

**University of South Bohemia
Research Institute of Fish Culture and Hydrobiology**



Biennial Report
2004-2005

Vodňany, 2006

Biennial Report 2004-2005

Edited by: Blanka Vykusová

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PREFACE

Since March 2005, the directorial organization of USB RIFCH was obviously changed by incorporating internal management offices to the office of dean. Upon changing, developmental conception of institution was started by attempts of the following defined organization chart;

- 1) Development of USB RIFCH (Dean)
- 2) Economies (Assistant manager in economical purposes)
- 3) Science (Scientific secretary)
- 4) Training and Education (Assistant manager in training and education)
- 5) Information Technology (Assistant of IT)
- 6) Coordinator (Assistant in parts of technical activity in USB RIFCH)

Upon establishing new systemic organization chart, institutional convention (agreement) promoted which manifest positive institutional economic record in 2005. Directional policy was to develop staff directory by utilization of comprehensive passive and active elements; in this regard incentive components was also used. General conception of exerted policies at USB RIFCH was to evaluate and establish international scientific research and corporation as well post-graduated educational program (Doctorate Philosophy in Fishery) in long term. Attempts were made to join the insinuation workplace to contemporary world's trend in researches that are related to aquaculture or fisheries sciences (e.g., aquaculture project in European Union and sturgeon conservation program in Chinese Academy of Sciences). In this way knowledgeable, technical and scientific information and records are interchanged. In fact, the directorial offices of USB RIFCH tried to collaborate both nationally in Czech Republic and internationally, to issue and spread scientific obtained results either by presentation in symposium and conferences or by publishing in journals indexed by ISI web of knowledge and to provide young researchers with scientific opportunities by student fellowships or scholarships.

Experimental and evolutionary activities of institute are subsidiary of an extensive research plan USB RIFCH MSM6007665809 that has been defined in 2005. These programs are composed of three basic parts that subsequently divided into 15 themes. As well, institutional departments (Fish Genetics and Breeding, Aquaculture and Hydrobiology, Aquatic Toxicology and Fish Diseases) opened new files for further research themes that some are applied as a project/grant proposal to Grant Agency of Czech Republic and Academy of Sciences of Czech Republic) and other are in preparation stage for application. All applied and ongoing research plans cover different research topics in fisheries sciences in Czech Republic. In term of Ph.D. program, USB RIFCH arranged some meeting to dispose conditions for registration and entry of talent students in fishery by contribution of member's fishery entrepreneurial sphere and civil department in Vodňany.

In part of non-academic activity, USB RIFCH cooperated with subjective tertiary sector education in region, especially with bachelor schools in Vodňany (College of Water Resource and Environmental Sciences and College of Fishery). Through sales executive USB RIFCH fast sought out project in the area structural fund with monitoring activities in regional and nation-wide institution. To reach parts of institutional defined aims, it was organized some meetings with members of different committees, organizations and directories characterized in ministries of Agriculture and Science. It was due to explain and evaluate financial status of determined plan in terms of policy, limitation, policy and solutions.

Office of Information technology tried to develop and modernize computer network, LAN and telephone network. Other aim was to create web site of institute on the world internet web (www.vurh.jcu.cz). Organization chart, research plan and activities and laboratory and filed facilities are introduced in Czech and English.

Some parts of ongoing activities of USB RIFCH that defined in 2006 are including;

- Mobilizing of Molecular Biology and Confocal Microscopy laboratories;
- Preparation of necessity for realization and establishment of "European Fishery Research, Technology and Education Centre" that would be held in USB RIFCH Vodňany; its financial aid would be supported by European Structural Fund in 2007-2011 for reconstruction (investment equal to 139 millions of CZ crowns);
- Collaboration of RIFCH and Faculty of Agriculture at USB to establish an International Fishery Program in Master of Science (M.Sc) for European student;
- Scientifically most considerable original work near young worker to the 35 years prize director of USB RIFCH;

- Seeking to talent students in fishery for their doctorate fishery study and providing them with academic carrier including field and laboratory research, studentship with capability of short-stay in related laboratories abroad, participating in symposium, conferences and meeting as well class with tribal worker of USB RIFCH;
- On line electronic publication of bulletin of USB RIFCH Vodňany at the web page of institute;
- Innovation of web page of institute by including of information about Ph.D. post-graduate program in fishery and related items;
- Beside basic university research permanent nurse region applied research in cooperation with business sphere;
- Gear institution to the programmed SOCRATES, ERASMUS MUNDUS etc., which wasn't before accreditation of doctorate fishery study.

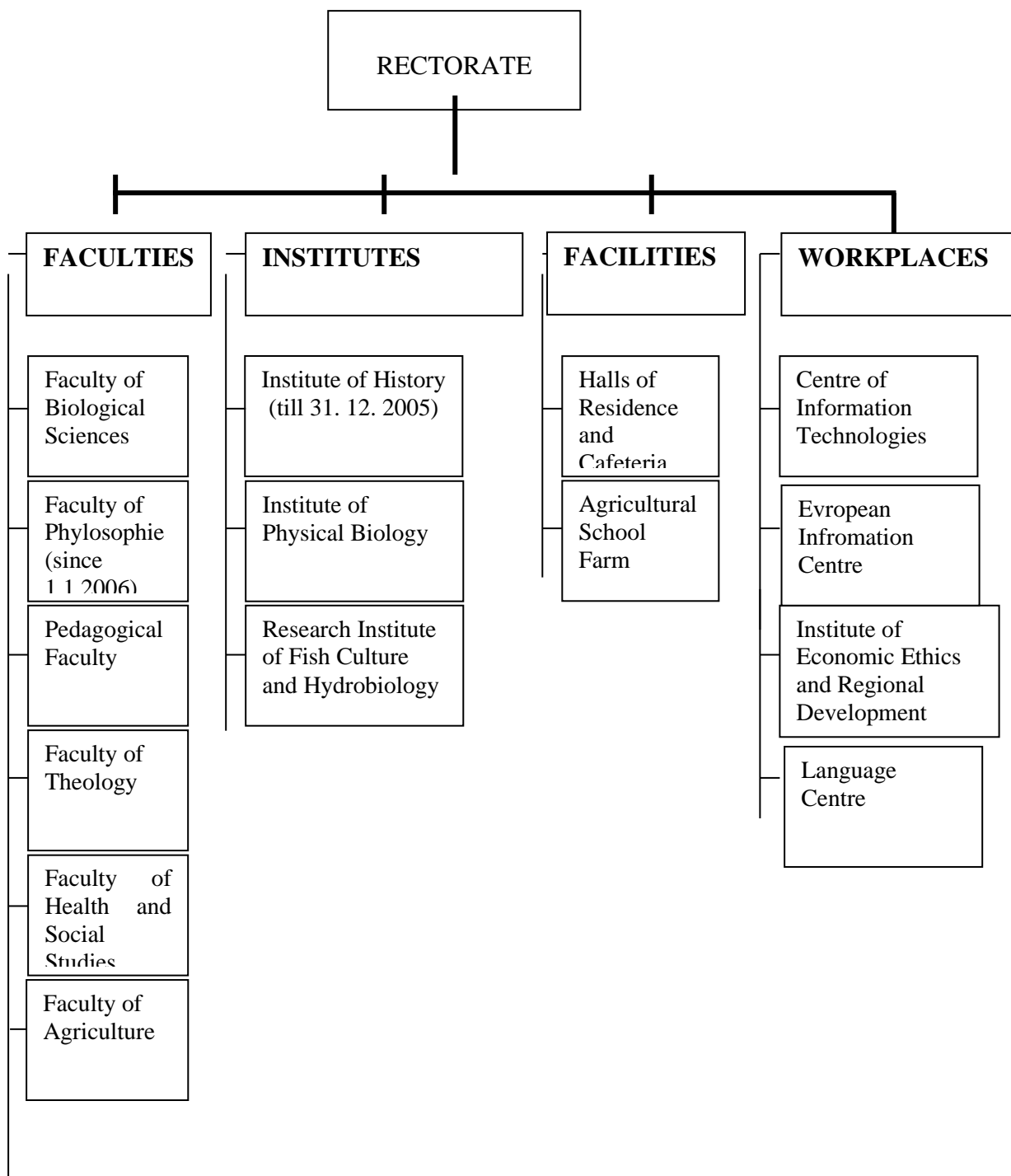
Prof. Dipl.-Ing. Otomar Linhart, DrSc.
director

CONTENTS

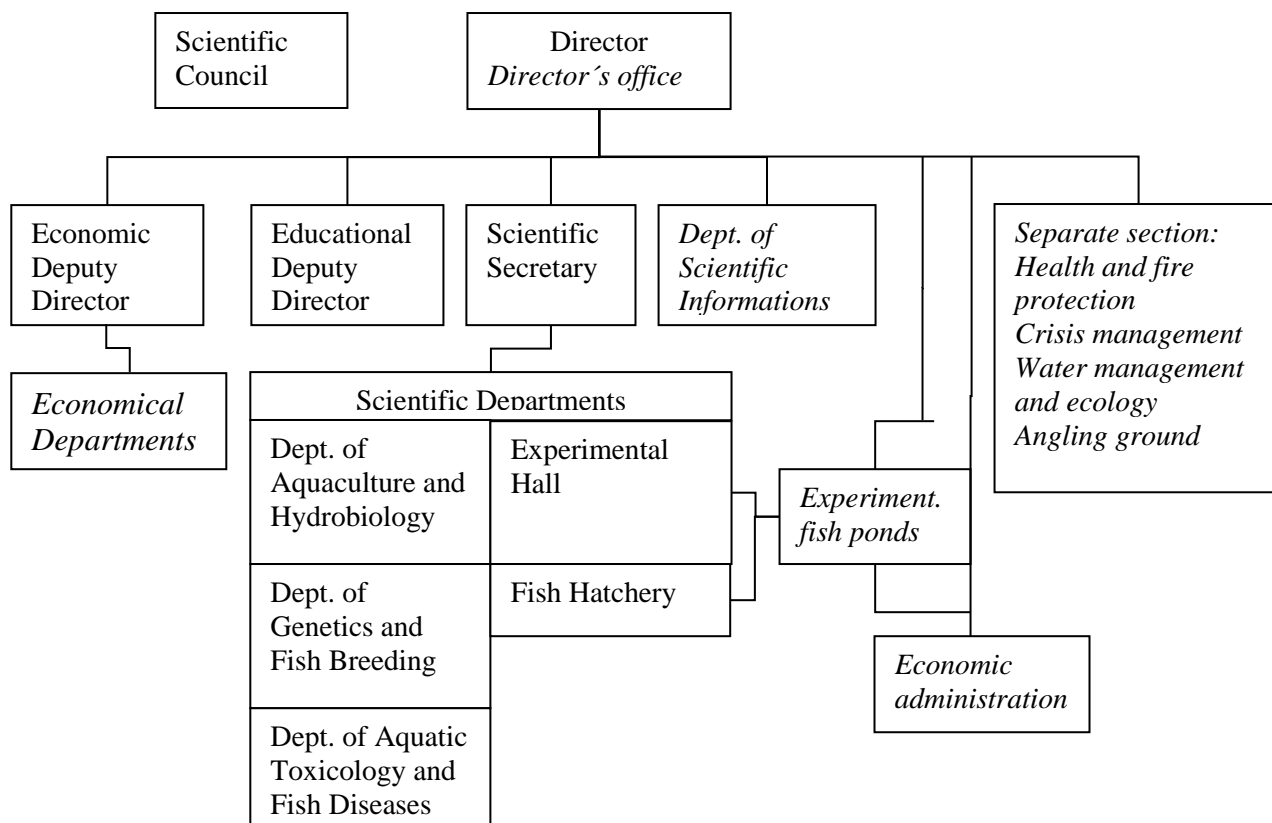
Preface	3
Organization structure	6
Addresses	7
Management and Services	8
Scientific Council	9
Research projects	10
List of publications	12
Scientific departments	22
International activities	43
Education and teaching activities	45
Editorial and popularization activities	47

ORGANIZATION STRUCTURE

University of South Bohemia České Budějovice



RIFCH USB



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Main building:

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MANAGEMENT AND SERVICES

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Dipl.-Ing. *Jan Kouřil*, Ph.D., since 1.3.2005
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Marta Polanská, head 1.5.2005 - 30.6.2005
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SCIENTIFIC COUNCIL

Till 18.11.2005:

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RIFCH USB Vodňany

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Faculty of Biological Sciences USB Č. Budějovice

RIFCH USB Vodňany

UWM WOSR Poland

RIFCH USB Vodňany

Faculty of Agriculture, USB České Budějovice

Faculty of Agriculture, USB České Budějovice

Faculty of Agriculture, MZLU Brno

RIFCH USB Vodňany

Institute of Hydrobiology ASCR České Budějovice

Institute of Vertebrate Biology ASCR, Brno

České Budějovice

Institute of animal physiology and genetics ASCR

Liběchov

Faculty of Agriculture, USB České Budějovice

Museum Silesiae, Opava

SPU AF, Slovak Republic

RIFCH USB Vodňany

Czech Fish Farmer Association, České Budějovice

Czech Fishing Union, Regional Union West

Bohemia, Plzeň

RIFCH USB Vodňany

RIFCH USB Vodňany

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Dipl.-Ing. Jiří Řehulka, CSc.

Assoc.Prof.Ing.Ivan Straňai, CSc.

Prof.MVDr. Zdeňka Svobodová, DrSc.

Dipl.-Ing. Václav Šilhavý

Dipl.-Ing. Jan Štípek

Assoc.Prof. Dipl.-Ing. František Vácha, CSc.

Dipl.-Ing. Blanka Vykusová, CSc.

Since 18.11. 2005::

Chairperson:

doc. Ing. *Otomar Linhart*, DrSc.

RIFCH USB Vodňany

Internal members:

Dipl.-Ing. *Jan Kouřil*, Ph.D.

Dipl.-Ing. *Pavel Kozák*, Ph.D.

Dipl.-Ing. *Martin Flajšhans*

Ing. *Martin Kocour*

RNDr. *Dalibor Štys*, CSc.,

Assoc.Prof. Dipl.-Ing. *Petr Hartvich*, CSc.

prof. RNDr. *Libor Grubhoffer*, CSc.

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IPB USB České Budějovice

Faculty of Agriculture, USB České Budějovice

Faculty of Biological Sciences USB Č. Budějovice

External members:

prof. MVDr. *Zdeňka Svobodová*, DrSc.,

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Assoc.Prof. Ing. *Petr Ráb*, DrSc.,

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RNDr. *Jan Pokorný*, CSc.

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Veterinary and Pharmaceutical University, Brno

Mendel University of Agriculture and Forestry

Brno

Institute of animal physiology and genetics ASCR

Liběchov

Institute of Hydrobiology ASCR České Budějovice

ENKI p.b.c., Třeboň

Agency for Nature Conservation and Landscape

Protection of the Czech Republic

Ministry of Agriculture of the Czech Republic

Praha

RESEARCH PROJECTS

2004

Projects supported by the Ministry of Education, Youth and Sport

Institutional research plans

- MSM126100001 – Biological bases of fresh water culture, 1999-2004.
- MSM126100003 – Assessment of interactions among danger factors in water environment and ecosystems, 1999-2004.

Bilateral projects

- Programme KONTAKT ME638 – Comparative biological study of chromosomally manipulated and selectively bred populations of tench in China and the Czech Republic – M. Flajšhans, 2003-2004
- Programme KONTAKT ME615 – Comparative study of reproduction biology and genetic differences in populations of tench in China and the Czech republic – O. Linhart, 2003-2004
- Programme BARRANDE (KONTAKT) 2004-044-1 – Determination of suitable selection scheme for improved growth in common carp – O. Linhart, 2004-2005.

