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6–11 August 2017, 8th International Symposium on Monogenea

Organisers

Local Organizing Committee
Chairman: Milan Gelnar
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Members: Andrea Vetešníková Šimková
Nikol Kmentová
Zuzana Kobíková
Maria Lujza Kičinjaová
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Welcome to the ISM8

On behalf of the Organising Committee, we wish to welcome you all, both delegates and accompanying persons, to the 8th International Symposium on Monogenea (ISM8). Without dedicated scientists willing to share their findings, this symposium would not be possible. It is hoped, that ISM8 will provide opportunity for delegates to present and discuss their new ideas, share their findings and to promote studies on fascinating monogenean parasites. Thank you very much for coming and contributing to the Symposium. We hope that you had a pleasant journey and will enjoy the conference, the atmosphere, and the opportunity to revive old friendships and built new links and acquaintances.

Previous ISM symposia have been of high academic standard and we trust that this symposium will be not an exception. We are very pleased to see an increasing number of students and young scientists attending the meeting. Students should grab this opportunity to rub shoulders with established scientists and establish contacts.

Academic program includes 56 oral and 34 poster presentations. Oral presentations have been grouped into broad topics that include monogenean taxonomy and diversity including morphology and ultrastructure, their evolution and phylogenetics, host-parasites interactions, evolutionary ecology, molecular interactions and immunity, for the first time also genomic and transcriptomic approaches and last, but not least monogenea in introduced, imported and commercially important fish.

However, scientific conferences are not only about science. In addition to arranging the academic program, the ISM8 Organising Committee has worked to balance the scientific and social activities. We hope you will enjoy the social events in Brno, the South Moravia Region and in the Czech Republic generally. On Sunday evening, we will kick the Symposium off with an icebreaker – Welcome Party and on Monday morning we will start with Opening Ceremony and Scientific Sessions. To allow everyone to enjoy a breakfast and recover from previous night, we will start the sessions at 9:00 each morning. On Wednesday we will have traditional Mid Symposium Trip, this ISM to Valtice Castle – part of UNESCO World Heritage Site. Here we will have lunch and wine tasting in the wine cellar. On Thursday evening, the atrium of Central European Institute of Technology (CEITEC) – a part of new Masaryk University Campus Bohunice – will be a venue of our Gala dinner and ISM8 closing ceremony.

We are particularly grateful to the generous support provided to ISM8 from sponsors and several learned institutions. Members of the ISM8 Organising Committee have given their time, enthusiasm and energy unselfishly in the preparation of this conference. We thank committee members for their commitment and efforts over very long time. Without their hard work, there would have been no ISM8.

We hope that you will have a great time in Brno and in the Czech Republic.

Iva Přikrylová and Milan Gelnar
Welcome to the Czech Republic

It is our great honour and pleasure to welcome you to the 8th International Symposium on Monogenea. The ISM is returning to the Czech Republic and to Brno after exactly 20 years (3rd International Symposium on Monogenea, 1997, Brno) and to the Czech Republic again after 30 years (1st International Symposium on Monogenea, 1988, České Budějovice) when the tradition of this scientific meeting has been established.

The Czech Republic (including former Czechoslovakia) has a very long tradition in monogenean research and was home for three famous monogenean specialists, Radim Ergens (1933–2007) Institute of Parasitology, Czech. Academy of Science, Prague and after 1985 České Budějovice, Zdeněk Lucký (1925–2011), Veterinary and Pharmaceutical University, Brno and Rudolf Žitňan (1930–1992) Institute of Helminthology, Slovak Academy of Science, Košice, Slovakia. We would like to use this meeting to make you more familiar with current monogenean research.

The Czech Republic

The small republic into the heart of the European continent is the youngest form of the state which has been in existence for almost twelve hundred years. In 1992, Czechoslovakia went through a divorce, creating two separate states, the Czech and Slovak Republics. The relations between the countries are very good and cooperation continues.

Brno

Brno, the metropolis of the South Moravia, is the second largest city in the Czech Republic with the population about 500 000. The location was first inhabited more then 70 000 years ago, and it is possible to trace long sequence of cultures up to the time of arrival of the Slavonic tribes. The name of the city is derived from the old slavonic word „brn“, meaning „clay“.

At present, Brno is prominent centre of higher education; the Masaryk University, the Technical University, the Veterinary and Pharmaceutical University, Gregor Mendel University of Agriculture, the Military University and Janáček Academy of Music and Drama are situated in Brno. Here J. G. Mendel defines his theory of genetics, V. Kaplan developed his first water turbine, L. Janáček composed his famous music, B. Fuchs and Mies van der Rohe built their work of modern architecture (e.g. villa Tugendhat) and M. Kundera wrote his first novels.

The city is located 200km (125 miles) from the Czech capital, Prague, and 130 km (60 miles) both from Slovak capital Bratislava and Austrian capital Vienna.

The Masaryk University

The Masaryk University was founded in 1919, a year after a free Czechoslovakia was reborn, as the second University in the country. It is named after the first president of Czechoslovakia, T. G. Masaryk, who strongly supported establishing of the University. At present, the Masaryk University, located in Brno, is the second-largest public university in the Czech Republic and the leading higher education institution in Moravia. For more information see: www.muni.cz
The Faculty of Science

The Faculty of Science at Masaryk University in Brno provides higher education in the scientific fields of Mathematics, Physics, Chemistry, Biology and Earth Sciences. There are also 153 accredited fields of study in Bachelor’s, Master’s and Doctoral degree programs, designed to educate specialized researchers in the science and the secondary school teachers. Intensive practical activity in science is one of the faculty’s main priorities. For more information see: www.sci.muni.cz

The ECIP team

The ECIP team – European Centre of IchtyoParasitology – was instigated as a mean of bringing together new innovation and improved complementarity to ichthyoparasitological research, encouraging new researchers and increasing mutual collaboration between:

- Department of Botany and Zoology, Faculty of Science, Masaryk University, Brno
- Institute of Parasitology, Biology Centre of the Czech Academy of Sciences, České Budějovice
- Institute of Vertebrate Biology of the Czech Academy of Sciences, Brno

ECIP’s research is organized into four research modules:

- Parasite diversity and adaptation to parasitism
- Phylogenetics and molecular taxonomy
- Host-parasite relationships and molecular integrations
- Evolutionary ecology of model parasite-host systems

Each module utilises a wide range of methodological approaches to study eight basic parasite groups (Apicomplexa, free-living amoebae, Myxozoa, Monogenea, Trematoda, Cestoda, Nematoda and parasitic Crustacea) including their fish hosts.

Almost 50 mostly young researchers including PhD students from 13 countries and four continents are involved. For more information see www.ecip.cz

ISM8 Venue information – Hotel Continental

Four-star Hotel Continental Brno is situated in a quiet part of town. Way to the centre takes something around 10 minutes by car. Every room is available free of charge unlimited Internet connection.

Advantages for the organization of congress and social events is that the premises are located in the city centre. Providing new modern facilities, space for accompanying services (serving refreshments, business presentations, registration of participants of events, etc.) bar in the immediate vicinity of the halls with a fine selection of snacks, air conditioning, dressing-room opposite the halls and comfortable availability by cargo lift directly from the hotel parking lot. Gastronomy services at a high level.

For more information see: www.hotel.continetal.brno
Registration and Information Desk

The IMS8 Registration and Information Desk will be open for registration on Sunday 6 August from 16:00–18:30 and on Monday 7 August from 9:00–10:00 both at Hotel Continental. From Monday 7 August until Thursday 10 August, the desk will be staffed throughout the conference each day from 9:00–16:00 at the Symposium site.

Badges

The IMS8 Organizing Committee request that delegates and accompanying persons wear their badges through the Symposium. Only badge holders will be admitted to the Scientific Sessions and Social events. The IMS8 organizers and their assistants will be easily identified according to blue colour of their badges. Do not hesitate and ask them for any help you will need during the Symposium.

Meals

Lunches, morning and afternoon coffees and teas are provided free of charge for all registered delegates and accompanying persons. Monday (7 Aug), Tuesday (8 Aug) and Wednesday (9 Aug) are free evenings and no provision has been made for dinner. Delegates are encouraged to explore the city’s night life. Within the city centre, there are a lot of nice and pleasant restaurants with Czech or international cuisine/food. For recommendations of the best places, please kindly see the Message Board on the Symposium site.

Health risk

When it comes to health in Brno/the Czech Republic, tourists can be assured that no health risk exist and that there is no need to for a tourist to be vaccinated before entering the Czech Republic. It is however advised for tourists to take medical insurance not only when visiting the Czech Republic but for other destinations as well. Brno is clean and safe and tape water of the Czech Republic is safe to drink, unless otherwise stated.

Local transportation

Several companies of taxis and city trams, buses and trolleybuses are available from just about anywhere to anywhere. Public transportation in Brno is of international standard and safe. If you wish to hire taxi, it is best to request taxi operators from the information desk at your hotel. There is no metro in Brno.

Banking

The Czech Republic has a world-class and modern financial sector. Banks and automatic teller machines (ATM) are available at the most shopping centres and many special places within the city. All major credit cards can be used in the Czech Republic, Master Card and
Visa are generally accepted. On the other hand, it is still not common to speak English in Banks and shops. Bear in mind, that your accent may also be tricky for local people to understand. So, don’t hesitate to ask the organizers for help. Our students will help you!

Currency, tipping,

Currency of the Czech Republic is Crown (koruna). Its value is generally about 26 and 22 that of Euro and US dollars respectively. A lot of international currencies can be exchanged at the various banks and bureaux de Exchange, but you may find that bureaux de change charges less for conversions that the banks. The Crown notes include C100 (green), C200 (orange), C500 (light grey), C1000 (violate) C2000 (dark green) and C 5000 (silver-blue). The Crown coins include C1, C2, C5, C10, C20 and C50.

Tipping is generally expected in the Czech Republic if you are being served by a waiter or waitress at a restaurant and it is not included in the bill. Recommended value of the tipping is usually 10% of the bill received. You are not obliged to tip for poor service.

Message Boards

Changes to the Scientific Program will be posted on a message board outside the lecture hall. Important announcements concerning all delegates will be given prior to Scientific Sessions.

Student and young scientists awards

Student presentation will be judged and prizes will be awarded to best student oral and poster presentations. Presentations will be judged on quality of the study and of the presentations. Young scientists will be also awarded for best presentation.

Accompanying person program

We have lined up memorable tours/sightseeing opportunities for the accompanying persons. On the occasion that the accompanying persons do not return to the conference venue for lunch, they will be provided with lunches on sites during the trip. Don’t forget your camera!

Beer contest

We will have delegates from all parts of the globe and want to invite you to bring with you a can or bottle of traditional beer from your country. We will have a contest during which a panel of experts will judge can/bottle and contents at the symposium.
Other information

For those bringing laptop computer or cell phones with chargers, Czech Republic electricity operates on 220 V. Plugs used here are typically the 3-pin standard round type, and 2-pin round type for smaller plugs.

If you are renting a vehicle, please remember that in Czech Republic we drive on the right, and that all pedestrians must first look left when crossing any of our roads. We have a rule that green light for pedestrians give right of way, however, be very vigilant.

Crime in Brno is rather low, however please do not venture out into dark street especially out of city centre at night and ensure that your valuables such as cameras and valets are kept out of sight. Passports should be locked away at your hotel for safe-keeping. If you are unsure, please speak to any organizers.
Welcome in Brno

You can enjoy the city of Brno or surrounding area, see our suggestions below:

**Labyrinth underneath Zelný trh** (“Vegetable Market”)/Ossuary underneath the Church of St. James/Mint Master’s Cellar - network of underground tunnels, corridors, shafts, crypts and cellars http://ticbrno.cz/en/underground (photo Michal Růžička, TICmB)

The Capuchin crypt - cellar of the monastery from the 18th century where the naturally mummified remains of members of the Capuchin order, their patrons and prominent personalities of Brno lie, (photo Michal Krajiček)


- **Vila Tugendhat** - part of UNESCO World Cultural Heritage as a monument of modern architecture, http://www.tugendhat.eu (photo David Židlický)

- **Boat trip on Brno Reservoir and/or Veveří Castle** - popular recreational area with opportunities not only for swimming and water sports, but also hiking and cycling, or you can enjoy boat trip to the Veveří Castle, founded in the middle of 11th century as a hunting lodge, https://www.veveri.cz/en (photo www.prygl.net)

- Beer tram (Šalina pub) - each Friday you can enjoy sightseeing ride with the beer in your hand, http://www.salina-pub.cz/cs-CZ/ (photo www.archiv.ihned.cz)

- VIDA Science centre - science amusement park with seventy interactive exhibits that give better understanding of the world around us, https://vida.cz/english/ (photo www.vida.cz)
● **Starobrno Brewery** - near the Mendel Museum you can savour beer and Czech meal in the garden of Starobrno Brewery, or participate a brewery excursion with beer degustation, http://pivovarskastarobrno.cz/english/ (photo www.pivovarskastarobrno.cz)

● **Swimming pool** or **Brno Observatory and Planetarium** in Kraví Hora, http://www.hvezdarna.cz/?page_id=28 (photo www.gotobrno.cz)

● **Shopping** (Galerie Vaňkovka, Olympia shopping park)
Time table at a glance

Sunday 6 August 2017
16:00 - 18:30   Registration desk will be opened at Hotel Continental
19:00 - 21:30   Welcome Reception, Hotel Continental Restaurant

Monday 7 August 2017
09:00 - 10:00   Registration Desk open at symposium site of Hotel Continental
10:00 - 10:30   Symposium opening
10:30 - 12:15   TAXONOMY AND DIVERSITY 1
12:30 - 13:30   Lunch - Hotel Continental Restaurant
14:00 - 16:00   EVOLUTION AND PHYLOGENETICS
16:00 – 16:30   Coffee Break
16:30 - 17:30   TAXONOMY AND DIVERSITY 2
17:30 – 18:30   Poster Session 1
19:00 - 22:00   Free evening

Tuesday 8 August 2017
09:00 - 10:30   HOST-PARASITE INTERACTIONS: EVOLUTIONARY ECOLOGY
10:30 - 11:00   Coffee Break
11:00 - 12:15   TAXONOMY AND DIVERSITY 3
12:30 - 13:30   Lunch
14:00 - 15:30   HOST-PARASITE INTERACTIONS: IMMUNITY AND MOLECULAR INTERACTIONS
15:30 - 16:00   Coffee Break
16:00 - 17:00   Session 7 - GENOMICS AND TRANSCRIPTOMICS OF MONOGENEANS
17:00 - 17:45   ISM9 - presentations and discussion regarding the next symposium
17:45 - 18:45   Poster Session 2
19:00 - 22:00   Free evening

Wednesday 9 August 2017
8:30 - 17:00   Mid symposium trip to Valtice – Beer Contest – Group Photo
19:00 – 22:00   Free evening

Thursday 10 August 2017
09:00 - 10:30   MORPHOLOGY AND ULTRASTRUCTURE OF MONOGENEANS
10:30 - 11:00   Coffee Break
11:00 - 12:15   DIVERSITY AND EVOLUTION
12:30 – 13:30   Lunch
14:00 - 15:30   MONOGENEANS IN INTRODUCED, IMPORTED AND COMMERCIALLY IMPORTANT FISH
15:30 - 16:00   Coffee Break
16:00 - 17:00   MONOGENEANS IN INTRODUCED, IMPORTED AND COMMERCIALLY IMPORTANT FISH
19:00   Departure for the gala evening – atrium of CEITEC, Masaryk University Campus
19:30 - 23:00   Gala dinner with the closing of the symposium

Friday 11 August 2017
Time to explore the City of Brno or small groups visit of Masaryk University Campus
Social and accompanying person program

Sunday 6 August 2017
19:00 - 21:30  Welcome Reception, Hotel Continental Restaurant

Monday 7 August 2017
Meeting point at 13:45 in the Reception Hall, Hotel Continental
City tour Brno - two-hour guided tour around the historical centre of Brno

Tuesday 8 August 2017
Meeting point at 8:30 at the parking place, Hotel Continental
Bouzov Castle - 14th-century fortress

Wednesday 9 August 2017
Meeting point at 8:30 at the parking place, Hotel Continental
Mid conference trip to Valtice Castle
10:00–12:00  Visit of Castle Valtice, UNESCO World Heritage site
12:00–14:00  Lunch in the horse stables with the concert of brass instruments quartet
             Moravské žestě
14:30–17:00  Visit of historical vine cellar with the degustation of Moravian vines

Thursday 10 August 2017
1. Meeting point at 8:30 at the parking place, Hotel Continental
Wallachian Open Air Museum
2. Meeting point at 19:00 at the parking place, Hotel Continental, Departure for the gala evening.
19:30 – 23:00  Gala dinner with the closing of the symposium
             International beer contest award
             Students and young scientists awards
             Closing of the ISM8 and the venue of the next ISM9
             Cimbalom band and great time

Friday, 11 August, 2017
Time to explore the City of Brno or small groups visit of Masaryk University Campus
Scientific programme

SUNDAY 6. 8.
16:00–18:30 Registration, Hotel Continental
19:00–21:30 Welcome Reception, Hotel Continental

MONDAY 7. 8.
9:00–10:00 Registration
10:00–10:30 Symposium opening

TAXONOMY AND DIVERSITY I
10:30–11:00 Madagascar: A polystome diversity hotspot
L. du Preez, O. Verneau
11:00–11:15 Taxonomy and phylogeny of oviparous Gyrodactylids
(Platyhelminthes: Monogenoidea), parasites of Loricariidae
(Ostariophysi: Siluriformes) from Guamá River, Pará, Brazil
A. R. Oliveira Rodrigues, M. V. Domingues
11:15–11:30 Morphometric and molecular characterization of Gyrodactylus
spp. (Platyhelminthes: Monogononta) from Antarctic nototheniid fish
I. Heglasová, V. Nezhybová, I. Přikrylová
11:30–11:45 Ancyrocephalinae (Monogenea: Dactylogyridae)
of fishes in Iraq: literature review and future research
F. Shihab Al-Nasiri, J. A. Balbuena
11:45–12:00 Morphological and molecular diversity of Characidotrema
(Monogenea: Dactylogyridae) parasitizing African tetras
(Characiformes: Alestidae)
M. L. Červenka Kičinj, M. Seifertová, E. Řehulková
12:00–12:15 How many monogenoid species are there on Indian notopterids
S. Rajvanshi, A. Tripathi, N. Agrawal, J. Verma, G. G. Agrawal
12:30–13:30 Lunch

EVOLUTION AND PHYLOGENETICS
14:00–14:30 Exploring the use of mitogenomics for phylogenetic reconstruction
of African monogenes (Gyrodactyliidea and Dactylogyridae)
M. P.M. Vanhove, A. Briscoe, W. Fannes, M. W.P. Jorissens,
D. T. J. Littlewood, T. Huyse
14:30–14:45 Dactylogyrus (Monogenea) parasitizing cyprinids from Balkan
Peninsula: phylogeny and relationships to central European species
M. Benovics, Y. Desdevises, A. Šimková (S1)
14:45–15:00 Preliminary phylogenetic analyses of Dactylogyrus spp. from
Enteromius hosts from the Limpopo and Olifants River systems, South Africa
M. Raphahlelo, M. Matla, S. Dippenaar, I. Přikrylová (S2)
15:00–15:15 Coevolution between Anacanthorus spp. (Monogenea:
Dactylogyroidea) and Serrasalmidae (Characiformes)
J. Moreira, A. Šimková, J. Luque (S3)
15:15–15:30 How three genera of Monopisthocotylean monogenes and their
fish hosts coevolved
R. Míguez-Lozano, A. Rodríguez-González, J. A. Balbuena
6–11 August 2017, 8th International Symposium on Monogenea

15:30–15:45  **Phylogenetic relationships among Diplectanum spp. and host-parasite associations in sciaenid fishes from the Mediterranean**
M. Villar-Torres, I. Blasco-Costa, F. E. Montero, J. A. Raga, A., Repullés-Albelda

15:45–16:00  **Gill parasites (Monogenoidea) of Neotropical cichlids (Cichlidae) and their phylogenetic relationships based on molecular data**
C. A. Mendoza-Palmero, I. Blasco-Costa, G. Pérez-Ponce de León (J1)

16:00–16:30  Coffee break

**TAXONOMY AND DIVERSITY II**

16:30–16:45  *Quadriacanthus* species (Monogenea: Dactylogyridae) parasitizing catfishes (Teleostei: Siluriformes) in Eastern Africa: morphological and molecular characterization
K. Francová, M. Seifertová, R. Blažek, M. Gelnar, Z. N. Mahmoud, E. Řehulková

16:45–17:00  **Cichlidogyrus** spp. (Monogenea: Dactylogyridae) communities of Burundese and Congolese cichlids (Lake Tanganyika): taxonomic study and phylogenetic approach
Ch. Rahmouni, M. P.M. Vanhove, A. Šimková (S4)

17:00–17:15  Checklist of monogenean parasites of freshwater fish from Iran with notes on their distribution
M. Raissy, M. Ansari

17:15–17:30  Three new species of Dactylogyrus Diesing, 1850 (Monogenea: Dactylogyridae), from Moroccan Cyprinids based on morphological and molecular data
I. Rahmouni, E. Řehulková, A. Pariselle, O. Berrada Rkhami, A. Šimková (S5)

17:30–18:30 Poster Session

**TUESDAY 8.8.**

**HOST-PARASITES INTERACTIONS: EVOLUTIONARY ECOLOGY**

9:00–9:30  Congeneric monogeneans in evolutionary and ecological studies
A. Šimková, M. Benovics, I. Rahmouni, J. Vukić

9:30–9:45  **The TILAPIA project: Parasite host switching in the Congo Basin?**

9:45–10:00  Monogenean parasite communities in asexual-sexual *Carassius auratus* complex
T. Pakosta, A. Šimková (S7)

10:00–10:15  *Trachemys scripta elegans* as a vector of polystome dispersal
O. Verneau, L. Héritier, L. du Preez

10:15–10:30  The maternal ancestry and monogenean distribution in hybrids of Cyprinidae fish
V. Krasnovyd, L. Vetešník, A. Šimková (S8)

10:30–11:00 Coffee break

**TAXONOMY AND DIVERSITY III**

11:00–11:15  Diversity of monogeneans from freshwater fishes in Africa
E. Řehulková, K. Francová, I. Příkrylová, M. Seifertová

11:15–11:30  Identification of a new species of genus *Paradiplozoon* Achmerov, 1974 from the gills of *Sikukia gudgeri* (Smith, 1931) (Cyprinidae) from China
L. Fan, J. Bai, F. Meng, J. Wang, W. Xu, X. Wang
11:30–11:45  **Diversity and distribution of species of *Gyrodactylus* von Nordmann, 1832 on African cichlids**  
P. Zahradničková, M. Barson, W. J. Luus-Powell, M. Gelnar, I. Přikrylová (S9)

11:45–12:00  **Preliminary studies on the monogenean from fishes of Sansha in the South China Sea**  
Z. Zhou, Y. Li, K. Yuan, Z. Xiao, L. Liu, X. Ding

12:00–12:15  **Monogenean Parasites Fauna from Iranian Freshwater Fishes**  
D. Azadikhah, S. Shamsi, B. Jalali Jafari

12:30–13:30 Lunch

**HOST-PARASITE INTERACTION: IMMUNITY AND MOLECULAR INTERACTIONS**

14:00–14:30  **The balance between immune reactions and immune tolerance of fish skin and gills to monogenean colonization**  
K. Buchmann

14:30–14:45  **Inhibition of proteolysis by the hematophagous *Eudiplozoon nipponicum***  
J. Ilgová, L. Jedličková, H. Dvořáková, L. Mikeš, M. Benovics, P. Roudnický, J. Vorel, L. Vojtek, P. Hyršl, J. Salát, M. Gelnar, M. Kašný (S10)

14:45–15:00  **EnKT1-Kunitz type inhibitor of *Eudiplozoon nipponicum* involved in the regulation of hemostasis**  
L. Jedličková, J. Dvořák, J. P. Dalton, I. Hrachovinová, M. Kašný, L. Mikeš

15:00–15:15  **A group of cathepsins L as predominant proteolytic enzymes of *Eudiplozoon nipponicum***  
L. Jedličková, H. Dvořáková, J. Dvořák, J. P. Dalton, M. Kašný, L. Ulrychová, J. Vorel, L. Mikeš (S11)

15:15–15:30  **Cathepsins D and B: the role of aspartic and cysteine peptidases in the life of *Eudiplozoon nipponicum* (Monogenea)**  
H. Dvořáková, L. Jedličková, M. Kašný, J. Bulantová, L. Mikeš (S12)

15:30–16:00 Coffee break

**GENOMICS AND TRANSCRIPTOMICS OF MONOGENEANS**

16:00–16:15  **Diplozoons (Monogenea): what is the molecular base of their interaction with the host?**  

16:16–16:30  **De novo assembly of transcriptome of *Neobenedenia melleni* (Platyhelminthes: Monogenea) and data analysis**  
Z. Zhou, X. Wang, Z. Xiao, K. Yuan, X. Ding, L. Liu

16:30–16:45  **Eudiplozoon nipponicum (Polyopisthocotylea: Diplzoidae): transcriptome and secretome analyses of hematophagous fish parasite**  

16:45–17:00  **Genomic insights into the evolution of *Gyrodactylus salaris* and related species**  
Ch. Hahn, L. Bachmann (J2)

17:00–17:45  **ISM9 – presentations of candidates for organisation of next symposium**

17:45–18:45 Poster session
WEDNESDAY 9.8.
08:30 – 17:00  Mid conference trip to Valtice Castle

THURSDAY 10.8.

