAPD-profiles. Three random primers for the detection of interpopulation differences between snails form different populations were selected.

SOME ASPECTS OF GENETIC POLYMORPHISM IN TWO RELICT PLANT SPECIES FROM BAIKAL SIBERIA

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The study was aimed at preliminary estimation of genetic polymorphism in relict plant species of Baikal Siberian flora: *Anemone baicalensis* Turcz. ex Ledeb. and *Eranthis sibirica* DC. Plants were collected along the northern foothills of the Khamar-Daban Ridge (southern coast of Lake Baikal).

Firstly, levels of spatial isolation of the studied species populations were estimated by the net mapping on the model area between Bolshoi Mamai and Maliy Mamai rivers. The mapping showed that populations of both species from adjacent floodplains strongly overlapped, and the gap between populations takes place only in the lower flow of the rivers.

Genetic polymorphism was estimated using comparative analysis of DNA sequences of ITS regions and chloroplast intergenic spacers *(rpl20-rps20* and *psbA-trnH*). The analysis of ITS1 and ITS2 regions in *E. sibirica* showed the existence of both inter- and intraindividual polymorphism; however, the level of interpopulation polymorphism was higher. The differences found between populations from Besymiannaya and Bolshoi Mamai Rivers were high and corresponded rather to the interspecies level. The interpopulation polymorphism of *rpl20-rps20* was very low. Therefore, we found much higher level of interpopulation polymorphism than we expected based on the geographical distribution of *E. sibirica* populations. This case can be explained by the limited gene flow through the populations and possibly limited concerted evolution of ITS regions.

The analysis of ITS polymorphism in *A. baicalensis* (populations from Besymiannaya and Bolshaya Rechka rivers) showed interpopulation polymorphism, which, however, did not exceed 1%. We also found a very high ITS similarities with the East Asian *A. flaccida* (*A. baicanensis* aggr.). The sequences of *psbA-trnH* in populations of *A. baicalensis* s.str. and in *A. flaccida* were identical. We can assume there exists intensive genetic homogenization between the populations of *A. baicalensis*. The obtained results showed the necessity of taxonomic revision of the *A. baicalensis*.

The reported study was partially supported by RFBR, research project No. 14-04-31350-mol_a.

EFFECTS OF EMBRYO TRANSFER AND IN VITRO CULTURE ON THE DEVELOPMENT OF HYPERTENSIVE PHENOTYPE IN RATS GENETICALLY PREDISPOSED TO HYPERTENSION (ISIAH STRAIN)

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The influence of assisted reproductive technologies (ART) on the development of hypertensive phenotype has not been determined so far. The aim of this study was to investigate long-term effects of embryo transfer at preimplantation stages of development either combined or not combined with in vitro culture on the genetically determined traits of the adult rats with inherited stress-induced arterial hypertension (ISIAH strain). ISIAH rat preimplantation embryos were collected at two-cell stage and cultured *in vitro* 48 h in rat one-cell culture medium (R1ECM) up to morula stage with or without epidermal growth factor (EGF) or were collected *in vivo* at morula stage; finally these *in-vitro*-derived and *in-vivo*-derived morulas were transferred to recipient female rats. The pups born after transfer of non cultured and cultured embryos were compared with naturally born ISIAH rats. Ultrasonic vocalization (USV) of pups in isolation test was estimated at the age of 13 days, blood pressure was measured in all groups with tail-cuff method at the age of 60 days; thereafter the animals were euthanized and Fkbp5 gene expression was measured in myocardium by RT-PCR. Two-cell ISIAH embryos developed up to morula stage during 48 hours of *in vitro* culture; however, some of them achieved blastocyst stage. Supplementation of culture medium with EGF improved the development of ISIAH embryos *in vitro*: more embryos developed and the rate of their development was accelerated, although this effect was mostly observed after 72 hours of culture. USV in pups born after transfer of *in-vivo*-derived embryos was at lower frequency as compared with all the other groups. At the age of 60 days all rats possessed hypertension. Expression of Fkbp5 gene in myocardium in adult ISIAH rats born after transfer of *in-vivo*-derived morulas (non cultured embryos) was significantly higher than in two other groups: naturally born ISIAH rats and rats born as a result of *in-vitro*-derived embryos transfer. Elevated Fkbp5 gene expression in myocardium as well as low frequency of USV during early postnatal development may be considered as markers of stress condition. Our data suggest that some combinations of ART, applied to ISIAH rats at preimplantation stages of development, affect their postnatal development and their adult phenotype.

Supported by the Russian Foundation for Basic Research (Projects No.13–04–00685; No. 15-04-05509).

INTRASPECIES GENETIC DIFFERENTIATION OF NORTHERN RED-BACKED VOLE POPULATIONS INHABITING THE URALS BASED ON mtDNA

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The northern red-backed vole (*Clethrionomys rutilus*, Pallas, 1779) is a wide-ranging Holarctic species encompassing 20 subspecies, designated based on variation in the color of the fur, body size and craniological features. Among them, hardly more than eight-ten subspecies fall into real taxonomic categories (Gromov, Erbaeva, 1995). A broad overlap of values in morphological traits, used to identify a subspecies, hampers subspecies differentiation, in particular, if information about a geographical sampling site is absent. Within the Urals and neighboring territories, the following zones are defined: steppe, forest-steppe, forest, forest-tundra and tundra along with distinct altitudinal belts. Therefore, the Urals might be a habitat for a few northern red-backed vole subspecies (Bolshakov, 1972). Genetic analysis of *C. rutilus* samples from different geographical zones is very important as, unlike morphological analysis, it is based on the variability of genetic markers, which are significantly less affected by environmental factors.

We conducted genetic studies of *C. rutilus,* based on the nucleotide variability of mtDNA cytochrome b (*cyt b*) fragment (866 bp) of 44 voles from eight localities within Polar (near Labytnangi town), Middle (Sverdlovsk region) and Southern (Chelyabinsk region, Bashkortostan) Urals along with Southern Yamal (the Yerkuta River). Phylogenetic analysis of *cyt b* sequences were conducted employing the Bayes method (program MrBayes 3.2). The evaluation of phylogenetic tree parameters was conducted with the help of Traser v 1.6 program. The sequences of *C. rufocanus cyt b*, obtained from GenBank, were used as an outgroup.

Among 44 analyzed nucleotide sequences of *cyt b* gene of *Cl. rutilus* we revealed 23 haplotypes including 17, which are not found in the GenBank database. The obtained phylogeographic data, however, do not point to the existence of a separate subspecies of the *C. rutilus* group within the Urals. According to the obtained phylogenetic tree of *C. rutilus,* representatives from different subspecies do not fall into separated monophyletic groups, and even voles from the same locality also do not group together. To conduct further, more specific phylogenetic analysis of *C. rutilus,* inhabiting the Urals, it is necessary to use a higher amount of samples per locality, include more sampling sites and employ new DNA markers, in particular nuclear gene(s).

Consequently, the obtained data on genetic variability of *cyt b* point to the absence of a distinct phylogeographic structure of *C. rutilus* inhabiting the Urals in the geographic gradient from the north to the south.

This study was supported by the Russian Foundation for Basic Research (projects nos. 14-04-00614 and 13-04-00847).