Projects supported by the Grant Agency of the Czech Republic

- 206/03/D064 – Biological study of invasive spiny-cheek crayfish *Orconectes limosus* Raf. under laboratory conditions – P. Kozák, 2003-2006 .
- 206/03/0532– Population ecology of terminal and resident populations of invasive crayfish species – P. Kozák, (coordinated by Faculty of Science, University of Ostrava, Czech Republic), 2003-2005.
- 524/03/0178 – Detail study of spermatozoa in model species of cartilaginous (*Chondrostei*) and bony (*Teleostei*) fishes – O. Linhart, 2003-2005.

Projects supported by Grant Agency of the Czech Academy of Science

- IBS4055006 – Utilization of biologically active compounds in reproduction of fish, J.Kouřil (coordinated by Institute of Organic Chemistry and Biochemistry, CAS, Prague), 2000-2004.
- KSK6005114 – Biodiversity and function of ecological systems – O. Linhart (coordinated by Entomological Institute CAS, České Budějovice), 2002-2004.

Projects supported by Ministry of Agriculture (National Agency for Agricultural Research)

- QF3028 – Development of new technologies in rearing of important river fish and crayfish species endangered by degradation of environment – P. Kozák, 2003-2007.
- QF4118 – Development of fish reproduction with utilization of technical aquaculture and their mutual combination with pond culture – J. Kouřil, 2003-2007.
- QF4117 – Influence of mass selection in common carp (*Cyprinus carpio* L.) on performance of growth-related traits its progeny with using of molecular methods – M. Kocour, 2003-2007.
- 1B44016 – Protection of common carp (*Cyprinus carpio* L.) culture systems before the disease caused with KOI herpes virus (KHV) – V. Piačková, 2004-2006.
- QF3029 – Harmonization with EU in implementation of principles of farmacovigilance in aquaculture of the Czech Republic – J. Kolářová, 2003-2006.

Projects supported by Ministry of Environment

- SA/650/5/03 – Labe IV – T. Randák (coordinated by T. G. Masaryk Water Research Institute, Prague), 2003-2006.
- SM/730/1/03 – Research on distribution of PCB-like compounds in food chains and their accumulation in organisms; methods of withdrawal of contaminated waste – J. Máchová (coordinated by AQUATEST, Prague), 2003-2004.

Project supported by European Commission

5th Framework programme

- IFIBO - Intensive Fish Culture Optimalization – J. Kouřil (coordinated by Czech Technical University, Prague), 2001-2004.

6th Framework programme

- COOP-CT-2004 512629 - Securing juvenile production of Eurasian perch by improving reproduction and larval rearing – O. Linhart (coordinated by University of Henri Poincaré, Nancy, France), 2004-2006.
- CSN-INTRAN- Creating Supporting Network for International Transfer of Innovative Technologies in European Aquaculture – Z. Adámek (coordinated by University of Sterling, Scotland, UK), 2003-2006.

Projects supported by the Ministry of Education, Youth and Sport*Institutional research plan*

- MSM6007665809 – Biological, environmental and breeding aspects in fish culture, 2005-2011.

Bilateral projects

- Programme KONTAKT 1P05 ME742 – Short term storage and cryopreservation of fish embryos – O. Linhart, 2005-2006.
- Programme KONTAKT ME743 – Comparative biological study of chromosomally manipulated and selectively bred populations of tench in China and the Czech Republic – D. Gela, 2005-2006.
- Programme BARRANDE (KONTAKT) 2004-044-2 – Determination of suitable selection scheme for improved growth in common carp – O. Linhart, 2004-2005.

Projects supported by the Grant Agency of the Czech Republic

- 206/03/D064 – Biological study of invasive spiny-cheek crayfish *Orconectes limosus* Raf. under laboratory conditions – P. Kozák, 2003-2006.
- 206/03/0532 – Population ecology of terminal and resident populations of invasive crayfish species – P. Kozák (coordinated by Faculty of Science, University of Ostrava, Czech Republic), 2003-2005.
- 524/03/0178 – Detail study of spermatozoa in model species of cartilaginous (*Chondrostei*) and bony (*Teleostei*) fishes – O. Linhart, 2003-2005
- 206/05/2159 – Genetic, population and reproductive variability of invasive fish, *Carassius gibelio*, with alternative bisexually/sexual reproduction in the Central Europe – M. Flajšhans (coordinated by Institute of Vertebrate Biology, CAS, Brno), 2005-2007.

Project supported by Internal Grand Agency of the University of South Bohemia

- 24/2004/P-VURH – Study of nitrites effect on fish with aim to minimize their negative influence – H. Kroupová, 2005.

Projects supported by Ministry of Agriculture (National Agency for Agricultural Research)

- QF3028 – Development of new technologies in rearing of important river fish and crayfish species endangered by degradation of environment – P. Kozák, 2003-2007.
- QF4118 – Development of fish reproduction with utilization of technical aquaculture and their mutual combination with pond culture – J. Kouřil, 2003-2007.
- QF4117 – Influence of mass selection in common carp (*Cyprinus carpio* L.) on performance of growth-related traits its progeny with using of molecular methods – M. Kocour, 2004-2007.
- 1B44016 – Protection of common carp (*Cyprinus carpio* L.) culture systems before the disease caused with KOI herpes virus (KHV) – V. Piačková, 2004-2006.
- QF3029 – Harmonization with EU in implementation of principles of farmacovigilance in aquaculture of the Czech Republic – V. Piačková, 2003-2007.
- QG50058 – Utilization of reproductive potential of *Pseudorasbora parva* as prey fish during rearing of pikeperch (*Sander lucioperca*) fingerlings – J. Kouřil, 2005

Projects supported by Ministry of Environment

- SA/650/5/03 – Labe IV T. Randák (coordinated by T. G. Masaryk Water Research Institute, Prague), 2003-2006.

Project supported by European Commission*6th Framework programme*

- COOP-CT-2004 512629 (6th FP) - Securing juvenile production of Eurasian perch by improving reproduction and larval rearing – O. Linhart (coordinated by University of Henri Poincaré, Nancy, France), 2004-2006.
- COOP-CT-2004 512575-PROTENCH - Intensive and sustainable culture of the freshwater species tench – M. Kocour (coordinated by GESINFIN, Spain), 2005-2006.
- CSN-INTRAN (6th FP) - Creating Supporting Network for International Transfer of Innovative Technologies in European Aquaculture – Z. Adámek (coordinated by University of Sterling, Scotland, UK), 2003-2006.

LIST OF PUBLICATIONS

2004

Papers in journal included in the database of the Web of Science

- Adámek Z.; Andreji J.; Henshaw A. 2004. Stripping fecundity of common bream (*Abramis brama* L.) from the rivers Trent and Sow (Nottinghamshire, UK). *Aquaculture International*. 12(1):133-137
- Brzuska E.; Kouřil J.; Adamek J.; Stupka Z.; Bekh V. 2004 The application of (D-Tle,ProNHEt) mGnRH (Lecilerin) with the dopaminergic inhibitor metoclopramide to stimulate ovulation in African catfish (*Clarias gariepinus*). *Czech journal of animal science: Živočišná výroba*. 49(7):297-306
- Buchtová H.; Smutná M.; Vorlová L.; Svobodová Z.; Flajšhans M. 2004. Fatty Acid Composition of Diploid and Triploid Populations of Tench (*Tinca tinca* L.). *Acta veterinaria*. 73(2):235-245
- Drastichová J.; Svobodová Z.; Lusková V.; Čelechovská O.; Kaláb B. 2004. Effect of cadmium on blood plasma biochemistry in carp (*Cyprinus carpio* L.). *Bulletin of Environmental Contamination and Toxicology*. 72(4):733-740
- Drastichová J.; Svobodová Z.; Lusková V.; Máchová J. 2004. Effect of cadmium on hematological indices of common carp (*Cyprinus carpio* L.). *Bulletin of Environmental Contamination and Toxicology*. 72(4):725-732
- Flajšhans M.; Cosson J.; Linhart O. 2004. The application of image cytometry for viability assessment of dual fluorescent - stained fish spermatozoa. *Cell Biology International*. 28(12):955-959
- Flajšhans M.; Kocour M.; Gela D.; Piačková V. 2004. The first results on interactions among diploid, gynogenic and triploid tench, *Tinca tinca* L. under communal testing. *Aquaculture International*. 12(1):103-118
- Hamáčková J., Lepičová A., Kozák P., Stupka Z., Kouřil J., Lepič P. 2004. The efficacy of various anaesthetics in tench (*Tinca tinca* L.) related to water temperature. *Veterinary medicine*. 49(12):467-472
- Koubek P., Elzeinová F., Linhart O., Pěkníková J. 2004. Study of fish sperm proteins using monoclonal antibodies. *American Journal of Reproductive Immunology*. 51(6):483
- Kozák P., Polícar T., Ďuriš Z. 2004. Migration ability of *Orconectes limosus* through a fishpass and notes on its occurrence in the Czech Republic. *Bull. Fr. de la Peche et de la Pisciculture*. 372/373:367-373
- Linhart O., Pěkníková J., Rodina M. 2004. Factors regulating the motility of fish sperm. *American Journal of Reproductive Immunology*. 51 (6):464
- Linhart O., Rodina M., Gela D., Kocour M. 2004. Optimalization of artificial propagation in European catfish, *Silurus glanis* L. *Aquaculture*, 235:619-632
- Polícar T., Simon V., Kozák P. 2004. Egg incubation in the noble crayfish (*Astacus astacus* L.): The effect of controlled laboratory and outdoor ambient condition on hatching success, growth and survival rate of juveniles. *Bull. Fr. Peche Piscic*. 372-373: 411-423
- Rábová M., Ráb P., Boron A., Bohlen A., Janko K., Šlechtová V., Flajšhans M. 2004. Cytogenetic of bisexual species and their asexual hybrid clones in European spined loach, genus *Cobitis*. I. Karyotypes and extensive polymorphism of major ribosomal sites in four parental species. *Cytogenetic and Genome Research*. 106(1):24
- Randák T., Žlábek V., Kolářová J., Svobodová Z., Hajšlová J., Šířoká Z. 2004. Contamination assessment of Elbe river and its tributaries by means of biomarker detection and chemical analyses in chub (*Leuciscus cephalus* L.). *Toxicology and Applied Pharmacology*. 197(3):188
- Rodina M., Cosson J., Gela D., Linhart O. 2004. Kurokura solution as immobilizing medium for spermatozoa of tench (*Tinca tinca* L.). *Aquaculture International*, 12(1):119-131
- Rzemieniecki A., Domagala J., Glogowski J., Ciereszko A., Trzebiatowski R., Kouřil J., Hamáčková J., Babiak I. 2004. Induced spermiation in 3-year-old sterlet, *Acipenser ruthenus* L. *Aquaculture Research*. 35(2):144-151
- Svobodová Z., Čelechovská O., Kolářová J., Randák T., Žlábek V. 2004. Assessment of contamination by metals in the upper reaches of the Tichá Orlice River. *Czech journal of animal science : Živočišná výroba*. 49(10):458-464
- Svobodová Z., Kolářová J. 2004. A review of the diseases and contaminant related mortalities of tench (*Tinca tinca* L.). *Veterinární medicína: Veterinary Medicine - Czech*. 49(1):19-34
- Svobodová Z., Žlábek V., Randák T., Máchová J., Kolářová J., Hajšlová J., Suchan P., Dušek L., Jarkovský J. 2004. Profiles of PCBs in tissues of marketable common carp and in bottom sediments of selected ponds in South and West Bohemia. *Acta veterinaria*. 73(1):133-142
- Šetlíková I., Adámek Z. 2004. Feeding selectivity and growth of Nile tilapia (*Oreochromis niloticus* L.) fed on temperate-zone aquatic macrophytes. *Czech J.Anim.Sci*. 49(6):271-278

- Šíroková Z., Krijt J., Randák T., Svobodová Z. 2004. Cytochrome P450 induction in fish as a biochemical marker of Labe river contamination. *Toxicology and Applied Pharmacology*. 197(3):189
- Vandeputte M., Kocour M., Mauger S., Dupont-Nivet M., De Guerry D., Rodina M., Gela D., Vallod D., Chevassus B., Linhart O. 2004. Heritability estimates for growth-related traits using microsatellite parentage assignment in juvenile common carp (*Cyprinus carpio* L.). *Aquaculture*. 235(1-4):223-236
- Velíšek J., Svobodová Z. 2004. Anaesthesia of rainbow trout (*Oncorhynchus mykiss*) with 2-phenoxyethanol: Acute Toxicity and Biochemical Blood Profile. *Acta veterinaria*. 73(3):379-384
- Velíšek J., Svobodová Z. 2004. Anaesthesia of common carp (*Cyprinus carpio* L.) with 2-phenoxyethanol: Acute Toxicity and Effects on Biochemical Blood Profile. *Acta veterinaria*. 73(2):247-252
- Žlábek V., Randák T., Kolářová J., Svobodová Z., Hajšlová J., Suchan P. 2004. Monitoring of endocrine disruption in chub (*Leuciscus cephalus*) from the Vltava River. *Toxicology and Applied Pharmacology*. 197(3):191

Papers in other journals

- Adámek Z.; Musil J.; Sukop I. 2004. Diet composition and selectivity in 0+ perch (*Perca fluviatilis* L.) and its competition with adult fish and carp (*Cyprinus carpio* L.) stock in pond culture. *Agriculturae Conspectus Scientificus*. 69(1):21-27
- Buchtová H.; Flajšhans M.; Svobodová Z.; Tremlová M. 2004. The induced polyploidy influence on a tench *Tinca tinca* (Linnaeus, 1758) gonadogenesis. *Veterinářství. Odborný a stavovský měsíčník*. 54(2):107-112
- Drastichová J.; Šíroková Z.; Žlábek V. 2004. Vitellogenin as a biomarker for exposure of fish to estrogenic chemicals. *Folia veterinaria*. 48(3):114-118
- Flajšhans M.: Conservation of rare breeds of freshwater fish. Six years of experience with the National Programme of Animal Gene Resources. Conservation in the Czech Republic. SAVE Foundation Report. 2002; :14-15
- Gavel A.; Maršálek B.; Adámek Z. 2004. Viability of *Microcystis* colonies is not damaged by silver carp (*Hypophthalmichthys molitrix*) digestion. *Algological Studies* 113, *Arch. Hydrobiol. Suppl.* 153:189-194
- Piačková V., Randák T., Žlábek P. 2004. The toxic metals contamination of tissues of fish and bottom sediments from Blanice river. *Bulletin VÚRH Vodňany*. 40(3):125-130
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Scientific Citation Index (SCI):
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Dept. of Aquaculture and Hydrobiology		
Dipl.-Ing. Pavel Kozák, Ph.D.	0	8
Dipl.-Ing. Jan Kouřil, Ph.D.	12	8
Assoc. Prof. RNDr. Zdeněk Adámek, Ph.D.	4	3
Dipl.-Ing. Jitka Hamáčková	9	10
Dipl.-Ing. Tomáš Polícar	0	7
Dipl.-Ing. Vlastimil Stejskal	nehodnocen	0
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Dipl.-Ing. Martin Kocour	0	2
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Dipl.-Ing. Martin Hulák	0	0
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Dept. of Scientific Informations		
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Dept. of aquaculture and hydrobiology – from the left standing: Jan Kouřil, Vlastimil Stejskal, Pavel Kozák, Tomáš Polícar a Jiří Musil; in the front line: Pavel Lepič, Petra Vorlíčková, Jitka Hamáčková a Petra Martinková

RESEARCH PRIORITIES

The department studies both the fundamental and applied researches and collaborates with university in training under-graduate students. The main objective of their researches is to optimize methods of artificial spawning and intensive culture of freshwater fish (including artificial and semi-artificial spawning, optimization of environmental conditions for nursing the early fry stages, optimization of food intake in fish reared under intensive culture, assessment of effectiveness of recirculation systems and modern methods of using of anesthetic in intensive culture). Fish laboratory models are barbus, perch, catfish, brook trout and sturgeons.

