UNIVERSITY OF VETERINARY SCIENCES BRNO

Mendel University in Brno

European Association of Fish Pathologists

THE 2ND MEETING OF THE CENTRAL AND EASTERN EUROPEAN EAFP BRANCHES

1st to 4th September 2026 Křtiny, Czech Republic

ABSTRACT BOOK

University of Veterinary Sciences Brno

Mendel University in Brno

THE 2ND MEETING OF THE CENTRAL AND EASTERN EUROPEAN EAFP BRANCHES

1st to 4th September 2026 Křtiny, Czech Republic

ABSTRACT BOOK

Editors: Ivona Toulová et al.

UNIVERSITY OF VETERINARY SCIENCES BRNO

Mendel University in Brno

2026

Acknowledgement:
Organizers thank to sponsoring organisations:
University of Veterinary Sciences Brno, Faculty of Veterinary Hygiene and Ecology
Mendel University in Brno, Faculty of AgriSciences

Editors:
Ivona Toulová, Miroslava Palíková, Ivana Papežíková, Ivana Mikulíková

© Mendel University in Brno, Zemědělská 1, 613 00 Brno, Czech Republic

ISBN 978-80-7701-115-0 (online ; pdf)
https://doi.org/10.11118/978-80-7701-115-0

Open Access. Publication *The 2nd Meeting of ohe Central and Eastern European EAFP Branches: Abstract Book* is licensed by Creative Commons Attribution-ShareAlike 4.0 International (CC BY-SA 4.0) license.
https://creativecommons.org/licenses/by-sa/4.0/deed.en

ORAL PRESENTATIONS

Petr Posel: Practical benefits of our participation in the EAFP meeting 2024 5

Krzysztof Rakus: Modeling tilapia lake virus (TiLV) and fancy-tailed guppy virus (FTGV) infections in zebrafish 6

Anju Sebastian: Integrated analysis of strain-specific host responses and gill microbiome dynamics during *Cyprinid herpesvirus*-3 infection in carp 7

Mikolaj Adamek: Patholomics of koi sleepy disease in common carp caused by the carp edema virus infection 8

Ľubomír Pojezdal: Koi herpesvirus disease in the context of the Animal Health Law – current status and diagnostic methods 9

Pavla Bartošová-Sojková: Lust for blood: Proteases-mediated hematophagy in the common carp pathogen *Sphaerospora molnari* 10

Terezie Maturová: Parasite fauna of non-native bullhead catfish of the genus *Ameiurus* in Czechia 11

Edit Eszterbauer: Biological control possibilities of fish-pathogenic water moulds (Oomycota, Saprolegniales) 12

Kateřina Matějíčková: Early eDNA detection and targeted oral metaphylaxis for the control of *Ichthyophthirius multifiliis* in salmonid fish 13

Alena Lövy: Fish-farm-associated amoebae in freshwater ecosystems revealed by eDNA metabarcoding . 14

Ivana Mikulíková: Mass mortality in bighead carp (*Hypophthalmichthys nobilis*) linked to amphizoic amoebae gill infection and metabolic derangements 15

Ľubomír Pojezdal: Mortality of resident eels following the stocking of juvenile eels – case report 16

Jitka Kolářová: Treatment of massive gyrodactylosis infection (*Gyrodactylus luciopercae*) in intensive farming (RAS) of juvenile pikeperch (*Sander lucioperca*) – case report 17

Patrícia Kóczán: Bacterial enteritis associated with *Aeromonas dhakensis* in African catfish (*Clarias gariepinus*) fingerlings in Hungary 18

Mikolaj Adamek: Not so fancy seeing you here – first detection of fancy-tailed guppy virus in Europe 19

Harriet Nketiah Birikorang: Case study of suppressed growth in the pacific oyster (*Crassostrea gigas*) 20

Alena Balážová: In field LAMP diagnostics of trout diseases 21

Lucie Komárková: Optimalization and stabilization of LAMP mastermix through systematic evaluation of reaction additives 22

Kristína Plevová: Immunohistochemical approaches in fish intestinal and hepatic research: a comparative analysis of methodological variability 23

Eliška Zusková: Treatment strategy of diplostomosis in grass carp 24

Veronika Piačková: Fish health management in Czech and Moravian ponds from the perspective of fish farmers 25

Tomáš Korytář: Untargeted flow cytometry of teleost blood for predictive profiling of systemic immune activation 26

Jiří Kyslík: Architects of Immunity: The FcR/FcRL profiling in common carp and zebrafish models 27

Mikolaj Adamek: Simultaneously measuring immune responses and the presence of multiple pathogens supports the diagnosis of gill diseases in common carp and rainbow trout 28

Fumio Takizawa: Thymic and mucosal distribution of distinct CD4-positive T cell populations in rainbow trout 29

Shinji Takeda: Detection of antigen-specific T-cell responses and antigen presentation in ginbuna crucian carp: an in vitro assay based on cytokine gene expression 30

POSTER PRESENTATIONS

Réka Vámosi: Comparative assessment of water mould (Oomycota, Saprolegniales) diversity using culture-based isolation and eDNA metabarcoding 31

Kateřina Karbusová: Effect of pollutants with thyroid receptor antagonistic activity on fish and amphibians 32

Bharath Raja: Circulating IgM form reveal temperature and vaccination dependent remodeling of the teleost antibody response 33

Ivona Toulová: *Edwardsiella tarda* infection in African catfish (*Clarias gariepinus*) under experimental conditions 34

Lucie Všetičková: Feed additives enhancing fish performance and gut health 35

Ivana Papežíková: Does genetic resistance of fish guarantee protection against infectious diseases? 36

Leona Nepejchalová: The use of medicinal products in fish veterinary practice 37

Miroslava Palíková: Are fish farms producing stock salmonid fish a source of pathogens to open waters? 38

Ryosuke Fujihara: Investigation of S1P-dependent lymphocyte migration mechanism in teleost fish using FTY720 39

Chenkai Shao: Fugu secretory IgD and its receptor on the basophils 40

Věra Vaibarová: Antimicrobial activity of essential oils against fish-pathogenic bacteria 41

Yasuhiro Shibasaki: Attenuation of IgM antibody response by splenectomy in rainbow trout 42

Thilini Hettiarachchi: Lectin-based immunophenotyping reveals species-specific glycosylation patterns and hidden leukocyte heterogeneity in teleost fish 43

Mikolaj Adamek: Black hole of the unknown: results on hyperpigmentation in the common dab (*Limanda limanda*) 44

Mikolaj Adamek: Risk assessment of fish diseases in Armenia: aquaculture growth, transboundary trade, and native fish conservation 45

Monika Němcová: Common carp leukocytes restimulated with carp edema virus produce significantly more cytokines in a memory-like response 46

Mikolaj Adamek: Characteristic behavioural traits associated with infection caused by the carp edema virus and predator attacks as indicators for automated early-warning systems 47

PRACTICAL BENEFITS OF OUR PARTICIPATION IN THE EAFP MEETING 2024

Petr Posel

PETRA – AQUA, s.r.o. Prague, Czech Republic

In 2024, I attended the EAPF Meeting for the first time. It proved to be an extremely valuable experience for both me and our company, and I would like to share how the knowledge we gained has influenced our production, husbandry, storage, and distribution of ornamental fish.

I attended the meeting with three main objectives: to learn about the latest developments in fish disease research, to establish new professional contacts, and to seek solutions to several specific health issues affecting particular species of ornamental fish.

I was genuinely impressed by the current state of fish disease research. I should mention that I had not been actively involved in this field for more than twenty years, and it was evident how far the discipline has advanced. Many of the presented findings have considerable potential for practical application in the ornamental fish industry. Our sector is highly fragmented and often underestimates the importance of fish health management. As a result, disease control still largely relies on traditional methods such as formalin–malachite green-methylene blue treatments (FMC) and salt baths. While these methods remain useful, they are far from sufficient for addressing all health issues. Modern diagnostic techniques, preventive measures, and treatment options have advanced significantly, yet they are still only rarely applied in the ornamental fish sector.

Another major benefit of the meeting was the opportunity to establish new professional contacts. Events such as the EAFP Meeting provide an ideal platform for connecting researchers with industry professionals. I would therefore like to thank the organizers for making the meeting accessible not only to scientists but also to practitioners interested in fish health. I am equally grateful to the participants for their willingness to answer questions from a non-academic perspective and for their efforts to translate the results of basic research into practical applications.

The most valuable outcomes, however, were the concrete changes we implemented in our company.

1. Complete restructuring of our angelfish breeding facility: Based on information obtained during the meeting and subsequent laboratory analyses made possible through our newly established contacts, we decided to completely discontinue our existing angelfish (*Pterophyllum scalare*) breeding unit. We built a new, fully separated breeding facility and restarted production using broodstock originating from a highly isolated source. Closing a commercial breeding operation with 150 breeding pairs was a difficult decision, but the practical results have clearly demonstrated that it was the right one.

2. Modification of our production system: We comprehensively revised our ornamental fish production system by improving biosecurity and hygiene standards, optimizing fish nutrition, and shortening the production cycle until market size. This reduced the period during which fish are exposed to potential pathogens within our facilities and has resulted in a significant improvement in their overall health status.

3. Improved monitoring of imported fish: We introduced a more systematic evaluation of imported fish from our international suppliers, with particular attention to guppies (*Poecilia reticulata*) and goldfish (*Carassius auratus*). Long-term monitoring enabled us to identify suppliers whose production repeatedly caused health problems during quarantine and subsequent distribution. As a result, we have been able to eliminate some of these suppliers from our supply chain.

Fish diseases remain a complex issue that requires continuous attention and further research. Nevertheless, I believe that our experience clearly demonstrates that transferring scientific knowledge into everyday ornamental fish practice is not only possible but also highly beneficial. Closer cooperation between research institutions and the ornamental fish industry can substantially improve fish health, production quality, and the long-term sustainability of the sector.

Key words: practical benefit; EAFP; ornamental fish production

MODELING TILAPIA LAKE VIRUS (TiLV) AND FANCY-TAILED GUPPY VIRUS (FTGV) INFECTIONS IN ZEBRAFISH

Krzysztof Rakus[1], Miriam Mojzesz[1], Maria Zawisza[1], Zaneta Baran[1], Justyna Starzyk[1], Matylda Cyganik[1], Mateusz Niemiec[1], Magdalena Chadzinska[1], Mikolaj Adamek[2]

[1]Jagiellonian University, Faculty of Biology, Institute of Zoology and Biomedical Research, Department of Evolutionary Immunology, Krakow, Poland; [2]University of Veterinary Medicine, Institute for Parasitology, Fish Disease Research Unit, Hannover, Germany

Zebrafish (*Danio rerio*) is a well-established model organism extensively used in biomedical research, including studies of immune responses and host-pathogen interactions. It also provides a valuable model for investigating viral diseases of significance to aquaculture. In recent years, we have established zebrafish models of infection with two recently emerged fish viruses: Tilapia Lake Virus (TiLV) and Fancy-Tailed Guppy Virus (FTGV). Both TiLV and FTGV are negative-sense, single-stranded RNA viruses belonging to the family *Amnoonviridae.* TiLV causes high mortality in Nile tilapia (*Oreochromis niloticus*), while FTGV has recently been identified as the etiological agent of disease in fancy-tailed guppies (*Poecilia reticulata*).

We demonstrated that both TiLV and FTGV replicate in zebrafish larvae after infection by microinjection and induce the upregulation of genes involved in the type I interferon (IFN) response, pro-inflammatory cytokines, and markers of macrophages/microglia and astrocytes. Furthermore, TiLV infection resulted in increased mortality of zebrafish larvae. In adult zebrafish, we established a TiLV infection model using intraperitoneal injection. We demonstrated that TiLV persists in the brain for at least 90 days, even when the virus is no longer detectable in peripheral organs. During infection, zebrafish exhibited pronounced sickness behavior, characterized by reduced locomotor activity, decreased food intake, and a preference for the bottom zone of the aquarium. Gene expression analysis revealed that TiLV induces a robust antiviral and inflammatory response in the adult zebrafish brain and upregulates the expression of genes associated with microglia/macrophage markers. Finally, using zebrafish larvae, we demonstrated that TiLV infection activates microglia, as evidenced by a morphological transition from a resting, ramified phenotype in mock-infected larvae to a highly amoeboid, activated phenotype in TiLV-infected larvae.

In conclusion, zebrafish represent a valuable model for studying host–pathogen interactions during TiLV and FTGV infections, providing new insights into viral biology and the mechanisms underlying antiviral responses.

Key words: host-pathogen interactions; viral persistence; aquaculture virology

Acknowledgement: This work was supported by the National Science Centre of Poland (Grant number UMO-2015/18/E/NZ6/ 00516 and UMO-2023/51/B/NZ6/02066).

INTEGRATED ANALYSIS OF STRAIN-SPECIFIC HOST RESPONSES AND GILL MICROBIOME DYNAMICS DURING CYPRINID HERPESVIRUS-3 INFECTION IN CARP

Anju Sebastian[1], Mikolaj Adamek[2], Anna Martyrosyan[1], Ľubomír Pojezdal[3], Kateřina Matějíčková[3], Veronika Piačková[1]

[1]University of South Bohemia in České Budějovice, Faculty of Fisheries and Protection of Waters, South Bohemian Research Center of Aquaculture and Biodiversity of Hydrocenoses, Vodňany, Czech Republic; [2]University of Veterinary Medicine Hannover, Fish Disease Research Unit, Hannover, Germany; [3]Veterinary Research Institute, Department of Infectious Diseases and Preventive Medicine, Brno, Czech Republic

Cyprinid herpesvirus-3 (CyHV-3), the causative agent of koi herpesvirus disease, is a major viral pathogen responsible for significant economic losses in carp aquaculture. Although differences in susceptibility among carp strains have been reported, the relationships between viral replication, immune responses, and microbiome alterations remain poorly understood. In this study, we compared the responses of koi carp, Hungarian mirror carp (M2), a hybrid of Hungarian mirror carp and Amur mirror carp (M2×AL), Amur wild carp (AS), and grass carp following the infection with CyHV-3 Viral load, immune-related gene expression (*igm, mx2, il1β,* and *gzmA*), virus-specific IgM antibody production, and gill bacterial microbiota were evaluated throughout the infection. Viral DNA was detected in all fish groups; however, clear strain-dependent differences were observed. Hungarian mirror carp (M2) exhibited the highest viral load during the early stage of infection, while koi and M2×AL maintained high viral loads at 14 and 21 days post-infection, indicating prolonged viral replication. In contrast, AS consistently displayed lower viral loads, and grass carp maintained minimal viral DNA throughout the experiment, suggesting strong resistance or limited viral replication.

Immune responses also varied among the carp strains. The antiviral gene *mx2* was strongly induced in all groups, with the highest expression observed in koi during the peak of infection. Expression of *igm* increased progressively throughout infection, whereas *il1β* showed sustained upregulation, particularly in M2 and koi, indicating prolonged inflammatory responses. The cytotoxic marker *gzmA* exhibited delayed but pronounced upregulation in AS and M2 during the later stages of infection. Virus-specific IgM antibodies gradually increased in all groups, with koi displaying the strongest humoral response and AS showing comparatively lower antibody levels by the end of the experiment. CyHV-3 infection also altered the gill bacterial community, resulting in shifts in the relative abundance of dominant bacterial phyla and species between pre-infection and 14 days post-infection, although the magnitude of these changes differed among carp strains. Collectively, these findings demonstrate distinct strain-dependent differences in susceptibility to CyHV-3, with grass carp and AS exhibiting greater resistance than koi, M2, and M2×AL. The coordinated differences in viral replication, immune activation, antibody production, and microbiome composition provide new insights into host–virus interactions and identify potential traits for breeding CyHV-3-resistant carp. strains.

Key words: CyHV-3, gill microbiome, carp strains, immune response

Acknowledgement: This study was financially supported by the Ministry of Education, Youth and Sports of the Czech Republic, the projects PROFISH (CZ.02.1.01/0.0/0.0/ 16_019 / 0000869) and OP JAK – project Aquaculture for Future (CZ.02.01.01/00/23_021/0012616).