MORPHOLOGY AND ULTRASTRUCTURE OF MONOGENEANS
9:00–9:30  Spermiogenesis and spermatozoon ultrastructure in basal polyplastriclytean monogeneans, Hexabothriidae and Chimaericolidae, and significance for the phylogeny of the Monogenea
J. Justine, L. G. Poddubnaya
9:30–9:45  Unique ultrastructural characteristics indicating a chimaera-like organisation for chimaericicolid monogeneans
L. G. Poddubnaya, D. I. Gibson
9:45–10:00  Ectoparasitic vocation: trematodes living as monogeneans
A. E. Ahuir-Baraja, F. E. Montero
10:00–10:15  Neodiplectanotrema helicoleni from different geographical Atlantic and Mediterranean locations: the attachment of an endoparasitic monogenean
M. Villora-Montero, J. S. Hernández-Orts, A. Pérez-del-Olmo, F. E. Montero (S14)
10:15–10:30  Ultrastructural aspects of surface and attachment clamps of Paradiplozoon homoin (Monogena, Diplozoidae)
V. Konstanzová, B. Koubková, M. Gelnar (S15)

10:30–11:00 Coffee break

DIVERSITY AND EVOLUTION
11:00–11:15  Monogenean parasites of endemic cyprinid species from the selected areas of Mediterranean
K. Vyčítalová, M. Benovics, B. Koubková, A. Šimková (S16)
11:15–11:30  Can worms tell us who their hosts are?
A. L. Schoeman, L. H. du Preez (S17)
11:30–11:45  How many species? How many forms? Hidden biodiversity of merizocotyline monogeneans in batoids (Chondrichthyes: Elasmobranchii) from the coasts of the Argentine Sea
M. M. Irigoitia, P. E. Braicovich, M. D. Farber, J. T. Timi (S18)
11:45–12:00  Highly host-specific gill Monogeneans (Pseudorhabdosynochus spp., Diplectanidae) on groupers (Perciformes) in the Mediterranean Sea: What do we know?
A. Chaabane, L. Neifar, J. Justine (J3)
12:00_12:15  The use of Gyrodactylus parasites in studying host migration
T. Huyse, G. A. Bristow, M. Oeyen, M. H.D. Larmuseau, B. Hellemans, F. A.M. Volckaert, M. A. Bell, A. MacColl, J. A.M. Raeymaekers

12:30–13:30 Lunch

MONOGENEA IN INTRODUCED, IMPORTED AND COMMERCIAL IMPORTANT FISH
14:00–14:30  Integrating biology with management: ongoing understanding of approaches to controlling monogeneans in aquaculture
M. R. Deveney
14:30–14:45  Fisheries targets in Lake Tanganyika under the magnifying glass: a look at their monogeneans
6–11 August 2017, 8th International Symposium on Monogenea

14:45–15:00 Co-introduction of ancyrocephalid monogeneans on their invasive host, the largemouth bass, *Micropterus salmoides* (Lacepéde, 1802) in South Africa
M. Truter, I. Přikrylová, O. L.F. Weyl, N. J. Smit (S20)

15:00–15:15 Studies on the biology of *Cichlidogyrus sclerosus* and the seasonal dynamics of monogenean populations and communities on three strains of tilapia in net cages
T. Yang, Y. Zhou

K. R. Tancredo, N. C. Marchiori, S. A. Pereira, M. L. Martins (S21)

15:30–16:00 Coffee break

16:00–16:15 Exotic monogenoids and Indian guidelines for the import of ornamental fish: review and perspective
A. Tripathi

16:15–16:30 Monogenean parasites infecting imported ornamental fish to Australia
A. Trujillo-González, J. A. Becker, D. B. Vaughan, K. S. Hutson (S22)

16:30–16:45 Environmental influence on infective stages of the monogenean *Sparicotide chrysophrii*, parasite of gilthead sea bream (*Sparus aurata*) in Mediterranean aquaculture
M. Villar-Torres, F. E. Montero, J. A. Raga, A. Repullés-Albelda

16:45–17:00 Monogenean parasites causing pathological impacts on commercially important Mediterranean Sea fishes in Egypt
M.M. Fahmy, N. E. Mahmoud

19:00 Departure for the gala evening
19:30–23:00 Gala dinner with the closing of the symposium

**FRIDAY 11.8.**

Time to explore City of Brno ☻ or small group visit of Masaryk University Campus
List of the posters

TAXONOMY AND DIVERSITY

P1 Description of a new gyrodactylid (Gyrodactylidae: Monogenea) genus from the gills of the bulldog, Marcusenius macrolepidotus (Peters) from Lake Kariba, Zimbabwe
I. Přikrylová, G. Paladini, M. Barson, A. Shinn

P2 Monogenean biodiversity in elasmobranchs from the Oceanogràfic aquarium of Valencia
C. Taurá Pardo, A. Repullés-Albelda, F. E. Montero, A. E. Ahuir-Baraja

P3 Three-dimensional images of rigid structures by Laser Scanning Confocal Fluorescence microscopy as a tool for taxonomic studies on Dactylogyridae (Monogenea)
M. M. Irigoitia, P. N. De Francesco, M. A. Rossin, J. T. Timi (S)

P4 Preliminary study on the monogenean parasites of cyprinid fishes from the river Sauer (Germany/Luxembourg)
E. Dzika, H. Taraschewski, F. Scharfenberger, F. Thielen

P5 Morphometric and scanning electron microscopy of Dactylogyrus minutus Kulwièc, 1927 (Monogenea: Dactylogyridae) from the Cyprinus carpio var. koi
K. R. Tancred, N. da Costa Marchiori, M. L. Martins (S)

P6 Diversity and distribution of parasitic monogenoids (Platyhelminthes) in a global biodiversity hotspot - Arunachal Pradesh, India
A. Tripathi, M. Yassa (S)

P7 Enteromius paludinosus (Peters, 1852): host for new species of Gyrodactylus von Nordmann, 1832 (Monogenea: Gyrodactylidae) in South Africa
M. Truter, I. Přikrylová, N. Smit (S)

P8 Monogeneans of marine fishes from Patagonia, Argentina: diversity and new host records
S. Georgieva, F. E. Montero, F. J. Aznar, N. A. García, E. A. Crespo, J. A. Raga, J. S. Hernández-Orts

P9 Introduction to “Fauna Sinica Platyhelminthes Monogenea II”
L. Liu, H. Li, J. Zhang, X. Ding

P10 Taxonomic Studies on Monogeneans from Elasmobranchs off South China Sea
F. Yang, X. Ding, L. Liu, K. Yuan, Z. Xiao (S)

P11 Monogenean diversity of western Mediterranean mackerels, bonitos and tunas (Scombridae)

P12 First record of Protocotyle grisea (Cerfontaine, 1899) (Monogenea: Hexabothriidae) infecting the bluntnose sixgill shark, Hexanchus griseus (Bonnaterre) from the Valencian Gulf, Spain
J. F. Palacios-Abella, A. Juan-Garcia, R. Kuchta, C. Mendoza-Palmero, F. E. Montero, J. S. Hernández-Orts

P13 Monogenean fauna of Japanese freshwater fishes and phylogeny of Dactylogyridae from Japan
M. Nitta, K. Nagasawa (S)

P14 Diversity of monogeneans from selected fish species from Haut-Katanga Province in the Democratic Republic of Congo
W. J Smit, W. J Luas-Powell, G. Kasembele, A. Chocha Manda, J. R. Sara, F. M. Bukinga, E. Ngoma, M. P. M. Vanhove (S)
P15 Morphometric and molecular characterisation of two new species of *Enterogyrus Paperna, 1963* (Dactylogyridae: Ancyrocephalinae) from *Oreochromis mossambicus* in South Africa
W. J. Luus-Powell, M. M. Matla, G. N. Madaniere-Moyo, S. Tavakol, I. Přikrylová

P16 The first record of dactylogyrid parasites from *Labeo horie* and *Labeo niloticus* from Sudan
M. Pravdová, E. Řehulková, M. Ondračková, I. Přikrylová, R. Blažek, M. Gelnar (S)

P17 Monogenean parasites of sparid fishes, from the western Mediterranean off Algeria
M. Douinzed, L. Lamia, R. Mohammed, B. Fatima, B. Mohamed

P18 Monogenea of largemouth bass in southern Africa: new records
M. M. Matla, M. N. Mokgalong, S. N. Mashego

P19 Dactylogyrids (Platyhelminthes, Monogenea) from the gills of *Hoplias malabaricus* (Characiformes: Erythrinidae) from coastal rivers of the Oriental Amazon Basin: Species of *Urocleidoides* and New Genus

P20 Monogeneoid parasites of *Acestrorhynchus falcatus* (Characiformes: Acestrorhynchidae) from Pará State, Brazil: species of New Genus and *Diaphorocleidus* (Monogeneida: Dactylogyridae)
J. F. dos Santos Neto, N. G. Souza Costa, M. V. Domingues

P21 Diversity of Monogeneida (Platyhelminthes, Neodermata) of two components of the neotropical ichthyofauna: Loricariidae (Siluriformes) and Gymnotiformes from coastal rivers of the Oriental Amazon Basin and Xingu Basin
B. Branches, M. V. Domingues

P22 Dactylogyrids (Platyhelminthes, Monogenea) from the gills of *Hassar gabiru* Birindelli, Fayal et Wosiacki, 2011 and *Hassar orestis* (Steindachner, 1875) (Siluriformes, Doradidae) from the Xingu Basin
G. Barbosa Soares, M. V. Domingues

HOST-PARASITES INTERACTIONS

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P24 Expression of genes encoding Toll-like receptors associated with infection by monogenean parasites
A. Potrok, A. Šimková, L. Gettová (S)

P25 Microscopic anatomy of monogenean parasite *Paradiplozoon homoion*
I. Hodová, R. Sonnek, M. Gelnar, A. Valigurová

P26 The immunity induced by *Dactylogyrus ctenopharyngodonid* can protect grass carp from infection of *D. ctenopharyngodonid* and *Ichthyophthirius multifiliis*
Q. Zhang, J. Li, Y. Fu

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N. E. Mahmoud, M. M. Fahmy

P28 Prevalence of monogenean species in *Oreochromis niloticus* from southern Brazil

P29 *Pyragraphorus hollisae* Euzet & Ktari, 1970 infecting pompano, *Trachinotus ovatus* (L.), in captivity
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Abstracts of oral lectures
In order of appearance
Madagascar: A polystome diversity hotspot

Louis du Preez¹, Olivier Verneau¹, ²

¹ Unit for Environmental Sciences and Management, North-West University, Potchefstroom, South Africa
² Univ. Perpignan Via Domitia, Centre de Formation et de Recherche sur les Environnements Méditerranéens, Perpignan, France

With more than 300 known species of frogs and probably many more to describe the island of Madagascar is a true amphibian hot spot. Madagascar is known for its exceptional degree of endemicity. Of all the species in Madagascar, only the Asian Toad Duttaphrynus melanostictus and the Indian Bullfrog Hoplobatrachus tigerinus are not endemic.

This unique diversity posed the opportunity to explore for the presence of polystomes. *Metapolystoma brygoonis* was described from Ptychadena mascareniensis in 1964 and *Uropolystomoides chabaudi* from the terrapin Pelomedusa subrufra in 1965.

Several visits were undertaken to Madagascar and of the 86 frog species screened we recovered polystomes from 25, all belonging to the Ptychadenidae and Mantellidae. A molecular phylogenetic analysis based on DNA sequences of the 18S and 28S rRNA genes uncovered an unexpected variety of polystome species that belonged into two separate clades: one forming a lineage within the genus *Metapolystoma*, with several species in the mantellid host genera Aglyptodactylus and Boophis, and the second corresponding to two newly described genera *Madapolystoma* and *Kankana* that was found in species in the family Mantellidae. Molecular dating suggested *Madapolystoma* colonized Madagascar 135-40 million years ago (Mya), whereas the origin of *Metapolystoma* in Madagascar was more recent, dating back to about 14-2 My. *Madapolystoma* appeared to be sister group to *Eupolystoma* that is known from pyxicephalid and bufonid hosts from Africa and India, and may have colonized Madagascar in the Late Mesozoic or early Cenozoic, either with the mantellid ancestors from India or with early, now extinct, hyloid frogs. In contrast, *Metapolystoma* is currently known only from Ptychadena in Africa and Madagascar. The polystome data strongly suggested that the ancestors of *P. mascareniensis* colonized Madagascar naturally by overseas dispersal, bringing along *Metapolystoma* parasites. These gave rise to *M. brygoonis* and, after a host switch, to a rapid radiation of *Metapolystoma* species specialized to mantellid hosts. The high diversity of polystomes in Madagascar provide a striking example how parasite data can provide novel insights into the biogeographic history of their hosts.
Taxonomy and Phylogeny of Oviparous Gyrodactylids (Platyhelminthes: Monogenoidea), Parasites of Loricariidae (Ostariophysi: Siluriformes) from Guamá River, Pará, Brazil

Allan Rodrigo Oliveira Rodrigues¹, Marcus Vinicius Domingues²

¹ Universidade Federal do Pará, Campus Universitário de Bragança, Instituto de Estudos, Bragança, Pará, Brazil
² Programa de Pós-Graduação em Biologia Ambiental, Universidade Federal do Pará

Five new species of oviparous gyroactylids are described from the body surface of fishes from Loricariidae (Siluriformes) family from Guamá river, Pará, Brazil: Oogyrodactylus sp. n.1 and Oogyrodactylus sp. n. 2 from Farlowella sp. and Rinoloricaria sp. respectively; Phanerothecium sp. n. from Lasiancistrus saetiger Ambruster; Nothogyrodactylus sp. n. from Ancistrus sp. A new genus is proposed to accommodate a new species from Cochliodon sp. This genus is characterized by possessing: a male copulatory organ with spines; accessory piece; and esclerotization in the opening of the fertilization chamber. A hypothesis on the phylogenetic relationship for genera of oviparous gyroactylids + Gyrodactylus based on morphological data is proposed (Length=28; Consistency Index: 89%; Retention Index: 83%). The present hypothesis is (Nothogyrodactylus (Aglaiogyrodactylus (Onychogyrodactylus (Oogyrodactylus, Phanerothecioides (Gyrodactylus, Hyperopletes, Phanerothecium, New Genus))). This hypothesis indicates that Gyrodactylus (i.e., viviparous gyroactylids) appears as a taxon associates with an oviparous clade, as already supported in previously hypotheses.
Morphometric and molecular characterization of *Gyrodactylus* spp. (Platyhelminthes: Monogenea) from Antarctic notothenioid fish

Ivana Heglasová¹², Veronika Nezhybová³⁴, Iva Přikrylová⁴⁵⁶

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² Institute of Parasitology, Slovak Academy of Sciences, Košice, Slovak Republic
³ Institute of Vertebrate Biology, Academy of Sciences of the Czech Republic, Brno, Czech Republic
⁴ Department of Botany and Zoology, Faculty of Science, Masaryk University, Brno, Czech Republic
⁵ Water Research Group, Unit for Environmental Sciences and Development, North West University, Potchefstroom, South Africa
⁶ Department of Biodiversity, School of Molecular and Life Sciences, University of Limpopo, Sovenga, South Africa

Parasites of the genus *Gyrodactylus* von Nordmann, 1832 are tiny worms being distributed worldwide. Nearly 500 species have been described so far, of which 466 species can be considered as a valid species. Currently, only nine species of *Gyrodactylus* are known from notothenioid fishes in Antarctic area.

A total number of 115 fish specimens of five species were collected during two Antarctic expeditions at the Czech Antarctic Station of Johann Gregor Mendel, James Ross Island (Prince Gustav Channel, Weddel Sea) between January 2013 and March 2014. Morphometric and genetic analysis of collected material were performed at the laboratory in the Czech Republic.

Species identification based on the morphometry of opisthaptoral hard parts in combination with nuclear rDNA sequences confirmed the presence of four species. The identified species were: *Gyrodactylus antarcticus* Gussev, 1967, *Gyrodactylus cf. byrdi* and *Gyrodactylus coriiceps* Rokicka, Lumme, Zietara, 2009. The fourth species recorded from *Trematomus bernacchii*, *Trematomus hansoni* and *Trematonus newnesi* represent new species. Based on the ITS rDNA sequences the newly identified species resulted as a sister taxon to *G. antarcticus*. The phylogenetic analysis based on the 18S rDNA showed that Antarctic species are more closely related to European *Gyrodactylus* spp. than those South American.

This research was funded by the Grant Agency of the Czech Republic (GBP505/12/G112)
Ancyrocephalinae (Monogenea: Dactylogyridae) of fishes in Iraq: literature review and future research

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² Cavanilles Institute of Biodiversity and Evolutionary Biology, University of Valencia, Valencia, Spain

The Ancyrocephalinae are gill parasites of fishes. In Iraq, 14 species have been reported so far from marine, brackish and freshwater fishes, namely, one species of Ancyrocephalus (A. polymorphus), three species of Cichlidogyrus (C. tiberianus, C. tilapiae, C. sclerosus), two species of Haliotrema (H. mugilis, H. mugilinus), seven species of Ligophorus (L. bantingensis, L. fluvialitis, L. heteronchus, L. imitans, L. lebedevi, L. sagmarius, L. vanbenedenii) and one species of Paradactylogyrus (P. bati). In addition, three unidentified species of Ancyrocephalus, Haliotrema, and Ligophorus have been reported.

These numbers probably do not represent the actual species richness of monogenean ancyrocephalines of fishes in Iraq, but only the number of species presently reported from some geographical region by researchers who have worked in these regions. Therefore, these described species represent a very small portion of the expected richness of parasitic fauna of fish in Iraq.

The identification of ancyrocephalines of Iraq was based on morphometrical data and differences in the sclerotized parts of the opisthohaptor and copulatory organ. However, scrutiny of the publications and their respective figures suggests that a number of species have been probably misidentified. In fact, a previous study has noted the erroneous identification of Dactylogyrids, which were described on grey mullets (Mugilidae) in Iraq. Unfortunately, molecular and genetic data are not available for all recorded species. Only one actual research paper had been published for identification of ancyrocephaline parasites in Iraq, but it is restricted on the diagnosis of Ligophorus spp. parasitizing grey mullets in Shatt Al-Arab River and Estuary in the South of Iraq, and it is still lack of molecular studies. Additional research of this genus is being carried out by us in grey mullets collected from the Tigris River passing through Baghdad city.

The aim of the present contribution is to summarize the ancyrocephaline monogeneans recorded so far in fish in inland water bodies from Iraq, with information of their host and geographic ranges. We discuss and highlight the possibility of making future morphometric and molecular researches on Ancyrocephalinae of fish for the understanding the actual identification of species previously recorded in the studied regions. In additions, future research in unstudied regions are necessary to provide more data on the species composition of these monogeneans in most areas of Iraq.
Morphological and molecular diversity of Characidotrema (Monogenea: Dactylogyridae) parasitizing African tetras (Characiformes: Alestidae)

Maria Lujza Červenka Kičinja¹, Mária Seifertová¹, Eva Řehulková¹

¹ Department of Botany and Zoology, Faculty of Science, Masaryk University, Brno, Czech Republic

African tetras are known to be parasitized by monogeneans representing three genera: Afrocleidodiscus (1 species), Annulotrema (39 spp.) and Characidotrema (10 spp.). To date, Characidotrema were found on the gills of four alestid genera (Alestes, Brycinus, Phenacogrammus, and Hemigrammopetersius) from seven African countries: Cameroon, Egypt, Ghana, Kenya, Tanzania, Togo and Uganda.

The main objectives of the present study are to investigate: (1) species diversity of Characidotrema species from alestids from four other African countries; (2) interspecific genetic variability in Characidotrema spp.; (3) the relationships between Characidotrema spp. and other representatives of Dactylogyridae parasitizing African freshwater fishes based on nuclear gene sequences.

Characidotrema species were collected from Brycinus imberi, B. leuciscus and B. nurse during field trips in the Democratic Republic of the Congo, Senegal, Sudan and Zimbabwe. Parasites were investigated using two complementary approaches: a morphological study of the hard sclerotized structures (haptor, male copulatory organ and vagina) and a molecular one, using ribosomal DNA sequence data (LSU and SSU).

This study revealed the presence of four known species: Ch. brevipenis, Ch. nursei, Ch. spinivaginus, Ch. zelotes and three species probably new to science. Preliminary phylogenetic analyses confirmed the monophyly of Characidotrema and its close relationships with representatives of Annulotrema. Close position within some Characidotrema species corresponds with the morphology of sclerotized structures of male copulatory organ and vagina.

Acknowledgements: Czech Science Foundation (project no. P505/12/G112)
How many monogenoid species are there on Indian notopterids?

Saroj Rajvanshi¹, Amit Tripathi, Nirupama Agrawal¹, Jyoti Verma¹, Girdhar Gopal Agarwal²

¹ Department of Zoology, University of Lucknow, India
² Department of Statistics, University of Lucknow, India

Indian notopterids comprise Notopterus notopterus and Chitala chitala (earlier known as N. chitala). Urocleidus notopterus was described from Notopterus notopterus by Jain (1955). Genus Notopterodiscoides was established by Lim and Furtado, 1986 with N. belidus and N. ornatohamus from Peninsular Malaysia. Agrawal and Vishwakarma (2000), while transferring Urocleidus notopterus (placed under the genus Silurodiscoides by Lim and Furtado (1986) and Lim (2001)) under Notopterodiscoides added N. indicus, N. curvihamulus, N. chitalai, N. cirri and N. lucknowensis in India. Agrawal and Vishwakarma (2000) further transferred T. notopterus (Jain, 1955) Lim and Furtado, 1986, T. platamauxili Lim and Furtado, 1986 and T. tasekensis Lim and Furtado, 1986 under Notopterodiscoides. Examination of Notopterus notopterus and Chitala chitala, commonly known as Patra, knife fish or Feather backs (single, feather like dorsal fin) from different localities in India, suggested that presence of different combination of characters in monogenoidean species complex parasitizing their gills could be separate genera/species. This prompted us to use statistical classifiers and molecular markers for differentiation.
Space for your NOTES
Space for your NOTES
Exploring the use of mitogenomics for phylogenetic reconstruction of African monogeneans (Gyrodactylidea and Dactylogyridea)

Maarten P.M. Vanhove, Andrew Briscoe, Wouter Fannes, Michiel W.P. Jorissen, D. Tim J. Littlewood, Tine Huyse

1 Department of Botany and Zoology, Faculty of Science, Masaryk University, Brno, Czech Republic
2 Invertebrate Section, Biology Department, Royal Museum for Central Africa, Tervuren, Belgium
3 Laboratory of Biodiversity and Evolutionary Genomics, Department of Biology, University of Leuven, Leuven, Belgium
4 Hasselt University, Centre for Environmental Sciences, Research Group Zoology: Biodiversity & Toxicology, Diepenbeek, Belgium
5 Capacities for Biodiversity and Sustainable Development, Operational Directorate Natural Environment, Royal Belgian Institute of Natural Sciences, Brussels, Belgium
6 Division of Parasites & Vectors, Department of Life Sciences, Natural History Museum, London, United Kingdom

The Gyrodactylidea and the Dactylogyridea are probably the most species-rich monogenean taxa, with representatives that can be harmful fish pathogens, especially in intensive aquaculture conditions or after anthropogenic co-introduction outside of their native range. In Africa, several cichlids and clariid catfishes have been relatively well-studied for monogenean parasites, among which the most important aquaculture fishes on the continent, many of which have also been widely introduced outside Africa. A limitation, however, is the small number of genetic markers available for these flatworms. The need for additional and higher-resolution markers is especially apparent when studying processes on a recent timescale, such as population genetics, phylogeography or barcoding. At the moment, a small set of established mitochondrial gene fragments constitutes the most variable markers available for such purposes, with advantages like the lack of recombination, the high rate of molecular evolution… In addition, these mitochondrial sequences also served in flatworm systematics on a higher, macro-evolutionary scale. The high (amino acid) sequence variability within the mitochondrial genomes of monogeneans (and other flatworms) has however hampered the availability of universal barcoding primers.

