

DGP2015

34th annual meeting
March 3rd–6th 2015
in Magdeburg



**PROGRAM
ABSTRACTS**

LIST OF PARTICIPANTS

Organizer:
Helge Norf
Ute Risse-Buhl
Markus Weitere

Department River Ecology
Helmholtz Center for Environmental Research
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LPB

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DEAR PARTICIPANTS OF THE DGP 2015 IN MAGDEBURG!

Welcome to the 34th annual meeting of the German Society for Protozoology. A total of 85 participants will join the meeting organized by the Department River Ecology of the Helmholtz-Centre for Environmental Research - UFZ. We are very pleased that about half of the contributions will be given by young researchers presenting their Bachelor's, Master's, PhD thesis or PostDoc projects. We are also very pleased to welcome one national and three international plenary speakers to this DGP meeting.

The meeting will cover a broad range of research topics within protozoology, including aquatic and terrestrial ecology, cell physiology, species interactions, evolution, diversity and molecular evolution. We are looking forward to fruitful discussions after the presentations. The numerous social activities will give room for discussions of research questions and the establishment of new collaborations.

The official conference language will be English, since the meeting becomes more and more attractive for non-German-speaking people.

Welcome also to Magdeburg, the capital of Saxony-Anhalt. The city's gothic cathedral (the oldest in Germany), the last building of the artist Hundertwasser constructed and the Elbe River are in close vicinity of the venue. With numerous research institutes and two universities, Magdeburg offers a good atmosphere for scientific meetings. We wish you a stimulating and enjoyable meeting!

ORGANIZING COMMITTEE

Helge Norf, Ute Risse-Buhl & Markus Weitere

HELPERS

Désirée Dietrich, Kathleen Kirschner, Christine Anlanger, Lino Parlow, Martin Diener, Nicole Oberhoffner, Steffen Geisthardt



OUR THANK GOES TO THE FOLLOWING SPONSORS AND SUPPORTERS

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<http://www.zeiss.de>

Schweizerbart'sche Verlagsbuchhandlung

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VENUE

The conference will be held at Roncalli-Haus, which is located in the city center of Magdeburg.

Roncalli-Haus e.V., Max-Josef-Metzger-Str. 12/13, 39104 Magdeburg

Tel.: (0391) 596 1400, Fax: (0391) 596 1440

Please check in your rooms at the reception of the Roncalli House.

Complementary WLAN access is provided on the 2nd and 3rd floor of the Roncalli-Haus. Please ask for a password at the check in or registration.

REGISTRATION

Members of the pre-conference workshop have the opportunity to register before the start of the workshop. Official registration starts **Wednesday 3rd March at 4 pm**. If the registration desk is unoccupied, please contact a member from the organizing committee. The fee for daily registration is 60 €.

PRESENTATIONS

All talks will be presented in the Lecture hall at the 5th floor of the Roncalli-Haus. The hall is equipped with computer (PC and Mac) and beamer technology. If you need any special equipment please contact a member of the organizing committee. Posters can be viewed at the 3rd floor during the whole meeting. There are special poster sessions for which we kindly ask presenters to be at their posters for discussions.

REFRESHMENT BREAKS

Coffee and refreshments will be made available during breaks outside the meeting room.



INSTRUCTIONS FOR POSTERS AND THEIR SHORT PRESENTATION

The poster format will be A0 portrait. You have the opportunity to present your poster to the entire audience on **Wednesday 3rd March**. Please prepare a 2 min presentation. You can hand in your presentation on CD or USB-stick in the Lecture hall latest in the break before the poster session.

INSTRUCTIONS FOR TALKS

The time for an oral presentation is 15 min. The presentations should not exceed **12 min to give room for 3 min discussion**. Beamer and computer (PC and Mac) will be available in the Lecture hall. Please bring your presentation on CD or USB-stick. You can hand in your presentation in the Lecture hall latest in the break before your session starts.

SOCIAL EVENTS

The get-together will be on Tuesday evening, **3rd March**, in the **Bar Chaplin** which is located in the basement of the Conference Venue.