PATHOLOMICS OF KOI SLEEPY DISEASE IN COMMON CARP CAUSED BY THE CARP EDEMA VIRUS INFECTION

Mikolaj Adamek[1], Maria Zawisza[2], Alberto Falco[3], Tomáš Korytář[4], Andrzej Ciereszko[5], Veronika Piačková[6], Verena Jung-Schroers[1], Krzysztof Rakus[2], Mariola Dietrich[5]

[1]University of Veterinary Medicine Hannover, Fish Disease Research Unit, Hannover, Germany; [2]Jagiellonian University, Faculty of Biology, Institute of Zoology and Biomedical Research, Department of Evolutionary Immunology, Krakow, Poland; [3]Spanish National Research Council (IATS-CSIC), Institute of Aquaculture Torre de La Sal, Fish Pathology Group, Cabanes, Castellón, Spain; [4]Czech Academy of Sciences, Biology Centre, Institute of Parasitology, Laboratory of Fish Immunology, České Budějovice, Czech Republic; [5]Polish Academy of Sciences, InLife Institute of Animal Reproduction and Food Research, Gamete Biology Team, Olsztyn, Poland [6]University of South Bohemia, Faculty of Fisheries and Protection of Waters, South Bohemian Research Center of Aquaculture and Biodiversity of Hydrocenoses, Vodňany, Czech Republic

Patholomics is an integrative approach combining pathology and high-throughput omics to improve our understanding of disease mechanisms at molecular, cellular and tissue levels. In the current study, we employed this framework to elucidate the host-pathogen-environment interactions during gill disease caused by carp edema virus, which serves as a unique model for investigating the impact of gill diseases in common carp.

CEV-infected fish were selected to fulfil the following criteria: a high viral load (>10^5 viral genome copies per 250 ng of DNA); the development of classic clinical signs such as lethargy; and the presence of physiological signs of gill dysfunction, such as a decrease in sodium levels below 100 mmol/L. TMT-based proteomics and phosphoproteomics, as well as non-targeted metabolomics and lipidomics, were performed on the gills and blood plasma of these individuals, and the results were compared with those of non-infected controls in order to evaluate the biological pathways affected by the disease.

Coordinated multi-omics analysis of CEV infection showed proteomic signatures of infection-associated cellular remodelling, characterised by increased RNA/protein processing, proliferation and immune cell interaction, alongside the suppression of apoptotic and necrotic death pathways. These results were reinforced by phosphoproteomics, which indicated enhanced motility, cytoskeletal and vesicle trafficking dynamics, survival/proliferation signalling and immune cell recruitment. This was confirmed by metabolome results indicating a state of metabolic stress and immune-associated remodelling. The strongest signals were found in altered mitochondrial fatty acid oxidation, branched-chain amino acid catabolism, arginine/nitric oxide metabolism, nucleotide turnover, redox imbalance and methylation/osmolyte metabolism. While, the lipidome pattern supports redox-sensitive membrane remodelling, polyunsaturated fatty acid (PUFA)-containing phospholipid turnover, lysophospholipid signalling, and sphingomyelin-associated stress responses.

Together, these changes suggest that CEV pathology is associated with a pro-survival, anti-apoptotic and immune-remodelling phenotype, which may lead to impaired gill homeostasis, dysregulated host defence and progression towards severe gill disease.

Key words: common carp; proteomics; lipidomics; metabolomics; gill disease

Acknowledgements: This research was supported by Deutsche Forschungsgemeinschaft (DFG project number 426513195), by the National Science Centre of Poland (NCN project number UMO-2018/31/F/NZ6/02311) and by European Union's Horizon Europe Research and Innovation Programme under grant agreement 101084204 Cure4Aqua.

KOI HERPESVIRUS DISEASE IN THE CONTEXT OF THE ANIMAL HEALTH LAW – CURRENT STATUS AND DIAGNOSTIC METHODS

Kateřina Matějíčková[1], Jitka Motlová[1], Miroslava Palíková[1,2,3], Ľubomír Pojezdal[1]

[1]Veterinary Research Institute, Department of Infectious Diseases and Preventive Medicine, Brno, Czech Republic; [2]University of Veterinary Sciences Brno, Faculty of Veterinary Hygiene and Ecology, Department of Ecology and Diseases of Zoo Animals, Game, Fish and Bees, Brno, Czech Republic; [3]Mendel University in Brno, Faculty of AgriSciences, Department of Zoology, Fisheries and Hydrobiology, Brno, Czech Republic

Common carp (*Cyprinus carpio*) is the most important farmed fish in the Central and Eastern Europe. It is, together with ornamental koi carp (*C. rubrofuscus*), the only species susceptible to the koi herpesvirus disease (KHVD) caused by the *Cyvirus cyprinidallo-3* (also cyprinid herpesvirus-3 [CyHV-3] or koi herpesvirus [KHV]). Due to severe economic impact on the farmed and wild carp populations, KHVD is a WOAH listed disease and a notifiable disease under European legislation.

The EU Regulation 2016/429 on transmissible animal diseases (Animal Health Law, AHL) applicable since April 2021 has listed KHVD as Category E disease, a listed disease for which there is a need for surveillance within the Union. The surveillance in member countries is therefore dependent on the national legislation.

The current situation in Czechia, where the confirmed presence of virus without clinical signs of infection is currently not viewed as an outbreak site and treated as healthy fish by the veterinary authorities, will be compared with the biological status of virus persistence and reactivation confirmed both by standard (real-time PCR) and novel (LAMP) diagnostic methods.

The situation in other Central and Eastern European countries will also be discussed.

Key words: *Cyvirus cyprinidallo*; PCR; LAMP;

Acknowledgments: This work was supported by the Technology Agency of the Czech Republic Grant TN02000017.

LUST FOR BLOOD: PROTEASES-MEDIATED HEMATOPHAGY IN THE COMMON CARP PATHOGEN *SPHAEROSPORA MOLNARI*

Pavla Bartošová-Sojková[1], Tamás Dobai[1,2], Sagar Nayak[1], Filip Dyčka[2], Martin Horn[3], Lucia Bieliková[3], Barbora Kaščáková[1], György G. Ferenczy[4], Astrid S. Holzer[5], Zdeněk Franta[2]

[1]Czech Academy of Sciences, Biology Centre, Institute of Parasitology, České Budějovice, Czech Republic; [2]University of South Bohemia, Faculty of Science, Department of Chemistry, České Budějovice, Czech Republic; [3]Czech Academy of Sciences, Institute of Organic Chemistry and Biochemistry, Prague, Czech Republic; [4]HUN-REN Research Centre for Natural Sciences, Medicinal Chemistry Research Group and National Drug Discovery and Development Laboratory, Budapest, Hungary; [5]University of Veterinary Medicine, Fish Health Division, Vienna, Austria

Myxozoans are highly reduced cnidarian endoparasites with complex life cycles and specialized strategies for exploiting their hosts. Despite expanding genomic resources, myxozoan metabolism and nutrient acquisition remain poorly understood. The heme biosynthesis pathway (HBP) is essential for aerobic metabolism, yet many blood-feeding parasites acquire heme from host hemoglobin (Hb). Hb degradation is mediated by cysteine, aspartic, and metallopeptidases. *Sphaerospora molnari*, a myxozoan pathogen of common carp *Cyprinus carpio*, proliferates as motile blood stages (SMBS) and forms spores in gills and skin, causing anemia and osmoregulatory failure. SMBS attach to red blood cells (RBCs) and internalize Hb, suggesting that host RBCs provide amino acids and heme. Here, we characterize the hemoglobinolytic cascade of *S. molnari*, assess its HBP, and identify druggable proteolytic targets.

We combined genome, transcriptome, and proteome mining with qPCR profiling to identify genes involved in HBP, heme/iron-related handling, and Hb degradation. SMBS lysate activities were assessed using fluorogenic substrates, specific inhibitors, activity-based probes, and Hb digestion assays. The dominant cathepsin L-like cysteine peptidase, SmolCathL2, was recombinantly produced and characterized for substrate specificity, pH optimum, and Hb-cleaving activity. Antibodies against recSmolCathL2 were used for confocal and TEM immunogold localization. Its structure was solved by X-ray crystallography and used with docking/simulations to identify parasite-selective compounds.

S. molnari encodes all core HBP enzymes but shows reduced HBP gene expression in SMBS relative to sporogonic stages, suggesting lower reliance on *de novo* heme production during blood parasitism. No canonical heme oxygenase was identified, whereas genes linked to heme/iron transport and heme-binding redox functions were detected. SMBS proteomics identified 3,588 proteins, including 79 peptidases from 8 enzyme classes. Activity assays and inhibitor profiling showed that C1 cysteine and A1 aspartic peptidases dominate SMBS proteolytic activity. Hb digestion required both cathepsin L-like and cathepsin D-like peptidases: single-class inhibition partially blocked degradation, whereas combined inhibition abolished Hb cleavage. SmolCathL2 was the most abundant and active cathepsin L-like enzyme in SMBS and recSmolCathL2 showed acidic pH optimum, cleaved Hb *in vitro*, and localized to lysosome-like compartments. Its 2 Å structure revealed high similarity to host cathepsin Ls and showed parasite-specific Ser-206 near the active site, used for selective inhibitor design.

S. molnari combines endogenous HBP with host Hb exploitation during SMBS development. Its hemoglobinolytic cascade depends on coordinated cysteine and aspartic peptidase activity, resembling strategies of other hematophagous parasites. SmolCathL2 is the first recombinant and structurally characterized protease from a myxozoan parasite and a promising target against this pathogen.

Key words: Proteases; Myxozoa; *Sphaerospora*; common carp; blood feeding

Acknowledgements: This study was supported by the Grant Agency of the Czech Republic (GAČR) project No. 24-13238L.

PARASITE FAUNA OF NON-NATIVE BULLHEAD CATFISH OF THE GENUS *AMEIURUS* IN CZECHIA

Terezie Maturová[1], Ivanna Dudliv[2], Ivona Toulová[1,3,4], Maria Yu Tkachenko[2], Miroslava Palíková[1,3,4], Markéta Ondračková[2]

[1]University of Veterinary Sciences Brno, Faculty of Veterinary Hygiene and Ecology, Department of Ecology and Diseases of Zoo Animals, Game, Fish and Bees, Brno, Czech Republic; [2]Czech Academy of Science, Institute of Vertebrate Biology, Brno, Czech Republic; [3]Mendel University in Brno, Faculty of AgriSciences, Department of Zoology, Fisheries and Hydrobiology, Brno, Czech Republic; [4]Veterinary Research Institute, Department of Infectious Diseases and Preventive Medicine, Brno, Czech Republic

Bullhead catfish of the genus *Ameiurus* are non-native fish species in Europe, originating from freshwater habitats across North America. The brown bullhead (*Ameiurus nebulosus* Le Sueur, 1819) and the black bullhead (*Ameiurus melas* Rafinesque, 1820) represent the most frequently recorded species of this genus in European waters. Their introduction into natural ecosystems has raised concerns regarding potential impacts on native fish communities from both ecological and health perspectives, including the transmission of diseases caused by various etiological agents, parasites among them. To date, a wide range of parasites of varying ecological and veterinary importance has been reported in these non-native catfish species.

The objective of this study was to evaluate the parasite fauna of non-native bullhead catfish (*A. nebulosus* and *A. melas*) in Czechia and to assess possible differences between the two host species. Fish were collected from three localities where both species co-occur (Mlazické tůně, Vehlovická tůň, Medkovy rybníky) during spring and autumn 2024 and 2025. All major organs and tissues (skin, musculature, head, eyes, brain, barbels, fins, gills, visceral organs and swim bladder) were examined following standard parasitological procedure. Collected parasites were preserved depending on taxonomic group, while protozoans were assessed in fresh mounts.

Parasitological examination revealed representatives of several taxonomic groups. The detected parasites included: Ciliophora (*Trichodina* spp., *Ichthyophthirius multifiliis*), Monogenea (*Gyrodactylus melas*, *G. nebulosus*, *Ligictaluridus pricei*, *L. monticelli*), Arthropoda (*Neoergasillus japonicus*, *Ergasilus sieboldi*, *Lernaea cyprinacea*, *Argulus foliaceus*), Nematoda (*Anguillicola crassus*, *Raphidascaris acus*,), Trematoda (*Diplostomum pseudospathaceum*, *D. spathaceum*, *Hysteromorpha triloba*), Cestoda (*Ophiotaenia europaea*, *Bothriocephalus claviceps*, *Proteocephalus* sp.). Most parasite taxa occurred in both black and brown bullheads, and no significant differences in parasite prevalence were detected between the host species.

In conclusion, our results showed that the parasite communities of sympatric brown and black bullheads in Czechia were largely comparable. Both species harboured parasites native to the Czech region (e.g. *R. acus*), parasites originating from their native North American range (e.g. *G. nebulosus*, *G. melas*), as well as parasites that are non-native to both regions (e.g. *A. crassus, N. japonicus*). The potential role of the bullhead catfish in the transmission of *A. crassus* to its definitive host, the European eel (*Anguilla anguilla*), has been discussed in recent years; however, their actual involvement in this process remains unclear.

Key words: *Ameiurus nebulosus; Ameiurus melas;* black bullhead; brown bullhead; invasive species

Acknowledgments: This work was supported by the project FVHE/Pikula/2026ITA24.

BIOLOGICAL CONTROL POSSIBILITIES OF FISH-PATHOGENIC WATER MOULDS (OOMYCOTA, SAPROLEGNIALES)

Edit Eszterbauer[1], Noémi Erdei[1], Patrícia Kóczán[1], Réka Vámosi[1], Milán Farkas[2], Győző L. Kaján[1], Dóra Sipos[1], Domonkos Sváb[1], Edit Kaszab[3]

[1]HUN-REN Veterinary Medical Research Institute, Budapest, Hungary; [2]Department of Molecular Ecology, Institute of Aquaculture and Environmental Safety, Hungarian University of Agriculture and Life Sciences, Gödöllő, Hungary; [3]Department of Environmental Safety, Institute of Aquaculture and Environmental Safety, Hungarian University of Agriculture and Life Sciences, Gödöllő, Hungary

Saprolegniosis, mainly caused by *Saprolegnia* spp., is one of the most important diseases affecting freshwater aquaculture worldwide. The withdrawal of several chemical treatments and increasing environmental concerns have intensified the search for sustainable alternatives. Biological control using naturally occurring bacteria represents a promising strategy for reducing the impact of fish-pathogenic water moulds.

We investigated the diversity and anti-water mould activity of 24 aquatic *Pseudomonas* isolates originating primarily from Hungarian fish farms and natural waters. Isolates were characterised using *16S rRNA* and *rpoD* gene sequencing, MALDI-TOF MS protein profiling, and phenotypic assays, including siderophore production and biofilm formation. *In vitro* inhibition assays were performed against *Saprolegnia parasitica* using both bacterial cultures and cell-free supernatants. In addition, selected strains were evaluated *in vivo* using rainbow trout (*Oncorhynchus mykiss*) eggs challenged with *S. parasitica*.

Six *Pseudomonas* species were identified, including *Pseudomonas paracarnis*, *P. shahriarae*, *P. salmasensis*, *P. canadensis*, *P. chlororaphis* subsp. *aurantiaca* and *P. danubii*. The strongest inhibitory activity was observed among several *P. danubii* isolates, one P. *chlororaphis* isolate and *P. canadensis* B379-HU. Exposure to inhibitory compounds induced characteristic morphological alterations in *S. parasitica* hyphae, including branching, nodule formation, fragmentation and eventual hyphal death. Two candidate strains, *P. danubii* PSE207KE and *P. canadensis* B379-HU, were further validated in rainbow trout egg experiments and significantly reduced oomycete colonisation without detectable toxicity to the developing embryos. Both strains exhibited siderophore production, moderate biofilm-forming capacity and strong extracellular inhibitory activity. The identification of the responsible metabolites is currently underway using HPLC-MS-based metabolomic analyses.

Our findings demonstrate that aquatic *Pseudomonas* species constitute a valuable source of biological control agents against fish-pathogenic oomycetes. The inhibitory effects observed in *in vivo* experiments highlight the practical applicability of these strains for future hatchery management. Extracellular inhibitory compounds, rather than the direct application of live bacteria, may offer a safe and environmentally sustainable alternative for controlling saprolegniosis in aquaculture.

Key words: saprolegniosis biocontrol; antagonistic bacteria; phylogeny; inhibitory activity; *in vivo* experiments.

Acknowledgement: This work was supported by the National Research, Development and Innovation Office (NKFIH), Hungary, under grant no. K141889.

