#### University of Plovdiv "Paisii Hilendarski" Faculty of Biology, Department of Zoology

#### INTERNATIONAL CONFERENCE ON ZOOLOGY AND ZOONOSES

#### PROGRAMME & ABSTRACTS

October 26 – 28, 2016 Hissar, Bulgaria

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#### **FOREWORD**

Dear colleagues and friends,

On behalf of the Organizing Committee it is our great pleasure to welcome you to the International Conference on Zoology and Zoonoses.

We are happy that the Conference, with its location at wonderful ancient town of Hissar, has attracted such an interest.

The International Conference on Zoology and Zoonoses is aimed at academics, researchers and students from various fields of zoology, as well as microbiologists, vets and epidemiologists.

We have a wide variety of presentations comprising the working programme from the areas of taxonomy, faunistics, phylogeny, ecology, applied zoology, vector-borne diseases, epidemiology, population genetics, molecular biology, biomonitoring, conservation zoology.

We hope the Conference will be a great forum for a dynamic exchange of information, ideas and recent scientific discoveries in light of recent climate changes and the associated epidemiological risk for public health, and yield positive contacts and future collaboration for all participants.

We wish you fruitful work, success and a pleasant stay in beautiful Hissar town and in Bulgaria!

#### Welcome!

**Organizing Committee** 

Department of Zoology, University of Plovdiv National Center of Infectious and Parasites Diseases Bulgarian Food Safety Agency Ministry of Health-Care, Bulgaria Southeastern European Centre for Disease Control

#### **COMMITTEES**

#### **Organizing Committee**

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Assoc. prof. Dr. Anelia Stojanova, University of Plovdiv

M.Sc., Viktor Ivanov - Web design and Book design

#### **SCIENTIFIC PROGRAMME**

#### International Conference on Zoology and Zoonoses October 26 – 28, 2016, Hissar, Bulgaria

#### Wednesday, 26 October 2016

14.00 – 18.00 Arrival, registration, posters set up. SPA Hotel "Hissar".18.30 Welcome cocktail at the Lobby bar, SPA Hotel "Hissar".

#### Thursday, 27 October 2016

- **8.00 9.00** Registration
- **9.00 9.45 Opening**. Maritsa hall in SPA Hotel "Hissar"
- 9.45 10.15 Plenary lecture: When the Balkans resembled Africa. Professor DSc Dimiter Ivanov, BAS, Sofia
- **10.15 10.45 Plenary lecture:** Southeast European Center for Infectious Diseases Control a success story of countries collaboration. **Teita Myrseli** representative of SECID, Tirana
- 10.45 11.00 Coffee-break

#### POPULATION GENETICS AND MOLECULAR BIOLOGY

(Trakiya hall)

**Session chair:** Professor DSc Paraskeva Michailova **Session secretary:** Assist. prof. Dr. Peter Boyadzhiev

- 11.00 11.15 Possibilities for discrimination of Bulgarian honey bees based on different genetic markers. E. Ivanova
- 11.15 11.30 Application of DNA barcoding in *Eurygaster integriceps* specimens to determine intraspecific genetic variation. **E. Koçak**

#### BIODIVERSITY, ECOLOGY AND CONSERVATION ZOOLOGY

(Trakiya hall)

**Session chair:** Professor DSc Georgi Markov **Session secretary:** Assist. prof. Dr. Vesela Mitkovska

11.30 – 11.45 Ex situ effects of priority substances on the lysosomal membrane stability and respiration rate in zebra mussel (Dreissena polymorpha) 1. Heavy metals. V. Yancheva

- 11.45 12.00 Ex situ effects of priority substances on the lysosomal membrane stability and respiration rate in zebra mussel (Dreissena polymorpha) 2. Pesticides. V. Yancheva
- 12.00 12.15 New data on fungus gnats (Diptera: Bolitophilidae, Keroplatidae and Mycetophilidae) in Bulgaria, Greece and Turkey. A. Pavlova
- 12.15 12.30 First record of the relict *Pentacyphona* stat. nov. in the Palearctic area (Diptera, Pediciidae) with discussion on relationship with their kin. L. Kolcsár
- 12.30 12.45 Bacteria associated with *Rhagoletis cerasi* (Diptera: Tephritidae). A. Tuncbilek

#### **ONE HEALTH** (Maritsa hall)

**Session chair:** Professor DSc Iva Christova **Session secretary:** Assoc. prof. Dr. Todorka Yankovska-Stefanova

- 11.00 11.30 Public Health challenges. MD Angel Kunchev, Chief State Health Inspector, Ministry of Health
- 11.30 13.00 Communicable diseases prevention and control in Europa: Avian Influenza. Dr. Cornelia Adlhoch, Respiratory Diseases expert, ECDC, Stockholm, Sweden - presentation and discussion
- 13.00 14.00 Lunch, Restaurant SPA Hotel "Hissar"

#### BIODIVERSITY, ECOLOGY AND CONSERVATION ZOOLOGY (Trakiya hall)

Session chair: Professor Dr. Roumiana Metcheva **Session secretary:** Assist. prof. Miroslav Antov

- 14.00 14.15 Ichneumon wasps (Hymenoptera: Ichneumonidae) reared from tortrix moths (Lepidoptera: Tortricidae) in the oak forests in Sofia region, Bulgaria. G. Zaemdzhikova
- 14.15 14.30 Red List of Bulgarian bombyces and sphinges (Insecta: Lepidoptera) and their distribution in Bulgaria. H. Hristova
- 14.30 14.45 An assessment of a hypersaline lake: case study of Atanasovsko Lake, Bulgaria. G. Gecheva
- 14.45 15.00 Fish based method for water monitoring and ecological classification of natural lakes on the Bulgarian Danube floodplain. L. Pehlivanov

- **15.00 15.15** Health status of *Pelophylax ridibundus* (Amphibia: Ranidae) in a rice paddy ecosystem in southern Bulgaria: body condition factor and fluctuating asymmetry Part 1. **A. Arnaudov**
- 15.15 15.30 Tracking methods for lesser kestrel (*Falco naumanni*) used in the course of the species' recovery as breeder in Bulgaria. **G. Gradev**
- **15.30 15.45** Ichneumonidae (Hymenoptera) fauna of Davraz Mountain in Isparta, Turkey. **O. Birol**

#### **ONE HEALTH** (Maritsa hall)

Session chairs: MD Angel Kunchev, Prof. Dr. Todor Kantardjiev Session secretary: Dr. Petya Petkova

- **14.00 14.15** Viral and bacterial zoonoses in humans (zooanthroponoses) with similar clinical manifestation. **I. Christova**
- 14.15 14.30 Anthrax the old-new challenge. A. Zdravkova
- 14.30 14.45 Monitoring and control of influenza in birds in Bulgaria.

  A. Miteva
- 14.45 15.00 New cases of rabies in foxes in Bulgaria. P. Kamenov
- 15.00 15.15 Dynamics of echinococcosis in Bulgaria for the period 2011 2015 year. I. Rainova,
- 15.15 15.30 Brucellosis in sheep and goats (*Brucella melitensis*).

  P. Dangov
- 15.30 15.45 Realization of the programme for prophylactics, supervision and control of the West Nile fever in Bulgaria.

  S. Chakarova
- 15.45 16.00 Coffee-break

#### **ONE HEALTH** (Maritsa hall)

**Session chair:** Assoc. prof. Dr. Iskra Raynova **Session secretary:** Assoc. prof. Dr. Atanas Arnaudov

- 16.00 16.15 Tuberculosis in large ruminants. Y. Yordanov
- **16.15 16.30** Ixodidae ticks in domestic ruminants in the Valley of Maritsa River in the Plovdiv region. **A. Arnaudov**
- **16.30 16.45** Supervision and control of salmonellosis in birds in Bulgaria. **S. Kolev**
- 16.45 17.00 Lumpy skin disease new challenge for Europe. A. Miteva

- **17.00 17.15** Food explosion of salmonellosis in Varna practical aspects. **K. Nikolov**
- 16.00 18.00 Poster session, Trakiya hall in SPA Hotel "Hissar"
- **19.30 Conference dinner**, Restaurant "Prestige" in SPA Hotel "Hissar"

#### Friday, 28 October 2016

(Maritsa hall)

- 9.00 9.15 Plenary lecture: Summarised review of the results from the 1959 complex expedition for investigating the natural outbreak of diseases in Bulgaria. Professor Dr. S. Gerasimov
- **9.15 10.00 Presentations of sponsors** presentations of vaccines, medicines and disinfectants used in practice.
- 10.30 11.00 Closing the conference, Maritsa hall
- 11.00 The tour to the Thracian temple complex Starosel
- 14.00 Departure

#### POSTER SESSION

- **Poster panel:** Scientific committee of International Conference on Zoology and Zoonoses
- P1 The response of Chironomidae (Diptera) genome to heavy metal pollution in two rivers of Southern Poland. **Julia Ilkova**, Paraskeva Michailova, Ewa Szarek-Gwiazda, Andrzej Kownacki, Dariusz Ciszewski
- P2 Genetic comparison between local *Apis mellifera macedonica*, selectively reared for production of bee queens and swarms in Bulgaria and honey bee colonies with indicative hygienic behavior. **Vida Georgieva**, Plamen Petrov, Teodora Staykova, Svilen Lazarov, Ivan Stoyanov, Evgeniya Ivanova
- P3 Genetic variability in populations of *Messor barbarus* (Hymenoptera, Formicidae) from Bulgaria based on isoenzyme analysis. **Ivan Stoyanov**, Teodora Staykova, Penka Vasileva, Evgeniya Iyanova
- P4 Genetic variability of *Eupelmus* species (Hymenoptera: Eupelmidae) based on allozyme markers. **Miroslav Antov**, Ivan Stoyanov, Anelia Stojanova, Teodora Staykova
- P5 Potential of PCR based molecular diagnostic methods for rapid detection of bacterial pathogens causing economically important diseases in fishes. Maria Gevezova-Kazakova, Feriha Tserkova, Ina Kirilova, Tania Hubenova, Angel Zaikov, Angelina Ivanova, Maria Yankova, Iliya Denev
- P6 Genetic diversity and geographic distribution of round goby *Neogobius melanostomus* (Pallas) as revealed by mtDNA cyt b gene haplotypes. Feriha Tserkova, **Maria Gevezova-Kazakova**, Galin Gospodinov, Ina Kirilova, D. Klisarova, Tania Hubenova, Iliya Denev
- P7 The difference in the some quantitatives parameters of the observed bee breeding lines from Montenegro and Serbia. Slađan Rašić, Mića Mladenović, Ljubiša Stanisavljević, Radica Đedović, Leonidas Charistos, Aleksa Božičković

- P8 Evidence of geno-toxicity induced by 60 Hz magnetic fields on mice bone marrow as assessed by *in vivo* micronucleus test. J. Antonio Heredia-Rojas, Abraham O. Rodríguez-de la Fuente, Omar Heredia-Rodríguez, **Michaela Beltcheva**, Roumiana Metcheva, Laura E. Rodríguez-Flores, Martha A. Santoyo-Stephano, Esperanza Castañeda-Garza
- P9 Allozyme genetic characterization of *Apis mellifera* colonies from Bulgaria with different hygienic behavior. **Svilen Lazarov**, Ivan Stoyanov, Vida Georgieva, Ivanka Zhelyazkova, Evgeniya Ivanova
- P10 Dynamic and ecological impact of marine zooplankton aliens in Varna Bay (Bulgarian Black Sea). **Kremena Stefanova**, Elitsa Stefanova, Valentina Doncheva
- P11 Summary of significant fungal infections in mollusca. **Stojmir Stojanovski**, Mišo Hristovski, Stoe Smiljkov, Dijana Blažeković-Dimovska, Georgi Atanasov, Lidija Velkova-Jordanovska
- P12 Summer rotifer assemblage in three Macedonian reservoirs (Konče 1, Konče 3 and Špilje). **Orhideja Tasevska**, Maria Špoljar, Dafina Gušeska, Goce Kostoski
- P13 Contributions to the study of earthworms (Oligochaeta, Lumbricidae) of the Pannonian region of Serbia (Vojvodina Province). Mirjana Stojanović, Jovana Sekulić, **Ralitsa Tsekova**, Tanja Trakić
- P14 Distribution and biogeographical significance of the endemic earthworm *Allolobophora robusta spasenijakaramani* (Blakemore, 2004) (Oligochaeta: Lumbricidae) on the Balkan Peninsula: first finding place in Macedonia. Mirjana Stojanović, Jovana Sekulić, **Ralitsa Tsekova**, Tanja Trakić
- P15 Mating behavior of millipede *Pachyiulus hungaricus* (Karsch, 1881) (Myriapoda, Diplopoda, Julidae) in laboratory conditions. **Sofija Pavković-Lučić**, Zvezdana Jovanović, Bojan Ilić, Vukica Vujić, Boris Dudić, Slobodan Makarov, Luka Lučić, Vladimir Tomić
- P16 Cuticular chemoprofile in the fruit fly, *Drosophila subobscura* Collin, 1936 (Diptera: Drosophilidae). **Sofija Pavković-Lučić**, Marina Todosijević, Tatjana Savić, Jelena Trajković, Ljubodrag Vujisić

- P17 Morphological analysis of *Branchipus* sp. from area of Stara Planina Mountains and Pannonian lowlands in Serbia. **Dragana Miličić**, Jelena Trajković, Sofija Pavković-Lučić, Tatjana Savić, Ljiljana Tomović
- P18 First finding of *Lepidurus couesii* Packard, 1875 (Crustacea, Triopsidae) in Serbia a record based on development of diagnostic body characters in males and females. Ivana Šaganović, Vladimir Tomić, Luka Lučić, **Dragana Miličić**
- P19 Study of contents of lead, cadmium and nickel in the water and in metabolically active organs of crayfish *Astacus leptodactylus* in Kardzhali dam. **Desislava Arnaudova**, Aneliya Pavlova, Atanas Arnaudov
- P20 First record of cysticercoids of *Microsomacanthus* (*Microsomacanthus*) abortiva (von Linstow, 1904) Lopez-Neyra, 1942 (Cyclophyllidea: Hymenolepididae) in *Gammarus* sp. (Amphipoda, Gammaridae) in Bulgaria. **Margarita Marinova**, Zlatka Dimitrova, Gergana Vasileva, Boyko Georgiev
- P21 Morphometric variability, allometric growth and sexual dimorphism in narrow-clawed crayfish (*Astacus leptodactylus*, Esch.) during the ontogenesis. **Penka Vasileva**, Tania Hubenova, Angel Zaikov, Ivan Stoyanov
- P22 A survey of natural enemies and associated arthropod pests in pomegranate orchards in Antalya province (South-western part of Turkey). Ismail Karaca, Özgür Şahin, **Fedai Erler**
- P23 Species of *Ooencyrtus* genus (Hymenoptera: Encyrtidae), egg parasitoids of *Thaumetopoea solitaria* (Lepidoptera: Notodontidae) in Bulgaria. **Peter Boyadzhiev**, Plamen Mirchev, Georgi Georgiev
- P24 A new species of the genus *Omphale* Haliday (Hymenoptera, Eulophidae, Entedoninae) from Bulgaria. Zoya Yefremova, Ekaterina Yegorenkova, Anelia Stojanova, **Peter Boyadzhiev**
- P25 Description of *Stepanovia fructirosae* sp. n. (Hymenoptera: Chalcidoidea: Eulophidae) from Turkey. Peter Boyadzhiev, Zoya Yefremova, Göksel Tozlu, Yusuf Mergen, **Özlem Mete**
- P26 The community members associated with rose gall wasp *Diplolepis fructuum* (Rübsaamen, 1895) (Hymenoptera: Cynipidae) in Tokat Province of Turkey. Özlem Mete, Yusuf Mergen

- P27 Detection and identification of *Wolbachia* endosymbiont from *Sitophilus granaries*. **Aydin Suzu Tuncbilek**, Sevgi Bakir, Fahriye Sümer Ercan, Ilhan Derin, Hüsniye Bilbil
- P28 Effect of non-ionizing radiation (UV) on prepupa and pupa stages of egg parasitoid *Trichogramma brassicae* Bezdenko (Hymenoptera: Trichogrammatidae). Fahriye Ercan, Sevcan Oztemiz, Nuri Ercan, **Aydin Suzu Tuncbilek**
- P29 Isolation and identification of some gram positive bacteria causing infections in silkworm (*Bombyx mori* L.). Kalinka Gurgulova, Petya Orozova, Mihail Panayotov, **Ivanka Zhelyazkova**, Sonya Takova, Radostina Gancheva
- P30 Aphid parasitoids (Hymenoptera, Aphidiinae) of the agro-ecosystems of the experimental fields in Plovdiv region. **Ognian Todorov**
- P31 The horseflies (Diptera: Tabanidae) of Rila Mountains, Bulgaria.