Research combines intensive culture and semi-intensive culture of fish in recirculation system and pond conditions, respectively. Applied fisheries hydrobiology issues (including quality of environment and biological assessment of pond environment degradation, food biology of zooplanktonivorous and piscivorous fish) are investigated under pond conditions.

Special attention is paid to losses caused by piscivorous predators in still and running waters in the Czech Republic.

In addition to studies on freshwater fish, a few researches are focused also on crayfish biology and culture. In this regard, researches carried out on the monitoring of autochthonous and alien crayfish species in inland water bodies in the Czech Republic, biology of crayfish species, various methods of crayfish reproduction and culture, restocking and/or re-introducing of native crayfish to new natural localities and consultation services in case of crayfish kills or crayfish transfers.

There are seven main programmes of research on the department:

- Controlled artificial and semi-artificial fish reproduction
- Nursing the early fry stages, including optimization of environmental conditions
- Fish culture under intensive conditions
- Use of anaesthetics in aquaculture
- Applied fisheries hydrobiology
- Losses caused by piscivorous predators
- Research in crayfish biology and culture

SELECTED RESEARCH RESULTS

The application of anaesthetics in aquaculture

The effect of three different anaesthetics including 2-phenoxyethanol (0.6 ml.l⁻¹), Propiscin (0.75 ml.l⁻¹) and clove oil (0.033 ml.l⁻¹) were studied on the time periods necessary for the induction of particular characteristic phases of anaesthetisation and recovery of both male and female of tench (*Tinca tinca* L.) with mean of body weight 260 g (66 – 583 g) at different water temperatures (17.9; 20.4; 22.6 and 25.1 °C). At all temperatures, the statistically longest induction of anaesthesia (p<0.05) and longest recovery (p<0.01) were observed when Propiscin has been used. With clove oil and 2-phenoxyethanol, the time period necessary for induction of phase II b anaesthesia statistically declined (p<0.05) by increasing water temperature. However, this phenomenon was not seen with Propiscin.

Hamáčková, J., Lepičová, A., Kozák, P., Stupka, Z., Kouřil, J., Lepič, P., 2004: The efficacy of various anaesthetics in tench (*Tinca tinca* L.) related to water temperature. *Vet. Med.-Czech*, 49(12):467-472. (IF=0,621)

The application of preparations containing of GnRH analogues with/without dopaminergic inhibitor to induction of ovulation in bighead carp *Aristichthys nobilis*, silver carp *Hypophthalmichthys molitrix* and grass carp *Ctenopharyngodon idella*

In artificial reproduction of fish, the synchronization of female spawning is limited and therefore ovulation must be stimulated. Over the last 30 years, fish farmers have been using several types of products. The first generation, called hypophysation, is based on the injection of crude or dry fish pituitary extract, a natural mixture of several biological hormones. The second generation of products was developed as a result of various GnRH peptides (mammalian and salmon GnRH and their synthetic analogues). And the last generation contains GnRH analogues (GnRHa) and dopamine inhibitor (DI). This study was carried out to induce ovulation in three cyprinid herbivorous fish species. Combined preparates Ovopel and Dagin, containing GnRHa and DI metoclopramide as complementary substance, were injected singly in standard (recommended) doses and tested for ovulation induction, enabling artificial propagation of fish. Carp pituitary extract (CPE) injected in two separate doses was used as a control. In bighead and grass carp, application of Dagin (10 µg/kg of salmon GnRHa D-Arg⁶, ProNH₂Et and 20 mg/kg DI) was successful to ovulate 100 % of females. In grass carp, the application of preparate Dagin induced the ovulation in 67-100 % of females. Females of silver carp did not ovulate after administration of Dagin. In grass carp, the application of Ovopel (mammalian GnRHa D-Ala⁶, ProNH₂Et and 2 mg/kg DI) was successful (56-99 % of females ovulated). Partial positive result was observed in grass carp administered by of Supergestran, containing mammalian GnRHa Lecirelin (GnRHa D-Tle⁶, ProNH₂Et) in 200 µg/kg dose (71 % of females ovulated). Injection of CPE induced ovulation in 100% of bighead and silver carps but just 67 % of grass carp.

Rzemieniecki, A., Domagala, J., Glogowski, J., Ciereszko, A., Trzebiatowski, R., Kouřil, J., Hamáčková, J., Babiak, I. *Indukce spermiace tříletých jeseterů malých *Acipenser ruthenus* L. (Induced spermiation in 3-year-old sterlet, *Acipenser ruthenus* L.). *Aquaculture Research*, 2004, 35, 144-151. (IF 0,746)*

Induced ovulation and artificial spawning of barbel (*Barbus barbus*) females using different doses of GnRH analogue

An experiment was conducted to induce ovulation in barbel (*Barbus barbus*) originating from the river by a single injection of preparate Supergestran containing GnRHa Lecirelin at three different doses and carp pituitary as a control. Water temperature was 19.0 °C. Using of carp pituitary at dose 6 mg.kg⁻¹, the ovulation was induced in 60% females 21.8 hours post-injection. At low doses of Supergestran (5 and 25 µg.kg⁻¹ of GnRH) ovulation was induced in 40% of females 36.1 resp. 38.1 hours post activation. The best results were found when Supergestran has been injected in dose 125 µg.kg⁻¹. In this case, 80% of females spawned 35.9 hours post-injection. The relative weight of eggs obtained from the females treated with pituitary and low doses of Lecirelin were 3.47%, resp. 5.71 and 3.97%, and in 10.54% females treated with the highest dose of Lecirelin.

Oxygen consumption and total ammonium nitrogen (TAN) excretion in brook trout (*Salvelinus fontinalis*) reared at low temperatures

Six groups of market-size brook trout of individual weight in range of 100–190 g were fed by pellet amounting to 0.14-0.48 %/days of current biomass. This study was carried out in flow-trough 720-L tanks, at stocking density 20.80–23.96 kg m⁻³ and lasting for 105 days. Commercial feed Biomar Ecolife 19 (contains 48% crude protein, 25% total fat, 3 mm size) was supplied in three daily partial doses (at 7 a.m., 11 a.m. and 14 p.m.). The diurnal course of oxygen consumption and total ammonium nitrogen (TAN; NH₃ - N and NH₄⁺ - N mg kg⁻¹ hour⁻¹) excretion was recorded at four different temperatures (1.0, 1.3, 2.7 and 3.2 °C) with three

hours intervals. Significant correlation was observed between the mean daily oxygen consumption per unit of fish biomass and SFR at different water temperatures. At the lowest tested temperature (1.0 °C) and the lowest SFR (0.15 % day⁻¹) the O₂ consumption amounted to 16 mg kg⁻¹ hour⁻¹, but in contrast, it increased to 60 mg kg⁻¹ hour⁻¹ at the highest monitored temperature (3.2 °C) and the highest SFR 0.47 % day⁻¹. The linear regression functions are $y = 49.506x + 9.0158$ ($R^2 = 0.8674$) and $y = 36.055x + 43.363$ ($R^2 = 0.9114$) at temperatures 1.0 °C and 3.2 °C where y and x are O₂ consumption and the SFR, respectively.

Biological filtration efficiency at start operation of the recirculation system with fish culture

The experiment was performed in a recirculation system at RIFCH USB for 84 days (at average water temperature 22.56±2.70 °C, pH 6.23±0.63). Biomass of introduced fish was 250-300 kg and they were daily feed at rate of 1%.day⁻¹ of current biomass. The samples for chemical analyses (COD_{Mn}, N-NH₄⁺, N-NO₃⁻, N-NO₂⁻, P-PO₄⁻³, P_{total}, pH) were collected weekly. In water supplying the trickling filter, following average values were recorded: COD_{Mn} – 14.4±3.1mg.l⁻¹; N-NH₄⁺ - 6.6±8.77 mg.l⁻¹; N-NO₃⁻ - 54.1±12.1 mg.l⁻¹; N-NO₂⁻ - 0.138±0.108 mg.l⁻¹; P-PO₄⁻³ – 3.33±1.01 mg.l⁻¹ and P_{total} – 3.58±0.974 mg.l⁻¹. Filtration efficiency was low– 3.5 and 5.1% in P-PO₄⁻³ and P_{total} respectively. Regarding COD_{Mn}, the initial filtration efficiency was negative but the amount of COD_{Mn} gradually decreased. A considerable decline of N-NH₄⁺ amount was recorded since the fourth week. The amount of N –NO₂⁻ raised until the third week (from 0,1 to 0,4 mg.l⁻¹) but filtration efficiency was low to negative. Biological filters started to operate properly six weeks after fish stocking.

Svobodová Z, Máchová J, Poleszczuk G, Hůda J, Hamáčková J, Kroupová H. 2005. Nitrite poisoning of fish in aquaculture facilities with water-recirculating systems. Acta Veterinaria Brno, 74(1):129-137, (IF=0,353)

New techniques of intensive culture in common barbel under controlled conditions

The effects of natural and artificial foods were monitored on the growth parameters and survival rates of larvae, juveniles and young fish in common barbel during 2003 - 2005. In this way, the effect of frozen plankton was studied on the development and maturation of gonads and gametes as a supplementary food. Main objective was to determine a technique for intensive rearing of barbel as well gonadal development in captive condition. The obtained results reveal successful gonadal development of barbel in captive condition. The reared barbels produced good quality of gametes, both sperm and eggs. Nevertheless, further studies are in going steps to investigate endocrine profile of hormones regulation gametogenesis in term of optimizing artificial reproduction in captive condition. This can be used to create sufficient stock of barbel with good quality and sufficient quantity of gametes for off-season spawning and subsequently, to release produced fingerlings to natural water bodies in term of biological conservation in Czech Republic.

Vorlíčková P., Polícar T., Hamáčková J., Kozák P., 2006: Vliv rozdílné potravy používané při odchovu palem obecných v kontrolovaných podmínkách na vývoj jejich gonád. Bulletin VÚRH Vodňany, 42(1): 25-32.

First domestication process of perch under controlled conditions in the Czech Republic

First adaptation process of perch originating from pond into controlled and intensive conditions of Czech aquaculture started in the autumn 2004. Successfully performed artificial reproduction of perch was the first step of its adaptation. After perch reproduction, the larvae were stocked into ponds. Larvae were reared under intensive and controlled pond conditions till the age two months. In this point (body weight and total length about 500 mg and 30 mm, respectively), perch were transferred into controlled recirculation system of fish farming facilities of RIFCH Vodňany and the adaptation of perch was commenced. During the period of adaptation, perch were trained to ingest artificial feed according by special semiwet artificial food. After this adaptation period when all perch individuals accepted semiwet artificial feed, their diet was shifted to dry artificial feed Biomar Ecolife 15 us in intensive culture of salmonids. After these steps of adaptation, very high losses of adapted perch were found. However, sufficient amounts of young perch adapted upon intensive and controlled conditions were obtained. These young perch will be reared till their mature under controlled conditions. After maturation, perch broodstock will be spawned and next generation of perch will be obtained. We suppose that new generations of perch obtained from controlled conditions will prove faster growth and higher survival rates for rearing under intensive and controlled conditions of aquaculture.

Stejskal V., Kouřil J. 2006: Potravní adaptace plůdku okouna na podmínky intenzivního chovu. Bulletin VÚRH Vodňany, 42(1): 18-24.

Production of autumn zander fingerlings, *Sander lucioperca* (L.) in pond culture

Advanced fry harvested from pond culture on June 23, were used in all experiments to produce autumn pikeperch fingerlings by fish farmers by two frequently applied methods (treatments 1 and 2). To test an effectiveness of early piscivory in 0 age pikeperch fingerlings, a small multiple spawner – topmouth gudgeon, *Pseudorasbora parva* (Temminck & Schlegel) was used as a prey fish. Therefore, two ponds (1)

were stocked with prey fish (in biomass of 100 kg.ha⁻¹) already in May 19 before the experiments began to support the development of sufficient available forage basis. We hypothesized, that zooplanktivorous cyprinid might cause earlier switch of zander fry to piscivory due to its availability as a prey itself and because of its size selective feeding causing a significant reduction of zooplankton simulating a timing of piscivorous stage. In contrast, the other ponds (2) were prepared following a traditional way to support the development of large zooplankton community as a food source. Finally, prey fish were regularly monthly stocked with one month delay (2). Significant differences in body lengths between 0+ pikeperch cultured in different treatments and over time were found. 0+ pikeperch grew faster in a treatment 1 for the whole period of observation and the most distinct differences between compared treatments were recorded since the first sample in 16 July. Other relevant characteristics such as piscivory, cannibalism rates and diet analyses were performed throughout the whole experimental period.

Musil, J., Peterka J., 2005: *Potrava 0+ okouna a candáta – Některé aspekty přechodu od planktivorie k piscivorii. Bulletin VÚRH Vodňany. 41(3): 99-106.*

Musil, J., 2006: *Metody odchovu násadového materiálu candáta obecného (Sander lucioperca L.) v rybníčních podmínkách České republiky – krátký souhrn. Bulletin VÚRH Vodňany. 42(1): 38-44.*

Musil, J., Kouřil, J., 2006: *Řízená reprodukce candáta obecného a odchov jeho plůdku v rybnících. Edice metodik VÚRH JU Vodňany 76: 1-14.*

Production of autumn perch fingerlings, Perca fluviatilis L., in a pond monoculture

Free swimming larvae originating from semi-artificial spawning still with rest of oil globule (14-17 days post hatch) were stocked into ponds on April 27 in afternoon hours in density of 120 000 individuals per hectare. Four ponds of different zooplankton quality, but similar in other aspects were selected for perch larvae rearing. Two of them proved higher share of rotifers, whilst in the two remaining, nauplii and copepodite stages of copepods and/or copepodite stages of copepods and cladocerans prevailed. Therefore, the first goal of this study was (1) to evaluate the importance of different zooplankton species (rotifers vs. nauplii and copepodite stages of copepods, and cladocerans) in the diet of perch larvae during the first days of its exogenous feeding to evaluate any exceptional food demands. Our results suggest that specific preparation of nursing ponds towards high abundance of rotifers and on the other hand low abundance of crustaceoplankton is not necessary in perch rearing, but rather contra productive in some respect. The second goal of the study was to test an effectiveness of potential piscivory in 0 age perch (2). In June 17, a total biomass of 40 kg per ha of cyprinid multiple spawners (*Pseudorasbora parva*) was introduced just into two of experimental ponds. Perch stock in these two different treatments was monitored monthly afterwards and a subsample of perch and its prey (in one treatment) was taken for stomach content analysis. Significant differences caused by perch piscivory were observed with consequences in size-structured dynamics, rate of cannibalism significantly stimulated by piscivory and thus mortality rate and higher growth rate of piscivores.