PHYLOGENY OF BAIKALIAN ENDEMIC AMPHIPODS BASED ON SEQUENCES OF THEIR MITOCHONDRIAL GENOMES

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Lake Baikal is the most ancient and the deepest freshwater reservoir, which is known as natural laboratory of adaptive radiation. Amphipods (Crustacea) are the most numerous (about 350 species) and diverse invertebrates’ species flock of the lake (Kamaltynov, 2001). However, the phylogenetic relationships in this group of animals remain poorly resolved due to extremely high intraspecific genetic diversity and relatively ancient origin of current amphipod species (McDonald et al., 2005). Here we report the phylogenetic inferences involving the sequences of mitochondrial (mt) genomes. Species of different lifestyles and occupying different ecological niches are presented on the tree.

Complete and near complete mtDNA sequences from nine Baikal amphipod species were obtained using next-generation sequencing (Illumina). Mt genomes of given species have different length which mainly due to the length variability at the putative control region. Three out of nine species have altered mt genes order in comparison to the pancrustacean background pattern (Kilpert, Podsiadlowski, 2006) and may be unique within the known ones of crustaceans.

Phylogenetic tree based on 13 concatenated protein-coding genes of mtDNA was entirely resolved with highly supported nodes. Topology of the tree corroborates with the modern taxonomy of the group and may be used in order to solve existing taxonomic problems.

We also estimated nucleotide diversity range for every mt protein-coding gene of species in this study. The highest value was observed at *nad*2, *nad*6 and at the last third of *nad*5 genes, and the lowest one was at *cox*1, *cox*3 and *cob* genes. Thus, we can recommend utilize the former set of mt genes as molecular markers for population studies and the later set for phylogenic inferences of families and maybe other high taxa. The comparison to other amphipod genomes sequenced up to date indicates dramatic difference in the diversity patterns among the genes.

DIVERSITY OF *APODEMUS PENINSULAE* (RODENTIA) B CHROMOSOMES DNA

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B chromosomes are dispensable components of the genome exhibitingnon-Mendelian inheritance. B chromosomes are additional to standard karyotypes (A) and may vary in size, number and morphology even between cells of the same individual. Korean field mouse (*Apodemus peninsulae*) is a species with high frequency of animals with B-chromosomes, which differ in number, morphology and DNA composition in different geographical regions.

We summarized the literature data on the DNA content of *A. peninsulae* B chromosomes after fluorescent in situ hybridization (FISH) of DNA probes with metaphase chromosomes of the samples from Siberia, Transbaikalia, and the Far East, including Russia, South Korea and Japan (Karamysheva et al, 2002; Trifonov et al., 2002; Matsubara et al., 2004; 2008; Rubtsov et al., 2004; 2005; 2015). DNA library includes microdissection DNA probes from B chromosomes and their regions, as well as from pericentromeric C-positive autosomal regions.

It was shown that B chromosomes and their regions contain various repeated sequences. Several B chromosomes contained active NORs. Repeats that were typical for the weakly condensed regions of macro and micro B chromosomes were detected. DNA composition of micro B chromosomes was different from that of macro B chromosomes. Repeated sequences that were the most typical for B chromosomes, are also presented in A chromosomes. Homology between pericentromeric DNA from B and A chromosomes was revealed. At least one part of the repeats characteristic for the arms of the majority of B chromosomes of species from the Far East are also present as dispersed repeats in C-negative chromosomal regions of the basic set. But some regions were described in several B chromosomes that have no DNA homologous to the repeats in DNA library probes.

We revealed that B chromosomes in population from the Russian Far East and South Korea exhibit low variability in DNA content; however, DNA composition of B chromosomes in populations of Siberia, Transbaikalia and Japan are highly variable. These data open up possibilities to discuss the pathway of origin of *A. peninsulae* B chromosomes in different regions.

PHYLOGENETIC RELATIONSHIPS OF *PARADICLYBOTHRIUM PACIFICUM* AND *DICLYBOTHRIUM ARMATUM* (MONOGENOIDEA: DICLYBOTHRIIDAE) INFERRED FROM 18S RDNA SEQUENCE DATA

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The Diclybothriidae (Monogenoidea: Oligonchoinea) includes specific parasites of fishes assigned to the ancient Order Acipenseriformes. Phylogeny of the Diclybothriidae is still unclear despite several systematic studies based on morphological characters. Together with the closely related Hexabothriidae represented by parasites of sharks and ray-fishes, the position of the Diclybothriidae in different taxonomical systems has been a matter of discussion. Here, we present the first molecular data on the Diclybothriidae. The SSU rRNA gene was used to investigate phylogenetic position of *Paradiclybothrium pacificum* and *Diclybothrium armatum* among the other Oligonchoinea. Complete nucleotide sequences of *P. pacificum* and *D. armatum* demonstrated high identity (98.53%) with no intraspecific sequence variability. Specimens of *D. armatum* were obtained from different hosts (*Acipenser schrenckii* and *Huso dauricus*); however, variation by host was not detected. The sequence divergence and phylogenetic trees data show that Diclybothriidae and Hexabothriidae are more closely related to each other than with other representatives of Oligonchoinea.

GENETIC BACKGROUND OF VARIABILITY THE PHENOTYPIC EXPRESSION OF NONSPECIFIC REACTIVITY OF AN ORGANISM IN THE INDIGENOUS GROUPS OF VARIOUS TERRITORIES IN THE VOLGOGRAD REGION, RUSSIA

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In the previously performed own researches it was justified the integrativity of structural and functional manifestations of the level of general non-specific reactivity of an organism (LGNRO). The pain threshold was used as an indicator of LGNRO. The LGNRO was differentiated by the degree of manifestation of pain sensitivity to "high", "medium" and "low", where a high LGNRO meets the minimum values of pain threshold, a low LGNRO meets maximum values and medium met the intermediate values of nociceptive reactivity. It was statistically proven that intraspecific distribution of pain threshold in humans and animals corresponds to the normal distribution. Significant differences between the distribution of the pain threshold in different populations within single species of animals and humans were obtained. As a result of a number of experimental researches the mechanisms of the central and vegetative organization and personalization of nociceptive reactivity of the organism were identify. Taking into account that population characteristics are strictly determined by constant environmental factors, the hypothesis that LGNRO distribution among different populations of one species will reflect the specificity of background exogenous influence has been suggested. The territory of Volgograd Oblast which has unique natural zones: the forest-steppe, steppe, and semi-desert zones that lead to significant differences in living conditions for population has been studied. As a result of complex analysis of climatic and geographic factors the territories with specific characteristics – Western, Central and Eastern were distinguished. Following above arrangements, the sample populations of aboriginal inhabitants of these territories are formed, and their LGNRO was estimated. This research has found out the presense of differences for population distributions of LGNRO. The Western population is mainly characterized by high LGNRO and the Central and Eastern are characterized by gradual decline of LGNRO. The conducted literature review revealed six SNPs in four genes:- rs1851048 and rs6777055 in voltage-gated calcium channel subunit α2δ3; rs2562456, which is in disequilibrium linkage with a gene encoding zinc finger protein 429; rs6923492 and rs362962 in metabotropic glutamate receptor type 1 gene; rs6314 in serotonin receptor type 2A gene. In order to identify the genetic basis that forming the level of general nonspecific reactivity of the organism the experimental research of polymorphism of several genes associated with pain sensitivity was undertaken**.** Our results indicate that people with high level of general nonspecific reactivity of the organism usually had AA variant of rs6777055 and rs1851048, allele C in rs362962 and allele T in rs6314 SNP. Individuals with low reactivity in most cases had AA variant of rs2562456 SNP. Thus, the genetic background of variability of the population manifestations LGNRO at aboriginals of certain areas of the Volgograd region is identified.