With this in mind, we want to apply a mitogenomic approach for primer development (by capturing the most variable mitochondrial regions). We here present the first mitogenomic data on African monogeneans (from cichlid and clariid hosts), based on Illumina technology, and explore questions on phylogenetic position and gene order.

Given the wide-ranging applicability of mitochondrial markers, we aim to open up future research avenues for the promising but relatively understudied African monogenean fauna.
Dactylogyrus (Monogenea) parasitizing cyprinids from Balkan Peninsula: phylogeny and relationships to central European species

Michal Benovics1, Yves Desdevises2, Andrea Šimková1

1 Department of Botany and Zoology, Masaryk University, Brno, Czech Republic
2 Observatoire Océanologique de Banyuls, Université Pierre et Marie Curie, Banyuls-sur-Mer, France

Species richness of parasitic taxa and their distribution in host species is usually closely related to history, dispersion and diversity of their hosts. The parasites, belonging to genus Dactylogyrus Diesing, 1850 (Monogenea), are known for the high degree of host specificity. Wide species richness of this genus (over 900 nominal species) could be linked to the multitude of their host species which are freshwater fish belonging to the family Cyprinidae.

Unlike in Central and Northern Europe, where the cyprinid fauna is relatively uniform, Southern European Peninsulas are extremely rich in endemic species and consists of several highly diversified genera. It is hypothesized that the endemic cyprinids harbour endemic Dactylogyrus species. So far, this parasite group is in Balkan Peninsula relatively unexplored. Therefore, we decided to investigate the endemic Balkan Dactylogyrus and their phylogenetic relationships to widely distributed species collected from cyprinids in Central Europe.

In years 2014–2017 we sampled 64 cyprinid species from 53 different localities in Balkan Peninsula and Czech Republic. A total of 51 Dactylogyrus species were identified and used for reconstruction of evolutionary history and further molecular analyses. The highest Dactylogyrus species diversity was found in the representatives of Squalius, Barbus, Chondrostoma and Rutilus genera. The phylogenetic reconstruction based on sequence data of partial ribosomal SSU combined with partial LSU revealed 4 strongly supported groups including endemic and widely distributed species. To resolve phylogenetic relationships within the group including the most of Dactylogyrus species we applied highly variable ITS1 region. The second phylogenetic reconstruction divided this group into 4 well or moderately supported clades.

The computation of genetic distances performed on specimens of generalist Dactylogyrus, such as D. vistulae, D. rarissimus and D. folkmanovae, revealed high interpopulation variability. The highest genetic distances were observed between specimens of D. folkmanovae collected from 7 Squalius species (p-distance 0.002-0.035). The Mantel test supports that geographically adjacent populations are more similar also at the molecular level.

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Preliminary phylogenetic analyses of *Dactylogyrus* spp. from *Enteromius* hosts from the Limpopo and Olifants River systems, South Africa

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Members of the genus *Dactylogyrus* are fish ectoparasites primarily infecting the gills of cyprinids. By far the most diverse species group among monogeneans consists of more than 900 nominal species. There have been numerous studies on the phylogenetic relationships of *Dactylogyrus* spp. from other countries, however, very little is known about their phylogeny in Africa. The studies of the phylogenies are important to understand the patterns of evolutionary relationships amongst species. Currently, only seven species are known from *Enteromius* hosts in South Africa, while the eighth is doubtful (herein discussed).

Fish hosts were collected from various dams and rivers within the Limpopo and Olifants River systems, using a combination of conventional angling gear, casts nets, seine nets, fyke nets and an electric shocker. Fish were identified using standard procedures. From these, three hosts were identified, *Enteromius afrohamiltoni*, *Enteromius unitaeniatus* and *Enteromius trimaculatus*. Parasites were collected from the gills and were preserved in 96% ethanol where after DNA was extracted. From the extracted DNA samples, the partial 18S rDNA region and the entire ITS1 was amplified using primers S1 and IR8, and the 28S rDNA region was amplified using CI and D2 primers. Phylogenetic relationships were estimated using PAUP* to employ parsimony (MP), neighbour joining (NJ), maximum likelihood (ML) and Bayesian inference methods.

This study represents preliminary estimation of the phylogenetic relationships among six species of *Dactylogyrus* from *Enteromius* hosts collected from the Limpopo and Olifants River systems. Although data collection and analyses is still on-going, this study provides the first phylogenetic relationship of *Dactylogyrus-Enteromius* from Africa.
Coevolution between *Anacanthorus* spp. (Monogenea: Dactylogyridae) and Serrasalmidae (Characiformes)

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*Anacanthorus* is a Neotropical genus of gill monogeneans which are known to parasitize Characiform fishes, mainly the members of Serrasalmidae and some species of Characidae. The host-parasite associations between the serrasalmids and the *Anacanthorus* parasites have been known for a long time. The family Serrasalmidae is restricted to South America and currently consists of 95 species representing 16 genera, which are commonly divided in three main clades (i.e. pacu, true piranhas and a Myleus-like pacus clade). Further, the majority of serrasalmids (69) are distributed in Brazil. To date there are 73 valid species of *Anacanthorus*, and more than half of the species (37) were described from serrasalmids. According to literature, some species within this host-parasite system can be considered generalists (e.g. *A. jegui* and *A. mesocondylus*, which can be found in 6 different hosts), but most of them (22) are strictly specialists (parasitizing a single host species). Previous studies have demonstrated that monogeneans may provide good model for the study of host-parasite coevolution, therefore the main aim of this study is to reconstruct phylogenies for the hosts (serrasalmids) and their monogenean parasites (*Anacanthorus*) to investigate host-parasite coevolution and with the use of molecular analyses (based on the sequences of 28S rDNA) to define the systematic position of these monogeneans among other dactylogyrids. During our parasitological surveys carried out from 2015-2016 in different Brazilian river basins, eleven species of serrasalmids (including representatives of the three clades), were dissected. Molecular identification of fish species was performed using cytochrome b gene. Nineteen species (8 unidentified) of *Anacanthorus* were found and the phylogenetic relationships of some of these species were assessed using molecular sequences. Preliminary results of our analyses are presented.

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How three genera of Monopisthocotylean monogeneans and their fish hosts coevolved

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The study of processes that determine the coevolutionary relationships between closely associated species has recently attracted much attention. Within this realm, host–parasite relationships are specially interesting, because of their tight relationships, which are particularly remarkable in many monogenean and their hosts. We studied the cophylogeny of species of three genera of Monopisthocotylea: Lamellodiscus, Dactylogyrus and Gyrodactylus, and their fish hosts, in order to determine whether there is a coevolution signal, and establish whether there is a common pattern of coevolutionary events in these three systems. We applied two relatively new cophylogenetic methods: PÂCo as a distance-based and Jane as an event-based. PÂCo revealed significant coevolutionary signal in all the three systems, whereas Jane pointed to some similarities between them: the number of coespeciation events was low, the number of losses was always high and relatively constant, and failure to diverge events was always the commonest event. Duplications and host-switchings showed both the highest variability between and within the three systems, but were relatively numerous in most situations, especially the latter. However, some differences were noted: host-switching seems more frequent in Lamellodiscus and Gyrodactylus than in Dactylogyrus, in which duplications were more numerous. Contrary to the classical coevolutionary processes postulated in terrestrial host-lice systems shaped by coespeciation events, our results suggest that duplication, host-switching and failure to diverge were the three most important coevolutionary events that shaped the actual diversity and evolutionary relationships between these monogeneans and their fish hosts. This also implies that the habitual host-specificity showed in these three genera of monogeneans is mostly driven by evolutionary events other than coespeciation.
Phylogenetic relationships among *Diplectanum* spp. and host-parasite associations in sciaenid fishes from the Mediterranean

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*Diplectanum* (Monopisthocotylea: Diplectanidae) is a worldwide-distributed genus parasitic in the gills of Perciformes, predominantly sciaenids and serranids. There are 25 valid species of *Diplectanum*, 8 of them restricted to sciaenids of the Mediterranean region, and 44 considered incertae sedis. The taxonomic classification of species in this group is mostly based on the morphology of sclerotized structures. Several species have been recently placed in new combinations, molecular data does not exist for any valid species and their phylogenetic relationships remain unexplored. We investigate the phylogenetic relationship among *Diplectanum* spp. and the degree of specificity in *Diplectanum* - sciaenids interactions occurring in the Mediterranean to understand their diversity and evolutionary association with these hosts, some of which are target or potential species for Mediterranean aquaculture. We collected *Diplectanum* specimens from four sciaenid species, *Argyrosomus regius*, *Sciaena umbra*, *Umbrina canariensis* and *U. cirrosa* off two localities in the Western Mediterranean. We extracted DNA from a small piece of the anterior end of the body and mounted the rest of the specimen (hologenophore) for morphological identification. We recognised 5 distinct morphotypes out of 40 specimens on the grounds of their copulatory organs. Our sequences for the 28S rDNA region of *Diplectanum* from Mediterranean sciaenids cluster in a monophyletic clade. *Diplectanum* appeared sister to *Sinodiplectanotrematrea*, in a clade together with *Paradiplectanum*. We discuss the phylogenetic results and highlight the need for a molecular re-evaluation of the diversity of species in *Diplectanum*.

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Gill parasites (Monogenoidea) of Neotropical cichlids (Cichlidae) and their phylogenetic relationships based on molecular data

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Neotropical cichlids (Cichlidae: Perciformes) represent one of most diverse freshwater fish groups in the American continent with over 500 species distributed from northern Patagonia in Argentina up to northern Mexico. To date, a total of 47 species of dactylogyrids (Dactylogyridae) have been recorded in American cichlids allocated in the genera *Sciadicleithrum* (26 species), *Gussevia* (17 spp.), *Trinidadactus*, *Tucunarella*, and the recently erected *Parasciadicleithrum*, with one species each. *Gyrodactylus geophagensis* (Gyrodactylidae) represents the only gyrodactylid currently described infecting these fishes. Phylogenetic relationships of gill parasites of Middle-American cichlids were evaluated for the first time using partial sequences of the 28S rRNA gene. A new genus and species, *Parasciadicleithrum octofasciatum*, was erected based on its phylogenetic position, genetic divergences (25-26%) with respect to other species of *Sciadicleithrum* and on morphological characteristics of internal organs and body size dimensions. The monotypic *Parasciadicleithrum* represents a case of morphological convergence with *Sciadicleithrum* and can be confidently distinguished from the former genus on the basis of molecular data. Regarding the phylogenetic of other gill parasites of Neotropical cichlids, this study showed, on the one hand, that four species of *Sciadicleithrum* (*S. bravohollisae*, *S. meekii*, *S. mexicanum* and *S. splendidae*) represent a monophyletic group closely related to marine taxa occurring mostly in percomorphs across the Pacific Ocean. On the other hand, *P. octofasciatum* showed a close phylogenetic affinity to freshwater dactylogyrids of African cichlids (*Cichlidogyrus* and *Scutogyrus*). The phylogenetic position of *P. octofasciatum* points towards shared biogeographic patterns of Gondwanan vicariance for both, cichlids and this clade of freshwater dactylogyrids. The results obtained in this study suggest that the parasite community of Middle-American cichlids (including monogenoids) is composed by species resulting from both, association by descent and association by colonisation.
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Quadriacanthus species (Monogenea: Dactylogyridae) parasitizing catfishes (Teleostei: Siluriformes) in Eastern Africa: morphological and molecular characterization

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Diversity of Quadriacanthus species parasitizing Clarias gariepinus, Heterobranchus bidorsalis (Clariidae) and Bagrus docmak (Bagridae) was investigated in the Lake Turkana (Kenya) and Nile River Basin (Sudan). Both morphological and molecular approaches were used for description of the new species found, and for a critical review of the previously described Quadriacanthus spp. The interspecific relationships among Quadriacanthus spp. infecting the siluriform hosts inferred from ribosomal DNA sequences were investigated for the first time.

Seven species (including four new) of Quadriacanthus were identified: Quadriacanthus aegypticus El-Naggar & Serag, 1986, Quadriacanthus clariadis Paperna, 1961, Quadriacanthus fornicatus n. sp., Quadriacanthus pravus n. sp., and Quadriacanthus zuheiri n. sp. from Clarias gariepinus; Quadriacanthus mandibulatus n. sp. from Heterobranchus bidorsalis; and Quadriacanthus bagrae Paperna, 1979 from Bagrus docmak.

For both 18S-ITS1 and 28S rDNA regions, Q. clariadis from a clariid fish was found to be most closely related to Q. bagrae from a bagrid host. Quadriacanthus mandibulatus n. sp. was observed to be the most distant species from the others. The separation of Q. mandibulatus n. sp. from the other species corresponds with the different morphology of its copulatory tube. The observed interspecific genetic relationships among Quadriacanthus spp. from clariids and Q. bagrae from a bagrid host suggest a possible host-switching event in the evolutionary history of the genus.

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**Cichlidogyrus** spp. (Monogenea: Dactylogyridae) communities of Burundese and Congolese cichlids (Lake Tanganyika): taxonomic study and phylogenetic approach

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In 2013 and 2016, a total of 284 cichlid individuals belonging to 43 Tanganyikan species were collected from the Burundese and Congolese lakehores, and examined for gill ectoparasites. Morphological and molecular identification of the gill monogeneans were performed, and phylogenetic analyses were carried out by employing different molecular markers (28S rDNA, 18S rDNA and ITS-1). Phylogenetic trees were generated using maximum likelihood and Bayesian inference algorithms. The taxonomic study of more than 800 *Cichlidogyrus* specimens has demonstrated the occurrence of 37 monogenean species including 11 *Cichlidogyrus* species previously described on Tanganyikan cichlids and 14 species that are considered new for science. Our investigation represents the first parasitological examination of representatives belonging to the Tanganyikan cichlid genera *Aulonocranus*, *Callochromis*, *Cyphotilapia*, *Cyprichromis*, *Eretmodus*, *Tanganicodus*, and *Xenotilapia*. Congolese *Aulonocranus dewindti* hosted two new *Cichlidogyrus* species exhibiting similar morphotypes as the monogenean species reported recently on this host in Burundi. Likewise, a few *Cichlidogyrus* species have already been recognized on the gills of *Petrochromis* spp., but no formal description has been provided yet. Our study allowed us to record 7 new *Cichlidogyrus* species on Congolese specimens of *Petrochromis*. Furthermore, the gills of *Tylochromis polylepis* revealed the presence of *Cichlidogyrus* sp. “*T. polylepis 3*”, an undescribed species previously reported from this host. Finally, phylogenetic trees show that *Cichlidogyrus* species clustered following the phylogeny of their Tanganyikan cichlids, and mirror the affinities revealed by morphological characters between monogenean species and among their fish hosts.

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Checklist of monogenean parasites of freshwater fish from Iran with notes on their distribution

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Monogenea are a large group of platyhelminthes mainly found on skin or gills of fish. A checklist of the monogenean parasites in 52 native and introduced cyprinid species from 21 genera from Iran was generated. The checklist is based on published data since 1983. Available information on fish host, the sites of occurrence and geographical distribution is provided. The checklist contains 94 nominal species, from 7 genera of monogenean parasites. The parasites were mostly belonged to Sarmatian fauna and then to the Mesopotamian fauna.
Three new species of *Dactylogyrus* Diesing, 1850 (Monogenea: Dactylogyridae), from Moroccan Cyprinids based on morphological and molecular data

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With more than 2400 species belonging to 220 genera, Cyprinidae represent one of the most diversified family of freshwater fishes, present in all continents, except for Australia and South America. Cyprinid fishes are known to be parasitized by species belonging to *Dactylogyrus* Diesing, 1850, representing one of the most diversified helminth genera. In Morocco, Cyprinidae represent the most frequent freshwater fishes, with four genera: *Luciobarbus* Heckel, 1843, *Carasobarbus* Karaman, 1971, *Labeobarbus* Rüppel, 1835 and *Pterocapoeta* Günther, 1902 and 18 species (all endemic). The examination of the gill arches of *Luciobarbus rifensis*, *L. moulouyensis* and *L. maghrebensis* revealed the presence of new *Dactylogyrus* spp., which were identified using morphological characters (haptor, male copulatory organs and vagina) combined with molecular data.

Apart *D. falsiphallus* with singular morphology associated with species-specific molecular data, we found a cryptic species complex (*D. benhoussai*, *D. scorpius* and *D. varius*) exhibiting a similar morphology but with differences in molecular data (i.e. variability in partial 18S rDNA, ITS-1 and 28S rDNA sequences). In addition, one of these species (*D. varius*) present three distinct morphological forms (*D. varius* forma vulgaris, *D. varius* f. magnus and *D. varius* f. dromedaries), all of them with identical molecular data.

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Congeneric monogeneans in evolutionary and ecological studies

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*Dactylogyrus* are gill monogeneans infecting mainly cyprinid species. *Dactylogyrus* exhibit high species diversity with more than 960 nominal species described. A high species diversity is also reported at the level of host species suggesting that *Dactylogyrus* species are able to coexist on one host species. *Dactylogyrus* are aggregated at the level of hosts and microhabitats. They occupy the different microhabitat positions determining the specific niches. The coexistence of *Dactylogyrus* species on one host species is facilitated by high intraspecific aggregation, morphological adaptation (haptor morphology), microhabitat segregation and reproductive isolation among species. Many *Dactylogyrus* species are host specific with the different levels of specificity ranging from strict host specificity through congeneric host specificity until phylogenetic host specificity.

Due to host-parasite coevolution, host specific parasites may be considered as a possible tool to elucidate at least partially the historical biogeography of hosts, or alternatively to shed a light on the more recent contacts among host species. Especially *Dactylogyrus* parasitizing cyprinids as primarily freshwater fish may help to clarify some questions related to biogeographical distribution of these fish with limited capacity of dispersion. Concerning the cyprinids of Mediterranean area, three main hypotheses of their historical dispersion may explain their actual distribution. All suggest that the cyprinids originated in Asia and reached the Mediterranean peninsulas via (1) a northern route, (2) a southern route via landbridges connecting continents, or (3) a southern dispersion through the Mediterranean Sea during its so-called Lago Mare freshwater phase. *Dactylogyrus* species were used to infer potential historical contacts between Northwest African and Iberian cyprinid species and the origin of *Dactylogyrus* parasitizing these cyprinid species was investigated. The phylogenetic positions of *Dactylogyrus* of *Carasobarbus* (Torini lineage) and *Luciobarbus* (Barbini lineage) from Northwest Africa and *Dactylogyrus* of *Luciobarbus* from Spain were investigated. Even if molecular phylogenetic studies of cyprinids widely support the southern route of Iberian *Luciobarbus* dispersion, phylogenetic reconstruction of *Dactylogyrus* evolution indicates the northern route of *Dactylogyrus* dispersion to Northwest African *Luciobarbus* species. The multiple historical contacts between Iberian *Luciobarbus* and two lineages of Northwest African cyprinids (Torini and Barbini) with different origins and historical dispersions was shown resulting in two independent diversifications of *Dactylogyrus* in Iberian *Luciobarbus*.

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The TILAPIA project: Parasite host switching in the Congo Basin?

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The main goal of the TILAPIA project is to trace parasite host switching (spill-over and spill-back effects) between introduced Nile tilapia, Oreochromis niloticus and native cichlids in the Congo Basin. For this, three regions are sampled (Lower Congo, Middle Congo and the Bangwuelu-Mweru region, the latter draining into the Upper Congo River). Each region has a distinct native cichlid and parasite fauna, and in each region Nile tilapia has been introduced for aquaculture purposes. The parasites which we focus on are monogenean gill parasites belonging to the Dactylogyridae (Cichlidogyrus, Onchobdella and Scutogyrus) and Gyrodactylidae (Gyrodactylus). These parasites provide a suitable study system for tracing host switching. This was done by sampling the present-day fauna of native and introduced cichlids from fresh field samples. This was then compared with a pre-introduction baseline parasite fauna, reconstructed through specimens of the ichthyology collection of the Royal Museum for Central Africa. No host switches were found. However, in Bangwuelu-Mweru we discovered that Nile tilapia shares multiple monogenean species with native O. mweruensis. All of these shared parasite species were already present in the baseline fauna, so are not introduced by Nile tilapia. However, of Cichlidogyrus halli an endemic morphotype was found on O. mweruensis, only present where Nile tilapia is not, but where both species of fish do co-occur, the non-endemic C. halli was found exclusively on both fish. So, it seems that this endemic morphotype is replaced by the introduced non-endemic morphotype of C. halli. To confirm this and unravel the geographic origin of these individuals a phylogeographic analysis of host and parasite is in order. Outside of this, one new species of cichlid and 14 new species of gill parasites were discovered. Also, for the majority of parasite species their geographic and host-range was extended, such as for C. papernastrema, which was found for the first time since its’ description in 1969 and this time on cichlids from Oreochromini and Coptodini in addition to the already known host range within Tilapiini. Also, in the whole Congo Basin C. rognoni, a typical Nile tilapia parasite, was not found, which possibly is an effect of the enemy-release hypothesis.
Monogenean parasite communities in asexual-sexual *Carassius auratus* complex

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Based on Red Queen hypothesis, the dynamics of host-parasite co-evolutionary interactions is predicted. The most common host genotype is the target of parasite adaptation and should present the most commonly parasitized genotype. By this way, genetically homogenous asexual organisms may be disadvantaged when compared to the genetically variable sexual organisms. *Carassius auratus* (Cyprinidae) is the complex of forms or species combining asexual (gynogenetic) and sexual reproduction. The combination of two reproduction strategies represents a high level of invasive success of this species complex. The aims of this study were to investigate the temporal changes in the intensity of infection of monogenean parasites in the metazoan parasite communities in asexual (gynogenetic triploid form) and sexual diploid form of *C. auratus* complex. All fish specimens were genotyped for mtDNA and only the specimens determined as *C. gibelio* (the most common form of *C. auratus*) genotypes were selected for the analyses. A total of 7 species of *Dactylogyrus*, 3 species of *Gyrodactylus* and *Paradiplozoon homoion* were identified in *C. gibelio*. The maximum prevalence and the highest intensity of infection were found for the following monogenean species: *Dactylogyrus dulkeiti*, *Dactylogyrus anchoratus*, *Gyrodactylus sprostonae* in both forms of gibel carp. In overall comparison, our analyses revealed similar values for monogenean infection in gynogenetic females and sexual individuals. However, our study revealed a temporal variation in parasite load in both forms which could potentially be explained by the dynamics of host-parasite interactions predicted by Red Queen hypothesis.