Adámek Z.; Musil J.; Sukop I. 2004. *Diet composition and selectivity in 0+ perch (Perca fluviatilis L.) and its competition with adult fish and carp (Cyprinus carpio L.) stock in pond culture. Agriculturae Conspectus Scientificus. 69(1):21-27*

Food selectivity in carp-pond commercial predatory fish species

The experiments aimed at prey selectivity in three important carp-pond predatory fish species – pike (*Esox lucius*), zander (*Stizostedion lucioperca*) and perch (*Perca fluviatilis*) were performed under experimental laboratory conditions. Prey fish (39-53, 24-37 and 37-51 % TL of pike, zander and perch respectively) were submitted to predators originating from a pond culture. No particular selectivity was registered in pike (139 ± 3 mm TL) which slightly preferred asp (*Aspius aspius*) fingerlings over chub (*Leuciscus cephalus*) and topmouth gudgeon (*Pseudorasbora parva*). The specific growth rate (SGR) and food conversion ratio (FCR) in pike fed live food were 1.09 ± 0.25 %·day⁻¹ and 6.93 ± 1.07 g·g⁻¹, respectively. Only chub and roach (*Rutilus rutilus*) fingerlings were preferred by zander (244 ± 13 mm TL) while other prey fish like perch, topmouth gudgeon and asp were avoided. The SGR and FCR values in zander fed live fish under experimental conditions were 1.67 ± 0.14 %·day⁻¹ and 3.41 ± 0.49 g·g⁻¹, respectively. Perch (148 ± 2 mm TL) preferred only topmouth gudgeon and avoided asp and chub fingerlings. Thus, the SGR and FCR values for perch were assessed as 1.07 ± 0.13 %·day⁻¹ and 6.55 ± 0.35 g·g⁻¹, respectively.

Adámek Z., Opačák A., 2005: *Prey selectivity in pike (Esox lucius), zander (Sander lucioperca) and perch (Perca fluviatilis) under experimental conditions. Biologia, Bratislava, 60(5): 567-570.*

Fish losses caused by cormorant (Phalacrocorax carbo sinensis) predation

Cormorant predation on fishponds is a serious problem requiring special attention. Recently, fishpond stocks suffer not only from direct predation but also from injuries and stress caused by hunting cormorant flocks. Cormorant grasp prey fish by sharp tip of the beak with the tendency to hit the proximal body area behind gill cover (*operculum*). They emerge with the prey on water surface before swallowing it head forwards. Fish, which escaped from cormorant's grasp and/or which could not be swallowed due to their big size, suffer from various deep injuries resulting in consecutive infections and subsequent mortality. The computer assisted image analysis was applied to describe the time dependent course of injury infection and progress. Two-year-old mirror carp ($\cong 250$ mm TL, 200 g W) which escaped from cormorant's beak grasp had injuries ranging over approximately 10% of the total body surface. Immediately after wounding, the damaged epithelium makes about 8 %, and deeper under-epithelial wounds caused by beak tip represent the area of 1- 2 % of the total body surface. With the time progress, these ratios change – deeper necroses represent up to 10 % body surface and healing epithelium makes just 1 – 2 %. Major parts of wounded fish die due to these injuries, particularly if the course of healing is complicated by saprolegniosis. Two- to three-year-old silver carp, *Hypophthalmichthys molitrix*, and bighead carp, *Aristichthys nobilis* ($\cong 350$ mm TL, 600 g W) are particularly susceptible to this disease already after a relatively small injury due to cormorant attacks despite the deeper sub-epithelial wounds are not so spacious in these species. The total proportion of damaged silver carp body surface is similar to mirror carp amounting to approximately 10 % but the share of sub-epithelial wounds usually does not exceed 0.5 % due to their compact scaly cover.

Adámek Z., Kortan J., Flajšhans M. 2005. Computer-assisted image analysis in evaluation of fish injuries caused by cormorant (*Phalacrocorax carbo sinensis*) attacks. *New Challenges in Pond Aquaculture*. 1:23

Some biological aspects of invasive crayfish

Some biological characteristics of *Orconectes limosus* were determined in known localities of occurrence in the Czech Republic. In this study, some reproductive characteristics were considered including mating time, time of extrusion of eggs, fecundity, time of eggs development, post-embryonic development of juveniles. Higher reproductive capacity and shorter embryonic development of *O. limosus* were confirmed in comparison to native crayfish. Diurnal activity and reaction of spiny-cheek crayfish on presence of fish predators were studied using audio-visual technique. Higher day activity of *O. limosus* was observed in comparison to noble crayfish *Astacus astacus*. The growth and survival of both species were compared under laboratory conditions. The faster growth was observed in *O. limosus* during the first growing season. The screening of *O. limosus* populations focused on prevalence of crayfish plague *Aphanomyces astaci* was provided in cooperation with Charles University in Prague. High prevalence of fungus *A. astaci* in all tested populations was confirmed.

Kozák, P., Polícar, T., Ďuriš, Z., 2004: Migration ability of *Orconectes limosus* through a fishpass and notes on its occurrence in the Czech Republic. *Bull. Fr. Peche Piscic.*, 372 – 373: 367 – 373. (IF=0,780)

Kozák, P., Máčková, J., Polícar, T., 2005: The effect of chloride content in water on the toxicity of sodium nitrite for spiny-cheek crayfish (*Orconectes limosus* Raf.). *Bull. Fr. Peche Piscic.* 376-377:705-714. (IF 2004=0,780)

Monitoring, rearing and reintroductions of the noble crayfish Astacus astacus

Hatching and rearing of noble crayfish juveniles were provided under controlled conditions for the stocking purposes. Big attention was focused on optimization of rearing conditions during the mating, eggs extrusion, hatching and rearing of juveniles. Monitoring of crayfish occurrence, their healthy conditions and quality of environment continued during the years 2004-5. Monitoring was focused mainly to region of the Šumava National park, Třeboňsko Protected Landscape area and the region of Písek. Confirmations of previous reintroductions and hatchery reared crayfish to new localities were done. Altogether, 2500 noble crayfish were stocked into new localities during the years 2005-6.

Polícar, T., Simon, V., Kozák, P., 2004: Egg incubation in the noble crayfish (*Astacus astacus* L.): The effect of controlled laboratory and outdoor ambient condition on hatching success, growth and survival rate of juveniles. *Bull. Fr. Peche Piscic.*, 372 – 373: 411 - 423. (IF=0,780)

Polícar, T., Kozák, 2005: Comparison of trap and baited stick catch efficiency for noble crayfish (*Astacus astacus* L.) in course of the growing season. *Bull. Fr. Peche Piscic.*, 376-377: 675-686. (IF=0,780)

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RESEARCH PRIORITIES

The department performs both the fundamental and applied research and university education in genetics and reproductive physiology of freshwater fish, conservation of fish genetic resources and enhancement of genetic potential of commercially important species, as e.g. common carp, tench, wels and sturgeons. Research combines laboratory analyses and experiments with pilot testing in experimental ponds or indoor/outdoor tanks, and/or with regular performance testing in ponds on productive fish farms. Many results of our studies have wide implications for applied aquaculture. There are seven main programmes of research for the department:

- Genetic, biological, physiological and reproductive aspects of polyploid and unisexual fish populations.
- Cytogenetic, haematologic and reproductive aspects of hybrid diploid-polyploid complexes of selected fish genera (*Cobitis*, *Carassius*).
- Selective breeding programmes based upon determination of heritability of performance traits.

- Endocrine effects on gametogenesis and gamete quality for controlled reproduction.
- Male competition during the fertilization process and its effects on population genetic variability.
- Short-term preservation and cryopreservation of fish spermatozoa and embryos.
- Sperm ultrastructure and characteristics of the fertilization process.



Department of Genetics and Fish Breeding –from the left standing: Otomar Linhart, Martin Flajšhans, Martin Kahanec, Marie Pečená, Zdeněk Elšnic, Ivana Samková, David Gela, Martin Kocour, Sayyed Mohammad Hadi Alavi, Martin Pšenička; in the front line: Filip Raab, Martin Hulák, Marek Rodina

Selected Research Results

Analysis of higher ploidy levels of fish

Routine diagnostics of ploidy level in our studies of spontaneous- and induced polyploid fish employed image cytometry and flow cytometry cell nuclei, as well as number of erythrocytes (RBC) and mean cell volume (MCV). Image cytometry of erythrocyte nuclei of evolutionarily diploid, spontaneous and induced triploid and tetraploid fish (*Lepisosteus osseus*, *Tinca tinca*, *Silurus glanis* and *Carassius auratus*) showed nuclear area from blood smears to be the best ploidy level predictor increasing in a linear relationship with the $2n - 3n - 4n$ level determined by karyotyping and/or flow cytometry. Similarly, RBC dropped and MCV increased with ploidy level. Recent studies of evolutionarily tetraploid sturgeons (*Acipenser ruthenus*, *A. stellatus*, *Huso huso*), penta-, hexa- and heptaploid sturgeon hybrids, evolutionarily oktaploid sturgeons (*A. baerii* and *A. gueldenstaedtii*), nona- and dodekaploid sturgeon hybrids showed that erythrocyte nuclear area still increased with $4n - 5n - 6n - 7n - 8n - 9n - 12n$ level but since $5n$ this relationship appeared to be nonlinear. Similar nonlinear relationships were found for dropping RBC and increasing MCV of the respective polyploids.

For ploidy levels higher than $5n$, neither the erythrocyte nuclear area from blood smears, nor the other haematological indices used were reliable ploidy level predictors. Comparative determination of ploidy level by FCM was thus recommended. However, karyotyping still remains the most precise reference method.

Ráb, P., Flajšhans, M., Ludwig, A., Lieckfeldt, D., Ene, A.-C. Rábová, M., Piačková, V., Paaver, T., 2004. The second highest chromosome count among vertebrates is associated with extreme ploidy diversity in hybrid sturgeons. Cytogenetic and Genome Research, 106,1: 16 (abs.) (IF=2,076)

Genome manipulations in common carp for production of all-female populations

The department of genetics and breeding works on genome manipulations in common carp with aim to determine the optimal methodology for induction of gynogenesis and to find a possible utilization of genome manipulated fish for fish production. One possibility how to utilise the gynogenic population of common carp is its reversal to neomales (masculinized females), which produce sperm but only with X chromosome. When crossing neomales with regular females, we receive all-female population, because common carp has the *Drosophila* system of sex chromosomes. It is known that females of common carp grow faster than males but nobody compared the all-female and mixed-sex populations in a performance test

from the beginning of larval rearing to market size in conditions of the Central Europe. Such trial was conducted at our institute in two different populations, namely, Northern mirror carp (M72) and scaly crossbred between M72 and scaly Ropsha carp (Rop). Results shows 6-8 % increase in production of common carp flesh, which can be attained by using all-female populations under the same pond culture condition. The marketing of all-female populations should be the most profitable in three-year-old fish (1.5-1.8 kg), because higher percentage of edible parts is mostly affected by a lower ratio of sexually mature fish (lower GSI) in all-female populations. However, the economical benefit from rearing of all-female carp populations will be decreased by higher expenses to establish, rear and renew neomale broodstock.

Kocour, M., Linhart, O., Gela, D., Rodina, M., 2005. Growth performance of all-female and mixed-sex common carp *Cyprinus carpio* L. populations in the central Europe climatic conditions. *Journal of the World Aquaculture Society* 36 (1): 103-113. (IF = 0,560)

Detailed analysis of spermatozoa in some models of teleostean and chondrosteian fish species with the determination of competition among males by means of colour and cryopreservation of spermatozoa

Research was focused to morphology and structure of spermatozoa of some models of teleostean and chondrosteian fish species and to factors which activating and inhibiting motility of intact and demembrated spermatozoa. Motility was evaluated from the point of view of the percentage of motile and velocity of spermatozoa. Spermatozoa competition and their qualitative/quantitative characteristics were studied at the level of paternal representation in progeny, using colour markers. Theoretical knowledge on spermatozoa motility and structure was applied in cryoconservation of spermatozoa of chondrosteian and teleost fishes.

In competition experiment, the percentage of sperm motility (92-100 %), spermatozoa velocity (112-163 $\mu\text{m}\cdot\text{s}^{-1}$) and control hatching rates (83-96 %) were evaluated for each of 6 gold and 5 green male common carp (*Cyprinus carpio*). In all 30 possible paired combinations of sperm competition tests, hatching rates of 90-97 % were achieved. The mean percentage of offspring sired was strongly influenced by the male used ($P < 0.001$, $R^2 = 0.91$). The best male sired an average of 88% of the offspring in its competition tests, while the worst male sired only 5%. Spermatozoa quality parameters could explain only part of the variation in male competitive ability. The male effects alone explained 91.4% of the observed variance, consisting of 17.1% explained by spermatozoa motilities and 32.5% by control hatching rates in single fertilizations. Undetermined male effects explained 41.8%. The velocity of spermatozoa had no effect on the outcome of sperm competition. Neither was there any link between spermatozoa velocity and hatching rate in a control hatching test, whereas there was an effect of motility on hatching rate in this same test.

Method of cryopreservation was elaborated for *ex situ* conservation of European catfish. The success of sperm cryopreservation was evaluated by post-thaw sperm motility and velocity, percentage of live spermatozoa and fertility (hatching rates) using frozen/thawed sperm. The best hatching rates of 82-86 % were obtained with sperm stored for 5 h before freezing in immobilizing solution and frozen with Me_2SO in concentrations of 8, 10 and 12 %, or with a mixture of 5 % Me_2SO and 5 % propandiole. These results did not significantly differ from the fresh sperm control sample. The percentage of live spermatozoa in frozen/thawed sperm did not correlate with hatching rate or motility of spermatozoa, but was negatively correlated with velocity of spermatozoa ($r = -0.47$, $P = 0.05$). The percentage of frozen/thawed sperm motility ranged from 8 to 62 %, when sperm was stored in immobilizing solution 5 h before freezing. The average value in the fresh sperm (control) was 96 %. The frozen/thawed sperm motility rate significantly correlated with the hatching rate ($r = 0.76$, $P = 0.0002$), but not with the percentage of live spermatozoa ($r = 0.16$, $P = 0.52$) or the sperm velocity ($r = 0.07$, $P = 0.79$). The velocity of frozen/thawed spermatozoa ranged from 37 to 85 $\mu\text{m}\cdot\text{s}^{-1}$, whereby methanol concentrations of 7.5 and 10% resulted in highest velocities. Freezing sperm volumes of 1 – 4 ml did not affect the quality of frozen/thawed sperm.

Linhart, O., Rodina, M., Gela, D., Kocour, M. and Vandeputte, M., 2005. Spermatozoal competition in common carp (*Cyprinus carpio*): What is the primary determinant of competition success. *Reproduction*, 130, 705-711 (IF=3,136).

Linhart, O., Rodina, M., Flajshans, M., Gela, D. and Kocour, M., 2005. Cryopreservation of European catfish *Silurus glanis* sperm: Sperm motility, viability and hatching success of embryos. *Cryobiology*, 51, 250-261 (IF=1,765)

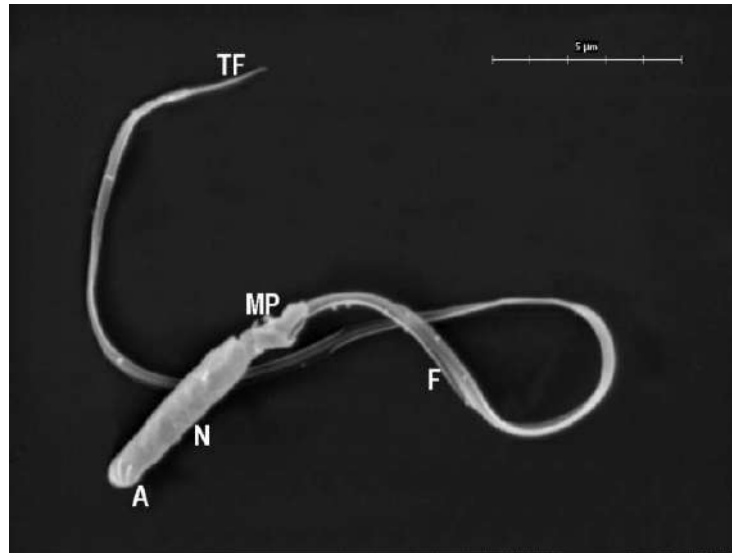
Ultrastructure of fish sperm

Ultrastructure of sperm was studied in tench *Tinca tinca* (a model of teleostean fish) and Siberian sturgeon *Acipenser baerii* (a model of chondrosteian fish) by means of scanning electron microscopy (SEM) and transmission electron microscopy (TEM).

