University of Plovdiv "Paisii Hilendarski"
Medical University of Plovdiv
National Center of Infectious and Parasitic Diseases

THIRD INTERNATIONAL CONFERENCE ON ZOOLOGY, ZOONOSES AND EPIDEMIOLOGY

PROGRAMME & ABSTRACTS

October 21 – 23, 2019
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 Third International Conference on Zoology, Zoonoses and Epidemiology.

The Conference provides the opportunity for academics, researchers, epidemiologists, microbiologists and vets to gather and discuss about the latest advances in the field of animal taxonomy, faunistics, ecology, applied zoology, vector-borne diseases, epidemiology, population genetics, molecular biology, biomonitoring, conservation zoology.

We hope the Conference will be a great platform 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.

We assure the Third International Conference on Zoology, Zoonoses and Epidemiology is going to facilitate future collaborations and develop ideas for further research 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 "Paisii Hilendarski"
Medical University of Plovdiv, Bulgaria
National Center of Infectious and Parasitic Diseases, Sofia
Institute of Biodiversity and Ecosystem Research, BAS
Bulgarian Scientific Society of Epidemiology of infectious and noninfectious diseases
South East European Center of Infectious Diseases Surveillance and Control (SECID), Tirana, Albania

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Prof. DSc **Boyko Georgiev**, Institute of Biodiversity and Ecosystems Research, BAS
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Assoc. Prof. Dr. **Anelia Stojanova**, University of Plovdiv "Paisii Hilendarski"

SCIENTIFIC PROGRAMME
Third International Conference on Zoology, Zoonoses and Epidemiology
October 21 – 23, 2019, Hissar, Bulgaria

Monday, 21 October 2019

13.00 – 18.00 Arrival, registration, posters set up. SPA Hotel "Hissar".

EPIDEMIOLOGY
(Maritsa hall)
Session chairs: Prof. Rumen Konstantinov, Dr. NadezhdaVladimirova

14.00 – 14.10 Communicable diseases in the human evolution. **Wiliam Monev**

14.10 – 14.20 Epidemiological forecasting – biosocial nature and methodology. **Yordanka Stoilova**

14.20 – 14.30 Contemporary aspects of the epidemiological surveillance of socially significant tick-borneinfections (TBIs) in Varna region. **Rumen Konstantinov**

14.30 – 14.40 Epidemiological aspects of anthrax in North-East Bulgaria. **Tsonko Paunov**

14.40 – 15.00 Benefits for the community from pneumococcal immunization programs. **Magdalina Mircheva, GSK**

15.00 – 15.10 Legionella infections in Bulgaria. **Iskra Tomova**

15.10 – 15.20 Zoonotic potential of *Helicobacter* spp. **Irena Mladenova**

15.20 – 15.40 Decontamination – principles and tendencies. Contemporary methods and tools in reprocessing of reusable medical devices. **Dragomir Ivanov, MTE**

15.40 – 15.50 Basic principles and approved decontamination method against biological warfare agents. **Milena Nikolova**

15.50 – 16.10 Coffee-break

EPIDEMIOLOGY

(Maritsa hall)

Session chairs: Assos. Prof. Nina Gatcheva, Prof. Irena Mladenova

16.10 – 16.25 Implementation of Medical standard on prevention and control of nosocomial infections: Conclusions based on questionnaire surveys of BAPIC BulNoso, 2012 – 2018. **Nina Gatcheva**

16.25 – 16.40 The pressure of vaccines on microorganisms and their response. **Andrey Galev**

16.40 – 17.00 Influenza and influenza vaccination in cardiovascular diseases. **Nikolay Badzhinerov, Sanofi-Pasteur**

17. 00 – 17.15 Measles in Europe and in Bulgaria during the 2-nd decade of 21 century. **NadezhdaVladimirova**

17.15 – 17.35 Immunizations for preterm infants. **Petya Galabova, GSK**

17.35 – 17.50 A review of global good practices to improve communication between pediatricians and parents on the topic of vaccines. **Savina Stoitsova**

17.50 – 18.00 Vaccine-preventable diseases in HIV (+) persons. **Nikolay Vatev**

19.30 **Welcome cocktail** at the Lobby bar, SPA Hotel "Hissar".

Tuesday, 22 October 2019

8.00 – 9.00 Registration

9.00 – 9.45 **Opening**. Maritsa hall in SPA Hotel "Hissar"

9.45 – 10.15 **Plenary lecture:** Public Health challenges. **Assoc. Prof. Dr. Angel Kunchev, MD**, Chief State Health Inspector, Ministry of Health

10.15 – 10.45 **Plenary lecture:** Satellite tracking of birds: science and practice. **MSc Stoycho Stoychev,** Conservation Director of the Bulgarian Society for the Protection of Birds /BirdLife in Bulgaria

10.45 – 11.00 **Coffee-break**

GENETICS AND MOLECULAR TAXONOMY

(Hissar hall)

Session chair: Prof. Dr. Teodora Staykova

Session secretary: Assist. Prof. Dr. Miroslav Antov

11.00 – 11.15 Molecular taxonomy study of genus *Alosa* (Clupeidae) in Black Sea based on variability in cytochrome b sequences. **Darinka Boyadzhieva-Doychinova**

11.15 – 11.30 Diagnostic markers for discrimination of the Bulgarian honey bee *Apis mellifera macedonica*, type *rodopica*, in accordance with its valuable ethological and productive qualities. **Evgeniya Ivanova**

11.30 – 11.45 New chromosomal form of mole rat in Bulgaria? Synaptonemal complex karyotype of *Nannospalax leucodon* (Rodentia) from the eastern area of Sofia. **Sergey Matveevsky**

BIODIVERSITY, ECOLOGY AND CONSERVATION ZOOLOGY

(Hissar hall)

Session chair: Assoc. Prof. Dr. Tsenka Chassovnikarova

Session secretary: Chief Assist. Prof. Dr. Vesela Mitkovska

11.45 – 12.00 Some parasitological features of *Paradiplozoon bliccae* (Monogenea: Diplozoidae) infestation in *Pseudophoxinus burduricus* – an endemic fish species from Doğanbaba Creek, Turkey. **Özlem Özmen**

12.00 – 12.15 Pathological effects of *Eustrongylides excisus* in four teleost fish species of Eğirdir Lake, with a new host record. **Özlem Özmen**

12.15 – 12.30 Factors influencing the expansion of the Pine processionary moth (*Thaumetopoea pityocampa*) in Central Bulgaria. **Gergana Zaemdzhikova**

12.30 – 12.45 The flight of the Pine processionary moth (*Thaumetopoea pityocampa*) in the Valey of Mesta, Bulgaria. **Gergana Zaemdzhikova**

12.45 – 13.00 Impact of egg parasitoids on Pine processionary moth (*Thaumetopoea pityocampa*) in a new habitat in Bulgaria. **Georgi Georgiev**

13.00 – 13.15 DNA damage in sperm cells – biomarker for genotoxicity in striped field mouse (*Apodemus agrarius* Pallas, 1771) from rice fields. **Vesela Mitkovska**

ONE HEALTH

(Maritsa hall)

Session chairs:

Assoc. Prof. Angel Kunchev, Prof. Dr. Todor Kantardzhiev

11.00 – 11.45 **Review of new and current Public Health Programs in Republic of Bulgaria**

- National Program for control and treatment of rotavirus infections in Republic of Bulgaria (2017 – 2021).
- National Program for primary prophylaxis of cervical cancer (2017 – 2020).
- National Program for improving seasonal influenza vaccination (2019 – 2022).
- National Program for elimination of measles and rubella (2019 – 2022).
- Draft of National Program for control of viral hepatitis in Republic of Bulgaria (2019 – 2024).

11.45 – 13.00 **National Immunization Calendar – News, Challenges, Reporting and Control**

- National Immunization Calendar 2020 – changes, trends, experience – Implementation of NIC 2019.
  Dr. Kremena Parmakova, Ministry of Health
- Spread of pneumococcal serotypes in Bugaria after the introduction of PCV10.
  Prof. DSc Todor Kantardzhiev,
  National Center of infectious and parasitic diseases.
- Vaccinations – the point of view of the GP.
  Assoc. Prof. Lyubomir Kirov, National association of general practitioners in Bulgaria.
- Anti-epidemic measures for measles epidemic in Sofia region, 2019. Alexander Zlatanov, **Bojidarka Kayrakova** (RHI, Sofia region), George Donkov (Department of Epidemiology, Medical University, Sofia).

- Legal aspects of the Immunization Calendar implementation – lawyer's point of view.
  Maria Sharkova, lawyer
- Vaccinations and anti-vaccine movement, attitude of the Bulgarians – joint study of Ministry of Health and University of Ruse "Angel Kanchev".
  Assoc. Prof. Nikola Sabev, University of Ruse
- MenB and Reverse vaccinology.
  Dr. Magdalina Mircheva, GSK

13.00 – 14.00 Lunch, Restaurant SPA Hotel "Hissar"

BIODIVERSITY, ECOLOGY AND CONSERVATION ZOOLOGY
(Hissar hall)
Session chair: Assoc. Prof. Dr. Ivelin Mollov
Session secretary: Chief Assist. Prof. Dr. Peter Boyadzhiev

14.00 – 14.15 Honey bee colony losses in Bulgaria during the period 2017 – 2019. **Iliyana Ilieva**

14.15 – 14.30 The parasitoid complex associated with *Larinus sibiricus* Gyllenhal, 1835 (Coleoptera: Curculionidae) in Bulgaria. **Miroslav Antov**

14.30 – 14.45 Atypical winter activity of fungus gnats (Diptera: Mycetophilidae) in critically endangered Mediterranean and sub-Mediterranean type habitats in Bulgaria. **Aneliya Pavlova**

14.45 – 15.00 Fish diversity and distribution in the Ceyhan River Estuary (Adana – Turkey). **Deniz Innal**

15.00 – 15.15 Seasonal changes in density and diversity of fishes in the Seyhan River Estuary (Mersin-Turkey). **Deniz Innal**

15.15 – 15.30 Land-management subsidies for shrub removal are a major threat for herpetofauna in Bulgaria. **Georgi Popgeorgiev**

15.30 – 15.45 Pilot line transect surveys of cetaceans in a Bulgarian MPA – BG0001007 Strandzha SCI. **Dimitar Popov**

15.45 – 16.00 Coffee-break

16.00 – 18.00 Poster session, Trakiya hall in SPA Hotel "Hissar"

19.30 **Conference dinner**, Restaurant "Prestige" in SPA Hotel "Hissar"

ONE HEALTH
(Maritsa hall)
Session chairs:
Assoc. Prof. Angel Kunchev, Prof. Dr. Todor Kantardzhiev

14.00 – 14.50

- Vector-borne zoonoses – current situation and future challenges to human and veterinary medicine.
- Vector-borne infectious diseases in humans in Bulgaria. **Prof. Iva Hristova**, National Center of Infectious and Parasitic diseases; **Assoc. Prof. Angel Kunchev**, MD, Ministry of Health, Republic of Bulgaria
- Current issues related to epizootic of African swine fever. **Prof. Georgi Georgiev**, Risk Assessment Center on Food Chain
- Role of different vectors for the outbreak of epidemics and epizootics in the country.

14.50 – 15.45

- Disinfections, disinsections, deratizations – new and established disinfectants and pest control substances, mode of action, in cooperation with local authorities and Bulgarian Food Safety Agency
- Principles of DDD activities in epidemiology. **Prof. Wiliam Monev**, National Expert Council of Surveillance and Control of Infection Diseases and Immunoprophylaxis, Ministry of Health
- Major mistakes in controlling mosquito populations in the country. **Chief Assist. Prof. Dr. Ognyan Mikov**, National Center of Infectious and Parasitic diseases
- DDD field activities – the experience of the health inspector in Sofia RHI. **Tsezarina Ilieva**, Sofia RHI
- BFSA's experience in organizing large-scale disinfestations in the country.

EPIDEMIOLOGY

(Trakiya hall)

Session chairs: Prof. Yordanka Stoilova, Assoc. Prof. Milena Karcheva

14.00 – 14.10 Chemical agents suitable for treatment of incubators. Disinfection of incubators – multicentre questionnaire study, 2019. **Milena Nikolova**

14.10 – 14.20 Efficacy and toxicological profile of repellents with IR3535 and DEET. **Sofia Bukovska**

14.20 – 14.40 Cleaning and disinfection in healthcare facilities – key aspect in the combat with healthcare-associated infections. **Mitko Sterev**, BioKomTrendafilov

14.40 – 14.50 Epidemiological assessment – a basic element of medical support planning of military operations. **Rostislav Kostadinov**

14.50 – 15.25 Evidence – based medicine in support of immunoprophylaxis. **Ani Kevorkyan**, MSD

15.25 – 15.35 Study of infectivity with *Borrelia burgdorferi* sl among *Ixodes ricinus* population in Pleven region, Bulgaria. **Alexander Blazhev**

15.35 – 15.45 Research of the wild pigs infected with the parasite *Trichinella* in the Blagoevgrad district for the period from 01.01.2018. to 01.01.2019. **Andrey Galev**

15.45 – 15.55 Epidemiological evaluation of *L. monocytogenes* circulation. **Svetla Staneva**

15.50 – 16.00 Coffee-break

16.00 – 18.00 Poster session, Trakiya hall in SPA Hotel "Hissar"

19.30 **Conference dinner**, Restaurant "Prestige" in SPA Hotel "Hissar"

ONE HEALTH

(Maritsa hall)

16.00 – 17.00 Closed meeting with the directors of Regional Health Inspectorates.

19.30 **Conference dinner**, Restaurant "Prestige" in SPA Hotel "Hissar"

Wednesday, 23 October 2019

9.00 – 11.00 **Round table** – discussion on current problems in the prevention and control of infectious diseases. **Maritsa hall**

9.30 – 11.00 **Presentations of sponsors** – presentations of vaccines, medicines and disinfectants used in practice. **Trakiya hall**

11.00 – 11.30 **Closing the conference. Maritsa hall**

11.30 – 12.00 General Assembly of Bulgarian Scientific Society of Epidemiology of infectious and noninfectious diseases. **Maritsa hall**

12.00 **Lunch, Restaurant SPA Hotel "Hissar"**

13.00 **Departure**

POSTER SESSION

(Trakiya hall)

ZOOLOGY AND ZOONOSES (P1 – P47)

Poster panel:

Assoc. Prof. Dr. Hristo Dimitrov, Assoc. Prof. Dr. Georgi Popgeorgiev

P1 New and additional records of earthworms (Annelida: Clitellata) from Kopaonik Mountain: First finding of *Allolobophora treskavicensis* (Mršić, 1991) in Serbia. Filip Popović, Mirjana Stojanović, Tanja Trakić, Jovana Sekulić, Slobodanka Sekulić, **Ralitsa Tsekova**

P2 Effects of a modern biorational insecticide spinosad on earthworm, *Eisenia fetida* (Savigny, 1826). Jovana Sekulić, Mirjana Stojanović, Tanja Trakić, Filip Popović, **Ralitsa Tsekova**

P3 The first detection of *Toxoplasma gondii* in rivers in Serbia. Vladimir Ćirković, Aleksandra Uzelac, Ivana Klun, **Dragana Miličić**, Olgica Djurković-Djaković

P4 Morphological variability of *Lepidurus apus* (Linnaeus, 1758) (Notostraca, Crustacea) from Bosnia and Herzegovina. **Dragana Miličić**, Đorđe Janković, Goran Šukalo, Dejan Dmitrović, Srećko Čolić, Ivana Šaganović, Bojan Ilić, Luka Lučić, Sofija Pavković-Lučić, Vukica Vujić

P5 Where to swim, where to eat? Laboratory report on behaviour of the ostracod *Heterocypris incongruens.* **Tatjana Savić**, Branka Petković, Sofija Pavković-Lučić, Dragana Miličić

P6 Influence of sex ratio and wing morphology on mating success in *Drosophila melanogaster*: lessons from female and multiple choice experiments. **Sofija Pavković-Lučić**, Jelena Trajković, Dragana Miličić, Tatjana Savić

P7 Population-genetic heterogeneity in *Messor structor* and *Messor barbarous* populations as an approach for their discrimination. **Ivan Stoyanov**, Teodora Staykova, PenkaVasileva, Teodora Popova, Evgeniya Ivanova

P8 Hygienic behaviour and fat body development in worker bees (*Apis mellifera* L.). **Svilen Lazarov**, Ivanka Zhelyazkova

P9 Hygienic behaviour of local honey bee (*Apis mellifera* L.) and differences in the dimensions of chitin body parts of worker bees. **Svilen Lazarov**, Ivanka Zhelyazkova

P10 Ontogenetic and caste differentiation in the expression of water-soluble proteins and some isozymes in *Reticulitermes lucifugus* (Rossi, 1792) (Isoptera: Rhinotermitidae). Evgeniya Ivanova, Ivan Vulchev, Teodora Staykova, Miroslav Antov, Teodora Popova, **Penka Vasileva**, Ivan Stoyanov

P11 Isoenzyme polymorphism of silkworm (*Bombyx mori* L.) breeds from germplasm resourses of Bulgaria. **Teodora Staykova**, Panomir Tzenov, Yolanda Vasileva, Nikoleta Takova, Evgenya Ivanova, Ivan Stoyanov, Penka Vasileva, Teodora Popova, Miroslav Antov

P12 New parasitoids of *Leucoptera sinuella* (Reutti) (Lepidoptera: Lyonetiidae) in Bulgaria. Maria Dobreva, Pencho Dermendzhiev, Nikola Kavardzhikov, Rumen Nachev, Peter Boyadzhiev, **Georgi Georgiev**

P13 Entomopathogenic fungi (Ascomycota, Hypocreales) as natural antagonists of the pine processionary moth, *Thaumetopoea pityocampa*, in Bulgaria. Marek Barta, Miriam Kádasi Horáková, **Margarita Georgieva**, Plamen Mirchev, Gergana Zaemdzhikova, Daniela Pilarska, Danail Takov, Milcho Todorov, Zdravko Hubenov, Plamen Pilarski, Georgi Georgiev

P14 *Syspastospora parasitica*, a parasite of entomopathogenic fungi *Beauveria pseudobassiana* and *B. varroae* on *Thaumetopoea pityocampa* – a new tritrophic associations in Bulgaria. **Margarita Georgieva**, Plamen Mirchev, Georgi Georgiev

P15 Sensitivity to climate change of selected zooplankton species. **Kremena Stefanova**, Valentina Doncheva, Nadejda Valcheva, Elitsa Stefanova, Snejana Moncheva, Nataliya Slabakova, Violeta Slabakova

P16 Zooplankton traits vs. environmental conditions within intra-decadal scale assessment in Lake Ohrid (North Macedonia, SE Europe). **Orhideja Tasevska**, Maria Špoljar, Dafina Gušeska, Goce Kostoski, ElizabetaVeljanoska Sarafiloska, Suzana Patcheva

P17 Modelling factor – Macrophyte communities for macrozoobenthos in Lake Prespa, Republic of North Macedonia. **Biljana Budzakoska Gjoreska**, Sasho Trajanovski, Sonja Trajanovska, Marina Talevska

P18 Diversity of macrozoobenthos in different type of Charophyta mono species communities in Lake Ohrid. **Sonja Trajanovska**, Marina Talevska, Biljana Budzakoska-Gjoreska, Saso Trajanovski

P19 Physical-chemical and biological indicators in function of the Lake Ohrid trophic state assessment. **Elizabeta Veljanoska-Sarafiloska**, Orhideja Tasevska, Lence Lokoska, Suzana Patceva, Jovica Leshoski

P20 Histochemical and biochemical alterations in Zebra mussel, *Dreissena polymoprha* (Pallas, 1771) after Cd and PAHs chronic exposure. **Stela Stoyanova**, Vesela Yancheva, Iliana Velcheva, Ilia Iliev, Tonka Vasileva, Veselin Bivolarski, Elenka Georgieva