On Wednesday **4th March** you have the opportunity to join a **guided tour** through Magdeburg's Old Town, which will take you into the history of the former Imperial City or through the ancient Magdeburg Cathedral. Tours will last approximately 1 hour. You will spend this time either outside or in an unheated cathedral, so **you will need warm clothes and shoes!**

On Thursday **5th March** we would like to welcome you to the **Conference dinner**, which will officially start with a guided tour through the Gruson greenhouses. The greenhouses are known as a unique collection of exotic plants. After this tour we will take a short walk to the 'Gesellschaftshaus', where the conference dinner will be held. This beautiful building from the 18th century offers us a sophisticated atmosphere for a tasty buffet and – for those who like – subsequent dance.



HELMHOLTZ CENTRE FOR ENVIRONMENTAL RESEARCH – UFZ

The Helmholtz Association contributes to solving major challenges facing society, science and the economy with top scientific achievements in six research areas: Energy, Earth and Environment, Health, Key Technologies, Structure of Matter, Transport and Space. With about 34,000 employees in 18 research centres and an annual budget of approximately 3.4 billion €, the Helmholtz Association is Germany's largest scientific organization. Its work follows in the tradition of the great natural scientist Hermann von Helmholtz (1821-1894).

The Helmholtz Centre for Environmental Research – UFZ was established in 1991 and has more than 1,100 employees that are divided in the three main locations Leipzig, Halle/Saale and Magdeburg. They study the complex interactions between humans and the environment in cultivated and damaged landscapes. The scientists develop concepts and processes to help secure the natural foundations of human life for future generations.

The Department of River Ecology at the UFZ addresses ecological interactions and chemical pollutions in running waters. The activities focus on central Germany with different streams and rivers in the Bode-Saale-Elbe network. Furthermore, other lotic systems in Europe as well as in other countries are subject of specific research projects. The scientific expertise of the department comprises ecological interactions in both microbial and macrobial communities with a focus on microbial biofilms as well as the dynamics of contaminants. For analysis of bulk water, planktonic and interfacial processes a broad range of tools and advanced techniques are available including: research vessel, mesocosm facilities, chemical analytical techniques (TXRF, GC-MS, HPLC-MS), isotope laboratory, flow-field flow fractionation, laser scanning microscopy (1-photon and 2-photon) and digital image analysis.



SCHEDULE

Tuesday, 3rd March 2015

12:30–13:00 Registration for participants of the workshop

Pre-Conference Workshop

Will take place in the seminar room at the UFZ Magdeburg, Brückstr. 3a, 39114 Magdeburg.

13:00–17:00 **Alexandra Jeuck & Hartmut Arndt**

A user-friendly guiding tour through the identification of morphotypes of heterotrophic flagellates in freshwater samples

16:00–20:00 Check-in and Registration at Roncalli-Haus

19:00–23:00 **Get-Together in Bar Chaplin (base of the Roncalli-Haus)**



Wednesday, 4th March 2015

09:00 Welcome by **Jens Boenigk & Markus Weitere**

Diversity of protists, Chair: Hartmut Arndt

09:15–10:00 **David Bass**

The expanding eDNA toolbox for protistan parasitology

10:00–10:15 **Heinz Martin Schumacher, Elke Heine-Dobbernack**

Deposit of protozoa at DSMZ – Considerations concerning accession and distribution

10:15–10:30 **Sascha Krenek, Thomas U. Berendonk, Sergei I. Fokin**

How to deal with cryptic diversity in protistology – a case example in genus *Paramecium*

10:30–11:00 **Coffee Break**

11:00–11:15 **Dominik Forster, Micah Dunthorn, Thorsten Stoeck, the BioMarKs Consortium**

Exploring and estimating microbial eukaryote diversity in distinct marine compartments along the European coastline

11:15–11:30 **Anna Gimmler, Ralf Korn, Thorsten Stoeck, the TARA Oceans Consortium**

Diversity and distribution of planktonic ciliates in the photic zone of the world's oceans