The study was done with financial support of *Russian Foundation for Basic Research* in the framework of a research project number 15-06-08034.

MITOCHONDRIAL DNA SEQUENCE VARIATION AND DEMOGRAPHIC HISTORY OF KALUGA AND AMUR STURGEON (ACIPENSERIDAE): A COMPARISON

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The variability of mitochondrial DNA control region (CR) of kaluga *Acipenser dauricus* Georgi, 1775 and Amur sturgeon *A. schrenckii* Brandt, 1869, endemics of the Amur River, classified as critically endangered by the IUCN Red List of Threatened species – was studied. Similar sample sizes in Kaluga (n=122) and Amur sturgeon (n=112) were analyzed. In Kaluga, there were less number of haplotypes (27 versus 73), and lower levels of haplotype (0.927±0.009 versus 0.976±0.009) and nucleotide (0.004±0.002 versus 0.019±0.010) diversity. Calculated in various ways, values of population parameter θ ($2N_{ef}\mu$) for kaluga were 2.8–9 times (5.3 times, on the average) lower than those of Amur sturgeon. Therefore, if rates of evolution of CR in these sturgeon species are the same, then it can be argued that the long-term female effective number (N_{ef}) in kaluga is about the same magnitude (~3–9.5 times, taking into consideration different length of their generations) lower than the N_{ef} in Amur sturgeon. Suchlike conclusion stems from the comparison of bayesian skylines: the N_{ef} in kaluga (ca. 2,800) is about 8 times less than the N_{ef} in Amur sturgeon (ca. 23,000). These estimates are consistent with calculations of the number of kaluga and Amur sturgeon made from the catch data obtained in the late 19th century. In contrast to Amur sturgeon, which is a typical benthophage, kaluga is a fish-eating predator, occupying the top of the trophic pyramid in the ecosystem of the Amur River. The abundance of top predators can not be high. Probably this is what induces kaluga's and Amur sturgeon's differences in the level of variation CR. It should be noted that if the current estimates of abundance of the Amur River sturgeons are correct (the ratio of mature kaluga to Amur sturgeon is ~1:1 (Koshelev et al., 2014)), then their dramatic decline in the 20th century was more due to Amur sturgeon than kaluga.

This work was supported in part by the Far Eastern Branch of Russian Academy of Sciences (grants no. 12-I-P30-04 and 15-I-6-079).

THE ROLE THE ISOLATES IN PHYLOGEOGRAPHICAL STRUCTURE OF *MICROTUS FORTIS* BUCHNER, 1889

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The study of variation at different levels provides insight to the fundamental problems in evolutionary biology. Phylogeography is a powerful tool for identifying the type of intraspecific structure. In recent years it has been used successfully in studies of different species.

The reed vole *Microtus fortis* Buchner, 1889 is a polytypical species widely distributed in Mongolia, Korea, China and Russia. Population has a mosaic distribution range, and on the periphery of the distribution area there exist several isolated populations. The reed vole is of great interest to phylogeography, since it lives with a wide area and has a number of different isolates.

Phylogeography of *Microtus fortis* was studied on the basis of the variability of mtDNA control region. A total of 368 specimens of the reed vole were sampled in 12 island and 40 mainland localities. High level of intraspecific variation and differentiation was observed in the species. Six phylogenetic lines were identified. Four lines are characteristic of the southeastern China and two for the Trans-Baikal area, the Russian Far East and northeastern China. Rare haplotypes in the northern part of the range were found in some of the reed vole peripheral isolates. The hypothesis on conservation of the ancient diversity in peripheral isolates was proposed.

Work was partly supported by grants RFBR 15-04-03871.

MOLECULAR CHARACTERIZATION OF *METAGONIMUS YOKOGAWAI* (TREMATODA: HETEROPHYIDAE) IN THE SOUTHERN RUSSIAN FAR EAST

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Metagonimus species cause severe gastrointestinal disorders and have important epidemiological significance. In the East and South-East Asia seven trematode species have been described in the genus *Metagonimus*. Three species of adult worms (*M. yokogawai* Katsuradai, 1912, *M. takahashii* Suzuki, 1930 и *M. miyatai* Saito, Chai, Kim, Lee & Rim, 1997) are morphologically similar. Also morphological similarity was shown for the cercariae and metacercariae stages of *M. yokogawai* and *M. takahashii.* Therefore, both morphological and genetic data are required for the species identification. Only *M. yokogawai* was found in the southern Russian Far East. The present genetic investigation of *M. yokogawai* from different regions of the southern Russian Far East has confirmed the results from the morphological species identification.

Adult flukes of *M. yokogawai* completely conformed to the morphological description. We analyzed 64 complete ITS1-5.8S-ITS2 (1,134 bp) and 5 partial 28S (1,297 bp) gene rDNA sequences for *M. yokogawai* from different regions in the southern Russian Far East. The obtained sequences were compared with available data in GenBank.

ITS1-5.8S-ITS2 rDNA sequences from Russia were similar to each other (99.7%), but sufficiently different (1.85%) from the unique complete ITS1-5.8S-ITS2 rDNA sequence from China (KJ631740). ITS1 sequences from China (KJ631740, KJ631742) and Korea (JN638319, JN638321) were different from Russian samples in 11 (1.6%) and 14 variable sites (2%), respectively. The intragenomic polymorphism was identified in two ITS2 sequences from Russia, and other nucleotide sequences were identical. Partial ITS2 sequences for Chinese, Korean and Japanese samples were different from the Russian ITS2 sequences by 2.3-2.9%. Genetic distances between the samples from China, Korea and Japan varied from 0.8% to 2%. The complete 5.8S gene and partial 28S gene rDNA sequences were identical in Russian samples. Unique sequences of 5.8S gene (KJ631740, China) and 28S gene (HQ832639, Japan) were different from Russian samples at 1 (0.6%) and 8 (0.6%) variable sites, respectively.

Described genetic distances correspond to the genetic distances between *Metagonimus* species. Therefore, there is a probability for mistakes in species identification. We can’t estimate the mistakes’ significance, since morphological description of *M. yokogawai* is absent in most of the genetic studies.

VARIABILITY OF mtDNA IN *MICROTUS ARVALIS OBSCURUS*: NEW DATA AND PROSPECTS FOR FUTURE RESEARCH

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New data on mitochondrial DNA variability (cytochrome *b* gene and control region) in the common vole (*Microtus arvalis obscurus*) are obtained and compared to the published data on the phylogenetic structure of the species (Tougard et al., 2013) with the addition of new findings (Zhumabekova et al., 2013). Our data include more than 50 samples from 20 localities in Pre-Urals, Urals and Trans-Urals (51°07’ - 60°38' N; 48°38' - 66°33' E) and encompass the area inhabited by the Sino-Russian (SR) clade of *M. arvalis obscurus*, which was previously established by Tougard et al. (2013).