P21 Cd and PAHs exposure changes the condition index and soft tissue wet ratio in *Dreissena polymorpha* (Pallas, 1771): a statistical perspective. **Vesela Yancheva**, Stela Stoyanova, Ivelin Mollov, Elenka Georgieva, IlianaVelcheva

P22 Comparative analysis of the oxidative stress in Bulgarian Black sea bivalves and their bioindicator potential. Lachezar Yakimov, Elina Tsvetanova, Almira Georgieva, Galina Nenkova, Nesho Chipev, **Albena Alexandrova**

P23 Evaluation of the impact of *Rapana* fisheries with beam trawl on juvenile stages of turbot in Western Black Sea. Elitsa Petrova, Vesselina Mihneva, **Feriha Tserkova**, Stoyko Stoykov, Stanimir Valchev, Philip Penchev

P24 Contribution to parasite fauna of Prespa roach *Rutilus prespensis* (Karaman, 1924) (Pisces: Cyprinidae) in Lake Prespa, North Macedonia. **Stojmir Stojanovski**, Dijana Blazhekovikj-Dimovska, Stoe Smiljkov

P25 Length-weight relationships for 5 cyprinid fish species from Lake Ohrid. **Blagoja Trajchevski**, Trajche Talevski

P26 Ecosystem pressure and impact on fish population in Channel Studencista (Lake Ohrid). **LidijaVelkova-Jordanoska**, Blagoja Trajcevski

P27 Insights into the genetic structure of the genus *Chondrostoma* (Pisces, Cyprinidae) across the central part of Balkan Penninsula. **LidijaVelkova-Jordanoska**

P28 Researches on reed belt from Lake Prespa as habitat and spawning ground for cyprinid fishes. **Marina Talevska**, Trajce Talevski

P29 The impact of alien fish species on native fish population from Lake Dojran and Lake Tikvesh (Aegean catchment area). **Trajce Talevski**, Dragana Milosevic, Marina Talevska, Blagoja Trajceski, Aleksandra Lesoska, Elena Pejoski

P30 Long-term changes of the ichthyofauna in the Iskar River, Danube basin. **Luchezar Pehlivanov**, Apostolos Apostolou, Stefan Kazakov, Boris Velkov

P31 Feeding ecology of the Green Toad (*Bufotes viridis* complex) in urban environment. Part 1. Trophic spectrum and niche. **Ivelin Mollov**, Anelia Stojanova, Peter Boyadzhiev

P32 Feeding ecology of the Green Toad (*Bufotes viridis* complex) in urban environment. Part 2. Prey Availability and electivity. **Ivelin Mollov**, Anelia Stojanova, Peter Boyadzhiev

P33 Pingers as cetacean bycatch mitigation measure in Bulgarian turbot fishery. **Dimitar Popov**, Galina Meshkova, Polina Hristova, Gradimir Gradev, Marina Panayotova, Hristo Dimitrov

P34 Hematology data of striped mouse (*Apodemus agrarius*) from Southeastern Bulgaria (Strandzha Mountain): indicators for environmental status. **Hristo Dimitrov**, Georgi Markov, Angel Kunchev, Tsenka Chassovnikarova, Vesela Mitkovska

P35 Strategies in open field behaviour and new object exploration in eastern broad-toothed field mouse, *Apodemus mystacinus* and yellow-necked mouse, *Apodemus flavicollis.* Krastio Dimitrov, **Daniela Simeonovska-Nikolova**, Veselina Sidova, Venislava Racheva

P36 Species diversity and population structure of small mammalian communities depending on habitat heterogeneity in Lozenska Planina Mountain, Bulgaria. Roumiana Metcheva, **Mihaela Beltcheva**, Iliana Aleksieva, José Antonio Heredia-Rojas

P37 Evidence of radioprotective effect of Resveratrol against extremely low frequency electromagnetic fields clastogenic effect. José Antonio Heredia-Rojas, **Mihaela Beltcheva**, Roumiana Metcheva, Abraham O. Rodríguez-De la Fuente, Omar Heredia-Rodríguez

P38 Natural and induced UV irradiation effects on some morpho-physiological and genetic characteristics of albino mice. **Michaela Beltcheva**, Roumiana Metcheva, Margarita Topashka-Ancheva, Nikolay Tyutyundzhiev, Iliana Aleksieva, Peter Ostoich, Christo Angelov, José Antonio Heredia Rojas

P39 Comparative analysis of age changes in the growth of lower canine teeth of wild boar (*Sus scrofa*) males in Bulgaria. **Damyan Damyanov**

P40 Dynamics of the number and shooting of wild boar (*Sus scrofa* L.) in Bulgaria and the danger of epizootic. **Damyan Damyanov**, Peter Genov

P41 Red deer (*Cervus elaphus* L.) as a biomonitor for contemporary heavy metal pollution of the environment in forest mountain regions in Bulgaria. **Georgi Markov**, Atidzhe Ahmed, Chavdar Zhelev

P42 Epigenetic variation and distinctness of red fox (*Vulpes vulpes*) populations in its European range. **Georgi Markov**, Milena Gospodinova

P43 The relationship between the fertility potential quality and leukocyte concentration in the sperm as a potential indicator for the mutagenic effect of infections. **Spas Dzhoglov**, Vesela Mitkovska, Evgeniya Ivanova

P44 Monitoring of micronuclei frequency in rodents with blood parasites. **Vesela Mitkovska**, Hristo Dimitrov, Angel Kunchev, Tsenka Chassovnikarova

P45 Rodent control in urban industrial areas in Plovdiv (Bulgaria): from research to action. **Eliza Koteva**, Hristo Dimitrov, Vesela Mitkovska, Nasko Atanasov, Tsenka Chassovnikarova

P46 Compensatory increase of the reproductive capacity of red fox (*Vulpes vulpes*) in sympatric coexistence with the golden jackal (*Canis aureus*). **Albena Vlaseva**, Tsenka Chassovnikarova, Vesela Mitkovska, Hristo Dimitrov

P47 General evaluation of the impact of invasive fish species on native freshwater fish fauna in Turkey. **Saniye Cevher Özeren**

EPIDEMIOLOGY (P48 – P61)

Poster panel:
Assoc. Prof. Tsonko Paunov, Assoc. Prof. Ani Kevorkyan

P48 Seroprevalence of Parvovirus B19 IgG antibodies among pregnant women. **Milena Karcheva**, Anton Petrov, Alexander Blazhev

P49 Seroepidemiological survey of Parvovirus B19 IgG in childbearing age women. **T. Petkova**, S. Pachkova, Tz. Doichinova

P50 Status and development of the anti-epidemic control system in the region of Varna. **Eliyana Ivanova**, Rumen Konstantinov

P51 Investigating the attitudes of parents to vaccines and immunizations. **Tsvetanka Mincheva**, Svetla Angelova, Jordanka Mitova, Victoria Doycheva, B. Velinova

P52 Study of hygienic disinfection of hands of staff and students in dental practice. **V. Stoeva**, A. Petrova, H. Batselova, D. Ivanov, B. Tilov

P53 Control of *Legionella* spp. in dental practice in England and Bulgaria. **V. Stoeva**, S. Ilieva, I. Tomova, A. Atanasovski, A. Kevorkyan, Y. Stoilova

P54 Varicella outbreak in a daycare in Plovdiv (2019). **H. Batselova**, N. Vatev, A. Galev

P55 Surveillance of Ventilator-associated pneumonia in neonatal intensive care unit. **V. Rangelova**, A. Kevorkyan, M. Krasteva, Y. Kalchev

P56 The main epidemiological paradigm in the context of viral hepatitis C: Comorbidity and co-infections. K.Terzieva, **M. Nikolova**, I. Popivanov, T. Doichinova, D. Shalamanov

P57 The role serological screening for determining of HBsAg among hospitalized and ambulatory patients. **A. Gotseva**

P58 Diagnosis and treatment of bisphosphonate-associated osteonecrosis of the jaws. **Meri Hristamyan**, Petya Pechalova, Ralista Raycheva, Veselka Hristamyan, Yordanka Stoilova

P59 Epidemiological surveillance – indicator for the effectiveness of the medical support in floods. **Rumyana Etova**, Rostislav Kostadinov

P60 Epidemiology and health education as disaster medical support elements. Rostislav Kostadinov, **Mariya Georgieva**

P61 Which measures to prevent the spread of measles at a hospital in Bulgaria are recommended when examining / hospitalizing a patient. **V. Voynova-Georgieva**, N. Gatcheva, T. Perchemlieva, A. Kevorkyan

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ORAL TASKS

GENETICS AND MOLECULAR TAXONOMY

Molecular taxonomy study of genus *Alosa* (Clupeidae) in Black Sea based on variability in cytochrome b sequences

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Abstract: The family Clupeidae is represented in Black Sea by nine species. Genus *Alosa* is one of the most challenging group because some molecular data demonstrated that the differences between *A. caspia* and *A. immaculata* are less than 0.8 which could means that these are not two different species or they very recently split and may have hybridization between them. The economic importance of Clupeidae species on one hand and the need to take adequate measures for the conservation and proper management of fishery resources in the Black Sea region determine the need for more detailed study of biodiversity and phylogenic relationships among species in this family. In this study studied one hundred and sixty samples of representatives of genus *Alosa* to isolate total DNA and study variability mitochondrial encoded gene of cytochrome b. For this purpose cytochrome b was isolated by PCR from twelve areas along Bulgarian coastal areas. The obtained sequences were processed by MEGA 7.2. The results displayed high similarity in studied representatives of *A. immaculata*, which clustered together in the resulting tree. The representatives of *A. caspia* ssp. *nodrmanni* were out grouped from *A. immaculata*. Although both species formed one joint cluster, the first was clearly separated from *A. immaculata*. Interestingly five representatives out grouped from above mentioned species and showed indications that they belong to *Alosa tanaica*. To our knowledge and according NCBI database this is the first time when sequences of this species are obtained and deposited in global genetic data base.

Key words: *Alosa*, Clupeidae, fish, Black Sea, PCR, molecular markers

Diagnostic markers for discrimination of the Bulgarian honey bee *Apis mellifera macedonica*, type *rodopica*, in accordance with its valuable ethological and productive qualities

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Abstract: The genetic polymorphism in honey bee populations from different Bulgarian regions and in European populations of *A. m. carnica*, *A. m. caucasica* and *A. m. macedonica* has been investigated by usage of DNA microsatellite analysis. Nine microsatellite loci (Ac011; A024; A043; A088; Ap226; Ap238; Ap243; Ap249 and Ap256) and 1443 individual worker bee samples have been included in the study. Polymorphism on all of the 9 analyzed microsatellite loci has been observed and compared for all of the studied populations. Totally, 121 alleles were found for the nine microsatellite loci in Bulgarian populations studied. The phylogenetic relationships between them and the grouping of the honey bee populations from the Eastern, Western and Central parts in Bulgaria have been discussed. Thirty five private alleles were observed in 19 of the studied local populations. On the other hand, the microsatellite polymorphism observed for the Bulgarian populations has been compared to those found in other European populations. The phylogenetic relationships between Bulgarian honey bee populations and European populations of *A. m. macedonica*, *A. m. carnica* and *A. m. caucasica* have been analyzed. UPGMA and Neighbor-joining dendrograms have been constructed on the base of the calculated Nei's genetic distance between the studied populations. Twenty two private alleles were detected for the Bulgarian *A. m. mcedonica* populations in comparisons to other studied European populations. Nine private alleles

were detected for the Greek populations of *A. m. macedonica*, 20 alleles – for the *A. m. carnica* populations and 6 – for the *A. m. caucasica* population. Clear diagnostic markers, appropriate for discrimination of local Bulgarian honey bee have been found and described. On the basis of the genetic differences established between the Bulgarian and other *A. m. macedonica* populations, the conclusion has been done that the local Bulgarian honey bee is a different ecotype of *A. m. macedonica – A. m. macedonica*, type rodopica. The described in this study diagnostic markers should be considered in accordance with the biological and productive valuable characteristics of the local Bulgarian honey bee and together with the complex of morphological and ethological indicators they should be taken into account when conducting activities for conservation of the gene pool of *Apis mellifera macedonica*, type rodopica in Bulgaria.

Key words: *A. m. macedonica*, type *rodopica*, microsatellites, genetic polymorphism, discrimination

New chromosomal form of mole rat in Bulgaria? Synaptonemal complex karyotype of *Nannospalax leucodon* (Rodentia) from the eastern area of Sofia

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Abstract: Blind mole rats *Nannospalax* have wide chromosome variability. In the Lesser mole rat *N. leucodon* (Nordman, 1840) superspecies, the diploid chromosome number (2n) varies between 46 and 58 and the chromosomal arms (NF) from 76 to 98. Ten chromosomal forms have been described in Bulgaria in 1983. No karyological studies of mole rats have been conducted ever since while the Southern part of the country has not been investigated at all.

Two male of mole rats were caught northwest of Ravno Pole village near Sofia in May 2019. Karyotyping was carried out on the basis of immunocytochemical analysis of spread synaptonemal complexes (SC). SC is a nucleoproteide skeleton formed between two homologous chromosomes in the meiotic prophase I. The number of SCs corresponds to the haploid number of the species chromosomes. The SCs analysis clearly identifies the centromere positions in each bivalent and can be used as an indicator of the sex chromosome behavior and the chromosomal heteromorphism in heterozygous animals.

An SCs study of *N. leucodon* has been conducted for the first time. It was found that the animals had 2n = 56, NFa = 84 and NF = 88. Fifteen biarmed chromosomes (2 metacentrics (M), 3 submetacentrics

(Sm) and 10 subtelocentrics (St)) and 12 acrocentrics (A) were identified in the SC's karyotype. The X chromosome is a medium Sm. The Y chromosome is a small St. A similar mole rat karyotype was found in Novo Selo near Plovdiv: 2n = 56, NFa = 84 and NF = 88, however with a different chromosome combination: 1M, 7Sm, 7St, 12A, X-Sm, Y-St. The distance between the Ravno Pole and Novo Selo populations is about 100 km. There are karyotypes 2n=54, NF=86 (Pazardzhik) and 2n = 6, NF = 90 (Ihtiman) between them. These two populations with same 2n, NFa, NF, but different chromosome structures are most likely independently separated chromosomal forms. Further G- and C- banding techniques should be performed.

Key words: lesser mole rat, chromosome variability, synaptonemal complexes, new chromosomal form

BIODIVERSITY, ECOLOGY AND CONSERVATION ZOOLOGY

Some parasitological features of *Paradiplozoon bliccae* (Monogenea: Diplozoidae) infestation in *Pseudophoxinus burduricus* – an endemic fish species from Doğanbaba Creek, Turkey

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Abstract: This study was aimed to identify *Paradiplozoon bliccae* (Monogenea: Diplozoidae) in an endemic fish species *Pseudophoxinus burduricus* (Teleostei: Cyprinidae) and to describe parasite's prevalence and intensity of infection. *P. burduricus* specimens were collected between January 2014 and February 2015 in Doğanbaba Creek (Yeşilova-Burdur). A total of 60 *P. burduricus* specimens were examined for monogenean parasites. During the course of surveys, diplozoid specimens were collected from the gills of these fish and examined microscopically for identification. *Paradiplozoon bliccae* specimens were isolated from the examined fish. Prevalence, intensity and seasonality of infection, age classes and sex compositions of infected population have been calculated. It has been recorded that the highest point of prevalence being in summer and the highest invasion value in the two-age class of fish. Hyperaemia, haemorrhage and atrophy of the gills were observed during pathological examination of the parasite infested fish individuals.

Key words: Diplozoidae, Doğanbaba Creek, Monogenea, Prevalence

Pathological effects of *Eustrongylides excisus* in four teleost fish species of Eğirdir Lake, with a new host record

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Abstract: This study was carried out to determine infection of *Eustrongylides excisus* in fish species inhabiting Egirdir Lake between January 2018 and October 2018. Host specimens belonging to *Aphanius iconii*, *Atherina boyeri*, *Knipowitschia caucasica* and *Sander lucioperca* were analyzed to investigate the parasite community. The highest prevalence of infection was determined in pike-perch. The nematode *Eustrongylides excisus* Jägerskiöld, 1909 was recorded for the first time on *Knipowitschia caucasica* Berg, 1916, a non-native fish species in Eğirdir Lake. Moreover, the present paper provides first microphotographs concerning histopathological aspects of *E. excisus* encysted in three fish species: big-scale sand smelt, caucasian dwarf goby and pike-perch. At the histopathological examination, hyperaemia, edema, microhaemorrhage, inflammatory reaction and necrosis were observed around the parasites.

Key words: Eğirdir Lake, *Eustrongylides excisus*, fish host, histopathology

Factors influencing the expansion of the Pine processionary moth (*Thaumetopoea pityocampa*) in Central Bulgaria

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Abstract: The Stara Zagora region is a dynamically developing new range of the pine processionary moth (*Thaumetopoea pityocampa*) in Central Bulgaria, which emerged 25 years ago and continues to expand. Available data on the expansion process are merged into an electronic data bank on the basis of which a factor analysis has been carried out. It has been shown that the attacks do not depend on the age of the pine plantations and are slightly dependent on the stocking rate. In Central Bulgaria, the attacks of the pine processionary moth are limited to altitudes up to 900 m, while in its old range in South Bulgaria it is also found at significantly higher altitudes. The most likely explanation is that this is due to the harsher climate of Central Bulgaria.

Key words: *Thaumetopoea pityocampa*, expansion, rate of spread, pine plantations, Bulgaria

The flight of the Pine processionary moth (*Thaumetopoea pityocampa*) in the Valey of Mesta, Bulgaria

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Abstract: In 2017 and 2018, the flight of *Thaumetopoea pityocampa* in the region of the town Gotse Delchev was traced with pheromone traps. It was found that during the cooler summer of 2018, the flight of the pine processionary moth (PPM) in the area under investigation was weaker. In both years, the flight began a week or two after the summer solstice on June 22 and ended on September 10. The active flight of PPM took place during the hottest time of the year – July and August, and the peak of the flight preceded with a few days the annual culmination of the temperatures in the region.

From the determined flight times it is possible to draw the preliminary conclusion that the area is occupied by a Mediterranean phenological form of the species. In both years a tridimodal distribution of flight over time was observed. There are three two-week active flight periods with two-week breaks between them, during which only single males are captured. The most likely explanation for this fact is the presence in the vicinity of the sample area of populations with different phenological calendars.

Key words: *Thaumetopoea pityocampa*, phenology, ecology, Bulgaria

Impact of egg parasitoids on Pine processionary moth (*Thaumetopoea pityocampa*) in a new habitat in Bulgaria

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Absract: In 2010, the pine processionary moth (*Thaumetopoea pityocampa*) appeared for first time in the land of Vetrino vill. on the territory of State Forest Enterprise Nevestino in South-Western Bulgaria. Studies on egg parasitoids were carried out in 2013-2016 in the new habitat of *T. pityocampa*. Egg batches from 2013, 2014 and 2016 host's generations were collected in *Pinus sylvestris* stands. They were transported to the Laboratory of entomology of Forest Research Institute in Sofia. The egg-batches were put singly in test tubes covered with cotton stoppers and kept at room temperature (20 – 22°C). As a result, four primary parasitoids (*Ooencyrtus pityocampae*, *Baryscapus servadeii*, *Anastatus bifasciatus*, *Trichogramma* sp.) and the hyperparasitoid *Baryscapus transversalis* were established. The most numerous was *O. pityocampae*, but its relative share gradually decreased in studied years from 87.9% (2013) to 70.0% (2014) and 23.5% (2016). In the individual batches, a wide range of combinations between the parasitoids was observed. All emerged specimens of *O. pityocampae* and *B. servadeii* were female. In *A. bifasciatus* the emerged specimens were male, and in *B. transversalis* – of both sexes, at a ratio of female to male individuals of 2:1. The longest emergence period in laboratory conditions was found in *O. pityocampae*, which reached 178 days, ending in July – August. A significant impact (17.3 – 23.0%) of the parasitoids on T. *pityocampa* numbers was established.