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Trachemys scripta elegans as a vector of polystome dispersal

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With less than 200 species reported, the Polystomatidae includes 25 genera that parasitize semi aquatic vertebrates, such as the Australian lungfish, amphibians, freshwater turtles and the common hippopotamus. Polystomes are globally distributed within frogs and terrapins all over the world. Whereas they are recovered mostly from the bladder of amphibian hosts, they can be found in the bladder, pharyngeal cavity or conjunctival sacs of chelonian hosts. Although polystomes were regarded mostly as host and site specific, some species infecting freshwater turtles were reported from more than a single host species in outdoor turtle enclosures. This was the case of Polystomoides coronatum, which was found from the pharyngeal cavity of Trachemys scripta elegans and Apalone spinifera, and Neopolystoma orbiculare, which was found from the bladder of T. s. elegans, Apalone ferox and Chrysemys picta in the zoological aquarium of New-York in the 20’s. More recently, in the early 2000’s, Polystomoides oris and N. orbiculare, which were described and reported respectively from the pharyngeal cavity and the bladder of C. picta in American aquatic ecosystems, were both recorded from European turtles, namely Mauremys leprosa and Emys orbicularis in outdoor turtle enclosures of Southern France. Overall these results suggested that chelonian polystome could be not as strictly host specific as hypothesized. These outcomes were also supported in European natural environments through the study of the polystome diversity within populations of M. leprosa and E. orbicularis. Because most of the M. leprosa and E. orbicularis parasites were regarded as American turtle’s parasites and because those parasites were also found within feral populations of T. s. elegans, it was assumed that this turtle could serve as a vector for a variety of not strictly host-specific parasites. If that hypothesis is correct, those parasites should be found within T. s. elegans in American freshwater ecosystems. Fieldwork surveys that were recently performed in several states of the USA, namely Florida, Indiana, Kansas, Maine and North Carolina, allowed the collection of four distinct polystome species in T. s. elegans: (i) N. orbiculare from the bladder of T. s. elegans in Florida and North Carolina; (ii) Neopolystoma sp from the bladder of T. s. elegans in Indiana and Kansas; (iii) Polystomoides soredensis from the pharyngeal cavity of T. s. elegans in Indiana, North Carolina and Maine; (iv) Polystomoides scriptanus from the pharyngeal cavity of T. s. elegans in Florida and North Carolina. Because all these parasites are reported from American turtles as well as from native European turtles, T. s. elegans can be considered as a vector of polystome dispersal all over the world.
The maternal ancestry and monogenean distribution in hybrids of cyprinidae fish

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The intermediate status of hybrids regarding to parental species in morphology, ecology and trophic biology was often documented in nature and confirmed empirically. Concerning host-parasite associations, the host-parasite co-adaptation is interrupt due to interspecific hybridization which may lead to the changes of the structure of parasite communities in the hybrids when compared to parental species. In addition, it is hypothesized that the maternal ancestry of hybrids affects monogenean infection. The monogenean parasites with specific co-adaptation to their hosts represent the suitable model for the investigation of the effect of hybridization on host specificity and host-parasite interaction. The aim of this study was to investigate the effects of the hybridization and maternal origin of hybrids on the composition of monogenean parasite communities in the hybridizing system of common bream (Abramis brama) and silver bream (Blicca bjoerkna). Pure lines of both species were prepared as well as F1 hybrids with different maternal ancestry were obtained by the artificial cross-breeding. Fish were infected by monogenean parasites from source specimens of both parental species. Maternal origin of the hybrids was checked using cytochrome b sequences. Experimental work proceeded in two consecutive years (2015 and 2016). Hybrid offspring possess higher parasite species richness when compared to common bream and silver bream. Monogenean abundance was higher in parental species. Hybrids with the maternal origin of common bream were less parasitized than the other fish groups. Interruption of the system of co-adapted genes might lead to the higher monogenean richness and presence of the parental species-specific parasites of the both parental species in the hybrids. The species-specific composition in parasite communities of hybrid fish with both maternal origins was shifted to the parasites of the silver bream. This seems to suggest the different degree of the host-parasite co-adaptation in A. brama and B. bjoerkna.

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Space for your NOTES
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Diversity of monogeneans from freshwater fishes in Africa

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Knowledge of monogeneans is still incomplete in Africa and the present numbers of these parasites on/in fishes in the region are likely an underestimate, as a relatively few hosts have been examined. Today, more than twenty years since the last compilation (see Khalil and Polling, 1997), a total of 455 species of polyonchoineans (32 genera in three families) and seven species of oligonchoineans (three genera in two families) have been described. Given the high diversity of freshwater fish species in Africa (approximately 2950 spp.), monogenean species are therefore still poorly known. At the family level, the Dactylogyridae are represented currently by the greatest number of species (402 spp.) belonging to 26 genera: Afrocleidodiscus (3), Ancyrocephalus (s.l.) (4), Annulotrema (46), Archidiplectanum (1), Bagrobdella (3), Birgiellus (3), Bouixella (10), Characidotrema (10), Cichlidogyrus (103), Dactylogyrus (100), Dogielius (21), Enterogyrus (8), Eutrianchoratus (5), Heteronchocleidus (5), Heterotesia (1), Insulacleidus (3), Nanotrema (2), Onchobdella (8), Paraquadriacanthus (1), Protoancylodiscoides (6), Quadriacanthus (30), Schilbetrema (14), Schilbetrematoides (2), Scutogyrus (7), Synodontella (5), and Urogyrus (1). Only a single species of diplectanids, Diplectanum lacustre, has been reported from Lates niloticus. The viviparous gyrodactylids are the second largest family, with 52 species attributed to five genera, i.e. Afrogyrodactylus (4), Diplogyrodactylus (1), Gyrodactylus (37), Macrogyrodactylus (9), and Mormyrgyrodactylus (1). The only representatives of the Oligonchoinea are six species belonging to Paradiplozoon (5), Afrodiplozoon (1) (Diplozoidae) and one species of Heterobothrium (Diclidophoridae).
Identification of a new species of genus *Paradiplozoon* Achmerov, 1974 from the gills of *Sikukia gudgeri* (Smith, 1931) (Cyprinidae) from China

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The study presents an identification of a new species *Paradiplozoon yunnenensis* sp. n. of genus *Paradiplozoon* (Monogenean: Diplozoidae) from the gills of *Sikukia gudgeri*, Smith 1931 (Cyprinidae) which collected from Jinghong basin, a main tributary of the international Mekong-Lancang River. And the new species is the first diplozoid species to be discovered from *S. gudgeri*, synchronously the present study raised the number of species of *Paradiplozoon* recorded in China to 27. The new species is distinguished from the others valid species of genus *Paradiplozoon* by the combination of morphological and molecular features. The posterior part of the species is without tegumental ridges, folds, cup-shaped or a saucer-like shape. The absences of which is typical for genus *Paradiplozoon*. The sclerites of the anterior and posterior jaws are distinct slender and the sclerites of posterior jaw are divided into two parts as in other species of the genus. The anterior end of the median plate is thickening at marginal area, and with a column of round particles sclerites in the middle line. The new species is built generally from fragile sclerites, the trapeze spur is narrow not wider than median plate. Even molecular characterization based on variability of the second internal transcribed spacer (ITS 2) of the ribosomal DNA region provided the most definable support of the separation of the new species from the records. The ITS 2 sequences (accession number KT781100) were compared with previously 22 submitted sequences of *Paradiplozoon* in GenBank for estimating the homogeneity and genetic distance. New species *P. yunnanensis* was supported to be a valid species based on the molecular description, whereas supplementary molecular research of Chinese recorded diplozoid is strongly needed to provide suitable indicator for precluding some morphological resemblance among these species.
Diversity and distribution of species of *Gyrodactylus* von Nordmann, 1832 on African cichlids

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Parasites of the genus *Gyrodactylus* von Nordmann, 1832 are distributed worldwide. In the recent past, studies focused mainly on hosts which are important for the aquaculture, but recently the interest expanded which enabled deeper knowledge on the variety and phylogenetic traits of this highly diverse parasitic group. An increasing amount of available data on the distribution of these parasites allows revalidation regarding host specificity, preferences of site of infection and relationship within phylogenetic lineages.

The present study includes data from several surveys. The majority of the samples were collected during 2011-2015 in Burundi, Democratic Republic of Congo, South Africa, Sudan and Zimbabwe. Collected hosts were examined for the presence of *Gyrodactylus*. Infected host tribes and species belongs to: Ectodini - *Ophalmotilapia nasuta*; Eretmodini - *Eretmodomus marksmithii*; Haplochromini - *Astatotilapia burtoni, Ctenochromis horei, Pharyngochromis acuticeps, Pseudocrenilabrus philander*; Tropheini - *Simochromis diaphagma*, and Tilapiini - *Oreochromis niloticus, Coptodon rendalli, Tilapia sparrmanii, Tilapia zillii* and *Tilapia* sp.

Identification of parasite species was based on the morphometric analyses of attachment organ hard parts and molecular analyses of nuclear ribosomal DNA internal transcribed spacer (ITS) sequences. In total, 16 species of *Gyrodactylus* were recognized, *Gyrodactylus* sp. 1-8, *G. chitandiri, G. nyanzae, G. occupatus, G. parisellei, G. sturmbeueri, G. thyisi, G. thlapi* and *G. yacatli*. Maximum likelihood (ML) and Bayesian interference (BI) were used for the evaluation of phylogenetic relationships between parasitic species. Morphometric characteristic were visualized via Principal Component Analyses (PCA) what allows simplified interpretation of crucial patterns in attachment organ hard parts of specimens.

Results of the present study demonstrate a relationship between hard parts of attachment organ and molecular sequences, patterns in host/geographical distribution and also the importance of using integrative approach during parasite idnetification.

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Preliminary studies on the monogenean from fishes of Sansha in the South China Sea

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Sansha Islands in the South China Sea is a typical coral reef ecosystem, which has abundant fish resources. However, there is no monogenean record in this area. In order to investigate the fauna of monogeneans in the South China Sea, we visited Sansha Islands by taking the fishery resources survey ship of the South China Sea Fisheries Research Institute, Chinese Academy of Fishery Sciences during the spring and autumn in 2016. Systematic scientific research has been performed. In total, 95 species of marine fishes including 842 specimens were harvested and checked from the Sansha areas, and 49 species of fishes were infected by monogenean. Taxonomic identification of monogeneas is progressing now. At present, 21 monogenean species have been preliminary identified. Among those, a new genus including a new species is proposed and 8 species are new records in China. At the same time, 16 monogenean species were used to amplify partial DNA sequence of 28S rDNA and the taxonomic status of the new species, *Paracaesicola nanshaensis* n. g., n. sp., were explored based on the DNA sequence. This project figured out the monogenean constitution in the Sansha area and accumulated the original material for the construction of molecular barcode database of monogenean. New species and new records are listed as follows:

*Paracaesicola nanshaensis* n. g., n. sp.
*Capsala martinierei* Bosc, 1811 n. rec. in China
*Capsala onchidiocotyle* Setti, 1899 n. rec. in China
*Capsala katsuwoni* (Ishii, 1936) Whittington, 2007 n. rec. in China
*Neothoracocotyle acanthocybii* (Meserve,1938) Hargis, 1956 n. rec. in China
*Tetrancistrum yamagutii* Kritsky, 2007 n.rec. in China
*Diplectanum fusiformis* Bychowsky, 1957 n. rec. in China
*Pseudopisthogynopsis lepidocybii* Yamaguti, 1965 n. rec. in China
*Allopseudopisthogynoe constricta* Yamaguti, 1965 n. rec. in China
Monogenean parasites fauna from Iranian freshwater fishes

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This study is based on the previously published records and original data from 72 species of fish. Altogether 142 species of monogeneans belonging to 9 families, including 14 genera, i.e., Ancyrocephalus, Ancylodiscoides, Dactylogyrus, Diclobothrium, Diplozoon, Dogielius, Eudiplozoon, Gyrodactylus, Ligophorus, Mazocraes, Microcotyle, Nitzschia, Octomacrum and Paradiplozoon were recorded from Iranian freshwater fishes. Genus Dactylogyrus was the most abundant in monogeneans. The data suggest occurrence of some exotic monogenean species such as D. anchoratus on Iranian native fish which seems was due to introducing non-native fish to the country. Also, regarding the abundance and diversity of monogenean species found in Iranian fishes, it is suggested that there must be many unknown species which need further investigation.
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The balance between immune reactions and immune tolerance of fish skin and gills to monogenean colonization

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The ancestors of the first monogeneans colonizing fish skin and gills more than 500 MYA were probably more or less free-living flatworms. Some may previously have inhabited sediments or may have been ectocommensals of invertebrates (molluscs, arthropods, echinoderms). By invading fish surfaces these browsers expanded their forage area, increased their mating chances, spreading potential and secured an aerobic microhabitat with a stable supply of nutrition. This new strategy of colonization required a trade-off between living spaces with easily accessible nutrients and the protective armament of fish. The ancestors of the early hosts, various agnathans (hagfish, lampreys) and gnathostomes (sharks, rays, bony fish), possessed a rather effective immune response and the colonizers had to cope with a series of innate and adaptive immune reactions in gills and skin of the early fishes. Recent studies on mucosal immune responses in fish show that the monogenean microhabitat on the fish surface is covered by not only a dense layer of mucus and mucous cells but also a wide spectrum of immune cells and proteins with a potentially adverse effect on the colonizers. Agnathans possess various leukocytes including lymphocytes with variable lymphocyte receptors and they produce protective proteins based on leucine rich repeats (LRRs). In addition, a series of complement like proteins and anti-microbial proteins (AMPs) assist in keeping surface pathogens under control. Gnathostomes carry a wide spectrum of leukocytes, including several subsets of B- and T-lymphocytes. They produce at least three classes of immunoglobulins, IgM, IgD, IgT (Z) which interact with a series of innate reactants (lectins including MBL, SAA, CRP, SAP, proteases and complement factors C1, C2, C3, C4, C5, C6 and MAC elements). The response is coordinated by networks of cytokines (among others TNF-α, IFN-γ, TGF-β, IL-1, IL-2, IL-4/13, IL-5, IL-6, IL-8, IL-10, IL-12, IL-17, IL-22). Monogeneans have developed strategies to protect themselves against this armament in the fish skin. They may minimize the contact between parasite and host by using large anchors and clamps placing merely gentle pressures on gill and skin epithelia. Behavioural strategies include vivid movements from one attachment site to others whereby induced inflammatory reactions at specific foci in fish epithelia will have minimal effect on the straying monogenean. Feeding techniques of blood feeding polyopistocotyleans may produce minimal gill tissue disturbance. Other browsing parasites such as monopithocotyleans may cause more severe reactions. In some cases secure attachment of the monogenean is secured by circumventing encapsulating cellular reactions to improved attachment. In these cases hard structures (anchors) may be reduced but immuno-suppressive elements (proteases, lectins, fatty acids) and even release of symbiotic microorganism including viruses may be even more important in these situations. Finally immunological tolerance occur in fish. Immuno-tolerance may be induced by light continuous infections making use of an evolutionary inherited mechanism of the host to cope with surrounding microbiota.
Inhibition of proteolysis by the hematophagous *Eudiplozoon nipponicum*

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*Eudiplozoon nipponicum* (Monogenea: Diplozoidae) is an obligatory hematophagous parasite of the common carp (*Cyprinus carpio*). The blood degradation by this species involves a cascade of cysteine and aspartic proteases hypothetically regulated by protease inhibitors (e.g. cystatins and stefins). These inhibitory molecules are also known to have impact on immunomodulation of the host and the repair of its tissue damaged by the parasite. Our study aims to reveal the biological function of the stefin of *E. nipponicum* which was detected in the transcriptome and excretory-secretory products of adult individuals. We prepared recombinant form of *E. nipponicum* stefin (rEnStef) in *E. coli* BL21 bacterial strain using pET19b expression plasmid vector. By adoption of fluorometric assay we observed efficient inhibition of cysteine peptidases (cathepsins L and B from *E. nipponicum* and mouse cathepsin L) via its conserved papain-binding domain. Surprisingly legumain (asparaginyl endopeptidase) inhibition was detected probably due to legumain-binding domain, untypical for stefins. rEnStef blocked proteolytic degradation of hemoglobin mediated by cysteine peptidases in the excretory-secretory products, soluble protein extracts from *E. nipponicum* and by recombinant cathepsins L3 and B of *E. nipponicum*, which manifests its role in blood digestion. rEnStef any effect on the activation of complement in carp’s plasma or oxidative burst in full blood studied using luminol-enhanced chemiluminescence. Significant downregulation of selected cytokines (IL-1β, IL-8, TNF-α, IL-6 and IL-10) by LPS stimulated porcine alveolar macrophages and monocyte derived macrophages caused by rEnStef might indicate possible role of *E. nipponicum* stefin in immunomodulation of the host.

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EnKT1- Kunitz type inhibitor of Eudiplozoon nipponicum involved in the regulation of hemostasis

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Serine peptidases are a group of proteolytic enzymes that participate in many physiological processes including hemostasis in vertebrates. The activity of these enzymes can be regulated by parasite inhibitors belonging to the families of serpins and Kunitz domain proteins. Such parasite bioactive molecules may have medical potential as new therapeutics. Only few Kunitz type inhibitors (KTI) have been recently described from endoparasitic helminths. However, no peptidase inhibitors have been reported from monogeneans, so far.

In this study, we focused on KTI from the blood-feeding monogenean Eudiplozoon nipponicum occurring on the gills of the common carp Cyprinus carpio. Twenty one sequences of KTI were predicted in the transcriptome of adult worms. One of them, provisionally named EnKT1, shares amino acid sequence similarity with textilinin-1, an antihemorrhagic factor from the venom of the Australian brown snake Pseudonaja textilis. It was produced in recombinant form in Escherichia coli bacterial system, and further biochemically and functionally characterized. EnKT1 inhibited the peptidolytic activity of factor Xa of the coagulation cascade, but it displayed even higher inhibitory potential towards plasmin and plasma kallikrein, i.e., serine peptidases that participate in fibrinolysis and production of bradykinin. The anticoagulation activity of EnKT1 was confirmed by tromboelastography with human blood. RNA transcripts of EnKT1 were localised by RNA in situ hybridization inside the digestive (haematin) cells of the worms. Blood-feeding monogeneans, like other hematophagous parasites, need to overcome hemostasis by means of antihemostatic molecules to complete feeding. The probable role of EnKT1 in the biology of the parasite will be discussed.
A group of cathepsins L as predominant proteolytic enzymes of *Eudiplozoon nipponicum*

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Cathepsin L-like peptidases of the hematophagous monogenean *Eudiplozoon nipponicum* (family Diplozoidae) seem to play a key role in the digestion of host blood proteins. We have identified at least 13 different transcripts coding for cathepsins L in the transcriptome of adult worms. Two of them, the most transcribed ones, named *EnCL1* [GenBank: KP793605] and *EnCL3* [GenBank: KP793606], were heterologously expressed in *E. coli* bacterial (*rEnCL1b*) and *P. pastoris* yeast systems (*rEnCL1y* and *rEnCL3y*). While *rEnCL3y* was obtained as a relatively stable zymogen (proenzyme) and could be autoactivated at low pH, *rEnCL1y* undertook self-processing in yeast medium and was very unstable. Both recombinant enzymes exhibited substrate preferences characteristic for cathepsin L-like peptidases, and were able to hydrolize hemoglobin, albumin, type I collagen, IgG, and fibrinogen. Specific RNA probes and monospecific antibodies localized the transcripts/enzymes inside the vesicles of haematin cells of the digestive tract. Moreover, both *EnCL1* and *EnCL3* were also detected in the gut lumen of adult worms. These results strongly support the idea that both CL endopeptidases studied participate in processing of blood. Additionally, bioinformatic sequence analyses were performed in order to compare the other cathepsin L-like sequences found in the transcriptome of adult *E. nipponicum*. One of the sequences (named *EnCL2*) has a conspicuously divergent composition of the S2 subsite, and could be expected to efficiently cleave the substrates with proline in P2 position, including the repeated Gly-Pro-Xaa motifs occurring within the amino acid sequence of collagen. Thanks to this, *EnCL2* might participate in disruption of host tissues, e.g. of gill lamellae.
Cathepsins D and B: the role of aspartic and cysteine peptidases in the life of *Eudiplozoon nipponicum* (Monogenea)

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Processing of haemoglobin in blood-feeding helminths (e.g., *Schistosoma* spp., *Fasciola* spp., *Ancylostoma duodenale*) and mites (e.g. *Ixodes ricinus*) generally relies on an evolutionary conserved network of cysteine (cathepsins B, L, C, legumain) and aspartic peptidases (cathepsins D). Recently, it has been found that similar enzymes and mechanisms occur inside the digestive tract of blood-feeding monogeneans, where the dominant activity of cathepsin L-like peptidases seemed to be supported by cathepsins D and B. Therefore, this study aimed to reveal the biological function of the cathepsins D (*En*CDs) and cathepsin B (*En*CB) expressed by adult *E. nipponicum*.

Two types of cathepsins D (*r*EnCD2 and *r*EnCD3) were produced as recombinant enzymes in both *E. coli* bacterial and *P. pastoris* yeast expression systems. Their activity towards the cathepsin D-specific peptide substrate KPAEFnFRL was verified, and this activity was partially inhibited by pepstatin A (reversible aspartic peptidase inhibitor). Both enzymes were able to cleave haemoglobin. Monospecific antibody immunolocalized the *En*CD2 inside the vesicles of haematin (digestive) cells of the parasite, which suggests participation of the enzyme in haemoglobinolysis. Moreover, a direct proof of *En*CD2 in homogenates of adult worms was brought by a pulldown technique employing iBody3 (a polymer conjugate containing pepstatin A), SDS-PAGE and mass spectrometry analysis.

The recombinant cathepsin B (*r*EnCB) of adult *E. nipponicum* exhibited substrate preferences characteristic for cathepsin B-like peptidases. Possessing an occluding loop, the enzyme has also exopeptidase (peptidyl-dipeptidase) activity, as confirmed with Hippuryl-His-Leu-OH substrate and detection by fluorescamine. Although *r*EnCB was able to cleave components of blood (haemoglobin, albumin, fibrinogen, and to a lesser extent also immunoglobulins), immunohistochemistry localized *En*CB outside the gut - in vitelline cells of the parasite, suggesting a possible role in reproduction.

In conclusion, it seems that cathepsins D, but not cathepsin B, contribute to the complex intestinal proteolytic network in the blood-feeding monogenean *E. nipponicum*. 
Space for your NOTES
Diplozoons (Monogenea): what is the molecular base of their interaction with the host?

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The representatives from the class Monogenea are predominantly ectoparasites of fish. We adopted the experimental organisms - *Eudiplozoon nipponicum* (Diplozoidae, Heteronchoinea); a blood-feeding monogenean inhabiting gills of common carp (*Cyprinus carpio*).

In contrast to the other groups of platyhelminths, the identity of genes, the level of their expression and spectrum of functional protein molecules produced by these parasites are very limited.

The pyrosequencing methodology (454/Roche, MiSeq/HiSeq) enabled us to generate robust genomic/transcriptomic data and combined them with the results of comprehensive mass spectrometry analysis (Orbitrap-Elite MS System) of excretory-secretory products isolated from the adult worms. The pipelines of bioinformatic data processing were optimized and number of functional molecules was annotated. The reached platform currently represents significant source of information available for further experimental research; up to now at least six functional protein molecules (peptidases and their inhibitors) have been molecularly / biochemically / immunochemically characterized.

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De novo assembly of transcriptome of *Neobenedenia melleni* (Platyhelminthes: Monogenea) and data analysis

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*Neobenedenia melleni*, a widespread pathogen of many economically important marine fishes, has caused serious loss of marine cultured fish. However, the information on its molecular biology, particularly genomics is relatively scarce. In our study, a high-throughout method of transcriptome sequencing was performed to obtain the comprehensive information about this species.

The first *N. melleni* transcriptomic data was yielded and 75329780 clean reads were obtained by the Illumina paired-end sequencing technology. Totally 10998 non-redundant unigenes were *de novo* assembled with an average length of 759 bp. About 6772 unigenes have functional annotation after the similarity search against public databases. 6749 and 5633 unigenes were respectively annotated in the NCBI non-redundant database (nr) and Swiss-Prot protein database. 2678 unigenes were grouped into 25 Cluster of Orthologous Groups (COG) functional categories and 3794 unigenes were clustered into 231 different Kyoto Encyclopedia of Genes and Genomes (KEGG) pathways. Using the transcriptome dataset, proteases family sorting was performed and 384 simple sequence repeats (SSR) were identified. Comparative analysis of other sequenced flatworms was performed. *N. melleni* may be lack of the ability to synthesize fatty acids, but has the expression of *vasa* and *piwi*. The pathways related to secretion of bio-adhesive materials were identified. The transcriptomic data provides the genomic basis of the species that is a valuable resource for further study including the phylogenetic analysis in monogenean and the novel gene discovery.
**Eudiplozoon nipponicum** (Polyopisthocotylea: Diplozoidae): transcriptome and secretome analyses of hematophagous fish parasite

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We performed the complex analyses of transcriptome and secretome of monogenean representative *Eudiplozoon nipponicum* Goto, 1891 (Polyopisthocotylea: Diplozoidae).