The internal genetic structure of SR clade of *M. arvalis obscurus* is investigated in this work in two ways:

1. Variability of cytochrome *b* gene.

Analysis of cytochrome *b* gene variability (original data and sequences presented in GeneBank (Haynes et al., 2002; Tougard et al., 2013; Zhumabekova et al., 2013)) confirms the conclusion made by Tougard et al. (2013) about the low genetic diversity within the SR clade of *M. arvalis obscurus*. Although the SR clade is strongly homogeneous, the sequences from northwestern China (Tougard et al., 2013) and northeastern Kazakhstan (Zhumabekova et al., 2013) form a slightly different group of haplotypes.

2. Variability of the two mtDNA markers.

The analysis of both cytochrome *b* gene and control region of mtDNA based on original data only suggests that, in the study area (Pre-Urals, Urals and Trans-Urals, see above), there are three different and partially sympatric mtDNA lineages found: Southern lineage (Southern Urals); Eastern lineage (Trans-Urals and partially north, middle and southern Urals); Western lineage (Pre-Urals and partially north and middle Urals).

Our results suggest that joint use of the two mtDNA markers (cytochrome *b* gene and control region) allows us to describe the three partially sympatric lineages within the SR clade of *M. arvalis obscurus*. This approach appears to be useful when investigating the species history and dispersal in the central part of the Northern Eurasia.

The work was supported by RFBR 14-04-00614 and by grant of President of the Russian Federation for support of the leading scientific schools НШ-2840.2014.4.

MECHANISM OF MITOCHONDRIAL GENOME SIMPLIFICATION IN EVOLUTION

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The set of genes in mitochondrial genome as well as the gene order varies strongly between major eukaryotic lineages. The repertoire of mitochondrial genes in most of cases is poorer than the repertoire of biochemical functions required. This is the result of gene traffic which leads from mitochondria to nucleus so that, after the commitment of a transfer, the gene products are transported into the mitochondrion to perform their function. So the latter undergoes changes in evolution which can be described as simplification. Transfer mechanism includes several steps, but the exact mechanism of deletion of mitochondrial “originals” coupled with the nuclear copies activation remains unknown. Transfers follow two general patterns: those that lead to resurrection of transferred gene function and those turning the transferred genes into mitochondrial pseudogenes. Here we attempt to describe some common elements of evolutionary pathway of the rearranging mitochondrial genome in invertebrates.

We compared gene annotations resulting Hidden Markov Model method implemented MITOS pipeline to the annotations of the same mitochondrial genomes meeting all GenBank requirements such as uninterrupted reading frame (not ultimately required by MITOS). GenomeView was used to compare prediction of genes based on HMM and MiTFi tRNA. The procedure was applied to the set of published invertebrate mitochondrial genomes. In course of the study many cases of likely non-functional mitochondrial gene copies were detected in mitochondrial genomes. Sometimes these loci were found on the opposite strand relative to a functional gene position in other taxa. The accumulation of nonsense mutations is believed to suggest that the gene is on its way to final extinction, and it's often nominated as pseudogenes. Such gene artifacts and gene rearrangements are evident signs of mitochondrial genome simplification in progress.

Some of the genes picked up by MITOS pipeline, especially tRNA genes, had e-value insufficient to believe that they were functional. MiTFi approach has found additional tRNA genes in every mitochondrial genome, mostly in inverted position. Recombination in the sites bearing combination of short reverse and direct repeats are hypothesized to increase the probability of strand swaps during recombination and thus serve as the hotspots for gene order rearrangements in mitochondrial genomes. The traffic mechanism of genes between mtDNA strands in invertebrates, its reduction in evolution and pseudogene generation in mitochondrial genome are discussed.

References:

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NEW DATA ON HYBRIDIZATION IN SCOTTISH BLUE MUSSELS ENABLE COMPARISON AMONG KNOWN HYBRID ZONES BETWEEN *MYTILUS EDULIS* L. AND *M. TROSSULUS* GOULD

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Blue mussels of the *Mytilus edulis* species complex are dominant members in the Northern seas coastal communities. The complex consists of three species: *Mytilus edulis* L., *M. galloprovincialis* Lamarck and *M. trossulus* Gould. Three *Mytilus* species fall into introgressive hybridization if their ranges overlap. Five hybrid zones between temperate *M. edulis* and *M. trossulus* are known in the North Atlantic: along the American coast, at the entrance to the Baltic Sea, along the coasts of Kola Peninsula (Russia), in Western Norway and in Scotland. The extent of hybridization varies among hybrid zones. The rate of hybrids is relatively low in Russia, Norway and America and high in the Baltic Sea. However, direct comparison among hybrid zones in America and Europe is complicated because they were studied using different sets of loci (diagnostic DNA markers and semi-diagnostic allozymes, respectively), and different statistical approaches. Situation in Scotland is most unclear because no formal analysis of the zone has been undertaken yet. The aim of our study is to analyze the Scottish material using both allozymes (phosphoglucomutase, *Pgm*, esterase-D, *Est-D*, octopine dehydrogenase, *Odh*, glucophosphate isomerase, *Gpi*) and DNA markers (ME15/16, Glu-5', ITS, MAL-I, EFbis, COIII) from the sets traditionally used for taxa delineation and to perform the analysis of hybridization using conventional statistical approaches. This enables indirect comparison between all hitherto studied hybrid zones between *M. edulis* and *M. trossulus*.

This work is supported by RFBR grant (13-04-00394_a) and grant of Saint-Petersburg State University (1.38.253.2014).

MICROSATELLITE ANALYSIS OF POPULATION STRUCTURE IN PACIFIC COD *GADUS MACROCEPHALUS*

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Pacific cod *Gadus macrocephalus* is widely distributed in the North Pacific and is of high interest for many consumer countries in this region as a commercially harvested species. According to the catch rates, Pacific cod is getting more valuable in the Russian Far East and its stocks are under severe pressure. Fishery management is based mostly on geographical and administrative features of this region. Currently, the number and boundaries of Pacific cod stocks are poorly studied, which makes long-term and sustainable fisheries impossible.

This study is one of the stages of a large project on Pacific cod population structure. According to the results of the previous study (genetic markers: gene of *Cyt b* and control region of mtDNA) Pacific cod populations in the Sea of Okhotsk can be divided into two groups: northern and southern. Also significant difference between cod samples from the northern part Sea of Okhotsk and samples from waters off the Kuril Islands (Pacific Ocean side) and Navarin-Anadyr region was revealed.

However, further molecular genetic studies were required in order to specify more accurately population structure, number and boundaries of Pacific cod stocks. Three multiplexes of microsatellites (total number is 12) are analyzed. Samples were collected from nine different regions of the North Pacific (Sea of Okhotsk, Bering Sea, waters off Kuril Islands, Canada and Sea of Japan). Results of this analysis are presented and discussed.

MOLECULAR ADAPTATION OF *CLONORCHIS SINENSIS* (TREMATODA: OPISTHORCHIIDAE) FROM RAT'S LIVER AND INTESTINE

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Clonorchis sinensis Cobbold, 1875, the Chinese liver fluke, is the epidemiologically important trematode from the family Opisthorchiidae. Clonorchiasis leads to liver and pancreas destruction. In the present research we studied molecular adaptation of *C. sinensis* to different digestive organs of definitive host using nuclear rDNA ITS1-5.8S-ITS2 region and complete mtDNA cox1 gene sequences. Unusual disposition of the parasite in rat's intestine was detected during dissection. We proposed that it can be a local adaptation of *C. sinensis*. The liver fluke populations from liver and intestine were analyzed using rDNA and mtDNA markers. Considerable distinctions between nucleotide sequences were not obtained, which indicates the absence of adaptation at the species level. However, we have found some features in genetic diversity for both markers.