  Diana Ganeya
- P32 Developmental time and mating success in *Drosophila* melanogaster strains maintained on apple and carrot substrates.

  Jelena Trajković, Vukica Vujić, Dragana Miličić, Sofija Pavković-Lučić, Tatjana Savić
- P33 Does diet affect mating preference and success in *Drosophila* melanogaster via changes in wing size and shape? Jelena Trajković, Sofija Pavković-Lučić, Tatjana Savić
- P34 Different effects of a strong static magnetic field (2.4 T) on *Drosophila melanogaster* wing size and shape. **Tatjana Savić**, Dajana Todorović, Jasna Ristić-Djurović, Sofija Pavković-Lučić, Branka Petković, Jelena Trajković
- P35 Habitat and sex-specific responses of *Drosophila subobscura* exposed to static magnetic field (2.4 T) revealed by analysis of morphological traits. **Tatjana Savić**, Dajana Todorović, Saša Ćirković, Sofija Pavković-Lučić, Branka Petković, Jelena Trajković
- P36 Mosquito (Culicidae, Diptera) and vector monitoring in Danube
   Delta most representative ecosystems 2014-2015 (România).
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- P37 Morpho-physiological profile and changes in blood parameters of *Carassius gibelio* (Pisces: Cyprinidae) as response to the anthropogenic pollution in two water basins in the region of Galabovo, Southern Bulgaria. **Zhivko M. Zhelev**, Katerina N. Georgieva, Stefan V. Tsonev, Peter S. Boyadziev, Mladen V. Angeliov
- P38 Effects of Ni and Pb on the respiration rate and histological structure of common carp (*Cyprinus carpio*, L.) gills. **Stela Stoyanova**, Iliana Velcheva, Vesela Yancheva, Ivelin Mollov, Elenka Georgieva
- P39 Fulton condition coefficient of 11 fish species from great natural and two artificial lakes in the Republic of Macedonia. **Trajče Talevski**, Dragana Milošević
- P40 Histopathology of kidney of *Salmothimus ohridanus* (Pisces, Salmonidae) caged in Lake Ohrid. **Lidija Velkova-Jordanoska**, Stojmir Stojanovski, Goce Kostoski
- P41 Macrophyte vegetation as spawning ground of some cyprinid fish from Lake Ohrid. **Marina Talevska**, Trajce Talevski
- P42 Seasonal changes in the red blood cell parameters in Ohrid roach blood from Ohrid Lake. **Blagoja Trajčevski**, Trajče Talevski, Dragana Milošević
- P43 Correlation between the choice of partner and the individual nesting territory in the lesser kestrel (*Falco naumanni*) and preconditions for polyandry. **Polya Mihtieva**, Petya Karpuzova, Simeon Marin, Pavlin Zhelev, Gradimir Gradev, Dimitar Marinov
- P44 Lesser kestrel (*Falco naumanni*) in Thrace distribution, numbers, and threats. **Gradimir Gradev**, Dimitar Demerdchiev, Simeon Marin, Ivaylo Angelov, Pavlin Zhelev, Elena Kmetova-Biro, Efterpi Patetsini, Dobromir Dobrev, Stoycho Stoychev
- P45 First telemetric study of the individual territories, seasonal movements, and habitat use of the Balkanic capercaillie (*Tetrao urogallus rudolfi*). **Dimitar Plachiyski**, Georgi Popgeorgiev, Stefan Avramov, Katerina Angelova
- P46 Characteristics of the territorial and hunting behavior of the redfooted falcon (*Falco vespertinus*) from South-Eastern Bulgaria. **Girgina Daskalova**

- P47 Organochloride pesticides in feathers of *Pygoscelidae penguins* from Livingston island, Western Antarctica. **Roumiana Metcheva**, Michaela Beltcheva, Ginka Kalinova, Margarita Marinova, J. Antonio Heredia-Rojas, Vesela Peneva
- P48 Bat fauna (Mammalia: Chiroptera) from Sakar-Mountain (South-Eastern Bulgaria). Ivan Pandurski, **Roumiana Metcheva**, Michaela Beltcheva, Yordan Yankov
- P49 Population epigenetic diversity versus subspecies detachment of the forest dormouse (*Dryomys nitedula*) in a long distance transect in Eurasia: implication for its conservation. **Georgi Markov**, Ercument Çolak, Nuri Yigit, Maria Kocheva, Milena Gospodinova, Hristo Dimitrov
- P50 Indication for genetic diversity of European roe deer (*Capreolus capreolus*) in Southeastern Europe revealed by mtDNA markers. **Georgi Markov**, Elena Zvychaynaya, Alexei Danilkin, Marina Kholodova, Laszlo Sugar, Hristo Dimitrov
- P51 Influence of tourists on the summer bat colonies in the Devetashka cave. **Svetlana Ivanova**, Boyan Petrov, Daniela Simeonovska-Nikolova
- P52 Copulatory behavior of *Lasiopodomys (Stenocranius) gregalis* (Rodentia, Mammalia). Tanya Zorenko, **Nasko Atanasov**
- P53 Karyotypic and craniometric characteristic of harvest mouse (*Micromys minutus* Pallas, 1771) (Mammalia: Rodentia) from South Bulgaria. **Tsenka Chassovnikarova**, Nasko Atanasov, Hristo Dimitrov, Vesela Mitkovska
- P54 Autumn-winter diet and food niche overlap between red fox (*Vulpes vulpes* L. 1758) and golden jackal (*Canis aureus* L. 1758) in Bulgaria. Albena Vlasseva, **Tsenka Chassovnikarova**, Nasko Atanassov
- P55 In vivo genotoxicity and cytotoxicity assessment of permissible concentrations of Ni and Pb based on comet assay and nuclear abnormalities in acridine orange stained erythrocytes of common carp (Cyprinus carpio L.). Vesela Mitkovska, Hristo Dimitrov, Tsenka Chassovnikarova

- P56 Determination of specific IgG avidity and anti-Toxocara IgA antibodies in patients with toxocarosis and their application to specify the stage of disease. **Eleonora Kaneva**, Desislava Velcheva, Iskra Rainova
- P57 Phytoplankton and chlorophyll a concentration in Lake Prespaduring 2014. **Suzana Patceva**
- P58 The water quality of the tributaries of the Lake Ohrid. **Silvana Vasilevska**, Elizabeta Veljanoska-Sarafiloska, Lence Lokoska
- P59 The water quality of the river Sateska from microbiological and chemical aspects. **Lence Lokoska**, Elizabeta Veljanoska-Sarafiloska, Silvana Vasilevska
- P60 Compartion between organochlorine pesticide residues, organic matter and lipophilic bacteria in sediment from Lake Ohrid and Lake Dojran. Elizabeta Veljanoska-Sarafiloska, Lence Lokoska
- P61 Antibiotic resistance the plague of today. **Teodora Stoyanova**

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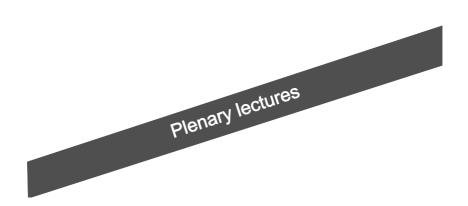
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#### When the Balkans resembled Africa

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#### ABSTRACT

The Miocene sediment deposits in Bulgaria are rich in various flora and fauna specimens, which show key aspects of the evolution of plants and animals on the territory of the Balkan Peninsula. Samples of great importance amongst the vast diversity of fossils are undoubtedly these of late Miocene vertebrate animals, as well as paleofloristic findings from the era. The fossilisation processes typically happened in rivers and on the banks of lakes and in most cases the conditions were beneficial for preservation of a large variety of fossils. The largest animal fossil deposits in west Bulgaria are in the regions of Hadjidimovo, Kalimatsi, Kromidovo, Gorna Shushitsa and Hrabarsko. Well preserved floral fossil deposits are also found in the same areas – Gotse Delchev region, Melnik region, Sofia region and others. Taphonomic and paleontologic analysis show predominance of species that inhabited open areas. They illustrate a relatively arid environment which resembles a wooded savannah. The paloefloristic data supports these findings, showing an increased distribution of grass species during the same time period. The summarization of information about fauna and flora in the environment during the paleo era significantly improves the quality and reliability of reconstruction of ecological systems during that time.

### **Southeast European Center for Infectious Diseases Control – a success story of countries collaboration**

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#### ABSTRACT

The SEEHN Network was founded in Sofia, Bulgaria, in April 2001 by the signatories of the Dubrovnik Pledge: Albania, Bosnia and Herzegovina, Bulgaria, Croatia, Romania, Serbia and Montenegro, and the former Yugoslav Republic of Macedonia.

This was the first ever political document on cross border, health development in the SEE region. The process was supported by the Council of Europe and WHO Regional Office for Europe in the framework of the Stability Pact.

The main purpose of the SEEHN is to provide leadership and to sustain project ownership by the countries in the region. WHO/Europe lends technical support to SEEHN's various health projects, after having supplied its secretariat, along with the Council of Europe, from 2001 to 2009. SEEHN has received political, technical and financial support from 10 partner countries and 5 international organizations.

For over a decade, SEEHN has been the undisputed vehicle of health development in the areas of mental health, communicable diseases, food safety and nutrition, blood safety, tobacco control, information systems, maternal and neonatal health, public health services and health systems.

Based on previous agreement in 2008 for the development of regional development centers, the RHDC on CDS and IHR implementation was established at Institute of Public Health, in Tirana, Albania in November 2010. The center is fostering safety and equity, reducing morbidity and mortality from infectious diseases and increasing joint

capacities in Southeast Europe to implement IHR and early warning systems through regional collaboration and maintenance of a network of communities of practice and by ensuring the regional leadership, ownership and partnership in one health initiative.

"Southeast European Center for Surveillance and Control of Infectious Diseases" – SECID, established on 04.02.2013, is an integral part of the Regional Development Center of Surveillance and Control of Infectious Diseases, RDC – CDC as well as the Administrative Partner of Institute of Public Health, with international vocation and will develop its activity in the territory of the Republic of Albania and can open its branches inside or outside the country.

SECID in cooperation with the University Hospital of Infectious Diseases "Dr. Fran Mihaljevic", in Zagreb, Croatia and with the kind support of WHO Euro and FAO, has been organizing a first Expert Group Meeting "The Assessment and Mitigation of Zoonosis in Southeast Europe – One Health Approach" meeting on June 2013, in Zagreb, Croatia.

The purpose of the meeting was to bring together all the SEE countries national experts in recognition of the importance of zoonotic diseases, having in focus One Health as one its important objectives and the need for extensive communication and cooperation between human and animal health workers.

Connecting Organizations for Regional Disease Surveillance (CORDS) started to explore the feasibility of developing an operational research project to increase Leishmaniasis surveillance in the Middle East and South Eastern Europe. MECIDS & SECID – decide to strengthen Leishmaniasis control in region establish jointly One Health Virtual Group for Leishmaniasis. The objective was to establish a platform for leishmaniasis expert from different sectors and countries, establish an innovative way for knowledge sharing for capacity building and problem solving, improve awareness and funding on Leishmaniasis and further project development.

A web-based platform, Leishmaniax.net has been developed to improve awareness, surveillance and control of cutaneous and visceral Leishmaniasis. Within the platform, a member's forum has been established where national specialists from a range of disciplines can collaborate, inform and share best practices with each other and with international experts.

#### 28 October 2016

## Summarised review of the results from the 1959 complex expedition for investigating the natural outbreak of diseases in Bulgaria

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#### ABSTRACT

The first complex expedition carried out by three units of BAS and two institutes – Institute of Ministry of Public Health and Ministry of farming was held in 1995. The expedition was initiated by Academician Yevgeny Nikanorovich Pavlovskyi during his visit at the Institute of Zoology with Museum of BAS in 1958.

It was organized and managed by Correspondent member, Professor Georgi Paspalev, director of the Institute of Zoology with Museum, BAS. The main findings of the study were published in 1961 in the BAS issued book "Complex research of the natural outbreaks of diseases in the region of Strandzha Mountain" and it contained 14 articles.

The publications cover information about the origin of collected samples, the number of examined mammals, birds, amphibians and reptiles, as well as infestations by helminths and ectoparasites. The extentsity and intensity of various infections were evaluated both in farmed animals and humans.

As a conclusion it was recommended to carry out complex expeditions for investigating the natural outbreaks of diseases in the same or other suitable regions of the country. Additionally, it was suggested to encourage further combined research on similar problems in the Balkan region.



# Population Genetics and Molecular Biology

Session chair: Professor DSc Paraskeva Michailova

Session secretary: Assist. prof. Dr. Peter Boyadzhiev

# Possibilities for discrimination of Bulgarian honey bees based on different genetic markers

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### **ABSRACT**

The genetic variability of honey bee populations from 24 Bulgarian provinces and populations of A. m. carnica, A. m. caucasica, A. m. ligustica and A. m. macedonica has been studied using 15 allozymic and microsatellite loci (MDH, ME, EST, ALP, PGM, HK, Ac011; A024; A043; A088; Ap226; Ap238; Ap243; Ap249 and Ap256). All of the studied allozymic loci were found to be polymorphic with three to six alleles in most of the populations. Polymorphism in all of the populations for all of the 9 analyzed microsatellite loci was observed and discussed. A presence of a total 121 alleles was reported. Thirtyfive private alleles were observed in 19 of the studied populations. UPGMA and Neighbor-joining phylogenetic trees were constructed on the base of the calculated Nei's genetic distance between studied populations. Genetic markers, appropriate for discrimination of Bulgarian honey bees were found and described. Some differences between Bulgarian and other A. m. macedonica honey bee populations were deliberated. It was suggested that local Bulgarian honey bees could be a different ecotype of A. m. macedonica.

**Key words:** *Apis mellifera*, allozymes, microsatellites, polymorphism, Bulgaria

# Application of DNA barcoding in *Eurygaster integriceps* specimens to determine intraspecific genetic variation

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### **ABSTRACT**

Eurygaster spp., commonly known as sunn pest is a major constraint to wheat and barley in North Africa through Middle East and Eastern Europe. In addition to feeding on both the vegetative and generative stages of the plant, sunn pest also affects the quality by releasing digestive enzymes that greatly reduce the baking quality of the flour. By far the most economically important species of the sunn pest are Eurygaster integriceps Puton, Eurygaster maura (L.) and Eurygaster austriaca (Schrank). Following species can be distinguished using traditional taxonomic methods based on keys and descriptions. However due to variety of color morphs especially in E. integriceps, examination of a male specimen's genitalia is the only certain method of determination. This approach has some drawback such as only use of male genitalia; nymphs will require rearing to adulthood for species determination and error-prone due to variation among populations. Therefore DNA barcoding analysis can overcome many of the aforesaid problems.

In this study, *E. integriceps* samples were collected from fifteen different locations of Turkey in order to develop a DNA barcoding tool among sunn pest populations. After genomic DNA isolation, mtDNA sequences were analyzed using cytochrome oxidase (COI) gene from morphologically identified specimens. The obtained sequences were analyzed in terms of nucleotide composition, nucleotide pair frequency. Genetic divergence among haplotypes was estimated by con-

structing genetic distance matrix using DNA sequence variations, by Kimura 2-parameter model. Variable sites and average variations of the sequenced 500 base pair long DNA fragment were calculated and a Neighbor Joining phylogenetic tree was generated. Moreover, the levels of gene flow among populations in different geographic areas are also discussed.

**Key words:** sunn pest, mtDNA, cytochrome oxidase I, phylogenetic analysis, genetic variation

Biodiversity, Ecology and Conservation Zoology

Session chair: Professor DSc Georgi Markov

Session secretary: Assist. prof. Dr. Vesela Mitkovska

# Ex situ effects of priority substances on the lysosomal membrane stability and respiration rate in zebra mussel (Dreissena polymorpha) 1. Heavy metals

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#### **ABSTRACT**

In the current study we aimed to investigate the lysosomal membrane stability in haemocytes of the invasive mollusk zebra mussel (Dreissena polymorpha) by applying the neutral red retention assay (NRRA), as well as changes in the respiration rate and survival under acute heavy metal exposure. The mussels were treated with different concentrations of Ni and Pb in laboratory conditions for a total period of 72 h. These metals are considered as priority substances in surface waters according to Directive 2008/105/EO. The metal concentrations were prepared as 100, 75, 50 and 25% of the maximum permissible levels set by national and EU law. We found out that after the first 24h of exposure, the lysosomes retained the dye between 60 and 90 min. in the mussels exposed to the higher Ni and Pb concentrations (100, 75 and 50%). We also registered a negative, statistically significant correlation between the metal concentrations and the average time the lysosomes retained the dye after the 24<sup>th</sup> and 72<sup>nd</sup> h. Moreover, we found that the lysosomes could not retain the dye more than 60 min. after the 72<sup>nd</sup> h of exposure compared to the control (100 min.). The respiration rate increased in a dose-dependent manner after the 24<sup>th</sup> and 72<sup>nd</sup> h. We can conclude that the acute metal exposure, including all metal concentrations below the allowable concentrations, lead to destabilization of the lysosomal membrane stability and changes in the respiration rate of zebra mussel, thus altered physiological functions.