The structure of tench sperm was primitive and similar to sperm of other teleosts. It had the following structure: no acrosome; a spherical or slightly elliptic nucleus located excentrically to the flagellum with electrolucent vesicle; two differently oriented centrioles under 140° angle; a short undifferentiated midpiece pervaded by the cytoplasmic channel and one flagellum inserted in an invagination

of the implantation fossa by means of the centriolar complex. This sperm is probably the smallest spermatozoon described among cyprinid fishes.

The sperm of Siberian sturgeon had a long head with a distinct acrosome terminated with 10 posterolateral projections, a midpiece and a single flagellum with 9+2 structure of microtubules and two lateral extensions. Three intertwining endonuclear canals bounded by membranes traversed the nucleus from the acrosomal end to the basal nuclear fossa region. The midpiece contained a centriolar complex, three to six mitochondria and two lipid droplets. Both centrioles were formed of nine peripheral triplets of microtubules.



Siberian sturgeon sperm cell structures using scanning electron microscopy. Acrosome (A), nucleus (N), midpiece (MP), flagellum (F) and terminal region of flagellum (TF) (scale bar = 5 μ m).

Assessment of gametogenesis and gamete quality during controlled reproduction of fish, including the strategy of fertilization.

In tench and European catfish, methods of gamete collection was optimized, as well as their short-term preservation under aerobic conditions with immobilization or incubation of gametes in immobilizing or incubating media with specific ionic composition and osmotic level. Spermatozoa: egg ratio was optimized for the process of artificial insemination. To check the compatibility of gametes, acrosome integrity and quality of spermatozoa in the course of fertilization, monoclonal antibodies to surface- and intra-acrosomal proteins of mammalian spermatozoa was used, as well as newly synthesized monoclonal antibodies specific to fish sperm proteins.

In this study, KUROKURA solution and its modifications by increasing NaCl content to 160, 180 and 200 mM were tested as immobilizing solutions for sampling and short-term preservation of potential motility of tench spermatozoa. The immobilizing solution is used because high possibility of contamination of sperm by urine during stripping, causing poor quality of spermatozoa with low motilities and velocities (almost 0) and subsequently resulting in low fertilization and hatching rates. Sperm was sampled into a syringe containing an immobilizing solution (IS) with IS: sperm ratio at 2: 1 under aerobic conditions and at 0 - 4°C. Sperm was stored in such for 10 h. and untreated sperm was collected prior to fertilization as a control. Spermatozoa quality was evaluated by determination of percentage of sperm motility, sperm velocity and fertilizing ability. Hatching rate was also measured as an indicator of sperm quality.

Obtained results reveal significant role of IS to achieve high fertilization success in artificial insemination. Significantly, higher percentage of sperm motility, sperm velocity, fertilizing ability and subsequently hatching rate were observed when stripped sperm collected in the various immobilizing solutions and used for artificial insemination after 10 h at 0 - 4 °C. Nevertheless, the best immobilizing solution was found to be KUROKURA 180 (180 mM NaCl, 2.68 mM KCl, 1.36 mM CaCl₂ . 2H₂O and 2.38 mM NaHCO₃), in which fertility and hatching rates of 41 % were observed 10 h post storage of sperm. In control condition (sperm collected without immobilizing solution) very low fertilization and hatching rates (6-7 %) were achieved.

The process of fertilization is accompanied by contact between the spermatozoon and the egg according to their affinity. Study of monoclonal antibodies contributes to understanding the gamete compatibility was developed. In mammals, monoclonal antibodies to acrosin and intracrosomal proteins are known. These are used for testing the status of acrosome in spermatozoa before- and after induction of acrosomal reaction in porcine, bull and canine spermatozoa. Monoclonal antibodies were used as tools for testing of the sperm quality, because of the status of acrosome and intra-acrosomal proteins that are

important for the linkage of spermatozoon to the egg. The first antibodies against chondrosteian and teleostean fish sperm were prepared and their physiological function was studied (Koubek et al.,2004).

Koubek P., Elzeinova F., Linhart O., Peknicova J., 2004. Study of fish sperm proteins using monoclonal antibodies. *American Journal of Reproductive Immunology*, 51, 483 abs. (IF= 1,416)

Rodina M., Cosson J., Gela D., Linhart O., 2004. Kurokura solution as immobilizing medium for spermatozoa of tench (*Tinca tinca* L.) *Aquaculture International*, 12, 119-131 (IF=0,568).

APPLICATIONS OF RESEARCH RESULTS

Conservation of fish genetic resources.

This conservation programme is targeted to rare breeds and populations of common carp, tench, rainbow trout, brown trout and European catfish, and to pure species of coregonids and sturgeons, altogether kept at in 45 shoals. Subvention by the Ministry of Agriculture of the Czech Republic Programme 6, Part B.1.16 Fish is annually administered to 13 fish farms. The programme is managed by our department in cooperation with the Research Institute of Animal Breeding in Prague-Uhřetěves, with the Institute of Animal Physiology and Genetics, Czech Academy of Sciences at Liběchov, and with the Fish Farmers' Association of the Czech Republic in České Budějovice. Our department is also responsible for cryoconservation of sperm of these breeds and for maintenance of our own live fish gene bank.

Selective breeding of fish, performance and heredity testing

Being specified in the Animal Breeding Act No. 154/2000 of the Code of Laws of the Czech Republic, performance and heredity tests of fish are uniformly designed, involve replicates for statistic assessment and employ mathematic models to compute corrected weight in order to eliminate the environmental components of genetic variation. During the 2004 – 2005 period, our department conducted four (2004) and two (2005) performance tests with common carp crossbreds, tench purebreds and/or chromosomally manipulated populations in own and contracted testing facilities. Apart from this, the department staff also conducted methodical control and assessment of performance tests carried out in testing facilities of other fish farms. Annual subsidy for performance and heredity testing of fish is provided by the Ministry of Agriculture of the Czech Republic Programme 2, Part 2.A.e.1. to authorized persons.

Kocour, M., Gela, D., Rodina, M., Linhart, O., 2005. Testing performance in common carp *Cyprinus carpio* L. under pond husbandery conditions I: top-crossing with Northern mirror carp. *Aquaculture Research* 36: 1207-1215. (IF = 0,746)

Optimization of artificial propagation in European catfish

We focused on technical aspects of immobilizing solution, activation solution, process of insemination, activation of gametes and elimination of eggs stickiness with a practical solution for artificial propagation in hatcheries. Spermiation was stimulated and could be sustained over a period of 1 month after weekly carp pituitary (CP) injection of 5 mg.kg⁻¹ b.w. Males produced significantly the largest quantity of sperm, 0.12-0.13. 10⁹ spermatozoa. kg⁻¹ b.w., after 3rd and 4th injections. The best immobilization solution for sperm was determined as solution containing 170 - 200 mM NaCl, 30 mM Tris-HCl, pH 7 and activation solution for activation of gametes containing 17 mM NaCl, 5 mM Tris-HCl, pH 7. The ratio of 1:2 rather than ratio of 1:1 in the control batch (volume of activation solution in ml: grams of eggs) increased the hatchability up to 68 %. The highest hatching success 82 - 88% from four individual females were found for sperm number of 800 – 80,000 spermatozoa per egg, in comparison to 26% hatching at 80 spermatozoa per egg. The best time for application of alcalase enzyme for elimination of egg stickiness was three minutes after gamete activation at concentration 20 ml of enzyme per liter with exposure of 2 minutes.

Linhart O., Rodina M., Gela D., Kocour M., 2004. Optimalization of artificial propagation in European catfish, *Silurus glanis* L. *Aquaculture*, 235, 619-632 (IF= 1,374)

INTERNATIONAL COOPERATION

Assessment of Spermatozoa Viability

The viability of spermatozoa has been assessed using SYBR 14 staining for DNA of living cells and propidium iodide staining for DNA of degenerate cells. This dual staining was performed on four fish species (Siberian sturgeon, *Acipenser baerii*; common carp, *Cyprinus carpio*; tench, *Tinca tinca* and wels, *Silurus glanis*) and the proportions of live and dead spermatozoa were assessed by epifluorescence microscopy and image cytometry. Ten phase contrast and epifluorescent images were recorded per sample, corresponding images were overlaid, and the blended images were evaluated for live and dead spermatozoa, represented by green and red fluorescence signals. Live/dead proportions were assessed, after dual thresholding, by imaging software that counted absolute numbers of objects and computed their

frequencies. All sperm heads were found to be labelled, emitting either green or red light. Standard deviations did not exceed 5% of the means. The image cytometric system using dual staining with SYBR 14 and propidium iodide was clearly suitable for assessing the viability of freshwater fish spermatozoa.

Flajšhans, M., Cosson, J., Rodina, M., Linhart, O., 2004. The application of image cytometry for viability assessment of dual fluorescent - stained spermatozoa of fish. Cell Biol. Int. 28, 12: 955 – 959. (IF=1,194)

Application of selection programme in common carp

On the base of mutual cooperation with laboratory of genetics of fish in INRA, France and their large experience with selection programmes in trouts and marine fish species, we have been testing the possibility to apply modified selection programme PROSPER for increasing of growth in common carp. Despite twice higher common carp annual production compared to all salmonids, no systematic mass selection breeding programmes in common carp are used. The situation is given due to results from past conducted in Israel, where no positive selection response was found after five generation selection programme. However, the way of carp rearing in ponds and former impossibility to use molecular genetic methods for pedigree construction could have caused environmental bias and did not allow to use high number of breeders. Using microsatellite markers for parentage assignment in a Hungarian synthetic mirror strain established at our department we estimated heritability for growth-related traits in 8 week-old progeny in range 0,3-0,4 when 10 females and 24 males were used for establishing of the experimental population. The results promise applying 2 % selection press the gain on the level of 20 % in weight for one generation. However, only running experimental selection programme will show the suitability of mass selection on growth for practical usage.

*Vandeputte M., Kocour M., Mauger S., Dupont-Nivet M., De Guerry D., Rodina M., Gela D., Vallod D., Chevassus B. and Linhart O., 2004. Heritability estimates for growth-related traits using microsatellite parentage assignment in juvenile common carp (*Cyprinus carpio* L.). *Aquaculture* 235: 223-236. (IF = 1,374)*

Securing juvenile production of Eurasian perch by improving reproduction and larval rearing

According the EU CRAFT project COOP-CT-2004-512629 the USB RIFCH and our department was involved in R&D project from Octobre 2004 as research partner to secure the production of Eurasian perch (*Perca fluviatilis*) juveniles (3-5 g) in order to sustain the development of European SMEs (6) which have funded in this new way of diversification (production of 10-15 g fillets for consumption markets corresponding to 100-150 g fish). Presently, the juveniles availability of perch is very low and limited to the annual cycle of reproductive period which occurs in early spring. Moreover, the quality of supplied juveniles is very variable. Thus, to support the sustainable development of Eurasian perch production, a R&D project is proposed to help the setting of a hatchery – nursery activity that other SMEs wish to develop. In order to respond to the demand, a first objective of this R&D project was focused on the optimisation of the broodstock management considering environmental, nutritional and population variables (i) to obtain delayed (extension of the natural reproductive period) and out-of-season spawning, (ii) to reduce breeders mortality during the spawning period and (iii) to improve the control of gametes and larval quality. At this level, a specific action was conducted on the cryopreservation of Eurasian perch semen for artificial reproduction. This objective required multifactorial and multidisciplinary studies. A second objective consisted of developing protocols for the production of juveniles populations with improved potentialities by allfemales production. Finally, an economical study was realised to define the cost of production of juveniles using different rearing systems (semi-intensive in ponds and intensive in tanks) and considering different socio-economic contexts.

Application of a temperature-dependent mitotic interval tau(0) for induction of diploid meiotic gynogenetic paddlefish.

We tested the application of mitotic interval (tau 0) unit in comparison with absolute time to help standardize preshock timing for a consistent production of diploid meiotic gynogenetic paddlefish *Polyodon spathula*. The diploid gynogenetic larvae were produced by applying heat shock (35 degrees C; 2 min) at different times after activation of paddlefish eggs with irradiated sperm of shovelnose sturgeon *Scaphirhynchus platyrhynchus* at two different preshock temperatures (15 degrees C and 20 degrees C). When the timing of heat shock (minutes after activation) was expressed in absolute time, the yield of gynogenetic diploid larvae was distinctly different between the two preshock incubation temperatures. At 20 degrees C, the highest yield was observed when the timing of the heat shock was 12-13 min after activation, whereas at 15 degrees C the highest yield was observed when the timing of the heat shock was 17-22 min after activation. However, when the timing of heat shock was expressed in terms of tau(0) the yield of

gynogenetic diploid larvae for the two preshock incubation temperatures coincided completely at each time point tested. The highest yield (about 30%) of diploid gynogenetic larvae was obtained when the timing of heat shock was between 0.22 tau(o) and 0.26 tau(o). Reporting of heat shock timing in terms of tau(o) provides the opportunity to standardize the preshock time interval for any preshock temperature. This could help to identify the optimal timing of heat shock for a specific experiment and allow comparison of data among different experiments in which different preshock temperatures were used.

Mims, S.D., Shelton, W.L., Linhart, O., Wang, C.Z., Gomelsky, B., Onders, R.J., 2005. Application of a temperature-dependent mitotic interval (tau(o) for induction of diploid meiotic gynogenetic paddlefish. North American Journal of Aquaculture, 67: 340-343 (IF= 0,570)

Intensive and sustainable culture of freshwater species tench

Many regions of Europe have a continental aquatic medium, very appropriated for the culture of autochthones cyprinid fishes, as tench. This fish is appreciated in Europe as a refined dish in the restaurant, a piece of angling and also as ornamental fish. Tench has been cultivated extensively (by natural reproduction system) in the ponds of Central and Eastern Europe for more than 500 years. However, extensive production cannot avoid large losses of fry during the first year, slow growth and long generation interval. Intensive tench culture (inducing the reproduction during the whole year, and optimising both survival and growth of juveniles) is largely dependent on reproduction issues (seasonality of spawning, asynchronous spawning), feeding affairs, and environmental factors (illumination, temperature, welfare). In order to create an optimum procedure for intensive production of tench, 4 RTD partners including USB RIFCH and 8 SMEs from Europe joined for optimalization 1) the method to assure a good rate of continuous fry production (reproduction and nursery aspects), 2) the food composition more adequate according tench requirements, 3) a protocol to guarantee the survival and adequate growth rate of juveniles, and 4) the study of the better conditions to commercialise them. The implantation of an intensive culture of tench responds to the need of promoting and diversifying the freshwater aquaculture, increasing the sustainable production of a wide range of species. Moreover, this will contribute to the creation of stable employment in rural depressed regions

Cryopreservation of embryos

The department started to be focused on embryo cryopreservation during 2005, according to bilateral project with Institute of Hydrobiology, Chinese Academy of Sciences. Systematic germ plasm cryopreservation can have a profound impact on aquaculture by 1) reducing inbreeding; 2) reducing pressure on wild populations; 3) sustainable productivity by minimizing the impact of live-culture failures resulting from human error, natural disasters, breeding failure and epidemics, etc.; 4) conservation benefit to endangered species; etc. To successfully cryopreserve embryo of cyprinids as our model species, osmotically active water must exit the cells and an appropriate cryoprotectant must enter the cells. The most basic functions of cryoprotectant were tested in order not be toxic to the embryo, help to stabilize the membranes and to reduce the formation of lethal intracellular ice crystal. We believe that there are many new approaches that will facilitate the understanding and successful cryopreservation of fish embryos.