NUMTS AS A SOURCE OF NEW MTDNA HAPLOTYPES: SIBERIAN RUBYTHROAT *LUSCINIA CALLIOPE* (MUSCICAPIDAE, AVES) CASE

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NUMTs are nuclear copies of mtDNA genes. For the first time the transfer of a large mtDNA fragment (7.9 kb) into the nucleus had been described for a domestic cat (*Felis catus*) and a hypothetical model was proposed for mtDNA fragment integration into the nuclear genome (Lopez et al. 1994). Today the mechanisms of genetic information transmission between the mitochondria and the nucleus are not well understood. Currently, many nuclear pseudogenes of mitochondrial genome have been detected and described in different groups of organisms (fungi, plants, arthropods, birds, mammals). Primarily NUMTs were considered as the hindrance for phylogenetic investigations (Bernt et al., 2013). Nevertheless, several NUMTs were identified as the cause of genetic diseases (Chen et al., 2005). A recent example shows that they can also become the source of new spliceosomal introns (Curtis, Archibald, 2010). Although the exchange of DNA between the nucleus and cellular organelles is considered a common phenomenon in all eukaryotes, the literature only describes the homologues of mitochondrial genes in the nuclear genome, and we don’t know any contradicting facts. We are presenting such evidence in our work for the first time now.

The research material is represented by museum samples of nesting birds of the nominate subspecies of the Siberian rubythroat *Luscinia calliope calliope* Pallas, 1776 (n = 21) from 18 points of the range. DNA isolation, amplification and sequencing were described previously (Spiridonova et al., 2013). Cloning of amplified fragments was carried out with a set of TA-cloning (Fermentas, Litva) according to the manufacturer's recipe. The preliminary analysis of mtDNA *cyt b* gene of several birds from the western and central parts of the range revealed a significant divergence, and provided the reason for a more detailed study.

Previously we have shown intraspecific division of the Siberian rubythroat into two highly differentiated haplogroups: the western one (*calliope*) and the eastern one which has two subgroups (*anadyrensis-camtschakensis* and *sachalinensis*) (Spiridonova et al., 2013). Since there are no geographical gaps between the ranges of the subspecies, and the entire eastern mainland part of the area (Anadyr, Kamchatka, Magadan region) is represented by populations with both variants of mtDNA, a significant divergence between haplotypes of *calliope* and *anadyrensis-camtschakensis* has no logical explanation. In addition, there is still an unresolved question of the origin of the *sachalinensis* haplotype found only on Sakhalin Island and missing in the close mainland part.

Analysis of numerous *cyt b* gene sequences for western samples of *L.c. calliope* revealed double peaks in the taxon-specific sites of sequences, indicating presence of second sequence (nuclear copies of the gene) in the amplified fragment of mtDNA. We carried out cloning of the amplified fragments of two Siberian rubythroat samples from Tomsk and Krasnoyarsk territory. It turned out that one of the clone variants from both samples was represented by the mitochondrial *cyt b* gene of nominate subspecies *calliope* itself, and the second – by a nuclear pseudogene *cyt b* haplotypes, specific for the eastern group *anadyrensis-camtschakensis* (98% similarity).

The discovered fact explains the origin of the eastern haplogroup by direct recombination of homologous regions of nuclear and mitochondrial genomes and by the phenomena of the founder effect. Since the probability of occurrence of identical events in several individuals is extremely low, we believe that the recombination event could occur at the early stage of the ovaries formation. Thereby the recombinant haplotype become inherited by all descendants of a particular female. Transfer of nuclear pseudogene in mtDNA, preservation and dissemination of recombinant haplotype probably occurred at the border area of the *calliope* range perhaps which is now the zone of intergradation with *anadyrensis.* Low numbers of birds allow for consolidating a new haplotype throughout the time and becoming taxon-specific.

Opposite assumption of the inclusion of the Eastern haplotype in the nuclear genome of *calliope* is unlikely in our opinion for several reasons. Firstly, there are great distances between locations where a nuclear pseudogene has been found (western Siberia) and the nesting habitat of *anadyrensis-camtschakensis* (Anadyr, Kamchtka); secondly, there is a well-known high phylopatry of Siberian rubythroat and its peculiar permanent migration routes. In addition, according to the observations by ornithologists (Nazarenko, 2004), the extension of the area of *L. calliope* takes place from west to east, and we did not find the eastern mitotypes themselves to the west of Blagoveshchensk (Amur region).

Therefore, we were the first to show the role of nuclear copies of mtDNA pseudogenes as a source of new taxon-specific haplotypes and their importance in microevolution, which is contrary to the opinion of NUMT as a hindrance. This fact being a natural phenomenon may explain the presence of several significantly different variants of haplotypes in many taxa.

GENETIC DIVERSITY IN *CLONORCHIS SINENSIS* (TREMATODA: OPISTHORCHIIDAE) FROM RUSSIA AND VIETNAM USING IRAP

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Clonorchis sinensis Cobbold, 1875 (Digenea: Opisthorchiidae) is an animal and human parasite from the class Trematoda, which inhabits South-East Asia. This fluke is the third in the world as to the number of infected people, and it has officially been listed as a biological carcinogen. For these reasons, *Clonorchis sinensis* is of great scientific and epidemiological significance. Earlier we explored the genetic diversity in the complete *cox1* gene sequence for five populations of *C. sinensis* in Russia and Vietnam (Chelomina et al., 2014). Korean researchers (Bae and Kong, 2003) used inter-retrotransposon amplified polymorphism (IRAP) method with *CsRn1* long-terminal-repeat (LTR) retrotransposon for *C. sinensis* from Korea. The IRAP patterns were not analyzed in this study. However, the researchers suggested that *CsRn1* can be useful for comparative genetic studies of Chinese liver fluke from various endemic areas. The aim of our study is the genetic diversity analysis of five isolated *C. sinensis* populations in Russia and Vietnam using IRAP PCR. Obtained data was used for estimation of genetic variability and reconstruction of intraspecific phylogenetic relationships. It is the first investigation of Russian and Vietnamese populations by IRAP method.

Parasitological material of *Clonorchis sinensis* was collected from two countries: from 3 localities in Russia and from 2 provinces in Vietnam (Chelomina et al., 2014). The IRAP PCR was carried out using primers for *CsRn1* LTR retrotransposon (Bae and Kong, 2003). Negative control was included. The PCR products were analyzed by electrophoresis in 1.5% agarose gel. The weight of DNA banding patterns were compared with O’GeneRuler^{TM} 100 bp Ladder Plus. The presence of a band was marked as 1, while the null (visually absent) allele was marked as 0.