DNA damage in sperm cells – biomarker for genotoxicity in striped field mouse (*Apodemus agrarius* Pallas, 1771) from rice fields

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Abstract: Comet assay is widely used in human biomonitoring and genotoxicity testing with plants and animals. While its application in somatic cells gives an idea of a short-term DNA damage (single-, double-stranded brakes and alkali-labile sites that can be repaired), the study of male germ cells evaluates the long-term effects that can affect offspring as a result of DNA breaks which cannot be repaired.

The aim of this study is to assess the degree of DNA damage in sperm of striped field mouse (*Apodemus agrarius* Pallas, 1771) from an area with anthropogenic pollution – rice fields. A total of 35 male individuals were investigated – 20 from the impact area of the rice fields and 15 individuals from the control region of Strandzha Nature Park. The neutral version of sperm comet assay was applied. The investigated comet parameters (% tail DNA – TDNA, tail length – TL, tail moment – TM and Olive tail moment – OTM) in mice from rice fields were statistically significantly higher than those of the background individuals (%TDNA, TM and OTM at $p \leq 0.05$; TL at $p \leq 0.001$).

The observed comet parameters show that the DNA damage in sperm cells of *A. agrarius*, reflects the genetic damage that occurred during spermatogenesis in mice inhabiting the rice fields. Significant differences with the control region are an indication of genotoxic agents presence in the impacted area. The results obtained are evidence of the

successful application of DNA damage in sperm as a biomarker in biomonitoring studies.

Key words: sperm comet assay, DNA damage, *Apodemus agrarius*, rice fields

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Honey bee colony losses in Bulgaria during the period 2017 – 2019

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Absract: The study presents the regional differences in rate of honey bee colony losses in Bulgaria during the period 2017 – 2019. The survey was carried out amongst members of the National Bee Breeding Association, owners of 68 apiaries (more than 7 300 bee colonies). By usage of a standardized in 2017 COLOSS questionnaire data collected by beekeepers from 16 administrative and four geographic regions in Bulgaria have been analyzed concerning the rate of colony losses obtained and the reported reasons for established mortality of honey bees.

Information regarding agrochemicals used against Varroa, the melliferous plants available in the regions studied and pesticides used in agriculture that have caused acute mortality in honey bee colonies is collected and analyzed. The rate of the bee colonies lost because of unsolvable queen problems, natural disaster or other reasons for mortality has been calculated. Totally about 3 to 5% of honey bee colonies included in the survey have been lost during the period 2017 – 2019. Among the melliferous flora there have been reported the presence of orchards, oilseed rape, maize, sunflower, heather and autumn forage crops most of which have been treated by different pesticides, including some neonicotinoid agrochemicals.

Key words: *Apis mellifera*, colony losses, reasons for mortality

Acknowledgments: This study was supported by the Research Fund of the Plovdiv University “Paisii Hilendarski” through the Contract № MU19-BF-005.

The parasitoid complex associated with *Larinus sibiricus* Gyllenhal, 1835 (Coleoptera: Curculionidae) in Bulgaria

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Absract: A total of 119 weevils *Larinus sibiricus* Gyllenhal, 1835 were reared from flowerheads of *Xeranthemum annuum* L. (Asteraceae) in Bulgaria. A rich complex of hymenopteran parasitoids associated with the weevil was reared and comments on it are presented. As a result, the following parasitoids were established: *Bracon urinator* (Fabricius, 1798) (Braconidae), *Exeristes roborator* (Fabricius, 1793) (Ichneumonidae), *Cyrtoptyx* sp., *Norbanus cerasiops* (Masi, 1922), *Pteromalus vibulenus* (Walker, 1839), *Stenoselma nigrum* Delucchi, 1956 (Pteromalidae), *Exopristus trigonomerus* (Masi, 1916) (Torymidae), *Leptomastix ephyra* Noyes & Hayat, 1994 (Encyrtidae), *Aprostocetus venustus* (Gahan, 1914), *Baryscapus* sp. near *carthami* (Eulophidae), *Eupelmus microzonus* Förster, 1860 (Eupelmidae) and *Eurytoma curculionum* Mayr, 1878 (Eurytomidae). Four of them (*B.* sp. near *carthami*, *E. curculionum*, *L. ephyra* and *N. cerasiops*) were recorded for the first time in Bulgaria and *L. ephyra* is a new record for the Balkan Peninsula and Europe. New host and plant associates were established.

Key words: *Larinus sibiricus*, *Xeranthemum annuum*, parasitoids, new records, Bulgaria, new host and plant associates

Atypical winter activity of fungus gnats (Diptera: Mycetophilidae) in critically endangered Mediterranean and sub-Mediterranean type habitats in Bulgaria

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Absract: Adult fungus gnats are typical inhabitants of moderately humid forest habitats, rare in drier forests, but also avoiding too wet habitats. In European temperate climatic conditions they are active mainly during the period from April to September. However, in the most Southwestern regions of Bulgaria with habitats of Mediterranean and sub-Mediterranean type, it turns out that the conditions during the winter of 2002 – 2003 have been established extremely high abundance of fungus gnats in the area. The study was conducted through tree and soil traps. The results show between 3000 and 10000 specimens per each winter month, as several species from genus Phronia, Coleosia and Boletina are with predominant abundance. Stigmatomeria crassicornis is reported for first time in Bulgarian fauna.

Key words: fungus gnats, winter activitiy, Bulgaria

Fish diversity and distribution in the Ceyhan River Estuary (Adana-Turkey)

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Absract: Seasonal changes in density, species composition, and life-style categories of the fish assemblage in a shallow Mediterranean estuary of southeastern Anatolia are described. Fishes were sampled seasonally by different nets in Ceyhan River estuary from December 2014 and June 2017 (11 sampling events). Water quality parameters were measured at each survey site at the start of each field. In total, 5254 individuals from 28 species were captured. *Liza aurata*, *Liza ramada* and *Mugil cephalus* were the dominant species and comprised over 60% of the total number. The most abundant order was Perciformes with 25% of all species. Mugilidae was the most dominant family contributing 6 species in 3 genera. The fish fauna comprised 14 marine, 11 freshwater, 2 brackish and 1 migrant species. The index of species richness, Shannon-wiener diversity, and evenness each showed a consistent seasonal pattern. This study also provides the information about environmental parameters.

Key words: fish assemblage, salinity, estuary, mediterranean

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Seasonal changes in density and diversity of fishes in the Seyhan River Estuary (Mersin-Turkey)

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Absract: The present study investigated the seasonal changes in the fish assemblage composition in relation to density and species number in a shallow Mediterranean estuary of southeastern Anatolia are described. Fishes were sampled seasonally by different nets in main channel of Seyhan River estuary from December 2014 and June 2017 (11 sampling events). In total, 6950 individuals from 30 species were captured. *Liza aurata, Liza saliens* and *Gambusia holbrooki* were the dominant species and comprised over 60% of the total number. Mugilidae was the most dominant family contributing 6 species in 3 genera. The fish fauna comprised 20 marine, 7 freshwater, 2 migrant and 1 brackish species. Fish community structure estimated by the ecological diversity indices showed variations between stations and seasons. This study also provides the information about environmental parameters.

Key words: fish assemblage, salinity, estuary, seyhan, Mediterranean

Acknowledgements: This research was financially supported by the TÜBİTAK (Scientific and Technological Research Council of Turkey) under the Project numbered KBAG, 114 Z 259.

Land-management subsidies for shrub removal are a major threat for herpetofauna in Bulgaria

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Absract: Since 2007 Bulgarian farmers have new financial opportunities for land management provided through the Common Agricultural Policy (CAP). Although some CAP and Agri-Environment Schemes (AES) goals are to support biodiversity, their implementation may result in significant loss of habitats suitable for herpetofauna (e.g. 9% decrease of open grasslands' area in four years).

We studied the impact of land management in grasslands on the herpetofauna in three Natura 2000 sites (S and SE Bulgaria) between 2016 and 2017. Twenty-six representative plots with five types of management (from animal grazing to mulching) were selected. Using satellite imagery we evaluated the percent of shrub cover per plot. We found 7 amphibian species (33% of all Bulgarian species, 49 observations) and 15 reptile species (44% of all species, 845 observations). A PCA analysis demonstrated that semi-open grasslands with 20 – 60% cover of shrub vegetation supported the highest herpetofaunal abundance and diversity. However, CAP subsidies for open pastures currently cause substantial loss of shrub vegetation (especially of important plant communities with Paliurus spina-christi) both in the studied areas and nationally. Although we observed extensive direct mortality (e.g. eight *Testudo hermanni* in ca. 2 ha), rapid loss of suitable microhabitats (e.g. for refugia, thermoregulation) is an even great-

er threat. Major drawbacks of the CAP and AES represent criteria for direct payments inappropriate to the country's natural conditions and a lack of a procedure for assessing the impacts of agricultural activities on biodiversity. Thus, currently CAP and AES in Bulgaria fail in supporting biodiversity and herpetofaunal conservation.

Pilot line transect surveys of cetaceans in a Bulgarian MPA – BG0001007 Strandzha SCI

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Abstract: All three Black Sea cetacean species are nominated as endemic subspecies for the basin listed in the IUCN Red List of Endangered species: Black Sea Harbour porpoise (*Phocoena phocoena relicta*) and Black Sea Bottlenose dolphin (*Tursiops truncatus ponticus*) as Endangered (EN) while Black Sea Common dolphin (*Delphinus delphis ponticus*) as Vulnerable (VU). First two species are listed in Annex II of EU Habitats Directive 92/43/EEC meaning member-states must protect their key habitats by designating that as sites of Community importance (SCIs) included in the Natura 2000 network. The paper presents the first effort for estimation of cetacean distribution, density and abundance in marine area of Strandzha SCI BG0001007 comprising total area of 376 sq. km. Data was collected during two line transect distance sampling surveys in April and May 2016 using a motor sailing yacht as platform. In May all three Black Sea cetacean species have been observed, while in April only Harbour porpoise and Bottlenose dolphin have been detected. In both surveys most abundant was found to be the Black Sea Harbour porpoise allowing estimation of density and abundance for that species for both months. Encounter rates during the surveys were as follows: Harbour porpoise – 0.25 in April and 0.14 in May; Bottlenose dolphin – 0.1 in April and 0.02 in May; Common dolphin – 0.02 in May.

Key words: Black Sea cetaceans, *Phocoena phocoena relicta*, *Delphinus delphis ponticus*, *Tursiops truncatus ponticus*, distance-sampling, abundance

EPIDEMIOLOGY

Communicable diseases in the human evolution

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Abstract: The humans are a manageable but no managing factors of evolution. The structure of human population is less as the structure of nature at all. The variability of human species forms under the effect of inter-species struggle of the agents of diseases. The infection may to be determining as main selection pressing on the human beings follows to polymorphism. The infection diseases contributed for genetic selection of europeids. The contemporary population in largely are a result of the effect of tuberculosis, malaria, plague. Under impact of selective pressing HLA-system has become the most polymorphous of all of the human systems. Through the systems "quorum sensing" and "cross-talk" the microorganisms complicated even more this process. These facts allowed to be considered the pathogenic microorganisms of the most powerful factor in natural selection, which actively influences on the evolution of humans. Perception of infectious pathology as a result of inter-species struggle of human beings and microorganisms, stimulates a new approaches to treatment and prophylaxis.

Key words: infectious diseases, evolution of humans

Epidemiological forecasting – biosocial nature and methodology

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Abstract: ***Background***. The epidemiological forecasting facilitates scientifically sound solutions to upcoming theoretical and practical issues, in development of public health management, in particular of infectious diseases. Combination of various epidemiological and mathematical approaches is in favor of building a science-based forecasting and organizing prevention measures, reducing human losses and economic damage.

Aim. To critically analyze the most recent scientific advances in the biosocial nature and methodology of epidemiological forecasting in order to present a real-life example of pertussis, a disease with shifting epidemiology.

Materials and methods. We performed systematic literature search of sources containing information on epidemiological forecasting with emphasis on ARIMA modeling methods – a high-precision short-term prediction model. To model pertussis morbidity we obtained official data from the Ministry of Health and the National Center for Infectious and Parasitic Diseases, since the beginning of disease registration in 1921 until 2017. We also analyzed the shifting epidemiology of pertussis.

Results. Historically, epidemiological forecasting is based on the complex biosocial nature of the epidemic examined for many years. Mathematical and statistical modeling methods have long remained under developed and applied, even underestimated and defined as "in-

fatuation". Hereafter, we are targeting the definition of categories: epidemiological forecast models – in particular ARIMA, as available and scientifically validated forecasting approach that accurately short-term forecasted pertussis morbidity.

Conclusion. The application of short-term forecasting models is aiding decision-making for rational allocation of health resources and favors successful planning of prevention and control of infectious disease.

Key words: short-term epidemiological forecasting, pertussis, ARIMA method

Contemporary aspects of the epidemiological surveillance of socially significant tick-borneinfections (TBIs) in Varna region

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Abstract: The aim of this article is to trace the evolution in parameters of the parasitic system in Lyme Borreliosis, Boutonneuse fever, Crimean-Congo hemorrhagic fever, and Q-fever in the Varna region for the period 2001-2017 and the activities of the epidemiological surveillance system.

Data from active epidemiological studies and analyses of TBIs in the region of Varna and Bulgaria for the period 2001-2017 were used; as well as epidemiological and entomological methods, statistical and graphic analyses, nosogeographic analyses and others.Data from the epidemiological analysis determined the continuing health and social significance of registered TBIs in the Varna region during the considered period. High levels of morbidity in Lyme disease (LD) and Boutonneuse fever (BF) with characteristic summer seasonality, correlated with the activity of vectors – reservoirs (*Ixodes* ticks), have been established. The system of epidemiological surveillance includes events for effective dispensary monitoring of the bitten by a tick; collecting complete epidemiological information about the area where the bite has occurred; preparation of epidemiological analysis of the morbidity of transmissible infections on the territory of the Varna region with emphasis on Lyme borreliosis, Boutonneuse fever, Q fever, Crimean-Congo hemorrhagic fever (CCHF), surveillance over the species composition and territorial distribution of tick populations and expanding the tick control activities in the region. There is a timely diagnosis and

treatment of the registered patients. It is necessary to expand and deepen the system of epidemiological surveillance with the proposed strategy for surveillance and control of socially significant TBIs in the Black Sea region of the Republic of Bulgaria.

Key words: epidemiological surveillance, tick-borne infections, Varna region

Epidemiological aspects of anthrax in North-East Bulgaria

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Abstract: ***Introduction***. Northeast Bulgaria is a territory whose climatic-geographical landscape and economic features predispose to the spread of anthrax. From the retrospective epizootological analysis it can be stated that in the 1950s anthrax affected villages in Bulgaria were 2253 (42.5% of all settlements). In the subsequent period the number of stationary anthrax foci with manifested activity decreased by more than 5 times. In all periods, the most intensive is the epizootic situation in Dobrich, Razgrad, Shumen, Targoviste and Yambol regions.

Goal and tasks. To study the epidemiological characteristics of anthrax in Northeast Bulgaria for the period 1985 – 2018. To study the registered anthrax diseases in Varna region for the period 1985 – 2018. To track the clinical characteristics of anthrax diseases in humans in 2015 and their outcome. To evaluate the general preventive, anti-epidemic, epizootic measures carried out during the different periods of the study.

Materials and Methods. We used data from our own epidemiological studies, registration data and case studies of anthrax from the RHI and veterinary services, data from registration journals and medical history of patients hospitalized at the Infectious Clinic, geographical maps, meteorological data; We used the following methods: documentary, epidemiological retrospective and prospective study, epidemiological and cartographic analysis.

Results. From the analysis for the period 1985 – 2018. we find that the most affected by anthrax settlements are in Shumen, Silistra, Varna

and Targovishte regions. The most diseased animals are in Varna and Shumen regions. In terms of the number of sick people, the Silistra region comes first.

Conclusion. The anthrax problem existed in North-East Bulgaria. Although cases of anthrax in humans and animals have declined significantly over the last decade, with certain influence of the climatic and socio-economic factors in the anthrax foci activity can be expected.

Key words: anthrax, focus, epizootic, epidemiology

Legionella infections in Bulgaria

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Abstract: The belief that Bulgaria is an island in Europe, spared from the "exotic" legionella infections is still present in our country (including among the medical community). At the background of data from the European Surveillance Network for Legionnaires' Disease (LD), we will present an epidemiological analysis of 150 cases of Legionella infections related to Bulgaria. Particular attention will be paid to:

- LD in Bulgarian citizens
- LD clusters and outbreaks in Bulgaria
- Problems in LD diagnosis
- Problems in the prevention and investigation of LD cases

Obviously the LD cases presented here are just the tip of the iceberg. Despite of the gaps in LD diagnosis and/or reporting and thanks to the lack of LD preventive measures in Bulgaria, the disease is here – on our seemingly quiet island.

Key words: epidemiology, legionella, Legionnaires' Disease

Zoonotic potential of *Helicobacter* spp.

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Abstract: The genus *Helicobacter* now includes more than 35 formally named species. *Helicobacter pylori* is the best known and the most important in terms of global impact on human disease. Over 50% of the world population is infected by this bacteria. *Helicobacter pylori* is the main cause of gastritis, peptic ulcers, and gastric cancer. Some of the species may also have zoonotic potential. *H. heilmannii* and H. *felis* are associated with gastritis in a variety of animals, including humans. Some of these non-pylori helicobacters (NPHS) – *H. canis, H. pullorum, H. cinaedi, H. fennelliae, H. canadensis, H. winghamensis, H. westmeadi*, and *H. rappini*, have been isolated from diarrhoeic and/or bacteraemic humans. *H. pullorum* has been isolated from humans and poultry, *H. canis* from dogs, cats, and humans, *H. cinaedi* from humans, non-human primates, dogs, and hamsters, and "H. rappini" from dogs, cats, mice, humans, and non-human primates. The findings concerning a potential zoonotic pathway for the transmission of *Helicobacter* spp, the ecology and epidemiology of this genus, deserve more attention to these emerging pathogens. In our study, in Bulgaria, the zoonotic potential was confirmed indirectly. We investigated the risk factors of acquisition and modes of transmission of *H.pylori* infection. The place of residence in childhood and the parent's place of living – village; consumption of heat unprocessed vegetables; the presence of domestic animals; the contact with pets; the consumption of uncooked (unpasteurized) milk (via just milking animal) in childhood; the presence of rodents near or in the houses (the

parameters of the impact of animals as a risk factor) – were significantly associated and strong predictors of *H. pylori* seropositivity in Bulgaria. It is necessary to prove this causal connection by long-term cohort studies.

Key words: *Helicobacter pylori*, *Helicobacter* spp., Zoonosis, chronic gastritis, peptic ulcer disease

Basic principles and approved decontamination method against biological warfare agents

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Abstract: ***Summary***. Disinfection, in the presence of biological warfare (BW) agents, is a standard procedure in the common set of biological defence measures. This activity must comply with the national legislation related to the prevention of infectious diseases and to the methodological principles and current requirements for decontamination. ***Objective***. To present the main methods and means for disinfection of contaminated with BW agents environment and to review the legal acts regulating this activity in the country. ***Methods***. A documentary research method was used for the review. National legislation was discussed in the part related to the defence of the population from BW agents. The same documentary research method was used in preparing an overview on methods and means for biological defence. ***Results***. The highlights of the national legislation with which the organization and conduction of disinfection in a BW attack area should be considered were outlined. The principles of disinfection treatments were systematized. The peculiarities of aerosol application of BW agents were considered especially when contaminating food and water supplies.