**Key words:** *Dreissena polymorpha*, heavy metals, lysosomes, neutral red retention, respiration rate

⇒ This study was supported by the NPD – Plovdiv University "Paisii Hilendarski" under Grant № NI15-BF-003, Integrated biological approaches for monitoring priority substances in water.

# Ex situ effects of priority substances on the lysosomal membrane stability and respiration rate in zebra mussel (Dreissena polymorpha) 2. Pesticides

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### ABSTRACT

The present study examines the lysosomal membrane stability in haemocytes of zebra mussel (*Dreissena polymorpha*) by applying the neutral red retention assay (NRRA), as well as the respiration rate and survival under acute pesticide exposure. The mussels were treated with different concentrations of chlorpyrifos in laboratory conditions for a total acute exposure period of 72 h. This organophosphorous substance is considered as a priority contaminant according to Directive 2008/105/EO. The pesticide concentrations were prepared as 100, 50 and 30% of the maximum permissible level set by the national and EU law. We found out that destabilization of the lysosomal membrane stability occurred at all tested concentrations and the respiration rate showed a time and dose-dependent pattern. In summary, we consider that the results from such experiments can be successfully applied in risk assessment, monitoring programs and water policy, and the use of pesticides such as chlorpyrifos should be controlled very cautiously in plant protection and agriculture.

**Key words:** *Dreissena polymorpha*, chlorpyrifos, lysosomes, neutral red retention, respiration rate

⇒ This study was supported by the NPD – Plovdiv University "Paisii Hilendarski" under Grant № NI15-BF-003, Integrated biological approaches for monitoring priority substances in water

# New data on fungus gnats (Diptera: Bolitophilidae, Keroplatidae and Mycetophilidae) in Bulgaria, Greece and Turkey

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#### **ABSTRACT**

New faunistic data on fungus gnats are presented: 1 species new to Bulgaria, 2 to Greece and 6 to Turkey. 14 new localities for some other species in these countries have been reported too.

# First record of the relic *Pentacyphona* stat. nov. in the Palearctic area (Diptera, Pediciidae) with discussion on relationship with their kin

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## ABSTRACT

Eocene period (55.8 – 33.9 My) was characterized by several land bridge connections between Europe and North America which allowed for temperate and later also for cool adapted species to cross the space between nowadays distant continents. There are some examples among opilionids, mammal and chestnut species which have demonstrated that migration in certain cases was possible not only via Beringian strait, but also between direct connection (the so called de Geer land bridge) between Europe and North America. In the present study the species collected by us in the Carpathians (Tarcau Mts.) belong taxonomically to Pediciidae, genus Pentacyphona stat. nov. which is the first record of the genus in the Palearctic area. Previously the Pentacyphona classified as the subgenera of genus Tricyphona or Pedicia. Our genetic and morphological comparison results suggest, that is represent a separate subgenera. The other members of this genus were recorded only from North America. A number of 12 species were identified from the Eastern and Western parts of North America. The species discovered by us wear a number of conspicuous morphological features which are characteristics only to *Pentacyphona*. However, the newly discovered species are deeply divergent from its kin by having a lobe on the dorsal side of the gonocoxite. Similar disjunct distribution was discovered in some other Pediciidae taxa, too like Nasiternella or Dicranota (Plectromyia) which should reflect similarly old and relic like divergences.

# Bacteria associated with *Rhagoletis cerasi* (Diptera: Tephritidae)

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### **ABSRACT**

The European cherry fruit fly, Rhagoletis cerasi (L.) (Diptera: Tephritidae), is a highly destructive pest of sweet and sour cherries in Turkey. Its management is becoming increasingly difficult as previously effective but broad-spectrum insecticides are removed from the market. There is a need suitable and environmentally safe alternative method. Environmental protection regulations and the disadvantages of using chemicals have prompted the development of environmentally friendly pest control methods. Microbes are important for cherry fruit fly nutrition, growth, reproduction and control. The population suppression approach, known as Incompatible Insect Technique (I.I.T.), has been successfully used for the population control of pest species. R. cerasi adults were screened by PCR with 16S ribosomal RNA gene for gut and reproductive bacteria. Klebsiella, Pantoea and Enterobacter were commonly associated with R. cerasi population. Although Cardinium, Hamiltonella, Ricketsia and Arsenophonus were not detected, Wolbachia were detected in the population. Our aim is to raise awareness that insect symbionts can be interesting sources of biological control.

**Key words:** *Rhagoletis cerasi*, symbionts, *Wolbachia*, molecular techniques.

# Ichneumon wasps (Hymenoptera: Ichneumonidae) reared from tortrix moths (Lepidoptera: Tortricidae) in the oak forests in Sofia region, Bulgaria

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## ABSRACT

During the period of 2011 – 2013, studies were carried out in the oak forests in Sofia region. Twenty-four species of parasitoids (Hymenoptera: Ichneumonidae) were reared by the host insects (Lepidoptera: Tortricidae). Among them, eight parasitoids are new records of the parasitoid complex of the hosts: Apechthis rufata (Gmel.), Theronia atalantae (P.), Lissonota saturator (Th.), Lissonota carbonaria H., Lissonota culiciformis Grav., Scambus calobatus (Grav.) – on Archips crataegana (Hüb.); Mesochorus punctipleuris Th. and Trichomma enecator (R.) - on Eudemis profundana (Den. & Schiff.). Seven parasitoid-hosts relationships are new for Bulgaria: Itoplectis maculator (F.), Phytodietus polyzonias (F.), Apechthis quadridentata (Th.) -A. crataegana; T. enecator, Exochus lictor H. – Archips xylosteana (L.); A. quadridentata – Tortrix viridana L. and A. rufata – Pandemis cerasana (Hüb.). Two ichneumon species – M. punctipleuris and Apophua genalis (Moll.) have been found only at faunistic studies in Bulgaria. No data about hosts from family Tortricidae, concerning M. punctipleuris, was found in entomological literature. During the studies, low rates of parasitoid infestation were established.

**Key words**: parasitoid-host relationships, Ichneumonidae, Tortricidae, oak forests, Bulgaria.

# Red List of Bulgarian bombyces and sphinges (Insecta: Lepidoptera) and their distribution in Bulgaria

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### **ABSRACT**

Currently 119 species of the bombyces and sphinges are unambiguously confirmed for Bulgaria. All these species were assessed against the IUCN Red List Criteria at national level. Four species (40%) of Cossoidea, three species (20%) of Drepanoidea, six species (29%) of Lasiocampoidea, 10 species (29%) of Bombycoidea and nine species (24%) of Noctuoidea: Notodontidae, are considered threatened (Vulnerable, Endangered or Critically Endangered) based on the criteria. The areas with the highest concentration of threatened species are situated in SW Bulgaria, Eastern Rhodopi Mts. and the Black Sea Coast. Generalized distribution maps are provided for the species in each category.

# An assessment of a hypersaline lake: case study of Atanasovsko Lake, Bulgaria

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### **ABSRACT**

Atanasovsko Lake (7,200 ha) is situated in Southeast Bulgaria, incorporates diverse habitats and lies along Europe's second largest migration route, the Via Pontica.

The status and trends of water quality (water chemical parameters and biological quality elements) and the factors controlling water quality, were studied for 3-years.

The key factor for the lake is the salt production mechanism (salt pans were created in 1906) and sea-freshwater exchange.

Baseline information contained in this research will be incorporated into future data analyses of the lake under the project Salt of Life, LIFE11 NAT/BG/000362.

# Fish based method for water monitoring and ecological classification of natural lakes on the Bulgarian Danube floodplain

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### **ABSRACT**

The natural water bodies along the Bulgarian Danube section belong to the lake type L5 according the System "B" as defined in WFD/2000/60/EC. The proposed method was developed based on the data of 15 years ichthyological studies in the Srebarna Lake which thanks to its highly protected status is the only lake on the Bulgarian Danube floodplain with close to the natural conditions regardless the existing human pressure.

This method is based on the analysis of information about the composition and structure of the ichthyocenose in comparison with the referent model fish community defined by historical data and by data of specific hydromorphological features of the water body. Quantitative sampling is performed with standard NORDIC type multimesh gillnets twice a year of at least 8 sampling sites covering proportionally all main fish habitats. The quantitative parameters (numbers and biomass of fish) are calculated as CPUE = 100 m<sup>2</sup> net per 1 hour conditional exposition.

Seven parameters characterizing both the fish community (species composition, total abundance, trophic structure and health status) and type specific species/guilds (frequency, dominance and size-age structure) with 11 indicators are used to determine the ecological status. Every indicator meets certain value (points) according to its significance and to the specific metrics. The maximum total score of points for referent conditions is 100. The assessment is based on the propor-

tion "real score/referent score" (EQR) varying from 1 (referent conditions) to 0.

The proposed method can be adapted also for the natural lakes situated on the Black Sea coast.

**Key words:** fish, model community, Danube floodplain, natural lakes, referent conditions, EQR

# Health status of *Pelophylax ridibundus* (Amphibia: Ranidae) in a rice paddy ecosystem in southern Bulgaria: body condition factor and fluctuating asymmetry – Part 1

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#### **ABSRACT**

Pollution effects on the morphological parameters were investigated for Pelophylax ridibundus populations, which were collected from two sites in southern Bulgaria: the rice fields (RF) Tsalapitsa and the Vacha River (Reference site - RS). Morphological analysis showed significantly different values of the index for fluctuating asymmetry – FAMI and body condition factor (CF) in frogs from RF, compared with those from RS. These findings provide information on the longterm background pollution of the habitat (RF). In our view, the lower values of body CF and the high levels of FA that were found in P. ridibundus populations from the rice fields Tsalapitsa, are consequences of the negative effects of the presence of xenobiotics in paddy cages – pesticides and fertilizers, imported by man in rice production. The present work proves the possibilities for practical application of integral indicator for developmental stability - the fluctuating asymmetry in the P. ridibundus populations in bioindication analyses, for assessing the ecological status of agroecosystems.

**Key words:** *Pelophylax ridibundus*, developmental stability, morphological parameters, pesticides, bioindication.

# Tracking methods for lesser kestrel (Falco naumanni) used in the course of the species' recovery as breeder in Bulgaria

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#### ABSRACT

According to IUCN requirements, in species reintroduction, a monitoring of the process, and its results are compulsory. Considering this, when Green Balkans started the lesser kestrel reinforcement in Bulgaria, the following key methods for marking and tracking were chosen – ringing, radio and satellite tracking. The species was considered extinct from nesting in this country for the past several decades, and the organization started reinforcing the specie. Little chicks were used, hatched ex-situ conditions in specialized centers. The chicks were released in a specialized Lesser Kestrel Release and Adaptation Module. Considering that young and inexperienced chicks are being released in the wild, the need for tracking for the subsequent adaptation, threat evaluation, and determination of other ecological aspects is of the utmost importance. Every single released bird has a standard ornithological ring on one leg and a specialized PVC one (orange with two or three symbol code, a combination of letter and/or numbers in black) on the other. In the 2013 – 2015 period we have marked a total of 289 individual birds. In addition, 6 birds varied in age as in gender, were marked with radio transmitters (2.38 gr. - PIP Ag393 Tag.). 4 Platform Terminal Transmitters - PTTs (5g Solar PTT-100 backpack) were used to track lesser kestrels. Radio and satellite tracking is used for the first time in this country in tracking lesser kestrels, and there has been no one to use this method for studying this specie.

**Key words:** lesser kestrel, *Falco naumanni*, tracking, ringing, reinforcement, re-stocking, recovery

# Ichneumonidae (Hymenoptera) fauna of Davraz Mountain in Isparta, Turkey

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#### **ABSTRACT**

This study was carried out to determine fauna of Ichneumonidae (Hymenoptera) in Davraz Mountain in Isparta province. 500 specimens belonging 19 subfamilies were collected at three stations with sweep net and Malaise trap according to altitude between May 2009 – September 2009 and 34 species belonging to 13 subfamilies have been identified. The distribution of species are each of 6 species from Tryphoninae and Pimplinae, each of 4 species from Cryptinae and Diplazontinae, 3 species from Banchinae, each of 2 species from Ctenopelmatinae, Ophioninae and Tersilochinae, each of 1 species from Adelognathinae, Anomaloninae, Collyriinae, Ichneumoninae and Orthocentrinae. The synonyms and general distribution of all species were given on the basis of literatures, as well as known host information.

Within this study identified two species are new records for the Turkish fauna. These are *Enizemum ornatum* (Gravenhorst, 1829) and *Lissonota* (*Lissonota*) pectinator Aubert, 1972 also *Enizemum* genus is recorded for the first time in Turkish fauna.

**Key words:** Hymenoptera, Ichneumonidae, Davraz Mountain, fauna, Isparta – Turkey.



Session chair: Professor DSc Iva Christova

Session secretary: Assoc. prof. Dr. Todorka Yankovska-

# Viral and bacterial zoonoses in humans (zooanthroponoses) with similar clinical manifestation

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#### **ABSTRACT**

Crimean-Congo hemorrhagic fever (CCHF) is a tick-borne human viral disease with fatality rate up to 30%. It is characterized by a sudden onset of fever, muscular pain, often progressing to hemorrhagic manifestations. Natural reservoir hosts include various wild and domestic mammals. Primary vector and reservoir hosts are ticks from the genus Hyalomma. Humans become infected through bite of infected tick or direct contact with tissues or body fluids of viremic animals or CCHF patients. Several hantaviruses cause hemorrhagic fever with renal syndrome (HFRS). Hantaviruses are maintained in rodents (order Rodentia, families Muridae and Cricetidae), insectivores (order Lipotyphla, families Soricidae (shrews) and Talpidae (moles)) and also bats (order Chyroptera). Hantaviruses can be transmitted to humans, most commonly through inhalation of infested rodent excreta. Each hantavirus is associated with a specific natural reservoir. HFRS caused by Dobrava hantavirus is endemic in the Balkan countries and Alpe-Adrian region. It is usually a severe illness presented with hemorrhages, fever, acute renal failure often requiring dialysis, and has a case-fatality rate up to 10%. Leptospirosis is endemic in warm-climate countries worldwide. Various species of wild and domestic animals serve as maintenance hosts. Humans can be infected by direct contact with urine or tissues of infected animal or indirectly by contaminated water. The severe form of the disease is often associated with multiple organ failure and fatality rate is up to 30%. Located on the Balkan Peninsula, Bulgaria is known as endemic for CCHF, HFRS and leptospirosis. Recent investigations of vectors, reservoirs and hosts are discussed.

# Anthrax - the old-new challenge

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#### **ABSTRACT**

In 2016 in Bulgaria there were five outbreaks of the disease, which affected four inhabited areas – three in Silistra regions and one in Yambol region.

For the timely control of the disease it is essential to educate animal owners and farmers about the clinical symptoms of anthrax, in order to ensure quick recognition of the illness. Furthermore, they need to be informed about their obligation to report changes in the health of animals and any deaths to their practitioning vet or to a representative of the Bulgarian Agency for Food Safety.

# Monitoring and control of influenza in birds in Bulgaria

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### **ABSTRACT**

Annually the Bulgarian Food Safety Agency performs a program of supervision and control of Influenza in domestic and wild birds on the territory of the entire country, approved and co-funded by the European Commission (EC).

The most recently found outbreaks of Influenza are from 2015 in Burgas region (in domestic and wild birds) and in Silistra region (in wild birds).

The disease was successfully eliminated in accordance with European legislations and now there are no cases of bird Influenza in Bulgaria.

# New cases of rabies in foxes in Bulgaria

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### **ABSTRACT**

This is the first year since 2014 in which two cases of rabies in foxes were found (in the regions of Sofia and Silistra). Immediate precautions were taken to inform the population and vaccinate domestic animals.

In October this year a campaign for oral vaccination of foxes against rabies will be undertaken in areas of risk.