Two sequencing methods (454/Roche and Illumina MiSeq platform) and mass spectrometry analysis (HPLC MS/MS) were used. Total RNA from a few *E. nipponicum* adults, in the form of cDNA, was sequenced and Illumina and 454/Roche raw reads were processed and assembled separately using specific bioinformatics tools. Based on statistical evaluation, selected assembly variants were pooled and filtered, in order to get high quality transcriptomics sequences for further annotation, into final contaminant-free 37,062 transcripts with corresponding translated proteins. Totally 19,539 (52.7 %) transcripts were homologous to some record in used public sequence databases (BLASTn and BLASTp algorithms were used, E-value cut-off: 1e-5) and 18,556 (50.1 %) of them were linked with sequences related to organisms only in phylum Platyhelminthes deposited in UniProtKB/TrEMBL database. All transcripts were deeply annotated by established approach.

Excretory-secretory products (ESP) – protein compounds present in parasite secretome, were obtained from ~100 living adults. HPLC MS/MS analysis of peptide mixture were done using RSLCnano System connected to Orbitrap Elite Hybrid spectrometer after tryptic digestion. The analysis of the mass spectrometric raw data was carried out using the Proteome Discoverer software tool with Mascot search engine utilisation and finally, 1,033 transcripts were identified in secretome.

The research was financially supported by Masaryk University, Brno (MUNI/A/1362/2016), Czech Science Foundation (GBP505/12/G112, P506/12/1258), Charles University, Prague (PRVOUK P41, UNCE 204017 and SVV 260432/2017) and the European Regional Development Fund (CZ.1·05/1·1·00/02·0068)
Genomic insights into the evolution of *Gyrodactylus salaris* and related species

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*Gyrodactylus salaris*, the notorious pathogen of European salmonids, is arguably the best studied gyrodactylid monogenean on the planet. Despite decades of research, however, the complex evolutionary history of the species is still surprisingly poorly understood. Existing molecular markers fail to unambiguously resolve the phylogenetic relationships between parasite lineages infecting host species as diverse as Atlantic salmon, Arctic charr, rainbow trout and European grayling. Especially the lack of phylogenetic resolution with respect to isolates from the latter host species (*Gyrodactylus thymalli*) fuels debates about the taxonomic status of these lineages, despite some evidence for host-specificity mediated pre-zygotic reproductive isolation. Based on whole-genome sequencing (WGS) data obtained for multiple lineages of *G. salaris* and *G. thymalli*, we have inferred fully resolved phylogenetic relationships that are incongruent with previous results including mitochondrial markers. The dataset includes isolates obtained from five salmonid host species and we used the population-level data to scan the gyrodactylid genomes for regions of increased divergence, i.e. putative candidates regions, sometimes called ‘genomic islands of speciation’, and key genes, under positive selection between the evolutionary lineages found on different host species.
Space for your NOTES
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Spermiogenesis and spermatozoon ultrastructure in basal polyopisthocotylean monogeneans, Hexabothriidae and Chimaericolidae, and significance for the phylogeny of the Monogenea

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Sperm ultrastructure is an important source of morphological characters, useful for understanding the phylogeny of the Neodermata. Although sperm ultrastructure has been described in many polyopisthocotyleans, no study was available for two basal branches, the Chimaericolidea and Dicybothriidea. We describe here spermiogenesis and mature sperm ultrastructure in Chimaericola leptogaster (Chimaericolidae) and Rajonchocotyle emarginata (Hexabothriidae), and sperm structure in Callorhynchocotyle callorhynchi (Hexabothriidae). Spermiogenesis in C. leptogaster and R. emarginata shows the usual pattern seen in most Polyopisthocotylea (and other Neodermata), with a typical zone of differentiation and proximo-distal fusion of the axonemes. In all three species, the structure of the spermatozoon is biflagellate, with two incorporated trepaxonematan 9+"1" axonemes and a posterior nucleus. However, interesting and unexpected structures were also seen. An alleged synapomorphy of the Polyopisthocotylea is the presence of lateral peripheral microtubules, which produces sections of spermatozoa with a continuous row of microtubules in the nuclear region. The mature sperm of C. leptogaster has a posterior part with a single axoneme, and the part with a nucleus is devoid of the continuous row of microtubules. The spermatozoon of R. emarginata has an anterior region with ornamentation on the membrane and a very wide nucleus in the posterior part; lateral microtubules are absent. The spermatozoon of C. callorhynchi was the most surprising, with transverse sections that mimic typical sections of digenean spermatozoa, i.e. with only dorsal and ventral microtubules. In addition, the spermatozoon of C. callorhynchi has a posterior “undulating membrane” containing a single axoneme and the nucleus. Implications for phylogeny and the definition of synapomorphies in the Neodermata are discussed. The major conclusion is that the synapomorphy “lateral microtubules in the principal region of the spermatozoon” does not define the Polyopisthocotylea as a whole but is restricted to the terminal branch Mazocraeidea.
Unique ultrastructural characteristics indicating a chimaera-like organisation for chimaericolid monogeneans

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Chimaericolids are basal polyopisthocotylean monogeneans associated with holocephalan fishes, a relict of the late Silurian era. Our detailed ultrastructural studies of a member of this family, Chimaericola leptogaster from Chimaera montrougesi, are intended to shed light on their phylogenetic relationships, origin and coevolutionary development with their host group. Several unique ultrastructural features have been revealed in the uterus, where three different regions are apparent. In the proximal uterus, the lining forms a so-called ‘single-layered multi-rowed cellular epithelium’, which includes two types of cells, tall and short, both lying on the basement membrane. Although known from some other bilaterian groups, this is the first record of this type of epithelium in the Neodermata. The lining of the middle uterine region comprises a single regular layer of columnar glandular epithelial cells, which produce numerous secretory granules; the presence of such glands in the uterus is an unusual feature for a monogenean, having previously been described only for basal orders of the Cestoda, i.e. the Gyrocotylidea, Caryophyllidea and Spathebothriidea. The epithelium of the distal uterus contains three areas of tall, single-layered columnar epithelium interspersed by three areas of flattened epithelium. This arrangement is quite different from that reported for other monogeneans and, indeed, other neodermatan groups. The vaginal configuration of this species is also unique, as there is a connection between each vagina and the intestine, a feature also known from turbellarians. A spine-like surface structure and three kinds of unicellular glands in the middle vaginal region are also unusual characteristics for a neodermatan vagina. Features remarkable for a monogenean also occur in the caecal epithelium, where digestive cells and a connecting syncytium are arranged alternately, and the nuclear regions of both cell types are sunk beneath the general caecal epithelial level. Furthermore, a concave depression on the apical margin of the digestive cells is covered by a lamellate bubble formed by thin projections emanating from the connecting syncytium. The haptor of C. leptogaster too has distinctive traits in comparison with all other monogeneans. Its clamps are simple and apparently archaic in nature, possessing a unique arrangement of their single median and paired lateral sclerites which differ in location, ultrastructure, chemical composition and origin. The unique structure of six previously unrecorded spine-like structures in these clamps appear to resemble structures found in proseriate turbellarians and may represent another piece of evidence for an ancient origin for chimaericolids. The unique ultrastructural characteristics of this group may represent evolutionary indicators for a polyopisthocotylean ancestor of the Neodermata. In one instance, the apparently unique presence of a single-layered multi-rowed cellular epithelium in the uterus requires investigation in relation to non-classical, molecular phylogenies of the Bilateria. Until more is known of the ultrastructure of related polyopisthocotyleans, the presence in chimaericolids of characteristics found in apparently phylogenetically distant groups do signal a chimaera-like status for this group.

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Neodiplectanotrema helicoleni from different geographical
Atlantic and Mediterranean locations: the attachment of an
endoparasitic monogenean

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A total of 71 blackbelly rosefish, Helicolenus dactylopterus (Delaroche, 1809), have been
analysed for parasites, 35 from the Spanish Mediterranean [12–20.1 cm in total length (TL);
27.16–126.1 g in weight (W); May 2017], 30 from North-eastern Atlantic [32.4–28.1 cm in
TL; 507–309 g in W; May 2017] and six from Patagonia, Argentina [25.9–32.5 cm in TL;
278–560 g in W; March 2007]. Gills, oesophagus and stomach were examined looking for
monogeneans, specifically for Neodiplectanotrema spp. For morphological identification,
some specimens were fixed in 70% ethanol, stained with iron acetic carmine, dehydrated in
an ethanol series, cleared in dimethyl phthalate and mounted on permanent slides with
Canada balsam. Additionally, some specimens attached in the fish tissue, were fixed in 4%
formalin for histological and Scanning Electron Microscope analyses, all the specimens have
been identified morphologically as Neodiplectanotrema helicoleni Gerasev, Gaevskaja &
Kovaleva, 1987, a species previously described in the blackbelly rosefish from the African
Atlantic coast. In Mediterranean fish, only 20% of the fish were infected with N. helicoleni
(mean intensity [MI] = 1.7±1.1). Parasite loads in Atlantic localities were much higher; all
North-eastern Atlantic fish were infected, with MI = 72.2±46.5, while 83% of the fish from
Patagonia were infected (MI = 26.6±39.1). These findings, together with previous records
in other species of Helicolenus from the American Atlantic Ocean, show the wide
distribution of this parasite in the Atlantic Basin. All the monogeneans were found attached
by the haptor at the middle section of the oesophagus length, being mostly concentrated in
depressions of the mucosa. Adult and post-larvae were found in the same habitat with
different degree of development, however, mature specimens were apparently smaller in
Mediterranean fish. No apparent damage or remarkable inflammation was detected in the
tissue where the haptors were attached, despite the high parasite loads detected. The
apparently minor adaptations to endoparasitic life of a monogenean with ectoparasite
structure are discussed.

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Ultrastructural aspects of surface and attachment clamps of *Paradiplozoon homoion* (Monogena, Diplozoidae)

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*Paradiplozoon homoion* (Bychowsky et Nagibina, 1959) is a representative of blood-feeding ectoparasites from family Diplozoidae with specific life-cycle, parasitizing mainly on gills of cyprinid fish, where they can cause mechanical damage to the gill filaments, initiating the development of secondary infections (bacterial, mycotic) and anemia. During the diplozoon life cycle two larvae, the diporpae, pair and subsequently fuse permanently producing the typical X-shaped diplozoon body arrangement, this is a unique character for Diplozoidae.

The samples of *P. homoion* were collected from the gills of bleak (*Alburnus alburnus*), which were caught in the littoral zone of the Mušov lowland reservoir. The fish gills were extracted and checked for the presence of all diplozoid ontogenetic stages. Parasites intended for transmission electron microscopy (TEM) were washed in freshwater to remove any remaining mucus and then fixed in 2% osmium tetroxide and dehydrated through an ascending acetone series. The dehydrated samples were embedded in Spurr resin.

In order to obtain a better orientation in basic diplozoon morphology several semi-thin sections (0.5 µm) of whole worm bodies were cut using a Leica EM UC6i. Sections were stained with toluidine blue. Ultrathin sections were contrasted with uranyl acetate, followed by citrate, and examined using a JEOL JEM-1010 TEM.

The presentation provides detailed ultrastructural data on the surface and haptoral attachment clamps of *P. homoion* and brings new insights into the ultrastructure of Diplozoidae. Our findings support the hypothesis that the structure of particular neodermal compartments is similar within the Platyhelminthes.

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Monogenean parasites of endemic cyprinid species from the selected areas of Mediterranean

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The Mediterranean area is distinguished from most of the other parts of Europe in occurrence of many unique endemic fish species (especially cyprinids). Studies focusing on the investigation of parasitofauna of endemic freshwater fishes are rather exceptional. In the present study, we analysed the parasite communities in endemic cyprinid species of a large part of the Balkan Peninsula. A total of 577 specimens belonging to 57 cyprinid species were sampled during 2014 – 2015 in selected localities in Albania, Bosnia and Herzegovina and Greece and examined for the presence of metazoan parasites. We analysed the infection level of parasites and the diversity of parasite communities. In addition, we focussed on host specificity and endemism of parasites. The highest species diversity was reported within Monogenea – a total of 103 species was recorded. We reported 38 species of Dactylogyrus (from which 11 were identified as likely new for science), 54 species of Gyrodactylus (from which 25 are likely new for science), and Paradiplozoon homoion and Paradiplozoon megan. Number of monogenean parasite species per endemic cyprinid species ranged from 1 to 16. Diversity of parasite communities was analysed at the levels of metacommunity and infracommunity using indices of diversity. Metacommunities of parasites on Luciobarbus graecus and Luciobarbus albanicus reached the highest dominance. The parasite metacommunity with the highest species diversity was found for Telestes montenigrinus, and the metacommunities with the highest species evenness were found for Rutilus panosi and Tropidophoxinellus spartiaticus. The highest mean infracommunity parasite diversity was found for Alburnus neretvae, Barbus rebeli and Barbus prespensis. Using multidimensional scaling on the similarity of parasite communities expressed by the indices of similarity, the parasite communities of cyprinids from Greece and Albania were similar, whilst parasite communities from Bosnia and Herzegovina were different from both of them. The similarity of parasite metacommunities of congeneric hosts belonging to genera Squalius and Barbus and the high dissimilarity of parasite metacommunities belonging to genera Alburnus, Rutilus and Telestes were found. Some monogeneans reported in endemic cyprinid species are commonly distributed in Europe, however, the majority of Dactylogyrus and Gyrodactylus species found in endemic cyprinid species are characterised by a high degree of host specificity and include taxa endemic to the Mediterranean area. Of all collected parasite species 66 were classified as genus specific, 40 as species specific and 63 as endemic. The majority of genus specific parasites occurred mostly on Barbus, the species specific parasites mostly on Alburnus neretvae, Pachychilon pictum and Aulopyge huegelii. The most generalist parasite species and nonendemic species were found on Telestes montenigrinus, Rutilus ohridanus and Squalius vardarenis.

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Can worms tell us who their hosts are?

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The African Clawed Frog, \textit{Xenopus laevis} Daudin 1802 (Anura: Pipidae), is the world's third most globally widespread amphibian, with established invasive populations on four continents. In its native range in southern Africa this frog is equally widespread. However, the parasitic fauna of \textit{X. laevis} is far from ordinary. Over 25 parasite genera from seven invertebrate groups have been associated with \textit{X. laevis} in its native range. In addition, many of these parasites are the sole representatives of their respective genera, hinting at the taxonomic isolation of the \textit{X. laevis}' parasitic community. An example of this taxonomic isolation is \textit{X. laevis}' two monogenean trematodes, \textit{Protopolystoma xenopodis} Price 1943 (Monogenea: Polystomatidae) from the excretory bladder and \textit{Gyrdicotylus gallieni} Vercammen-Grandjean 1960 (Monogenea: Gyrodactylidae) from the buccal cavity and pharynx. Both of these worms exhibit unique adaptations to their host that are absent in their closest relatives. However, this is not the complete picture of these species' coevolutionary history with \textit{X. laevis}. There are indications of considerable genetic variation between the different populations of \textit{X. laevis} in southern Africa and this influences the radiation of their parasites. This study aims to address the question of whether there are significant differences, both genetic and morphological, between the two monogenean parasites from hosts of different regions. In turn, this tells us something about the taxonomic status of their hosts. By studying the monogeneans, a clearer picture will emerge of the distinctness of different \textit{X. laevis} populations in southern Africa and whether genetic distinctness actually amounts to ecological isolation. This is not only applicable to native southern African \textit{X. laevis} – both these monogeneans have followed their host out of Africa and are to be found in some of the feral populations on other continents.
How many species? How many forms? Hidden biodiversity of merizocotyline monogeneans in batoids (Chondrichthyes: Elasmobranchii) from the coasts of the Argentine Sea

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In order to study monogenean diversity from elasmobranch hosts in the Argentine Sea, olfactory sacs of 310 batoids, corresponding to six species of Arhynchobatidae and one species of Myliobatidae were examined. Representatives of the genus *Empruthotrema* spp. were found in seven host species. Haptor morphology of all worms was similar to that of *E. raiae*, a species known from European batoids. However, when the male copulatory organs (MCO) were thoroughly studied under SEM, after digestion with Proteinase K, three groups were identified according to the size and morphology. A first group with MCO short and curved, parasitizing *Psammobatis normani*, *P. rudis*, *P. bergi* and *Atlantoraja castelnaui*; a second group with MCO long and slightly curved, from *Sympterygia bonapartii* and *S. acuta*; and a third group with MCO long, straight and blunt from *Myliobatis goodei*. In order to corroborate these results by genetic studies, sequences of 28S ribosomal DNA were obtained from specimens of each host species. Generated sequenced were compared for similarities against sequences retrieved from Genbank and phylogenetic analysis were performed including *E. quindecima* and *E. dasyatidis*. Genetic and morphological results agreed in determining the same groupings of specimens from each host species, whereas species retrieved from Genbank clustered in a different clade, providing evidences on the existence of 3 putative new species of *Empruthotrema* in the Argentine Sea. Co-evolutionary processes giving rise to the obtained phylogenies and the effect of environmental factors (depth, temperature, salinity) on them are discussed.
Highly host-specific gill Monogeneans (*Pseudorhabdosynochus* spp., Diplectanidae) on groupers (Perciformes) in the Mediterranean Sea: What do we know?

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Gills of groupers harbour many diplectanid Monogeneans, among which more than 80 species are assigned to the genus *Pseudorhabdosynochus* Yamaguti, 1958. These species are known to be highly host-specific. The comparative morphology of the sclerotized vagina is one of the principal characters for their identification. Groupers in the Mediterranean Sea are no exception. Before our studies, a few species of *Pseudorhabdosynochus* were known from three host species, including *P. beverleyburtonae* and *P. riouxi* on *Mycteroperca marginata*, *P. bouaini*, *P. enitsuji*, *P. dolicocolpos*, *P. sosia* and *P. sinediscus* on *M. costae*, and *P. americanus* and *P. hargisi* on *Epinephelus aeneus*. We studied newly collected material from Tunisia and Libya and specimens from Museum collections. Monogenean COI sequences were used to complement the morphological analysis of parasites. Fish COI sequences were used to confirm the morphological identification of hosts. Three additional species of *Pseudorhabdosynochus* from *M. rubra*, *M. marginata*, and *Hyporthodus haifensis* were added to the list: respectively, *P. regius* Chaabane et al., 2015, *P. hayet* Chaabane et al., 2016, *P. oliveri* Chaabane et al., 2016, and *P. sulamericanus* Santos et al., 2004. The latter was previously described from two hosts off the American coast of the Atlantic Ocean, making this finding in the Mediterranean unexpected. Two groups of *Pseudorhabdosynochus* species were proposed for parasites from *Mycteroperca* spp., the “beverleyburtonae group” and the “riouxi group”.

The use of *Gyrodactylus* parasites in studying host migration

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Most studies on host-parasite co-speciation focus on the phylogenetic level, examining co-evolutionary relationships between species pairs. However, speciation is a continuous process and processes acting at the population-level influence patterns of co-speciation. Phylogeography offers the tools to unravel co-evolutionary relationships in different host and parasite populations across their shared distribution range. In the case of host-specific parasites with a vertical transmission mode or low dispersal abilities, this may lead to a strong concordance among the phylogeographic patterns of distant host and parasite taxa (co-evolution hypothesis). In this scenario, parasite genetic analyses can shed more light on host evolution on the condition that these parasites have a short generation time and a small effective population size. This is the so-called ‘magnifying glass’ hypothesis. Here we test this hypothesis in two fish parasite models. We focus on *Gyrodactylus alexanderi* from the three-spined stickleback (*Gasterosteus aculeatus*, L.) from British Columbia, Alaska and Japan, and on *Gyrodactylus gondae* from European sand goby populations (*Pomatoschistus minutus* (Pallas, 1770)). We sequenced the complete ITS rDNA region and a partial cox2 mitochondrial DNA (mtDNA) fragment and compared the constructed network with the one obtained for the host based on partial cyt b mtDNA.
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Integrating biology with management: ongoing understanding of approaches to controlling monogeneans in aquaculture

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Monogeneans are problematic parasites in aquaculture. Their life-cycle is simple and can theoretically be interrupted by two strategically timed effective treatments, but impacts continue in aquaculture industries worldwide. Information is lacking for most host-parasite systems in several key areas: methods for assessing parasite abundance; effects of environmental variables on egg hatching rhythms, time to maturity and fecundity; optimising treatment; assessing treatment outcomes; and integrating information and providing a framework for adjusting the approach to control through experience. Developing management approaches requires activities to be integrated with animal husbandry practices that improve health. Optimising management efficiency is important for the increasing role aquaculture plays in world seafood supply.
Fisheries targets in Lake Tanganyika under the magnifying glass: a look at their monogeneans

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Lake Tanganyika (LT) is one of the African biodiversity hotspots and its fishes are an important food source for local people. While most previous studies have focused on the lake’s spectacular largely endemic cichlid fauna, knowledge about the economically most important species is still poor. The fisheries effort is concentrated mainly on the lake’s pelagic zone, with two clupeids (Limnothrissa miodon, Stolothrissa tanganicae) and four latid species (Lates angustifrons, L. mariae, L. microlepis, L. stappersii) as dominant targets.

We examined the abovementioned hosts for the presence of monogeneans to explore 1) which monogeneans infect clupeids and latids in LT, 2) freshwater parasite versus marine host origin and 3) seasonal, environmental and geographical gradients in intraspecific morphological variation and infection parameters.

Samples originated from 25 localities in LT including all three historical subbasins. Monogenean species identification and delineation was based on the sclerotised structures, combined with molecular characterisation using nuclear and mitochondrial markers. The effect of host species, environmental, geographic and fixative on monogenean morphological intraspecific variability was tested.

In total, 800 fish specimens were examined and infection of three different monogenean species was detected. While clupeids were infected by typically freshwater monogeneans assigned to the new genus called Kapentagyrus, three of the four latid species were parasitized by a representative of the marine genus Diplectanum. Phenomenon of lower monogenean host-specificity in pelagic zone of LT compared to littoral habitat was therefore enhanced by two monogenean genera from non-cichlid fishes. Seasonality in parasite prevalence on clupeids related to host species was observed. Moreover, morphological analyses of K. tanganicae indicated that phenotypic variation depended on host species. No shared infection trends of collected monogenean species in last 80 were detected. The parasite population structure inferred from part of the COI gene shows no north-south gradient. For future studies, the phylogeography of these parasites can reflect historical events that are too recent to be inferred from host genetics.
Co-introduction of ancyrocephalid monogeneans on their invasive host, the largemouth bass, Micropterus salmoides (Lacepède, 1802) in South Africa

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Largemouth bass, Micropterus salmoides (Lacepède, 1802) were sampled from four provinces (Eastern Cape, EC; North West, NWP; KwaZulu-Natal, KZN; Western Cape, WC) in South Africa to assess for parasite diversity and community composition. Morphological evaluation of the sampled parasite specimens provided evidence for the first record of five monogeneans from the family Ancyrocephalidae: Clavunculus bursatus (Mueller, 1963), Onchocleidus dispar (Mueller, 1936), Onchocleidus furcatus (Mueller, 1937), Onchocleidus principalis (Mizelle, 1936) and Syncleithrium fusiformis (Mueller, 1934) from the African continent. Community composition differed between localities. Clavunculus bursatus were only sampled from the EC and KZN, O. dispar and O. principalis were only sampled from the EC and WC, O. furcatus was only sampled from the NWP and KZN populations and S. fusiformis only from KZN. Prevalence was 100% at all sites, except in the WC where one of the two localities had a prevalence of 93%. Data from this study support the enemy release hypothesis as many of the parasites reported from the native range of M. salmoides were not collected.
Studies on the biology of *Cichlidogyrus sclerosus* and the seasonal dynamics of monogenean populations and communities on three strains of tilapia in net cages

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Tilapias are widely cultured in the world, and monogeneans are common parasitic pathogens of these fishes in aquaculture. The study on the ecology of monogeneans included the indoor experiments focusing on the egg production and hatchment, larval infection and development mainly related to temperature, and the seasonal dynamics of monogenean communities and populations on three strains of host in the outdoor experimental cages.