Blood sampling of tench, Wuhan, China

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RESEARCH PRIORITIES

The department performs fundamental and applied research and participate in academic program in courses of fish diseases and aquatic toxicology at University of South Bohemia České Budějovice. Aquatic Toxicology and Fish Diseases Department usually focuses on laboratory researches in parallel with aquacultural and environmental studies. There are six main research programmes in which staffs are studying on:

- Surveillance of xenobiotics effects which loading the aquatic environments on fish.
- Assessment of contamination of fish and other components of aquatic ecosystems with xenobiotics.
- Toxicity assessment of substances and wastes for aquatic organisms.
- Improvement of prevention and elimination of health risks in fish culture – Principles of pharmacovigilancy and implications for aquaculture in Czech Republic (harmonisation with EU): Protection of common carp (*Cyprinus carpio* L.) from KHV disease.
- Assessment of technological properties of fish flesh, fish processing, organoleptic assessment, and determination of polyunsaturated fatty acids (PUFA, EPA, DHA), amino acids and fat deposition, and effects of fish farming technology to fish flesh quality.
- Management of brown trout and grayling culture, interaction between wild and reared fish.



Department of Aquatic Toxicology and Fish Diseases – from the left standing: Tomáš Randák, Jana Máchová, Anna Kocová, Jindra Čížková, Vladimír Žlábek; in the front line: Veronika Piačková, Jitka Kolářová, Olga Valentová.

SELECTED RESEARCH RESULTS

Pollution of aquatic environment with toxic metals

Research was focused to assess contamination of fish and aquatic environment of polluted surface waters.

The aim of the study (Svobodová et al., 2004) was to assess metal contamination in the reaches of the Tichá Orlice river, and thus to help explain unsatisfactory results in the reproduction of salmonid fish. The contamination assessment was based on measuring metal concentrations in the brown trout (*Salmo trutta morpha fario*) and bottom sediment samples. The samples were collected at two collection sites from the Tichá Orlice River and its tributary Kralický Brook in June 2000 and 2001. The AAS method was used to determine the total mercury, arsenic, cadmium, lead, copper, zinc, chromium and nickel contents in the muscle tissue of the fish and in aquatic sediments. The Kralický Brook is an important anthropogenic source of metal contamination (particularly of mercury and copper) for the Tichá Orlice River. The highest concentrations of mercury in muscles of brown trout were found at the Kralický Brook and Lichkov and the lowest concentration at Červená Voda in 2000 and 2001. Significantly higher concentrations of arsenic were found in the muscle tissue of the brown trout collected at Červená Voda than at the downstream site Lichkov. The authors hypothesize that the difference was due to different environmental conditions (principally water temperature). It seems reasonable to assume that unsatisfactory results in the reproduction of fish from the upper reaches of the Tichá Orlice River are due not only to organic pollutants but also to mercury compounds that are classified among suspect endocrine disrupters.

The aim of the following study (Čelechovská et al., 2005) was to evaluate arsenic content in tissues (muscle, liver, kidneys, gonads) of fish caught in selected localities along the River Elbe and its tributary the Vltava in 1999, 2002 and 2003, and in pools of the River Elbe in 2000. A total of 118 bream and 151 perch were examined by AAS. The highest muscle tissue arsenic concentrations were found in Zelčín in bream ($0.17 \pm 0.08 \text{ mg.kg}^{-1}$) in 2003 and in perch ($0.084 \pm 0.012 \text{ mg.kg}^{-1}$) in 2002. The highest arsenic concentrations in perch were found in the kidneys, followed by the liver, milt, muscle and eggs. A significant increase ($P < 0.01$) was found in case of arsenic concentrations in 2003 (after floods) compared with 2002 in bream muscle tissues in Zelčín and in perch muscle tissues in Podolí. A decrease in arsenic concentrations was demonstrated in perch musculature in Verdek, Němčice, Hřensko ($P < 0.01$) and in bream musculature in Hřensko ($P < 0.05$). Arsenic contamination in the localities under study does not pose a major environmental or health risk.

Svobodová Z., Čelechovská O., Kolářová J., Randák T., Žlábek V. 2004: Assessment of metal contamination in the upper reaches of the Tichá Orlice River. Czech J. Anim. Sci., 49 (10):485-464 (IF 0,254)

Čelechovská, O., Svobodová, Z., Randák, T., 2005. Arsenic content in tissues of fish from the River Elbe. *Acta Vet. Brno*, 74 (3): 419-425. (IF= 0,353)

Hygienic quality of marketable common carp from selected ponds of the Czech Republic

The aim of the study was to evaluate the extent of contamination of six selected ponds in South and West Bohemia by PCBs. Seven indicator congeners were determined in the tissues of common carp and bottom sediments. The ponds were sampled during autumn 1999-2001. Five to seven specimens of three- to four-years-old carp were sampled from every pond. The results demonstrated comparable indicative ability of fish tissues and bottom sediments for evaluation of pond load by the monitored PCBs. Considering concentrations of PCBs in fish and bottom sediments, the most contaminated pond was Tovaryš followed by Buzický, Dřemliny, Regent, Bezdrév and Horusický. As regards the distribution of PCBs in fish, the highest values of indicator congeners were found in liver followed by ovaries, muscle and testes. A significant correlation was found between the PCB levels and the content of fat in individual tissues of carp. The pattern of individual indicator congeners in fish was almost the same as that in bottom sediments. This observation is corresponding with slow biotransformation rate of PCBs in fish. The study contributed to the assessment of quality of carp (which is the main representative of marketable fish in the Czech Republic) in terms of its safety as food as well as to evaluation of the pollution of respective aquatic environment.

Svobodová Z., Žlábek V., Randák T., Máchová J., Kolářová J., Hajšlová J., Suchan P., Dušek L., Jarkovský J. 2004: Profiles of PCBs in tissues of marketable common carp and in bottom sediments of selected ponds in South and West Bohemia, *Acta Vet. Brno*, 73:133-142. (IF= 0,353)

Mercury pollution of the aquatic environment

The main aim of the studies was to assess the mercury contamination of the Elbe River (Žlábek et al., 2005) and Skalka reservoir (Maršálek et al., 2005) in Czech Republic.

The samples were collected at 6, 2 and 1 collection sites from the Elbe River, the Vltava River and the Blanice River in 1999, 2002 and 2003 respectively. Samples of bream (*Abramis brama* L.), perch (*Perca fluviatilis* L.) and chub (*Leuciscus cephalus* L.) muscle were collected at the monitored sites. The single-purpose mercury analyzer AMA 254 was used to determine the total mercury content in the muscle tissue of the fish. In total 290 fish were sampled. The highest values of total mercury content were found in fish from the localities Obříství and Lysá nad Labem. Decreases of total mercury contamination have occurred in the localities Děčín and Hřensko in period 1999-2003. Findings in 2003, indicated the catastrophic floods in 2002 left so far no significant effect on mercury contamination in fish. The highest value of total mercury content (2.56 mg.kg⁻¹) in the fresh muscle of chub was in the Obříství vicinity. This presents high-risk for consumers, considering average consumption of 10 kg fish per capita and year.

The aim of other study was to investigate the total mercury and methylmercury contamination of fish from the Skalka reservoir. The reservoir had been polluted with mercury containing sewage water for several decades. In total, 30 fish was sampled in August 2003. The fish caught included roach (*Rutilus rutilus*), bream (*Abramis brama*), silver bream (*Blicca bjoerkna*), big head carp (*Aristichthys nobilis*), asp (*Aspius aspius*), eel (*Anguilla anguilla*) and wels (*Silurus glanis*). Muscle tissue, the liver and the gonads were used to determine mercury. Total mercury in muscle tissue of the asp (as the representative of predatory species) and the bream (as the representative of non-predatory species) averaged 3.11 ± 0.20 mg.kg⁻¹ and 0.96 ± 0.22 mg.kg⁻¹, respectively. The results showed that mercury contamination of the Skalka reservoir continues to be very high, that consumption of predatory fish in particular poses a major health risk and that methylmercury was the predominant contaminant in fish tissues.

Žlábek V., Svobodová Z., Randák T., Valentová O. 2005: Content of mercury in muscle of fish from the Elbe River and its tributary. *Czech J. Anim. Sci.*, 50 (11): 528-534. (IF= 0,254)

Maršálek, P., Svobodová, Z., Randák, T., Švehla, J., 2005. Mercury and methylmercury contamination of fish from the Skalka reservoir: A case study. *Acta Vet. Brno*, 74 (3): 427-434. (IF= 0,353)

Influence of aquatic contamination on fish organism

This study deal with determining the primary cause of reproductive disturbances in salmonids from the Tichá Orlice river. Three sites at the Tichá Orlice river were monitored during 2000-2003. Samples of fish muscle, bile and bottom sediment were collected on monitored sites for chemical analyses of PCB, HCB, DDT and its metabolites, HCH, OCS, 1-OHPY (bile) and PAH (bottom sediment) concentrations. Vitellogenin (Vtg) concentration in brown trout (*Salmo trutta fario*) blood plasma was assessed using ELISA (Rainbow trout vitellogenin EIA kit). In comparison with the control site, locality at the source of pollution and on another locality downstream were found considerably polluted. This contamination was

manifested also by repetitious findings of high Vtg concentrations in blood plasma, resulting in adverse effects on fish reproduction.

Kolářová J., Svobodová Z., Žlábek V., Randák T., Hajšlová J., Suchan P. 2005: Organochlorine and PAHs in brown trout (*Salmo trutta fario*) population from Tichá Orlice River due to chemical plant with possible effects to vitellogenin expression. *Fresenius Environmental Bulletin*, 14:1091-1096. (IF= 0,509)

Contamination assessment by means of multiple biomarker detection

The Elbe River is the most polluted aquatic ecosystem in territory of Czech Republic. The most important contaminants are heavy metals and persistent organic compounds. A whole series of contaminants may affect negatively the endocrine systems of aquatic animals. The chub (*Leuciscus cephalus* L.) was selected as a bioindicator for the field study. Blood plasma and tissue samples were collected from wild chub (*Leuciscus cephalus* L.) from 9 localities of the Elbe and Vltava rivers and from 1 locality of the Blanice river (control locality) in 2003. Biochemical markers were used to evaluate the influence of aquatic pollution on fish population. Fish muscle samples were analysed for toxic metals (Hg, Cd, Pb, As), persistent organochlorine pollutants (PCB, DDT, HCH, HCB, OCS) and alkylphenols (Randák et al., 2006; Široká et al., 2005).

In 2004, the effect of three main chemical plants (Synthesia Pardubice, Spolana Neratovice and Spolchemie Ústí nad Labem) on chub (*Leuciscus cephalus* L.) was studied along the Elbe River. Sampling sites were chosen upstream and downstream the respectively mentioned sources of pollution respectively. Control site was chosen upstream the Lipno reservoir located on the Vltava River. Samples of blood plasma were analyzed for the yolk protein precursor vitellogenin (VTG) and 11 – ketotestosterone (11-KT). Gonadosomatic index (GSI) was also measured. Liver samples were analysed for cytochrome P450 (CYP1A), EROD activity, glutathione (GSH) and glutathione S-transferase (GST). Bile samples were analysed for 1-hydroxypyrene (1-OHPY). Fish muscle samples were analysed for mercury (Hg), persistent organochlorine pollutants (PCB, DDT, HCH, HCB, OCS) and alkylphenols (4-terc-nonylphenols, 4-terc-oktylphenol). Results of all analysed biochemical markers except GST were sex related and significant for males. Therefore, further evaluation was concerned males. Results of health status evaluation, analyses of VTG, 11-KT and GSI parameters proved higher load of endocrine disrupters in localities downstream from chemical plants compared to localities upstream sources of pollution. Chemical monitoring clearly showed higher contamination of localities downstream the sources of pollution in comparison with upstream located localities. Biomarkers studied and chemical analyses proved significant negative influence of chemical plants on load of pollutants in the Elbe River. Contamination of monitored sites presents significant risk to aquatic organisms (Randák et al., 2005).

Randák T., Žlábek V., Kolářová J., Svobodová Z., Hajšlová J., Široká Z., Jánková M., Pulkrabová J., Čajka T., Jarkovský J. 2006: Biomarkers Detected in Chub (*Leuciscus cephalus* L.) to Evaluate Contamination of the Elbe and Vltava Rivers, Czech Republic. *Bull. Environ. Contam. Toxicol.* 76:233-241. (IF= 0,626)

Široká, Z., Krijt, J., Randák, T., Svobodová, Z., Pešková, G., Fuksa, J., Hajšlová, J., Jarkovský, J., Jánková, M., 2005. Organic Pollutant Contamination of the River Elbe as Assessed by Biochemical Markers. *Acta Vet. Brno*, 74: 293 – 303. (IF= 0,353)

Randák, T., Žlábek, V., Kolářová, J., Široká, Z., Svobodová, Z., Pulkrabová, J., Tomaniová, M., 2005. Influence of the main pollution sources of Czech part of the Elbe River on the fish. *Book of Abstracts, 12th International Conference Diseases of Fish and Shellfish 2005, Copenhagen, Denmark*, 46.

Nitrite toxicity for aquatic animals

Study of the effect of nitrite to aquatic organisms is of great importance, since elevated concentrations of nitrite which can be found in the surface waters cause great problems in intensive culture of commercial fish species and ornamental fish. The aquatic animals are at higher risk of nitrite poisoning than terrestrial animals as nitrite in the ambient water can be actively taken up across the gill epithelium and can accumulate to very high concentrations in the body fluids. One critical consequence of nitrite accumulation is the oxidation of haemoglobin to methaemoglobin, compromising blood oxygen transport. Furthermore, nitrite is a disrupter of multiple physiological functions including ion regulatory, respiratory, cardiovascular, endocrine and excretory processes (Kroupová et al., 2005).

Nitrite toxicity for fish is known to depend greatly on chloride concentration in ambient water. The protective effect of chloride on the health of fish was assessed in 2-years-old common carp (*Cyprinus carpio* L.) exposed to elevated nitrite concentration. Four groups of carp were exposed to different concentrations of nitrite and chloride for 96 hours (group E1: 67 mg.l⁻¹ NO₂⁻, 11 mg.l⁻¹ Cl⁻; group E2: 67mg.l⁻¹ NO₂⁻, 100 mg.l⁻¹ Cl⁻; group E3: traces mg.l⁻¹ NO₂⁻, 100 mg.l⁻¹ Cl⁻ and group C: traces mg.l⁻¹ NO₂⁻, 11mg.l⁻¹ Cl⁻). The main haematological response of carp to an acute exposure to nitrite (group E1) was a significant decrease in haemoglobin concentrations, haematocrit, erythrocyte count, leucocyte count and lymphocyte count, and a

significant increase in methaemoglobin concentration and mean corpuscular haemoglobin concentration. At higher chloride concentrations (group E2), a lower nitrite toxicity was observed. Erythrocytes in carp exposed to nitrite showed qualitative changes. Compared with the control group C, group E1 showed a significant higher number of elongated erythrocytes, with the nucleus located at one cell pole. All erythrocytes of group E1 had remarkably clear cytoplasm compared with the cytoplasm in the control group. The biochemical values found were comparable with those found in controls. The main histological lesions were found in the gills of carp exposed to nitrite and consisted of hyperplasia and an elevated number of chloride cells (Svobodová et al., 2005b).