# Dynamics of echinococcosis in Bulgaria for the period $2011-2015\ year$

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### ABSTRACT

Parasitic zoonoses have important health and social significance in Bulgaria. Echinococcosis, one of the most serious ongoing helminth infection often leads to prolonged inability to work, not infrequently to permanent disability, recurrences and is also possible a fatal outcome. Since 1992, the incidence of hydatid disease increased sharply and reached 8.47%000 in 1998 and 8.32%000 in 2002. So in 2004 the country's government adopted a National Programme for Control of echinococcosis in humans and animals, which lasted until 2008. After the implementation of the medical part of the programme the incidence of echinococcosis was lowered to 3.95%000 in 2010. An increase in the incidence was observed again after 2011 from 4.09%000 till 4.35%000 in 2015. Total for the period from 2011 to 2015 were registered 1643 cases of hydatid disease, from which 1488 were primary and 155 relapses. The majority of reported cases were found in adults, but the disease is not rare among children and adolescents. For the same period were registered 282 cases of hydatid disease in children from 1 to 19 old or 17% of all diseased. The high proportion of children infected by hydatid disease indicates the presence of active transmission of the parasite.

# Brucellosis in sheep and goats (Brucella melitensis)

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### **ABSTRACT**

In June 2006, in Bulgaria were established the first epizootic outbreaks of brucellosis in sheep and goats (*Brucella melitensis*) since 1941. By the end of 2008 this disease is registered in 16 populated areas in 4 regions – Smolyan, Yambol, Haskovo and Stara Zagora. To eradicate this zoonosis 496 goats, 117 sheep and 7 large cattle were killed.

In 2015 epizootic outbreaks of the disease were found in Smochevo village, Rila town, Padala village, Rila Monastery, Rila municipality, Mursalevo village, Kocherinova municipality from the Kyustendil region, Kochan village and Satovcha municipality from the Blagoevgrad region.

To date the outbreak of brucellosis in sheep and goats in Kochan village, Satovcha municipality from the Blagoevgrad region remains active.

# Realization of the programme for prophylactics, supervision and control of the West Nile fever in Bulgaria

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#### **ABSTRACT**

The 60s of the XX century was the first time antibodies against the West Nile fever were found in Bulgaria. In 1978 using the immunofluorescent method the virus is found in bloodsucking insects.

During the period 2010 - 2014 the circulation of the infection is determined in 4 regions in the country with high risk: Durankulak and Shabla (Dobrich region), Tutracan (Silistra region) and Staro Orjahovo (Varna region).

In 2015 antibodies against the West Nile fever in Bulgaria were found in sentinel animals in 4 regions with proven circulation, as well as in the newly added for the current year risk regions: Ruse, Vidin, Lom, Kozloduy and Svilengrad. Additionally, the West Nile Fever virus to line two was found in samples of wild birds from the Corvidae family collected in Sofia. Serological testing shows the presence of antibodies in wild birds, equids and dogs.

# **Tuberculosis in large ruminants**

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### **ABSTRACT**

The main priority of the Bulgarian Food Safety Agency (BFSA) is rapid eradication of active outbreaks of tuberculosis in large cattle in Bulgaria and acquiring a status a disease-free country. For this purpose, the National Programme is annually performed, and since 2016 a strategy for faster and effective eradication of epizootic outbreaks has been undertaken.

# Ixodidae ticks in domestic ruminants in the valley of Maritsa River in the Plovdiv region

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### **ABSTRACT**

The species composition, the distribution, the seasonal dynamics and the indices of invasion of Ixodidae ticks invading sheep (Ovis aries), goats (Carpa aegagrus hircus) and cattle (Bos taurus) in four municipalities near Maritsa river in Plovdiv region were studied. During 2011 – 2016 were examined 584 sheep, 521 goats and 114 cattle from localities throughout investigated region. (The species composition, distribution, seasonal dynamics and invasion indices of Ixodidae ticks which invade sheep, goats and cattle were studied in 4 municipalities along the river Maritsa in the Plovdiv region. In 2011 - 2016 584 sheep, 521 goats and 114 cattle were studied in this area). The invaded animals were examined clinically. Eight types of Ixodidae ticks which invade domestic ruminants were established in the researched area – Rhipicephalus bursa, Rhipicephalus sangiuineus, Hyaloma plumbeum, Dermacentor marginatus, Ixodes ricinus, Boophylus calcaratus, Haemaphysalis sulcata and Haemaphysalis punctata. Rhipicephalus bursa is the predominant invasive species in sheep and goats, and Hyaloma plumbeum predominates in cattle. Some differences in the indices of invasion and seasonal dynamics of the spread of Ixodidae ticks were observed in the different types of domestic ruminants. There were no serious deviations observed in the health status of the invaded animals.

# Supervision and control of salmonellosis in birds in Bulgaria

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### **ABSTRACT**

Supervision and control of salmonellosis in birds is carried out in accordance with the following programs:

- Programme for control of salmonella in breeding poultry flocks;
- Programme for control of salmonella in broilers;
- Programme for control of salmonella in laying hens from the species *Gallus gallus*;
- Programme for control of salmonella in turkeys;

Testing of samples is carried out and further measures are taken according to the requirements of Regulation (EO) № 2160/2003 of the European Parliament and the Council from 17 November 2003 about the control of salmonella and other specific agents which cause zoonosis and are present in the food chain.

# **Lumpy skin disease – new challenge for Europe**

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### ABSTRACT

For Europe, Lumpy skin disease was an exotic sickness widespread in many African countries. In 2012 however, it started to observe a serious trend of virus invasion in the Middle East countries and in 2015 the disease spread in Turkey, regions of Russia, Georgia, Cyprus and Greece.

In early April 2016, days after the Greek authorities announced an outbreak of Lumpy skin disease in town Sérres (less than 10 km of the Bulgarian border). Within the clinical examination performed in the country, the sickness was registered for the first time in Bulgaria in two farms in the central southern part of Bulgaria (Dimitrovgrad Municipality). In only 3 months, 217 outbreaks were recorded in 17 regions of the country. In parallel, the virus began to invade the northwest and spread to almost all countries of the Balkan Peninsula.

# Food explosion of salmonellosis in Varna – practical aspects

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### **ABSTRACT**

For the period May – June 2015 on the territory of Varna and the region, several cases of sickness in organized children's collectives have been registered. In all cases, Salmonella was proven as the cause – groups D, enteritis, group E and B. Together with employees of Regional Health Inspectorate – Varna a thorough epidemiological investigation was conducted and the necessary actions to limit the disease were identified.

In the central management (CM) of BFSA on the basis of these results a "Procedure for reaction to a crisis situation – suspected explosion of food illness" was developed and approved. By order of the executive director of the BFSA the procedure is approved since December 2015.



### The response of Chironomidae (Diptera) genome to heavy metal pollution in two rivers of Southern Poland

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#### **ABSTRACT**

The effects of heavy metals pollution in two Polish rivers on the genome of phylogenetical different species of family Chironomidae were studied. The concentrations of Pb, Zn, Cd, and Cu in the channel sediments of Chechło and Biała Przemsza Rivers were many times higher than reference data, as a result of Pb-Zn mining activities. In the studied species: Prodiamesa olivacea Meigen, P. bureshi Michailova, Chironomus riparius Meigen, C. annularius Meigen genome instability was evaluated by somatic and inherited chromosome aberrations. On the basis of somatic alterations the somatic index (S) has been calculated, and shows highest value in genus Chironomus: (S=2 in C. annularius). This index is a good biomarker for assessing the genotoxic effect of contaminants in the sediments. In both Prodiamesa species from Biała Przemsza River heterozygous somatic aberrations occurred significantly more often (P<0.001) than in the same species in unpolluted region previously described. The same response was found in *Chironomus* species. Different types of ectopic conjugations between chromosome arms appeared with very high frequency in P. bureshi (61.15%) and P. olivacea (81.95%) but in *Chironomus* species occurred in single cells only (about 3%). The mouthpart deformities were detected with highest frequency in P. olivacea (26.9%). The results showed that the species from different genera has a species – specific genome reaction to stress agents in the environment. The data obtained support the idea these species to be usefully explored in biomonitoring studies in aquatic ecosystems.

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Genetic comparison between local *Apis mellifera* macedonica, selectively reared for production of bee queens and swarms in Bulgaria and honey bee colonies with indicative hygienic behavior

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#### **ABSTRACT**

The genetic polymorphism of MDH-1 and EST-3 loci in selectively reared for production of bee queens and swarms in Bulgaria local honey bee populations of Apis mellifera macedonica was studied in comparison with honey bee colonies with indicative hygienic behaviour. Totally 851 worker bees collected from the Selection bee rearing bases and 414 worker bee individuals from colonies with manifested hygienic behaviour were used for this comparative study. Both of the studied loci were found to be polymorphic in almost all populations represented the Selection bee rearing bases and in almost all hygienic colonies, with the exception of totally six investigated populations where the EST-3100 allele was fixed. Polymorphism with two and three alleles was found for MDH-1 locus and with four and five alleles – for EST-3 locus for compared hygienic colonies and populations under selective control. High levels of polymorphism were calculated for all colonies and populations. The observed and expected heterozygosities (Ho and He), mean FST and Nm values were calculated. The observed differences and similarities among hygienic colonies and populations under selective control were discussed.

# Genetic variability in populations of *Messor barbarus* (Hymenoptera, Formicidae) from Bulgaria based on isoenzyme analysis

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#### ABSTRACT

The genetic heterogeneity in 13 ant populations of *Messor barbarus* (Linnaeus, 1767) from Bulgaria has been studied using analysis of seven enzyme and protein systems which have been found to be appropriate genetic markers for characterization of genetic variability within and between populations. A comparative analysis of gene pool and genotypic structure of the tested populations have been carried out. Moderate to high percentage of polymorphism and low levels of heterozygosity have been calculated. Deviations from Hardy-Weinberg equilibrium in almost all analyzed loci, in favor of homozygotes, have been established. The mean value of inbreeding coefficient (FIS) has been found to be high (0.8212), demonstrating a high level of inbreeding in the studied populations. The obtained data of the genetic variability provide new information concerning polymorphism and phylogenetic relations between the studied populations.

**Key words**: *Messor barbarus*, isoenzymes, genetic variability, phylogenetics

# Genetic variability of *Eupelmus* species (Hymenoptera: Eupelmidae) based on allozyme markers

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#### **ABSTRACT**

Four enzyme systems (MDH, ME, PGM and HK) were studied of three parasitoid species of the genus *Eupelmus* (*E. vesicularis* (Retzius, 1783), *E. urozonus* Dalman, 1820 and *E. microzonus* Förster, 1860) using PAGE. Totally five populations were investigated as three of them belong to *Eupelmus urozonus*. Polymorphism with four alleles of Mdh-1 locus, three of Me loci, four of Pgm loci, four of Hk-1 and Hk-2 loci were observed. Mdh-1<sup>59</sup> and Pgm<sup>123</sup> alleles were diagnostic to *E. vesicularis* and Me<sup>136</sup> allel – to *E. microzonus* and *E. urozonus*. The degree of polymorphism and heterozygosity of each species were calculated. UPGMA cluster analysis confirmed that *E. vesicularis*, which belongs to subgenus *Macroneura* is divergent from *E. urozonus* and *E. microzonus*, which belong to subgenus *Eupelmus*. *E. urozonus* population from Granitsa Village is more similar with *E. urozonus* population from Marchaevo Village than the same of Eremiya Village.

Key words: Hymenoptera, Eupelmus, genetic variability, allozymes

# Potential of PCR – based molecular diagnostic methods for rapid detection of bacterial pathogens causing economically important diseases in fishes

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#### **ABSTRACT**

Aquacultures are the fastest growing sector of food production worldwide, providing half of the fish needed for human consumption and has a significant potential for further growth. The intensive development of aquacultures however created also conditions for rapid spreading of diseases caused by bacterial pathogens. They cause high mortality and significant economic losses. Classic methods often require visible symptoms and significant time for diagnosis. Faster more accurate and reliable methods for detection and identification of key pathogens during the initial stages of diseases outbreaks are needed. Molecular biology can offer new techniques as the best alternative to traditional methods. Using molecular tools pathogens can be detected in at very early stages of outbreaks, infected fishes can be isolated and spreading of diseases can be prevented. Thus, antibiotic treatment may be extended to a limited number of fishes and thereby reduce treatment costs, production losses and reduce the likelihood for appearance of antibiotic resistant bacteria. In this study we tested PCR - based methods on the most common gram-negative pathogenic bacteria Flavobacterium psychrophilum, Yersinia ruckeri and Aeromonas salmonicida. Initially designed primers were tested on isolated cultures of *F. psychrophilum*, *Y. ruckeri* and *A. salmonicida* by colony – PCR to prove they work properly. Next several methods where tested – simple PCR, multiplex PCR, qReal-Time PCR. The multiplex PCR proved to be most cost and time efficient method. The results of this research, although still preliminary, demonstrated the power of molecular diagnostic methods for bacterial pathogens in fish farms.

### Genetic diversity and geographic distribution of round goby *Neogobius melanostomus* (Pallas) as revealed by mtDNA cyt b gene haplotypes

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#### **ABSTRACT**

Round gobies are relatively small soft-bodied fishes, characterized by a distinctive black spot on the first dorsal fin. They are native for central Eurasia. They are euryhaline fishes and can be found both in marine and freshwater habitats including the Black Sea, Sea of Azov, Sea of Marmara and Caspian Sea, costal lakes, lagoons and river from their basins. N. melanostomus is an invasive species, which recently established large non-native populations in the Baltic Sea, several major Eurasian rivers, and the North American Great Lakes threatening native species and local biodiversity. Two expansion mechanisms were proposed: 1) "corridor expansion" via European rivers and human-made channels and 2) "jump dispersal" via ballast waters from international shipping. Molecular phylogeny analyses of marker genes can provide information about origin and of each non-native population and gene exchange rate with other population. Cytochrome b is considered to be the most useful marker in determining phylogenic relationships between closely related organisms e.g. species, populations, due to its sequence variability. For this purpose, we analyzed variability in 184 Cyt b sequences. 71 were isolated by us from of N.

melanostomus captures in Black Sea, rivers Rhine and Mosel Germany and the rest were downloaded from NCBI: accessions KF 549989 – 549990, EU 331156 – 331236, EU 564119 – 564125, KC 814168 – 814174, KC 886276 – 886278, NMU 53673 – 53677, KJ 654330 – 654332, HQ 452491 – 452492, AY 884582 – 884583and KC 800809. Analyses revealed twenty-two haplotypes. Observed value of Haplotype Diversity, Hd was 0.361. Polymorphic sites were 21 according to DnaSP Ver. 5.10.01. Phylogenic analyses revealed that is 80% of the Cyt b sequences belong to haplotype 1. It is widely spread both in West European and US basins. Others form local populations. The geographic distribution, phylogenic links and frequency of polymorphic sites in Europe and North America are discussed.

# The difference in the some quantitatives parameters of the observed bee breeding lines from Montenegro and Serbia

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#### **ABSTRACT**

The main scientific objective of this study was to investigate the difference in the some quantitatives parameters between geographically distant, selected honeybee lines from Serbia and Montenegro. These lines are taken from the Centre for the of selection queen bees in Vrsac, and Vranje (Serbia) and Bijelo Polje and Sutomore (Montenegro). Lines from Bijelo Polje and Vranje are grown in mountainous conditions. The line from Sutomore has been selected in terms of coastal climate, a line from Vrsac belonged flatland region. Were monitored following traits: the estimated number of bees, estimated surface of brood, estimated area of honey, estimated area of pollen, assessment of the quality of the brood, ratings for temperament. Testing differences between the tested lines was performed by analysis of variance using the statistical package Statistica 6 (StatSoft, Inc. 2003). ANOVA was established significance of differences between monitored lines:

• Bee line from Vrsac has the best results in terms of the number of bees in all control checkups. This line is the first autumn examina-

tion is very significantly different (p < 0.01) of all lines in both autumnal control review.

• In terms of the number of brood cells, line of Vrsac is a very significant difference (p < 0.01) of both lines from Montenegro, a significant (p < 0.05) from the line from Vranje in both autumn views.

# Evidence of geno-toxicity induced by 60 Hz magnetic fields on mice bone marrow as assessed by *in vivo* micronucleus test

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#### **ABSTRACT**

Modern life implies a constant exposure of living organisms to electromagnetic fields (EMFs) generated by human made technology. The question of whether or not EMFs in the non-ionizing extremely low frequency (ELF) range can affect cellular functions, increasing the risk of cancer or a variety of pathologies is currently a subject of interest. In the present study, we investigated the potential genotoxic effect induced in mice exposed to ELF-EMFs. The evaluated cytological endpoints included the frequency of micronucleated polychromatic erythrocytes in bone marrow and ratio of polychromatic erythrocytes (PCEs). Three independent experimental conditions were carried out: (a) animals exposed to 60 Hz sinusoidal magnetic fields for 72 h at 1.0, 1.5, and 2.0 mT of magnetic flux density, (b) mice exposed for 10 days/8 h daily at 60 Hz and 2.0 mT, and (c) 72 h exposure to ELF-EMFs at 60 Hz and 2.0 mT plus 5 mg/kg of Mitomycin-C (a well-known clastogenic agent). Statistically significant differences

indicative of ELF-EMF genotoxic effect were observed for micronucleus frequency when compared ELF-EMF exposed and control animals at 1.5 and 2.0 mT exposure conditions. In addition, an opposite effect between ELF-EMF exposure and Mitomycin-C treatment in terms of micronucleus frequency was observed for this co-exposure condition. Regarding PCEs percentages, no statistically significant differences were observed among groups. In conclusion, the present study suggests an association between DNA damage and ELF-EMF exposure as assessed by *in vivo* micronucleus assay in mice.