11:30–11:45 **Frank Nitsche, Helge Thomsen, Daniel Richter**

Choanoflagellate systematics and taxonomy

11:45–12:00 **Leonardo Fernández, Enrique Lara, María Romina Schiaffino, Irina Izaguirre**

A lake-dwelling microalga (Bathycoccaceae) exhibits population genetic structure among lacustrine populations in the southern Argentinean Patagonia

12:00–12:15 **Jingyun Chi, Micah Dunthorn**

An inventory of meiotic genes in Alveolates reveals an independent loss in ciliates and dinoflagellates

12:30–14:00 **Lunch break**



Ecology of terrestrial protists, Chair: Edward Mitchell

14:00–14:45 **Michael Bonkowski, Maike Hünninghaus, Kenneth Dumack, Dörte Dibbern, Tillmann Lueders, Tim Ulrich, Anna Maria Fiore Donno, Stefan Geisen**

Species description of the soil dwelling *Lecythium terrestris*, a fungi and algae feeding protist based on morphology and SSU sequence data (Chlamydomphryidae).

14:45–15:00 **Kenneth Dumack, Marina E.H. Müller, Michael Bonkowski**

Species description of the soil dwelling *Lecythium terrestris*, a fungi and algae feeding protist based on morphology and SSU sequence data (Chlamydomphryidae).

15:00–15:15 **Wilhelm Foissner**

Protist distribution: 100 new neotropical soil ciliates emphasize moderate ciliate endemism

15:15–15:30 **Leonardo Fernández, Reinaldo Rivera, Bertrand Fournier, Cristián Hernández, Enrique Lara, Edward A. D. Mitchell**

Soil testate amoeba diversity follows a unimodal latitudinal pattern in southwestern South America (18°–56° S) caused by short- and long-term processes.

15:30–16:00 Coffee Break

16:00–16:15 **Manfred Wanner, Barbara Seidl-Lampa, Axel Höhn, Daniel Puppe, Michael Sommer**

Culture growth of testate amoebae under different silicon concentrations

16:15–16:30 **Monika K. Reczuga, Christophe Seppey, Ildikò Szelez, Bertrand Fournier, David Singer, Enrique Lara, Edward A. D. Mitchell**

Response of soil micro-eukaryotes to cadaver decomposition as assessed by high throughput sequencing

16:30–17:15 **Poster presentations**

17:15–18:15 **Poster session & Beer**

20:00–21:30 **Guided tours through**

Magdeburgs Old Town or Magdeburgs Cathedral

**Thursday, 5th March 2015****Ecology of aquatic protists, Chair: Thomas Berendonk****09:00–09:45 Anna Romani**

Responses of fluvial biofilms to temperature: the relevance of microbial interactions

09:45–10:00 Markus Weitere, Martina Erken, Helge Norf, Jennifer Wey

Microbial food web interactions within biofilms

10:00–10:15 Helge Norf, Steffen Geisthardt, Sebastian Hasselhuhn, Norbert Kamjunke, Thomas R. Neu, Markus Weitere

Spatial competition between microalgae and stalked protozoa in biofilms developed under different light regimes

10:15–10:30 Ute Risse-Buhl, Christine Anlanger, Christian Noss, Andreas Lorke, Thomas R. Neu, Markus Weitere

The role of hydrodynamics in shaping the composition of biofilms in two streams of contrasting trophic

10:30–11:00 Coffee Break**11:00–11:15 Claudia Seiler, Tamara Thieser, Ursula Gaedke, Thomas U. Berendonk, Markus Weitere**

Role of protozoans in plankton - biofilm systems: Dynamics in morphology of bacteria and their processing within the food web

11:15–11:30 Sebastian Hess

Hunting for agile prey: Two novel leptophryid amoebae (Vampyrellida, Cercozoa) devouring planktonic freshwater algae



Cell physiology, Chair: Michael Schweikert

- 11:30–11:45 **Helmut Plattner**
Calcium signaling in protozoa and beyond
- 11:45–12:00 **Anna Busch, Sebastian Hess, Michael Melkonian**
The cytoskeleton of viridiraptorid amoeboflagellates in free-living and trophic states
- 12:00–12:15 **Lisa Siegmund, Hendrike Dürichen, Anke Burmester, Johannes Wöstemeyer**
Ingestion of bovine-serum-albumin-methacrylate microparticles in the ciliate *Tetrahymena pyriformis*
- 12:15–12:30 **Sebastian Dirren**
Promiscuous or conservative symbiont acquisition in the genus *Nuclearia*?