EARLY eDNA DETECTION AND TARGETED ORAL METAPHYLAXIS FOR THE CONTROL OF *ICHTHYOPHTHIRIUS MULTIFILIIS* IN SALMONID FISH

Ľubomír Pojezdal[1], Jitka Motlová[1], Zuzana Mikulková[2], Karel Tittl[2], Martin Jeřábek[2], Kateřina Matějíčková[1]

[1]Veterinary Research Institute, Department of Infectious Diseases and Preventive Medicine, Brno, Czech Republic; [2]Tekro, spol. s r.o., Prague, Czech Republic

The ciliate *Ichthyophthirius multifiliis* causes severe losses in intensive salmonid aquaculture worldwide. The lack of registered veterinary medicinal products requires the development of new preventive and therapeutic approaches. This study evaluates a combination of non-invasive monitoring using environmental DNA (eDNA) and targeted oral metaphylaxis. Early diagnosis of the parasite was tested using a qPCR assay targeting eDNA in water and sediment. Within oral therapy, the efficacy of five active substances was first evaluated. A subsequent experiment focused on verifying the efficacy of doxycycline in experimentally infected brook trout (*Salvelinus fontinalis*), comparing a standard formulation (2 mg/g) with an innovative microencapsulated controlled-release formulation (0.8 mg/g).

Environmental DNA analysis reliably detected the pathogen in the rearing environment up to five weeks before first identification via light microscopy and the onset of clinical signs in the fish. Among the five evaluated substances, praziquantel and doxycycline showed the highest therapeutic potential, whilst the remaining substances did not provide a sufficient protective effect. In the trials with brook trout, the administration of microencapsulated doxycycline resulted in a survival rate of 84.2%, representing a statistically significant increase in efficacy ($P = 0.004$) compared to the standard formulation, which yielded a 73.7% survival rate. A 100% mortality rate was recorded in the untreated control group. The innovative formulation achieved higher fish survival at a 2.5-fold lower dose of the drug in feed, which limits the release of residues into the aquatic environment and minimises the risk of antimicrobial resistance development. Combining early eDNA monitoring and targeted application of microencapsulated therapeutics under the veterinary cascade system offers a functional tool for the control of *Ichthyophthirius multifiliis* in aquaculture.

Key words: ICH; PCR; environmental DNA; doxycycline

Acknowledgments: This work was supported by the Ministry of Agriculture of the Czech Republic project No. RO0523 and by the Operational Programme Fisheries project CZ.08.02.01/01/25_024/0000774.

FISH-FARM-ASSOCIATED AMOEBAE IN FRESHWATER ECOSYSTEMS REVEALED BY EDNA METABARCODING

Alena Lövy[1], Eliška Lazárková[2], Martina Lisnerová[1], Hana Pecková[1], Ivan Fiala[1]

[1]Czech Academy of Sciences, Biology Centre, Institute of Parasitology, České Budějovice, Czech Republic; [2]University of South Bohemia, Faculty of Science, České Budějovice, Czech Republic

Free-living amoebae are a diverse assemblage of unicellular eukaryotes characterized by amoeboid movement and the formation of pseudopodia. Most freshwater amoebae belong to Amoebozoa, one of the major evolutionary lineages of eukaryotes, while additional amoeboid diversity is represented by groups such as Heterolobosea. They are widespread in freshwater ecosystems, where they regulate microbial communities through grazing on bacteria and other microorganisms and contribute to nutrient cycling. Some amoebae are also associated with fish and aquaculture environments, either as opportunistic pathogens or as reservoirs of microorganisms. Despite their ecological importance, the diversity and distribution of amoebae in fish-farming landscapes remain poorly understood.

We investigated amoebae communities in freshwater habitats subjected to different levels of aquaculture influence in South Bohemia, Czech Republic. Environmental DNA (eDNA) metabarcoding was applied to water and sediment samples collected from the Lužnice River and the Zlatá stoka channel, which is closely connected to the Třeboň fishpond system. Ten primer pairs targeting SSU rRNA and mitochondrial *cox1* markers were evaluated for their ability to detect amoebae. Sequencing was performed using Oxford Nanopore Technologies followed by bioinformatic and phylogenetic analyses.

The highest detection efficiency and specificity were achieved with *cox1*-targeting primers. Amoebozoa dominated the detected communities, particularly representatives of the genera *Parvamoeba* and *Cochliopodium*, while Heterolobosea were represented by a diverse assemblage of *Naegleria* species. Amoebae diversity was consistently higher in riverine sites than in locations influenced by intensive fish farming. In contrast, the Zlatá stoka channel exhibited reduced diversity, suggesting that aquaculture-associated environmental conditions significantly affect amoebae community structure. Phylogenetic analyses further indicated that many recovered lineages likely represent previously undescribed species.

Our results demonstrate that fish-farming activities can shape freshwater amoebae communities and highlight the value of eDNA metabarcoding for monitoring microbial eukaryotic diversity in aquaculture-impacted ecosystems. The detected diversity of amoebae emphasizes the need for further studies on their ecological roles and potential relevance to fish health.

Key words: amoebae; eDNA metabarcoding; aquaculture; freshwater ecosystems

Acknowledgement: This study was supported by the Ministry of Education, Youth and Sports of the Czech Republic (project no. LUAUS24281 to I.F.).

MASS MORTALITY IN BIGHEAD CARP (*HYPOPHTHALMICHTHYS NOBILIS*) LINKED TO AMPHIZOIC AMOEBAE GILL INFECTION AND METABOLIC DERANGEMENTS

Ivana Mikulíková[1,2,3], Jiří Pikula[1], Ivana Papežíková[1,2,3], Ivona Toulová[1,2,3], Ľubomír Pojezdal[3], Jitka Motlová[3], Miroslava Palíková[1,2,3]

[1]University of Veterinary Sciences Brno, Faculty of Veterinary Hygiene and Ecology, Department of Ecology and Diseases of Zoo Animals, Game, Fish and Bees, Brno, Czech Republic; [2]Mendel University in Brno, Faculty of AgriSciences, Department of Zoology, Fisheries and Hydrobiology, Brno, Czech Republic; [3]Veterinary Research Institute, Department of Infectious Diseases and Preventive Medicine, Brno, Czech Republic

We report a case of mass mortality involving 15 tons of market bighead carp (*Hypophthalmichthys nobilis*). Disease was first detected in early March 2026 in overwintered fish. Affected fish showed lethargy and gathered below the water surface in the central area of the storage pond. Necropsy revealed corneal opacity, periocular haemorrhages, haemorrhages on the ventral surface of the body, saprolegniosis, necrotic gills, and splenomegaly. In addition, viscous fluid was present in the body cavity, and the hepatopancreas exhibited yellow discoloration with haemorrhages.

Diseased specimens sampled in March exhibited significantly lower haemoglobin, mean corpuscular haemoglobin, total protein, albumin, cholesterol, and alanine aminotransferase levels. There were significant differences between control healthy fish and diseased fish sampled in April in mean corpuscular volume, methaemoglobin, white blood cell count, phagocytosis peak and integral, calcium, creatinine, triglycerides, lactate, lactate dehydrogenase, and alanine aminotransferase.

In March, a mixed bacterial population was isolated from the gill samples, with no single species predominating. Bacteriological cultivation performed in April detected mixed microbiota in the gill samples including *Aeromonas sobria*, which may act as an opportunistic pathogen.

Virological examination detected no cultivatable viruses on the EPC and BF-2 cell lines at 16 °C and 24 °C. The samples were negative for the presence of SVCV and Grass carp reovirus using PCR.

Microscopic lesions were characterized by proliferation of the gill epithelium which resulted in adhesions between secondary lamellae and the formation of compact cellular blocks in most gill filaments. The extensive disruption of gill architecture likely impaired respiratory function, explaining the mass mortality observed. Histopathological changes were consistent with an advanced or terminal stage of amoebic gill disease. However, at the time of sampling, the gill samples contained only a minimal number of surviving amoeboid organisms.

Two scenarios may account for amoeba colonisation of the gills: 1) colonisation occurred at the end of the active season, and fish with affected gills survived the winter period by shifting to a resting state with minimal metabolic requirements, 2) massive colonisation occurred during the overwintering period at the bottom of the tanks due to the spread of amoebae from the surrounding environment.

Key words: cyprinid fish; haematology, biochemistry; gross and microscopic pathology

Acknowledgments: Financial support was provided by the Ministry of Agriculture of the Czech Republic project No. RO0523 and the project FVHE/Pikula/V3U2026.

MORTALITY OF RESIDENT EELS FOLLOWING THE STOCKING OF JUVENILE EELS – CASE REPORT

Ivana Mikulíková[1,2,3], Ľubomír Pojezdal[3], Kateřina Matějíčková[3], Ivana Papežíková[1,2,3], Ivona Toulová[1,2,3], Miroslava Palíková[1,2,3]

[1]University of Veterinary Sciences Brno, Faculty of Veterinary Hygiene and Ecology, Department of Ecology and Diseases of Zoo Animals, Game, Fish and Bees, Brno, Czech Republic; [2]Mendel University in Brno, Faculty of AgriSciences, Department of Zoology, Fisheries and Hydrobiology, Brno, Czech Republic; [3]Veterinary Research Institute, Department of Infectious Diseases and Preventive Medicine, Brno, Czech Republic

Approximately one month after juvenile eels with a mean body weight of 5 g were introduced to several stocking sites, mortality primarily affecting larger resident eels measuring 20–60 cm, but not the freshly stocked fish, was observed. Affected eels were lethargic and frequently congregated near water inflows. At some sites, mortality was restricted to eels, while at others several fish species were affected. Environmental conditions varied among the investigated sites. Water temperature above 28 °C, low dissolved oxygen concentrations and high organic loads were recorded at some locations, whereas these factors were less pronounced or absent at others.

Examined live fish underwent parasitological, bacteriological and virological examinations. Parasitological examination of three live fish revealed the presence of *Argulus* sp. on the skin, *Pseudodactylogyrus* sp. and *Trichodina* sp. on the gills and *Anguillicola crassus* in the swim bladder at low to moderate intensities. Bacteriological culture predominantly yielded *Aeromonas veronii* from the skin, gills and spleen. Some of the specimens were delivered decomposing and therefore examined only for the presence of *Anguillicola crassus* in the swim bladder. The presence of the parasite was confirmed, but without reliable intensity measurements.

To assess potential infectious agent introduction, live eels originating from the same source batch as stocked fish underwent comprehensive examination. Parasitology revealed presence of *Pseudodactylogyrus* sp. in the gills and bacteriological culture yielded *Aeromonas hydrophila* from the gills of most examined fish and from the spleen of some fish.

Virological examination is ongoing, with all samples testing negative for cultivatable viruses on EPC and BF-2 cell lines at both 16 °C and 24 °C and preliminary PCR results showing the presence of *Birnaviridae* in one of the samples.

Key words: *Anguilla anguilla*; fish stocking; monogeneans; *Aeromonas* sp.; *Birnaviridae*

Acknowledgement: This work was supported by the Ministry of Agriculture of the Czech Republic project No. RO0523 and the project FVHE/Pikula/2026ITA24.

TREATMENT OF MASSIVE GYRODACTYLOSIS INFECTION (*GYRODACTYLUS LUCIOPERCAE*) IN INTENSIVE FARMING (RAS) OF JUVENILE PIKEPERCH (*SANDER LUCIOPERCA*) – CASE REPORT

Jitka Kolářová[1], Tomáš Pěnka[1], Oleksandr Malinovskyi[1], Václav Kučera[1], Andrea Lepičová[1], Markéta Ondračková[2], Tomáš Policar[1]

[1]University of South Bohemia in České Budějovice, Faculty of Fisheries and Protection of Waters, South Bohemian Research Centre of Aquaculture and Biodiversity of Hydrocenoses, Vodňany, Czech Republic; [2]Institute of Vertebrate Biology of the Czech Academy of Sciences, Brno, Czech Republic

In this study, we present a case of treating a severe outbreak of gyrodactylosis in juvenile pikeperch (*S. lucioperca*) reared in a recirculating aquaculture system (RAS). During routine monitoring of juvenile pikeperch weighing 15.8 ± 1.78 g, a natural massive parasitic infection with the monogenean *Gyrodactylus luciopercae* was diagnosed. The nursing staff repeatedly applied a long-term formaldehyde bath, but the treatment proved ineffective. This prompted the development and evaluation of an alternative treatment protocol tailored to this specific outbreak. The first step was a fish safety and efficacy test using selected antiparasitic drugs recommended for the treatment of monogenean infections (salt, formaldehyde, fenbendazole, levamisole hydrochloride (LHC), Indian almond tree leaves). The results showed that, in terms of both antiparasitic effect and fish safety, the LHC bath was the most suitable option for treating the current gyrodactylosis outbreak in this pikeperch breeding. During the 40-day monitoring of the fish health status, it became that the application of LHC bath followed by water change was not sufficient, and reinfection occurred. Our experience from this case indicates that managing a massive infection of *G. luciopercae* requires combining an effective therapeutic bath with thorough water treatment in the rearing system, ideally through repeated water exchange. Although the short-term treatment LHC bath (2 hours) effectively removed parasites from the fish, some individuals were probably only immobilized and subsequently contributed to reinfection.

Key words: RAS; pikeperch; monogenean infection; antiparasitic treatment; levamisole

Acknowledgements: This study was supported by the Ministry of Agriculture of the Czech Republic (project NAZV QL26020013), and by the Ministry of Education, Youth and Sports of the Czech Republic, project Biodiversity (CZ.02.1.01./0.0/0.0/16_025/0007370).

BACTERIAL ENTERITIS ASSOCIATED WITH *AEROMONAS DHAKENSIS* IN AFRICAN CATFISH (*CLARIAS GARIEPINUS*) FINGERLINGS IN HUNGARY

Patrícia Kóczán[1], Noémi Erdei[1], Réka Vámosi[1], Péter Bársony[2], Andor Doszpoly[1], Edit Eszterbauer[1]

[1]HUN-REN Veterinary Medical Research Institute, Budapest, Hungary; [2]University of Debrecen, Faculty of Agricultural and Food Sciences and Environmental Management, Institute of Animal Science, Biotechnology and Nature Conservation, Department of Animal Nutrition, Debrecen, Hungary

Enteritis is among the most common health disorders and is frequently associated with improper feeding practices or deteriorating water. In addition, various pathogenic microorganisms, particularly bacteria such as *Aeromonas* spp. and *Clostridium* spp., as well as environmental toxins, may affect fry and fingerlings, especially when their immune systems are compromised. In Hungary, the African catfish (*Clarias gariepinus*) and its hybrids represent one of the most important species cultured in intensive aquaculture systems, primarily due to their high adaptability, stress tolerance, and robust survival capacity.

Following the increased mortality in an African catfish hybrid stock (*C. gariepinus* × *Heterobranchus longifilis*) in Hungary, affected fingerlings (7–8 cm total length) were submitted to our laboratory for diagnostic examination. Necropsy revealed noticeable intestinal distension, multifocal petechial haemorrhages of the intestinal wall, and extensive inflammation involving a large portion of the intestine. Samples obtained from the affected intestinal mucosa were subjected to bacteriological examination by cultivation on Columbia blood agar under both aerobic and anaerobic conditions. After subculturing, biochemical profiling was carried out with the API20NE test kit. Molecular species identification was performed using *16S rRNA* and *rpoD* gene sequencing, and phylogenetic relationships were analysed using the maximum-likelihood method with RAxML-ng. The isolates were identified as *Aeromonas dhakensis*: *16S rRNA* gene analysis showed 100% identity with the *A. dhakensis* strain B88 (KU041796) from striped catfish in Vietnam, *A. dhakensis* strain b2-100 (ON533364) from zebrafish in China and strain AHAPRM2 (PZ263896) from a sick fish in India. While *rpoD* gene analysis confirmed complete identity with the *A. dhakensis* strain UGH41 (LC547029), a human case from Japan.

Our findings suggest that the enteritis observed in African catfish fingerlings was most likely associated with *A. dhakensis* infection. This represents the second reported isolation of *A. dhakensis* from African catfish worldwide, with the isolate showing 99.21% sequence similarity to strain C83c (OK058317), previously identified in African catfish from Nigeria. Notably, the clinical manifestations of enteritis were comparable in both cases. As one of the most virulent species within the genus *Aeromonas*, *A. dhakensis* appears to be widely distributed across diverse aquatic environments. To our knowledge, this is the first report of *A. dhakensis* isolated from African catfish in Europe and the first documented occurrence of this pathogen in Hungary.