# Allozyme genetic characterization of *Apis mellifera* colonies from Bulgaria with different hygienic behaviour

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#### **ABSTRACT**

The genetic variability in 25 honey bee colonies from different regions of Bulgaria with diverse hygienic behavior (super hygienic, hygienic and not hygienic) has been studied. Alloenzymic analysis of two systems (MDH-1 and EST-3) corresponding to 2 loci was used in order to characterize the colony polymorphism. Totally 1150 worker bees were included in this investigation. MDH-1 locus was found to be polymorphic in all of the studied colonies, having two alleles – MDH-165 and MDH-1100. The EST-3 locus was fixed in ten of the investigated colonies. Polymorphism with total presence of four alleles of this locus (EST-380, EST-388, EST-3100 and EST-3118) was found in the other studied colonies. The calculated polymorphism was between 50% and 100% with mean level -80%. The mean levels of observed and expected heterozygosities were 0.320 and 0.274, respectively. The calculated mean Fst level was 0.151. Allele frequencies of all studied loci were used to estimate Nei's (1972) genetic distance, which was established to range between 0.000 and 0.319 among the studied colonies with different hygienic behavior. Dissimilarities between levels of polymorphism, heterozygosity, Fst, Nm and allele frequencies in the studied groups of colonies with different hygienic behavior were found and discussed. The results of the present study provide new information concerning relations between hygienic behavior and allozyme characteristics which could be used for future selection with honey bees in Bulgaria.

## Dynamic and ecological impact of marine zooplankton aliens in Varna Bay (Bulgarian Black Sea)

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#### **ABSTRACT**

The introduction of non-native species, respectively invasive species, especially into coastal waters is among the four highest risks for the marine environment, exaggerated further in relation to the global climatic changes projections and caused extremely severe environmental, economic and public health impacts. An issue for the alien species management is that, once a marine organism has been introduced to its new environment, it is nearly impossible to eradicate the invader, if it has established to the area. Recently four zooplankton species are important for the zooplankton community structure, two species of subclass Copepoda (Acartia tonsa and Oithona davisae) and two species of phylum Ctenophora Mnemiopsis leidy and Beroe ovata. As a key factor for the development of mesozooplankton, Mnemiopsis leidyi becomes a reliable indicator of the pelagic Black Sea ecosystem dynamic, functioning of the pelagic food web, ecosystem stability and health. Investigations were carried out in springsummer-autumn period of 2015 - 2016 in conformity of ESENIAS project. Study area was Varna Bay Seasonal dynamic of nonindigenous zooplankton species were presented and analyzed. Large aggregates of M. leidyi formed "hot spots" along the coast during the summer and ecological state according to the biomass of M. leidyi varied from bad to good in respect of Water Frame Directive indicators. The WFD did not require the zooplankton as a biological quality element but zooplankton was included as a complementary element at National monitoring programme of coastal marine waters in Bulgaria.

### Summary of significant fungal infections in mollusca

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#### ABSTRACT

The adverse social, economic and environmental consequences of uncontrolled movement of live aquatic animals and their products have increased global awareness of the need for improved health management standards. The serious impact of unrestricted international movement of aquatic animals has led to the development of health certification and risk reduction methodologies.

Increasing development of shellfish aquaculture, and recent advances in diagnostic techniques, along with diversification of cultured species, continue to provide a seemingly inexhaustible reserve of new or emerging infectious disease problems.

Numerous species of fungi have been described from various shellfish, especially representatives of the Mollusca. Some fungi have had a serious impact on wild populations and shellfish aquaculture production.

Key words: fungi, infections, Mollusca

# Summer rotifer assemblage in three Macedonian reservoirs (Konče 1, Konče 3 and Špilje)

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#### **ABSTRACT**

The effects of environmental factors on rotifer assemblage in reservoirs that differ in size and trophic state: eutrophic reservoirs Konče 1 and Konče 3 and mesotrophic reservoir Špilje was examined. Qualitative analyses of summer rotifer composition in Konče 1, 3 and Špilje showed the presence of 13 and 10 taxa, respectively. Rotifer assemblage significantly differed in their community structure, population densities and the occurrence pattern of dominant species, due to the significant differences observed in the environmental parameters measured between Konče 1, 3 and Špilje. The average density of rotifers in Konče 1 and 3 was  $50.049 \pm 7.825$  ind 1<sup>-1</sup>, whereas  $2.898 \pm 4.08$ ind 1<sup>-1</sup> were recorded in Špilje. In Konče 1 and 3 rotifers were dominated by Brachionus falcatus, Brachionus angularis and Hexarthra mira, contributing 28%, 27% and 23% to rotifer abundance, respectively. In contrast, in Špilje was dominant species Kellicottia longispina, comprising 47% of the total rotifer density. The results suggest that Rotifer abundance, as well as species composition and distribution, often reflect the trophic status of aquatic ecosystems.

Key words: Rotifera, reservoir, structure, density, dominant species

# Contributions to the study of earthworms (Oligochaeta, Lumbricidae) of the Pannonian region of Serbia (Vojvodina Province)

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#### ABSTRACT

Vojvodina is a province in the northern Serbia, situated in the part of the southern Pannonian Region. The Province of Vojvodina (44°38' – 46°10'N; 18°10' – 21°15'E) is predominantly a flat region, and more than 75% of its territory is used for agriculture. The aim of this paper is to present new data on the earthworm fauna of the Vojvodina Province. During the study from 2013 to 2016 in the western and south parts of Vojvodina eleven earthworm species were recorded in the several new localities. On the basis of existing information on the earthworms from the entire territory of Vojvodina, we have summarized all published data and have established the definitive list of known earthworm taxa of the Province. The list comprises 33 species and subspecies, belonging to 11 genera of the family Lumbricidae. Majority of the recorded species belong to the genera Aporrectodea and Allolobophora. Our study shows that the degree of endemism is quite low compared with the endemic taxa recorded from the territory of Serbia situated on the area of the Balkan Peninsula.

Distribution and biogeographical significance of the endemic earthworm *Allolobophora robusta spAssenijakaramani* (Blakemore, 2004) (Oligochaeta: Lumbricidae) on the Balkan Peninsula: first finding place in Macedonia

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#### **ABSTRACT**

The objective of this paper is to analyze the whole list of records in order to present a general overview of distribution of *Allolobophora robusta spassenijakaramani* (Blakemore, 2004) on the Balkan Peninsula. It belongs to the larger endemite on the Balkan Peninsula. During the last 20 years this subspecies has been recorded from many localities in central and southeastern part of Serbia. Until the present, the northernmost finding of the species has been in the central part of Serbia, while the southernmost point is in southern part of Serbia. However, after the investigation in last two years *Allolobophora robusta spassenijakaramani* recorded in the south part of the Balkans. Gostivar (Šar Mts.), in the state of Macedonia, is the southernmost locality reported so far, extending its known distribution area more than 280 km to the south. Its area of occupancy is around 250 km². Such a discovery is of a considerable faunistic interest: it represents the first record for Macedonia and the southernmost one reported so far.

# Mating behavior of millipede *Pachyiulus hungaricus* (Karsch, 1881) (Myriapoda, Diplopoda, Julidae) in laboratory conditions

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#### ABSTRACT

Pachyiulus hungaricus (Karsch, 1881) represents one of the largest and the most robust European diplopods, distributed mainly in the Balkan Peninsula. Its morphology, development, taxonomic and biogeographical peculiarities were the subject of some investigations. Some behaviors of this species were also described mostly according to the observations in nature. However, many behavioral traits are still insufficiently known, probably because long-term growing of millipedes in the laboratory conditions is very difficult.

We performed a set of experiments in order to obtain more information about mating behavior of *P. hungaricus*. Millipedes were collected at Mt. Avala, near Belgrade, Serbia, and maintained separately by sex in laboratory conditions for seven days until mating assays were conducted. Considering that *P. hungaricus* is characterized by polygynandry, three types of mating experiments were performed: test in "mating arena" (multiple mating combinations in plastic boxes with a layer of decomposed litter), "female choice" test and "male choice" test. Precopulatory, copulatory and postcopulatory behaviors of both sexes were monitored, described, and statistically analyzed. In all mating assays, millipedes were scored for mating type, mating latency and copulation duration. Obtained results should contribute to a better understanding of *P. hungaricus* mating strategy.

# Cuticular chemoprofile in the fruit fly, *Drosophila* subobscura Collin, 1936 (Diptera: Drosophilidae)

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#### **ABSTRACT**

In Drosophila, chemical communication takes place via the cuticular hydrocarbons (CHCs), which represent a complex mixture of organic compounds. CHCs have different biological roles, starting from preventing water loss and desiccation resistance, up to complex behavioral roles in mating, learning, aggregation and social behavior. Their composition may be affected by sex, age, developmental conditions, social and mating experience, and different ecological factors. Regarding to the literature and available databases, contrary to the most exploited Drosophila melanogaster, epicuticular chemical profiles were insufficiently investigated in obscura group of species, including eight species that were recorded in Serbia. In this work, cuticular chemoprofile of Drosophila subobscura Collin, 1936 was characterized using gas chromatography (GC) and gas chromatography coupled with mass spectrometry (GC-MS). This species, frequently found in Serbia in wild and semidomestic habitats, is commonly used in genetic, ecological and evolutionary studies. Since D. subobscura is marked as an important model system in monitoring of global climate changes, it is important to identify its epicuticular chemoprofile in the context of geographic variability and relate it with different behaviors.

## Morphological analysis of *Branchipus* sp. from area of Stara Planina Mountains and Pannonian lowlands in Serbia

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#### **ABSTRACT**

Morphological variability of specimens belonging to the genus Branchipus originated from six localities in Serbia has been studied. Branchipus sp. is known from a number of localities in Serbia, especially frequently founded in the area of the Pannonian lowlands. In the southern parts of the country, populations were reported in the area of Stara Planina Mountains. In order to establish a degree of variability of Branchipus sp. morphological traits from different parts of the country, multivariate analyses of great number of characters of both sexes have been applied. The results have shown that morphometric characters contribute most to morphological differentiation, while meristic characters have not shown statistical significance. Primary and secondary sexual characteristics were the most discriminant both in males and females. Specimens from different localities have been better defined on the basis of male morphometric characters, than on those in females. There was considerable difference in morphological features of individuals from the northern and southern localities. Specimens from hillymountainous area of Serbia (Stara Planina Mountains) have the most complex level of differentiation. Having in mind the high percentage of correct identification it may be stated with certainty that this analyzed population represents a separate entity.

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First finding of *Lepidurus couesii* Packard, 1875 (Crustacea, Triopsidae) in Serbia – a record based on development of diagnostic body characters in males and females

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#### **ABSTRACT**

Lepidurus couesii Packard, 1875, a new large branchiopod species for Serbia is reported. In Europe, L. couesii has been presented in the steppe zone of the Eastern Balkan region in the area of Danube delta, and from Mediterranean basin. This taxon was unknown to the Serbian fauna until recently, when specimens of both sexes were collected on the periphery of the city of Belgrade in northern Serbia. A bisexual population was found in the flooded area of the Sava River, and is recognized as highly female-biased. Occurrence of new species is based on the analysis of the mature males and females. The evidence is derived according to development of body parts which are considered as diagnostic for determination of this species: the overall body size, appearance and size of carapace, telson and caudal lamina, arrangements of spines, pairs of the legs, number of body rings, form and placement of the nuchal organ, characteristics of first trunk appendage. Presented results are in line with morphological observations of this species published by several authors, and support the existence of new notostracan species L. couesii in Serbia and in the region of Western Balkans.

### Study of contents of lead, cadmium and nickel in the water and in metabolically active organs of crayfish Astacus leptodactylus in Kardzhali dam

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#### ABSTRACT

It is examined the contents of lead, cadmium and nickel in the water of Kardzhali dam, as well as muscle and hepatopancreas of *Astacus leptodactylus*. In water samples is taken into account increasing of lead at one point of study. In tissue samples is reported increasing of cadmium in samples of hepatopancreas. The analyses give reason crayfish to be defined as a biomarker for contamination of any water.

**Key words**: heavy metals, *Astacus leptodactylus*, crayfish, contaminated water

First record of cysticercoids of *Microsomacanthus* (*Microsomacanthus*) abortiva (von Linstow, 1904) Lopez-Neyra, 1942 (Cyclophyllidea: Hymenolepididae) in *Gammarus* sp. (Amphipoda, Gammaridae) in Bulgaria

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#### **ABSTRACT**

Cysticercoids isolated from amphipod crustaceans from the Poda Protected Site (Burgas Region) in the summer of 2015 were studied. Up to 50 cysticercoids of *Microsomacanthus* (*Microsomacanthus*) abortiva (von Linstow, 1904) were found in the body cavity of 3 specimens of *Gammarus* sp. These data indicate the local transmission of the parasite in the conditions of Burgas Wetlands. *M. abortiva* is parasitic in caeca of domesticated and wild waterfowl (Anatidae). Previously adults of this species were reported from 4 aquatic bird species in Bulgaria: *Anas platyrhynchos*, *A. platyrhynchos* f. *dom.*, *A. acuta* and *Anser anser* f. *dom.* (Vasilev, 1962; Kamburov and Vasilev, 1972; Vasilev, 1973). This study is the first record of cysticercoids of hymenolepidid cestodes in *Gammarus* sp. in Bulgaria.

### Morphometric variability, allometric growth and sexual dimorphism in narrow-clawed crayfish (Astacus leptodactylus, Esch.) during the ontogenesis

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#### **ABSTRACT**

The study on variability of morphometric characteristics of 927 one-summer-old, one-year-old and two-summer-old narrow-clawed cray-fish, cultivated in Institute of Fisheries and Aquaculture in Plovdiv was carried out. The weight-length relationships of two sexes on different age groups were determinated using graphic equations. Allometric growth was observed in all individuals. Significant sexual dimorphism on 12 of studied exterior characteristics was established. Higher values of the features body weight, carapace length, carapace width, chela length, chela width, chela corpulence, dactyl length, palm width and chela length from the carpal joint to the its tip were measured in males. Concerning the females, in connection with the reproduction higher values on the features abdomen length, abdomen width and telson width were measured.

A survey of natural enemies and associated arthropod pests in pomegranate orchards in Antalya province (South-western part of Turkey)

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#### **ABSTRACT**

A survey of natural enemies and their associated arthropod preys/hosts occurring in the pomegranate orchards in Antalya province (Southwestern part of Turkey) was carried out during the years, 2011 and 2012. Samples were collected by aspirator, hand picking with a fine forceps or a fine brush for tiny arthropods and beating, or jarring, of vegetation. Additionally, small and medium sized branches infested with arthropod pests were collected for subsequent close examination or rearing of immature stages of some predators. Pest-infested plant samples containing parasitoids were also collected to obtain adult parasitoids in emergence boxes. All the collected samples were taken to the laboratory and separated soon after each sampling. The results from the study revealed that 22 species of predators (Coleoptera -Coccinellidae: 13; Hemiptera – Anthocoridae: 2, Nabidae: 2; Dermaptera - Forficulidae: 1; Neuroptera - Chrysopidae: 2; Thysanoptera -Thripidae: 1 and Acarina - Phytoseiidae: 1) and 2 species of hymenopteran parasitoids (Encyrtidae) were detected to be associated with 15 arthropod pest species in the pomegranate orchards in Antalya province. Among the predators found, the coccinellids were the most abundant predator group, followed by Chrysoperla carnea (Stephens) (Neuroptera: Chrysopidae), and both of them were generally associated with the aphids, Aphis gossypii Glover and Ap. punicae (Passerini) (Hem.: Aphididae). Of the two encyrtid parasitoids, *Anagyrus pseudococci* Girault was associated with citrus mealybug [*Planococcus citri* (Risso) (Hem.: Pseudococcidae)] whereas the other [*Cheiloneurus paralia* (Walker)] associated with the Florida wax scale, *Ceroplastes floridensis* Comstock (Hem.: Coccidae).