The indoor experiments revealed that the larva of *Cichlidogyrus sclerosus* on the host *Oreochromis niloticus* reached sexual maturity in vivo on day 12 and 6 post infection at 20 °C and 25°C respectively. The egg-laying peaks observed on day 23-26, 13 ± 3 eggs/day at 20°C, and 14-18 days post infection, 16 ± 2 eggs/day at temperature 25°C. Eggs production at 30 °C was highest among three temperatures. Results from herd infection were consistent with those by individual infection. Salinity of 1 ppt can promote in vitro eggs production. Moderate low temperature (at 20°C) can prolong the lifespan of parasites in vitro and delay the egg laying. As for the egg hatchment, the first larvae was observed on 5-6, 2, 3 days, with hatching peak at 7, 3, 4 days at 20°C, 25°C, 30°C separately. Eggs could tolerate low temperature to a certain extent although hatchment rate decreased after low temperature treatment.

The outdoor experiments on the seasonal dynamics of monogenean populations and communities on three strains of tilapias caged in the west lake of south campus of SYSU revealed that *C. sclerosus* on the three strains of host, had significant seasonal population variation, showing as higher mean intensities and abundances in winter months than those of summer months. It usually became dominant species of gill parasite communities; The *C. tilapia* had higher prevalences, abundances and mean intensities on the three strains of tilapias in summer seasons, and generally became dominant species of gill parasites communities in months between summer and autumn. *Scutogyrus longicornis* had low and less fluctuant prevalences, mean intensities throughout the whole year, nearly disappeared in summer months. (3) There were not strong interspecific and intraspecific competitions detected, neither found preferences of their distribution on the gills in relation to both left/right sides and front/back gill plates. However, it has been found that *C. sclerosus* and *S. longicornis* have preference to the proximal position of gill filaments, while *Cichlidogyrus tilapia* tends to choose the distal portion of the gill filaments.
Minimum inhibitory concentration (MIC-test) of formalin for *Dactylogyrus minutus* Kulwièc, 1927 (Monogenea: Dactylogyridae): *in vitro* test

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The formaldehyde has been widely used in its diluted form as an alternative to malachite green in the treatment of parasites and fungi. It has been used as anti-fungal and against parasites, especially protozoans and monogeneans from the gills and body surface of fish and immature amphibians like tadpoles. The minimum inhibitory concentration (MIC-test) of formalin *in vitro* that controls the *Dactylogyrus minutus* Kulwièc, 1927 of *Cyprinus carpio* var. *koi* was determined in this study. Samples of carp var. *koi* (1.73±0.25 g mean weight and 4.7±0.46 cm total length) were obtained in September 2016 from a fish farm located in Biguaçu, Santa Catarina, Brazil and transported live to Aquatic Organisms Health Laboratory. Eight concentrations of formalin were tested: 50, 75, 100, 125, 150, 175, 200, 250 ppm. In addition, two controls were prepared, one with water from the recirculation system and another with distilled water. Gill filaments from parasitized gill arches were collected to obtain the adult specimens of *D. minutus*. Parasites were placed into 6-well flat plates for cell culture in a volume of 3 ml per well, where each set of parasitized filaments received a formalin concentration. Mortality observations were recorded every 5 minutes at the highest concentrations or 10 minutes at the lowest concentrations using a stereomicroscope. The parasites were considered dead, when the absence of movements to the stimulus with needle was verified, as well as typical characteristics of mortality as the wrinkling of the body. Data were submitted to analysis of variance (ANOVA) and Tukey’s test was used to compare means (p<0.05). The concentration of 200 ppm was the most lethal to *D. minutus*, killing all the parasites in 16 minutes. However, for 250, 175, 125 and 75 ppm there was no statistical difference when compared to the two most lethal concentrations. The concentration of 100 ppm took longer to act on all monogeneans, however the time (47 minutes) was not higher than the time normally recommended in fish treatment. As some protocols indicate baths of at least 1 hour or even continuous baths, with this study, we could guarantee the real efficacy of the formalin directly in the parasite by monitoring the time and concentration.

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Space for your NOTES
Exotic monogenoids and Indian guidelines for the import of ornamental fish: review and perspective

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Aquarium trade is a potential pathway for the global translocation of exotic aquarium fish and their parasites, especially the parasitic monogenoids. The Government of India has recently enacted the ‘Guidelines for the import of ornamental fishes into India’ to effectively control the ornamental fish and associated disease introduction into the country. The guidelines appear, prima facie, stringent as they impose a number of import procedures and requirements, including pre-quarantine, quarantine and post-quarantine. However, despite these measures, there have been continued incursions of exotic monogenoids along with the imported ornamental fish, revealing potentially serious shortcomings in the guidelines. This communication discusses the effectiveness of these guidelines within the context of key issues surrounding exotic monogenoids. The priority research areas to tackle current challenges include 1) developing a national database on parasites, including monogenoids, of exotic/invasive aquarium fish, 2) probing whether the exotic monogenoids can infect the native Indian fish (parasite spillover) and whether the native Indian monogenoids can infect the introduced exotic fish (parasite spillback).
Monogenean parasites infecting imported ornamental fish to Australia

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The ornamental fish trade is a known pathway for the global translocation of aquatic parasites. Australian import conditions require all ornamental fish to be inspected seven days prior to importation to be free of parasites and diseases, however only goldfish consignments are required to be treated with parasiticides for the presence of Dactylogyrus extensus (Mueller and Van Cleave, 1932) and Dactylogyrus vastator Nybelin, 1924. We examined 37 ornamental fish consignments imported from Singapore, Malaysia, Thailand, and Sri Lanka to Australia (including freshwater and marine fish species) for monogenean parasites following inspection at Australian border control. Parasite species were identified based on haptoral and copulatory organ morphology, and representative samples of each species were used to amplify the ITS1 gene region for comparison. More than 950 monogenean parasites were collected from 15 consignments, representing a total of 14 species from five freshwater ornamental fishes. Seven Dactylogyrus spp. (including Dactylogyrus vastator Nybelin, 1924), and three Gyrodactylus spp. infected goldfish, Carassius auratus Linnaeus, 1758, from Malaysia, Singapore and Thailand; Dactylogyrus ostraviensis Réhulka, 1988, infected rosy barb, Pethia conchonius Hamilton, 1822, from Singapore, Sri Lanka and Thailand; two Trianchoratus spp. infected three spot gourami, Trichopodus trichopterus Pallas, 1970 and pearl gourami T. leeri Bleeker, 1852, from Sri Lanka and; Urocleidoides reticulatus Mizelle and Price, 1964, infected guppy, Poecilia reticulata Peters, 1859, from Sri Lanka. The discovery of D. vastator in goldfish consignments from Malaysia and Thailand suggests that the requirements of import health certificates for goldfish consignments prior to exportation to Australia are not being met. It is particularly concerning that other freshwater fish species require no prior treatment for parasites, as U. reticulatus and D. ostraviensis displayed high prevalence and mean infection intensities (prevalence=86.6% with mean intensity ± S.E.=18.30 ± 3.57, and 73.3% with 6.05 ± 0.74, respectively). The outcome of this study suggests that pre-export conditions and visual inspection at border control were insufficient to detect monogenean parasites infecting ornamental fish. Inspection of fish consignments prior to exportation and at border control must consider how highly cryptic in nature monogenean parasites can be, and consider alternative detection methods to visual inspection.
Environmental influence on infective stages of the monogenean *Sparicotyle chrysophrii*, parasite of gilthead sea bream (*Sparus aurata*) in Mediterranean aquaculture

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*Sparicotyle chrysophrii* is a monogenean causing significant losses of gilthead sea breams (*Sparus aurata*) in Mediterranean aquaculture. Infective stages of monogeneans (eggs and oncomiracidia) are directly exposed to environmental variability which affects their development, survival and transmission. This is particularly relevant in aquaculture facilities of gilthead seabream where seasonal and recurrent infections of *S. chrysophrii* hamper the production. Since improving management and reducing treatment applications is currently of main concern, this study focus on elucidate the effects of environmental variability on *S. chrysophrii* development, survival and transmission. With this aim, three abiotic factors, temperature, light and salinity, were experimentally tested on different *S. chrysophrii* egg-sets and the emerging larvae. Results showed that temperature was the most relevant abiotic factor since affected all the variables analysed. Developmental and survival times reduced as temperature increased while hatching success was quite high in a wide range of intermediate temperatures. Variations in photoperiod did not cause significant differences in hatching regimes or in developmental and survival variables. Moreover, effects of salinity were meagre even at the boundaries tested. The high tolerance of *S. chrysophrii* to temperatures and salinities occurring within and beyond the ranges of Mediterranean Sea underlies its wide-spread distribution and recurrence. Knowledge about environmental influence on *S. chrysophrii* development and survival should be implemented to improve treatment design and management of infection.

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Monogenean parasites causing pathological impacts on commercially important Mediterranean Sea fishes in Egypt

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Monogenea is the most common and abundant ectoparasites of fish, with greater diversity of species. Most of them are parasites mainly found on the gills, skin or fins of both freshwater and marine fishes causing mortality and economic losses. In the present study, three hundred fish species (100 Dicentrarchus labrax, 100 sparus aurata and 100 Argyrobs filamentus) were seasonally collected from the coastal region of Mediterranean Sea, Egypt from December 2015 to November 2016. One hundred eighty five samples (61.66%) were found infested with 6 species of monogenea belonging to four genera; Furnestinia, Encotylabe, Diplectanum and Sparicotyle. The morphological as well as morphometric characters of the isolated species were described using light microscopy. The prevalence, mean intensity, seasonal dynamics of the detected species were studied with the relation between rate of infestation and host length. Results revealed that Sparus aurata showed the highest rate of infestation (62%) and the maximum rate was recorded during summer. The most common histopathological alterations caused by the detected monogenean species were necrosis of gill epithelium with mononuclear cell infiltration.
Taxonomy and diversity of monogenean parasites of marine catfishes (Ariidae) from Visakhapatnam Coast, Bay of Bengal

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The present study deals with identification of monogenean parasites of two species of marine catfishes employing taxonomic and molecular techniques. Altogether 973 fish belonging to the family Ariidae were examined, out of which 835 were *A. jella* and 135 were *A. dussumieri*. Parasites collected from fishes were fixed in FAA for morphological studies and in 100% ethanol for molecular studies. Altogether 11 species of monogenean parasites were recorded. Preliminary identification of parasites was done employing various staining techniques and based on morphological characters. For molecular studies, DNA was amplified by using 28S rDNA primers i.e. F C1 [5'-ACC CGC TGA ATT TAA GCA-3'] AND 28S – R d2 [5'-TGG TCC GTG TTT CAA GAC-3'] (Hassouna et al., 1984). PCR amplification was performed in a final volume of 20 µl PCR reaction. The resultant PCR product was separated by electrophoresis, purified, sequenced and sequence analysis was done at GATC, Germany. All sequences were submitted to gene bank and accession numbers were obtained. Attempts were also made to analyse phylogenetic relationships of these parasites in relation to other groups to understand the processes of parasite speciation and diversification. All the 11 species of monogenean parasites were found to be distributed under four genera viz. Hamatopeduncularia with five species *H. arii* Yamaguti, 1953, *H. elongatum* Lim, 1996, *H. thallasini* Bychowsky and Nagibina 1969, *H. bifida* n.sp. (Present study) and *H. madhaviaei* n.sp. (present study), Chauhanellus with four species *C. chauhani* Venkatanarsaiah and Kulkarni,1990, *C. poculus*, Lim,1994, *C. hanumantharaoei* n.sp. (present study), and *C. visakhapatnamensis* n.sp. (present study), genus Neocalceostomoides with one species *N. spinivaginalis* Lim, 1995, and the genus Thysanatohaptor being represented by *T. rex* Krisky, et al. 2012. Out of these, four species with two each under the genus Hamatopeduncularia and Chauhanellus were considered as new species and one new genus was created with a single species under the family Neocalceostomatidae. Sharp and clear PCR bands were obtained for all species except *C. visakhapatnamensis* n.sp. which was not included in sequence analysis. The 28S rDNA sequence data was utilized for estimating base compositions and to determine the phylogenetic relationships of monogenean parasites belonging to different families under the order dactylogyridae. The outcome of the analysis as represented by the tree construction, clearly indicated a strong monophyly of the order dactylogyridae with 100% bootstrap values. Phylogenetic studies using the sequence data further revealed the close relationship of these parasites as they formed a single cluster in all phylogenetic trees tested. Bayesian inference analyses with transition model revealed the same tree topology as obtained by Maximum Parsimony, Neighbour Joining and Maximum Likelihood methods. The present study yielded a well-resolved phylogenetic relationship between monogenean species within each family. The results were further compared with respective fish host families and it is interesting to note that all species obtained from Ariidae fishes formed a single cluster in all the three relationship trees with slight positional changes. The study revealed that the two groups of monogenean parasites i.e. the Monopisthocotylea and the Polyopisthocotylea formed a monophyetic sister group and thus appeared as a clad.
Abstracts of posters

In order of appearance
Description of a new gyrodyactylicd (Gyrodactylidae: Monogenea) genus from the gills of the bulldog, Marcusenius macrolepidotus (Peters) from Lake Kariba, Zimbabwe

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There is a rich diversity of fish and amphibians in Africa’s inland water systems that serve as hosts for at least seven gyrodyactylicd genera. Currently, Africa’s gyrodyactylicd fauna includes species belonging to Afrogyrodactylus (3 species), Cithradactylus (1 species), Diplogyrodactylus (1 species), Gyrdicotylus (1 species), Gyrodactylus (35 species), Macroyrodactylus (9 species), and, Mormyrogyrodactylus (1 species).

In August, 2011, gyrodyactylicd material was collected from the gills of two specimens of bulldog, Marcusenius macrolepidotus, from Lake Kariba. The parasites collected presented a unique morphology and are regarded to represent a new genus. The monogeneans are viviparous, containing at least one developing embryo within in utero. The attachment apparatus consists of a single pair of hamuli with prominently flattened roots and a notable constriction one third along the length of the hamulus point. The hamuli are connected by a simple and thin dorsal bar. There are sixteen marginal hooks of one morphological type but of three different sizes. The male copulatory organ consists of a muscular pouch armed with approximately 30 gracile spines.

Phylogenetic analyses constructed on partial sequences of 18S rDNA using Maximum Likelihood and Bayesian Inference using a selected phylogenetic model (GTR+I+γ) did not place the new genus alongside other viviparous genera within the Gyrodactylidae. A Neighbour Joining analysis based on the uncorrected p-distances, however, clustered the new genus with Diplogyrodactylus martini, which possesses marginal hooks with two different morphologies.

A morphological and molecular description of the monogeneans collected from M. macrolepidotus is presented and discussed.

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Monogenean biodiversity in elasmobranchs from the Oceanogràfic aquarium of Valencia

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Elasmobranchs are especially delicate fish for their maintenance in aquaria; therefore, the study of their parasitoses becomes a fundamental issue to keep healthy elasmobranchs in captivity. Among the parasites, ectoparasites as monogeneans stand out as they produce severe epizootics (Thoney & Hargis, 1991; Bullard et al., 2001; Chisholm & Whittington, 2002; Janse & Borgsteede, 2003; Vaughan et al., 2016). Despite most of them are quite sporadic, those surviving captivity are particularly insidious, often parasitizing more vulnerable non-habitual hosts. In the Oceanogràfic aquarium in Valencia (Spain) the "Oceans" facility has numerous species of elasmobranchs, representing an important part of the biodiversity showed at the aquarium. During deworming routines several species of monogeneans are detached, collected and analyzed. As a result of the analysis of six species of Batoidei: Myliobatidae: Myliobatis aquila (L.), Aetobatus ocellatus (Kuhl, 1823), Aetomylaeus bovinus (Geoffroy Saint-Hilaire, 1817) and Rhinoptera bonasus (Mitchill, 1815); Dasyatidae: Pteroplatytrygon violacea (Bonaparte, 1832); and Glaucostegidae: Glaucostegus cemiculus Geoffroy Saint-Hilaire, 1817, seven different species of monogeneans have been identified: Neoentobdella diadema (Monticelli, 1902) Kearn & Whittington, 2005; Benedeniella posterocolpa (Hargis, 1955) Yamaguti, 1963; Thaumatocotyle pseudodasybatis Hargis, 1955; Decacotyle octona (Young, 1967); Monocotyle sp., Monocotylidae gen. sp.1 and Monocotylidae gen. sp. 2.

The presence of Neoentobdella diadema has been observed in five of the six species of elasmobranchs mentioned above. However, this species had only been reported previously on two of them: M. aquila and P. violacea to date (WoRMS; Host Parasite Data Base NHM). Pathology associated to these non-habitual hosts might be more severe and is being analyzed.

The study of host-parasite interactions will help us to design better deworming protocols to increase the welfare of elasmobranchs.

References:
Three-dimensional images of rigid structures by Laser Scanning Confocal Fluorescence microscopy as a tool for taxonomic studies on Dactylogyridae (Monogenea)

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One of the main problems in the study of the structural and functional morphology of small organisms, such as dactylogyrid monogeneans, is the lack of a comprehensive three-dimensional description of the diagnostic structures, since bi-dimensional line drawings and microphotographs provide partial pictures of their structural complexity. Dactylogyridae is overwhelmingly the most abundant and diverse taxon among monogeneans in continental waters of South America. Their small body size requires a considerable sampling effort and training to collect and identify worms from gills, skin, nasal cavities and other microhabitats. In fact, diagnostic features such as sclerites and the male copulatory complex are generally less than 100 μm in length and are essential for the taxonomic description and identification of species. The objective of this study is to propose a simple and routine method for morphological studies by Laser Scanning Confocal Fluorescence Microscopy, combining enzymatic digestion with Proteinase K for freeing sclerotized structures followed by laser confocal microscopy. This method is applicable to fresh or fixed specimens and does not require staining or dehydration. In fact, stable autofluorescence emission was detectable at 500-530 nm for bars, anchors and male copulatory complex when excited with argon laser. Readily available software for image processing was used for three-dimensional reconstruction of sclerotized structures generating 3D models and 360-degree full-rotation views, as well as orthogonal projections and morphometric analysis. The advantages of this protocol over previous methodologies for confocal laser imaging are discussed.
Little is known about the monogenean fauna of fishes in Germany and Central Europe. More than 30 years ago Kritscher (1988) investigated 20 species of fish of Lake Neusiedl (Austria), 13 of which were found infected with monogeneans. Later on roach (Dzika et al. 1999) and finally carp, rainbow trout (Dzika et al. 2009a,b) from two waters in Bavaria were studied in this respect. Now we conducted a preliminary study on the monogenean fauna of 7 species of fish. 65 fishes were examined in total. They belonged to the following species: loach (41 individuals), barbel (10 individuals), chub (6 individuals), ruffe (3 individuals), ide (2 individuals), perch (2 individuals) and tench (1 individual). The fish were caught by electro fishing in July 2014 from the river Sauer at the border between Germany and Luxembourg. Body surface, the fish gills and fins were placed in Petri dishes with water and inspected for monogeneans. Monogenea were gathered by scraping mucus with the use of a needle. The parasites were fixed in GAP (Malmberg 1970). Measurements and photographs of the haptor, copulatory and vagina apparatus were taken with an Olympus type light microscope using phase contrast and program cell B (Basic Imagining Software). Monogenea were determined to their species on the basis of skeletal elements, as well as the copulatory and vagina apparatus according to Lom and Ergens (1970), Gusev (1985) and Dzika (2008). The specimens of barbel, tench and perch were uninfected. A total of 44 parasite specimens belonging to 7 species were collected. These were: Dactylogyrus amphibothrium, D. folkmanovae, D. vistulae, Gyrodactylus prostae, G. jiroveci, G. sedelnikovi, G. pavlowskyi. The loach harbored 3 monogenean species, but in ruffe and ide only 1 species. The intensity of fish infection ranged from 1 to 4 specimens / fish. Among all of the species found, most belonged to specialists. The finding reveal that investigated cyprinid fishes of a river in Germany/Luxembourg were fairly well infected by 1 to 3 species of Monogenea. The number of parasites species being highest on loach might be related to the number of investigated fishes. Studies have shown that D. amphibothrium, G. prostae, G. jiroveci, G. sedelnikovi, G. pavlowskyi are new species for the fish fauna of Germany and Luxembourg. For further conclusion more comprehensive studies are required.
Morphometric and scanning electron microscopy of
*Dactylogyrus minutus* Kulwièc, 1927 (Monogenea: Dactylogyridae) from the *Cyprinus carpio var. koi*

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Carp from the coloured koi variety is commonly used for both aquarism and earthen-pond culture. However, its culture has been affected by dactylogyrid species, causing massive mortality for more than four species of *Dactylogyrus* genus in the same host. In the present study, *Dactylogyrus minutus* specimens living on the gills of *Cyprinus carpio* were identified using light, confocal and scanning electron microscopy to characterize its morphological and morphometrical characters by the first time in Brazil. Samples of carp var. *koi* (weight of 1.73±0.25 g; total length of 4.7±0.46 cm) were obtained in September 2016 from a fish farm located in Biguaçu, Santa Catarina, Southern Brazil and transported live to Aquatic Organisms Health Laboratory. After acclimatization period, thirty fish were anesthetized in clove oil (75 mg/L) and were examined for the infestation intensity of parasites on the gills using a stereomicroscope. Specimens of *Dactylogyrus* were isolated and fixed in 70% ethanol for morphological identification, 10% formalin for confocal microscopy and 2.5% phosphate-buffered glutaraldehyde for scanning electron microscopy. The monogeneans were identified as *Dactylogyrus minutus* Kulwièc, 1927 (P 75%, MII 3.55±2.91, MA 2.66±2.95). In general, the measurements (µm) of 20-Hoyer’s mounted specimens were close to those described by Kulwièc (1927): body 196-247 (216) long and 46-63 (53) wide at midbody, anchor total length 39-44 (42) long, with base length 33-37 (35) and point length 13-15 (14), anchor inner root 12-14 (13) long, outer root 5-6 (6) long. One single bar 28-32 (30) long, 14 marginal hooks 20-23 (22) long, male copulatory organ (MCO) 29-38 (34) total length. Oval egg 59-60 (60) long, 40-41 (40) total wide with one small appendage 5-6 (6). A pair of long anchors supported by a single bar longer rounded in the extremities at the dorsal side. In images obtained using confocal microscopy the shape of the bar was improved, evidencing the point in the extremities with a slight groove. The cirrus is a thin long tube which tapers anteriorly. There are two MCO accessory piece, one has a side branch and diverges into two points, and has approximately the same size as cirrus. Another piece is shorter originating at the opposite side. The vagina opens on the right side of the body anteriorly to the middle of the body, as shown by scanning electron microscopy. There was a previous report of *D. minutus* from Brazil in the same host but no morphological data were related. It can be considered that this study supports all the previous characterization, not only by the details of the haptor but also by the dorsal bar observation in a confocal microscopy and scanning electron microscopy. In fact, using these techniques the vaginal pore and specimens attached to the gills were observed. Morphometric measurements are given for the first time from koi carp collected in a fish farm from Brazil, which is important to highlight in case of a cosmopolitan fish parasite species.