Key words: biological defence, biological agents, disinfection, methods and means

Implementation of Medical standard on prevention and control of nosocomial infections: Conclusions based on questionnaire surveys of BAPIC BulNoso, 2012 – 2018

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Abstract: In the 9-year period since the introduction of Medical standard on prevention and control of nosocomial infections the BulNoso experts, part of the Standard authors, organized and carried out several questionnaire surveys aiming to gather information on its implementation in hospitals. In this report we present the conclusions derived from 2012 – 2018 studies. The achievements and problems met during the process of implementation of Standard key recommendations are analyzed. The successive stages indicators of the recommendations implementation are compared. It is established that a significant part of the hospitals still do not follow important rules and requirements both concerning the patient care and personnel protection. The average values of one of the most important indicators – the consumption of alcohol-based hand disinfectants does not show a positive trend and stay substantially behind the similar European data (6-7 procedures for a patient-day against 2 for Bulgaria). The average level of hepatitis B vaccine staff coverage does not differ significantly from the level established in our previous studies (71% in doctors, 61% in nurses and 56% in hospital attendants). The necessary conclusion is that the unfavorable and highly variable results correlate with shortage in IC specialists qualification.

Key words: Medical standard, prevention and control, nosocomial infections, implementation, hospitals

The pressure of vaccines on microorganisms and their response

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Abstract: Diseases that have killed thousands of people have been eliminated today or are in a demarcation stage, due precisely to the effectiveness of vaccines. By defending themselves, people defend indirectly and others by forming herd immunity. In parallel, there is a cross-reaction between the pathogenic microorganisms not included in the vaccine and the vaccine strains. The reaction of microorganisms against vaccine pressure is the change in isolate strains or emergence of new diseases. The application of vaccines has a number of advantages over the indiscriminate administration of antibiotics, which should be used only when necessary.

Key words: vaccine, pressure on microorganisms, antibiotics

Measles in Europe and in Bulgaria during the 2-nd decade of 21 century

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Abstract: ***Introduction.*** In 2010 the World Health Organization (WHO) has declared that" the global measles eradication is biologically, technically and operatively possible". Although most countries in the European region reached the status of measles elimination, sporadic cases or epidemic outbreaks have been reported. Intensive movement of people to and from measles endemic countries, creates a risk for reestablishment of virus spread in places where vaccination coverage of measles vaccine is inadequate.

Scope. To present the spread of measles in the European region(ER) of WHO and in Bulgaria, as well as the challenges towards measles elimination.

Materials and methods. The epidemiological and laboratory surveillance data of measles in the ER of WHO and in Bulgaria, presented within the last decade, have been reviewed and analyzed. The annual reports (WHO, 2013-2018) issued by the European Regional Verification Commission for Measles and Rubella Elimination have also been reviewed.

Results. Based on the data for 2017, 37 countries in the ER of WHO have provided evidence to demonstrate the elimination of endemic measles, 6 have provided evidence for the interruption of measles transmission for a period of 12(1 country) or 24(5 countries) months, and 10 countries were considered endemic for measles in 2017. Three measles epidemic outbreaks, each with different dimension and burden, arose in Bulgaria during the last decade (2009 – 2010, 2017 and 2019). Measles elimination achieved in the country between 2015 –

2018, illustrates the benefits of the non- epidemic period welfare, maintenance of which is possible through improvement of vaccination coverage against measles and adequate epidemiological surveillance.

Conclusion: Measles elimination in Europe will possibly remain a goal beyond 2020.

Key words: measles, elimination, surveillance

A review of global good practices to improve communication between pediatricians and parents on the topic of vaccines

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Abstract: ***Introduction***.Vaccine hesitancy among parents is one of the current challenges in public health. It is fueled by various processes. An important factor in recent years is the online distribution of misleading information about risks, associated with vaccination. Parents still perceive pediatricians to be a chief source of information among the various available information sources. Both pediatricians and other health workers need to meet the challenge of combining their routine work with well targeted effective activities, aimed at providing evidence-based information regarding vaccines to parents.

Aim. The aim of this review is to summarize global good practices for the optimization of communication between pediatricians and parents on the topic of vaccines. The study is part of the project "Childhood immunizations: a challenge to contemporary Bulgarian society", funded by the National science fund (NSF) under Contract № КП-06-ОПР03/15 from 19/12/2018 between NSF and the National Center of Infectious and Parasitic Diseases.

Materials and methods. The review consists of two parts: (1) A summary of guidelines related to communication on the topic of vaccines, published by national institutes of European Union and European Economic Area member states and international expert organizations; (2) A systematic review of publications, indexed in Scopus, which

provide specific examples of measures, taken to improve the communication on the topic of vaccines between pediatricians and parents.

Results and conclusion. The guidelines and specific examples identified deal with varied approaches to communication and organizational arrangements, facilitating more efficient sharing of information with parents. The applicability and effectiveness of measures to improve communication depend on the local context.

Key words: immunization, communication

Vaccine-preventable diseases in HIV (+) persons

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Abstract: ***Introduction.*** The risk for acquiring of infections in HIV(+) persons is significantly higher in comparison with HIV(-). That's why measures for their prevention have to be undertaken. The medical practice has effective instrument for this prevention, concerning the infections against which there are effective vaccines. ***The aim*** is to be analyzed the possibilities for application of vaccines to HIV-infected. Materials and methods: The recommendations of WHO, ECDC and CDC for the possibilities to be applied vaccines to HIV(+) have been analyzed, also these vaccines which have not to be applied. ***Results***. HIV(+) are with 4 – 8 times higher risk to be infected with influenza and 1.5 time greater risk to die. They are 35 – 100 times more risk group to acquire invasive pneumococcal disease and 5 – 13 times – for invasive meningococcal disease. The risk of infection with pertussis, hepatitis A, B, C and human papilloma virus is significantly higher for them. That's why it is recommended to HIV(+) persons to be applied the vaccines for which they have not contraindications. To HIV-infected may be applied inactivated, polysaccharide, recombinant vaccines and toxoids. Passive immune prophylaxis may also be applied to HIV(+): with normal human immune globulin and specific immune globulins against hepatitis B, rabies and varicella-zoster virus. Live vaccines have not to be applied to HIV-infected. ***Conclusion.*** As soon as possible after the diagnose HIV infection is confirmed, we have to check the immune status of this person for the routine immunizations. If there are lacks – to be done the necessary immunizations. And then it is appropriate to be done the recommended vaccines.

Key words: HIV(+) persons, vaccine-preventable diseases, immune prophylaxis

Chemical agents suitable for treatment of incubators. Disinfection of incubators – multicentre questionnaire study, 2019

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Abstract: ***Summary***. The register of authorized biocides includes 18 products for disinfection of incubators in the medical area. The method of decontamination of incubators is an element of the biocide authorization. The documents submitted to the Ministry of Health are difficult to evaluate in the part related to the use of disinfectants for incubators. There is no regulatory act describing the methods and chemical agents suitable for decontamination of this type of specific medical devices. ***Objective***. To establish the current practices for the decontamination of incubators. ***Methods***. A multicentre questionnaire survey was performed to analyze cleaning and disinfection practices for incubators in hospitals. Responses were received from 88 hospitals in the country with neonatal units. ***Results***. The results show different treatment methods and the need to develop instructions or guidance for decontamination of incubators to assist the staff in neonatal units.

Key words: incubators, disinfection, decontamination, chemical agents

Efficacy and toxicological profile of repellents with IR3535 and DEET

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Abstract: ***Introduction***. Some of the measures for prevention of vector-borne infections include reduction of the vector population density and personal protection against hematophagous arthropods. The use of repellents is recommended as a personal protective measure.

Aim. To prepare an updated review of the range of products authorised for use as repellents in Bulgaria with focus on their efficacy and toxicological profile. Data on two of the most commonly used active substances of repellents (IR3535 and DEET), as well as the main recommendations for safe application will be presented in this report.

Materials and methods. During the last 10 years (2008-2018) 20 products with different concentrations and different forms of application were studied under laboratory conditions. The products were tested against the mosquito *Aedes aegypti*, laboratory strain. The repellents were evaluated regarding complete protection time on human skin. Based on toxicological assessment within the EU Review Programme for active substances of biocides and on exposure and risk assessment, a number of recommendations and restrictions have been introduced for the use of repellents.

Results. The repellents (ready-to-use) were tested at concentrations 5%, 7%, 7.5%, 8.8%, 10%, 15% and 20%. The repellents with DEET at concentrations between 5% and 20% provided protection time lasting from 1 to 8 hours against mosquitoes. The repellents with IR3535 at concentrations between 10% and 20% provided protection from 3 to 8 hours against mosquitoes.

***Conclusion*.** In order to reduce dermal exposure to acceptable levels, it is advisable to give preference to repellents with the lowest content of active substances, which provide sufficiently effective and prolonged protection. When choosing repellents, it is necessary to estimate the specific situation, the time that the people spend outdoors and the level of the risk of being bitten by infected insects.

Key words: repellent, IR3535, DEET

Epidemiological assessment – a basic element of medical support planning of military operations

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Abstract: ***Introduction***. War accompanies human development from its earliest stages. The weapons, the tactical and operational activities, the combat units, equipment, planning and support of the military contingents and operations have changed and improved through the ages. From the down of military art, the role of the biological factor (the usage of biological weapons, morbidity among own troops, local population, the enemy) has been considered because it affects the abilities of the soldiers and the success of the campaign. Despite that, many military operations that changed the history, have been resolved by the impact of the biological factor on the troops.

Aim. The aim of the study is to evaluate the impact of the epidemiological assessment on the medical support planning for military operations.

Materials and methods. Descriptive and comparative methods are used to analyze the impact of infectious diseases on the outcome of military campaigns, that prove to be a turning point in the history of the country, the region or the continent. By the means of deductive and cluster analyses the importance of timely and accurate epidemiological assessment for the effectiveness of medical support planning is evaluated.

Results and discussion. Performed analyses highlight the profound negative impact from the disregard of the assessment of the epidemiological situation before and during military operations in the plans for medical support.

Conclusions. The obtained results determine epidemiological assessment as a basic element of military medical planning.

Key words: Epidemiological situation, Military medical support, Epidemiological assessment, Military medical planning

Study of infectivity with *Borrelia burgdorferi* sl among *Ixodes ricinus* population in Pleven region, Bulgaria

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Abstract: ***Introduction.*** Ticks-borne transmited infections occupy a significant part in human pathology. These are an infection with natural foci that can not be eradicated. The leading transmissive infection in Bulgaria is Lyme borreliosis, the cause of which is spirochetes from the *Borrelia burgdorferi* sl complex, distributed by ticks of the genus Ixodes.

Aim. To establish areas in the Pleven region with a high density of ticks of the species *Ixodes ricinus* and their contamination with *Borrelia burgdorferi* sl.

Material and methods. The study was conducted in 5 localities in the Pleven region for a period of four years (2016 – 2019). All ticks are collected in the phase of questing from vegetation by flag. For each collection, common phenology data (temperature, relative humidity, time, area of flags, and geographic coordinates) were measured. Identification of the ticks has been performed to species, sex and stage of development. Some of the collected *Ixodes ricinus* ticks were observed by dark-field microscopy for the presence of spirochetes from the *Borrelia burgdorferi* sl complex.

Results. Of ticks collected for four years (n = 937), 765 were categorized as *Ixodes ricinus*. Of these, 391 were examined by darkfield microscopy. Spirochetes from B.*burgdorferi* sl. were found in 130 of the observed.

***Conclusion*.** Five new areas of high population density of ticks of the species *Ixodes ricinus* have been established, as well as the presence of *B.burgorferisl* in their intestinal contents.

Key words: *Borrelia burgdorferi* sl, *Ixodes ricinus*, Pleven region, Lyme borreliosis

Research of the Wild pigs infected with the parasite *Trichinella* in the Blagoevgrad district for the period from 01.01.2018. to 01.01.2019

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Abstract: Trichinellosis is widespread zooanthroponosis in Bulgaria, often transmitted to humans by eating undercooked meats and local game meat products.

A survey was carried out on the *Trichinella* infestation of the wild pigs shot for the period 01.01.2018 to 01.01.2019 in the Blagoevgrad region. Samples of 2764 pigs were taken, of which 36 samples were positive (1.3% infestation). *Trichinella britovi* has been demonstrated in 80% of the samples and 20% in *Trichinella spiralis*. Samples are taken from the diaphragm (about 10 grams). The trichinoscopic and enzyme method was used for the studies.

This study proves that in the Blagoevgrad region the trichinosis in wild pigs is relatively high, which is a risk of contamination mostly in hunters and the spreading of disease among the people.

Key words: *Trihinella britovi*, *Trihinella spiralis*, Trichinellosis, Infestation, Wild pigs

Epidemiological evaluation of *L. monocytogenes* circulation

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Abstract: Listeriosis is an acute infectious disease that is rarely diagnosed and presents a serious medical and social problem. It is severely on going and has a high lethal rate especially for newborns and the elderly. It is caused by *Listeria monocytogenes*, a microorganism with wide adaptive capabilities. The ecological aspects of the circulation of this pathogen have not been enough studied. There were thorough studies about spreading of *L. monocytogenes* and an assessment of the ecological and epidemiological characteristics of contamination of various food and environmental objects. The features of the saprophytic distribution cycle of the infectious agent are discussed. We suggest a spatial model of the circulation of *Listeria monocytogenes*.

Key words: Listeriosis, epidemiological evaluation

POSTER SESSION

P1

New and additional records of earthworms (Annelida: Clitellata) from Kopaonik Mountain: First finding of *Allolobophora treskavicensis* (Mršić, 1991) in Serbia

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Abstract: The aim of this paper is to present new data on the earthworms in the study area situated in Kopaonik Mountain, Serbia. The Kopaonik Mt. is a part of the Dinaric mountain chain situated in the central part of southern Serbia. By analyzing the newly reported species together with literature records, we establish the list of known earthworm taxa. from family Lumbricidae. The list underlines earthworm diversity and provides a general overview of their distributions and zoogeographical positions. The list comprises 25 taxa, belonging to 10 genera of the family Lumbricidae. The genera with the largest number of the registered taxa are *Aporrectodea* (5) and *Dendrobaena* (5), while the genera *Bimastos*, *Eiseniella*, *Octolasion* and *Perelia* are represented by one taxon. There are literature data for 3 taxa that have not been found by our field research. With respect to the zoogeographic situation, the majority of them belong to peregrine species (11). The endemic species represent with 3 taxa and belong to the genus *Allolobophora*. It is interesting that endemic species *Allolobophora treskavicensis* (Mršić, 1991) proved to be new species in the fauna of Serbia, including the study area. So far, *All. treskavicensis* has been found only on one locality in Macedonia. Therefore, for now, the new sites from Kopaonik Mt. represent the northernmost limit of the species' natural range.

Key words: earthworm, Kopaonik Mountain, Serbia, zoogeography distribution

P2

Effects of a modern biorational insecticide spinosad on earthworm, *Eisenia fetida* (Savigny 1826)

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Abstract: This is report about effects of spinosad on the life-cycle parameters (survival, growth, cocoon production and hatching juveniles) of the non-target organism, earthworm *Eisenia fetida*. We used to artificial soil supplemented with different concentrations of the examined insecticide based on the recommended agricultural doses (RAD). The laboratory test was conducted according to the OECD guidelines. Our results show that insecticide spinosad has not an impact on mortality. However, for four weeks assessment the insecticide effects on weight were significant in 4×RAD. For six weeks, statistical significance exists in RAD, 2×RAD and 4×RAD. After eight weeks, statistical analyses have shown a significant difference in all concentrations, except in the lowest concentration (1/4 RAD), compared with the control. Negative growth inhibition was observed in the 1/4 RAD, 1/2 RAD and RAD during the four weeks experiment. Positive growth inhibition was observed in all other weeks and concentrations. The results of cocoon production and hatching juveniles showed no significant difference between the control and the treatments. This study showed that although spinosad had no impact on the most important sub-lethal effect (reproduction), had an impact on the weight after a long-time exposure, even in a concentration that is less than recommended.

Key words: Growth, earthworm, mortality, reproduction, spinosad

P3

The first detection of *Toxoplasma gondii* in rivers in Serbia

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Abstract: Infecting all warm blooded animals including humans, *Toxoplasma gondii* is one of the most successful parasites in the world. Acute maternal infection in pregnancy can result in congenital infection, and reactivated infection can be life-threatening in immunocompromised patients. The parasite life cycle includes three distinct infectious life stages, of which the oocyst, shed by Felidae as the definite host, is environmentally resistant, even in water. During the past decades several waterborne epidemics of toxoplasmosis worldwide emphasized the importance of water as a route of *T. gondii* infection. In Serbia, there are virtually no data on the presence of *T. gondii* in any type of water. We thus commenced a study in which surface water samples were collected from three major rivers and their tributaries. The samples (ā 10 L) were concentrated by filtration and total DNA was extracted from the pellet using a commercial kit, and screened for *T. gondii* DNA by amplification of the 529bp repeat fragment. Three of the first 20 samples were positive, and the specificity was confirmed by amplification of GRA6 and SAG2 genes. This is the first detection of *T. gondii* in rivers in Serbia. Importantly, the finding of 15% positive samples indicates a significant level of contamination of surface waters with *T. gondii* (oocysts), confirming water in Serbia as a potential source of infection. In addition, this ongoing study has shown that PCR can be successfully used for the detection of *T. gondii* in water samples.

P4

Morphological variability of *Lepidurus apus* (Linnaeus, 1758) (Notostraca, Crustacea) from Bosnia and Herzegovina

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Abstract: A crustacean presented here, *Lepidurus apus* (Linnaeus, 1758), is a representative of tadpole shrimps of the order Notostraca, within the class Branchiopoda. Notostracans are a group with restricted distribution in the world, but their presence can be very rich locally. A Golden Tadpole Shrimp, *L. apus*, lives in inland freshwaters, mostly the temporary ponds. Analysed sample consisted of 102 females from a highly female-biased population. Individuals were collected in the flood area near the village Bajinci in the northern part of Bosnia and Herzegovina. Descriptive statistics and regression analysis were performed in order to explore morphological variability and correlation between some traits in *L. apus*, as follows: carapax length and width, carapax area, number of posterior segments not covered with carapax, telson length, length of abdomen and cercopode, total body length, size of body segment. Results demonstrated that strong positive correlations exist between: carapax length and width ($r = 0.67$, $p < 0.0001$), length and area of carapax ($r = 0.89$, $p < 0.0001$), total length and length of carapax ($r = 0.68$, $p < 0.0001$), carapax width and area of carapax ($r = 0.93$, $p < 0.0001$), area of carapax and telson length ($r = 0.50$, $p < 0.0001$), area of carapax and total length

($r = 0.63$, $p < 0.0001$), abdomen length and size of body segment ($r = 0.79$, $p < 0.0001$), cercopode length and total length ($r = 0.81$, $p < 0.0001$). Although morphological variation has an important role in evolution, studies about morphological plasticity in notostracan crustaceans are still scarce. To the best of our knowledge, this is the first report considering morphological variability of some traits in the species *L. apus* from Bosnia and Herzegovina.

P5

Where to swim, where to eat? Laboratory report on behaviour of the ostracod *Heterocypris incongruens*

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Abstract: *Heterocypris incongruens* represents a cosmopolitan crustacean species inhabiting continental waters of temperate climate. They use the first and the second antennae and some body limbs for swimming, and to capture food particles. Our previous investigation revealed different behavioural activities and general food preferences of this species. The aim of the present study is to further follow and quantify locomotor and feeding activities of this species, in which individuals had free access to different items in the experimental conditions. In conditions that partially simulate its natural environment, we measured time that the individuals spent on two aforementioned activities. In order to measure feeding activities, four different types of food were offered: two of the plant origin and two of the animal origin. Also, some non-food items were included into experiment (neutral aquarium sand similar in colour with food). Behavioural activities of *H. incongruens* and their duration will be discussed in the context of presence or absence of food.

P6

Influence of sex ratio and wing morphology on mating success in *Drosophila melanogaster*: lessons from female and multiple choice experiments

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Abstract: *Drosophila* species express a variety of reproductive strategies. In this respect, models of sexual selection, mate preferences, and different levels of pre- and postcopulatory competition were often studied in these species. Ecological impact on sexual selection in *Drosophila* could be monitored through changes in population density and sex ratio, since they affect the dynamics of mate acquisition.