Key words: *Aeromonas* spp; bacterial enteritis; African catfish; phylogeny, *16S rRNA* gene, *rpoD*; biochemical profiling

NOT SO FANCY SEEING YOU HERE – FIRST DETECTION OF FANCY-TAILED GUPPY VIRUS IN EUROPE

Mikolaj Adamek[1], Tamara Ramirez Stoll[1], Dipa Debnath[1], Torben Krebs[1], Alice Westphal[1], Tomáš Korytář[2], Miroslava Palíková[3,4,5], Ľubomír Pojezdal[5], Alberto Falco[6], Esteban Soto[7], Win Surachetpong[8], Krzysztof Rakus[9], Verena Jung-Schroers[1]

[1]University of Veterinary Medicine Hannover, Institute for Parasitology, Fish Disease Research Unit, Hannover, Germany; [2]Czech Academy of Sciences, Biology Centre, Institute of Parasitology, Laboratory of Fish Immunology, České Budějovice, Czech Republic; [3] University of Veterinary Sciences Brno, Faculty of Veterinary Hygiene and Ecology, Department of Ecology and Diseases of Zoo Animals, Game, Fish and Bees, Brno, Czech Republic; [4]Mendel University in Brno, Faculty of AgriSciences, Department of Zoology, Fisheries and Hydrobiology, Brno, Czech Republic; [5]Veterinary Research Institute, Department of Infectious Diseases and Preventive Medicine, Brno, Czech Republic; [6]Spanish National Research Council (IATS-CSIC), Institute of Aquaculture Torre de La Sal, Fish Pathology Group, Cabanes, Castellón, Spain; [7] University of California, School of Veterinary Medicine, Department of Medicine and Epidemiology, Davis, California, USA; [8] University of Stirling, Institute of Aquaculture, Stirling, Scotland UK; [9] Jagiellonian University, Faculty of Biology, Institute of Zoology and Biomedical Research, Department of Evolutionary Immunology, Krakow, Poland

The Fancy-Tailed Guppy Virus (FTGV) is a pathogen of the guppy (*Poecilia reticulata*) and most likely a new member of the emerging *Amnoonviridae* family of viruses, which are proving to be some of the most virulent and dangerous for fish. Until now, the virus has been detected in the USA in fish populations originating from Asia and South America, in both clinically affected and healthy individuals. As the guppy is widely used as an ornamental pet and a biological model, health surveillance programmes are needed.

Samples were collected from guppies presented for examination over one month after a disease outbreak leading to a large-scale die-off. These fish were collected from an undisclosed ornamental fish retailer and originated from an ornamental fish breeder in Asia. Additional samples were collected from clinically healthy, unrelated, laboratory-raised experimental populations with no history of health problems, originating from South America. Classical virological and molecular methods were employed in combination with metabarcoding of bacterial populations and immune response profiling to identify the agent causing the disease and mortality.

Two populations originating from the disease outbreak were confirmed to be positive for FTGV, with high or moderate Ct values indicating persistent subclinical infection. The virus was re-isolated using E-11 cells. Partial genome sequencing and phylogenetic analysis revealed significant variation in the FTGV isolates. Furthermore, an indication of the involvement of pathogenic bacteria was detected. Evaluation of immune responses was not helpful due to the late subclinical phase of infection(s). The laboratory-raised experimental populations were confirmed to be FTGV-free.

This study confirms the presence of FTGV in guppy populations associated with a major mortality event. It highlights the potential for persistent subclinical infection and significant genetic variability of the virus and emphasises the need for targeted surveillance and biosecurity measures in the ornamental fish trade.

Key words: *Amnoonviridae*; guppy; *Poecilia reticulata*

Acknowledgements: The research was supported by European Union's Horizon Europe Research and Innovation Programme under grant agreement 101084204 Cure4Aqua.

CASE STUDY OF SUPPRESSED GROWTH IN THE PACIFIC OYSTER (*CRASSOSTREA GIGAS*)

Harriet Nketiah Birikorang, Mengjia Jiang, Mike Williams, Ernest O. Chuku, Andrew Trotter, Ivona Mladineo

Institute for Marine and Antarctic Studies (IMAS), University of Tasmania, Hobart, Tasmania, Australia

Introduction: Declining oyster production and unexplained mortality in Tasmania, Australia, have intensified investigations into oyster health. Interactions among the haemolymph microbial community, the host and the environment can hold cues to this decline in oyster farming productivity. This study characterises the haemolymph microbiome as an indicator of oyster health across thriving and declining oyster farms.

Methodology: Oysters were deployed to Pipe Clay Lagoon (PCL) sites 1 and 2, Pitt Water (PW), and Boomer Bay (BB) in December 2024, and haemolymph was sampled during performance assessments in January, March, and April 2025. DNA was extracted from 60 samples using a high-salt lysis protocol with proteinase K digestion and RNase treatment, followed by purification and quantification (Qubit). A subset of 24 samples (analysed in duplicate per time point) was selected for sequencing. Nanopore whole-genome sequencing was performed using a ligation-based native barcoding approach (Native Barcoding Kit 24 V14; SQK-NBD114.24), and taxonomic classification of reads (~8.4 million) was conducted using Kraken2 within the Galaxy platform. Microbial diversity analyses were conducted in Galaxy using taxonomically classified reads. Alpha diversity was assessed using the Shannon index, and beta diversity using Bray–Curtis dissimilarity to evaluate spatial and temporal variation in microbial community structure.

Results: Taxonomic profiling revealed site-specific variation in bacterial communities across sites and over time (Jan–Apr), though this was unaligned with growth, which followed BB>PCL2>PW>PCL1. *Escherichia coli* and *Acinetobacter baumannii* were ubiquitous, detected at all sites, but their relative abundances varied across samples. Higher counts were common in PCL and PW. The two PCL sites showed similar community structures, characterised by recurrent detection of *E. coli*, *Acinetobacter*, and *Actinobacteria*, whereas PW was distinguished by a higher contribution of marine-associated taxa such as *Halomonas* spp. Boomer Bay exhibited greater temporal variability, with taxa such as *Vibrio* appearing sparsely across time points. Other genera, including *Pseudomonas*, *Streptococcus*, and *Actinomyces*, were detected at low and variable abundances across sites and time points, whereas *Agrobacterium* was more consistently present at moderate levels. Viral sequences were detected at low levels, and approximately half of the reads remained unclassified. No single pathogenic taxon dominated the communities, and overall assemblages were consistent with commensal microbiota reported in healthy oysters. Alpha diversity (Shannon index) ranged from 0.67 to 1.09 and was comparable across sites and time points, with only minor temporal fluctuations. Beta diversity (Bray–Curtis) indicated substantial variation among samples (≈0.06–0.99), with lower dissimilarity within sites (particularly between PCL1 and PCL2) and higher dissimilarity between sites and time points. Despite this heterogeneity, the persistence of shared dominant taxa suggests an overall stable haemolymph microbiome, with no clear shifts associated with the observed suppression of growth in PCL oysters.

Conclusion: Collectively, these findings suggest that observed performance patterns are unlikely to be associated with infectious agents, as no dominant or pathogenic taxa were detected and the microbial assemblages were consistent with typical haemolymph microbiota. The relatively stable microbial community structure across sites and time points further supports the absence of infection-driven shifts, suggesting other environmental or physiological stressors could underlie the variable oyster performance. Physicochemical data confirmed that environmentally driven stress was associated with suppressed growth in PCL oysters.

Key words: *Pacific oyster (Crassostrea gigas)i*; haemolymph microbiome; suppressed growth; aquaculture health; environmental stress

IN FIELD LAMP DIAGNOSTICS OF TROUT DISEASES

Alena Balážová[1,2], Miroslava Palíková[1,2,3]

[1]University of Veterinary Sciences Brno, Faculty of Veterinary Hygiene and Ecology, Department of Ecology and Diseases of Zoo Animals, Game, Fish and Bees, Czech Republic; [2]Mendel University in Brno, Faculty of AgriSciences, Department of Zoology, Fisheries and Hydrobiology, Brno, Czech Republic; [3]Veterinary Research Institute, Infectious diseases and preventive medicine, Brno, Czech Republic

Proliferative Kidney Disease (PKD) is a parasitic disease of salmonid fish caused by the myxozoan *Tetracapsuloides bryosalmonae*. It primarily affects species like rainbow trout and brown trout, leading to kidney enlargement, anaemia, and high mortality, especially when water temperatures rise above 12–15 °C.

Infectious Hematopoietic Necrosis (IHN) is a highly contagious viral disease of salmonid fish, caused by a rhabdovirus targeting the kidney and spleen. It leads to severe disease outbreaks, especially in young fish, with mortality often reaching 90% or more in fry.

The aim of this research is to develop a reliable tool for in situ detection of both pathogens using loop-mediated isothermal amplification (LAMP) method. It is an amplification method that works at a constant temperature (60–65 °C), eliminating the need for thermal cycling. It uses 4–6 primers targeting an area of DNA forming single strand loop formation. Compared to traditional end-point PCR, LAMP method is much faster (typically the result is observed in 20–30 minutes).

For both detection methods we used portable Genie II machine (OptiGene, UK) and ISO-004 mastermix (OptiGene, UK) containing fluorescence dye. This mastermix can provide both DNA and RNA amplification. In PKD detection we used six LAMP primers targeting small sub-unit ribosomal RNA gene designed by El-Matbouli et al. (2005) This method was tested in laboratory using DNA isolate of fish confirmed to be positive by qPCR. We encountered a problem with inhibition in the samples isolated by standard kit and the samples have to be diluted 100× for the best assay performance. For the detection of IHN virus, we used a set of six primers designed by Suebsing et al. (2011) aiming at glycoprotein gene. Positive control was isolated using PrepMan^{TM} reagent from cell culture inoculated with the virus.

In the case of PKD sensitivity test was performed with diluted positive control (up to 10^{-6} dilution) to evaluate the assay´s sensitivity. The PKD method performed well even for samples diluted 10^{-5}. Both methods will be further tested during the diseases´ outbreaks to evaluate different in field isolation methods and their overall applicability in rapid detection. We also plan tests on detection of the pathogens´ e-DNA from infected ponds.

Key words: proliferative kidney disease; infectious hematopoietic necrosis; loop-mediated isothermal amplification

Acknowledgement: This work was supported by project TAČR TN02000017 (NaCeBiVet).

OPTIMIZATION AND STABILIZATION OF A LAMP MASTERMIX THROUGH SYSTEMATIC EVALUATION OF REACTION ADDITIVES

Lucie Komárková[1], Adam Norek[2], Petra Stejskalová[1], Ondřej Zítka[1]

[1]Mendel University in Brno, Faculty of AgriSciences, Department of Chemistry and Biochemistry, Brno, Czech Republic; [2]Veterinary Research Institute, Department of Infectious Diseases and Preventive Medicine, Brno, Czech Republic

Loop-mediated isothermal amplification (LAMP) is a rapid and sensitive nucleic acid amplification method with great potential for point-of-care diagnostics. However, the performance of a LAMP assay is strongly influenced by the composition of the reaction mixture. The aim of this study was to optimize and stabilize a LAMP mastermix through the systematic evaluation of reaction additives.

First, seven additives were evaluated individually to assess their effect on LAMP amplification performance. Each additive was tested at five different concentrations across a temperature gradient. Based on the screening results, the additives were classified into two functional groups: amplification enhancers and protective/stabilizing additives. Subsequently, the combination of the two most effective enhancers, betaine and formamide, was optimized, and sugar-based stabilizers were incorporated into the optimized formulation to increase mastermix stability while maintaining its performance.

Betaine exhibited the strongest amplification-enhancing effect, and its combination with formamide further improved the amplification efficiency of the LAMP assay. The incorporation of trehalose and sucrose increased mastermix stability, while both sugars demonstrated quantification performance. The combination of amplification enhancers and sugar-based stabilizers resulted in a LAMP formulation with improved amplification efficiency and stability. The optimized and stabilized LAMP mastermix represents a promising basis for the development of robust LAMP assays for point-of-care diagnostics.

Key words: LAMP; reaction additives; point-of-care; diagnostics

IMMUNOHISTOCHEMICAL APPROACHES IN FISH INTESTINAL AND HEPATIC RESEARCH: A COMPARATIVE ANALYSIS OF METHODOLOGICAL VARIABILITY

Kristína Plevová, Eliška Zusková

University of South Bohemia in České Budějovice, Faculty of Fisheries and Protection of Waters, South Bohemian Research Center of Aquaculture and Biodiversity of Hydrocenoses, Research Institute of Fish Culture and Hydrobiology, Vodňany, Czech Republic

Immunohistochemistry (IHC) is an essential tool in fish biological and pathological research, but unlike in mammalian systems, no standardised protocols exist for fish tissues. The scarcity of fish-specific antibodies forces researchers to adapt mammalian-derived reagents, introducing variability that undermines reproducibility and cross-study comparability. Yet this variability remains largely uncharacterized in the literature.

Since IHC protocols in fish research are rarely the primary focus of published studies but are instead embedded within broader research as part of the study design, a systematic extraction and comparison of these protocols has not previously been attempted.

We expect to find substantial methodological variability across studies, alongside significant gaps in protocol reporting that further limit reproducibility. Using a systematic comparative approach, IHC protocols applied to fish intestinal and hepatic tissues are extracted from published studies, both methodologically focused and those where IHC was employed as a tool, and key parameters, including tissue fixation, antigen retrieval, antibody selection and dilution, blocking strategies, and detection systems, are identified and compared.

This study will present a structured comparative overview of IHC protocols specific to fish intestinal and hepatic tissues and to highlight critical gaps in protocol reporting that currently limit reproducibility and cross-study comparability in fish histopathological research.

Key words: immunohistochemistry; aquatic animals; fish tissues; methodology; protocols

Acknowledgement: The study has been funded by Czech Ministry of Education, Youth and Sports through the project PROFISH (CZ.02.1.01/0.0/0.0/16_019/0000869) and operational program The Johannes Amos Comenius (OP JAK CZ.02.01.01/00/ 23_021/0012616). The authors would like to thank the CENAKVA research infrastructure for providing laboratory facilities and technical support during the study.

TREATMENT STRATEGY OF DIPLOSTOMOSIS IN GRASS CARP

Eliška Zusková, Kristína Plevová, Veronika Piačková

University of South Bohemia in České Budějovice, Faculty of Fisheries and Protection of Waters, South Bohemian Research Center of Aquaculture and Biodiversity of Hydrocenoses, Research Institute of Fish Culture and Hydrobiology, Vodňany, Czech Republic

Diplostomosis caused by the eye fluke *Diplostomum pseudospathaceum* is an important parasitic disease of freshwater fish that can impair vision, growth, and survival, particularly in juvenile grass carp (*Ctenopharyngodon idella*). This study proposes a treatment strategy based on the use of praziquantel (PZQ), a broad-spectrum anthelmintic effective against trematodes. The efficacy of medicated feed containing 30, 50, and 100 $mg \cdot kg^{-1}$ body weight of PZQ, administered for 7 days, was evaluated. Treatment efficacy increased with dosage, resulting in average reductions in viable metacercariae of 47%, 84%, and 88%, respectively, while repeated administration of 50 $mg \cdot kg^{-1}$ achieved complete parasite elimination. Bath treatments with praziquantel concentrations ranging from 2 to 10 $mg \cdot L^{-1}$ also demonstrated therapeutic potential by significantly increasing the proportion of non-motile metacercariae. The treatment protocol is complemented by preventive measures aimed at disrupting the parasite's life cycle by managing intermediate hosts and limiting piscivorous bird access. The strategy combines chemotherapy with preventive measures aimed at disrupting the parasite's life cycle by managing intermediate hosts and reducing bird-mediated transmission. The findings demonstrate that praziquantel is an effective and safe option for controlling diplostomosis in grass carp, providing a practical therapeutic tool for freshwater aquaculture.

Key words: diplostomosis; *Diplostomum pseudospathaceum*; grass carp; praziquantel; aquaculture, fish health

Acknowledgement: The study has been funded by Czech Ministry of Education, Youth and Sports through the project PROFISH (CZ.02.1.01/0.0/0.0/16_019/0000869) and operational program The Johannes Amos Comenius Programme (OP JAK CZ.02.01.01/00/23_021/0012616).

FISH HEALTH MANAGEMENT IN CZECH AND MORAVIAN PONDS FROM THE PERSPECTIVE OF FISH FARMERS

Veronika Piačková

University of South Bohemia in České Budějovice, Faculty of Fisheries and Protection of Waters, South Bohemian Research Center of Aquaculture and Biodiverzity of Hydrocenoses, Vodňany, Czech Republic

In the Czech Republic, the obligations of animal breeders with regard to the health of their animals are set out in the Veterinary Act (No. 166/1999 Coll.). Section 4 of this Act stipulates that a breeder must monitor the health of their animals, provide timely first aid where necessary and seek professional veterinary assistance. They must also prevent the occurrence and spread of infectious diseases and other animal illnesses. The Act also states that the Regional Veterinary Administration approves a private veterinarian for registered aquaculture facilities if such approval is required by a European Parliament and Council regulation. However, these regulations do not specify exactly which facilities are subject to the requirement to have their own approved private veterinarian.