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Diversity and distribution of parasitic monogenoids (Platyhelminthes) in a global biodiversity hotspot Arunachal Pradesh, India

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Arunachal Pradesh, the extreme northeast corner of India, being at the confluence of three major bio-geographical realms of the world-India, Indo-Malayan and Indo-Chinese- is extremely rich in biodiversity with several endemic species. Although this region has no less than 213- recorded fish species representing 20.72% of the total Indian freshwater fish fauna there is complete lack of information about the diversity of fish parasites. We characterized, using morphological approaches, the diversity and distribution of parasitic monogenoids from fish communities of five major river basins in Arunachal Pradesh. Over a 5-year sampling period from March 2011- December 2016, 93 species of fish belonging to 31 genera, 13 families, and 6 orders were collected and examined for monogenoids. 55 species (59%) were found infected with about 128 species of parasitic monogenoids giving an average infection of about 2.32 monogenoid per infected host fish species. Apparently, of these 128 species, 24 were known and 104 species new to the science. All 128 species belong to 15 known genera, (and apparently 11 undescribed genera), 4 families (Dactylogyridae, Gyrodactylidae, Sundanonchidae, and Diplozoidae), 3 orders (Gyrodactylidea, Dactylogyridea, and Mazocraeidea), and 2 subclasses (Polyonchoinea and Oligochainea). 15 known genera are: Dactylogyrus (48), Thaparocleidus (13), Bychowskyella (12), Paradiplozoon (12), Gyrodactylus (8), Dogielius (6), Cornodiscoides (4), Ancyrocephalus (3), Thaparogyrus (3), Mastacembelocleidus (02), Sundanonchus (02), Bifurcohaptor (01), Mizellius (01), Neodiplozoon (01), and Xenentocleidus (01). The monogenoids from the family Polyonchoinea represented the greatest number of species (115 species from 14 genera). The only representative of the family Oligochainea were 12 species belonging to Paradiplozoon and 1 species belonging to Neodiplozoon (Diplozoidae). Genus Dactylogyrus had the highest species diversity (48 species) followed by Thaparocleidus (13 species), Paradiplozoon (12 species), Bychowskyella (12 species), New genera (11 species), and Gyrodactylus (8 species). The cyprinids had the highest monogenoid species diversity (64), followed by Cobitidae (11), Bagridae (11), and Sisoridae (9). Among cyprinids, the highest diversity of monogenoids was found on Barilius bendelisis (8 species), followed by Bangana dero (7 species), and Garra kempi, Schizothorax richardsonii and Tor tor (5 species each). The study also records 32 new fish host records for monogenoids. The data generated herein are essential for studies on biogeography and provide a baseline for the conservation of biodiversity as a whole.
Enteromius paludinosus (Peters, 1852): host for new species of Gyrodactylus von Nordmann, 1832 (Monogenea: Gyrodactylidae) in South Africa

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Viviparous monogeneans of the genus Gyrodactylus are tiny parasites mostly parasitising bony fish. Nearly 500 Gyrodactylus species have been described worldwide, of which only 36 is known from the African continent. Parasitological survey of E. paludinosus at the Barberspan Ramsar site in South Africa in October 2015, recorded the presence of a new Gyrodactylus species. Sixty percent of the hosts were infected on the fins with up to three of these specimens. This new species of Gyrodactylus constitutes the first description of this parasite from E. paludinosus and, the 37th species described from the African continent. The morphologically new species represents a typical gyrodactylid specimen with noted similarities in the shape of haptoral sclerites. This specimen closely resembles to Gyrodactylus kyogae (Paperna, 1973) from Enteromius neumayeri (Fischer, 1884). Dissimilarities from G. kyogae are however evident in the overall larger size of the hamuli, ventral bar and marginal hooks. The elongate root of the hamuli and presence of a prominent membrane on the ventral bar also justify this as a new species. Furthermore, larger marginal hooks and definite disparity in the shape of the sickle is evident.
Monogeneans of marine fishes from Patagonia, Argentina: diversity and new host records

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Monogeneans are an integral part of the dynamic and complex fish parasite assemblages, often considered as demographic regulators of fish. Little is known about the monogenean diversity in the South Atlantic waters off Patagonia and very few recent contributions on the occurrence of these parasites in fish hosts have been published. The present study aims to contribute to the knowledge by exploring the diversity of monogeneans in marine fishes from off the Patagonian coast. A total of 557 teleost fishes belonging to 20 species of 17 families were collected from the neritic zone of the South-Western Atlantic (42°45'–47°19'S, 61°01'–64°25'W) between 1999 and 2007. A total of 11 species of monogeneans of three orders and six families were collected from 11 of the 20 fish species: (i) Capsalidea: Capsala martinieri Bosc, 1811 (Capsalidae) from Mola sp. and Tetrasepta dischizosepta Suriano, 1975 (Capsalidae) from Acanthistius patachonicus; (ii) Dactylogyridea: Neodiplectanotrema helicoleni Gerasev, Gaevskaja & Kovaleva, 1987 (Ancyrocephalidae (sensu lato)) from Helicolenus dactylopterus; (iii) Mazocraeidea: Anthocotyle merlucii van Beneden & Hesse, 1863 (Discocotylidae) from Merluccius hubusi, Euryorschis australis Manter & Walling, 1958 (Diclidophoridae) from Seriolella porosa, Microcotyle nemadactylus Dillon & Hargis, 1965 (Microcotylidae) from Nemadactylus bergi and Microcotyle pseudopercis Amato & Cezar, 1994 from Pseudopercis semifasciata, Neogrubea seriolellae Dillon & Hargis, 1968 (Mazocraeidae) from Stromateus brasiliensis and S. porosa, Neoheterobothrium paralichthyi Suriano & Labriola, 1999 (Diclidophoridae) from Xystreurys rasile and Orbocotyle marplatensis Euzet & Suriano, 1975 (Diclidophoridae) from Prionotus nudigula. Most of the monogeneans were collected from the gills, except for C. martinieri from the skin of Mola sp. and N. helicoleni from the oesophagus of H. dactylopterus. The present study provides a new host record for N. paralichthyi (i.e. X. rasile) and new locality records for five species: M. nemadactylus, N. helicoleni, N. paralichthyi, O. marplatensis and T. dischizosepta. The fact that more than 50% of analysed fin fish harbour at least one monogenean species points to a high presence of these parasites in the Patagonian waters, motivating further studies on this highly diverse environment.

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Introduction to “Fauna Sinica Platyhelminthes Monogenea II”

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More than 6000 species belonging to 53 families of monogeneans were described around the world based on published records. In China, Prof. Wu Baohua and other Chinese scholars published the book “Fauna Sinica Platyhelminthes Monogenea” in 2000, in which 584 monogeneans species, belonging to 9 families 42 genera were reported and most of them parasite on freshwater fishes, amphibians and chelonians. With the support of the National Natural Science foundation of China, we launched the project to edit “Fauna Sinica Platyhelminthes Monogenea II” started from 2011. The volume includes 432 species in total, belonging to 166 genera and 34 families. Among them, 28 families were not included in the book edited by Wu et al. (2000), and most of them were collected from marine fishes. The morphological characteristics of species were described, the features of genus and family were summarized, and the information about their host, parasitic sites and the geographical distribution were provided in this book. Each species was provided with the line graph, and microscope photos were shown for some species. This book, “Fauna Sinica Platyhelminthes Monogenea II”, comprehensively and systematically summarized the fauna of monogenean at present in China.
Taxonomic Studies on Monogeneans from Elasmobranchs off South China Sea

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About 200 species of monogeneans parasitizing on elasmobranchs were described around the world, while only 20 species record in China. The investigation of monogenean from elasmobranchs off South China Sea has carried out from 2012-2015, and 11 species of elasmobranchs including 196 specimens have been checked. 16 species belonging to 12 genera and five families of monogeneans were collected. Among them, 11 species belonging to Monocotylidae, two species to Hexabothriidae, and other three species belonging to Acanthocotylidae, Loimoidae and Amphibdellatidae respectively. Four new species are proposed and two families, five genera and seven species are first recorded in China. The new information are listed as follows:

**Acanthocotylidae Price, 1936 n. rec. in China**
*Pseudacanthocotyla* Yamaguti, 1963 n. rec. in China
*Pseudacanthocotyla williamsi* (Price, 1938) Yamaguti, 1963 n. rec. in China

**Loimoidae Bychowsky, 1957 n. rec. in China**
*Loimos* MacCallum, 1917 n. rec. in China
*Loimos shanweiensis* n. sp.

**Hexabothriidae Price, 1942**
*Squalonchocotyle* Cerfontaine, 1899 n. rec. in China
*Squalonchocotyle sinensis* n. sp.
*Erpocotyle sphyrnae* MacCallum, 1931 n. rec. in China

**Monocotylidae Taschenberg, 1879**
*Heterocotyle orientalis* n. sp.
*Neoheterocotyle tetraspina* n. sp.
*Calicotyle* Diesing, 1850 n. rec. in China
*C. australis* Johnston, 1934 n. rec. in China
*Monocotyle tritestis* Young, 1967 n. rec. in China
*M. caseyae* Chisholm et Whittington, 2005 n. rec. in China
*Merizocotyle macrostrobus* Chisholm & Whittington, 2012 n. rec. in China
*Empruthotrema* Johnston & Tiegs, 1922 n. rec. in China
*E. stenophallus* Chisholm & Whittington, 2005 n. rec. in China
Monogenean diversity of western Mediterranean mackerels, bonitos and tunas (Scombridae)

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Scombrids are important commercial species in fisheries worldwide and in particular in the Mediterranean Sea. Moreover, due to the incipient cultures of scombrids, it is imperative to know the parasite fauna which is potentially pathogenic. The aim of this work is to explore the metacommunity diversity of the monogeneans of the scombrids in the western Mediterranean describing and comparing the monogenean assemblages infecting fishes of the tribes Sardini (bonitos), Scombrini (mackerels) and Thunnini (tunas). Samples were taken from 1092 specimens of nine scombrid species caught in the western Mediterranean Sea (Sardini: 111 Sarda sarda, 120 Scomber colias; 33 Sc. scombrus; Thunnini: 57 Auxis rochei; 23 Euthynnus alletteratus; 22 Katsuwonus pelamis; 104 Thunnus alalunga, 3 T. obesus and 619 T. thynnus). Seventeen species of monogeneans were found: two in S. sarda, Capsala gregalis (prevalence, 12%) and Hexostoma thynni (5%); four in Sc. colias, Grubea cochlear (4%), Kuhnia scombercolias (1%), K. scombri (2%) and Pseudokuhnia minor (39%); two in Sc. scombrus, G. cochlear (12%) and K. scombri (3%); six in A. rochei, Allopseudaxine macrova (9%), C. gregalis (9%), C. manteri (2%), Churavera triangula (4%), H. auxisi (5%) and H. keokeo (4%); two in E. alletteratus, C. manteri (13%) and H. thunninae (17%); one in T. alalunga, C. paucispinosa (3%); one in T. obesus, H. thynnii (67%); six in T. thynnus, C. magronum (24%), C. paucispinosa (3%), Gastrocotylinae gen. sp. (0.2%), H. thynnii (46%), Nasicola hogansi (0.2%) and Tristomella onchidiocotyle (2%). No monogeneans were found in K. pelamis. The records of C. gregalis, C. manteri and H. keokeo in A. rochei, C. gregalis and H. thynnii in S. sarda, H. thynnii in T. obesus and Gastrocotylinae gen. sp. and N. hogansi in T. thynnus are new for the Mediterranean Sea. The monogenean species in S. sarda were also present in Thunnini fishes, relating both tribes. Hexostoma thynni was the most generalist parasite, in three host species (Sardini and Thunnini). The capsalids C. gregalis and C. manteri were found in S. sarda and species of Thunnini. Scomber spp. share exclusive Scombrini monogenean species. The results obtained also suggest that there are differences in parasite fauna between western Mediterranean Sea and Atlantic Ocean, and these data could be useful as fish population indicators. Capsalids and H. thynnii can be considered as potential pathogens for aquaculture; moreover wild bonitos and tunas can act as parasite reservoirs. Projects: IEO GPM1213, AGL2010-20892, AGL2015-68405-R and PROMETEO/2015/018. PhD student grants: FPU13/00274 for JR-L and “Atracció de talent, UVEG” for JFP-A. SM benefits of the grant n. 153/2016.2213 of the University of Sassari.
First record of Protocotyle grisea (Cerfontaine, 1899) (Monogenea: Hexabothriidae) infecting the bluntnose sixgill shark, Hexanchus griseus (Bonnaterre) from the Valencian Gulf, Spain

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The bluntnose sixgill shark, Hexanchus griseus (Bonnaterre) is a huge deep shark with a global distribution. In the eastern coast of the Spanish Mediterranean, records of bluntnose sixgill sharks are considerably numerous, which suggest that this shark species is relative abundant in the area. During a survey of parasites of deep-sea elasmobranchs from off the western Mediterranean coast of Valencia, Spain, we examined two specimens of H. griseus (68–102.5 cm in total length and 1,067–3,550 g in weight). Freshly sharks were collected in the Valencian Gulf (Balearic Sea) (38°04'–40°09'N, 0°04'–0°25'E; depth 400 m) during May 2010. After the parasitological analyses, several hexabothriids were collected from the gills, stained with iron acetocarmine or alum carmine, dehydrated through an ethanol series, cleared in dimethyl phthalate, and mounted in Canada balsam. Additionally, some specimens were examined in detail by scanning electron microscopy. A total of 56 monogeneans identified as Protocotyle grisea (Cerfontaine, 1899) Euzet & Millard, 1974 were collected from both sharks (intensity = 28 worms per fish). Adults found in the bluntnose sixgill sharks from Spain are somewhat smaller (e.g., anterior sclerites 876–1,641 μm; median sclerites 904–1,675 μm; posterior sclerites 1,086–1,536 μm; hamulus outer length 71–91 μm; hamulus inner length 53–69 μm) than specimens of P. grisea described from France and Algeria. This is the first report of P. grisea from the bluntnose sixgill shark from the Spanish Mediterranean coast. Voucher specimens of P. grisea from Spain are available at the Helminthological collection of the Institute of Parasitology, Czech Academy of Sciences, České Budějovice, Czech Republic.
Monogenean fauna of Japanese freshwater fishes and phylogeny of Dactylogyridae from Japan

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In total, 4,370 species of fishes are known to occur in Japan, where 229 species of monogeneans have so far been reported from only 172 species of fishes, 3 species of reptiles, 9 species of amphibians, and 3 species of invertebrates. Of these monogeneans, 78 nominal species have been reported from about 30 species of freshwater fishes, and most of these fishes are commercially important species from a viewpoint of parasitic diseases in aquaculture. Thus, much remains to be studied on the monogenean fauna of wild freshwater fishes of Japan.

Since 2012, we have examined about 170 species of freshwater fishes in Japan and collected many monogeneans including undescribed or unreported species. Based on our estimate, one species of Japanese freshwater fish harbors 1.3–1.8 species of monogeneans on average. About 500 species of freshwater fishes occur in Japan, and it is thus estimated that 650 to 900 species of monogeneans inhabit Japanese inland waters. About 90 endemic freshwater fish species occur, and monogeneans infecting those endemic fishes are also likely to be endemic. Dactylogyrus bicorniculus and D. oryziasi described by us are such endemic species and most likely faces the danger of co-extinction with its host. A total of 167 species of freshwater fishes are listed in the Red Data Book of Japan, extinction and sudden decrease of wild hosts and alteration of local ecosystems have been suggested to cause co-extinction of their parasites, and host-specific monogeneans may be under the same situation. Thus, it is important and necessary to understand the monogenean fauna of Japanese freshwater fishes in terms of conservation of biodiversity.

Currently, we tried to clarify the phylogenetic relationship of dactylogyrideans collected from Japanese freshwater fishes. Preliminary analysis based on 28S rDNA sequences shows that “Ancyrocephalus” parasitic on cyprinids and “Ancyrocephalus” parasitic on percichthyids and odontobutida are separated from each other and form two clades. The former is a sister group of Dactylogyrinae, and the latter clade is a part of Pseudodactylogyrinae. The species of “Ancyrocephalus” reported from primary freshwater fishes in Japan and neighboring Far East Asian countries need for a taxonomic revision.
Diversity of monogeneans from selected fish species from Haut-Katanga Province in the Democratic Republic of Congo

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The Haut-Katanga Province, Democratic Republic of Congo, is characterised by several lakes and rivers with abundant and diverse fish species. Despite this large fish diversity, few records on fish parasite research are available from this province. Fish were collected in March 2016 with seine and gill nets from Lubumbashi Zoo, an aquaculture farm and two lakes, Tshangalele and Koni, some 100 km from Lubumbashi. Seventeen fish species were examined for the presence of monogeneans. Standard procedures were followed for collection and preservation of monogeneans; prevalence of infestation is indicated. Eight fish species were collected from the Zoo with only two species infested with monogeneans; a diplozoid (100%) from Barbus paludinosus and Macrogyrodactylus sp. (50%) from Clarias gariepinus. Macrogyrodactylus sp. (40%) and Quadriacanthus sp. (40%) were recorded from C. gariepinus while Cichlidogyrus sp. (40%), Enterogyrus sp. (20%), and Gyrodactylus sp. (20%) were recorded in low numbers from Oreochromis niloticus at the aquaculture farm. Six fish species were collected from Lake Tshangalele with Cichlidogyrus spp. (66.6%) recorded from Coptodon rendalli, Cichlidogyrus sp. (40%) from Oreochromis macrochir, Cichlidogyrus sp. (50%) from Tilapia sparrmanii and Quadriacanthus sp. (100%) from C. gariepinus. Eight fish species were collected from Lake Koni with Enterogyrus sp. (20%) recorded from O. macrochir, Macrogyrodactylus sp. (50%) and Quadriacanthus sp. (50%) from C. gariepinus. Monogeneans were recorded from six of the seventeen fish species examined with none displaying high intensity levels. No severe pathology due to monogenean infestation was evident.
Morphometric and molecular characterisation of two new species of Enterogyrus Paperna, 1963 (Dactylogyridae: Ancyrocephalinae) from Oreochromis mossambicus in South Africa

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Enterogyrus Paperna, 1963 is one of 16 genera of Dactylogyridae with an endoparasitic life strategy. To date, 12 Enterogyrus spp. have been described mainly from the African continent, with only some species having a cosmopolitan distribution. To date, only one species, i.e. Enterogyrus cichlidarum Paperna, 1963 has been reported from the Mozambique Tilapia Oreochromis mossambicus (Peters, 1852).

The study was undertaken from April 2008 to April 2009 at Nwanedi-Luphephe Dam (22°39.492’S; 30°25.342’E) of the Limpopo River System (South Africa), where 45 specimens of O. mossambicus were collected using gill nets. Morphometrical evaluation of collected material revealed the presence of two unknown Enterogyrus species. In November 2014 additional sampling was carried out in order to obtain material for molecular characterisation. The partial SSU+ITS1 and LSU rDNA were sequenced and compared to data available in the nucleotide database.

The two new species can be differentiated from known species based on the dimensions and spiral formula of the MCO. Enterogyrus sp. 1 has a 5-2-3 formula with Enterogyrus sp. 2 having a unique 8/9-2-4 formula. The newly identified species differ from each other in their SSU+ITS sequences in 15 of 869 bp. The LSU sequences of both new species resulted as the closest to an undescribed Enterogyrus species described from Senegal.

The scarcity of molecular data for Enterogyrus spp. in the nucleotide database should serve as motivation to collect different species of the genus to perform a proper phylogenetic comparison to better understand interspecific relationships.

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The first record of dactylogyrid parasites from *Labeo horie* and *Labeo niloticus* from Sudan

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Dactylogyrid monogeneans of *Labeo horie* (Heckel, 1847) and *Labeo niloticus* (Forsskål, 1775) (Cyprinidae) were surveyed at two sites of the Sudanese stretch of the River Nile. We report the presence of ten *Dactylogyrus* species and three *Dogielius* species, including *Dactylogyrus nathaliae* Guégan, Lambert & Euzet, 1988, *D. rastellus* Guégan, Lambert & Euzet, 1988, *D. retroversus* Guégan, Lambert & Euzet, 1988, *D. senegalensis* Paperna, 1969, *D. yassensis* Musilová, Řehulková & Gelnar, 2009, *Dogielius flosculus* Guégan, Lambert & Euzet, 1989, five undescribed *Dactylogyrus* species and one undescribed *Dogielius* species. We also provide a description of *Dogielius sennarensis* n. sp., which resembles *D. intorquens* but differs from this species (and other congeners) by longer ventral bar and anchor points and absence of sclerotized vagina. Finally, we redescribe *Dogielius flosculus* based on the morphology of specimens recovered from *L. horie*. This redescription has determined a single large vagina in all specimens studied, in contrast to vagina composed of two unconnected parts provided in original description. All parasite species observed represent new host and geographic records.
Monogenean parasites of sparid fishes, from the western Mediterranean off Algeria

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The monogenean fauna of parasites infecting fishes of the family Sparidae off the Algerian coasts is poorly known. The study was carried out on sparid fishes of commercial importance which represent an important biological resource in marine food webs and for the human population along the Mediterranean coasts of Algeria. The purpose of this study is to advance the knowledge of the monogenean fauna of the sparid fishes that are considered as promising targets for aquaculture. The fish were sampled from different regions off the Algerian coast from 2013 to 2016. We examined the gills of six species: Oblada melanura, Spicara maena, Diplodus vulgaris, Sparus aurata, Pagellus bogaraveo and Pagrus pagrus. A total of 20 monogenean species belonging to 7 families was identified in the fishes sampled: Diplectanidae Monticelli, 1903 (Lamellodiscus elegans Bychowsky, 1957; Lamellodiscus ignoratus Palombi, 1943; Lamellodiscus kechemirae Kechemir-Issad, 2005; Lamellodiscus tomentosus Amine & Euzet, 2004; Lamellodiscus knoepfleri Oliver, 1969; Lamellodiscus baeri Oliver 1974 and Lamellodiscus virgula Euzet & Oliver 1967); Capsalidae Baird, 1853 (Encotyllabe vallei Monticelli, 1907; Encotyllabe sp.); Gotocotylidae (Gotocotyla Parona & Perugia, 1891); Microcotylidae Taschenberg, 1879 (Polylabris sp.; Atriaster seminalis Euzet & Maillard, 1973; Atrispinum sp.; Furnestinia echeneis (Wagener, 1857) Euzet & Audouin, 1959; Bivagina alcedinis Parona & Perugia, 1889; Microcotyle erythrina van Beneden & Hesse, 1863 and Microcotyle sp.); Diclidophoridae (Cyclocotyla bellones Otto, 1823; Choricotyle chrysophrii Van Beneden & Hesse, 1863; Choricotyle sp. In conclusion, the present results bring an important insight into species richness of the monogenean parasites of sparinds along the Algerian coasts. Our results will allow further investigation on the geographical distribution of the monogenean within the ranges of their hosts.
Monogenea of largemouth bass in southern Africa: new records

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Largemouth bass \textit{Micropterus salmoides} is an alien fish species introduced in South Africa in 1928. Like many biological invasions, they pose global threats as they may transfer their parasites to indigenous species and thus affect their health. The limited African literature on this fish species reveal parasites other than monogeneans. The aim of the study was to investigate the presence of monogeneans in specimens of largemouth bass introduced as fingerlings in a subtropical manmade lake in the Limpopo Province of South Africa mainly for angling tournaments by enthusiasts. Hosts (n=32) donated by anglers during bass-fishing tournaments were examined for monogeneans using a standard protocol. The investigation yielded three species of dactylogyrid monogeneans and all the specimens were mount on slides using glycerine jelly. Two species, \textit{Haplocleidus furcatus} (n=432) and \textit{Syncleithrum fusiformis} (n=12) cohabited the gills. The third species, \textit{Acolpenteron ureteroecetes} (n=3) inhabited the ureter-urinary bladder complex. The infection levels with regard to seasons, host size and host sex were determined. \textit{Haplocleidus} had the highest prevalence in summer, medium hosts and females with the highest abundance in spring, medium hosts and females. \textit{Syncleithrum} had the highest prevalence in summer, large hosts and females with the highest abundance in spring, medium hosts and females. \textit{Acolpenteron} had the highest prevalence in summer, large hosts and males with the highest abundance in summer, large hosts and males. The results serve as the first records of these three monogenean species in African freshwater bodies.
Dactylogyrids (Platyhelminthes, Monogenoidea) from the gills of *Hoplias malabaricus* (Characiformes: Erythrinidae) from coastal rivers of the Oriental Amazon Basin: Species of *Urocleidoides* and New Genus

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Five species of *Urocleidoides* (1 new) and 2 new species of **New Genus** are described in this study. All were collected from the gills of *Hoplias malabaricus* (Characiformes, Erythrinidae) captured in six localities of coastal rivers of the northeastern sector the State of Pará (Oriental Amazon): *Urocleidoides brasiliensis* Rosim, Mendoza-Franco & Luque, 2011, *Urocleidoides n. sp. 1*; *Urocleidoides cuiabai* Rosim, Mendoza-Franco and Luque, 2011, *Urocleidoides eremitus* Kritsky, Thatcher & Boeger, 1986, *Urocleidoides malabaricus* Rosim, Mendoza-Franco & Luque, 2011, New Genus n. sp. 1, and New Genus n. sp. 1. This is the first reported occurrence of the four previously described species of *Urocleidoides* parasitizing *Hoplias malabaricus* from streams in the Oriental Amazon Basin. The analysis of voucher specimens of *Urocleideoides eremitus* parasitizing the gills of *Hoplias malabaricus* from the Upper Paraná River floodplain in the limits of States of Paraná and Mato Grosso do Sul, Brazil, indicates that these specimens are members of a new species of *Urocleidoides*, described here as *Urocleidoides n. sp. 2* **New Genus** is proposed for the species with a male copulatory organ sclerotized, coiled, clockwise; ventral anchor with elongate superficial root, inconspicuous deep root; dorsal anchor with inconspicuous roots, and a constriction at the intersection between the shaft and the point. The host-parasite diversity scenario and host specificity of the species of **New Genus** and *Urocleidoides* from the gills of *Hoplias malabaricus* are also discussed in this study.
Monogenoidean parasites of *Acestrorhynchus falcatus* (Characiformes: Acestrorhynchidae) from Pará State, Brazil: species of New Genus and *Diaphorocleidus* (Monogenoidea: Dactylogyridae)

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Two new species of *Diaphorocleidus* and a new species of New Genus are described from the gills of *Acestrorhynchus falcatus* Bloch from coastal rivers of the northeastern sector the State of Pará (Oriental Amazon). *Diaphorocleidus* sp. n. A is characterized by possessing a male copulatory organ comprising a counterclockwise tube of about 3 rings; anchors similar with subtriangular superficial roots; ventral bar with posteromedial projection; and hook pairs 1, 4 and 7 approximately three times bigger than hook pair 5. *Diaphorocleidus* sp. n. B differs from the other congeneric species by having the accessory piece composed by two ramus; a tubular vagina, slightly sclerotized, bottle shaped. The monotypic, New Genus is proposed and characterized by the following features: male copulatory organ, sclerotized, coiled, clockwise; accessory piece sclerotized, articulated with the male copulatory organ; vagina, muscular, sinistro-dorsal; ventral anchor with conspicuous roots; dorsal anchor with superficial root five times bigger than deep root; and dorsal bar absent.