**Key words**: Pomegranate orchard, natural enemy, predator, parasitoid, Antalya, Turkey

Species of *Ooencyrtus* genus (Hymenoptera: Encyrtidae),

egg parasitoids of *Thaumetopoea solitaria* (Lepidoptera: Notodontidae) in Bulgaria

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#### **ABSTRACT**

The study was conducted during the period 2010 – 2015 in 6 sites located in the Eastern Rhodopes: Madzharovo, Dam Ivaylovgrad, Dupkata, Quarry, Liikanen and Meden buk. A total of 424 Thaumetopoea solitaria egg clusters were collected from these sites and were kept at room temperature. Out of a total of 52,628 eggs in the samples, 3 species of parasitoids from Ooencyrtus genus, O. masii, O. pityocampae and *Ooencyrtus* sp. nr. indefinitus, were found. The dominant species was O. masii, and the least numerous, with only two emerged female individuals - O. pityocapae. With the other two species, individuals from both sexes were established. With O. sp. nr. indefinitus, the ratio  $\mathbb{Q}$ : 33 was almost 1:1, whereas with *O. masii* the female individuals predominated. In laboratory conditions, the period of emergence of parasitoids lasted 41 days. With both species, O. masii and O. sp. nr. indefinitus, a similarity in the character of the emergence dynamics was observed. In 2014 it was more extended, and in the next year more explosive. The peak of emergence of O. masii was 20 - 25 days before that of the other parasitoids.

# A new species of the genus *Omphale* Haliday (Hymenoptera, Eulophidae, Entedoninae) from Bulgaria

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#### **ABSTRACT**

A new species of the genus *Omphale* Haliday (Hymenoptera: Eulophidae: Entedoninae), *Omphale rhodopensis* Yefremova, Yegorenkova & Boyadzhiev, sp. n. is described from the Mts. Rhodope, Bulgaria. This is the first newly described European species of *Omphale* after revision (includes 37 species) by Hansson & Shevtsova in 2012.

Description of *Stepanovia fructirosae* sp. n. (Hymenoptera: Chalcidoidea: Eulophidae) from Turkey

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#### **ABSTRACT**

Stepanovia fructirosae sp. n., reared from galls of Diplolepis fructuum (Hymenoptera: Cynipidae) in the NE Turkey, is described and illustrated. This species is morphologically close to *S. rosae* Boyadzhiev & Todorov. A key for identification of them is presented.

The community members associated with rose gall wasp *Diplolepis fructuum* (Rübsaamen, 1895) (Hymenoptera: Cynipidae) in Tokat Province of Turkey

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#### **ABSTRACT**

Rose gall wasps (Hymenoptera: Cynipidae: Diplolepidini) is a well-known gall inducing insect group on *Rosa* species, and their galls support species-rich communities. During this investigation, field studies were carried out in April, October, and November 2013 and March and October 2014 in Tokat Province of Turkey. Gall specimens of *Diplolepis fructuum* (Rübsaamen, 1895) (Hymenoptera: Cynipidae) on host plants from the genus *Rosa* were collected. Thirteen species from six families were reared. Among them, *Eurytoma caninae* Lotfalizadeh & Delvare, 2007 and *Aximopsis collina* (Zerova, 1984) are new records for Turkey. The most common parasitoids are *Eupelmus urozonus Dalman*, 1820 and *Stepanovia eurytomae* (Nees, 1834), respectively.

**Key words**: *Diplolepis fructuum*, parasitoid, new record, community members.

# Detection and identification of Wolbachia endosymbiont from Sitophilus granaries

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#### ABSTRACT

Arthropods are frequently infected with several micro-organisms, including symbiotic bacteria. Endosymbiotic bacteria belonging to the genus Wolbachia are maternally inherited and can cause various effects on the reproductive system of their invertebrate hosts. Due to their critical effects on insect reproduction, Wolbachia are potential tools for future insect pest control strategies. The diversification and popularization of molecular tools have led to the increasing use of molecular techniques to identify symbionts. Here, we explored reproductive symbionts in Sitophilus granarius populations. Polymerase chain reactions (PCRs) and sequencing of Cardinium, Arsenophonus, Spiroplasma and Wolbachia. S. granarius populations were not infected with Cardinium, Arsenophonus and Spiroplasma but infected with Wolbachia. In addition, we used Wolbachia multilocus sequence typing (MLST) to explore of Wolbachia subgroups. Detection and characterization of Wolbachia subgroups from this pest provide us with basic information for future pest control strategies using these endosymbiotic bacteria.

**Key words**: *Sitophilus granarius*, reproductive symbionts, *Wolbachia*, molecular techniques.

# Effect of non-ionizing radiation (UV) on prepupa and pupa stages of egg parasitoid *Trichogramma brassicae* Bezdenko (Hymenoptera: Trichogrammatidae)

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#### **ABSTRACT**

Determination of the effect of non-ionizing radiation (UV) on prepupa and pupa stages of egg parasitoid, Trichogramma brassicae Bezdenko (Hymenoptera: Trichogrammatidae) was aimed in this study. Experiments were conducted in rearing rooms constructed with 70±5% humidity, 27±1°C temperature and 16 hours light and 8 hours dark conditions. Ephestia kuehniella Zeller (Lepidoptera: Pyralidae) eggs were used as a host. The same number ( $50\pm 5$  eggs) and fresh host eggs (0-24 hours) were given to the *T. brassicae* newly emerged from the host eggs. For prepupa stage, four days after parasitization, parasitized eggs were exposed to the UV for 2, 4, 6, 8 and 10 minutes. For pupa stage, eight days after parazitization, parazitized eggs were exposed to the UV in the same way. The results were compared with a control group that there was no any application. The sensitivity to the UV was found different in both biological stages. The prepupa stage of parasitoid was more sensitive than pupa stage. There were not any differences in terms of getting dark colour of E. kuehniella eggs (development of parasitoid to the pupae stage) parasitized that exposed to the UV for both stages. Adult emergence decreased with increasing of the radiation duration. The same result was observed in both female and male emergence.

**Key words**: non-ionizing radiation, *Trichogramma brassicae*, *Ephestia kuehniella*, prepupa, pupa, adult emergence

Isolation and identification of some gram positive bacteria causing infections in silkworm (*Bombyx mori* L.)

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#### **ABSTRACT**

From the environment in a silkworm rearing room were taken 58 samples from air and various surfaces of shelves (wood), grids (metal) wall (wallpaper) and floor (linoleum). Thirty-six bacterial isolates were identified as Gram positive – spore-forming micro-organisms, staphylococci and streptococci. According biochemical identification of isolates by Micronaut-Scan were confirmed as *Paenibacillus macerans*, *Bacillus sphaericus*, *Bacillus circulans*, *Bacillus cereus*, *Staphylococcus aureus*, *Staphylococcus epidermidis* and *Streptococcus* spp. Susceptibility of some selected isolates to different drugs was determined. Resistance to important groups of antibacterials: penicillin, chloramphenicol, quinolones, macrolides, tetracyclines and sulfonamides were detected. Contamination of various surfaces with these bacteria is a potential risk of infection in silkworm.

Key words: silkworm (Bombyx mori L.), bacteria, drug resistance

# Aphid parasitoids (Hymenoptera, Aphidiinae) of the agro-ecosystems of the experimental fields in Plovdiv region

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#### ABSTRACT

Species from the family Aphidiidae (Hymenoptera) parasitize exclusively aphids and are used in biological control programs on protected crops in several countries. Development and use of parasitoids for biological control of aphids in Bulgaria is new area and may reduce the rate of increase of aphid populations.

Eleven Aphidiidae genera and 23 species from 197 aphidiids taxons were identified. Six species of them are new from Bulgarian fauna. It reported 32 plant – aphid – parasitoid associations. The studied and identified material is deposited in the collection of the author.

# The horseflies (Diptera: Tabanidae) of Rila Mountains, Bulgaria

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#### **ABSTRACT**

The study was carried out at 47 localities on the territory of the Rila Mountains, Bulgaria during the active seasons of tabanids in 2010 – 2015. Totally, 1401 tabanid specimens were collected and processed. A total of 34 species from 8 genera were identified: Silvius (1), Chrysops (3), Atylotus (2), Hybomitra (7), Tabanus (15), Haematopota (3), Dasyrhamphis (1) and Philipomyia (2).

The species *Hybomitra caucasica* (Enderlein, 1925), *Hybomitra muehlfeldi* (Brauer, 1880) and *Hybomitra solstitialis* (Meigen, 1820) are reported for the first time for the Bulgarian tabanid fauna.

At this study are not registered the species *Nemorius vitripennis* (Meigen, 1820), *Hybomitra aterrima* (Meigen, 1820) *Hybomitra montana* (Meigen, 1820), *Haematopota grandis* Meigen, 1820 and *Haematopota italica* Meigen, 1804, known for the Rila Mountains tabanid fauna from literature sources. Thus, based on the summary of the literature data and the results of this study have been established 39 species in the Rila Mountains tabanid fauna. The horsefly fauna of the studied region is predominated by elements of the Boreal-Eurasian faunal type (61.54% of the species).

# Developmental time and mating success in *Drosophila melanogaster* strains maintained on apple and carrot substrates

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#### **ABSTRACT**

The food directly affects development and reproduction rate of animals. It provides energy sources necessary for different biological functions, starting from those involved in developmental processes up to those included in growth and reproduction.

We have established, in laboratory conditions, several *Drosophila melanogaster* strains which were fed on different substrates from 2000. In this work, we used two of them, one containing apple and one containing carrot. Flies of these two strains were scored for developmental time and subsequently tested in series of mating experiments. Developmental time was calculated as an average time weighed by the total number of eclosed adults, while mating success was estimated as a number of copulations achieved during 1 hour observing period.

Flies maintained on carrot substrate developed significantly faster than flies developed on apple substrate. In upcoming mating assays, males maintained on carrot substrate were more successful in achieving copulations than males developed on apple substrate, when they were offered to females of their own strain. When chosen by females developed on apple substrate, males from both strains were equally successful in mating.

# Does diet affect mating preference and success in *Drosophila* melanogaster via changes in wing size and shape?

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#### **ABSTRACT**

Mating behavior in *Drosophila melanogaster* is very complex. It starts with a male courtship ritual, and is followed by copulation if female is receptive and willing to accept the courting male. Variability in traits important for intra- and intersexual selection in this species is influenced by both genetic background and environmental factors.

Effect of nutrition, as an important environmental factor, on *Drosophila* mating behavior may be observed, among other things, over its influence on body size, which is usually approximated using wing size. Otherwise, *Drosophila* adult size is entirely regulated by growth during the larval stages.

The goal of this research was to determine whether two *D. melanogaster* strains reared about 14 years on substrates containing tomato ("tomato strain") and banana ("banana strain") differ in mating preferences and mating success. In addition, we were interested if behavioral differences were influenced by nutritional effects on wing size and shape. Namely, wings provide important visual and acoustic stimuli in *Drosophila* courtship behavior.

Mate choice and mating success were monitored throughout the series of multiple choice tests. Results pointed that there were no differences in mating preferences and mating success between these two strains, despite significant differences in wing size and shape. Although different diets contributed to modification of wing parameters, they did not influence behavioral traits under investigation.

# Different effects of a strong static magnetic field (2.4 T) on *Drosophila melanogaster* wing size and shape

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#### **ABSTRACT**

The geomagnetic field has influence on all living organisms. Static magnetic fields affect evolution pattern of red blood cells, blood flow, orientation of macromolecules, oxygen dissolution in tissues, and embryonic development. Growth and differentiation of *Drosophila* are also susceptible to the influence of static magnetic fields. Changes in duration and stability of development can be estimated through variations in wing size and shape.

The aim of this study was to determine whether static magnetic field of 2.4 T can induce changes in wing size and shape of *Drosophila melanogaster*.

The first instar larvae were placed near the north (N) and the south (S) pole of the magnet (2.4 T, VINCY Cyclotron magnet) for 2 hours. The effect of a strong static magnetic field on *Drosophila melanogaster* was monitored through analysis of wing size and shape, which was performed by using a method of geometric morphometry. Obtained results revealed differences in wing size only between sexes. On the other hand, differences in wing shape were found between sexes and treatments. Detailed analysis further proved that only treatment has influence on wing shape, while differences between sexes are caused by differences in centroid size. In that way, strong static magnetic field affected wing shape, but did not change wing size in this species.

Habitat and sex-specific responses of *Drosophila subobscura* exposed to static magnetic field (2.4 T) revealed by analysis of morphological traits

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#### **ABSTRACT**

At the present time, with increased technological development, we are confronted with a growing influence of various forms of magnetic fields that can affect living organisms. Biological effects represent measurable responses to a change in the environment that are not necessarily harmful. Adult phenotype is determined by an individual's genotype and environmental influences, and can be analyzed using measurable morphological traits.

The main goal of this research was to consider how a strong static magnetic field (2.4 T) affects wing size and shape of males and females from two *Drosophila subobscura* populations (collected in beech and oak forest), after placing the first instar larvae near the north (N) and the south (S) pole of the magnet (VINCY Cyclotron magnet) for 2 hours.

Applied static magnetic field (2.4 T) induced habitat – and sex-specific response in *Drosophila subobscura*. Results revealed presence of sexual dimorphism in both wing size and shape, in magnetic field and control groups of two *Drosophila subobscura* populations. Significant differences are found for both wing size and shape only within beech population. Comparing morphological traits between "beech" and "oak" populations, wing shape was significantly different in all treatments for both sexes, while wing size considerably differed only between females exposed to the South Pole.

Mosquito (Culicidae, Diptera) and vector monitoring in Danube Delta most representative ecosystems 2014 – 2015 (România)

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#### ABSTRACT

Climate variability has a direct influence on the epidemiology of mosquito-borne viruses which should increase the frequency of a series of exotic diseases transmitted by viruses hosted in some highly mobile animals like migratory birds. A large number of Culicidae species feeding on these birds are known as potential transmitters of important diseases to humans like Dengue fever, West Nile or Japanese encephalitis. Some parts of Europe are of particular interest by accumulating a large number of wet habitats with a diverse bird fauna, but also a diverse, yet underestimated Culicidae diversity, and the Danube

Delta is one of the most important of them. Our research aims was to monitoring mosquito species and vector species, focusing on the Danube Delta, as a large and important migration place for a large number of birds. Used standard collection methods CO2-baited EVS traps. conducted between 2014 and 2015, from April to September, analyzing mosquito community's composition. A total number of 535 768 mosquito specimens were collected in the four ecosystems (aquatic, marsh, forest and urban ecosystems). All mosquitoes were identified using morphological characteristics and molecular character mitochondrial COI (658 bp) gene. Our integrative approach has the result a number of 17 different species, we found 13 vector species which 7 species were the most common species with highest number of specimens: Cq. richiardii (~334 999), An. hyrcanus (~106 872), An. maculipennis (~25 734), Cx. pipiens (~20 654), Cx. modestus (~11 187), Oc. caspius (~17 873), Ae. vexans (~8265). These common species are also potentially the most important vector-borne species for number of viruses, like West Nile Virus, Batai-, Tahyna-, and Usutu Viruses. The most height number of specimens was collected in aquatic ecosystem (311084 specimen), then marsh ecosystem (102 437), urban ecosystem (70056) and forest ecosystem (51 441). Finally, this information can also help to implement vector control programs depending on the life cycle of some mosquito species and to adjust the timing of interventions. This research was supported by a grant of the Ministry of National Education, CNCS- UEFISCDI, project nr. PN-II-ID-2012-4-0595, furthermore, this work was financially supported by the German Federal Ministry of Food and Agriculture (BMEL) through the Federal Office for Agriculture and Food (BLE) with the grant nr. 2819104315 and STSM program by EurNegVec.

Morpho-physiological profile and changes in blood parameters of *Carassius gibelio* (Pisces: Cyprinidae) as response to the anthropogenic pollution in two water basins in the region of Galabovo, Southern Bulgaria

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#### **ABSTRACT**

Basic morphological, physiological and hematological parameters were studied for adult, sexually mature individuals of *Carrasius gibelio*, living in three water basins in southern Bulgaria. Two of them are situated in the region of the town of Galabovo: the river Sazliyka, near the sluice gates, where water is diverted to TPP "Brikell" (site A) and the Rozov Kladenets reservoir (site B). The third site – the river Vacha (site C) is less disrupted (conventional control). For *C. gibelio* specimens, inhabiting site A, the lowest values of parameters: standard length, body weight and condition factor, as well as high values of spleen-somatic index and ren-somatic index were found. The hematological profiles of specimens of *C. gibelio* from site A showed erythrocytosis, leukocytosis and hyperchromia, and differential blood count – neutrophilia, eosinophilia, monocytosis and lymphocytopenia. These

changes indicate a great stress that is caused by the anthropogenic pollution of domestic-sewage typeand by the industrial waste water of the nearby TPP "Brikell".