As there are very few private veterinarians working on fish farms in our country, and breeders use the services of diagnostic laboratories at State Veterinary Institutes and research facilities only very rarely, I am interested in how fish farmers deal with fish diseases themselves. To this end, I created a multiple-choice questionnaire and asked fish farmers, via their association, to complete it anonymously. A total of 21 farmers were willing to participate, 19 of whom raise fish in ponds. Analysis of the survey results shows, for example, that the most commonly used remedy for health problems on fish farms is chlorinated lime; that most respondents use State Veterinary Institute services; and that no one calls State Veterinary Administration inspectors in the event of increased fish mortality.

Due to inconsistencies in the responses, the results of the questionnaire cannot provide a fully objective picture of the current situation in fish farming with regard to veterinary care. Nevertheless, it is clear that considerable improvements are needed in this area, from both fish farmers and diagnostic and research institutions. Thanks to the younger generation taking on leadership roles in fish farming enterprises, there is hope that communication and cooperation between fish producers and the veterinary sector could improve in the future. This would be in the interest of maximising the survival rate of farmed fish and maintaining the profitability of the fishing industry as a whole.

Key words: veterinary care; carp ponds; survey

Acknowledgement: This study was financially supported by the Ministry of Education, Youth and Sports of the Czech Republic, the projects PROFISH (CZ.02.1.01/0.0/0.0/ 16_019 / 0000869), and OP JAK – project Aquaculture for Future (CZ.02.01.01/00/23_021/0012616).

UNTARGETED FLOW CYTOMETRY OF TELEOST BLOOD FOR PREDICTIVE PROFILING OF SYSTEMIC IMMUNE ACTIVATION

Thilini Hettiarachchi[1], Jiří Kyslík[1], Tomáš Korytář[1,2]

[1]Czech Academy of Sciences, Biology Centre, Institute of Parasitology, České Budějovice, Czech Republic; [2]University of South Bohemia in České Budějovice, Faculty of Fisheries and Protection of Waters, South Bohemian Research Centre of Aquaculture and Biodiversity of Hydrocenoses, Vodňany, Czech Republic

Assessment of immune status in fish is typically based on pathogen detection, mortality, histopathology, gene expression profiling, or antigen-specific antibody responses. While these approaches remain indispensable, they offer limited insight into the cellular dynamics that drive immune activation, progression, and resolution. Here we present untargeted flow cytometry of teleost whole blood as a rapid and quantitative strategy to monitor systemic immune responses during infection and vaccination. Using minimally processed samples, we develop a simple gating framework based on forward and side scatter, pulse geometry, nucleic acid staining, and separation of erythroid and leukocyte compartments. This approach resolves major circulating cell populations without the need for extensive antibody panels, including erythrocytes, thrombocytes, and distinct leukocyte-like subsets such as lymphocyte-like, monocyte-like, and granulocyte-like cells, as well as cellular aggregates. When combined with volumetric counting or fixed blood-volume acquisition, this workflow enables estimation of both relative population frequencies and absolute cell numbers in circulation. Applying the method to experimental vaccination and viral infection models, we show that whole-blood flow cytometry captures dynamic and individualized patterns of immune cell mobilization. Early responses are characterized by shifts in granulocyte abundance, changes in scatter properties of innate cell compartments, and activation-associated phenotypes. Later phases reveal redistribution within lymphocyte-like populations, modulation of IgM-positive B-cell compartments, and signatures associated with recovery, persistence, or immune dysregulation. Coupled with functional assays such as reactive oxygen species production and proliferation readouts, this framework further allows assessment of effector activation and proliferative capacity during systemic immune responses.

Key words: untargeted flow cytometry; teleost immunology; leukocyte profiling; blood cell dynamics

Acknowledgements: This research was supported by the Czech Science Foundation (grant no. 23-08042K).

ARCHITECTS OF IMMUNITY: THE FCR/FCRL PROFILING IN COMMON CARP AND ZEBRAFISH MODELS

Jiří Kyslík[1], Alexander Rebl[2], Roman Franěk[3], Tomáš Korytář[1,3]

[1]Czech Academy of Sciences, Biology Centre, Institute of Parasitology, České Budějovice, Czech Republic; [2]Research Institute for Farm Animal Biology (FBN), Working Group Fish Genetics, Dummerstorf, Germany; [3]University of South Bohemia in České Budějovice, Faculty of Fisheries and Protection of Waters, South Bohemian Research Centre of Aquaculture and Biodiversity of Hydrocenoses, Vodňany, Czech Republic

The evolution of Fc receptors (FcRs) and Fc receptor-like (FcRL) molecules in teleost fishes is characterized by a dynamic interplay of genomic expansion and functional diversification. While these receptors are central to vertebrate adaptive immunity, their evolutionary trajectory in common carp (*Cyprinus carpio*) and zebrafish (*Danio rerio*) remains a distinct case study in how lineage-specific genetic mechanisms drive immunological adaptation. Here, we present a comparative study of these Fc receptor molecules, revealing that their respective repertoires (13 in *D. rerio* and 19 in *C. carpio*) have been shaped by complex events, including whole-genome duplication, tandem duplication, and transposition. Comparative immune cell type and tissue transcriptomic and ontogeny profiling demonstrates that these genetic expansions have facilitated both the maintenance of a conserved immune core and the emergence of specialized, lineage-specific adaptations. By moving beyond simple genomic description, this work provides a high-resolution framework for understanding how genomic redundancy serves as a primary driver for the diversification of immune detection mechanisms, highlighting the divergent strategies employed by the carp and zebrafish lineages to optimize adaptive immune system.

Key words: teleosts; Fc Receptors (FcR); adaptive immunity; expression

Acknowledgements: This research was supported by the Czech Science Foundation (grant no. JSPS-25-17).

SIMULTANEOUSLY MEASURING IMMUNE RESPONSES AND THE PRESENCE OF MULTIPLE PATHOGENS SUPPORTS THE DIAGNOSIS OF GILL DISEASES IN COMMON CARP AND RAINBOW TROUT

Mikolaj Adamek[1], Tamara Ramirez Stoll[1], Dipa Debnath[1], Torben Krebs[1], Julia Bauer[2], Peter Steinbauer[3], Ivona Mladineo[4], Ľubomír Pojezdal[5], Elisabeth Nardy[6], Krzysztof Rakus[7], Alexander Rebl[8], Alberto Falco[9], Marek Matras[10], Michal Reichert[10], Magdalena Stachnik[10], Tomáš Korytář[11], Verena Jung-Schroers[1]

[1]University of Veterinary Medicine Hannover, Institute for Parasitology, Fish Disease Research Unit, Hannover, Germany; [2]Veterinary Task Force: Control of Fish Epizootics, Lower Saxony State Office for Consumer Protection and Food Safety, Hannover, Germany; [3]Tiergesundheitsdienst Bayern e.V., Poing, Bayern, Germany; [4]Czech Academy of Sciences, Biology Centre, Institute of Parasitology, Laboratory of Functional Helminthology, České Budějovice, Czech Republic; [5]Veterinary Research Institute, Infectious Diseases and Preventive Medicine, Brno, Czech Republic; [6]Chemical and Veterinary Investigations Office (CVUA) Stuttgart, Fellbach, Germany; [7]Jagiellonian University, Faculty of Biology, Institute of Zoology and Biomedical Research, Department of Evolutionary Immunology, Krakow, Poland; [8]Institute for Farm Animal Biology (FBN), Fish Genetics Unit, Dummerstorf, Germany; [9]Spanish National Research Council (IATS-CSIC), Institute of Aquaculture Torre de La Sal, Fish Pathology Group, Cabanes, Castellón, Spain; [10]National Veterinary Research Institute, Laboratory of Fish Diseases, Pulawy, Poland; [11]Czech Academy of Sciences, Biology Centre, Institute of Parasitology, Laboratory of Fish Immunology, České Budějovice, Czech Republic

Monitoring immune responses in non-lethal epithelial tissue samples, combined with multi-pathogen detection, could be a powerful approach for diagnosing mucosal diseases in fish. We applied and evaluated this strategy in several case studies, including multifactorial gill diseases in common carp (*Cyprinus carpio*) and rainbow trout (*Oncorhynchus mykiss*). The aim was to evaluate the diagnostic value of epithelial immune profiling in conjunction with the molecular detection of pathogens in the fish and their environment.

The samples included gill biopsies, gill swabs, and water collected during natural outbreaks in aquaculture in Germany, Poland, and the Czech Republic. Pathogens and host immune responses were detected/profiled using RT-qPCR or Fluidigm/Standard BioTools microfluidic arrays.

Epithelial immune profiling revealed pathogen-class-specific signatures, with antiviral responses observed in viral gill diseases and proinflammatory profiles in bacterial infections. Gill swabs were found to be as effective as biopsies in detecting these signatures. Interestingly, rainbow trout samples carried large numbers of Ca. *Piscichlamydia salmonis* and Ca. *Branchiomonas cysticola*, indicating that proliferative/complex gill inflammation/disease caused by these bacteria is highly prevalent. Together with the near-omnipresence of *Flavobacterium psychrophilum*, this suggests that bacterial diseases are becoming a significant issue in rainbow trout aquaculture in Central Europe. Furthermore, immune response profiling showed that there might be an unknown virus present in rainbow trout aquaculture in Germany, which requires further investigation.

Measuring immune responses in epithelial tissues alongside multi-pathogen detection allows for the early, non-lethal diagnosis of gill diseases in a variety of fish species. This integrated approach can be used for differential diagnosis (e.g., distinguishing between viral and bacterial gill diseases), surveillance, and outbreak management, particularly in settings where post-mortem sampling is impractical.

Key words: common carp; rainbow trout; gill disease

Acknowledgements: The research was supported by European Union's Horizon Europe Research and Innovation Programme under grant agreement 101084204 Cure4Aqua.

THYMIC AND MUCOSAL DISTRIBUTION OF DISTINCT CD4-POSITIVE T CELL POPULATIONS IN RAINBOW TROUT

Fumio Takizawa[1], Ryuto Iguchi[1], Ryosuke Fujihara[2], Shota Watanabe[2], Yasuhiro Shibasaki[2], Tomáš Korytář[3], Ryuichiro Miyazawa[4], J. Oriol Sunyer[4], Hiroaki Suetake[1]

[1]Fukui Prefectural University, Faculty of Marine Science and Technology, Obama, Fukui, Japan; [2]Nihon University, College of Bioresource Sciences, Fujisawa, Kanagawa, Japan; [3]Czech Academy of Sciences, Biology Centre, Institute of Parasitology, České Budějovice, Czech Republic; [4]University of Pennsylvania, School of Veterinary Medicine, Philadelphia, USA

CD4-positive T cells play central roles in adaptive immunity, including the regulation of B-cell activation, antibody production and protective responses against infectious diseases. In teleost fish, two CD4 homologues, CD4-1 and CD4-2, have been identified; however, the diversity and functional specialization of CD4-positive T-cell populations remain poorly understood. In rainbow trout, CD4-1/CD4-2 double-positive (DP) cells and CD4-2 single-positive (SP) cells can be distinguished using specific monoclonal antibodies. Whether these populations represent different maturation stages or distinct T-cell subsets with different immunological functions is an important question for understanding T-cell-mediated immunity in fish.

In this study, leukocytes from the thymus, head kidney, spleen, gills and intestine of rainbow trout were analyzed by flow cytometry using anti-CD4-1 and anti-CD4-2 antibodies. DP and SP cells were detected in immune-related tissues and were particularly abundant in the thymus and gills. The presence of both populations in the thymus suggests that DP and SP cells are already distinguishable during thymic T-cell development. To further characterize these subsets, gill DP and SP cells were sorted and subjected to RNA-seq analysis. DP cells showed higher cd4-1 expression, consistent with their phenotypic identity, whereas SP cells displayed an activation- and effector-like transcriptomic profile. These findings indicate that rainbow trout possess distinct CD4-positive Th cell subsets with different tissue distribution and gene expression features. Their enrichment in the gills, a major mucosal interface exposed to aquatic pathogens, suggests that these subsets contribute to local immune surveillance and protective responses. These findings provide a basis for evaluating T-cell-mediated immunity during infection and vaccination in aquaculture.

Key words: rainbow trout; CD4+ helper T cells; mucosal immunity; gill; thymus

Acknowledgements: This work was supported by the JSPS Bilateral Program with the Czech Academy of Sciences (CAS), grant number JPJSBP120252503.

DETECTION OF ANTIGEN-SPECIFIC T CELL RESPONSES AND ANTIGEN PRESENTATION IN GINBUNA CRUCIAN CARP: AN IN VITRO ASSAY BASED ON CYTOKINE GENE EXPRESSION

[1]Shinji Takeda, [1]Daichi Oku, [2]Fumio Takizawa, [3]Tomáš Korytář, [1]Tadaaki Moritomo, [1]Fumihiko Katakura

[1]Nihon University, Graduate School of Veterinary Medicine, Laboratory of Comparative Immunology, Kanagawa, Japan; [2]Fukui Prefectural University, Faculty of Marine Science and Technology, Obama, Fukui, Japan; [3]Czech Academy of Sciences, Biology Centre, Institute of Parasitology, České Budějovice, Czech Republic

We have previously reported that antigen-sensitized kidney leukocytes from clonal ginbuna crucian carp (*Carassius auratus langsdorfii*) express T cell-associated cytokine genes (e.g., *ifng1*, *il4/13b*) following *in vitro* re-stimulation with the same antigen. In the present study, we investigated the cellular mechanism underlying this antigen-specific cytokine expression using keyhole limpet hemocyanin (KLH) as a model antigen. Kidney leukocytes isolated 4–6 weeks post-immunization were cultured with KLH for 24 h. To identify the specific cytokine-expressing cell types, 4×10^4 cells were sorted from each population, including CD4-1$^+$ T (Th), CD8α$^+$ T (Tc), IgM$^+$ B cells, macrophages (MΦ), and granulocytes, and subjected to RT-qPCR analysis. Both *ifng1* and *il4/13b* were predominantly expressed in Th and Tc cells, whereas minimal expression was observed in the other cell types. These findings suggest that T cells undergo *in vitro* activation mediated by antigen-presenting cells (APCs). To identify the responsible APCs, we examined APC-related gene expression in isolated B cells, MΦ, and granulocytes. *mhc I* was constitutively expressed in all three cell types, whereas *mhc II*, *cd83*, and *cd80/86* were detected mainly in B cells and MΦ, and only weakly in granulocytes. Next, to determine which cells were essential, KLH-sensitized leukocytes depleted of either B cells or MΦ were prepared and re-stimulated with KLH. The upregulation of *ifng1* and *il4/13b* was maintained after B cell depletion but abolished after MΦ depletion. To confirm the direct role of MΦ, 5×10^4 sorted Th or Tc cells were co-cultured with 2×10^5 MΦ in the presence of KLH for 24 h, which resulted in a significant upregulation of both cytokine genes. These results demonstrate that MΦ function as the primary APCs in this system; they not only present the exogenous antigen KLH to Th cells via MHC II, but potentially also present it to Tc cells via MHC I through cross-presentation.

Key words: ginbuna crucian carp; antigen-specific T cell response; antigen-presenting cell; macrophage; cytokine gene expression

Acknowledgements: This work was supported by the JSPS Bilateral Program with the Czech Academy of Sciences (CAS), grant number JPJSBP120252503.

COMPARATIVE ASSESSMENT OF WATER MOULD (OOMYCOTA, SAPROLEGNIALES) DIVERSITY USING CULTURE-BASED ISOLATION AND eDNA METABARCODING

Réka Vámosi[1], Erdei Noémi[1], Kóczán Patrícia[1], Ferincz Árpád[2], Martina Lisnerová[3], Ivan Fiala[3], Eszterbauer Edit[1]

[1]HUN-REN Veterinary Medical Research Institute, Budapest, Hungary; [2]National Federation of Hungarian Anglers, Budapest, Hungary; [3]Czech Academy of Sciences, Biology Centre, Institute of Parasitology, České Budějovice, Czech Republic

Water moulds are fungal-like oomycetes that are widespread in freshwater ecosystems and represent significant threats to both natural aquatic environments and aquaculture. Species belonging to the genera *Saprolegnia*, *Leptolegnia*, and *Achlya* are commonly associated with saprolegniosis in fish and fish eggs, while *Aphanomyces* species cause severe diseases such as epizootic ulcerative syndrome (EUS) in fish, and crayfish plague in freshwater crayfish. These pathogens can lead to substantial ecological damage and economic losses, highlighting the need for effective monitoring and management strategies.