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Diversity of Monogenoidea (Platyhelminthes, Neodermata) of two components of the neotropical ichthyofauna: Loricariidae (Siluriformes) and Gymnotiformes from coastal rivers of the Oriental Amazon Basin and Xingu Basin

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A total of 284 fish belonging to the order Gymnotiformes and the family Loricariidae (Siluriformes) from the basins of the Marapanim River, Maracana River; Caeté River; Piriá River; Guama River; Quatipuru and Xingu River were investigated for monogenoidean parasites. Fifty-eight specimens (30 loricariids, 28 gymnotiforms) were parasitized by at least one species of monogenoid. Ten species of dactilogyrid and two species of gyrodactylid previously described from studied hosts are reported were reported for the first time parasitizing loricariids and gymnotiforms from the streams of the Northeast Coast of the State of Pará. Two new species of monogenoids from the gill arches of loricariids collected Low Xingu- Iriri river and one new species of monogenoid parasitizing gill arches of gymnotiform host collected in Maracanã basin, are described herein. *Unilatus* sp. nov. A from *Scobinancistrus aureatus* Burgess, 1994 (Loricariidae), *Unilatus* sp. nov. B from *Panaque armbrusteri* Lujan, Hidalgo and Stewart, 2010 (Loricariidae) and *Urocleidoides* sp. nov. from *Brachyhypopomus* sp. (Gymnotiformes). *Unilatus* sp. nov. A is distinguished from other cogeners by possessing: (a) anterior anchor with well-developed root, inconspicuous or reduced deep root; (b) male copulatory organ spiraled with 16 to 18 rings; and (c) hooks pair 1 with shank inflated proximally (2/3 of the length of the hook). *Unilatus* sp. nov. B is characterized by having: (a) anterior anchor with well-developed superficial root, inconspicuous or reduced deep root, shaft bent at midpoint, forming angle of approximately 45º, evenly short curved point; (b) male copulatory organ spiraled with 18 to 19 rings with proximal portion with small bulb, distal portion thin, elongate (1/4 of the male copulatory length). *Urocleidoides* sp. is characterized by having copulatory organ with 2 laps and whole accessory piece, plate-shaped, dorsal anchor with elongated superficial root and tip trifurcated.

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Dactylogyrids (Platyhelminthes, Monogenoidea) from the gills of *Hassar gabiru* Birindelli, Fayal et Wosiacki, 2011 and *Hassar orestis* (Steindachner, 1875) (Siluriformes, Doradidae) from the Xingu Basin

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Four new species of monogenoids are reported parasitizing the gills of doradid catfishes (Siluriformes) from Xingu river and related rivers: *Cosmetocleithrum* sp. n. A from *Hassar orestis*; *Cosmetocleithrum* sp. n. B from *H. gabiru*; *Cosmetocleithrum* sp. n. C and Vancleaveus sp. n. from *H. orestis* and *H. gabiru*. *Cosmetocleithrum* sp. n. A possesses: (1) ventral anchor with well-developed superficial root, triangular, narrow; (2) dorsal anchor with superficial root slightly developed, broad triangular; inconspicuous deep root; (3) male copulatory organ (MCO) coiled with 2½ rings, base of the MCO with a wide sclerotized flap; (4) elongated accessory piece with distal region flabeliform; (5) slightly sclerotized vagina, vaginal canal long, sigmoid, slightly sclerotized. *Cosmetocleithrum* sp. n. B has: (1) ventral and dorsal anchors with a broad base, inconspicuous superficial, deep roots; (2) ventral bar slightly straight with inflated ends; (3) MCO coiled with 4½ rings; (4) accessory piece elongated, sigmoid, with distal region cup-shaped. *Cosmetocleithrum* sp. n. C presents: (1) ventral anchor with well-developed superficial root, subtriangular; (2) dorsal anchor with surface root subtriangular, inconspicuous deep root; (3) MCO J-shaped with proximal portion bulbous; (5) accessory piece elongated with sharp distal region. *Vancleaveus* sp. n. differs from the congeneric by possessing (1) ventral anchor with divergent roots; surface root well developed, triangular with a conspicuous fold; deep root well developed; (2) dorsal anchor with divergent roots, rounded surface root with a remarkable fold; deep root developed; (3) MCO coiled with 3½ rings, base of the MCO with sclerotized flap. We also reported the first occurrence of *Cosmetocleithrum bifurcum* into Brazilian waters is reported.

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Space for your NOTES
Serpin from *Eudiplozoon nipponicum*

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The properties of proteins (e.g. functions) of the members from the family Monogenea are among the less investigated in whole phylum Platyhelminthes. We chose *Eudiplozoon nipponicum* as our experimental organism to address this issue and complete the mosaic of its functional proteins equipment.

*E. nipponicum* (family Diplozoidae, Polyopisthocotylea) is hematophagous ectoparasite which lives on the gills of common carp (*Cyprinus carpio*).

The main aim of our current work is to understand the regulation of peptidase activity related to many physiological processes. Among the key regulatory factors could be included the peptidase inhibitors, such as serpins - serine peptidase inhibitors. These functional proteins are generally known as important regulators of the coagulation cascade, complement, fibrinolysis, angiogenesis, inflammation etc. The inhibition, mediated by serpins, is typically irreversible, comprising conformational changes in serpin molecule leading to distortion of peptidase tertiary structure. Except the inhibition, some of them may have other functions like e.g. protein transporters or chaperones and they are relatively abundant also in secretions, body lysates and genomes/transcriptomes of helminths.

We identified serpin gene/protein in transcriptome of *E. nipponicum* and investigated properties of its recombinant form – molecular and biochemical characterization was performed. The tertiary structure was predicted, antigenic properties evaluated and inhibitory effect measured.

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Expression of genes encoding Toll-like receptors associated with infection by monogenean parasites

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When invaded by pathogens, host organism mobilizes the immune system lines of defence. Study involving targeting of immune-related genes and monitoring changes in their expression levels is one of the possibilities how to study responses of host to pathogen infection. Toll-like receptors (TLRs), a group of transmembrane protein receptors, play a crucial role in innate immunity response to pathogens. Expression of genes coding for TLRs in fish was previously shown to be influenced by parasite infection. In our study, we aimed to compare TLRs genes expression patterns in different tissues (spleen, head kidney and gills) of Abramis brama x Rutilus rutilus hybrid system. Fish were collected from Hamry reservoir (Brno, Czech Republic) and parasitological dissection for metazoan parasites was performed. Monogenean parasites represented the most dominant (90%) group of metazoan parasites in studied fish. Considerably higher abundance of monogenean parasites was revealed in A. brama in comparison with R. rutilus and hybrids. At the same time, significantly higher TLR21 expression was observed in A. brama than in remaining fish groups. Significant effect of monogenean abundance and fish group (i.e. A. brama, R. rutilus and hybrids) on TLR21 expression was subsequently supported by GLM test. No effect of monogenean abundance was revealed on TLR21 expression in remaining tissues and TLR2 expression. However, in A. brama significant negative correlation was observed between the abundance of strict specialist Dactylogyrus zandti and expression of TLR21 in gills. Since effect of remaining parasite groups on levels of TLR21 expression cannot be excluded, experimental study involving infection by monogenean parasites is further needed in order to confirm these findings.

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Microscopic anatomy of monogenean parasite

*Paradiplozoon homoion*

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The species *Paradiplozoon homoion* (Bychowsky et Nagibina, 1959) belongs to the family Diplozoidae (Monogenea), representing blood-feeding gill ectoparasites of freshwater fish. Diplozoids exhibit extraordinary morphology and life cycle involving permanent fusion of two larval worms and their subsequent transformation into one individual. Samples of *P. homoion* were collected from the gills of roach (*Rutilus rutilus* L.), bleak (*Alburnus alburnus* L.) and gudgeon (*Gobio gobio* L.) caught by electrofishing or seine netting in the Mušov reservoir (southern Moravia, Czech Republic).

Using a combined approach of confocal and electron microscopy, adults of monogenean parasite *P. homoion* were investigated for the body organization and adaptation to the ectoparasitic lifestyle, with emphasis on worm’s forebody and hindbody. Special attention was given to the parasite’s tegument, digestive, reproductive, and excretory systems. Labelling of filamentous actin with fluorescent phalloidin revealed the musculature of the body wall, buccal suckers, the pharynx as well as some parts of reproductive tract. Staining with chloride carmine and counterstaining with DAPI proved to be powerful tools for confocal microscopic detection of various cell types including putative gland cells. Immunofluorescent detection of α-tubulin showed to be suitable for visualization of excretory system and some parts of nervous system.

This study was supported by ECIP (European Centre of Ichthyoparasitology); centre of excellence program on the Czech Science Foundation; project No. P505/12/G112
The immunity induced by Dactylogyrus ctenopharyngodonid can protect grass carp from infection of D. ctenopharyngodonid and Ichthyophthirius multifiliis

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*Dactylogyrus ctenopharyngodonid* and *Ichthyophthirius multifiliis* are two important ectoparasites of fish. Both parasites could induce fish immune response to decrease the parasites infection intensity and develop resistance ability against parasites re-infection. The present study aimed to study the protective immunity of grass carp survived from *D. ctenopharyngodonid* infection against *D. ctenopharyngodonid* or/and *I. multifiliis* infection. The results demonstrated that when grass carp were infected by *D. ctenopharyngodonid*, the red blood cell (RBC) and the percentages of thrombocytes, monocytes, and neutrophils significantly increased at the early stage of infection. The percentage of lymphocytes increased with the prolonging of infection period. The mean intensity of *D. ctenopharyngodonid* decreased to 0 on day 28. The activities of serum acid phosphatase (ACP), alkaline phosphatase (AKP), lysozyme (LZM), and superoxide dismutase (SOD) significantly increased after *D. ctenopharyngodonid* infection. In addition, the survived grass carp could completely resist *D. ctenopharyngodonid* re-infection and partially protect fish from *I. multifiliis* infection.
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Benedeniine capsalid monogeneans causing disease problems and mortality among cultured Mugil species in Egypt

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Mugilid fish are of the most important fish species that have successfully cultured in Egypt and commonly exposed to parasitic diseases because of the poor water quality. The object of this study is to estimate cases of disease problems and mortality among cultured Mugil capito and Mugil saliens in 2 private farms in Egypt and describe the morphological and morphometric characteristics of the causative parasitic agent. In the present investigation, live and freshly dead Mugil species samples from both farms were collected and examined by the standard parasitological method with detailed data concerning number of fish examined, their weight and body size also water samples were taken and examined for investigating the pollution level and quality parameters. Two species of capsalid monogenea were detected during the study which are Benedenia sp. and Neobenedenia sp. and both of species were identified according to the available literatures. Results of water analysis revealed significant correlation between pollution and parasitism in the examined farms.
Prevalence of monogenean species in *Oreochromis niloticus* from southern Brazil

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Due to its species richness, ubiquity and epizootic potential, monogenean parasites are considered one of the most important in captive fish breeding, including Nile tilapia *Oreochromis niloticus* whose association, not rarely, results in significant economic losses due to high mortality rates. Among the studies necessary for the elaboration of control strategies for the group, there is a highlight for parasitology. Therefore, the knowledge of their indices and forms of distribution are fundamental. The aim of this study was to report the prevalence of monogenean species in the gills and stomach of *O. niloticus* collected in the southern Brazil. A total of 55 fish (weight of 21.65 ± 12.62 g, total length of 10.97 ± 1.92 cm) were collected from a fish farm in earthen pond and transported alive to the Aquatic Organisms Health Laboratory (AQUOS) of the Federal University of Santa Catarina (UFSC). 24 h after acclimation period, the fish were anesthetized with eugenol (75 mg / L) and euthanized by cerebral commotion. The gill arches were removed, bathed in hot water (55°C) and fixed in 70% ethanol for further quantification and identification. With the help of a stereomicroscope, all Monogenea specimens were mounted in Hoyer's and identified according to specialized literature. Prevalence indices were calculated according to Bush et al. (1997). A total of 93 parasites specimens were found, with the highest prevalence reported for *Cichlidogyrus sclerosus* Prince and Kirk, 1967 (61%), followed by *C. tilapia* Paperna, 1960 (16%), *C. dossouï* Döuellou, 1993 (10%) *Enterogyrus cichlidarum* Paperna, 1963 (8%), *C. halli* Prince and Kirk, 1967 (4%) and *Scutogyrus longicornis* Paperna and Thurston, 1969 (1%). Parasitism by monogenean is common in fish farms around the world. Therefore, in order to contain economic losses caused by massive mortalities, cultivation environments must provide adequate water quality conditions in order to avoid eutrophic water with excess of organic matter and favorable to the proliferation and spread of these parasites. In a study conducted by Lesssuttichawal Theerawoot in Thailand with *O. niloticus* a total of seven species of monogenean were identified from cyprinid and clariid fish. In the state of Santa Catarina, southern region of Brazil, *E. cichlidarum* was reported parasitizing the stomach of cultured tilapia for the first time in 2007. In the same region and culture conditions, specimens of *C. sclerosus*, *C. halli*, *C. thurstonae* Ergens, 1981, *C. tilapia* and *S. longicornis* were found parasitizing the gills of *O. niloticus*. Parasitic surveys already carried out in fish farms in this region have shown that *C. sclerosus* is commonly the most prevalent species of the genus, corroborating with the results obtained here. In the present study, nine specimens of *C. dossouï* were found parasitizing the gills of *O. niloticus* grown in earthen ponds. *C. dossouï* has a wide geographic distribution and host specificity, and has been reported so far in nine different cichlid species and six countries, none of them in South America. Therefore, this work is the first record of this species in Brazil and, consequently, in the South American continent.
Pyragraphorus hollisae Euzet & Ktari, 1970 infecting pompano, *Trachinotus ovatus* (L.), in captivity

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Species of the genus *Trachinotus* (Teleostei, Carangidae) have been considered as candidates for mariculture (Cunha *et al.*, 2013). Several studies have been developed for contributing to increase its knowledge (Crabtree *et al.*, 2002; Tutman *et al.*, 2004). One of the main possible compromises to take into account are the high parasitological loads in these species. For *Trachinotus ovatus* (L.) (pompano), preliminary studies in the Oceanogràfic aquarium of Valencia revealed the presence of several Pyragraphorus hollisae Euzet & Ktari, 1970 (*Monogenea: Polyopisthocotylea*) on gills. The 35% of the analyzed fish (n=60) showed the parasite (mean intensity=18.6±13.2). Despite the mild parasitation observed, the histopathological study of this species shows particularly severe alterations due to the use of long peduncled clamps (“fire-tong”-shaped) which can induce important damages on fish. Moreover, the peduncles of the clamp provide an increased motility, projecting clamps to grasp distant gill lamellae.

References:
Identification of ectoparasites of Oscar (*Astronotus ocellatus*, Cuvier 1829) in Urmia city, Iran

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Fish parasites and their effects have become increasingly visible during the latest decades in connection with the development of fresh water ornamental fish industries throughout the world. The current study was done with the aim of identification of ectoparasites of Oscar in Urmia city, Northwest of Iran. For this purpose during 1 year, 60 fish were transferred to the laboratory and examined. Each fish was inspected and examined by routine methods. The obtained results revealed that the studied fish were infested by *Ichthyophthirius multifiliis*, Trichodina sp. and Dactylogyrus sp. Overall, 15 of 60 studied fish (25%) were found to be infested by parasites and the remaining were not infested.
Survey of pathological lesions induced by *Silurodiscoides vistolensis* (Sivak, 1932) in European Catfish from North-west of Iran

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Parasitic infection of fish is a serious problem for both natural resources and fish culture farms. It can cause some losses in growth and even mortality of infected fish. This survey was carried out to determine the pathologic lesions induced by *Silurodiscoides vistolensis* in European catfish (*Siluris glanis*) from North-west of Iran. A total of 360 S.glanis(120 fish for each dam) were collected from Aras, Mahabad and Zarrineh dam reservoirs in 2015. In gross examinations the gills were pale and anaemic. Also they were covered by thick mucus and worms were found attaching to the filaments. Pathological study demonstrated that the haptor of the parasite and the hooks adhesiveness to the gill of catfish and caused severe mechanical damages on it. General gill lesions comprised degeneration, tissue defects, haemorrhages, necrosis, atrophy and cell proliferation.
Molecular phylogeny of monogeneans (Monogenea: Dactylogyridae) parasitizing African freshwater catfishes (Siluriformes)

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This study constitutes the first evolutionary investigation of monogenean gill parasites of freshwater catfishes in Africa. Our main aim was to reconstruct molecular phylogeny of dactylogyrids specific to African siluriform fishes and determine their phylogenetic position within other dactylogyridean species parasitizing siluriforms from Africa (Ethiopian region), Asia (Oriental region) and Neotropical region. In 2014, representatives of six dactylogyridean genera (Bagrobdella, Protoancylodiscoides, Quadriacanthus, Schilbetrema, Schilbetrematoides and Synodontella) were collected from ten host species belonging to six families (Bagridae, Claridae, Claroteidae, Malapteruridea, Mochokidae and Schilbeidae) during the field expedition in Sudan (White Nile and Blue Nile). Species identification was performed based on morphological analysis of sclerotized structures and molecular characters (partial 18S rDNA, entire ITS1, partial 28S rDNA). Phylogenetic analyses were conducted using maximum likelihood (ML, RaxML) method.

Our results showed the monophyletic nature of the six dactylogyridean genera. Species infecting African, Asiatic and Neotropical siluriforms were split into two strongly supported clades. Clade A included the four genera of dactylogyrids from Neotropical region, while clade B comprised parasites of siluriform fishes from all three regions. Monogeneans of African catfishes formed two separate clades. Species of Schilbetrema, Schilbetrematoides, Synodontella, Bagrobdella and Protoancylodiscoides clustered as a strongly supported monophyletic group together with Asiatic species Chauhanellus, Hamatopeduncularia and Neocalceostomoides. Quadriacanthus species from Africa formed clade with Asiatic species Q. kobilensis and Bychowskiella pseudobagri.

This study was supported by ECIP (European Centre of IchthyoParasitology); centre of excellence program on the Czech Science Foundation; project No. P505/12/G112
Do the two bagrids *Mystus* Scopoli, 1777 and *Sperata* Holly, 1939 have only *Cornudiscoides* Kulkarni, 1969 (Monogenoidea: Dactylogyridae) or can we split it into two?

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Genus *Cornudiscoides* was established by Kulkarni 1969, from *Mystus tengara* (Hamilton, 1822) at Hyderabad, Andhra Pradesh with *C. heterotylus* as its type species. It has a very narrow host specificity, infecting Bagrids mainly *Mystus* Scopoli, 1777 and *Sperata* Holly, 1939 (earlier also known as Mystus). To date, 22 species have been reported in Southeast Asia, including India. Of 16 valid Indian species, three are from *M. tengara* (Hamilton, 1822), namely *C. heterotylus* Kulkarni, 1969, *C. microtylus* Kulkarni, 1969 and *C. megalorchis* Kulkarni, 1969; five from *M. vittatus* (Bloch, 1794) *C. proximus* Gusev, 1976, *C. geminus* Gusev, 1976, *C. vittati* Dubey, et al., 1992, *C. gomtiai* Agrawal and Vishwakarma, 1996 and *C. agarwali* Agrawal and Vishwakarma, 1996 and four in *M. bleekerae* (Day, 1877), *C. bleekerae* Agrawal and Vishwakarma, 1996, *C. susanae* Agrawal and Vishwakarma, 1996 and *C. tukarami* Agrawal and Vishwakarma, 1976; one species from *Mystus cavasius* is *C. sclerovaginalis* Devak and Pandey, 2007; three species from *Sperata aor* (Hamilton, 1822) are *C. mystusi* (Rizvi, 1971) Dubey, et al., 1992, *C. longicirrus* Agrawal et al., 2016 and *C. aori* Agrawal et al., 2016. During survey of these cat fishes, several new species are being encountered in Indian subcontinent. It is most likely that several new species are added and the worms parasitizing two genera *Mystus* and *Sperata* could be different as their morphology is not consistent with each other. A check list is being prepared, besides establishing the validity by using molecular markers.
The use of de novo developed microsatellite markers for population genetic study of *Dactylogyrus vistulae* populations

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Widespread species with preserved gene flow among populations are supposed to display higher levels of genetic variability in contrast to endemic species with small and fragmented populations. *Dactylogyrus vistulae* represent a generalist monogenean parasite with a high number of fish host species. Will genetic variability differ between *D. vistulae* infecting widespread and endemic species?

Microsatellite markers for *D. vistulae* were *de novo* developed using Illumina MiSeq platform. QDD program was used for selecting suitable microsatellite markers and primer design. Based on 30 polymorphic microsatellites, STRUCTURE and GenAlEx programs were applied for the study of genetic structure and variability of *D. vistulae* populations parasitizing widespread fish species sampled in Czech Republic with Euro-Asian distribution and geographically isolated fish species of Mediterranean region (i.e. Greece, Albania, Bosnia and Herzegovina, Italy and Croatia).

Based on preliminary results, populations of *D. vistulae* clustered into two groups separating *D. vistulae* populations of Bosnia and Herzegovina and Italy from those of remaining countries. Overall, no considerable differences in genetic variability were revealed when *D. vistulae* infecting fish originating from Czech Republic and Mediterranean region were compared. However, low variability in *D. vistulae* populations was detected in several particular endemic host species. Genetic variability of *D. vistulae* populations might, therefore, reflect genetic variability of their hosts. Study involving genetic variability of parasite hosts is further needed in order to support this hypothesis. However, microsatellite markers seem to represent valuable tools for the study of genetic structure and variability of *D. vistulae* populations.

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**Eudiplozoon nipponicum**: "Love at first sight?"

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In our study we used 11 samples of experimental model *Eudiplozoon nipponicum* from 2 different localities; family Diplozoidae isolated from common carp (*Cyprinus carpio*). The life cycle of this ectoparasite represents a rare biological phenomenon - two immature individuals (diporpa) meet on the gills of fish, subsequently permanently fuse together and perform X-shape body. The fusion of two individual larvae evokes many fundamental genetic, biochemical and immunological questions, which need to be addressed. Thus far, this topic was rather overlooked and there is no satisfactory explanation how the two partners find, recognize and stimulate each other for fusion. We applied whole genome scanning method - amplified fragment length polymorphism (AFLP) to find out if the permanently fused worms share genetic pattern specific for the “couple” or if they pair up randomly. Using this approach we were able to discover strong geographical pattern that might be key to understand the mystery behind diporpae paring. It seems that individuals from the same locality do not only share geographical pattern but also have inner population substructuring, which is showing the level of genetic similarities between fused individuals.

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