A total of 18 bands were determined for 59 *C. sinensis* samples with molecular weight 120-980 bp. All loci were polymorphic in the total dataset. Five loci were unique for Vietnamese populations, while only one unique locus was obtained in all Russian populations. These distinctions can reflect local adaptation of the parasite. The percentage of polymorphic loci in Russia and Vietnam were 72 and 83, respectively. The expected heterozygosity was 0.179 for the total dataset. Vietnamese samples had more polymorphic loci with low frequency (60% vs. 54% in Russia). Values of the sequence divergence were higher in Vietnamese samples, but average values inside Russia and Vietnam localities were almost identical. MST had a difficult structure with no clear substructures. We detected 42 haplotypes in the total dataset, and almost all of them (81%) were unique. Ancestral haplotype comprised the three samples from the northernmost Russian population. Two major haplotypes with five and seven copies included samples from different localities in Russia and Vietnam. Therefore, intraspecific phylogeny based on IRAP PCR was not resolved. Haplotypes from Russia and Vietnam on *cox1* gene sequences were not well subdivided in contrast to MST. A single ancestral haplotype and absence of phylogeographic subdivision suggested a relatively recent population expansion and probably high level of the gene flow among geographical localities. At the same time, common and ancestral haplotypes differed from the complete *cox1* gene data.

Therefore, our study using IRAP PCR did not resolve statistically supported population subdivisions. However, this genetic marker provided new data for studies on intraspecific diversity in *C. sinensis* from different geographical regions.

EVOLUTIONARY HISTORY OF ENDEMIC TRICHOPTERA FROM LAKE BAIKAL

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Endemic caddis flies are represented in Lake Baikal by one family, seven genera and 14 species, which is about 26% of trichoptera species inhabiting the lake. These organisms are very numerous and by their abundance are one of the dominating groups in the lake.

We sequenced the gene fragment CO1 in 11 endemic species and one palaearctic species *Apatania majuscula*. Phylogenetic analysis was performed based on molecular data of Bayesian Markov Chain Monte Carlo approach. Phylogeny of the Baikalian Apataniidae is inferred as well as their evolutionary relations to the non-baikalian sister taxa. Application of a statistically supported hypothesis on molecular clock enabled us to estimate time of existence of the most recent common ancestor of the extant baikalian trichoptera. We hypothesize that it corresponds approximately to the time of the first reproductive isolation between Baikalian caddisfiles and their continental sisters. Interestingly, the Baikalian endemics are shown to be evolutionary close to the taxa currently inhabiting the North America. Possible explanations to this finding are discussed.

STRUCTURAL FEATURES AND VARIATION OF MITOCHONDRIAL CONTROL REGION AMONG EELPOUTS (COTTOIDEI: ZOARCALES)

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The control region (CR) is a major non-protein-coding domain of mitochondrial DNA, which contains transcription promoters for both strands of mitochondrial genome as well as binding sites for transcription-replication machinery whereby the *D*-loop is formed, a vital structure for the existence of mitochondrial genome as a replicator. The origin and diversity of structure-functional elements of CR have been intensively investigated with massive recent involvement of new diverse taxa. In this study, we report the preliminary results on the analysis of eelpouts (the fishes of infraorder Zoarcales) CR structure and variation as well as apparent trends of their evolution.

Both intraspecific (28 sequences from *Stichaeus grigorjewi* Herzenstein, 1890) and infraorder (4 families from Zoarcales) levels have been covered in the analysis. CR sequences in *S. grigorjewi* as well as in other eelpouts are restricted by tRNAs *Pro* and *Phe*, which is consistent with the classic scheme among other vertebrates. The mean intraspecies genetic distance is 0.26±0.25. The nucleotide diversity landscape is not uniform with complete absence of variation in the central part. CR length is 862 bp. No length variation was observed. Three classic domains have been found in the CR structure of *S. grigorjewi*. In total 4 non-tandem putative termination associated sequences (TAS) are present at the 5' end of the CR. Central conservative domain contains full set of sequences previously observed in other vertebrates (represented in classic order CSB-A, CSB-B, CSB-C, CSB-D, CSB-E and CSB-F, in 3'-5' direction). Conservative sequences domain includes 3 common sequences (CSB-I, CSB-II and CSB-III, in 5'-3' direction).

At the infraorder level, the parallel decrease of CR length is observed from the basal to terminal branches, based on following scheme: *Stichaeus grigorjewi* (862 bp) → *Pholis fungi* (853 bp) + *Pholis crassispina* (853 bp) and *Lycodes toyamensis* (863 bp) → *Anarhichas denticulatus* (838 bp) → *Anarhichas minor* (837 bp) → *Anarhichas lupus* (835 bp). Therefore, the apparent trend among eelpouts CR structural evolution, taking into account the taxonomic representation, is the change of length, which is caused by the loss of the first (5' end) TAS sequence.

IN SEARCH OF REPRODUCTIVE ISOLATION BETWEEN *CRICETULUS BARABENSIS* AND *C. GRISEUS*: HYBRIDIZATION AND CYTOGENETIC ANALYSIS

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The superspecies *Cricetulus barabensis* – striped hamsters include: *C. barabensis* (2n=20), *C. griseus* (2n=22), *C. pseudogriseus* (2n=24) and *C. sokolovi* (2n=20). The taxonomic status of these four species is debatable. They can be considered as species (Malygin et al., 1992; Gromov, Erbaeva, 1995; Kartavtseva, 2002; Korablev et al., 2013; Vakurin et al., 2014), which have allopatric distribution and differences in the karyotype structure (Radjabli, Kryukova, 1973; Orlov, Iskhakova, 1975; Kral et al., 1984; Orlov, Malygin, 1988; Romanenko et al., 2007). Same forms are considered as subspecies (karyoforms) of polymorphic *C. barabensis* by craniometric characteristics (Lebedev, Lisovskii, 2008) and molecular analysis of mtDNA cytochrome *b* gene and Y-chromosome DBY1 gene (Poplavskaya et al., 2011, 2012), *C. sokolovi* confirms as independent species.

The several variants of hybridization of *C. barabensis*, *C. griseus* and *C. pseudogriseus* were conducted early and F1 hybrids were produced (Poplavskaya et al., 2012).

We will continue to evaluate the reproductive barriers between the two karyoforms. The crossing of ♂*C. barabensis* (2n=20, from natural population) and ♀*C. griseus* (2n=22, from laboratory line) was conducted. The hybrid animals were produced at Laboratory of Evolutionary Zoology and Genetics by Korablev V.P. and Pavlenko M.V.

The F1 (n=3), F2 (n=7) and F3 (n=6) hybrids were obtained. All hybrids were viable. The expected splitting of the diploid chromosome number: the uniformity of 2n=21 for F1 hybrids, the splitting corresponds to 2n=20:21:22 as 1:2:1 for F2 hybrids. At the initial stage, the cytogenetic characteristics and the karyotype structure of hybrid animals from different generations were analyzed without counting splitting. The cytogenetic analysis was conducted for F1 (n=3), F2 (n=3) and F3 (n=2) hybrids. The disturbances of mitosis were not found. There are various combinations of sex chromosomes of different morphology for F2 and F3 hybrids.

F1 hybrids (1♂; 2♀♀) have 2n=21, NF=38 (4lM, 5mM, 6sM, 4mA, XX/XY)

F2 hybrids (2♀♀) have 2n=21, NF=38 (4lM, 5mM, 6sM, 4mA, XX); F2 hybrid (1♂) has 2n=22, NF=38 (4lM, 4mM, 6sM, 6mA, XY)

F3 hybrids (2♂♂) have 2n=20, NF=38 (4lM, 6mM, 6sM, 2mA, XY)

Notation: lM – large metacentric, mM – medium metacentric, sM – small metacentric, mA – medium acrocentric.