This study aimed to develop and optimize an environmental DNA (eDNA) metabarcoding approach for detecting pathogenic water moulds in freshwater environments and compare its performance with conventional culture-based isolation methods. Water samples were collected from two sites representing different epidemiological conditions: (A) without visible disease outbreaks and (B) during the spawning season of pike perch, when water mould infections were present on fish eggs. For culture-based analysis, water moulds were isolated from water using hemp-seed baiting and from infected fish eggs, followed by DNA extraction of pure cultures. For eDNA metabarcoding, water samples were filtered and DNA was extracted directly from environmental samples. Species identification was based on amplification of the ITS region of ribosomal RNA gene cluster. Cultured isolates were identified by Sanger sequencing, while eDNA amplicons (ITS1 and ITS2) were sequenced on Illumina platform. Analyses included bioinformatic processing, taxonomic assignment, and phylogenetic reconstruction.

At site A, culture-based isolation recovered only a few species, including *Saprolegnia australis* and *S. ferax*. In contrast, eDNA metabarcoding detected a broader range of taxa, including important pathogens such as *S. parasitica*, *Aphanomyces astaci*, and other oomycetes. At site B, culture-based isolation was successful from all examined locations, particularly from fish eggs, and predominantly recovered *S. parasitica*. Meanwhile, eDNA metabarcoding detected several pathogenic species simultaneously, including *S. parasitica*, *S. australis*, *S. ferax*, and *S. delica*, revealing greater community diversity.

Overall, the traditional culture-based isolation was effective for detecting viable pathogens and active infections, but was limited by cultivation bias and species selectivity. On the other hand, eDNA metabarcoding provided a more comprehensive assessment of water mould diversity and detected taxa that could not be recovered through cultivation. Although it offers a more accurate estimate of oomycete diversity, its inability to distinguish viable and infective organisms limit its use for confirming active infections or predicting disease outbreaks. Therefore, combining eDNA metabarcoding with traditional isolation methods may provide the most effective approach for monitoring pathogenic water moulds in aquatic ecosystems.

Key words: water mould; saprolegniosis; *Saprolegnia parasitica*; environmental DNA; metabarcoding

Acknowledgement: This research was supported by the National Research, Development and Innovation Office under grant number K141889.

EFFECT OF POLLUTANTS WITH THYROID RECEPTOR ANTAGONISTIC ACTIVITY ON FISH AND AMPHIBIANS

Kateřina Karbusová[1], Michal Pech[1], Veronika Piačková[1], Kateřina Grabicová[1], Tomáš Korytář[1], Pavel Šauer[1], Ilona Prokopová[1], Hana Kocour Kroupová[1]

[1]University of South Bohemia in České Budějovice, Faculty of Fisheries and Protection of Waters, South Bohemian Research Center of Aquaculture and Biodiversity of Hydrocenoses, Czech Republic

Endocrine disruptors are widely recognized as significant environmental contaminants capable of interfering with hormonal regulation and, increasingly, with immune function. Among these, compounds affecting the hypothalamus-pituitary-thyroid axis are of particular concern due to the central role of thyroid hormones (THs) in development, metabolism, and immune regulation. The immune and endocrine systems are closely interconnected through complex signalling pathways, and disruption of this crosstalk may lead to altered immune competence. This interaction is especially critical during early development, when the immune system is highly sensitive to external influences and long-term effects can arise from even low-level exposures. Thyroid hormones, primarily triiodothyronine (T3) and thyroxine (T4), regulate key components of both innate and adaptive immunity, including lymphocyte proliferation, differentiation, apoptosis, and cytokine production. Consequently, chemicals that disrupt thyroid signalling may indirectly modulate immune responses. One important mechanism involves antagonism of thyroid hormone receptors (TRs), particularly TRβ. Competitive antagonists bind to these receptors without activating them, thereby blocking TH-mediated gene expression and disrupting normal physiological regulation. Such interference may impair immune cell function and reduce the organism's ability to mount effective defence responses. A variety of environmentally relevant contaminants, including pharmaceuticals, pesticides, industrial chemicals such as bisphenols and phthalates, and per- and polyfluoroalkyl substances, exhibit TR antagonistic activity. These compounds have been detected in multiple environmental matrices, particularly in aquatic ecosystems. Our screening study indicates that TR antagonistic activity is frequently present in surface waters, suggesting continuous, low-level exposure of aquatic organisms.

Fish and amphibians are especially vulnerable due to their permeable body surfaces and external development, which lacks maternal protection. Early-life exposure is of particular concern, as developmental immunotoxicity may lead to persistent alterations in immune function. These changes can manifest later as increased susceptibility to infections, impaired immune responses, or dysregulated inflammation. Given the evolutionary conservation of immune mechanisms across vertebrates, findings from aquatic organisms may also provide insights relevant to other species, including humans. Despite growing evidence of thyroid disruption in the environment, the immunological consequences of TR antagonism remain insufficiently understood. Current knowledge suggests that interference with TH signalling may suppress adaptive immunity and alter innate responses in a context-dependent manner. This highlights the need for deeper investigation into how environmental contaminants influence immune competence and disease susceptibility. Such understanding is essential for improving ecological risk assessment and addressing the broader health implications of endocrine disruption.

Key words: aquatic environment; early-life stages; endocrine disruptors; immune system

Acknowledgement: This work was supported by the Czech Science Foundation (Project No. 26-20993S).

CIRCULATING IGM FORM REVEAL TEMPERATURE AND VACCINATION DEPENDENT REMOELING OF THE TELEOST ANTIBODY RESPONSE

Bharath Raja[1], Jiří Kyslík[1], Tomáš Korytář[1,2]

[1]Czech Academy of Sciences, Biology Centre, Institute of Parasitology, České Budějovice, Czech Republic; [2]University of South Bohemia in České Budějovice, Faculty of Fisheries and Protection of Waters, South Bohemian Research Centre of Aquaculture and Biodiversity of Hydrocenoses, Vodňany, Czech Republic

Immunoglobulin M (IgM) is found in several states of covalent polymeric state. In teleost fish, the predominant circulating form is typically tetrameric compared to mammalian IgM which is most commonly pentameric. The total IgM concentration measured using ELISA does not reveal structural state in which the IgM circulates and therefore provides limited information on the antibody maturation, stability and functional quality. To investigate this overlooked aspect of the antibody response, we examined circulating IgM patterns in two independent experiments: fish infected with parasite *Sphaerospora molnari* and maintained at different temperatures, and orally vaccinated fish with plant-produced G-protein vaccine against spring viraemia of carp virus (SVCV). Immunoblot analysis revealed a dominant polymeric IgM form together with several distinct circulating IgM forms whose relative abundance varied among experimental conditions. In the *S. molnari* infection experiment, lower temperatures were associated with a predominance of polymeric IgM and fewer detectable IgM forms, whereas warmer conditions showed a broader IgM distribution, potentially reflecting temperature-dependent effects on IgM folding. Oral vaccination followed by SVCV challenge also altered the circulating IgM profile, indicating that vaccine priming may influence not only total IgM production but also IgM assembly, polymer stability Together, these findings indicate that both temperature and vaccination influence the overall glycosylation, disulfide-bond formation, polymer assembly, ER–Golgi trafficking and circulating stability. Also, total IgM measurements alone may overlook important changes in antibody structural organization. Assessing circulating IgM forms and polymerization status alongside ELISA may therefore provide a more complete evaluation of humoral immune quality during infection, vaccination, and temperature stress in teleost fish.

Key words: teleost IgM; polymeric immunoglobulin; antibody assembly; vaccination; glycosylation

Acknowledgements: This research was supported by the Czech Science Foundation (grant no. 24-13026S).

EDWARDSIELLA TARDA INFECTION IN AFRICAN CATFISH (*CLARIAS GARIEPINUS*) UNDER EXPERIMENTAL CONDITIONS

Ivona Toulová[1,2,3], Věra Vaibarová[4], Ivana Papežíková[1,2,3], Hana Novotná[1,2,3], Ivana Mikulíková[1,2,3], Zuzana Lepková[1], Miroslava Palíková[1,2,3]

[1]University of Veterinary Sciences Brno, Faculty of Veterinary Hygiene and Ecology, Department of Ecology and Diseases of Zoo Animals, Game, Fish and Bees, Brno, Czech Republic; [2]Mendel University in Brno, Faculty of AgriSciences, Department of Zoology, Fisheries and Hydrobiology, Brno, Czech Republic; [3]Veterinary Research Institute, Department of Infectious Diseases and Preventive Medicine, Brno, Czech Republic; [4]University of Veterinary Sciences Brno, Faculty of Veterinary Medicine, Department of Infectious Diseases and Microbiology, Brno, Czech Republic

Edwardsiella tarda is an important bacterial pathogen responsible for edwardsiellosis, a disease causing substantial economic losses in aquaculture, including the African catfish (*Clarias gariepinus*). The establishment of a reliable experimental infection model represents an important step towards a better understanding of *E. tarda* infection in African catfish. Therefore, a series of preliminary experiments was performed to compare different routes of infection and optimize the conditions for immersion challenge. Based on these findings, a reproducible immersion infection model was established using different bacterial concentrations and exposure times. Fish were monitored for clinical signs and mortality, and samples were collected for bacteriological, haematological, immunological, and pathological examinations. The optimised challenge protocol consistently induced experimental infection, with the course of the disease depending on both the bacterial concentration and the duration of exposure.

The established infection model was subsequently applied to evaluate the response of African catfish fed diets supplemented with functional feed additives. Fish were fed control or supplemented diets containing combinations of fatty acid monoacylglycerides, phytogenic compounds, yeast-derived components, and other functional ingredients. Following experimental *E. tarda* challenge, fish were evaluated for bacterial persistence, haematological, plasma biochemical and immunological responses, including phagocytic activity and specific antibody responses. Experimental edwardsiellosis under controlled conditions provides valuable insights for further studies evaluating different dietary strategies, immunostimulants, and other approaches aimed at understanding and managing bacterial infections in African catfish.

Key words: edwardsiellosis; challenge model; immunostimulants

Acknowledgements: This study was supported by Technology Agency of the Czech Republic, Project No. TN02000017 (NaCeBiVet) and by University of Veterinary Sciences Brno, Czech Republic, Project No. FVHE/Pikula/2026ITA24.

FEED ADDITIVES ENHANCING FISH PERFORMANCE AND GUT HEALTH

Lucie Všetičková[1], Eva Poštulková[1], Miroslava Palíková[1,2,3], Ivana Papežíková[1,2,3], Ivana Mikulíková[1,2,3], Zuzana Lepková[2], Ivona Toulová[1,2,3], Jan Mareš[1]

[1]Mendel University in Brno, Faculty of AgriSciences, The Department of Zoology, Fisheries and Hydrobiology, Brno, Czech Republic; [2]University of Veterinary Sciences Brno, Faculty of Veterinary Hygiene and Ecology, Department of Ecology and Diseases of Zoo Animals, Game, Fish and Bees, Brno, Czech Republic; [3]Veterinary Research Institute, Department of Infectious Diseases and Preventive Medicine, Brno, Czech Republic

In animal production, increasing attention is being paid not only to feed efficiency in terms of growth performance and productivity, but also to its effects on animal health and overall welfare. This approach is particularly important in aquaculture, where fish health directly influences production efficiency as well as the economic and environmental sustainability of farming systems. Therefore, the optimization of feed formulations through the targeted use of feed additives represents an effective strategy for enhancing immunity, supporting a healthy gut microbiome, reducing stress, and preventing diseases, thereby contributing to a reduction in the use of antibiotics and other chemotherapeutic substances. Feed additives commonly used in aquaculture include natural immunostimulants, probiotics, prebiotics, micronutrients such as antioxidants, organic acids, and phytobiotics (plant-derived extracts). Their main benefits include stabilizing fish health, enhancing feed utilization efficiency, and reducing production-related risks. In modern aquaculture, feed additives represent an important preventive tool that helps to maintain high production levels while ensuring good fish welfare. In addition, they increase the resistance of fish to adverse environmental conditions, stress, and the occurrence and effects of various pathogens.

Key words: aquaculture; nutrition; probiotics; antibiotic alternatives

Acknowledgements: This study was supported through Technology Agency of the Czech Republic as part of the National Center of Competence Program no. TN02000017 (NaCeBiVet).

DOES GENETIC RESISTANCE OF FISH GUARANTEE PROTECTION AGAINST INFECTIOUS DISEASES?

Ivana Papežíková[1,2,4], Ivana Mikulíková[1,2,4], Věra Vaibarová[3], Ľubomír Pojezdal[4], Kateřina Matějíčková[4], Ivona Toulová[1,2,4], Miroslava Palíková[1,2,4]

[1]University of Veterinary Sciences Brno, Faculty of Veterinary Hygiene and Ecology, Department of Ecology and Diseases of Zoo Animals, Game, Fish and Bees, Brno, Czech Republic; [2]Mendel University in Brno, Faculty of AgriSciences, Department of Zoology, Fisheries and Hydrobiology, Brno, Czech Republic; [3]University of Veterinary Sciences Brno, Faculty of Veterinary Medicine, Department of Infectious Diseases and Microbiology, Brno, Czech Republic; [4]Veterinary Research Institute, Department of Infectious Diseases and Preventive Medicine, Brno, Czech Republic

This work summarizes the results of three experiments evaluating the resistance of different rainbow trout (*Oncorhynchus mykiss*) genetic lines to infections caused by the bacterium *Flavobacterium psychrophilum* and the parasites *Ichthyophthirius multifiliis* and *Tetracapsuloides bryosalmonae*. Two of the fish lines tested carried quantitative trait loci (QTLs) associated with resistance to selected pathogens (*F. psychrophilum*, and *I. multifiliis* + *F. psychrophilum,* respectively). Mortality, selected immune parameters (leukocyte counts and phagocytic activity), and pathogen loads were assessed. Surprisingly, during the *F. psychrophilum* infection, the highest mortality was recorded in the line with declared genetic resistance, suggesting the influence of other factors, such as varying strain virulence or genotype-environment interactions. In the case of *I. multifiliis* infection, the genetically resistant line initially exhibited a higher parasite load but subsequently demonstrated more effective parasite elimination. For *T. bryosalmonae* infection, no significant differences were found between the experimental groups. These findings demonstrate that disease resistance in fish is a complex trait dependent on the interaction between genotype, pathogen, and environment. Therefore, practical application of resistant lines should always be preceded by testing under the specific conditions of the given aquaculture facility.

Key words: genetic resistance; rainbow trout fry syndrome; *Ichthyophthirius multifiliis*; white spot disease; proliferative kidney disease

Acknowledgments: This work was supported by the Ministry of Agriculture of the Czech Republic through research projects No. QK21010030, “Globalization, modern technology and climate change as sources of new opportunities and threats for salmonid fish breeding management.

THE USE OF MEDICINAL PRODUCTS IN FISH VETERINARY PRACTICE

Leona Nepejchalová

Institute for State Control of Veterinary Biologicals and Medicines (ISCVBM), Brno, Czech Republic

Veterinary practice in the aquaculture sector has been and is facing long-term challenges due to the unavailability of authorised veterinary medicinal products. Therefore, veterinarians rely on approaches permitted by current legislation, namely Regulation (EU) 2019/6 on veterinary medicinal products and its implementing regulations. In cases where authorised veterinary medicinal products are unavailable for the relevant aquatic target species and indications, this Regulation provides the rules for the prescription of a medicinal product under the so-called "cascade", which corresponds to the provisions of Article 112 for non-food-producing aquatic species and Article 114 for food-producing aquatic species. This is a tool for the exceptional use of a medicinal product under the direct personal responsibility of a veterinarian, which prevents unacceptable suffering of the animal that would result from a lack of treatment. After treatment in accordance with the rules of Art. 114, the veterinarian must establish an appropriate withdrawal period in accordance with the provisions of Art. 115 and inform the breeder or owner of the animals, who is responsible for ensuring that the withdrawal period is respected.