The aim of this study was to determine if differences in mating success of *Drosophila melanogaster* males is related to sex ratio. Further, in different sex ratio conditions, wing morphology was monitored, since it may contribute to variation in male courtship. Flies were allowed to mate in two types of mating assays, in female and multiple choice tests. In female choice tests, females of one strain were allowed to choose between males belonging to two strains (F:M ratio was 1:2, thus simulating competitive conditions). In multiple choice tests, flies of both sexes of two strains were present in equal sex ratio (F:M ratio was 1:1).

Results revealed differences in mating success between males belonging to different strains in competitive conditions, while no difference in mating success was observed in multiple choice tests. Using geometric morphometric analysis, differences in wing size and shape between strains and sexes were recorded in both experimental designs. Influence of sex ratio and wing morphology will be discussed in the context of sex ratio dependent sexual selection.

P7

Population-genetic heterogeneity in *Messor structor* and *Messor barbarous* populations as an approach for their discrimination

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Abstract: The genetic heterogeneity in *Messor structor* and *Messor barbarus* ant populations has been investigated on the basis of isoenzyme and soluble protein analysis. The genetic control of seven polymorphic enzyme and protein groups has been identified, suggesting appropriate markers for characterizing intra- and interpopulation genetic variability. A comparative analysis of the gene pool and genotypic structure of the studied populations has been carried out. Specific genetic markers have been identified to distinguish *Messor structor* and *Messor barbarus* species. The phylogenetic relationships among the studied populations at the intra- and interspecies levels have been analyzed. The comparative analysis of dendrograms demonstrates a clear differentiation between populations of the both species studied in the *Messor* genus.

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

P8

Hygienic behaviour and fat body development in worker bees (*Apis mellifera* L.)

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Abstract: Testing for the level of manifestation of the hygienic behaviour of 32 bee colonies from the local honey bee (*Apis mellifera* L.) from three apiaries has been carried out. Depending on the level of manifestation of the cleansing instinct, two groups of bee colonies have been formed – hygienic (these clean over 95% of the cells with dead brood up to the 48th hour after killing the brood) and non-hygienic (these clean less than 95% of the cells with dead brood up to the 48th hour). Worker bee samples have been taken from the tested be colonies to determine the level of fat body development. The fat body, in addition to being a fat and carbohydrate storage depot, is also considered to be the center of metabolism in insects. The analysis of the results from the present study revealed that the mean value of the level of fat body development in worker bees from the group of hygienic bee colonies was 2.76±0.038. In the group of nonhygienic colonies this value was 2.55±0.079 or 7.61% lower. This difference is statistically significant at ($p \leq 0.05$). The results obtained suggest that there is a correlation between the level of fat body development and the hygienic behaviour in bees.

Key words: *Apis mellifera*, hygienic behaviour, honey bees, fat body

P9

Hygienic behaviour of local honey bee (*Apis mellifera* L.) and differences in the dimensions of chitin body parts of worker bees

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Abstract: In the present study a total of 28 bee colonies from the local honey bee (*Apis mellifera* L.) have been tested for degree of manifestation of their hygienic behaviour. On the basis of the test, two groups of bee colonies have been identified – hygienic (these clean over 95% of the cells with dead brood up to the 48th hour after priercing) and non-hygienic (these clean less than 95% of the cells with dead brood up to the 48th hour). Morphometric measurements of the chitin body parts of the worker bees have been carried out on 12 features. Three of the studied morphological features (forelegs, upper jaws, and proboscis) relate to the cleaning and polishing of the beehive cells and maintaining hygiene in the bee nest. The upper jaw and the foreleg parts are new features not included in the measuring methods used in beekeeping. High degree of reliability ($p \leq 0.001$) between hygienic and non-hygienic bee colonies has been determined for 7 morphological features – length of the front wing and the first part of the cubital cell, length of thigh and width of first foot digit of the foreleg, length of proboscis and length and width of the upper jaw. For the feature number of hooks on the rear wing a low degree of reliability ($p \leq 0.05$) has been established. The results obtained in the present study concerning differences between morphometric signs of bee workers from hygienic and non hygienic bee colonies complement the information available so far on the factors influencing the manifestation of hygienic behaviour in bees.

Key words: *Apis mellifera*, hygienic behaviour, chitin body parts

P10

Ontogenetic and caste differentiation in the expression of water-soluble proteins and some isozymes in *Reticulitermes lucifugus* (Rossi, 1792) (Isoptera: Rhinotermitidae)

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Abstract: Ontogenetic and caste differentiation in the expression of water-soluble proteins, non-specific esterases, malic enzymes, lactate dehydrogenases and superoxide dismutases in the species *Reticulitermes lucifugus* (Rossi, 1792), spread out in Bulgaria has been investigated.

A total of 650 individuals have been included in the study: larvae, pseudo nymphs, nymphs, male and female imago forms – as a stage in ontogenesis and workers (pseudergates), pre-soldiers, soldiers and reproductives – secondary reproductives, neotenics and queens – as a caste in the termite nest.

A total of 41 loci – 30 for water-soluble proteins, 6 – for esterases, 2 – for malic enzymes, 2 – for lactate dehydrogenases and one – for superoxide dismutase have been analyzed. In the study on *R. lucifugus*, stage and caste specific protein and isoenzyme expressions have been established as follows: 11 of the protein loci analyzed have been characterized by a permanent activity without varying of the product expression in all the castes; for seven of the protein loci, a constant action has been found in ontogenesis but with a caste-specific variability in their expression; for four of the protein loci analyzed differential

gene regulation, expressed in gene activity in some of the castes and inactivation in the others; eight of the detected protein loci have demonstrated a caste-specific expression affecting different castes of the species; four of the sixth esterase loci have been characterized by a permanent action in the course of the ontogenesis; one of the two malic enzyme loci has been expressed only in reproductive caste (neotenics and queen) and the other one has been expressed in all stages of individual development; both lactate dehydrogenase loci have been expressed specifically in ontogenesis, depending on caste differentiation; superoxide dismutase locus has been found in all castes and stages of ontogenesis of *R. lucifugus*. The present investigation brings a new clarity about the specific expression of the protein and enzyme systems studied and could be used as a basis for future population-genetic analyzes of the species *Reticulitermes lucifugus* in Bulgaria.

Key words: *Reticulitermes lucifugus*, soluble proteins, isoenzymes, stage and caste specificity

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P11

Isoenzyme polymorphism of silkworm (*Bombyx mori* L.) breeds from germplasm resourses of Bulgaria

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Abstract: Polyacrilamide gel electrophoresis (PAGE) was used to study the isoenzyme polymorphism of nonspecific esterases (EST), malate dehydrogenase (MDH) and acid phosphatase (ACP) from haemolymph, phosphoglucomutase (PGM) and hexokinase (HK) from silk glands and alkaline phosphatase (ALP) from midgut of mulberry silkworm (*Bombyx mori* L.). Genetic variations within and among five breeds kept in Bulgaria were found out. A total of 7 polymorphic loci with 19 alleles were established. For the Spanish breed named AES 1-zebra it was found out a variability in all loci. Whereas in Bulgarian breed Hebar 2 a polymorphism only at one of the loci was established. The observed polymorphism demonstrated breeds specificity. Inter-breeds differences regarding the degree of polymorphism and heterozygosity were established, too. The degree of polymorphism ranged from 77.80% to 11.10%. Low levels of observed heterozygosity and deviations from Hardy-Weinberg equilibrium in the most of analyzed loci were found. The lowest values for polymorphism and heterozygosity were found in the Bulgarian breed Hebar 2, which was the most homogeneous while the highest values were recorded for the

Spanish breed AES 1-zebra. The dendrogram constructed with the values of genetic distance showed that Hebar 2 formed one main claster, while the rest breeds studied formed the other one. The results of the present study complement the data on the genetic heterogeneity of breeds from the silkworm genetic resources of Bulgaria and could help the breeders in the process to identify suitable parents for the breeding programs for yield improvement.

Key words: *Bombyx mori* L., isoenzyme variability, phylogenetic relationships

P12

New parasitoids of *Leucoptera sinuella* (Reutti) (Lepidoptera: Lyonetiidae) in Bulgaria

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Abstract: In 2018, a parasitoid complex of *Leucoptera sinuella* (Reutti, 1853) (Lepidoptera: Lyonetiidae) was studied in two localities (town of Banya and Voinjagovo vill.) near Stryama River in Central South Bulgaria. Larvae and pupae of *L. sinuella* were periodically collected in poplar (*Populus* spp.) plantations and studied in laboratory conditions. As a result, four hymenopteran parasitoids were established: *Baryscapus endemus* (Walker, 1839), *Chrysocharis pentheus* (Walker, 1839), *Cirrospilus pictus* (Nees, 1834), *Pediobius bruchicida* (Rondani, 1872) (Hymenoptera: Eulophidae). Three species (*B. endemus, C. pentheus* and *P. bruchicida*) were established for first time and are new records for parasitoid complex of *L. sinuella.* The most abundant was *B. endemus* (87.1%), followed by *P. bruchicida* (0.8%), C. *pictus* (0.3%) and *C. pentheus* (0.1%). The mortality of the host caused by parasitoids in different samples varied between 0 and 85.0%, with an average of 7.0%.

P13

Entomopathogenic fungi (Ascomycota, Hypocreales) as natural antagonists of the pine processionary moth, *Thaumetopoea pityocampa*, in Bulgaria

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Abstract: *Thaumetopoea pityocampa* (Lepidoptera, Thaumetopoeidae), a moth naturally distributed in pine ecosystems of Mediterranean region, is considered as the most dangerous defoliator in Bulgarian pine forests. Caterpillars of this species also represent a health hazard because their hairs, which contain an urticating protein – thaumetopoein, are responsible for painful skin irritations, rashes and, in some cases, allergic reactions in some people. Therefore, *T. pityocampa* is not only a serious forest pest, but also a public health problem. Populations of the pine processionary moth are regulated by a complex of natural enemies. However, there is only limited information about a parasitic activity of entomopathogenic fungi. Entomopathogenic fungi are natural antagonists of insects helping in control of host population and prevention of outbreaks formation. More than 700 fungal species are estimated as insect pathogens and a majority of per-

spective fungi for mass production and use in biological control are those from the Hypocreales order (Ascomycota). The goal of the present study was to identify species of entomopathogenic fungi in natural populations of *T. pityocampa* in Bulgaria. During the study, 27 caterpillars or pupae showing characteristic symptoms of mycosis were collected from two sites (Fotinovo and Kandalikavils.) in the Eastern Rhodopes and *in vitro* isolates were obtained. The cultures were microscopically identified and subsequently analysed by a sequencing study of internal transcribed region of rDNA and a partial sequence of TEF1-α gene. Altogether, two *Beauveria* species (*B. pseudobassiana* – 7 isolates and *B. varroae* – 6 isolates) and *Purpureocillium lilacinum* (14 isolates) were identified.

P14

Syspastospora parasitica, a parasite of entomopathogenic fungi *Beauveria pseudobassiana* and *B. varroae* on *Thaumetopoea pityocampa* – a new tritrophic associations in Bulgaria

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Abstract: *Thaumetopoea pityocampa* (Lepidoptera: Notodontidae) is the most harmful pest defoliator of pine forests in Bulgaria. The entomopathogenic fungi are natural antagonists of insects helping in control of host population and prevention of outbreaks formation. Species of *Beauveria* are determined as potential biological control agents of *T. pityocampa*.

In the present study, new tritrophic associations were established. The hyperparasitic fungus *Syspastospora parasitica* attacked two *Beauveria* species that had infected the larvae and pupae of *T. pityiocampa*. A number of 1502 *T. pityocampa* pupae were collected from three sites in the Eastern Rhodopes: Fotinovo vill. – 1120 pupae, Srandakvill. – 117 and Kandilkavill. – 178), 60 from Sandanski (Southwestern Bulgaria) and 27 from Klisura (Srednagora Mt.). The entomopathogenic fungi *Beauveria pseudobasiana* and *B. varroae* were isolated from 16% of *T. pityocampa* pupae. Fruit bodies (perithecia) of *S. parasitica* appeared on 47% of infected by *Beauveria* spp. pupae. Perithecia covered completely the *Beaveria mycelium* two months after their appearing and stopped the entomopathogenig fungi development.

The hyperparasitic ascomycete *S. parasitica* is known to attack Deuteromycetes infecting insects. This hyperparasite has been detected in Bulgaria on *Beauveria bassiana* on ash weevil (*Stereonychus fraxini*) and European pine sawfly (*Neodiprion sertifer*).

P15

Sensitivity to climate change of selected zooplankton species

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Abstract: Plankton fauna is a key link between primary producers and larger predators. Recently, zooplankton has been used as an indicator of global changes because it strongly influenced by climatic features. Multi-year zooplankton time series provide useful tools for examining climate-ecosystem interactions. The aim of the current study was to assess the state and long-term trends of zooplankton communities of the Western Black Sea (Bulgarian region) as a response to anthropogenic and environmental shifts. Available in IO datasets were used to extract an overall trend in zooplankton dynamics in the Western Black Sea (Bulgarian part) in a period of 1967 – 2017. Time-series data of zooplankton at Cape Galata transect were collected mainly seasonally. The standard statistic procedure was used to detect temporal changes of variable mean at one location. Prior to the series analyses we tried to extract indicator species as a function of the climate signals in terms of horizontal and vertical temperature fields. The taxa and species which were subjected to the analyses were cold water copepod species (*Calanus euxinus, Pseudocalanus elongatus*) and thermophilic once – cladocerans (*Evadne spinifera, Pseudevadne tergestina*, *Penilia avirostris*) and copepods (*Centropages ponticus)*.

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P16

Zooplankton traits vs. environmental conditions within intra-decadal scale assessment in Lake Ohrid (North Macedonia, SE Europe)

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Abstract: Multiple anthropogenic impacts, especially tourism, urbanization and fishery, could affect ecosystem integrity even in nature-protected area, as is recently observed in Ohrid-Prespa Transboundary Biosphere Reserve (North Macedonia and Albania). Zooplankton assemblages, i.e. Rotifera, Cladocera and Copepoda, are significant indicators of environmental alterations due to their essential role in aquatic food webs, as pivotal organisms between nutrient resources, primary producers and higher trophic levels. Main traits, diversity, abundance, biomass and functional feeding guilds of pelagial zooplankton assemblages in oligotrophic Lake Ohrid was assessed in relation to main environmental drivers. Two endemic copepods, calanoid *Arctodiaptomus steindachneri* (Richard, 1897) and cyclopoid *Cyclops ochridanus* Kiefer, 1932. Both species are phytoplankton grazers and enhanced balance within food webs. However, increasing of organic matter indicate on higher abundance of detritivorous, rotifer and cladoceran species. The most abundant rotifer was microphagous species *Kellicottia longispina* (Kellicott, 1879). Our results suggested the sig-

nificant importance of zooplankton for the essential preservation of globally unique ecosystem, supporting sustainable development and the conservation of species.

Key words: assemblages, functional feeding guilds, indicators, nutrient resources, trophic levels

P17

Modelling factor – Macrophyte communities for macrozoobenthos in Lake Prespa, Republic of North Macedonia

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Abstract: The macrophyte vegetation and the macrozoobenthos are two components mutually interconnected and contributing in the overall stability of the water ecosystem of Prespa Lake. Macrophyte communities as modeling factor for the benthic faunafrom 3 different depth pointsusing the methods of transects on four sites along the littoral region from Prespa Lake was subject on investigations. We used sampling methods in accordance to WFD and ISO standard for sampling of macrophytes and macroinvertebrates. According to the results, 32 macrozoobenthos species have been identified from 8 systematic groups (Turbellaria, Oligochaeta, Hirudinea, Gastropoda, Bivalvia, Isopoda, Amphipoda and Insecta). The following benthic species: *Gammarus triacanthus prespensis*, *Dreissena presbensis*, *Pyrgohydrobia prespaensis* and *Chironomus plumosus* qualitatively predominates in the samples from the littoral region of the Macedonian part of Lake Prespa. The quantitative analyses (according to their presence on m^2), has shown that *Gammarus triacanthus prespensis* (7250ind/m^2) and *Dreissena presbensis* (7025ind/m^2) are dominant species in the sampling sites. Concerning the macrophyte communities, 13 species have been identified on two sampling depth points (*Phragmites australis* (Cav.) Trin. ex Steud., *Typha angustifolia* L., *Potamogeton perfoliatus* L., *Potamogeton lucens* L., *Potamogeton*

crispus L., *Potamogeton pectinatus* L., *Potamogeton pusillus* L., *Myriophilum spicatum* L., *Ceratophillum demersum* L., *Ceratophillum submersum* L., *Vallisneria spiralis* L., *Zannichellia palustris* L., and *Najas marina* L.).

Key words: macrozoobenthos, macrophyte communities, Lake Prespa

P18

Diversity of macrozoobenthos in different type of Charophyta mono species communities in Lake Ohrid

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Abstract: One of the most impressive faunistic characteristics of Lake Ohrid is it rich and endemorelict benthic fauna. During our researches, we have compared a different monospecies communities of Charophyta as a habitat and food soursce for numerous macrozoobenthos species. In general, the investigations on the structural characteristic of the macrozoobenthos communities showed that the indices of richness and diversity rich the highest values in the mixed macrophyte associations developing on sandy bottom. The monospecies communities built by *Chara tomentosa*, *Chara globularis*, *Nitella opaca* and *Chara ohridana* in Lake Ohrid occur in deeper parts of the littoral, at the lower limit of distribution of macrophytic vegetation. The highest abundance of the macrozoobenthos species has been recorded on the communities of *Chara tomentosa* (9 species from 6 classes). The communities of *Nitella opaca*, which appear very rare in the Lake are found to be least attractive for the macrozoobenthos species i.e. only 4 species from 4 classes have been registered.

Key words: Macrozoobenthos species, Monospiecies communities, Charophyta, Lake Ohrid

P19

Physical-chemical and biological indicators in function of the Lake Ohrid trophic state assessment

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Abstract: The littoral region of an aquatic ecosystem is exposed to the most intense anthropogenic impact. The aim of this study was to estimate the trophic status of Lake Ohrid at 10 littoral sites that are under different external influence: near the river mouths (Sateska Littoral, Daljan, Grashnica), central city area (Mazija, Saraishte and Ohrid Bay), settlements (Radozda and Kalishta) and attractive touristic places (Metropol and Park). During the summer season 2017, several physical, chemical (biodegradable organic matter, total phosphorus and total nitrogen concentrations) and biological indicators (heterotrophic bacteria, total number of coliform bacteria, concentration of chlorophyll a and rotifers) have been measured. The Carlson's Trophic State Index (TSI), based on the concentrations of total phosphorus and chlorophyll a, showed mainly oligotrophic state of the water, with exception in Sateska littoral, Pristanishte and Grashnica, where TSI belongs to mesotrophic and meso-eutrophic state. The biological parameters are also correlated with physical-chemical parameters. The highest number of heterotrophic bacteria, as indicators of organic pollution, and coliform bacteria as indicators of faecal contamination are

registered at Grashnica (31 980 heterotrophic bact/ml; 81 900 coliform bact/100 ml) and Sateska Littoral (29 280 heterotrophic bact/ml; 52 200 coliform bact/100 ml). Saprobic Index based on rotifers ranged from 1.5 to 1.97, which correspond to oligosaprobic, oligo-β-mesosaprobic and β-mesosaprobic waters. In the region of Grashnica, the saprobic index is 2.63, which correspond to α-mesosaprobic water. The littoral sites Grashnica and Sateska Littoral, are under the pressure of communal waste water rich with pollutants and nutrients, due to the influence of the rivers Sateska and Velgoshka that flow into those regions.