Nitrite toxicity to spiny-cheek crayfish (*Orconectes limosus*) was also assessed in relation to the concentration of chlorides diluted in water. Results of the tests confirmed a favourable effect of chloride content in water on the tolerance of spiny – cheek crayfish to nitrites. A significant linear correlation was observed, $y = 0.23 x + 7$ $R=0.995$, where x and y are chloride concentration in water and 96hLC50, respectively (Kozák et al., 2005).

Furthermore, three cases of severely compromised fish health and death in newly commissioned aquaculture facilities with water-recirculating systems were described. The observed damage and death was due to increased concentrations of water-borne nitrites and the subsequent methaemoglobinemia. The increased NO_2^- concentrations in water were caused by impaired functionality of biological filters in the second stage of nitrification, i.e. the conversion of NO_2^- to NO_3^- . Chloride concentrations in water were considered the main factor influencing NO_2^- toxicity in all of the cases described. In the case of death of catfish and tench, the Cl^- to N-NO_2^- weight ratios were in the range of 13–28 and 11-19, respectively. In case of tilapia health impairment without symptoms of toxicity, the ratios were between 50 and 150. In the water tank inflow, the Cl^- to N-NO_2^- weight ratios were between 2000 at 10000. Blood methaemoglobin levels of catfish and tench (severe symptoms of poisoning) and of tilapia (no signs of impairment, only brownish discolouration of gills) were over 80% and 21%, respectively). In order to minimise risks in culture of fish in a water-recirculating systems, it is necessary to choose a proper stock of fish and a proper feeding ratio, not to treat the fish with antibiotics in the form of baths, to check meticulously the quality of water. In case of increasing concentration of nitrites, to administer sodium chloride to get the chloride concentration increased at least to 100 mg.l^{-1} Cl^- . Better operation of a biological filter can be speeded up by inoculation with activated sludge (Svobodová et al., 2005a).

Kroupová, H., Máchová, J., Svobodová, Z., 2005. Nitrite influence on fish – a review. *Vet. Med. – Czech*, 50, 2005 (11): 461–471. (IF = 0,621).

Svobodová, Z., Máchová, J., Drastichová, J., Groch, L., Lusková, V., Poleszczuk, G., Velíšek, J., Kroupová, H., 2005b. Haematological and biochemical profile of carp blood following nitrite exposure at different concentration of chloride. *Aquac. Res.*, 36 (12), 1177-1184. (IF= 0,746).

Kozák, P., Máchová, J., Polícar, T., 2005: The effect of chloride content in water on the toxicity of sodium nitrite for spiny-cheek crayfish (*Orconectes limosus* Raf.) *B. Fr. Peche Piscic.*, 376-377: 705-713. (IF= 0,780).

Svobodová, Z., Máchová, J., Poleszczuk, G., Hůda, J., Hamáčková, J., Kroupová, H., 2005a. Nitrite poisoning of fish in aquaculture facilities with water-recirculating systems. *Acta Vet. Brno*, 74: 129 – 137 (IF= 0,353).

Koi Herpesvirus (KHV)

A disease called koi carp causing serious losses in culture of common carp (*Cyprinus carpio* L.) and namely of its ornamental form has spread nearly all over the world since 1998. Koi herpesvirus (KHV) was designated as its agens. The outbreak of the disease is always accompanied with high mortality of fish (80-90%) and water temperature ranging from 18 to 28°C is a predisposition factor for emergence of the disease. The KHV is clinically manifested by fish disorientation, irregular swimming and respiration, and by mass kill. Large necrosis of gill lamellae is the most important pathological alteration. The virus can be cultivated on KF-1 cell line, creating a cytopathic effect (CPE). The approach of polymerase chain reaction (PCR) appears to be the best method for virus detection in tissues. World research workplaces are looking for possibilities of carp culture protection. A protocol of natural fish immunisation has been elaborated, application of attenuated vaccine has been tested, and resistant carp strains are being bred.

The Department of Aquatic Toxicology and Fish Diseases, the Research Institute of Veterinary Medicine in Brno and Pharmaceutical University in Brno co-operate on a grant project supported by Ministry of agriculture of Czech Republic “Protection of common carp culture from disease caused by KHV”. The project is focused on the implementation of PCR diagnostic of KHVD (Koi HerpesVirus Disease) into the praxis of National reference laboratory for fish diseases and application of this diagnostic in field monitoring of infection situation into common carp- and koi culture in Czech Republic. Within the scope of this project, the literature review was compiled which has summarised published available information about KHV (Pokorová et al., 2005). More than hundred specimens of fish were investigated during the year 2005. All results of investigation were negative.

Anaesthetics

Assessment of the effect of anaesthetics (2-phenoxyethanol and clove oil) was performed on the three important fish species including common carp (*Cyprinus carpio* L.), trout (*Oncorhynchus mykiss*) and wels (*Silurus glanis*). The effect was assessed on the basis of acute toxicity tests and effects of anaesthetics on haematological, biochemical blood plasma profile and histological examination of tissues.

The fish were divided into four groups for haematological and biochemical examinations of blood and histological examinations of tissues. The groups were as follows: Control I (before the anaesthetic administration), Experiment I (immediately after 10 min anaesthesia), Experiment II (24 hrs after 10 min anaesthesia) and Control II (controls examined in parallel with Experimental II). The indices used to evaluate the haematological profile included the erythrocyte count (Er), haemoglobin concentration (Hb), haematocrit (PCV), mean erythrocyte volume (MCV), mean corpuscular haemoglobin concentration (MCHC), mean corpuscular haemoglobin content (MCH), leukocyte count (Leuko) and the differential leukocyte count (Leukogram). Biochemical indices determined in blood plasma included glucose (GLU), total protein (TP), albumins (ALB), total globulins (GLOB), ammonia (NH₃), triacylglycerols (TRIG), aspartate aminotransferase (AST), alanin aminotransferase (ALT), lactate dehydrogenase (LDH), creatinkinase (CK), calcium (Ca²⁺) and inorganic phosphate (PHOS). Samples of gills, skin, liver, cranial and caudal kidney and spleen were histologically examined.

Results of the examinations suggest that the use of anaesthetics 2-phenoxyethanol (0.30 ml.l⁻¹) and clove oil (30 mg.l⁻¹) are safe in the recommended concentration for common carp (*Cyprinus carpio* L.), rainbow trout (*Oncorhynchus mykiss*), and wels (*Silurus glanis*).

Velíšek J., Svobodová Z. (2004a): Anaesthesia of Common Carp (*Cyprinus carpio* L.) with 2-phenoxyethanol: Acute Toxicity and Effects on Biochemical Blood Plasma. *Acta Vet. Brno*, 74: 247-252. (IF= 0,353)

Velíšek J., Svobodová Z. (2004b): Anaesthesia of Rainbow Trout (*Oncorhynchus mykiss*) with 2-phenoxyethanol: Acute Toxicity and Biochemical Blood Profile. *Acta Vet. Brno*, 74: 379-384. (IF= 0,353)

Velíšek J., Svobodová Z., Piačková V. (2005a): Effects of Clove Oil Anaesthesia on Rainbow Trout (*Oncorhynchus mykiss*). *Acta Vet. Brno*, 74 (2): 139-146. (IF=0,353)

Velisek J., Svobodova Z., Piackova V., Groch L., Nepejchalova L. (2005b): Effects of clove oil anaesthesia on common carp (*Cyprinus carpio* L.). *Vet Med-Czech*, 50 (6): 269-275. (IF= 0,621)

Biocide Diazinon 60 EC in fish farming

Approximately 80 % fish ponds are strongly in range of eutrophic and hypertrophic in Czech Republic. Fish-farming in such highly eutrophised ponds has its indisputable importance from the environmental point of view (nutrients which would serve no useful purpose, or, rather, be considered undesirable, are purposefully made use of in this arrangement). In spring months, the ponds often suffer from excessive production of phytoplankton, marked increases in pH, oxygen regimen fluctuations and excessive propagation of coarse zooplankton. Subsequently, fish may be damaged because of present unsuitable environment, and, as a rule, fish predation is unable to curb zooplankton propagation effectively. The existing oxygen deficit is then further aggravated by excessively rapidly propagating zooplankton, which may result in the death of large amounts of fish. There are approaches that will minimise such risks, although to completely prevent such high-risk situations from occurring is impossible. In such high-risk situations, Soldep (active substance trichlorfon), which was able to selectively eliminate daphnian zooplankton, caused no damage to fish and then decomposed in the aquatic environment, has been used until recently. However, the active substance of Soldep, i.e. trichlorfon, was put on the list of prohibited substances, and the use of Soldep in the Czech Republic was banned in 2002. There was need to replace Soldep preparation, which was used to control coarse zooplankton in eutrophic ponds.

We looked for a preparation with properties as follows: high and specific toxicity for daphnian zooplankton, low toxicity for fish and other aquatic organisms, easy and rapid biological degradability, minimal accumulation in fish and other components of ecosystem. Twelve different preparations have been tested in laboratory conditions and Diazinon 60 EC was found to be the best of them.

Diazinon 60 EC (chemical insecticide, organophosphate, active substance diazinon at the concentration of 600 g.l⁻¹) is used in fish farming in well-founded cases as a biocide to suppress excessive propagation of coarse daphnian zooplankton (used concentration of Diazinon 60 EC 0.010 mg.l⁻¹). Diazinon 60 EC is highly toxic for daphnian zooplankton (48hEC₅₀ = 0.00029 mg.l⁻¹ by the using of artificially prepared adulterating water according to the standard and 48hEC₅₀ = 0.0029 mg.l⁻¹ by the using of pond water). Toxicity of Diazinon 60 EC is markedly lower for other water organisms (96hLC₅₀ for *Poecilia reticulata*, *Cyprinus carpio* and *Oncorhynchus mykiss* are in the range 3 - 10 mg.l⁻¹ and 72hIC₅₀ for *Desmodesmus subspicatus* = 10 mg.l⁻¹). At the concentration of 0.01 mg.l⁻¹ (i.e. 100 g per 1 ha at 1 m mean

depth of the pond), Diazinon 60 EC can highly selectively eliminate daphnian zooplankton, causes no harm to fish and is relatively quickly decomposed in the aquatic environment. As demonstrated by embryonic and larval tests of toxicity for the common carp, Diazinon 60 EC at this concentration causes no harm to the tested individuals even in these sensitive (critical) stages of ontogeny.

Svobodová, Z., Máchová, J., Kroupová, H., Smutná, M., 2005: Ammonia autointoxication of common carp: three case studies. . In: Adámek, Z. (Ed.). New Challenges in Pond aquaculture. Book of Abstracts., 19.

Processing technology and hygienic quality of fish flesh

The aim of the research work was to evaluate the effects of transglutaminase (0.5 – 1.0 – 1.5%) combined with salt (0 – 1.0 – 2.0%) on biogenic amines (BA) formation in processed flesh of common carp (*Cyprinus carpio*) and the water holding capacity (WHC). Total content of PUT (putrescin) + CAD (kadaserin) in samples with additives combined with NaCl did not exceed 20 mg/kg. In samples without NaCl the critical contents (20 mg/kg) was reached within 8-9 days. Rapid elevation of PUT+CAD continued and the contents of 45 mg/kg were exceeded 10-11 days accompanied by bad organoleptic properties.

The other part of research was focussed on supplemental cereal feeding (maize, wheat and triticale) compared to control group with natural food and its effect on fatty acids (FA) expression in the flesh during long-lasting storage of common carp (*Cyprinus carpio*). The content of fatty acid was investigated in the flesh of carp during 8 months long-lasting storage without an additional feeding. The weight of the fish (marketable fish) ranged from 1358 to 2221 gr. Fatty acid (PUFA, n-3) content and composition in fish flesh were determined by gas chromatography (VARIAN 3300). Supplemental cereals caused lower level of PUFA and n-3 PUFA in fish fat. The content of these fatty acids did not decrease even during eight months of fish storage. The average percentage of PUFA in total fat from edible parts was as follows: 13.7% ± 1.58 for maize, 11.6% ± 1.17 for wheat and 10.7% ± 1.00 for triticale. The percentage of n-3 PUFA for maize was 2.5% ± 0.36, for wheat 3.38% ± 0.44 and for triticale 3.1% ± 0.39.

Křížek, M., Vácha, F.: Biogenic amines in fish mince with additives. In: Health implications of dietary amines. Coimbra (Portugal), University of Coimbra, 2005, s.22.

Křížek, M., Vácha, F.: Content of biogenic amines in fish roe. In: Health implication of dietary amines. Vilnius, Food Institute Kaunas, University of Technology, (Lithuania), 2005, ISBN 9955-675-00-4, s.33.

Management of salmonids culture

A program of rearing brown trout (*Salmo trutta m. fario* L.) spawners was established at the Husinec hatchery local unit belonging to the Czech Anglers' Union in 1999 in order to enhance the production of material for restocking the open waters of the southwest of Czech Republic. Part of progeny derived from artificial propagation (1999) of wild brood-stock of original brown trout population of Blanice River has been reared up to the brood stock stage (2002). The three-, four- and five-years-old spawners were artificially reproduced in 2002, 2003 and 2004 and some reproductive indicators (weight of eggs, total fecundity, relative fecundity, egg diameter, egg weight, sperm concentration) were measured and compared to those of the original wild population. In order to assess and compare the biological quality of eggs from farmed and wild females, fertilization rate, duration of incubation period, egg losses during incubation and mortality of starving hatched fry were assessed. The results showed that farming conditions did not significantly affect the reproductive parameters studied in the first generation of farmed brood-stock. The same biological quality of eggs were observed in farmed and wild females. Establishing a parallel broodstock under controlled conditions, the production of eggs of local origin population of brown trout was greatly enhanced, creating an important prerequisite for enhancing the production of stocking material for open water restocking in the region.

INTERNATIONAL CO-OPERATION

The department co-operated with following foreign institutions:

Faculty of Environmental Science and Fisheries, University of Warmia and Mazury Olsztyn, Poland (T. Wlasow and P. Gomulka) – Anaesthetics.

Szczecin University, Szczecin, Poland (G. Poleszczuk) – Nitrite toxicity for fish.

APPLICATION OF RESEARCH RESULTS AND OTHER PRIORITIES

- Monitoring of surface water pollution.
- Eco-toxicological assessment of preparations, substances, and wastes in toxicological laboratory accredited by the Czech Institute of Accreditation registered under No. 1052 (Head – Dipl.-Ing. J.

Máchová, Quality manager – Dipl.-Ing. O. Valentová). Acute toxicity test are carried out on fish (guppy *Poecilia reticulata*, zebra fish *Brachydanio rerio*, common carp *Cyprinus carpio*, and rainbow trout *Oncorhynchus mykiss*), green chlorococcal alga *Scenedesmus subspicatus*, aquatic arthropod *Daphnia magna* and plant *Sinapis alba*.

- Chronic toxicity test on fish – common carp (*Cyprinus carpio*) and rainbow trout (*Oncorhynchus mykiss*).
- Basic chemical analysis of drinking-, surface-, waste water and determination of total mercury in biological materials in Central Hydro-chemical Laboratory. Production and service of field kits for quick basic chemical analysis of water (Head – Dipl.-Ing. O. Valentová).
- Parasitological, pathological, biochemical and haematological examination of fish (V. Piačková, DVM; J. Kolářová, DVM and Prof. Z. Svobodová, DVM).
- Management of Blanice Vodňanská 4 B special trout angling ground.
- Teaching of Eco-toxicology at Higher Professional School of Water Management and Ecology by Fishery High School in Vodňany.