12:30–14:00 **Lunch break**

14:00–15:00 **Poster session & Coffee**

Presentations of Grell Awards, Chair: Jens Boenigk

- 15:00–15:30 **Agnes Weiner**
Genetic diversity and biogeography of living planktonic foraminifera
- 15:30–16:00 **Sebastian Hess**
Algivoracious freshwater protists of the phylum Cercozoa (Rhizaria) – Structure, life histories and evolution
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- 16:15–17:15 **General meeting of the DGP**
- 17:15–17:30 **General meeting of the DGPF**
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- 18:00–19:00 **Guided tour through Gruson greenhouses**
- 19:00 **Social evening in the 'Gesellschaftshaus'**
and awarding of the prices for the best posters and best talks
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**Friday, 6th March 2015****Parasitology, Chair: Julia Walochnik**

09:30–10:15

Craig Roberts

Toxoplasma gondii induced neuro-metabolomic, neuro-immunological and behavioural changes in mice

10:15–10:30

Marcel Deponte

From bacteria to parasites - prokaryotic ancestry and gene fusion of a dual localized peroxidase in malaria parasites

10:30–10:45

Adeline Loyau, Dirk Schmeller

Altitudinal distribution of zooplankton and the occurrence of *Batrachochytrium dendrobatitis*

10:45–11:15

Coffee Break

11:15–11:30

Dirk Schmeller, Adeline Loyau

Micropredators dictate the dynamics of a fungal disease

11:30–11:45

Julia Walochnik, Ute Scheikl, Eva-Maria Haller-Schober

Monitoring *Acanthamoeba* infections in Austria

11:45–12:00

Sabina Wodniok, Elif Demir, Jens Boenigk, Martin Lohr

Molecular evidence for plastids in *Spumella* (Heterotrophic Chrysophyceae)

12:00–12:15

Farewell

12:30–14:00

Lunch break



PRE-CONFERENCE WORKSHOP

A USER-FRIENDLY GUIDING TOUR THROUGH THE IDENTIFICATION OF MORPHOTYPES OF HETEROTROPHIC FLAGELLATES IN FRESHWATER SAMPLES

Alexandra Jeuck & Hartmut Arndt

University of Cologne, Germany, alexandra.jeuck@uni-koeln.de, hartmut.arndt@uni-koeln.de

The workshop will introduce to the main morphotypes of the most abundant groups of heterotrophic flagellates occurring in freshwater habitats (e.g. choanoflagellates, free-living kinetoplastids, euglenids, bicosoecids, chrysoomonads, ciliophryids, dinoflagellates, cercozoans, apusomonads, ancyromonads, cryptophyceans) and will refer to the current knowledge of the systematic position of relevant groups. We will use cultures to practice the identification of the groups using light microscopy (with video-recording). In addition, we will examine field samples from the River Elbe and participants are encouraged to bring also their own environmental samples to get an idea of flagellate community structure in the field using live-observations. Our workshop is especially meant for non-experienced participants.



PLENARY LECTURES

THE EXPANDING EDNA TOOLBOX FOR PROTISTAN PARASITOLOGY

David Bass

Natural History Museum, London, Great Britain

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Parasites occur on all main branches of the whole eukaryotic tree of life, and are likely account for many of the uncultured protistan lineages that are 'discovered' by environmental DNA sequencing (eDNA) studies. However, many parasites are missed by these broad eukaryotic surveys for a range of regions – for example evolutionary divergence, strong associations with hosts, highly discontinuous distributions in environmental matrices, and scarcity. After a phylogenetic overview of protistian parasites I will present case studies of how various eDNA and complementary methods have and can be used to characterize protistan parasites, detect emerging diseases, and understand how these parasites function in ecosystems. I will also discuss some of the pitfalls of large-scale taxonomic annotations of high throughput sequencing datasets, relevant both to parasites and free-living protists, and suggest some basic requirements to make these more reliable, informative, and comparable across studies.