The prescription of medicinal products under Article 114 will soon be further impacted by forthcoming list of substances which, if contained in EU-authorised veterinary medicinal products for a food- producing terrestrial species or EU-authorised human medicinal products, may be used in exceptional cases for treatment under "the cascade", as they have been assessed in terms of environmental risk, the risk of antimicrobial resistance and the risk related to the unavailability of appropriate treatment or the inability to implement preventive measures. The list must be implemented at the latest within five years from 28 January 2022, meaning it must enter into force no later than 28 January 2027.

Depending on this upcoming list of substances, Implementing Regulation (EU) 2024/1973 (establishing a list of antimicrobials that shall not be used in "the cascade" or that shall only be used in "the cascade" under certain conditions) will be amended to include rules on the antimicrobial treatment of food-producing aquatic species within its scope and to establish conditions for the therapeutic administration of certain antimicrobial substances used in these animals in accordance with Art. 114 of Regulation (EU) 2019/6. It is important to note that, in addition, a member state may further restrict or prohibit the use of certain antimicrobial substances in animals on its territory if the treatment of animals with such substances is inconsistent with the implementation of its national policy on prudent use of antimicrobials.

Key words: legislation; cascade; veterinary medicinal product; antimicrobials

ARE FISH FARMS PRODUCING STOCK SALMONID FISH A SOURCE OF PATHOGENS TO OPEN WATERS?

Miroslava Palíková[1,2,3], Ivana Mikulíková[1,2,3], Ivana Papežíková[1,2,3], Ľubomír Pojezdal[3], Ivona Toulová[1,2,3], Tomáš Doležal[2]

[1]University of Veterinary Sciences Brno, Faculty of Veterinary Hygiene and Ecology, Department of Ecology and Diseases of Zoo Animals, Game, Fish and Bees, Brno, Czech Republic; [2]Mendel University in Brno, Faculty of AgriSciences, Department of Zoology, Fisheries and Hydrobiology, Brno, Czech Republic; [3] Veterinary Research Institute, Department of Infectious Diseases and Preventive Medicine,Brno, Czech Republic

Stocking of rivers with hatchery-reared salmonids is an important management tool, but it may also represent a pathway for the introduction of pathogens into natural aquatic ecosystems. This study evaluated the occurrence of selected viral and parasitic pathogens of salmonid fish originating from fish farms supplying stocking material to rivers in the Czech Republic.

A total of 132 fish from 13 fish farms were examined between March and May 2025. Fish were screened for proliferative kidney disease (PKD; *Tetracapsuloides bryosalmonae* DNA) using real-time PCR, for viral haemorrhagic septicaemia virus (VHSV), infectious hematopoietic necrosis virus (IHNV), infectious pancreatic necrosis virus (IPNV) and piscine orthoreovirus (PRV-3) RNA using established conventional and real-time PCR assays, and for a range of ecto- and endoparasites using parasitological examination.

PKD-positive fish were detected in 6 of 12 evaluated farms, with prevalence ranging from 10 to 100 %. PRV-3 was detected in three farms, while all other tested farms were negative. Parasitological examinations revealed frequent occurrence of common salmonid parasites, particularly *Gyrodactylus* spp., which were found in most facilities and reached 100% prevalence in several farms. High parasite intensities of Gyrodactylus derjavinoides were recorded, with maximum counts exceeding 250 individuals per fish sample. Other commonly detected taxa included *Apiosoma* sp., *Trichodina* sp., *Chilodonella* sp., *Diplostomum* sp., *Tylodelphys* sp., and *Proteocephalus* sp. One facility was completely negative for all examined parasites.

The results demonstrate that hatcheries and fish farms producing stocking salmonids can harbor both viral and parasitic pathogens capable of entering open waters through stocking activities. Although most detected parasites are common and widespread in freshwater ecosystems, the occurrence of PKD and PRV-3 in a proportion of facilities highlights a potential epidemiological risk. Regular health monitoring and pathogen screening of stocking material should therefore be considered an essential component of fisheries management to minimize disease transmission between aquaculture facilities and wild fish populations.

Key words: PKD; PRV-3, *Gyrodactylus*; aquaculture; river ecosystems; fish health

Acknowledgments: This work was supported by the project FVHE/Pikula/2026ITA24.

INVESTIGATION OF S1P-DEPENDENT LYMPHOCYTE MIGRATION MECHANISM IN TELEOST FISH USING FTY720

Ryosuke Fujihara[1], Haruma Hirose[1], Kaede Matsuda[1], Fumio Takizawa[2], Tomáš Korytář[3], Nobuhiro Mano[1], Yasuhiro Shibasaki[1]

[1]Nihon University, College of Bioresource Sciences, Fujisawa, Kanagawa, Japan; [2]Fukui Prefectural University, Faculty of Marine Science and Technology, Obama, Fukui, Japan; [3]Czech Academy of Sciences, Biology Centre, Institute of Parasitology, České Budějovice, Czech Republic.

Upon infection and vaccination, lymphocyte migration among tissues and the peripheral circulation is essential for antigen encounter and the establishment of systemic immune responses. In mammals, sphingosine-1-phosphate (S1P), a lipid mediator, regulates lymphocyte egress from lymphoid tissues into the blood. Fingolimod (FTY720), an immunosuppressant, is known to inhibit this S1P-dependent lymphocyte migration. In teleost fish, the spleen has been shown to play a crucial role in antigen-specific immune responses; however, the mechanisms by which lymphocytes egress from immune-related tissues, including the spleen, remain unclear. Whether S1P regulates lymphocyte migration in teleost fish, and whether FTY720 is similarly effective in fish, are also unknown. Furthermore, because teleost fish lack lymph nodes, it is unknown in which tissues lymphocytes are retained when S1P-dependent lymphocyte egress is inhibited.

In this study, we investigated the effects of FTY720 on lymphocyte migration in rainbow trout to determine whether S1P-dependent lymphocyte migration mechanisms are present in teleost fish. After administration of FTY720, the dynamics of B lymphocytes and $CD4^+$ T lymphocytes in peripheral blood were analyzed by flow cytometry. In addition, various tissues were collected to identify where lymphocytes were retained after FTY720 treatment. FTY720 administration reduced the number of B lymphocytes and $CD4^+$ T lymphocytes in peripheral blood, while lymphocytes were retained in the trunk kidney, suggesting that FTY720 modulates lymphocyte dynamics in teleost fish. These findings provide new insight into S1P-dependent lymphocyte migration mechanisms in teleost fish and suggest that FTY720 may serve as a useful tool for studying immune responses.

Key words: rainbow trout; FTY720; S1P; lymphocyte migration

Acknowledgements: This work was supported by the JSPS Bilateral Program with the Czech Academy of Sciences (CAS) (grant number JPJSBP120252503) and by the Japan Science and Technology Agency (JST) FOREST Program (grant number JPMJFR233M).

FUGU SECRETORY IGD AND ITS RECEPTOR ON THE BASOPHILS

Chenkai Shao[1], Fumio Takizawa[1], Tomáš Korytář[2], Hiroaki Suetake[1]

[1]Fukui Prefectural University, Faculty of Marine Science and Technology, Obama, Fukui, Japan; [2]Czech Academy of Sciences, Biology Centre, Institute of Parasitology, České Budějovice, Czech Republic.

Basophils are leukocytes primarily involved in allergic and anti-parasitic responses in mammals. Mammalian basophils display IgE and IgD on their cell surface. In teleost fish, we found that basophils can harbor IgM, and other gsecretory IgD has been reported in rainbow trout. However, its characterization and receptors remain unclear in fugu (*Takifugu rubripes*), a key model for fish basophil research.

This study investigated gene identification and expression analysis of secretory IgD and its potential receptors (galectin-9 and CD44) in fugu. Using 3' RACE, we found that fugu IgD lacks membrane- or secretion-specific exons; instead, soluble and membrane-bound forms are generated via alternative splicing, similar to rainbow trout. RT-qPCR showed that secretory IgD was predominantly expressed in secondary lymphoid tissues, leukocytes, and gills. Then, we identified galectin-9 and CD44 in fugu. While *galectin-9* expression was detected in all tissues examined, *CD44* expression was significantly higher in basophils than in peripheral blood leukocytes, suggesting its crucial role in basophils.

In conclusion, fugu can express secretory IgD transcripts, and its candidate receptor is highly expressed in basophils. These findings suggest that fish basophils may mediate specific immune responses through secretory IgD binding, highlighting an evolutionarily conserved mechanism.

Key words: Secretory IgD; Basophils; CD44; Galectin-9

Acknowledgements: Acknowledgements: This work was supported by the JSPS Bilateral Joint Research Projects between Japan and the Czech Academy of Sciences (CAS), grant number JPJSBP120252503, and Grants-in-Aid for Scientific Research, 25K02100.

ANTIMICROBIAL ACTIVITY OF ESSENTIAL OILS AGAINST FISH-PATHOGENIC BACTERIA

Monika Haiserová, Věra Vaibarová, Alois Čížek

University of Veterinary Sciences Brno, Faculty of Veterinary Medicine, Department of Infectious Diseases and Microbiology, Brno, Czech Republic

Bacterial infections represent a primary cause of economic losses in aquaculture. While control currently relies on antibiotics, their use leads to antimicrobial resistance and concerns regarding residues in fish tissue and the environment. This necessitates the search for alternative treatments, such as essential oils (EOs). In this study, the antibacterial activity of five EOs (eucalyptus, cinnamon, sage, basil, and oregano) was evaluated *in vitro* at four concentrations (1%, 5%, 25%, and 100%) using the disk diffusion method. Testing was conducted against six fish pathogens: *Aeromonas salmonicida, A. hydrophila, Edwardsiella tarda, Flavobacterium psychrophilum, F. columnare*, and *Yersinia ruckeri*. Significant differences in antimicrobial activity were observed. Cinnamon EO exhibited the highest efficacy, producing the largest inhibition zones across all pathogens and concentrations, followed by oregano EO. Conversely, eucalyptus EO showed the lowest efficacy, particularly at lower concentrations. *F. psychrophilum* and *F. columnare* were the most susceptible species, with inhibition zones exceeding 50 mm at 100% EO concentration. *Yersinia ruckeri* demonstrated the highest resistance. Cinnamon and oregano EO showed significant *in vitro* antibacterial activity against all tested fish pathogens. These findings suggest they could serve as effective alternatives to antibiotics. However, further *in vivo* testing is necessary to confirm their safety and practical applicability in fish farming.

Key words: alternative treatment; bacterial infections; phytochemicals; antibacterial effect

Acknowledgements: This research was a part of Monika Haiserová's Bachelor thesis.

ATTENUATION OF IGM ANTIBODY RESPONSE BY SPLENECTOMY IN RAINBOW TROUT

Shota Watanabe[1], Shunpei Hashiba[1], Ryosuke Fujihara[1], Fumio Takizawa[2], Tomáš Korytář[3], Nobuhiro Mano[1], Yasuhiro Shibasaki[1]

[1]Nihon University, College of Bioresource Sciences, Fujisawa, Kanagawa, Japan; [2]Fukui Prefectural University, Faculty of Marine Science and Technology, Obama, Fukui, Japan; [3]Czech Academy of Sciences, Biology Centre, Institute of Parasitology, České Budějovice, Czech Republic

Background & Objective: Teleost fish were, until recently, thought to lack organized lymphoid structures. However, our previous study revealed that melanomacrophage center-associated lymphoid aggregates (M-LAs) in the rainbow trout spleen function as germinal center analogs, supporting antigen-specific IgM^+ B cell proliferation. To test whether the spleen is indispensable for systemic IgM induction, we investigated immune responses using a splenectomy model.

Methods: Rainbow trout were surgically splenectomized or sham-operated, and subsequently immunized with DNP-KLH. Over a 12-week period, we analyzed total and antigen-specific IgM levels, as well as the distribution of IgM^+ and IgT^+ B cells across various tissues (blood, spleen, head kidney, anterior and posterior trunk kidney, gut, and gills).

Results: Splenectomized fish exhibited a marked decrease in DNP-specific IgM levels and IgM^+ B cell numbers compared to sham-operated controls. While the spleen contained the highest proportion of antigen-specific IgM^+ B cells in sham-operated fish, a compensatory increase in these cells was specifically observed in the anterior trunk kidney at later time points following splenectomy. The proportion of antigen-specific IgT^+ B cells remained highest in mucosal tissues (gut and gills), regardless of splenectomy.

Conclusion: The spleen is the primary site for systemic antigen-specific IgM responses in rainbow trout. Although the anterior trunk kidney can partially compensate for its absence, splenectomy markedly attenuates systemic IgM production while IgT-mediated mucosal immunity is preserved.

Key words: rainbow trout; antibody response; spleen; splenectomy; B cell; IgM; IgT

Acknowledgements: This work was supported by the JSPS Bilateral Program with the Czech Academy of Sciences (CAS) (grant number JPJSBP120252503) and by the Japan Science and Technology Agency (JST) FOREST Program (grant number JPMJFR233M).

LECTIN-BASED IMMUNOPHENOTYPING REVEALS SPECIES-SPECIFIC GLYCOSYLATION PATTERNS AND HIDDEN LEUKOCYTE HETEROGENEITY IN TELEOST FISH

Thilini Hettiarachchi[1], Jiří Kyslík[1], Bharath Raja[1], Tomáš Korytář[1,2]

[1]Czech Academy of Sciences, Biology Centre, Institute of Parasitology, České Budějovice, Czech Republic; [2]University of South Bohemia in České Budějovice, Faculty of Fisheries and Protection of Waters, South Bohemian Research Centre of Aquaculture and Biodiversity of Hydrocenoses, Vodňany, Czech Republic

Current immunophenotyping in teleost fish is constrained by the limited availability of lineage-specific antibodies, restricting the identification of functionally distinct immune-cell populations. As cell-surface glycosylation changes accompany immune-cell differentiation and activation, fluorochrome-conjugated lectins may provide a complementary, non-targeted approach for resolving leukocyte heterogeneity. We evaluated the lectins Concanavalin A (ConA), Wheat Germ Agglutinin (WGA), and Peanut Agglutinin (PNA) in combination with IgM immunostaining by flow cytometry. In rainbow trout peripheral blood leukocytes, WGA co-staining with IgM resolved five reproducible leukocyte populations, comprising $IgM^{+}WGA^{+}$, $IgM^{-}WGA^{+}$, $IgM^{-}WGA^{-}$ lymphocytes, and WGA^{+} and WGA^{-} myeloid populations, whereas ConA showed intermediate binding and PNA exhibited minimal reactivity. In contrast, none of the lectins produced prominent staining of carp leukocytes, revealing pronounced species-specific differences in leukocyte glycosylation. Ongoing work is extending this approach to zebrafish, Nile tilapia, European perch, and sturgeon species using a multiparametric multicolour lectin panel to establish comparative glycosylation signatures across teleosts. The identified lectin-defined populations will subsequently undergo functional validation to determine whether distinct glycosylation phenotypes correspond to differences in immune function, thereby establishing the utility of non-targeted glycan profiling for resolving hidden leukocyte heterogeneity in fish.

Key words: lectins; glycosylation; flow cytometry; immunophenotyping; teleosts

Acknowledgements: This research was supported by the Czech Science Foundation (grant no. 23-08042K).

BLACK HOLE OF THE UNKNOWN: RESULTS ON HYPERPIGMENTATION IN THE COMMON DAB (*LIMANDA LIMANDA*)

Mikolaj Adamek[1], Torben Krebs[1], Alice Westphal[1], Filip Średniawa[2], Michael Gabel[3], Verena Jung-Schroers[1], Jörn Peter Scharsack[3]

[1]University of Veterinary Medicine Hannover, Fish Disease Research Unit, Hannover, Germany; [2]Jagiellonian University, Faculty of Biology, Institute of Zoology and Biomedical Research, Department of Evolutionary Immunology, Krakow, Poland; [3]Thünen Institute of Fisheries Ecology, Bremerhaven, Germany

In recent years, the increasing incidence of hyperpigmentation in the common dab (*Limanda limanda*) has been investigated in connection with environmental stressors, pollution and pathogens. This pigment anomaly has been linked to hyperplasia of pigment cells (melanophores and iridophores). Heavily pigmented dab exhibited a higher incidence of an active immune response, characterised by dermal lymphocytic infiltration. However, this immune response could not previously be linked to the infectious agents investigated, and the underlying cause remains unclear. Therefore, we have used this currently idiopathic disease to trial new diagnostic approaches based on metabarcoding of bacteria and further immune response profiling.

Skin samples were taken from dab in the North Sea during a scientific expedition and preserved in RNAlater. Where possible, samples were taken from both hyperpigmented and normal skin on the same individuals. The bacterial microbiome was investigated and determined using ONT sequencing of the complete 16S rRNA. RNA-seq and RT-qPCR was performed to identify any immune response leading to the hyperpigmentation changes.