Key words: littoral, nutrients, bioindicators, bacteria, rotifers, water quality

P20

Histochemical and biochemical alterations in Zebra mussel, *Dreissena polymoprha* (Pallas, 1771) after Cd and PAHs chronic exposure

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Abstract: Cd and PAHs are contaminants which are priority pollutants in surface waters according to Directive 2013/39/EC. They are toxic, persistent and tend to bioaccumulate in high levels in aquatic organisms. Zebra mussel, *Dreissena polymorpha* is proposed as a freshwater bioindicator in polluted waters. Biomarkers are biological tools which show the negative effects of different toxicants at a cellular, tissue or organism level. The aim of the present *ex situ* experiment was to study the impact of short-term (96h) and long-term (31 days) exposure to Cd and PAHs on the gills and digestive gland of Zebra mussel. For this purpose we studied the histochemical structure of the gills by applying the PAS-reaction, as well as the biochemical changes in the digestive gland by measuring the activity of (CAT and ChE). Thetested concentrations were prepared according to the legislation and represented the average annual concentration (AAC, 100%) and 50% above and below AAC. In sum, we found alterations both, in the gill structure and enzymatic activity at all tested concentrations, including the one below the allowable concentration according to EU legislation. These results confirmed the toxicity of Cd and PAHs. Furthermore, Cd was more toxic compared to PAHs in terms of the studied parameters. Last, the applied biomarkers and the proposed bioindicator (Zebra mussel) could be used for future monitoring and risk assessment programs.

Acknowledgements: The study is carried out with the financial support of project MU-19-BF-014, Assessment of the impact of Cypermethrin and Chlorpyrifos on Zebra mussels (*Dreissena polymorpha* Pallas, 1771) through application of complex biological approaches according to Directive 2013/39/EC, financed by the Department of Scientific Research, University of Plovdiv, Bulgaria.

P21

Cd and PAHs exposure changes the condition index and soft tissue wet ratio in *Dreissena polymorpha* (Pallas, 1771): a statistical perspective

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Abstract: Cd and PAHs are priority pollutants in surface waters according to Directive 2013/39/EC. They are toxic, persistent and tend to bioaccumulate in high levels in aquatic organisms. In the present Zebra mussel, *Dreissena polymorpha* is confirmed as a bioindicator for contaminated freshwater ecosystems. We aimed to study the effects of short-term (96h) and long-term (31 days) exposure to Cd and PAHs on the condition index and soft tissue wet ration in Zebra mussel. Overall, we found alterations both, in the condition index and soft tissue ratio at all tested concentrations, including the one below the allowable concentration according to EU legislation compared to the control group. These results confirmed the toxicity of both contaminants. Furthermore, we confirmed that the studied parameters could be successfully applied as biomarkers, as they are easy, fast and low-costin future monitoring programs and risk assessment programs in polluted freshwater ecosystems.

Acknowledgements: The study is carried out with the financial support of project MU-19-BF-014, Assessment of the impact of Cypermethrin and Chlorpyrifos on Zebra mussels (*Dreissena polymorpha* Pallas, 1771) through application of complex biological approaches according to Directive 2013/39/EC, financed by the Department of Scientific Research, University of Plovdiv, Bulgaria.

P22

Comparative analysis of the oxidative stress in Bulgarian Black sea bivalves and their bioindicator potential

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Abstract: Marine bivalves are essential part of the ecosystems filtering large quantities of water. Nowadays different species of bivalves are widely recognized as biomonitors. As with all aerobic organisms, reactive oxygen species (ROS) are generated in bivalves, that are neutralized by the evolutionary developed antioxidant defense system. Increased ROS production under the influence of various factors results in disturbing the balance between pro- and antioxidant processes, i.e. oxidative stress (OS). Studies confirm that pollutants induce OS in bivalves and this could serve for assessing anthropogenic impacts. The aim of the present study was to assess oxidative stress biomarkers in different species of bivalves (*Mytilus galloprovincialis*, *Donax trunculus*, *Chamelea gallina*) from different habitats of the Bulgarian Black sea from the viewpoint of their bio-indicative potential. It was established that *M. galloprovincialis* had higher values of LPO, GSH, GR and G6PD. Relatively low LPO levels were measured in *D. trunculus*. The antioxidant enzymes SOD, CAT and GPX in *M. galloprovincialis* and *D. trunculus* were similar. The SOD and CAT activities in *Ch. galina* were higher. The higher antioxidant potential in *Ch. galina* could be due to higher enzymatic activities of SOD and CAT, leading to low levels of LPO. In contrast, the higher LPO in *M. galloprovincialis* is related to the low enzymatic defense activity. Enzyme protection is a determining factor for the functional state of the bivalves studied.

Key words: Bulgarian Black sea, *Chamelea gallina*, *Donax trunculus*, *Mytilus galloprovincialis*, oxidative stress biomarkers.

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P23

Evaluation of the impact of Rapana fisheries with beam trawl on juvenile stages of turbot in Western Black Sea

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Abstract: The study aims to research bycatch in commercial beam trawls used in the *Rapana venosa* fishery during 2017 in the Bulgarian territorial waters. Four commercial boats, using beam trawls, are chartered and catch data are recorded from a total of 104 hauls. The hauls are performed at a depth of 14-38 meters for duration of 20 – 220 min and the hauling speed varies between 2.8-3.6 knots. The average rapana catch is 49.20 kg/trawl, while the catches vary from 10 to 160 kg/trawl. The total catch for the whole study period amount at 5117 kg. About 60 species are identified as bycatch, among them 29 turbot individuals are gathered, with a total weight of 14.46 kg. The average turbot weights from all trawls attain 0.13903 kg/trawl with a maximum of 2.22 kg/trawl. The current study discusses the impact of commercial beam trawls on the juvenile stages of turbot in Western Black Sea.

P24

Contribution to parasite fauna of Prespa roach *Rutilus prespensis* (Karaman, 1924) (Pisces: Cyprinidae) in Lake Prespa, North Macedonia

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Abstract: Eight out of the eleven indigenous fish species of Prespa are endemic, i.e. they exist only in the Prespa region and in no other place on earth. One of these endemic fish species is Prespa roach (*Rutilus prespensis*). The parasitological examination from the Macedonian part of the Lake Prespa showed that 175 specimens of Prespa roach fishes were infested of the 183 examined (95.63%). In our case study the presence of 14 parasite species was established: *Myxobolus cyprini*, *Myxobolus dispar*, *Chilodonella piscicola*, *Dactylogyrus sphyrna*, *Dactylogyrus erhardovae*, *Paradiplozoon zeller*, *Nicolla testiobliquum*, *Posthodiplostomum cuticola* (larva), *Caryophyllaeus laticeps*, *Ligula intestinalis* (plerocerkoid), *Philometra ovata*, *Contracaecum microcephalum* (larva), *Metechinorhynchus truttae* and *Ergasilus sieboldi*. Individually, by the parasite species, the highest prevalence was with *Posthodiplostomum cuticola* (larva) (84.70%), *Ligula intestinalis* (plerocerkoid) (77.04%) and *Dactylogyrus sphyrna* (38.80%). The lowest one was with *Caryophyllaeus laticeps* and *Philometra ovata* (0.55%). In our case study the parasite *Dactylogyrus erhardovae* is recorded for the first time in the ichthyoparasitofauna of Lake Prespa and North Macedonia.

Key words: Parasites, *Rutilus prespensis*, Lake Prespa

P25

Length-weight relationships for 5 cyprinid fish species from Lake Ohrid

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Abstract: Length-weight relationships (LWR) were estimated for 5 species of cyprinid fishes from Lake Ohrid, including *Alburnus scoranza, Barbus rebeli*, *Pachychilon pictum*, *Rutilus ohridanus* and *Squalius squalus*. From the measured data of length and weight, the a and b parameters were calculated and ranged from 0.0028 to 0.0187, and from 2.726 to 3.6038 respectively. The condition factor was also calculated and ranged from 0.9477 to 1.3424. This study provides first complete LWRs for some of these species, which are lacking in FishBase.

Key words: LWR, fish growth, condition factor, FishBase, Lake Ohrid

P26

Ecosystem pressure and impact on fish population in Channel Studencista (Lake Ohrid)

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Abstract: The Studencista channel has created the water supply from Biljana's sources and is in close communication with Studencista wetland and Ohrid Lake. The Studencista channel is an important winter residence for the fish *Alburnus scoranza* (Pisces, Cyprinidae). In the last decade, the channel has been intensively used as a marina for small craft, which causes major changes in the aquatic ecosystem. Similarly, in recent years, when the water level of the lake and the channel has decreased, fishing pestilence are often recorded. In our paper was presented results of histological and hematological investigation on *Alburnus scoranza* population from Channel Studencista. Our results display good health condition of fish population, but oxygen saturation of water is crucial factor for fish pestilence during winter period 2017 and 2018 years.

Key words: Studencista Channel, Lake Ohrid, histology, hematology, *Alburnus scoranza*

P27

Insights into the genetic structure of the genus *Chondrostoma* (Pisces, Cyprinidae) across the central part of Balkan Penninsula

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Abstract: The taxonomy of the species which belongs to genus *Chondrostoma* is rather problematic and the introduction of molecular data in the recent times began to yield in increased quality of the determination of the philogenetic connections between the members of this very heterogeneous group. Up until today different molecular-genetic techniques were applied during the identification process of the species of genus *Chondrostoma*.

In this paper has been analyze a total of 45 fish individuals collected from different localities in Republic of North Macedonia: River Bregalnica – tributary of River Vardar and two natural lakes-Ohrid Lake and Prespa Lake. Samples affiliated with the three previously mentioned fish species: *Chondrostoma ochridanum, Chondrostoma vardarense* and *Chondrostoma prespensis*.

Key words: *Chondrostoma* genus, molecular genetic techniques, Lake Ohrid, Lake Prespa, River Vardar.

P28

Researches on reed belt from Lake Prespa as habitat and spawning ground for cyprinid fishes

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Abstract: Lake Prespa is second big lake in North Macedonia situated in the south-western part of the country at an altitude of 853 meters. It is a transboundary lake with surface area of 274 km^2, maximal length about 28 km, and maximal width about 17 km. In the littoral region of Lake Prespa are present different populations of macrophyte vegetation (emerged, flotant and submersed). Dominant emerged plant species in Lake Prespa is reed *Phragmites australis* (Cav.) Trin. Ex Steud. which form a natural discontinuous belt. This belt along Macedonian coastline is the most developed along the north and east coast, while along the western coast it is less developed. The reed in belt is high (up to 4.40 meters) and dense (with abundance 4 and 5). The reed belt from Lake Prespa provide habitat, shelter and food for a large number of fishes. Also, it represent spawning ground for some cyprinid fishes, and have a large role in protection from their predators. In Lake Prespa are evidented 11 native fishs species of which in the belt of reed live and performed spawning *Alburnoides prespensis* Karaman, 1924, *Alburnus belvica* Karaman, 1924, *Chondrostoma prespense* Karaman, 1924, *Cyprinis carpio* Linnaeus, 1758, *Pelasgus prespensis* Karaman, 1924, *Rutilus prespensis* Karaman, 1924, and *Squalius prespensis* Fowler, 1977. In recent years oscillations of the water level, agricultural intensification with burning and cutting the reed and pollution cause significantly changes in the reed belt from Lake Prespa. Inadequate management with reed belt may result in reduction of the natural habitats and the natural spawning grounds of cyprinid fishes.

Key words: Lake Prespa, Reed belt, Cyprinid fishes

P29

The impact of alien fish species on native fish population from Lake Dojran and Lake Tikvesh (Aegean catchment area)

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Abstract: Introduction of alien fish species has resulted in great global change, damaging native fish species and communities throughout the world. The main objectives of this paper were to researches the species composition, ecology, and distribution of fish species in natural and artificial water bodies and to evaluate the role and contribution of alien species to socio-economic development in some parts of North Macedonia. Since the early twentieth century, the rate of introduction and the number of alien fish species introduced into local habitats have greatly increased. Differences in fish population structure in various sites were related with differences in physico-chemical parameters, composition of macrophyte vegetation, change of habitat composition and the other living conditions. The occurrence of alien species has not only changed the structure of native ichthyofauna, but has also caused ecological damage in water ecosystems, as well as economic harm to local fishermen. The researches of species composition, distribution and movement of alien fish in natural habitats revealed that these species have quickly spread and are now distributed in diverse aquatic habitats in Aegean catchment area, Dojran-natural

lake and Tikves-artificial lake. Also, in the Adriatic watershed where a large part of Macedonian waters belong, the situation is similar. This, provided evidence that are no limitations to the spreading of alien fish species in the natural habitats of Macedonian waters.

Key words: alien fish species, Macedonian waters, Aegean watershed, native fish fauna

P30

Long-term changes of the ichthyofauna in the Iskar River, Danube basin

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Abstract: Ichthyofauna of the Iskar River (the longest Bulgarian tributary of the Danube) has been studied since 1920s what enables to trace its alterations over the past nearby 100 years. A total of 48 fish species have been reported in the Iskar River so far. No significant changes of the species composition has been found in the river ichthyofauna as a whole throughout the 100 year period but decrease of the total fish abundance, and alterations of both the community structure and longitudinal species distribution occurred. Already in early 1920s significant decline of fish abundance and reduction of the mean fish size has been reported as effects of predatory fishing practiced by local people and of damaged fish habitats by the floods and sediment deposition on the river bottom due to catchment area deforestation and because of water abstraction for irrigation. Construction of massive dams and barrages has limited fish movements upstream. The river bed modifications and embankment of the flooding terraces have lead to increasing of the flow speed, alterations of the sediment features and loss of habitats of limnophylous fish species. Furthermore, until recently the fish fauna in the middle and lower courses of Iskar River was strongly affected by water pollution from industrial and household sources. Currently hydromorphological features along the Iskar River benefit the development of predominating fish complex typical for the semi-mountain sections of the Danube tributaries which was found to be distributed substantially wider than until the mid 20th Century.

Key words: Iskar River, ichthyofauna, human pressures, community structure, species composition, distribution

P31

Feeding ecology of the Green Toad (*Bufotes viridis* complex) in urban environment. Part 1. Trophic spectrum and niche

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Abstract: The current study presents data about the feeding ecology of the Green Toad (*Bufotes viridis* complex) from two of the hills of Plovdiv – "Mladezhki hulm" Hill and "Hulm Bunardzhik" Hill. The diet of the Green Toad consists mainly of insects, predominantly Hymenoptera, Formicidae and the Coleoptera order, with some differences between the number and volume ratios of the taxa between sexes and seasons. When comparing the quantitative composition of the trophic spectrum between the two populations, we did not detect statistically significant differences. However, the value of the trophic niche breadth of the population from "Mladezhki hulm" Hill is significantly greater than that of the population from "Hulm Bunardzhik" Hill, indicating that the first population uses a more diverse food. The overlapping of food niches between the two populations is low, due to the different conditions and micro-habitats of the two studied hills. The Green Toad can be classified as a polyphage (zoophage), which plays an important role in the food chains, both in natural and urban ecosystems.

Key words: green toad, *Bufotes viridis* complex, diet, trophic spectrum, trophic niche, Plovdiv hills.

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P32

Feeding ecology of the Green Toad (*Bufotes viridis* complex) in urban environment. Part 2. Prey availability and electivity

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Abstract: The second part of our study on the feeding ecology of the Green Toad (*Bufotes viridis* complex) from two of the hills of Plovdiv – "Mladezhki hulm" Hill and "Hulm Bunardzhik" Hill is focused on prey availability and electivity. Prey availability in the habitat was determined using pit-fall traps in September 2017. For the population at "Mladezhki hulm" Hill we recorded high preference for Hymenoptera, Formicidae with both indices used (Jacob's index – 0.70; Strauss' index – 0.41). We also recorded high preference to the following taxa: Coleoptera, Curculionida and Myriapoda. For the population at "Hulm Bunardzhik" Hill we recorded the highest preference to Hymenoptera, Formicidae (Jacob's index – 0.33; Strauss' index – 0.138) and Hemiptera (Jacob's index – 0.45; Strauss' index – 0.037). It seems that the Green Toad shows slight preference towards the ants, similar to other toad species.

Key words: green toad, *Bufotes viridis* complex, diet, prey availability, prey electivity, Plovdiv hills

P33

Pingers as cetacean bycatch mitigation measure in Bulgarian turbot fishery

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Abstract: The Black Sea turbot, *Scopthalmus meaoticus* Pallas, 1811 is one of the most valuable commercial species in the Black Sea. In Bulgaria quota is set each year and official type of fishing is bottom set gillnets (mesh size 400 mm). This type of fishery is considered one of the most important threats for small cetaceans due to bycatch. By-catch (incidental catch) of small cetaceans is a major problem in a number of gillnet fisheries around the World and Harbour porpoise is one of the most heavily affected species. Few studies on cetacean by-catch rates in turbot fishery have been implemented in the Black Sea (Turkey – Tonay, Ozturk 2003 and Gönener, Bilgin 2009; Ukraine – Birkun Jr. et al. 2009, Bulgaria – Mihaylov 2010) but all of these have reported largest share of Harbour porpoise – 90 to 98%. Even less are experiments with pingers (acoustic deterrent devices) as mitigation measure to decrease bycatch rate. First large-scale use of pingers (Future Oceans 10 kHz and 70 kHz models) was made during standard turbot fishing operations in Bulgarian waters in 2019 during two seasons – spring and summer respectively before and after turbot fishing ban (15 April – 15 June). Three vessels have been involved with part of the nets being without pingers – control and other parts fitted with pingers – active. Bycatch rates in active and control nets have not shown significant difference in both seasons. Though bycatch rates in

summer (2.30 ind/km) were significantly higher compared to spring (0.11 ind/km).

Key words: Black Sea cetaceans, *Phocoena phocoena relicta*, cetacean bycatch, pingers, Harbour porpoise, turbot fishery, gillnets.

P34

Hematology data of striped mouse (*Apodemus agrarius*) from Southeastern Bulgaria (Strandzha Mountain): indicators for environmental status

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Abstract: Haematology parameters are important indicators for the status of both individuals and populations of wild small animals that are affected by toxicants or diseases and must be studied with a view to possible influence of natural or anthropogenic harmful factors. We report for the first time the values of the following serum biochemistry parameters Number of Erythrocytes [10^{12} / l]; HGB) – Hemoglobin ([g / l]; (HCT) – Hematocrit [%]; (MCV) – Mean Corpuscular Volume of erythrocyte [fl]; (MCH) – Mean Corpuscular Hemoglobin of Erythrocyte [pg]; (MCHC) – Mean Corpuscular Hemoglobin Concentration [g / l]; (WBC) – Number of Leukocytes [10^9 / l]; (LYM) – Number of Lymphocytes [10^9 / l]; (MID) – Number of Middle cells [10^9 / l]; (GRA) – Number of Granulocytes [10^9 / l]; (LY) – Lymphocytes – percentage of total leukocytes [%]; (MI) – Middle cells – percentage of total leukocytes [%]; (GR) – Granulocytes – percentage of total leukocytes [%]; (PLT) – Number of platelets [10^9 / l]; (MPV) – Platelet volume [fl] obtained from adult males and females individuals of striped mouse (*Apodemus agrarius*) which are resident in Strandzha region of Northeastern Thrace in Bulgaria. The statistical analysis of the established hematological parameters has defined the initial norm of variation of the mean values described by the ± 95% confidence interval.

They can be used for the evaluation of the physiological condition of the striped mouse individuals, as a bio-indicative marker for evaluating the quality of the natural environment in Southeastern Bulgaria.