**Key words**: Prussian carp, morphological parameters, hematological parameters, anthropogenic pollution, bio-indication.

# Effects of Ni and Pb on the respiration rate and histological structure of common carp (*Cyprinus carpio*, L.) gills

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#### **ABSTRACT**

This work aimed to study the effect of heavy metal exposure on the histological structure of common carp (Cyprinus carpio, L.) gills. The fish were treated with different soluble concentrations of Ni and Pb in laboratory conditions for a total period of 72 h. These metals are considered as priority substances in surface waters according to Directive 2008/105/EO of the European parliament and the Council. The metal concentrations were prepared as 100, 75, 50 and 25% of the maximum permissible levels set by law. The results showed a higher index of respiration rate in all test tanks for both metals, compared to the control in the beginning of the experiment, but there was no pattern of increase or decrease in relation to the metal concentrations. After 72 hour of exposure we observed the same pattern, but in addition the respiration rate of the fish in the tanks treated with Pb showed an increase in a dose-dependent manner. We also observed different histological changes in the gill epithelium, which included proliferative and degenerative changes, as well as changes in the circulatory system. We found severe proliferative changes such as lamellar lifting, edema, proliferation of the glandular cells and epithelium, proliferation of the epithelium of the secondary lamellae, proliferation of the cartilage tissue and fusion of the secondary lamellae. These changes activated compensatory-adaptive mechanisms in order to prevent the entry of the metals through the gill surface. In addition, the degenerative changes were more pronounced in fish, treated with Pb concentrations, and the blood circulatory system showed mainly vasodilatation, which caused pathological changes in the gills. In sum, we can conclude that Ni and Pb have severe effects on the respiration rate and gills histology of the common carp, even at concentrations, which were lower than the allowable ones.

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## Fulton condition coefficient of 11 fish species from great natural and two artificial lakes in the Republic of Macedonia

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#### **ABSTRACT**

This study provides data on the Fulton condition coefficient (FCF) of 11 fish species from great natural (Prespa, Ohrid and Dojran) and two artificial lakes (Debar and Tikveš) in the Republic of Macedonia. In addition, this paper provides the first comprehensive data of the FCF for six endemic freshwater fish from the above mentioned lakes. Samples were collected from October 2008 through to August 2014. Average values of FCF are ranged from 0.862 to 1.394. The FCF and total length relationships for all examined species have been determined. The results of this study show that the FCF depends on the species, period of catch, age of the individuals and their sex. Also, the results show that FCF is different for each ecosystem and depends on many parameters. It should be noted that the differences arise due to the various physical and chemical parameters and environmental conditions that are different in each lake, then trophic state of lake, altitude, rich with fish food and more. The development of a management plan and a regular monitoring for good understanding of the biology of this species, especially endemic, is recommended.

**Key words**: fisheries, trophic state, Lake Prespa, Lake Ohrid, Lake Dojran, Lake Tikveš, Lake Debar

# Histopathology of kidney of *Salmothimus ohridanus* (Pisces, Salmonidae) caged in Lake Ohrid

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#### **ABSTRACT**

Histopathological changes have been widely used as biomarkers in the evaluation of the health of fish exposed to contaminants, both in the laboratory and field studies. One of the great advantages of using histopathological biomarkers in environmental monitoring is that this category of biomarkers allows examining specific target organs, including kidney, that are responsible for vital functions, such as excretion of xenobiotics in the fish. The alterations found in these organs are normally easier to identify than functional one, and serve as warning signs of damage to animal health.

In kidney of fishes caged in Lake Ohrid histopathological lesions were detected like: enlargement of the glomerulus, occlusion of the tubular lumen, etc.

**Key words**: histopathological biomarkers, kidney, Lake Ohrid.

# Macrophyte vegetation as spawning ground of some cyprinid fish from Lake Ohrid

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#### ABSTRACT

This paper is result of long-term researches of macrophyte vegetation and cyprinid fish from Lake Ohrid performed by the Department of Hydrobotany and Department of Cyprinid fauna from Hydrobiological Institute – Ohrid, R. Macedonia.

The researches were performed in 10 localities from eastern coastline of Lake Ohrid: Ajvan plaza, Park, Inex, St. Stephan, Granit, Metropol, Lagadin, Eleshec, Krusha, and Peshtani.

The aim of this paper was to research the spawning ground of the Ohrid cyprinid fish. Those are: chub, moranec, roach, carp, barbel, gudgeon, undermouth, bleak and others. The different cyprinid fishes are spawning in particular month in the spring-summer period, in different regions of Lake Ohrid and on different substrates.

The obtained results show that in the researched localities along eastern shoreline of Lake Ohrid were evidenced different macrophyte species. Their number varied from 4 (locality Inex) to 14 macrophyte species (locality Park). Also, it was evidenced that the beginning of cyprinid fish spawning is at the same time with the development of macrophyte vegetation.

It should be mentioned that in recent years macrophyte vegetation from Lake Ohrid is destroyed for building of hotels and beaches. This leads to changes in the qualitative and quantitative composition of cyprinid fish and their spawning grounds. Therefore, in the future it is necessary to take measures to protect macrophyte vegetation from Lake Ohrid from the negative anthropogenic impacts.

**Key words**: macrophyte vegetation, eastern coastline, spawning ground, Lake Ohrid

# Seasonal changes in the red blood cell parameters in Ohrid roach blood from Ohrid Lake

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#### **ABSTRACT**

The blood investigation of Ohrid roach, *Rutilus ohridanus* Karaman 1924, from Ohrid Lake, Republic of Macedonia, collected in two different seasons (late November/early December 2015 – late May 2016), in the litoral area at Vel Dab showed no difference in the red blood cell count, but significant difference in every other parameter. The hemoglobin concentration (p < 0,0001), hematocrit (p < 0,001), mch (p < 0,0001), mch (p < 0,001), mcv (p < 0,05) all have lower values in the May probe. This implies that the addaptible mechanisms of the blood physiology, throughout different seasonal periods, along with the different stressors in the environment, affects the hemoglobin concentrations and the volume of the red blood cells, and not their number.

Correlation between the choice of partner and the individual nesting territory in the lesser kestrel (*Falco naumanni*) and preconditions for polyandry

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#### ABSTRACT

In the period 2015 - 2016 within a project for recovering the population of the lesser kestrel (Falco naumanni) in Bulgaria were conducted daily observations and closely monitoring the nesting process in the newly formed colony. The nesting territory of the colony is centered around a releasing on which the artificial feeding of released birds was taking place. From the observations during the breeding season of 2016 was established a correlation between the ratio of the male and female birds returned from migration, the choice of a partner and the occupation of the individual nesting territory. This dependence was established in a situation of shortage of breeding female birds and excess of male birds, in which the gender roles in choosing the nest and partner were clearly visible. In 2015, the sex ratio is more aligned, which contributed to the rapid formation of couples and occupation of the territory. In 2016 due to a significant shortage of female birds and the gradual filling of the colony mainly from returning adult males, we had the opportunity to observe in detail the formation of the breeding couples. On the day of the appearance of a new returned female, she has a choice between several free male birds with already established nesting territories. She forms a short-term pairs with them until she finds the one whose territory is the most favorable. This indicates a priority of the nest site in front of the breeding partner. The ratio of sexes in 2016 created a prerequisite for polyandry, which further enhance the above view.

Lesser kestrel (*Falco naumanni*) in Thrace – distribution, numbers, and threats

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#### **ABSTRACT**

There are a lot of detailed data for the distribution and numbers of the lesser kestrel (Falco naumanni) from Spain, Italy, and Greece (southern part), where are the largest colonies of this species in Europe. There was no data, until now, for the area under this study, constituting of territories in South-eastern Bulgaria (there the species was restored as nesting in 2014), the European part of Turkey, and Northeastern Greece. At the same time the Thracian (South-eastern Balkan) population of the species is a veritable bridge between the numerous populations of the species in South Europe (Portugal, Spain, France, Italy, Greece), and Asia (Turkey, Israel, Kazakhstan, etc.). There is data for more than 15 colonies in Thrace, gathered by us in the target territories – 5 in North-eastern Greece, at least 10 in the European part of Turkey, and 2 colonies in South-eastern Bulgaria. The number of the colonies surveyed is from 2 to 40 nesting pairs, situated, predominantly, in residential housing, rural houses and mosques. 3 of the colonies use natural habitats for nesting, such as seaside cliffs (two colonies), and rock formation in mainland European Turkey (1 colony). The main threats, registered, are the destruction of nesting habitats by roof and building renovations. Potential threats that need additional research are agricultural intensifying, pastures conversion into arable land, change in the crops and electrocution. The main part of the research is carried out in the scope of projects under the European Union's LIFE Programme, implemented by Green Balkans and BSPB.

**Key words**: lesser kestrel, *Falco naumanni*, distribution, numbers, Thrace, Balkan Peninsula

First telemetric study of the individual territories, seasonal movements, and habitat use of the Balkanic capercaillie (*Tetrao urogallus rudolfi*)

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#### ABSTRACT

The capercaillie is a polygamous forest-dwelling bird with large spatial requirements, but is highly selective to the habitat characteristics. *Tetrao urogallus rudolfi* is a glacial relict, endemic subspecies with two isolated metapopulations – mainly in the Rilo-Rhodopes mountains and the southeastern Carpathian mountains. The species is included in Annex I of the Birds Directive and has been identified as Threatened in Bulgaria. For the first time in Bulgaria, in 2014 – 2015 we attached GPS transmitters on three adult males, associated with one lek, in a coniferous forest in Rila Mountain. The objectives were to estimate home range, seasonal movements and habitat use. We used backpack GPS from the company e-obs GmbH (Germany), weighing 82 g, with a storage capacity of 25 000 positions.

Individual territories were identified on a yearly and seasonal basis using the Minimum Convex Polygon estimation and the Kernel index. The sizes of the yearly individual territories of the three marked males, based on the MCP estimation were 1139.5, 739.0 and 276.5 ha. The differences are based predominantly on considerable differences in the degree of fragmentation of the habitats of the three birds. During the spring season, when birds have the smallest individual territories, their diel activity is highest. The birds actively moving, using a high amount of energy within a small perimeter, this makes them highly susceptible to disturbance during the breeding period. The longest displacement for 24 h is 4.1 km during the winter season, and the shortest distance is 6.5 m during the fall season.

# Characteristics of the territorial and hunting behavior of the red-footed falcon (*Falco vespertinus*) from South-Eastern Bulgaria

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#### **ABSTRACT**

The present study is focused on different types of hunting strategies of the red-footed falcon ( $Falco\ vespertinus$ ), dependent from the preferred prey items, on the length of hovering, number of series of hovering and differences in the hunting success in according to the sex of the studied birds, the period of reproduction and the weather conditions. Seven major types of hunting behavior of the species were separated which can shift quickly from one to another which is mostly connected with changes of the available prey and the speed and direction of the wind. Differences were found also in the average length of the hovering series and the number of these series between migrating and local breeding red-footed falcons and also between males and females. The total percent of completed attacks after hovering was 27.7; in females -25%, in males -33.3%. The percent of the successful attacks was 68.6%, respectively for the females -66.7%, and for the males -71.4%.

Territorial behavior of the red-footed falcons was studied separately for intraspecific and the interspecific conflicts – the last mostly attacks of territorial birds against other species of falcons breeding together with them in mixed colonies –such as kestrels (*Falco tinnunculus*) and hobbies (*Falco subbuteo*). Territorial conflicts were registered also with other bird species as lesser grey shrike (*Lanius minor*), booted eagle (*Aquila pennata*), long-legged buzzard (*Buteo rufinus*), corvids etc. Red-footed falcons found in the mixed colonies in SE-Bulgaria

were definitely territorial and highly aggressive birds. In the near past the red-footed falcons nested also in monospecific colonies in Bulgaria (mostly in NE Bulgaria) and such aggressive behavior was not reported. Agressive behavior showed by the pairs (often only by males) was connected with the distance between neighbouring nests. In colonies with distance between the nests greater than 50 - 60 m. intraspecific aggression was not observed. In a mixed colony where the all three mentioned species of falcons nested in artificial nest boxes the cases of aggressive behavior between different pairs were common and this lead to permanent disturbance, later start of the nesting, change of nest positions and even loss of chicks. In the same time it was found that red - footed falcons and kestrels despite the attacks between them sometimes can defend themselves cooperatively against a larger enemy. The obtained data have great practical importance for the conservation of the red-footed falcon which is a species endangered with extinction as a breeder in Bulgaria. Furthermore these data can be used as a basis of other ethological studies of that species.

## Organochloride pesticides in feathers of Pygoscelidae penguins from Livingston island, Western Antarctica

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#### ABSTRACT

The aim of the present work is to determine the presence of isomers αHCH, βHCH, γHCH, 4.4'DDE, 4.4'DDD and 4.4'DDT that compound the commercial forms of insecticides Lindan and DDT in molting feathers of Pigoscelis antarctica and Pigoscelis papua from Livingston and Peterman Islands, Western Antarctica.

The analyses of the total quantity of the chosen chlorinated pesticides shows significant differences between DDT and HCH concentrations where quantities of DDT were about six times more than this for HCH. In contrast there were not established any differences neither between investigated localities nor among penguin species despite the certain differences in the food spectrum of both of them. The concentrations of  $\Sigma$ HCH were as follows: for *P. antarctica* from Livingston island  $-0.419 \mu g/g$ ; for *P. papua* from Livingston island  $-0.356 \mu g/g$ and for P. papua from Peterman island – 0.437 µg/g. The corresponding values for ΣDDT were: for P. antarctica from Livingston island – 2.596  $\mu$ g/g; for *P. papua* from Livingston island – 2.507  $\mu$ g/g and for P. papua from Peterman island – 2.628 µg/g.

Percentage composition of the HCHs compounds shows that the value of  $\beta$ -HCH dominates in all investigated samples. It is between 55 and 70% of the total quantity of the analyzed HCHs. The major component DDT isomer composes about 2/3 from the total sum or between 72% in the feathers of *P. papua* from Livingston Island and 87% for *P. papua* from Peterman Island. No statistically significant differences were found when compared investigated species, nor between two localities.

# Bat fauna (Mammalia: Chiroptera) from Sakar-Mountain (South-Eastern Bulgaria)

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#### **ABSTRACT**

The data about bat fauna in the region of Sakar-Mountain are insufficient till now. In this study information based on the visits of appropriate shelters and analyses of ultrasounds of bats is presented. The study covered the period of early spring (April), summer (August) and autumn (October) 2011, which practically matched the spring, breeding period and autumn migration of the bats.

A total of 20 bat species are known till now from which 6 species are newly recorded. The habitat preferences, territorial distribution and relative activity are presented and discussed.

Population epigenetic diversity versus subspecies detachment of the forest dormouse (*Dryomys nitedula*) in a long distance transect in Eurasia: implication for its conservation

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#### **ABSTRACT**

Geographic variation and intraspecific taxonomy of forest dormouse (Dryomys nitedula) has not yet been investigated adequately, and its population epigenetic uniqueness and the status of many subspecies require clarification. To advance knowledge on the interspecies variety of forest dormouse, the epigenetic cranial polymorphism was studied in eight forest dormouse populations, located along a transect line from Central Anatolia region to Central Southeast Europe and inhabiting an area of suspected different subspecies taxa. The mapping of the population epigenetic diversity of forest dormouse within the area of the classically described subspecies manifested some overlaps of their epigenetic distances with the geographic remoteness between them. The revealed pattern of population epigenetic diversity in the territories of the classically described subspecies taxa of the forest dormouse expands the knowledge of the species diversity in its areal. It highlights the need to conduct future large-scale study of its population differentiation and uniqueness of several subspecies. A better understanding of biodiversity of forest dormouse in Palearctic scale would provide an important tool for the development of management plans aimed at preserving its biodiversity and population genetic differentiation.

⇒ This study was partly supported by bilateral project between Bulgarian academy of sciences, Bulgaria and TÜBİTAK, Turkey (113Z822).