The investigation of the microbiome of paired (hyper-pigmented/normally pigmented) skin samples from selected individuals revealed over 450 bacterial species. The microbiomes of individual fish were highly unique, with hypermelanistic and non-melanistic skin samples from the same animal showing similarity. Furthermore, there were no indications of an clear antiviral/bacterial response in the hypermelanistic samples. Also, levels of immune markers for red and hypermelanistic focal muscle changes in salmon did not yield conclusive results.

Our study showed that there are no differences in the microbiomes of normal and hyperpigmented dab skin. Furthermore, there were no signs of an immune response to an infection. Further sampling and investigative methods are therefore necessary to solve the mystery of the causes of hyperpigmentation in dab; however, the evidence increasingly suggests that it is not pathogen-related.

Key words: common dab; hyperpigmentation; skin disease

Acknowledgements: The research was supported by European Union's Horizon Europe Research and Innovation Programme under grant agreement 101084204 Cure4Aqua.

RISK ASSESSMENT OF FISH DISEASES IN ARMENIA: AQUACULTURE GROWTH, TRANSBOUNDARY TRADE, AND NATIVE FISH CONSERVATION

Mikolaj Adamek[1], Roza Barseghyan[2], Hripsime Kobelyan[2], Verena Jung-Schroers[1], Alice Westphal[1], Tigran Vardanyan[2], Nelli Barseghyan[2]

[1]University of Veterinary Medicine Hannover, Fish Disease Research Unit, Hannover, Germany; [2]Scientific Centre of Zoology and Hydroecology, Institute of Hydroecology and Ichthyology, Laboratory of Ichthyology, Yerevan, Armenia

Armenian aquaculture has developed into a freshwater production sector that is oriented towards export and dominated by rainbow trout, although common carp, sturgeons and other species are also important. While its growth creates economic opportunities, it also introduces biosecurity risks due to the movement of farmed fish, hatchery materials, transport water, farm effluents and escaped fish. This is not only an aquaculture problem, but it can also threaten natural waters and native fish fauna.

As disease surveillance is in its infancy in Armenia, the project 'Diagnostics and mitigation of diseases of Armenian wild and farmed fish: towards preserving biodiversity and creating sustainable aquaculture and fisheries', hosted by the Scientific Centre of Zoology and Hydroecology, was initiated. The first step was to perform a risk assessment and SWOT analysis, combining trade-pathway pathogen threat analysis, farm biosecurity evaluation, access to molecular diagnostics, and the expertise of a fish health specialist.

Current evidence indicates that the highest-priority hazards for Armenian aquaculture and natural waters are bacterial diseases of trout, particularly enteric redmouth disease, flavobacteriosis and furunculosis, as well as high-consequence viral diseases of salmonids and cyprinids, including VHSV, IHNV, CyHV-3 and CEV. Parasitic hazards, including *Ichthyophthirius multifiliis*, monogeneans, *Ligula* and trematodes, are particularly important at the interface between aquaculture and wild fish.

As Armenia currently has limited fish-specific pathogen data, the most defensible approach is a risk-based surveillance framework, which is set to begin within the project. This should combine molecular-based diagnostics, bacteriology of fish and farm effluent, eDNA monitoring, farm biosecurity audits, and regional data sharing. This framework will protect farm productivity, preserve confidence in target export markets, and reduce the risk of spillover to Armenia's native and conservation-sensitive fish fauna.

Key words: risk assessment; savan trout; rainbow trout; disease

Acknowledgements: The research was supported by the Higher Education and Science Committee of MESCS RA (Research Project № 25RL-1F016).

COMMON CARP LEUKOCYTES RESTIMULATED WITH CARP EDEMA VIRUS PRODUCE SIGNIFICANTLY MORE CYTOKINES IN A MEMORY-LIKE RESPONSE

Monika Němcová[1], Lucie Kováčová[1,3], Šárka Bednaříková[1], Ivona Toulová[1,2,4], Ivana Papežíková[1,2,4], Ivana Mikulíková[1,2,4], Miroslava Palíková[1,2,4], Jiří Pikula[1]

[1]University of Veterinary Sciences Brno, Faculty of Veterinary Hygiene and Ecology, Department of Ecology and Diseases of Zoo Animals, Game, Fish and Bees, Brno, Czech Republic; [2]Mendel University in Brno, Faculty of AgriSciences, Department of Zoology, Fisheries and Hydrobiology, Brno, Czech Republic; [3]Department of Animal Protection and Welfare & Veterinary Public Health, University of Veterinary Sciences Brno, Brno, Czech Republic; [4]Veterinary Research Institute, Department of Infectious Diseases and Preventive Medicine, Brno, Czech Republic

Carp edema virus disease (CEVD) is one of the most economically important viral diseases of common carp worldwide. High mortality and the lack of treatment and prophylaxis tools compel veterinarians to research the immune response of infected fish. Here, we aimed to study the common carp (*Cyprinus carpio*) leukocyte response to carp edema virus (CEV) antigens under *in vitro* conditions at temperatures of 24 or 28 °C, simulating different metabolic levels in the poikilothermic carp. Whole blood leukocytes derived from fish with different CEV infection histories were treated with infectious fish gill homogenate, as a source of CEV particles, for 24 hours. Then, the expression of cytokine-coding genes was determined. Leukocytes obtained from individuals exhibiting symptoms of acute CEVD produced significantly higher levels of pro-inflammatory interleukin 6 but almost no anti-inflammatory interleukin 10 (IL10) in response to CEV treatment. On the contrary, leukocytes obtained from healed fish produced high levels of IL-10. Besides IL-10, the gene coding for interferon alpha was upregulated after CEV treatment, despite its low total expression level compared to healthy fish. The level of the immunoglobulin M-coding gene was higher in the culture derived from healed fish compared to healthy fish, but not necessarily in response to repeated infection. Overall, the cellular response against CEV was stronger at 28 °C than at 24 °C. Based on these results, we suggest that there is potential for immune cell memory to control the acute phase response, in which, after repeated infection, IL-10 may play an essential role in protecting the organism from self-damage. A certain level of circulating antibodies can protect the fish immediately after antigen recognition, although full immunoglobulin production develops with a delay of several days.

Key words: innate immunity; immune memory; repeated infection; interleukin; interferon

Acknowledgement: This study was funded by the Ministry of Education, Youth and Sports of the Czech Republic, the projects PROFISH (CZ.02.1.01/0.0/0.0/ 16_019 / 0000869).

CHARACTERISTIC BEHAVIOURAL TRAITS ASSOCIATED WITH INFECTION CAUSED BY THE CARP EDEMA VIRUS AND PREDATOR ATTACKS AS INDICATORS FOR AUTOMATED EARLY-WARNING SYSTEMS

Mikolaj Adamek[1], Christianah Oluwaseun Ojelade[2], Magdalena Marcinkowska[3,4], Krzysztof Rakus[3], Sebastian Wille[5], Verena Jung-Schroers[1]

[1]University of Veterinary Medicine Hannover, Institute for Parasitology, Fish Disease Research Unit, Hannover, Germany; [2]Federal University of Agriculture, College of Environmental Resources Management (COLERM) Phase I., Department of Aquaculture and Fisheries Management, Abeokuta, Nigeria; [3]Jagiellonian University, Faculty of Biology, Institute of Zoology and Biomedical Research, Department of Evolutionary Immunology, Krakow, Poland; [4]Jagiellonian University, Doctoral School of Exact and Natural Sciences, Krakow, Poland; [5]Wille Engineering, Hattersheim am Main (Eddersheim), Germany

Behavioural monitoring offers a non-invasive approach to identifying relevant health and animal welfare risks in aquaculture. In this study, the behaviour of carp during infection with the carp edema virus (CEV) and during a simulated predator attack was quantified using the automated behavioural analysis software Noldus. The objective was to investigate whether an infection and a predator attack elicit different behavioural patterns that can be used for the automated detection of various threats.

In the experiments, movement parameters, social behaviour, and behavioural reactivity were analysed. The predator attack was simulated by rapidly introducing a fast-moving object into the experimental tank to mimic a cormorant attack. Locomotor and social parameters were analysed and included total distance moved, mean velocity, maximum acceleration, duration of movement and inactivity, and cumulative duration of body contact and absence of body contact between fish. Behavioural responsiveness was additionally evaluated using a novel-object test.

The CEV infection was primarily characterized by a progressive decrease in locomotor activity. Infected fish exhibited a reduction in total distance travelled, mean swimming speed, and duration of movement. In contrast, the simulated predator attack triggered an immediate flight response, with maximum acceleration representing the most pronounced behavioural change.

The results show that biologically distinct threats can be distinguished from one another based on quantitative behavioural parameters. The CEV infection led to a sustained pattern of reduced activity, while the simulated predator attack triggered an acute, high-intensity movement response. As part of the MyDigiFish project, these behavioural traits contribute to the development of an automated early-warning system that alerts fish farmers to potential disease outbreaks or predator attacks.

Key words: behaviour, common carp, aquaculture, predators, detection

Acknowledgements: Funded by the Bundesanstalt für Landwirtschaft und Ernährung / das Bundesministerium für Landwirtschaft, Ernährung und Heimat. COO was supported by the German Research Foundation.

	country	e-mail address	page of presentation
Adamek Mikolaj	Germany	Mikolaj.Adamek@tiho-hannover.de	7; 8; 9; 20; 29; 45; 46
Balážová Alena	Czech Republic	balazovaa@vfu.cz	22
Bartošová-Sojková Pavla	Czech Republic	bartoska81@gmail.com	11
Birikorang Harriet Nketiah	Australia	harrietbirikorang@yahoo.com	21
Eszterbauer Edit	Hungary	eszterbauer.edit@vmri.hun-ren.hu	13; 19; 32
Fujihara Ryosuke	Japan	brry26501@g.nihon-u.ac.jp	30; 40; 43
Hettiarachchi Thilini	Czech Republic	thilinihtm@gmail.com	27; 44
Karbusová Kateřina	Czech Republic	kkarbusova@frov.jcu.cz	33
Kóczán Patrícia	Hungary	koczan.patricia@vmri.hun-ren.hu	13; 19; 32
Kolářová Jitka	Czech Republic	kolarova@frov.jcu.cz	18
Komárková Lucie	Czech Republic	lucik095@gmail.com	23
Korytář Tomáš	Czech Republic	tkorytar@paru.cas.cz	9; 20; 27; 28; 29; 30; 31; 33; 34; 40; 41; 43; 44
Kyslík Jiří	Czech Republic	jiri.kyslik@paru.cas.cz	27; 28; 34; 44
Lövy Alena	Czech Republic	lovy@paru.cas.cz	15
Matějíčková Kateřina	Czech Republic	katerina.matejickova@vri.cz	8; 10; 14; 17; 37
Maturová Terezie	Czech Republic	maturovat@vfu.cz	12
Mikulíková Ivana	Czech Republic	mikulikovai@vfu.cz	16; 17; 35; 36; 37; 39; 47
Němcová Monika	Czech Republic	nemcovam@vfu.cz	47
Nepejchalová Leona	Czech Republic	nepejchalova@uskvbl.cz	38
Palíková Miroslava	Czech Republic	palikovam@vfu.cz	10; 12; 16; 17; 20; 22; 35; 36; 37; 39; 47

Papežíková Ivana	Czech Republic	papezikovai@vfu.cz	16; 17; 35; 36; 37; 39; 47
Piačková Veronika	Czech Republic	piackova@frov.jcu.cz	8; 9; 25; 26; 33
Plevová Kristína	Czech Republic	kplevova@jcu.cz	24; 25
Pojezdal Ľubomír	Czech Republic	lubomir.pojezdal@vri.cz	8; 10; 14; 16; 17; 20; 29; 37; 39
Posel Petr	Czech Republic	petr.posel@aqua.cz	6
Raja Bharath	Czech Republic	bharath.raja@paru.cas.cz	34; 44
Rakus Krysztof	Poland	krzysztof.rakus@uj.edu.pl	7; 9; 20; 29
Sebastian Anju	Czech Republic	sebastian@frov.jcu.cz	8
Shao Chenkai	Japan	s2574004@g.fpu.ac.jp	41
Shibasaki Yasuhiro	Japan	shibasaki.yasuhiro@nihon-u.ac.jp	30; 40; 43
Takeda Shinji	Japan	brsh23508@g.nihon-u.ac.jp	31
Takizawa Fumio	Japan	takizawa@g.fpu.ac.jp	30; 31; 40; 41
Toulová Ivona	Czech Republic	toulovai@vfu.cz	12; 16; 17; 35; 36; 37; 39; 47
Vaibarová Věra	Czech Republic	vaibarovav@vfu.cz	35; 37; 42
Vámosi Réka	Hungary	vamosi.reka@vmri.hun-ren.hu	13; 19; 32
Všetičková Lucie	Czech Republic	lucie.vsetickova@mendelu.cz	36
Zusková Eliška	Czech Republic	zuskova@frov.jcu.cz	24; 25

INSTITUTE OF ZOOLOGY, FISHERIES, HYDROBIOLOGY AND APICULTURE OF MENDEL UNIVERSITY IN BRNO

DEPARTMENT OF FISHERIES AND HYDROBIOLOGY

The Department of Fisheries and Hydrobiology at the Faculty of Agriculture of Mendel University in Brno has been providing teaching and research since 1949.

The education of students focuses on freshwater fisheries (aquaculture and fish health, ichthyology, management of open waters) and the related hydrobiological issues, including ecotoxicology.
The studies in fisheries and hydrobiology are part of the three-year bachelor program and two-year master studies program. Graduated students are also eligible to apply for the PhD studies.

Research activities are focused on fish breeding (development of intensive fish culture technologies in controlled conditions), monitoring of the changes in aquatic environments due to anthropogenic activities (heavy metal contamination, contamination of freshwaters by cyanobacterial blooms, eutrophication of surface waters), and the study of natural fish populations. The students are closely involved in solving these issues during the writing of their final paper for the Bachelors studies, their Master's and PhD thesis.

The department is equipped with:

- a recirculation system for fish farming (accredited experimental facility)
- a hydrobiological laboratory which provides analyses of aquatic invertebrates
- a hydrochemical and toxicological laboratory that provides analyses of water and sediments
- equipment for sampling and basic processing of fish blood and tissues
- equipment for fish sampling in open waters (electrofishers, nets)

MENDELU Faculty of AgriSciences

rybarstvi.eu

Oddělení rybářství a hydrobiologie

rybarimendelu

RYBÁŘSTVÍ & HYDROBIOLOGIE TRADICE 1949 MENDELU v Brně

UNIVERSITY OF VETERINARY SCIENCES BRNO

FACULTY OF VETERINARY HYGIENE AND ECOLOGY

The Faculty of Veterinary Hygiene and Ecology (FVHE) is one of the two faculties of the University of Veterinary Sciences Brno, which as the only university in the Czech Republic educates future veterinarians and not only them. The FVHE specializes in veterinary hygiene, food safety and quality, veterinary public health, animal protection and welfare, environmental protection, and ecological aspects of all these areas.

Veterinary education at the university is bound by a more than 100-year tradition and the Faculty of Veterinary Hygiene and Ecology has been included in the prestigious list of international educational level assessments of the European Association of Establishments for Veterinary Education (EAEVE) for more than 20 years, which clearly demonstrates the quality of the education provided and ranks the faculty among the best that any European veterinary school offers to students.

Applicants can choose from four bachelor's, five master's and five doctoral study programs or from the offer of lifelong learning events and the University of the third age.

- **VETERINARY HYGIENE AND ECOLOGY**
- **VETERINARY PUBLIC HEALTH**
- **FOOD SAFETY AND QUALITY**
- **ANIMAL PROTECTION AND WELFARE**
- **WILDLIFE ANIMAL HEALTH AND ENVIRONMENT PROTECTION**

www.fvhe.vetuni.cz

@vetuni_brno

www.facebook.com/VetuniBrno

Title: **The 2nd Meeting of the Central and Eastern European EAFP Branches Abstract Book**

Editors: Ivona Toulová, Miroslava Palíková, Ivana Papežíková, Ivana Mikulíková

Publisher: Mendel University in Brno, Zemědělská 1, 613 00 Brno, Czech Republic
Edition: 1st from 2026

The authors are responsible for the language and scientific content of their contributions.
The editors have made only those editorial revisions necessary for publication.

ISBN 978-80-7701-115-0 (online ; pdf)
https://doi.org/10.11118/ 978-80-7701-115-0