Key words: *Apodemus agrarius*, haematology parameters

P35

Strategies in open field behaviour and new object exploration in eastern broad-toothed field mouse, *Apodemus mystacinus* and yellow-necked mouse, *Apodemus flavicollis*

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Abstract: The eastern broad-toothed field mouse, *Apodemus mystacinus* and the yellow-necked mouse, *Apodemus flavicollis* are closely-related rodent species, which in some habitats in Southwest Bulgaria are syntopic. Their exploratory behaviour is of great importance for understanding how individuals occupy the habitats. The purpose of this work is to compare the behavioural response of individuals of each of the two rodent species to a new environment and to a new object in order to establish their behavioural strategies and the plasticity of their behaviour. To achieve this, the animals were tested in both natural and laboratory conditions. The eastern broad-toothed field mice and yellow-necked mice were captured from wild populations in two syntopic habitats in Southwest Bulgaria in the spring-summer period of 2018 using Sherman traps. Animals were tested immediately after their capturing, and a day or two after their transfer to the laboratory. The experiments were carried out in an open field arena. First, the individuals were allowed to explore the open field arena without the presence of a novel object and after that in the presence of a novel object. Each test lasted 5 minutes. A camcorder was positioned above the arena in order to record the animals' reaction. The results showed that in natural conditions *A. flavicollis* is more active in exploration of the new arena and the new object than *A. mystacinus*. The behaviour of *A. mystacinus* remains almost

unchanged in both natural and laboratory conditions. These different strategies seem to reflect their lifestyle and adaptations to the environment.

Key words: behavioural strategies, open field, new object exploration, rodents.

P36

Species diversity and population structure of small mammalian communities depending on habitat heterogeneity in Lozenska Planina Mountain, Bulgaria

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Abstract: Recent data indicate that biodiversity in mosaic habitats play a huge role in keeping environments stable. The aim of this paper is to assess the small rodent community as a key components of agrarian and semi-natural ecosystems. Small mammals were caught on the territory of Lozenska planina Mountain, West Bulgaria in different natural habitats and agricultures. For this purpose natural areas such as forest, grassland and their border areas, as well as corn and alfalfa were selected. Species composition varied significantly according to habitat with a total catch of four rodent and one shrew species. Population number, sex and age structure and some morphophysiological-paramethrs of the house mouse (*Mus musculus*), wood mouse (*Apodemus* sp.), bank vole (*Clethrionomys glareolus*), and common vole (*Microtus arvalis*) so as the common shrew (*Sorex araneus*) were investigated. Correlations were found between abundance of rodent species and habitat variables, particularly vegetation structure and cover and microhabitat.

Key words: small mammals, habitats, vegetation structure

P37

Evidence of radioprotective effect of Resveratrol against extremely low frequency electromagnetic fields clastogenic effect

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Abstract: The living organisms have never before in its evolutionary history been exposed to electromagnetic radiation, especially to extremely low frequency electromagnetic fields (ELF-EMFs) that are ubiquitous in the modern environment. This radiation is considered as a non ionizing radiation. There are some investigations that suggest that such fields can interact with cells in many ways to produce damage at several levels. On the other hand, thare are many attempts to develop radioprotective agents in order to prevent deleterious effects caused by radiation. In the current study, we show evidence indicating that Resveratrol, a natural product isolated mainly from grapes, is capable to inhibit the clastogenic effect induced by an exposure to ELF-EMFs on mice bone marrow micronucleus (MN) frequency. BALB/c mice were exposed, in controlled laboratory conditions, to a critical dosage (previously tested) of 2.0 mT ELF-EMFs at 60 Hz frequency and 72 h exposure, and compared with sham-exposed mice as negative controls. The clastogenic effect was assessed by an *in vivo* MN test. The exposed animals were treated with Resveratrol (Sigma-Aldrich) dissolved in saline isotonic solution (15 mg/Kg), and the controls were treated with saline alone. The obtained results indicated that animals exposed to magnetic fields but treated at the same time with

Resveratrol, showed, in spite of the radiation clastogenic effect, lower frequencies of MN when compared with those exposed animals but without the presence of Resveratrol. These findings suggest the potential use of Resveratrol for radioprotection purposes.

Key words: Extremely low frequency electromagnetic fields, clastogenic effect, radiation protection, micronucleus, Resveratrol

P38

Natural and induced UV irradiation effects on some morphophysiological and genetic characteristics of albino mice

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Abstract: This is the first study on Albino mice (ICR), chronically exposed to natural UV radiation in comparison with a parallel laboratory experiment in order to verify the naturally obtained effects on the animal organism. The mice were exposed at Moussala BEO station (2925 m s.l.) in Rila Mountain and in the laboratory, simulating the same radiation intensity. Results of the genotoxic effect of UV background on the experimental animals are presented. Different types of early mutagenic effects in blood cells like Howell-Jolly bodies in erythrocytes and diverse chromosome aberrations – breaks, fragments, centromeric and telomeric attachments in the bone marrow cells were observed. The analysis shows a gradual increase of Howell-Jolly bodies from the 15th to the 40th day of the experiment. The percentage of metaphases with aberrations in the samples from both peak of Moussala and laboratory varies closely in a similar fashion from about 8.75 ± 0.75 % on the 15th day of the beginning of the impact and increased slightly on the 30-th-day, reaching a maximum value of 10.75 ± 0.92% on the 40th of

the experiments. These experiments lead to the conclusion that the environmental impact of natural UV radiation induces extraordinary DNA changes, harmful for the animal organism, especially at the higher altitude.

Key words: Albino mice, UV radiation, High Mountain

P39

Comparative analysis of age changes in the growth of lower canine teeth of wild boar (*Sus scrofa*) males in Bulgaria

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Abstract: Wild boar (*Sus scrofa*) trophies were measured in order to study the lower teeth growth of male specimens in different regions of Bulgaria. In South Bulgaria, the trophies were obtained on the territory of Sofia, Kystendil and Smolyan Regions (the State Hunting Enterprises "Vitoshko – Studena" and "Kormisosh"), and in North Bulgaria – in the region of Ludogorie and Lovech Region. A total of 227 and 67 wild boar trophies were measured in South and North Bulgaria, respectively.

The comparative analysis of the percentage of visible section over the entire length of the tooth varies between age and area, but increases steadily with increasing age. There is also a difference in the erosion of the lower teeth, which is generally greater for the southern part of the country and for the mountain populations in poor habitats. The ratio between the measured width at the base of the tooth and the alveolar is different in different ages, but is similar in different age populations. When comparing different methods for estimating the age of wild boar (growth and wear of the molars, length of erosed part of the lower teeth, ratio of the total length and the exposed part of the lower teeth as well as Brandt's number), similar results are obtained with an acceptable for practice accuracy.

Key words: *Sus scrofa*, males, canine teeth (tusks), age changes

P40

Dynamics of the number and shooting of wild boar (*Sus scrofa* L.) in Bulgaria and the danger of epizootic

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Abstract: Changes in human society over the past 30 years have affected wildlife. The elimination of borders in Europe have allowed free migration of wild ungulates and they penetrated in all habitats suitable for them. The most adaptable species was the wild boar, which, thanks to man's resettlement, warm winters, lack of predators and desolate farmlands, penetrated the British Isles and northern parts of the Scandinavian Peninsula. In Bulgaria, the formal reporting of the number of wild boar continued after the social changes in 1990. There was no change in the way the numbers and shooting were determined. The last activity is without strict control, but the number of wild boar remains between 1991 and 2000 at a level between 38 and 42 thousand specimens. However, the shooting remained within the range of 20.4% (1991) to 26.0% (1997) of the spring number. The control over the numbers has decreased and by 2010, it has reached 70 680 specimens. The shooting increased from 21.6% (2001) to 37.8% (2005), but this did not affect the steady increase in the number of wild boar. In the spring of 2019, 97681 wild boar specimens were counted in the country. It is obvious that the shottingcan not stop the growth of this species, which is threatened by the African swine fever virus that arose in Europe. The epizootic is massive in Romania, from where, despite all the measures taken, it has infiltrated our country. On 31.08.2018, the first case of a positive sample of African swine fever was reported. The outbreak was recorded in the region of Tutrakantsi vill., Provadia province. The latest data on African swine fever oc-

curred during the period February – May 2019 in the State Hunting Enterpises Tervel and Karakuz, and Silistra State Forest Enterprise. The process is dynamic and the likelihood of African swine fever being stopped is small.

Key words: wild boar, number, shooting, epizooty, African swine fever virus

P41

Red deer (*Cervus elaphus* L.) as a biomonitor for contemporary heavy metal pollution of the environment in forest mountain regions in Bulgaria

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Abstract: The aim of the study was to check the current levels of toxic (Pb, Cd) and essential with a concentration-dependent toxic effect (Cu and Zn) heavy metals in the liver of European red deer (*Cervus elaphus*) inhabiting typical forest mountain regions in Bulgaria – Rhodope Mountains and in Central Stara Planina Mountain. For determination of analytic concentrations of the studied heavy metals was used an inductively coupled plasma atomic emission spectrometry (ICP-AES) and they were represented in [mg/kg dry weight]. The average values of metal concentration in the liver of the red deer inhabited in the Rhodope Mountains follow: for Cu – 176.7; for Zn – 599.3; for Pb – 2.2 and for Cd – 0.9. Respectively in Central StaraPlanina Mountain, they are: for Cu – 115.6; for Zn – 224.7; for Pb – 5.0 and for Cd – 0.9. The established results could be used to create a baseline for the estimation of the current heavy metal accumulation in red deer in the trophic area of the big game in the forest mountain regions of Bulgaria. They also offer an opportunity that red deer be used as a biomonitor of future potential anthropogenic negative impact on the environment in forest regions of the country, under the conditions of modern anthropogenic activities therein.

P42

Epigenetic variation and distinctness of red fox (*Vulpes vulpes*) populations in its European range

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Abstract: Herein we explore the variation of non-metric epigenetic cranial features in 25 red fox (*V. vulpes*) local populations in Europe, including populations inhabiting the Scandinavian, Iberian and Balkan Peninsula, and the territory of Central Europe. Our results revealed the presence of well-expressed polymorphism in all studied 14 cranial traits. The comparison of the red fox populations based on their epigenetic morphological variations shows that in its European area they differentiate in different degrees. The established epigenetic distances between the local populations were consistent with previous research of the subspecies biodiversity of the red fox in its European range based on craniometrics and biochemical genetic markers and are in agreement with the presence of three red fox subspecies in continental Europe – *V.v. vulpes* inhabiting the Scandinavian Peninsula, *V. v. crucigera*, spreading in Central and South-eastern Europe and *V. v. silaceus* typical of the Iberian Peninsula. The evaluation of the epigenetic similarity between these subspecies shows a low degree of epigenetic distance between *V. v. crucigera* and *V. v. silaceus* and the relatively higher of each of them with *V.v. vulpes*. The established epigenetic distances reflecting the population epigenetic characteristics of the red fox in the continent form the substantial source of biodiversity information of this species in its European range. The further research of the epigenetic skull characters of the red fox populations combined with molecular genetic studies will reveal the detailed genetic structure and phylogenetic relationships among its European populations.

P43

The relationship between the fertility potential quality and leukocyte concentration in the sperm as a potential indicator for the mutagenic effect of infections

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Abstract: DNA integrity in spermatozoa nuclei is a reliable indicator for the fertility potential quality. The present study focuses on the search for associations and analysis of the interaction between possible DNA damages, worsened sperm parameters, and increased leukocyte concentration in the sperm. By usage of an acridine orange fluorescence test, 87 men with diagnosis normosoospermia, asthenozoospermia, oligoasthenozoospermia, oligoasthenoteratozoospermia and asthenoteratozoospermia were analyzed. Descriptive statistics, nonparametric methods, Pearson Chi-Square and Gamma tests were used to characterize the groups compared. Statistically significant relationships between sperm quality parameters (concentration, motility and morphology) and the levels of DNA damage in spermatozoa nuclei were found, as well as that with decreasing of spermatozoa concentration, motility and number of cells with normal morphology, the frequency of DNA fragmentations in spermatozoa is increasing. Data from the study indicate a statistically significant correlation between leukocyte concentration and the DNA damage level in the group analyzed ($P = 0.02$). The positive value of the Gamma indicator demonstrates that raising the leukocyte concentration increases the risk of the DNA fragmentations in the spermatozoa nuclei. The analysis of this dependence shows that 59.5% of the men with a reported leukocyte concentration above the norm were found to be with fair (16.3%) or poor (43.2%) fertility potential. The results obtained and the conclu-

sions reached could be used in activities point at prophylactics and prevention for improving male reproductive health.

Key words: DNA damages, sperm parameters, acridine orange test, leukocyte concentration

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P44

Monitoring of micronucleus frequency in rodents with blood parasites

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Abstract: Rodents are vectors of many transmissible diseases. It was found that some parasites cause a genotoxic effect and induce micronuclei in their host cells. The purpose of this study is to assess the degree of presence of blood parasites in wild rodent populations and their potential link to the increase of micronucleus frequencies in erythrocytes. Populations of Macedonian mous (*Mus macedonicus* Petrov & Ružić, 1983), yellow-necked mouse (*Apodemus flavicollis* Melchior, 1834) and common vole (*Microtus arvalis* Pallas, 1778) have been investigated in region contaminated with heavy metals, as well as populations of striped field mouse (*Apodemus agrarius* Pallas, 1771) and *A. flavicollis* in an area with low anthropogenic influence (Strandzha Nature Park). Blood smears were prepared and after staining with a fluorescent dye acridine orange, presence of micronuclei in erythrocytes and blood parasites of the investigated animals was recorded.

The highest percentage of individuals with blood parasites was established in *Microtus arvalis* (57%) and *Mus macedonicus* (41%). In all *Apodemus* populations we registered bacterial infections with *Bartonella* sp., in *M. arvalis* – protozoan infection with *Babesia microti*

and bacterial with *Bartonella,* and in *M. macedonicus* – infection with tripanozoma (*Tripanosoma musculi*) and *Bartonella* sp. Only in the yellow-necked mice from the KCM region there was a statistically significant difference in the mean micronucleus frequency of infected and non-infected animals ($U = 12$, $p = 0.046$), however higher in non-infected animals. As for the other species, forest mice from the Strandzha region inclusive, no reliable difference was established ($p \leq 0.05$) between the average micronucleus frequency of the infected and non-infested animals. This illustrates the lack of a clear correlation between the presence of registered parasites and the value of the mean MN-frequency.

The results of our research show that the observed blood parasites cannot be associated with the induction of micronuclei in erythrocytes of the studied rodents. This provides justification to include rodents with the recorded protozoan and bacterial infections in biomonitoring studies for assessment of genotoxicity in contaminated areas.

Key words: micronuclei, rodents, blood parasites, *Tripanosoma*, *Babesia*, *Bartonella*

Acknowledgments: This study was supported by the National Program "Young Scientists and Postdocs", 2018, funded by the Ministry of Education, Republic of Bulgaria

P45

Rodent control in urban industrial areas in Plovdiv (Bulgaria): from research to action

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Abstract: Pest control measures become more and more important. With the growing economic activity and urbanization lots of businesses confront pest activity. The use of rodenticides is a very common method in rodent's pest control, but the bait consumption depends on the environmental alterations in different industry areas. They can lead to considerable changes in size and behaviour of rodent's populations. The rodents may have versatile feeding behavioural habits in different environmental conditions. Therefore, the aim of the study was on one hand to monitor and control rodents in facilities with different food industries using rodenticide bait sand on the other hand to determine those environmental factors that could directly influence the bait consumption.

In the city of Plovdiv (Bulgaria), 89 bait stations were monitored in different industries sites: glass factory, croissant factory and dairy. The bait consumption was studied using the electronic monitoring *PestScan* method. The effectiveness of the performed pest control was revealed through both bivariate and multivariate analysis. The results showed that factors such as industry type, home-range, vegetation, harbourage and water supplies influence the decision for the exact bait disposal. Another important factor for effective pest control is restraining of alternative food sources vying with the bait. All these factors may affect the rodent urban control planning with rodenticides.

Highlighting the important environmental conditions which influence the bait consumption could help planning of pest control for specific sites by matching adequate with specific strategies.

Key words: rodents pest control, public health, urban rodents, rodenticide

P46

Compensatory increase of the reproductive capacity of red fox (*Vulpes vulpes*) in sympatric coexistence with the golden jackal (*Canis aureus*)

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Abstract: Main reproductive parameters of the most common mesopredators in Bulgaria – a golden jackal (*Canis aureus*) and a red fox (*Vulpes vulpes*) were investigated in two regions – the Upper Thracian value and Southern Dobrudzha, where both species coexist in sympatry. The reproductive activity and the average age of the first birth were calculated in both species. Through a life table based on female individuals, are defined: gross reproductive rate (GRR), net reproductive rate (Ro), generation time (T) and the internal rate of populations growth (r). The testicular weight (g) of the male individuals was measured and the gonadosomatic index (GSI) was calculated.

In the investigated areas 41% of the studied female jackals and 65% of the studied female foxes reproduced themselves. The maximum number of placental spots in jackals was 9, and in foxes 8. The minimum number of placental spots in jackals was 2, and in foxes 4. The ratio between net reproductive speed (R0) and the time between two successive generations (T) was greater for the fox (40.98) than for the jackal (25.16). The gonadosomatic index was lower for the jackal (GSI = 0.08) than for fox (GSI = 0.19) which is a criterion for a higher degree of monogamy for the jackal.

The reproductive parameters studied in both species show a compensatory increase in the breeding capacity of the fox in the conditions of sympatry in Bulgaria and as a result of the faster increase in the number of jackals compared to the red fox.

Key words: red fox, golden jackal, reproductive parameters, sympatry

P47

General evaluation of the impact of invasive fish species on native freshwater fish fauna in Turkey

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Abstract: Currently, about 410 fish species belonging to 16 orders and 32 families have been recorded in the inland waters of Turkey. Of them, 195 fish species (44.1%) are endemic. A total number of 30 inland fish species have been considered invasive and alien species. The invasive alien species have been introduced to the freshwater systems for various purposes and through different pathways, such as aquaculture, biological control, pet trade, and fisheries. Some of the invasive alien species, such as *Pseudorasbora parva* and *Carassius gibelio*, have become dominant and have negatively affected the native fish populations. Three freshwater fish species which are endemic in Turkey are extinct according to the IUCN Red List 2019. The endemic fish species, *Alburnus akili*, *Pseudophoxinus handlirschi*, and *Alburnus nicaeensis*, which inhabited lakes Beyşehir, Eğirdir, and İznik, respectively, have become extinct after the introduction of the invasive species, such as *Sander lucioperca* and *Atherina boyeri*.

Key words: native freshwater fish, endemic fish, invasive fish, Turkey

P48

Seroprevalence of Parvovirus B19 IgG antibodies among pregnant women

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Abstract: ***Introduction***. Lack of observation and sufficient virological and epidemiological studies have led to inaccurate assessments of embryonic problems and associated viral infections in pregnant women. One of these is an infection with Parvovirus B19 (B19V), which may be without of symptoms in the mother, and the fetal problems associated with B19V may not be suspected.

Aim. Our objective was to determined the seroprevalence of Parvovirus B19 IgG antibodies among pregnant women.

Material and methods. A prospective seroepidemiological study was carried out. The Parvovirus B19 IgG antibodies were determined in serum samples. Enzyme-Linked Immunosorbent Assay (ELISA) was applied. NovaLisa Parvovirus B19 IgG ELISA Kit (NovaTecImmunodiagnosticaGmbH, Germany) and UVmax kinetic microplate reader were used. According to supplier instructions antibody levels greater than 11NTU were considered as positive, 9 – 11 – equivocal and lower than 9 NTU as negative. Serum samples from 242 pregnant women hospitalized in a Clinic of Obstetrics and Gynecology, University Hospital-Pleven, Bulgaria were analyzed.

Results. Seventy-three (30.17%) of the samples tested were positive (over 11 NTUs), negative (less than 9 NTU) samples were 168 (69.42%), and one (0.41%) was equivocal (9 – 11 NTU). Highest frequency (35.48%) was detected in women of less 20 years of age and

the lowest prevalence (28.47%) was detected in women between 20 and 30 years of age. No significant difference was found depending of age ($p > 0.05$).

Conclusion.This study found a high susceptibility of pregnant women to the B19V. We recommend conducting serological survey in pregnant women with complications and adverse outcomes of pregnancy, as well as in other high-risk groups.