COPY NUMBER VARIATION OF HUMAN RIBOSOMAL GENES IN HEALTH AND DISEASE

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Human ribosomal genes (RG) are located on five pairs of acrocentric chromosomes as clusters of tandem repeats. Human diploid genome contains 300 to 700 copies of RG (average: 420 RG copies per diploid genome). The basic RG function in the cell nucleus is the synthesis of 18S, 5.8S and 28S rRNA required for the formation of ribosomes, the organelles for protein synthesis. Mean RG copy numbers in the genomic DNA are statistically higher in the groups of patients with Down syndrome and schizophrenia, whilst lower in case of Roberstsonian translocations and infantile autism. Formerly established that ribosomal DNA copy number is coupled with gene expression variation and mitochondrial abundance in humans (Gibbons et al., 2014). In human body the RG are also contained within cell-free circulating blood DNA, or extracellular DNA (ecDNA). The abundance of RG copies in the ecDNA of healthy individuals is 2-3 times as much as the RG copy number in the cell nuclear DNA of the same individuals. The abundance of RG copies within the ecDNA is elevated by a factor of 5 to 20 in comparison with its genomic content in some diseases, such as ischemic heart disease and rheumatoid arthritis, at pregnancy or after a chronic exposure to low doses of ionizing radiation. The RG nucleotide sequence contains many binding sites for TLR9 and Gn motifs, in which deoxyguanosine is easily oxidized to 8-oxo- deoxyguanosine. That is why the RGs within ecDNA have an ability to induce synthesis of proinflammatory cytokines and to provoke increased generation of reactive oxygen species (ROS) by the cells. Thus, the RG copy number in the cells can influence the rate of protein synthesis, and the RG copy number within the ecDNA can have an effect upon the ROS level and the inflammation intensity in the human body.

APPLICATION OF ECOLOGICAL GENETICS FOR SURVEILLANCE OF *LISTERIA MONOCYTOGENES*

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Listeriosis is the second after salmonella the most frequent cause of death from food-borne bacterial infections. The genetic diversity of *Listeria monocytogenes* isolates were early studied for the Far East Region (FER) of Russia. The aim of this research was evaluation of genotypes of the environmental sources isolates, basically from the European Region (ER) of Russia, comparison with FER data for greater understanding of phylogenetic and regional features as well as for potential dangers of bacteria to human health.

For analysis of isolates we used the multilocus sequence typing method (MLST); genotypes (sequence types, STs) were determined by use of *L. monocytogenes* MLST database. The characteristics of internal profile (IP) were designed for genes *inlA*, *inlB*, *inlC*, *inlE*. Whole genome sequencing (WGS) made by 454 Roche technology. Annotation was carried out by RAST service.

By MLST data isolates from ER were subdivided into 12 STs, three of which (ST756, 757, 758) were newly detected and presented the single locus variants (SLV) of formerly known. At least five STs belonged to the previously established dangerous for humans epidemic clones III, V, VI and VII. The six new IPs were revealed. Comparison of our and previously obtained data of MLST and IP, demonstrated the prevalence of the global spread STs. However, the phylogenetic lineage II genotypes predominated in the ER, while the higher pathogenic for human lineage I genotypes – in the FER.

Therefore, two closely related representatives of global spread STs, included in phylogenetic lineage I, were studied by WGS. Isolates VIMVR081 (*Myodes rufocanus*, Primorsky Krai, ST145) and VIMHA007 (stillborn, Khabarovsk Krai, ST2) had STs, which were SLV. These genomes were compared with geographic distant reference genome *L. monocytogenes* str. ATCC 19117 (accession FR733643, sheep, ST2). IPs of all three strains were equal.

As a result, three genomes demonstrated high similarity. The same insertion sequence elements (IS) of IS3 family and phage region (e.g. gp61, antigen B, lysin) were found in these genomes. By RAST data, the similarity of str. ATCC 19117 and VIMVR081 proteins was 96.2%. The main virulence-associated proteins (e.g. internalin A-E, listeria antigen Lma A-D, metalloprotease Mpl, regulatory factor PrfA, and actin assembly-inducing protein ActA) were identical in three genomes. The differences in amino acid sequence were found only in 3.8% proteins, which may be important for host specificity adaptation.

The results obtained emphasize the significance of ecological genetics data for identification of potentially dangerous *L. monocytogenes* strains and for the surveillance of infection in the natural foci.

RESTITUTION AND GENETIC DIFFERENTIATION OF *NANOPHYETUS* POPULATIONS

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Trematoda is a class within the phylum Platyhelminthes. The subclass Digenea shows the highest trematode diversity, and includes obligate parasites of both mollusks and vertebrates. Trematodes are commonly referred to as flukes and were noted to be associated with the strong infectious diseases named trematodiasis. Nanophyetiasis is a zoonotic disease caused by the digenetic trematodes *Nanophyetus salmincola* and *Nanophyetus schikhobalowi* from the family Nanophyetidae, which are localized in the small intestine. These trematodes are distributed mainly in various species of canids (dog, fox), other carnivores (bear, badger etc.) and humans.

Nanophyetus salmincola was considered as a synonym or sister species or a subspecies of *Nanophyetus schikhobalowi*, and therefore, *Nanophyetus schikhobalowi* is rarely mentioned in the scientific literature. However, high level of invasion intensity with *N. schikhobalowi* metacercaria was reported in salmon-like fishes (summer chum, pink salmon, and trout) from the mountain tributaries (Khor, Anyui, and Manoma) of the Amur River. The subspecies of *N. schikhobalowi* was also reported from wild carnivores in the coastal US Pacific Northwest and Canada.

Mitochondrial genes provide excellent molecular markers for defining population groups, for tracing the genetic history of an individual or a particular group of related individuals, and for constructing deep-branch of taxonomic phylogenies (Boore et al., 1995; Rollinson et al., 1997; Boore, 1999). Analyzed are 20 sequences of 639 bp fragment of the *n*ad1 gene of *Nanophyetus salmincola* that were obtained by PCR amplification and direct sequencing. The phylogenetic analysis was carried out through Bayesian inference (BI) as implemented in the MrBayes program using own data and data available in GenBank: 91 sequences of 639 bp fragment of the *n*ad1 gene of *Nanophyetus salmincola* (Criscione, Blouin, 2005). The results point out to a deep phylogenetic break between *N. salmincola* and *N. schikhobalowi*.

The coding regions of the ribosomal DNA (18S and 28S) and Internal transcribed spacers (ITS) are widely used in molecular genetic studies. We combined our mitochondrial data set by complete gene sequences of the small subunit of 18s rRNA gene (rDNA). On the basis of this gene wide phylogenetic utility at many levels have revealed; regions of relatively high sequence variability are acompined by regions of high sequence conservation, allowing easy alignment and the establishment of base-position homology between taxa (e.g. Hasegawa et al., 1985; Hillis et al., 1996: 337).