RESPONSES OF FLUVIAL BIOFILMS TO TEMPERATURE: THE RELEVANCE OF MICROBIAL INTERACTIONS

Anna M. Romani

Institut d'Ecologia Aquàtica, Universitat de Girona, Girona, Spain

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Biofilms are dynamic players in biogeochemical cycling in rivers, mediating a significant proportion of the carbon turnover and dominating the ecosystem metabolism in many aquatic systems, being a major component for the uptake, storage and cycling of available organic matter. Aquatic biofilms are subjected to environmental stressors like those provoked by climate change. The different microorganisms composing a biofilm (bacteria, protozoa, algae, small metazoans) may show a differential response to temperature. Since in a biofilm there is a physical proximity and usually a strong relationship between the different components, a shift in their biomass distribution may affect their interactions and the whole biofilm metabolic outcome. It is then relevant to discern whether the predicted global change scenario would affect microbial biofilm structure and function. Several experiments have been performed by analysing the effect of 2.2–4.3°C increase (predicted for climate change scenarios by 2100) on biofilm structure and function. Results show that biofilm responses to warming are highly depending on nutrient and organic matter availability, major responses occurring under eutrophic conditions. Also, the biogeochemical potential of the stream biofilm is modified by warming, enhancing the decomposition of recalcitrant carbon compounds and the use of peptides, which seems to be also linked to changes in the biomass contribution of the different biofilm components as well as changes in prokaryote community composition. Studies using a larger range of temperatures (10–26°C) indicated a high sensitivity of biofilm grazers (rotifer) which indirectly affect the whole biofilm biomass. Results suggest a highly significant effect of microbial biofilm interactions on its response to warming, the temperature cascade effect on the biofilm microbial food web (from small metazoans to bacteria) modifying the biofilm functioning.



THE DIVERSITY AND FUNCTIONS OF PROTISTS IN SOIL: PROBLEMS AND PROGRESS

Michael Bonkowski, Maïke Hünninghaus, Kenneth Dumack, Dörte Dibbern, Tillmann Lueders, Tim Urich, Anna Maria Fiore Donno, Stefan Geisen

University of Cologne, Germany

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Protists are at the base of soil food webs, connecting the flows of energy from plants through bacteria and fungi to higher trophic levels in the soil food web. Laboratory experiments demonstrated strong effects of protists on plant growth, but despite their functional importance, our knowledge on the community composition of protists in soil is still highly contradictory. Despite environmental sequencing studies based on the SSU rRNA gene have revealed a huge diversity of previously unknown protists, new biases are being introduced by molecular techniques, obscuring the true protist diversity in soils. Fundamental problems are created by (i) the lack of SSU rRNA reference sequences for major protist clades, (ii) vast mislabelled sequences in public databases, and (iii) the enormous phylogenetic diversity of protist taxa, all commonly resulting in erroneous species assignments. Further biases are introduced by the PCR step that usually precedes high-throughput sequencing (HTS) of SSU rRNA gene studies. “General” eukaryotic primers are often applied to decipher the community structure of protists, but these primers are in fact far from being truly universal. While some taxa within this subset will be overrepresented due to preferential PCR amplification, other (dominant) taxa are entirely lacking, resulting in a strongly biased view of the protist community in soils. I will present some recent data on progress in molecular methods to describe the protist diversity in soils, and on linkages of the protist micro-food web to the root microbiome of plants.