The examination of fish health



View to toxicological laboratory

INTERNATIONAL ACTIVITIES

Cooperation

International co-operation runs on several levels:

- Cooperation in scope of mutual exchange of publications, experience, results and short-term stays during solution of close research subjects. This co-operation is based on the signed contracts between USB RIFCH and the concrete foreign institution.
- Bilateral cooperation in scope of countries which signed contracts on the governmental level regarding the mutual co-operation in research and development. These programmes are available through Ministry of Education, Youth and Sport of the Czech republic (see capture “Research projects”)
- Direct cooperation of different partners from Europe focused on research and development in scope of programmes of European union (see capture “Research projects”)

International scientific meetings organised by the USB RIFCH

- New Challenges in Pond Aquaculture, České Budějovice, Czech Republic, April 26-28, 2005

Over the years, the international conferences and workshops convened by the Research Institute of Fish Culture and Hydrobiology Vodňany (University of South Bohemia České Budějovice) on various aspects of fish culture with particular attention to pond farming technologies, have become an established tradition in South Bohemia – the region of thousands of ancient ponds. The workshop on “New Challenges in Pond Aquaculture” held in České Budějovice – the University of South Bohemia Campus – consisted of following sessions:

- Biological Background of Pond Aquaculture
- Interactions with Protected Plant and Animal Species
- Multipurpose Pond Usage
- Pond Management Strategies
- Pond Aquaculture Economics, Marketing, Networking and Product Development

The sessions were selected with respect to fish production under conditions of sustainable development of the pond environment. These issues cover a wide range of approaches associated with pond production, such as integrated farming systems, organic farming, nature friendly control mechanisms of pond environment, animal welfare in reproduction, pond harvesting, fish transportation etc.

Thus, the workshop addressed the wide ranging concept of pond aquaculture, although particular importance was given to less intensive and environmentally friendly approaches. Semi-intensive and extensive fish farming technologies in earthen ponds, usually exceeding several hectares in area, are the major types of fish production in the Czech Republic and in some regions of Central and Eastern Europe. Recently, the pond aquaculture sector has been facing increasing demands for implementation of non-production pond usage, notably landscape formation and protection, recreation, water protection and pond management issues.

An open debate on recent advances in different technologies and their impact upon pond-fish quality and environmental consequences were useful tool for the pond aquaculture sector to increase knowledge, exchange ideas and draw conclusions and recommendations on the challenges required for the present and future of pond aquaculture.

The conference was a joint event co-organised by the Research Institute of Fish Culture and Hydrobiology (University of South Bohemia) and the Aqua-Innovation Network as a part of the EC supported project “Creating Supporting. The conference was attended by 76 participants from 16 countries. The participants enjoyed also the social programme that included

- Technical meeting of partners of EU project COOP-CT-2004-512629-PERCATECH: Securing juvenile production of Eurasian perch by improving reproduction and larval rearing, Vodňany and Nové Hradky, Czech Republic, August 17-18, 2005
- 7th Czech Ichthyological Conference, Vodňany, Czech Republic, May 6-8, 2004

Participation on international conferences, workshops and symposiums

2004

- 4th International Workshop on Biology and Culture of the Tench, Wierzba, Poland, 20-23 September
- 3rd Thematic meeting of Craynet, Innsbruck, Austria, 8-11 September
- 15th Symposium of International association of astacology, London, UK
- PROFET workshop “Inland Fisheries and Aquaculture”, 20-21 February, Budapest, Hungary
- III. Conference „Karpíowate ryby reofilne“, Wars 30.06.-02.07.2004 Warsaw, Poland, 30 June-2 July, SGGW,
- 10th International Congress of Toxicology, Tampere, Finland, 11-15 July
- 11th Magdeburg seminary on waters in central Europe: Assessment, Protection, Management, Leipzig, Germany, 18-22 October.
- 39th Croatian Symposium on Agriculture, Opatija, Croatia, 17-20 February.
- FARMER EXPO, International Conference on Aquaculture and Fisheries, Debrecen, Hungary, 18 August
- Biotechnologies for Quality, EAS, Barcelona, Spain, 20-23 October.

2005

- New challenges in Pond Aquaculture.“ Book of Abstracts (ed. Z. Adámek), České Budějovice, April 26-28 2005, 45.
- Sixth International Crustacean Congress. Glasgow, Scotland, UK, July 18-22
- Craynet-Final Conference, Firenze, Italy, May 2-5, 2005.
- New Aspects in Aquaculture, Plovdiv, Bulgaria, 10-12 October
- 12th International Conference “Diseases of Fish and Shellfish”, Copenhagen, Denmark, 11-16 September
- 5th International Symposium on Sturgeons, Teheran, Iran, 9-13 May
- Aquaculture America 2005, New Orleans, Louisiana, USA, 17-20 January
- River bottom, Brno, Czech Republic, 19-24 September
- Workshop on Recirculation Aquaculture Technology, Trondheim, Norway, 9-10 August

Memberships in international organisations

Adámek Z.	European Aquaculture Society, member of board
Flajšhans M.	Network of Tropical Aquaculture Scientists
Kozák P.	International Association of Astacology
Svobodová Z.	The European Association of Fish Pathologists OECD – Ecotoxicology

Memberships in editorial boards

Adámek Z.	Aquaculture International Ribarstvo Zagreb Agriculae Conspectus Scientificus Zagreb
Kouřil J.	Czech Journal of Animal Science
Svobodová Z.	Acta Veterinaria
Linhart O.	Bulletin VÚRH Vodňany
Kozák P.	Bulletin VÚRH Vodňany
Randák T.	Bulletin VÚRH Vodňany
Vykusová B.	Bulletin VÚRH Vodňany

EDUCATION AND TEACHING ACTIVITIES

Since 2004, doctoral fishery study was accredited at USB in České Budějovice, whereas its pursuance was vice-chancellor charged Research Institute of Fish Culture and Hydrobiology with three research departments including Aquaculture and Hydrobiology, Fish Genetics and Breeding and Aquatic Toxicology and Fish Diseases (Ichthyopathology) in collaboration with Department of Fishery, faculty of Agriculture, USB.

Purposes of established doctoral study in fishery were to train specialist and promote the multidisciplinary study in different fields of fisheries Sciences. All students are assumed to obtain basic and practical knowledge in different region of fishery and especially in terms of their Ph.D. thesis. The student must defend the dissertation in the Dissertation Defence oral examination after completing the research and writing a dissertation. In addition, all students in the Ph.D. Program must take the Ph.D. courses defined by his/her supervisor. This program defined as a graduate program with duration of 3 years. Approximately 10 students are currently enrolled in the Ph.D. program.

Our current Ph.D. student and their specialization are as follow as:

Name	Entry date	Field of study	Supervisor
Dipl. ing. Martin Hulák	2004	sex in common carp	Assoc. Prof., Dipl. Ing. Otomar Linhart, DSc
Dipl. Ing. Hanka Kroupová		influence nitrite on fish	prof. MVDr. Svobodová Zdeňka, DSc.
MSc Sayyed Mohammad Hadi Alavi	2005	physiology of gametes and reproduction fish	Assoc. Prof., Dipl. Ing. Otomar Linhart, DSc
Dipl. ing. Jan Kohout		genetic monitoring and repatriation of common salmon	Assoc. Prof., Dipl. Ing. Petr Ráb, CSc.
Mgr. Eva Nyklová		hydrobiology	Assoc. Prof., RNDr. Zdeněk Adámek, CSc.
Dipl. Ing. Tomáš Pávek		intensive breeding of trout	Dipl. Ing. Jan Kouřil, PhD.
Dipl. ing. Martin Pšenička		morphology gametes and insemination	Assoc. Prof., Dipl. Ing. Otomar Linhart, DSc
Dipl. ing Vlastimil Stejskal		biology and breeding of perch and recirculation systems	Dipl. Ing. Jan Kouřil, PhD.
MVDr. Eva Sudová		problems of health in fish population and farmakovigilanc	prof. MVDr. Svobodová Zdeňka, DSc
Mgr Petera Vorlíčková		reproduction of fish	Assoc. Prof., Dipl. ing. Otomar Linhart, DSc

The Research Institute of Fish Culture and Hydrobiology lays great emphasis on education and teaching activities. In 2004-2005 years, we gave lectures at 5 universities and supervised 32 undergraduates and 20 postgraduates from 3 universities. Another important fact is that 7 and 2 students supervised by the staff succeeded in obtaining their MSc and PhD degrees, respectively. We have accreditation from the Ministry of Education, Youth and Sports to perform post-gradual studies in Fisheries (from year 2004).

1. Teaching at universities

<i>Lecturer</i>	<i>Subjects</i>	<i>2004 hour</i>	<i>2005 hour</i>	<i>University /Faculty</i>
Z. Adámek	Hydrobiology	56	56	1
	Fisheries	48	-	2
	Ichthyology and fisheries	48	48	3
M. Flajšhans	Genetic and breeding of fish	56	56	1
	Genetic and breeding of fish	4	4	4
J. Kouřil	Aquaculture	28	76	1
	Intensive aquaculture	4	4	4
P. Kozák	Conservation and breeding of freshwater crayfish and moluscs	24	24	1
	Conservation and breeding of freshwater crayfish and moluscs	4	4	4
O. Linhart	Fish reproduction	56	56	1
T. Policar	Intensive culture of ornamental fish	48	48	1

	Biology of ornamental fish	-	42	1
Z. Svobodová	Fish diseases	56	56	1
	Toxicology	48	48	5
	Food toxicology	56	56	5
	Fish biochemistry	2	2	5
	Judicial toxicology	4	4	5
F. Vácha	Fish processing	56	56	1
	Aquaculture commodities	112	112	1
	Gastronomy of aquaculture products	-	56	1
	Conservation and breeding of freshwater crayfish and moluscs	24	24	1
Total 8	18	730	828	5

1 University of South Bohemia, Faculty of Agriculture, České Budějovice

2 Masaryk University, Faculty of Science, Brno

3 Palacky University, Faculty of Sciences, Olomouc

4 Czech Agriculture University, Faculty of Agriculture, Prague

5 University of Veterinary and Farmacy, Brno

2. Undergraduate students working in the Institute and/or supervised by the Institute's fellows in 2004-2005

Supervisor	Student	2004	2005	Defended the theses	University/Faculty
Z. Adámek	K. Bratršovský			2005	1
	P. Kabilka		+		1
	J. Kortan	+	+		1
	K. Kořínek	+	+		1
	J. Ťuk		+		1
	J. Sikora		+		1
	J. Zeman		+		1
M. Flajšhans	J. Kašpar	+	+		1
J. Kouřil	P. Bolha	+	+	2005	1
	L. Mašek	+	+	2005	1
	V. Stejskal	+	+	2005	1
	J. Štícha	+	+	2005	1
	P. Benedikt	+	+		1
	J. Nocar	+	+		1
	J. Turek	+	+		1
	J. Škeřík	+	+		1
	J. Votrubec	+	+		1
	T. Borkovec		+		1
	J. Hájek		+		1
	J. Mráz		+		1
P. Kozák	M. Buřič	+	+		1
	M. Musil	+	+		1
	J. Kanta		+		1
	A. Kouba		+		1
O. Linhart	R. Slabý		+		1
	M. Aldorf		+		1
	V. Kašpar	+	+		1
	R. Vlach	+	+		1
J. Máchová	H. Kroupová	+		2004	2
	P. Dvořáková	+	+		1
	Š. Hrabánková	+	+		1
T. Polícar	V. Simon	+		2004	1
Total 8	32	18	24	7	2

1 University of South Bohemia, Faculty of Agriculture, České Budějovice

2 University of Chemical Technology, Faculty of Environment Protection Technology, Prague

3. PhD students working in the Institute and/or supervised by the Institute's or other fellows in 2004-2005

<i>Supervisor</i>	<i>Student</i>	<i>2004</i>	<i>2005</i>	<i>Defended the theses</i>	<i>University/Faculty</i>
Z. Adámek	I. Kalistová	+		2004	2
	A. Lepičová	+	+		2
	E. Nyklová		+		1
	D. Kortan	+	+		2
	J. Musil	+	+		2
	V. Prášek	+	+		3
	J. Sychra	+	+		3
J. Kouřil	H. Kroupová	+	+		1
	Z. Stupka	+	+		2
	T. Pávek		+		1
	V. Stejskal		+		1
O. Linhart	M. Kocour	+	+		4
	M. Rodina	+	+		1
	H. Alavi		+		1
	P. Vorlíčková		+		1
P. Ráb	J. Kohout		+		1
Z. Svobodová	V. Žlábek	+	+	2005	2
	J. Velíšek	+	+		2
	E. Sudová		+		1
	T. Randák	+	+		1
Total 5	20	13	19	2	4

1 University of South Bohemia, České Budějovice, Research Institute of Fish Culture and Hydrobiology, Vodňany

2 University of South Bohemia, Faculty of Agriculture, České Budějovice

3 Masaryk University, Faculty of Science, Brno

4 University of South Bohemia, Faculty of Biology, České Budějovice

EDITORIAL AND POPULARIZATION ACTIVITIES

In 2004-2005 we published following three titles from the *Edition Metodik (Methodologist)*:

Hartman P., 2004. Ecological friendly way of pond liming. Edice Metodik, VÚRH JU Vodňany, No. 73, 11 s.; Davidov O.N., 2004. Fish and human diseases (helminthosis). Edice Metodik, VÚRH JU Vodňany, No. 74, 23 s. (translate from Russian B. Vykusová); Anonymous, 2004. Rearing of european catfish and tench in ponds. Edice Metodik, VÚRH JU Vodňany, No. 75, 11 s. (translate from Germany O. Lhotský)

Bulletin VÚRH Vodňany - The Bulletin VURH Vodnany is a national publication and published papers dealing with different aspects of Aquaculture, Fish Biology, Aquatic Toxicology and other related topic to Fishery. Since 1965, publication of this bulletin had been started in 4 issues per year. Language of publication is Czech and each paper including an English Abstract. **Dokumentary Bulletin of Library** - All information on library available data in VURH is collected and classified in a guidebook. It make easy to look for exact literature as soon as possible. The information office is thinking of developing system using "Computerized Search System". **Proceedings:** Vykusová B. (ed.), 2004. VII. Czech ichthyological conference. Proceedings, 7th conference with international participation. VÚRH JU Vodňany, pp. 300.

Vykusová B. (ed.), 2005. Toxicity and biodegradability of matters important in water management. Proceedings, 11th conference . VÚRH JU Vodňany, 164 s.

Fisheries Days, Vodňany 2004, 2005

The Fisheries Days was traditionally prepared by RIFCH USB Vodňany, Fishery High School Vodňany and Civil Department of City Vodňany. The Czech Fish Farmer Association supported this regional event. The 14th volume of Fisheries Days holded on 2004 with **FISHFILM** show (the old films about fisheries and institute). Professional program included **7th Czech Ichthyological Conference**. Additional program contained exhibition Fishery status in Vodňany, performance of the band Babouci (with tasting of fish meals), traditional **Fish market and fair**. On the next year (2005), the rich program was open by international conference **New Challenges in Pond Aquaculture** (was hold at the campus of USB České Budějovice). The national conferences **Fish Health Protection** and **Rearing of fingerlings of predatory fish species** happened at Vodňany. The biennial exhibition **FISHTECH** and **Fish market** visited about 1000 visitors.

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