Key words: Parvovirus B19, pregnancy, B19 IgG antibodies, prevalence

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Seroepidemiological survey of Parvovirus B19 IgG in childbearing age women

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Abstract: ***Introduction***. The infection with Parvovirus B19 is widespread worldwide. The risk of infection with Parvovirus B19 during pregnancy is approximately 1% to 5% with subsequent transplant transmission in 24 – 33% of cases. According to literature data, 30% to 60% of childbearing age women are seronegative. They are a risk group for infection with the virus.

The aim of the study was to determine Parvovirus B19 IgG in childbearing age women.

Materials and methods. A prospective seroepidemiological study was carried out, examining 90 women from 19 to 40 years in Medical Center "Clinical Institute for Reproductive Medicine" – Pleven in 2018. The presence of antibodies was determined by a standardized ELISA kit (Anti-Parvovirus B19 (IgG) (EUROIMMUN, Germany).

Results. Of the 90 serum samples tested for Parvovirus B19 IgG, 34 (37.78%) were positive, 55 (61.11%) negative and 1 (1.11%) borderline. Detection of antibodies increased from 27.27% to 45% with age.

Conclusion. The results of our study confirmed the data of other researchers of high susceptibility of childbearing age women to Parvovirus B19. We recommended active monitoring of risk groups pregnant women to prevent fetal complications.

Key words: seroepidemiological survey, Parvovirus B19 IgG, childbearing age women.

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Status and development of the anti-epidemic control system in the region of Varna

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Abstract: ***Introduction***. After the Liberation in 1878, the modern state – public health in Bulgaria was founded. Effective organizational structures and sanitary legislation are introduced and implemented, in order to support the epidemiological surveillance and control of the widespread socially significant infectious diseases.

Purpose. To analyze the state and development of the anti-epidemic control system in Varna region in the period after the Liberation (1878) to the present in the context of changes in the public health system.

Materials and methods. The data of registered infectious diseases used is in the period from 1878 to 2018 under the form of legal documents. Epidemiological, descriptive, documentary and statistical methods have been applied.

Results. In the years after the Liberation, the epidemiological control in the country and in the Varna region is carried out in accordance with the current global requirements. After 1944, a network of hygienic and epidemiological structures has been set up, which, in cooperation with medical centres, organize and carry out the anti-epidemic and prophylactic activities by region. The massive immunization policy in the country in the 1970s and 1980s significantly reduced the incidence of vaccine-preventable infections to the stages of eradication (variola) and elimination (diphtheria and poliomyelitis). In the transition years after 1989 the health system crisis had a negative impact on the epidemiological surveillance and immunization. Because of this, conditions helping the accumulation of non-

immune contingents and the emergence of epidemics of vaccine-preventable diseases were created.

Conclusion. The changes in the public health system have a huge impact on the system of anti-epidemic control and determine the spread of infectious diseases at different historical stages.

Key words: anti-epidemic system in Bulgaria, immunoprophylaxis, infectious diseases.

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Investigating the attitudes of parents to vaccines and immunizations

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Abstract: ***Introduction***. Vaccines development and their application as prophylactic toolin the struggle against infectious diseases, is one of the greatest achievements ofhumanity, whose efficiency was proven by the elimination and eradication ofsome diseases. Almost simultaneously, with E. Jenner's first successfulattempts to vaccinate smallpox, anti-vaccination movements occur. Over recentyears, epidemics of vaccine-preventable infections are demonstrative example ofconsequences of low immunization rate as a result of abstaining fromimmunization.

Aim. To establish the attitude of parents regarding vaccines andimmunizations.

Materials and methods. Questionnaire survey, statistical processing and dataanalysis.

Results. For the period 18-21.06.2019 at SBALAG "Maichindom" 20 womeninlabour were interviewed. The age of the respondents is between 26 and 43. All respondents have a positive attitude towards immunization. None of themwouldrefuse to immunize their children. Only 20% of the women in labor thinkthey are well informed about the mandatory vaccines and the diseases fromwhich they protect their children. The main source of information is the mediaand the Internet. The prevailing opinion is that medical professionalistsmustinform the parents more actively.

Conclusion. All respondents have a positive attitude towards immunization.The problem that needs to be solved is giving more information about thebenefits of the vaccine and the consequences of the refusal of immunization.

Key words: mandatory immunizations, vaccines, awareness

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Study of hygienic disinfection of hands of staff and students in dental practice

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Abstract: ***Introduction***. The transient microbial flora on the hands of the medical staff may cause infections related to medical care. Unlike permanent, this microflora is superficial and can be removed by washing and disinfection. The hands of the medical / dental staff are the first factor for the transfer of micro-organisms: from patient to patient, as well as to various objects from the hospital / dental environment.

Aim. To investigate microbiological samples from the hands of dental students of the 5th course and medical staff before starting work with a patient to take into account the quality of the hygienic disinfection.

Materials and methods. With a sterile pad with a transport environment, the hands of 115 dentits and students from the Faculty of Dental Medicine of the Medical University – Plovdiv were treated after a disinfectant treatment. The samples were sown on blood and Levin agar and the results were recorded at the 24th and 48th hours after cultivation at 37˚C. Some of the isolates (20 pieces) were identified by Vitek MS (BioMérieux, France) using MALDI-TOF technology. The data is processed with Excel, Microsoft and SPSS 19.9, IBM.

Results. From 115 samples, 21 (18.3%) were sterile. With the highest relative share (69.6%), coagulase-negative staphylococci with a microbial number 103 – 104 were shown. Ten samples (8.7%) found the presence of coagulase-negative staphylococci with a microbial number ≥ 105. Single corineform bacteria, Streptococcus viridans, enterococci, and Gram-positive sporadic rods were recorded. The most common isolates identified with Vitek MS are: *Staphylococcus hominis* (4.35%), *Staphylococcus epidermidis* (3.48%) and *Micrococcus luteus* (2.61%).

Conclusion. The absence of *S. aureus*, as well as representatives of Enterobacteriales and Pseudomonas aeruginosa as a "transient microflora", is a good indicator of the quality of hygienic disinfection of hands with an alcohol-containing antiseptic. Coagulase-negative staphylococci are normal skin inhabitants, but their presence in a high microbial number is undesirable, especially in patients with immunosuppression. These results highlight the need to increase knowledge about hand disinfection and its actual application before working with a patient.

Key words: dental practice, hygiene hand disinfection, transient microbial flora

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Control of *Legionella* spp. in dental practice in England and Bulgaria

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Abstract: ***Introduction***. In dental practice it is easy infections caused by *Legionella* spp. and others such as *Pseudomonas* spp. and atypical mycobacteria to be spread, when working with multiple devices that use water and generate a fine aerosol as well as prolonged periods of water stagnation in the dental units waterlines (eg at night). All this, as well as lack of measures or insufficient measures, lead to the formation of biofilm on the inner surface of the waterlines – an environment that provides the bacteria with optimal living conditions.

Aim. To compare the monitoring and control measures applied in Bulgaria in the dental practice with good medical practices in England and to make recommendations for optimizing the quality of dental water in our country.

Materials and methods. Instructional Materials for the Prevention and Control of Bacterial Contamination and the Quality of Water in Dental Practice, according to the Code of Practice: Legionnaire disease, The Control of Legionella Bacteria in Water Systems (L8), HTM 01-05 (England and Northern Ireland) and WHTM 01-05 (Wales) and HTM 04-01 in England are compared with the guidance materials in Bulgar-

ia in dental practices. 56 water samples from dental units and the water distribution network in dental practice were studied.

Results. From preventive measures in England:

Usage of drinking water; flushing the waterways for 2 minutes at the beginning of the working day and for 20 – 30 seconds between the patients; mandatory disinfection of the dental bottle, washing of waterways on chairs that are rarely used at least twice a week; bacterial contamination test for water; the addition of a biocide to the water in the bottle and the washing of the waterways. The use of drinking water for treatment is only in Bulgaria.From 56 water samples from dental units and waterways from different sites 18 (32.14%) were positive for *Legionella* spp.

Conclusion. The comparative analysis shows that in Bulgaria there is no program to prevent the contamination with *Legionella* spp. of water in dental offices. The risk of infecting patients and dental staff is real.

Key words: *Legionella* spp., waterlines in dental practice, bacterial contamination

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Varicella outbreak in a daycare in Plovdiv (2019)

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Abstract: ***Introduction***. Varicella is the most commonly registered children's infectious disease in absence of a universal vaccine. Although the disease manifests more often with more lightly forms, complications and death are possible. The first vaccine against Varicella was produced in 1974 in Japan but for now it is not included in the Vaccine schedule of Bulgaria. In the absence of a routine vaccination in Kindergartens a periodic epidemic blast of Varicella occurs.

Aim. to conduct an epidemiologic research in an all-day Kindergarten of Varicella outbreak and analyze the epidemiologic characteristics.

Materials and methods. A prospective study on Varicella outbreak has been done in a child establishment (Kindergarten and a nursery) for the period November 2018 – June 2019. The establishment consists of 3 buildings, housing 105 children from the age of 1 year and 6 months to six year olds. In the nursery building 23 kids are taking care of, divided into 2 groups. In the first base of Kindergarten the children are 52, divided into 4 groups of 13 children each. In second base, the children are 15 divided into 2 groups. Complex epidemiological and statistical methods have been used. Age, gender, epidemiological connections between the cases and parents' temporarily work incapacity have been analyzed.

Results. All in all for the researched period in RZI Plovdiv have been registered 62 (59%) cases of Varicella. The first case in registered in November 2018 and the last one in June 2019. During this period the

child establishment has been under permanent quarantine. The average age of the sick children is 2.5 years old, 53.2% boys and 64.8% girls. The most of the infected children are listed during the months of April and May respectively 20 and 19 cases. Amongst the contacted children in the child establishment there are 4 of them with 2 doses of the Varicella vaccine applications and 2 children with one dose application. From them a 5-year-old got infected, with one dose of the vaccination application. From the staff of the establishment, 13 women are in fertile age and one of them is vaccinated with 2 doses. There are none infected among these women.

Conclusion. For containing this highly infectious disease, targeting mostly kids to 6 years of age and to avoid complications and death cases , we consider for appropriate to include the Varicella vaccine. It is necessary to be applied on a scheme of 2 doses and a range of over 80% of the children in immunization age to avoid epidemiological spread.

Key words: Varicella, outbreak, vaccine, Kindergarten

P55

Surveillance of Ventilator-associated pneumonia in neonatal intensive care unit

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Abstract: ***Introduction***. Ventilator-associated pneumonia (VAP) is a common complication of critical illnesses in neonatology intensive care units (ICUs).The main pathogenic bacteria of VAP are Gram-negative pathogens,which show a general decline in sensitivities to commonly used antibiotics.

Aim.The aim of the present study was to determine the clinical characteristics of the patients with VAP and the main causative agents.

Materials and methods. A prospective epidemiological study was conducted in the Neonatal intensive care unit of University Hospital "Saint George" Plovdiv, Bulgaria from January 2017 to June 2018. Information for 507 newborns (239 full-term and 268 premature newborns) hospitalized in the NICU was analyzed. For the aims of the study are used conventional microbiological methods for identification of the isolated microorganisms from tracheal aspirates. The rates of nosocomial infections (NI) and VAP are calculated on the base of 100 hospitalized newborns.

Results. The NI incidence was 9.5%. Ventilator-associated pneumonia was the most frequent NI – 67.86%, followed by bloodstream infec-

tion – 23.21% ($p < 0.001$). 107 of the infants were intubated for more than 48 hours, 33 of whom had VAP (30.84%). The mean birth body weight in the VAP group was statistically significantly lower than in the group without VAP (1617.58±817.561 vs 1967.30±935.144). In the etiological structure of VAP the proportion of Gram-negative microorganismis statistically significantly higher than the Gram-positive microorganisms (86.36% to 13.64% $p < 0.001$). The leading agents of VAP were *Klebsiella pneumoniae ESBL+* (27.27%), followed by *Acinetobacter baumannii* (13.64%) and *Pseudomonas aeruginosa* (10.61%). From the Gram-positive microflora the leading pathogen was *Enterococcus faecalis* (6.07%).

Conclusion.VAP remains a serious NICU problem. We recommend strict adherence to the infection control measures to lower the incidence of VAP.

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The main epidemiological paradigm in the context of viral hepatitis C: Comorbidity and co-infections

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Abstract: ***Summary***. The epidemic process in viral hepatitis C differs significantly from the universal characteristic of epidemic process in many trivial infections. The difference is due to clinical and epidemiological factors. Among the clinical factors is the chronicity of the infection, leading to the manifestation of the infection in later phase and elderly age, mainly as complications and in comorbidity with other diseases. Another feature is the disguise of the disease as a co-infection of some sexually transmitted or blood-borne infection. In these cases, attention is drawn to a more popular disease, such as AIDS or HBV.

Objective. To provide a brief literature review of the problem of comorbidity and co-infections with HCV and to present a typical case.

Methods. A documentary research method and clinical method have been used.

Results. Two cases, hospitalized in Clinic of Infectious Diseases at University Hospital "Dr Georgi Stranski"– Pleven, have been described and interpreted from a clinical and epidemiological point of view. One of the cases reported was with comorbidity with several concomitant non-infectious diseases and very severe clinical course. The other was a carrier of HCV, subsequently co-infected with HBV and developed acute hepatitis B infection.

Discussion. In the literature review we made, the problem was analyzed according to the basic epidemiological paradigm for the trinity of the epidemic process, including in this case predominantly hidden / unrecognized sources of infection, various mechanisms of transmission and more or less isolated risk groups.

Key words: epidemic process, viral hepatitis C, comorbidity, co-infection

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The role serological screening for determining of HBsAg among hospitalized and ambulatory patients

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Abstract: It commonly occurs in the clinical practice that HBsAg seropositivity is determined by chance prior hospitalization for operative treatment/intervention or after prophylactic tests.

Aim: The aims of study is to determine the prevalence of Hepatitis B surface antigen (HBsAg) among hospitalized and ambulatory patients of Uni Hospital – Panagyurishte.

Materials and Methods. During the period from July 2016 to July 2019 a total of 4 905 patients. Detection of the HBsAg was performed in human serum specimen by ELFA (Enzyme Linked Fluorescent Assay) method with a VIDAS PC (Biomerieux, France) immunological analyzer.

Results. 4 905 serum samples were tested in 2 578 (53%) women and 2 327 (47%) men in the age range from 10 to 89 years. 4 761 (97%) were negative for HBsAg. Positive HBsAg values were detected in 144 (3%).

Conclusion. Our study confirmed importance for early diagnosis of Hepatitis B virus (HBV) infection of the serological screening to detection of HBsAg.

Key words: HBV infection, HBsAg, ELFA

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Diagnosis and treatment of bisphosphonate-associated osteonecrosis of the jaws

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Abstract: ***Introduction***. Bisphosphonate-associated osteonecrosis of the jaws (BAONJ) is a complication of great medical significance. Some of the most discussed and controversial aspects are the diagnosis and treatment of the condition.

Aim. To investigate patient diagnosed with BAONJ in order to evaluate the methods of diagnosis and treatment.

Material and methods. A prospective epidemiological study of 112 patients diagnosed with Bisphosphonate-associated osteonecrosis of the jaw in 2016 and 2017 was conducted in the Clinic of maxillo-facial surgery of UMHAT "St. George", Plovdiv, Bulgaria, based on anamnesis, clinical examination, hospital documentation, imaging and paraclinic studies. SPSS Statistics v.24 was used for statistical analysis, at a significance level $p < 0.05$.

Results. The main diagnostic methods for all patients were the clinical examination and anamnesis. 91.96% had additional X-ray performed, and 1/3 – additional computer thomograpy or MRI scan. Half of the patients were diagnosed at stage II, 29.46% at stage I, 17.86% at stage III and 1.79% at stage 0. Pathogenic microorganisms were isolated in

25.89%, most commonly *E. coli*, *E. cloacae* and *Klebsiella pneumoniae*. Histologic examination was performed to 24.10%. Of the patients, 41.96% had multiple hospitalizations. Treatment included an antibiotic, in 54.45% from the β-lactam antibiotics group, followed by a significantly lower proportion of the treated with Lincosamides (31.25%) ($p < 0.05$). Debridement was preformed to 32.14% of patients, followed by other surgical manipulation in 18.75%, and resection in 4.56% of cases.

Conclusion. BAONJ is diagnosed late, which correlates with repeated hospitalizations due to relapse and necessity of surgical treatment.

Key words: bisphosphonates, osteonecrosis, diagnosis, treatment, epidemiology

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Epidemiological surveillance – indicator for the effectiveness of the medical support in floods

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Abstract: ***Introduction***. Floods are the most common disasters in Bulgaria. Despite that, medical support activities that are performed still are unable to face many of the challenges regarding protection and restoration of population's health. Epidemiological surveillance of the emerging infectious diseases in response and recovery phases could be used as an indicator for the effectiveness of the performed medical activities. Epidemiological surveillance can also identify required actions for mitigation of the impact of the floods in the future.

Aim. The aim of this study is to evaluate the role of epidemiological surveillance after floods as an indicator for the effectiveness of the medical support to the population in the affected area.

Materials and methods. Retrospective analysis of the epidemiological situation in Plovdiv and Pazardzik region after the flooding of Maritza river basin in 2005 is used. By the means of deductive and heuristic analyses the influence of epidemiological data on medical support planning in floods is predicted.

Results and discussion. The characteristic curves for morbidity from diseases with fecal-oral route of transmission after the flood categorically display the insufficiency of the preventive measures, applied in the prodromal phase, as well as inefficiency of the health education among the population after the flood.

Conclusions. The undertaken retrospective study proves the necessity for detailed analysis of the epidemiological situation after the flood

and the use of the obtained data for extension of the preplanned preventive measures and health education of the population.

Key words: floods, disaster medical support, epidemiological surveillance, preventive measures

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Epidemiology and health education as disaster medical support elements

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Abstract: ***Introduction***. Disaster medical support in and near to the area of damage is a set of activities that are performed before, during and after the onset of a natural or man-made disaster. In order to plan, organize and perform the disaster medical support activities, an effective and sufficient medical information exchange is needed.

The aim of this study is to assess the role and importance of epidemiology and health education in disaster planning and medical information exchange management.

Materials and methods. By the means of descriptive and comparative methods the type and nature, as well the sources of medically relevant data in the disaster planning and medical management processes, are analyzed. The regulatory documents that determine the form and contents of the different types of disaster medical plans are studied. Cluster analysis is used to identify the epidemiological data that is mandatory for the effective prevention and eradication of the impact of the damaging factors.

Results and discussion: The results of the performed analyses highlight that in order to fulfil medical information exchange, every element of the information chain has to process and exchange meaningfully unified medical data, some of which are related to the epidemiological situation. For health education success epidemiological knowledge is needed to be transmitted in an appropriate, understandable way during health education.

Key words: epidemiological situation, disaster medical support, medical information, health education

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Which measures to prevent the spread of measles at a hospital in Bulgaria are recommended when examining / hospitalizing a patient

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Abstract: From a practical point of view, different precaution options are considered that would increase the readiness of a healthcare facility to isolate a patient with (suspected) measles, in order to prevent the spread of infection among staff, other patients and visitors, especially those with low immunity and uninsured for different reasons vaccination-coverage. Such measures are: providing a self-contained ventilation and air filtration room with a HEPA filter, or an adapted one with natural ventilation and air dilution, and the ability to draw air from the patient's room to an out-of-hospital area without risk from infecting other people. Other precautions are also discussed, such as wearing different types of respiratory protection masks, vaccine prophylaxis and immunoprophylaxis, etc. All these measures are recommended by the world's leading institutions for infection prevention and control. The provision of some of these measures and the familiarity of the personnel would increase the readiness for early response to other epidemic situations in the hospital in case of airborne infections, as well as the correct behavior to prevent the spread of healthcare associated infections / HCAI, including those caused by hospital strains with antimicrobial resistance.

Key words: measles, airborne infections, precautions, prevention, control, hospital

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