# Indication for genetic diversity of European roe deer (*Capreolus capreolus*) in Southeastern Europe revealed by mtDNA markers

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#### ABSTRACT

A molecular-genetic analysis of a control region (936 np) and cytochrome b gene (1140 np) of the mtDNA roe deer from Pannonian mixed forests ecoregion in Central south-eastern Europe (Hungary) and roe deer of Rilo-Rhodope biogeographical region in Balkan Peninsula (Bulgaria) has been carried out. It was found that 97.02% of the samples from Pannonian mixed forests ecoregion had a mitotype typical for European roe deer (Capreolus capreolus). In the roe deer population inhabiting this region in the 2.98% of them possessed a typical for Siberian roe deer (C. pygargus) mitotype. In the same time, the variation pattern of the studied mtDNA fragments in roe dear from this biogeographicall region was in general similar to the overall distribution of the mitochondrial lines of the European roe deer. Both of the widely and chaotically spread in Europe, haplotypes were found; neither the unique South-Spanish and South Italian, nor Crimean and Krasnodar (Russian) sequences were found. The roe dear inhabiting in Rilo-Rhodope biogeographical region in Balkan Peninsula also had typical for the European roe deer mtDNA sequences, but they were represented by one haplogroup only. Because of the assessment of biochemical-genetic uniqueness of the investigated roe dear was postulated that its populations in Pannonian mixed forests ecoregion Rilo-Rhodope biogeographical region represent a distinct gene pools. This conclusion formed the basis of incurred recommendation — no admixed its populations in both studied European bio-geographical region by introductions of roe deer from abroad.

## Influence of tourists on the summer bat colonies in the Devetashka cave

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#### **ABSTRACT**

Microchiropteran bats, in particular cave dwellers, are adversely affected by human activity. The microbats are especially sensitive when they assemble in maternity colonies to raise their young, as they require a narrow range of microclimates and no disturbance. In this connection, we assessed the state of the bat colonies in relation to tourist abundance and behavior assessed the abundance, mortality and movement of the bat colonies in the Devetashka cave, Bulgaria. The cave is of European importance for bat conservation and houses more than 10 000 bats in the summer. However, during the weekends it is subject to an enormous tourist flow -500 - 1000 people per day. We assessed the state of the bat colonies in relation to tourist abundance and behavior. In order to estimate the tourist factor, the number of people, whether flashlights and photo flashes were used, the maximum noise produced and how deep tourists go into the cave galleries were recorded during observation sessions. The number of colonies. the number of bats within a colony, whether the colonies have moved/disappeared and the mortality rate of juveniles and adults were recorded in order to track the condition of the bat colonies. We discovered that often tourists go beyond the prohibitive barriers on each of the two galleries and that the most prolific disturbance they produced was photo flashes. Strong bat activity and movement was detected and a high number of juvenile mortality. The extent of tourist influence and the necessary measures to reduce it are discussed. Probably cave tours will need a special consideration so that they can be designed to minimize as much as possible the detrimental effect human activity has on bats.

# Copulatory behavior of *Lasiopodomys* (*Stenocranius*) gregalis (Rodentia, Mammalia)

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#### ABSTRACT

Narrow-headed vole *Lasiopodomys* (*Stenocranius*) *gregalis* was previously assigned as a species from the subgenus *Stenocranius*, genus *Microtus – Microtus gregalis* Pallas, 1779. However, molecular genetic analyses showed the closeness of this species to the genus *Lasiopodomys* s. str., so now its taxonomic status was defined as *Lasiopodomys* (*Stenocranius*) *gregalis*.

The purpose of the study was to establish how many markers of mating behavior of narrow-headed vole were consistent to the results of molecular genetic investigations. Analysis of sexual behavior parameters of narrow-headed vole compared with the parameters of some other species from *Lasiopodomys*, *Microtus* and *Alexandromys* genera was done.

The results showed the thrust stereotype of copulation, which includes separate intromissions and a very first mount with intromission, which ending with ejaculation. The number of ejaculations was small – from 3 to 5 and the genital stimulation rate averages 100 - 106 thrusts. During the courtship the singing and waltzing were moderately pronounced. Marking of the territory was realised by the application of the secretion of a specific flank skin glands.

The dendrogram of similarity of sexual behavior demonstrates that *L. gregalis* is close to the species of the genus *Lasiopodomys*. Clear difference between sexual behavior of narrow-headed vole in comparison with the species of the genus *Microtus* (*Microtus* s. str. and *Sumeriomys*) was noted. The level of inter-population differences in narrow-headed vole was much more and exceeds these of many other species.

Key words: Lasiopodomys gregalis, mating behavior, scent marking

Karyotypic and craniometric characteristic of harvest mouse (*Micromys minutus* Pallas, 1771) (Mammalia: Rodentia) from South Bulgaria

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#### **ABSTRACT**

The study presents the first cytogenetical and morphometrical investigations on harvest mouse (Micromys minutus Pallas, 1771) in Bulgaria. Population of harvest mouse from the rise fields near Plovdiv, Bulgarian part of the Thracian valley, was examined. Five individuals (3 males and 2 females) were cytogenetically analyzed. The established diploid chromosome number was 2n = 68. Autosomes consisted of 1 pair of large and 1pair of small metacentrics, 2 pairs of middle sized submetacentrics, 28 pairs of subtelocentrics and 1 pair of acrocentric chromosomes. The X chromosome was the largest subtelocetric and the Y chromosome was small acrocentric. The FN of the chromosomes was estimated to be 133, as the NFa was 130. All chromosomes except the largest metacentric pair had well expressed heterochromatin blocks in the centromeric regions, which also extended into the whole short arms in the most biarmed chromosomes. Block of centromeric heterochromatin was also found in the X chromosome, while Y chromosome appeared to be entirely heterochromatic. Four somatometrical and twelve craniometrical features were analyzed.

Key words: Micromys minutus, karyotype, C-banding, craniometrics

Autumn-winter diet and food niche overlap between red fox (*Vulpes vulpes* L. 1758) and golden jackal (*Canis aureus* L. 1758) in Bulgaria

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#### **ABSTRACT**

The main goal of this study was to identify the diet and the food niche overlap of the red fox (*Vulpes vulpes*) and the golden jackal (*Canis aureus*) in the areas with the greatest number in Bulgaria – Upper Thracian Plain and Northeastern Dobrudza. The stomach contents of 234 foxes and 256 jackals collected during the period 2008 – 2011 were analyzed.

The variety of food items in both species was almost equally. Twenty food components were found in the stomach content of the red fox and twenty-two in the stomach content of the golden jackal. This indicates that both species were not opportunists but facultative specialists in food habits. Small mammals, mainly rodents, were dominant in the diet of the both species. The herbaceous plants and poultry played a secondary role in the diet of red fox, while in the diet of jackal the additional food components were spread evenly. The distribution of food components in the diet of both predators was roughly equal in the two regions.

Significant differences were identified about food niches breadth in both species, as a wider was the niche breadth of the jackal. The level of overlap of the food niches was high in the two investigated regions.

Key words: Vulpes vulpes, Canis aureus, food habit, food niche overlap

In vivo genotoxicity and cytotoxicity assessment of permissible concentrations of Ni and Pb based on comet assay and nuclear abnormalities in acridine orange stained erythrocytes of common carp (Cyprinus carpio L.)

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#### **ABSTRACT**

The aim of the present study is the detection of a possible in vivo genotoxicity of heavy metal concentrations, considered safe by regulatory agencies. Nickel and lead are referred to as priority substances in surface waters according to Directive 2008/105/EO. In order to evaluate their genotoxic and cytotoxic potential, young specimens of common carp (Cyprinus carpio L.) have been exposed to 100, 75, 50 and 25% of the maximum permissible Ni and Pb concentrations (MPC) for 7h in laboratory conditions (MPC values are set by the Bulgarian and EU laws). The alkaline comet assay in circulating erythrocytes was applied and blood smears stained with acridine orange were observed for the presence of micronuclei and other nuclear abnormalities (NAs). The comet parameters % tail DNA (%TDNA) and tail moment (TM) indicated a statistically significant genotoxic damage for both metals at 100, 75 and 50% of the MPC compared to controls. The most intensive migration of DNA for Ni (%TDNA = 26.72±4.84;  $TM = 2.43\pm1.09$ ) and Pb (%TDNA = 26.37±8.12;  $TM = 3.35\pm1.60$ ) was observed at 100% MPC with no statistical difference between the metals. The lowest values were registered at 25% and the lack of statistical differences with controls showed the harmlessness of this concentration. Increased levels of micronuclei were not observed in carp erythrocytes, but we found out significant increase of other nuclear abnormalities – notched nuclei (NN), blebbed nuclei (BN), lobed nuclei (LN), eightshaped nuclei (EB) and nuclear buds (NB). The 72 h exposure time of the investigated concentrations was not effective for the formation of micronuclei, but demonstrated evident cytotoxic effect and confirmed NAs as effective biomarker. In general, the metal exposed groups showed significant variation in the frequency of nuclear abnormalities as well as an extent of a DNA damage compared to the control group. These frequencies increased significantly (p < 0.05) in a concentration dependent manner. The obtained results proved the geno- and cytotoxic effects of Ni and Pb, even at low permissible levels and illustrated the need of additional investigations in order to reduce MPC of heavy metals in water basins.

**Key words:** genotoxicity, cytotoxicity, Ni, Pb, *Cyprinus carpio*, comet assay, nuclear abnormalities

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Determination of specific IgG avidity and anti-Toxocara IgA antibodies in patients with toxocarosis and their application to specify the stage of disease

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#### ABSTRACT

Toxocarosis in humans is a parasitic zoonosis caused by migration of the larvae of animal nematodes -T. canis and T. cati. The clinical symptoms of the disease are nonspecific and highly variable, which complicates the diagnosis. Currently it is based on serological methods - ELISA and Western blot to determine the specific anti-Toxocara IgG antibodies in serum samples from suspected patients. Persistence of these antibodies for long period doesn't allow determining stage of the disease and evaluating the treatment. Therefore, in recent years are developed methods which made differentiation between acute and chronic infection. One of these is the determination of the avidity of specific IgG antibodies. Also it is examined the role of specific IgA antibody as a marker for fresh infection. Data of the use of these two methods in toxocarosis are insufficient. The aim of our study is testing samples from patients with a positive result in routine tests for the presence of specific IgG antibodies in ELISA, and to demonstrate the anti-Toxocara IgA antibodies, and developed by us, test to determine the avidity of IgG. 130 samples were examined, and the results indicate a low avidity (40%) at 7.3% of tested and a higher proportion of children and adolescents. Presence of anti-Toxocara IgA antibodies were found in 26.2% of those with toxocarosis, but positive result prevalent in people over 18 years. Comparing data from two tests showed that patients with low avidity have highest index of positivity of specific IgA antibodies. Inclusion in the diagnostic practice of these two new serological method will assist in the diagnosis of toxocarosis for determining the stage of infection and the need for treatment.

# Phytoplankton and chlorophyll a concentration in Lake Prespa during 2014

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#### ABSTRACT

Lake Prespa located in southeastern part of Europe between Macedonia, Greece and Albania is among the few ancient, long-lived of the world. Phytoplankton is the most sensitive indicator of the water trophic state that responds first to changes in nutrient concentrations. Chlorophyll a is a photosynthetic pigment that serves as a measurable parameter for all phytoplankton production and it is on average, 1.5% of algal organic matter. Phytoplankton samples from the pelagic and littoral zone of Lake Prespa were taken during eleven sampling campaigns in 2014 from April to December. Sampling and analyses were done according to the protocols for phytoplankton. The total abundance of phytoplankton was considerably higher in summer than in other seasons which is characteristic for meso-eutrophic lakes. Most phytoplankton species identified belong to the Bacillariophyta and Cyanophyta. Investigations showed distinct seasonal differences in phytoplankton composition. Similar to total phytoplankton abundance chlorophyll a concentrations at surface level were generally higher in summer than in other seasons. In the deeper layers, from 15 to 30 m, results were reverse. This kind of seasonal chlorophyll a distribution in the pelagic zone of Lake Prespa corresponds with the typical distribution of chlorophyll a in mesotrophic lakes from temperate zones. The overall composition of phytoplankton communities at Lake Prespa is typical for lakes in the process of eutrophication. According to the trophic state index (TSI), all sampling points at most times of the year indicated eutrophic I conditions.

**Key words**: Lake Prespa, phytoplankton, chlorophyll a, trophic state, pelagic zone, littoral

## The water quality of the tributaries of the Lake Ohrid

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#### **ABSTRACT**

The aim of this paper is to determine the water quality of the rivers Sateska, Koselska, Velgoshka and Cerava, representing major surface inflows in the Lake Ohrid from the Macedonian side. For this purpose, water samples were collected from six points for the determination of concentration of dissolved oxygen, biodegradable organic matter through KMnO<sub>4</sub> consumption, concentration of total nitrogen and concentration of total phosphorus. Out of the microbiological parameters there have been investigated the following: heterotrophic bacteria and total number of coliform bacteria with seasonal dynamics. The results of a survey of physicochemical and microbiological parameters indicate increased anthropogenic impact on the sampled water from the rivers. This situation is especially evident in the water samples collected from rivers Velgoshka and Koselska, where during the research period increased organic and nutrient load is registered. The water of rivers Cerava and Velgoshka is the most polluted by fecal and organic contaminants. The water quality of the tributaries of the lake varies from second to fourth category based on the Regulation on classification of waters of the Republic of Macedonia and is highly dependent on the season of collection of the samples.

Key words: rivers, anthropogenic, water quality, nutrients, bacteria

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## The water quality of the river Sateska from microbiological and chemical aspects

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#### **ABSTRACT**

Environmental problems in rivers, lakes and coastal waters are often a result of human pollution of nutrients or toxic substances. Since the nature is very complex it is hard to understand and calculate how these substances will spread in the ecosystem and which effect they will have. The purpose of these investigations was to establish the River Sateska water quality from microbiological and chemical aspects. Water samples were collected seasonally from spring 2013 to winter 2014, at three different localities (River Sateska before redirection, middle course and R. Sateska inlet). The following microbiological indicators for water quality were analysed: Total Coliforms, Total faecal coliforms, Enterococci, Escherichia coli, Aeromonas and heterotrophic bacteria. The chemical analyses done were: dissolved oxygen, dissolved biodegradable organic matter as permanganate consumption, total nitrogen, and total phosphorus. For all analyses were used standard limnological methods. All three sampling stations at River Sateska (before redirection, middle course and inlet) have the same water quality, low variability of all bacterial numbers and slight increase toward inflow. Presence of heterotrophic bacteria indicated moderate pollution by organic matter. Critical concentrations of enterococci were observed in all season (class III) and E. coli during in summer period (class III). All sampling points are good supplied with oxygen, with relatively high concentration of organic matter, total nitrogen and total phosphorus. Generally, more values for concentration of total phosphorus during the investigated period belong to mesotrophic state. According total nitrogen water quality of River Sateska at all localities belong from IV – V class (meso-eutrophic state).

**Key words**: River Sateska, water quality, microbiological and chemical indicators

#### P60

# Compartion between organochlorine pesticide residues, organic matter and lipophilic bacteria in sediment from Lake Ohrid and Lake Dojran

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#### **ABSTRACT**

Organochlorine pesticides as members of the persistent organic pollutants group, with their properties such as: toxicity, lipophilicity, stability and bioaccumulation pose a potential risk to the environment and all living things, including human as the top link in the trophic food chain. Once introduced into the water ecosystem, the persistent organic substances forever remain in it. They are introduced in the living organisms through the food or are accumulated on suspended particles, and due to their high affinity for binding to organic matter, they eventually accumulate in sediments that are a secondary source of contamination. The purpose of this paper is to show the correlation between the content of the total recorded organochlorine pesticides and organic matter and the number of lipophilic bacteria in the sediment samples collected from the littoral zone of two natural lakes in Macedonia, Lake Ohrid and Lake Dojran. The conclusion from the research results is that the highest values for all parameters analyzed were determined in sediment samples collected from Lake Dojran. This is because of the different trophic state of these two lake ecosystems. Lake Ohrid is oligotrophic ecosystem, while Lake Dojran shows eutrophic character. It can also be concluded that the content of the registered organochlorine pesticides is positively correlated with the content of organic matter and the number of lipophilic bacteria in the sediment samples of the surveyed sites.

**Key words**: organochlorine pesticides, lipophilic bacteria, trophic status, Lake Ohrid, Lake Dojran

#### P61

## Antibiotic resistance – the plague of today

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#### **ABSTRACT**

Antibiotic resistance is a major public-health problem worldwide, and international efforts are needed to counteract its emergence. Antibiotics are among the very few drugs that cure, rather than just reduce the symptoms of a disease. Over prescription, unfinished treatment courses and use in animal farming are some of the major reasons for the spread of resistance genes amongst bacteria. Even though big efforts are put into new antibiotic development and screening of soil and other environments for organisms that produce substances with antimicrobial activities there hasn't been much progress in the area and new antibiotics are hard to come by. The danger of a return to a pre- antibiotic era, where a simple infection can cause death is now becoming a serious threat. Scientists are looking for alternative methods of combating antibiotic resistant bacteria. For the past 60 years, we have conducted a global experiment in evolutionary selection and now we are seing the detrimental results. Strict world-wide measures need to be enforced to ensure antibiotic resistance does not develop further.

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