For the first time the complete nucleotide sequence of *Nanophyetus* ribosomal RNA gene cluster ~ 6,980 bp was determined, molecular analysis was conducted, secondary structures of the variable domains of the gene 18S rDNA were modeled both for the representatives of the Far Eastern and American flukes populations. Therefore, populations of *Nanophyetus* from the Russian Far East and North America may represent distinct species.

CHROMOSOMAL AND MITOCHONDRIAL DNA VARIABILITY IN THE STRIPED FIELD MOUSE (*Apodemus agrarius* Pall., 1771, MURIDAE, RODENTIA) IN THE URALS

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Geographical location of the Ural region determines the diversity of its climatic conditions and the uniqueness of its fauna, comprising both the European and Asian elements as well as widespread species of mammals. However, this area remains a "blind spot" in genetic studies of many species, including *A. agrarius*.

The results of studying the chromosomal and mitochondrial DNA variability (complete sequence of the cytochrome b gene (1140 bp)) in *A. agrarius* in the Ural region are presented. Karyotypes of 180 individuals from 12 localities in the Southern, Middle and Northern Urals up to the northern limit of the species distribution in the region (60º24'N, 59º59'E) are studied using routine Romanowsky staining of metaphase chromosomes of the bone marrow cells. No departures from the standard karyotype (2n=48, NFa=54, M(the number of pairs of small metacentric chromosomes)=8) are revealed. An exception is the case of the mosaicism detected in one male in the locality exposed to significant chronic radiation.

An analysis of 31 sequences of cytochrome b gene in *A. agrarius* from 11 localities in the Urals reveals 17 haplotypes, 15 of which are original and have not been described previously. Published data show that there exist several well-differentiated groups in the eastern part of the disjunctive range of *A. agrarius* (Pereverzeva, Pavlenko, 2014; Sakka et al., 2010), while similar studies are not known in the western part of the species range. Our results suggest that the Ural haplotypes (original data) fall into one haplogroup with the samples from the Russian Far East, north-western China and with some single specimens from the European part of the species' range (data from the GenBank). Obtained results could be the basis for further research aimed to clarify the history of the western part of the species range.

Data on chromosomal and mitochondrial variation in *A. agrarius* in the Urals obtained for the first time expand significantly the geography of investigations of genetic variability in *A. agrarius*.

This study was supported by RFBR project №14-04-00614.

PHYLOGENETIC INTERCOMMUNICATIONS OF TREMATODES OF THE GENUS *LEUCOCHLORIDIUM*

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Today molecular genotyping plays an important role in resolving taxonomic status of organisms and evaluating intraspecific variability. Molecular genetic analysis of rDNA of sporocyst of *Leucochloridium* sp. trematodes proved the objectiveness of morphological criteria for identification of *L. paradoxum* and *L. perturbatum* (Zhukova et al., 2014). A nucleotide sequence of rDNA including 4444 bps has been obtained (Zhukova et al., 2014) and used for the analysis of phylogenetic reletionships in the genus *Leucochloridium.*

Chromosome DNA has been obtained from sporocysts of *L. paradoxum, L. perturbatum, L. vogtianum* gathered in the Leningradskaya oblast (Russia). The research on intraspecific polymorphism of rDNA was performed in regions of ITS1 and ITS2 sequences. Segments encoding 18S, 5.8S and 28S of of rDNA were also used as markers of genetic polymorphysm. Amplicons of all these regions were obtained by PCR with specific primers, designed on the basis of *Leucochloridium* sp. sequences annotated in GeneBank, and self-obtained sequences of these trematodes (Zhukova et al., 2012). The analysis of ITS1 and ITS2 rDNA of *L. paradoxum* и *L. perturbatum* sporocyst with identical coloring showed 100% homology, while interspecies differences of these segments equaled 2.6% and 6.7%, respectively. The homology of *L. vogtianum* with *L. paradoxum* equaled 94.7%, and with *L. perturbatum* 95.4%.

Dendrogram of phylogenetic relationships of *Leucochloridium* species and other trematodes has been constructed on the basis of ITS1 and ITS2 sequences.

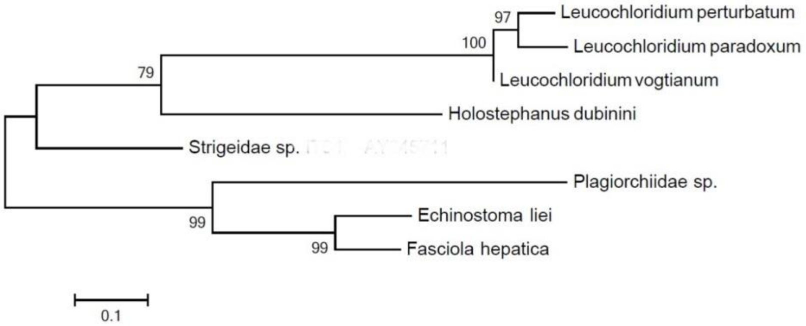

Figure 1. Phylogenetic relationships of *Leucochloridium* species and other trematodes (on the base of ITS1 and ITS2 sequences). Numbers indicate bootstrap support values on the *Leucochloridiidae* family tree.

The nucleotide sequences of rDNA, including ITS1, ITS2, 5,8S and a segment of 28S rDNA, have been registered at Gene Bank (JN639011.1; JN639012.1).

MOLECULAR GENETIC CHARACTERISTICS OF *PLANTAGO MEDIA* L. SUBPOPULATIONS FROM THE ENVIRONS OF KACHKANARSKY ORE MINING AND PROCESSING ENTERPRISE

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Anthropogenic impact on modern ecosystems is high, however, the genetic consequences of this influence is still not clear. In this study, the population structure of *Plantago media* L. was studied in a gradient of heavy metal pollution. Plants were picked up from two areas near Kachkanarsky Ore Mining and Processing Enterprise (Sverdlovsk region, Russia) with different values of total pollution index (S_i (1) = 2.65; S_i (2) = 4.62). Background area (S_i (3) = 1.5) was in 13.5 kilometers from contaminated habitats.

Genetic diversity of *P. media* subpopulations was evaluated by inter-simple sequence repeat loci method. Seven UBC primers showed 82 bands from 200 to 2,200 bp. Three loci were unique. The average number of loci per primer was 11.7, while totally they varied from 9 (UBC808) to 15 (UBC834). The number of bands per plant positively correlated with the level of anthropogenic impact. *P. media* plant clones were not found.

As a result of the UPGMA algorithm, clusters were identified, which are mainly made up of plants of one habitat. This differentiation was confirmed by the result of principal component analysis using Jacquard similarity index. Plants from the second habitat were distributed among others. Perhaps, the deficiency of genotypes from area 2 didn't allow them to unite in a single group.

Thus, the results obtained allow suppose that subpopulations of *Plantago media* L. are considerably less genetically diverse in contaminated habitats then in background territories.

РФФИ

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Современные достижения в популяционной,
эволюционной и экологической генетике
Международный симпозиум
Владивосток: Морская биологическая станция “Восток”
1-10 сентября 2015
Программа и тезисы докладов

Ответственный за выпуск Ю.Ф. Картавцев

Объем 10,1 усл.-печ. Л., 5,7 уч.-изд.л. Тираж 150 экз. Формат 60x84/8

Институт биологии моря им. А.В. Жирмунского ДВО РАН
690059 Владивосток, ул. Пальчевского, 17

Отпечатано: Институт биологии моря им. А.В. Жирмунского