TOXOPLASMA GONDII INDUCED NEURO-METABOLOMIC, NEURO-IMMUNOLOGICAL AND BEHAVIOURAL CHANGES IN MICE

Craig Roberts

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Toxoplasma gondii infects approximately 30% of the world's population and is responsible for ocular disease, neurological disease and abortion. In recent years, the ability of the parasite to manipulate the behaviour of infected mice and rats and alter personality attributes of humans has been reported. There is now compelling evidence that *T. gondii* infection is associated with the development of schizophrenia and potentially depressive disorders. The work presented demonstrates the role of arginine metabolism in control of *T. gondii* in the acute phase of infection. We also demonstrate that the behaviour of BALB/c mice, which are resistant to toxoplasmic encephalitis, and only develop small numbers of cysts is altered during *T. gondii* infection. In spite of relatively little pathology in the brains of these mice, transcripts for a number of immunological mediators are raised. Metabolomic analyses of brains by Liquid Chromatography Mass Spectrometry (LCMS) demonstrates alteration of key metabolites that might be responsible for the observed behavioural changes. Together this work demonstrates multifarious effects of immune induced changes to host cell metabolism.



GRELL AWARDS

GENETIC DIVERSITY AND BIOGEOGRAPHY OF LIVING PLANKTONIC FORAMINIFERA

Agnes Weiner

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In my PhD thesis with the title “Genetic diversity, biogeography and the morpho-genetic relationship in extant planktonic foraminifera” I studied the diversity, distribution and evolution of living planktonic foraminifera in order to advance our biological knowledge on this important group of protists. Planktonic foraminifera are exclusively marine protists that are found globally in the ocean. They construct calcite shells that accumulate in the sediment and are used as proxies in micropaleontological studies. Traditionally, modern planktonic foraminifera have been classified into about 40 species on the basis of morphological characteristics of their calcite shell. Molecular investigations however uncovered an unexpectedly high genetic diversity within the morphologically defined species, revealing a high number of cryptic species and implying that the biodiversity of the group has been largely underestimated. These cryptic species show distinct biogeographic patterns and ecological adaptations, shedding new light on the processes of evolution in this group of plankton. Importantly, the excellent fossil record of this group makes it possible to place cryptic diversification into a historical context. To this end, I applied a single cell approach to survey the extent of sequence diversity within the SSU rDNA of individual morphospecies and to examine their biogeography, habitats and ecological adaptations. I used phylogenetic reconstructions and molecular clock estimates to track the relationships and evolution of cryptic species. In addition, I tried to establish a connection between genetic diversity and morphological variability of the calcite shells by morphometric analysis in order to taxonomically revise the morphospecies and to create a connection between living specimens and the fossil record. From a biogeographic perspective most morphospecies show a cosmopolitan distribution in the world ocean throughout their preferred temperature province. On the level of cryptic species, however, my studies revealed the presence of complex distribution patterns, like depth parapatric separation or competitive exclusion with adaptations to different environmental parameters. These distribution patterns allowed me to get an understanding of the processes that generate the extensive genetic diversity and to identify factors that promote divergence in a group of plankton with cosmopolitan distribution and high potential dispersal rates.



ALGIVOROUS FRESHWATER PROTISTS OF THE PHYLUM CERCOZOA (RHIZARIA) – STRUCTURE, LIFE HISTORIES AND EVOLUTION

Sebastian Hess

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In freshwater ecosystems various elusive protists exist that consume microalgae in a selective and specialised manner. Some of these ‘parasitoid’ micro-organisms have been known since the mid of the 19th century and were studied by several naturalists. However, difficulties in long term maintenance and the convoluted taxonomic history of such forms strongly impeded further research. The present knowledge about protistan parasitoids of freshwater algae, in particular concerning phylogenetic affiliations and ecology, is extremely poor. During the studies presented, amoeboid and flagellate protists feeding on zygmatophycean and chlorophycean green algae (Viridiplantae) have been isolated from natural freshwater samples and resulted in single cell derived cultures. Based on culture material, cell morphology, feeding processes, locomotional behaviour and life histories have been studied by means of light microscopy and time lapse photography. Feeding experiments were conducted to reveal the food range specificity and SSU rDNA sequences of the strains were subjected to molecular phylogenetic analyses to allocate the organisms of interest in the tree of life.

The phenotypic and phylogenetic information about the algivorous protists investigated, resulted in a comprehensive characterisation of vampyrellid representatives (Vampyrellida, Cercozoa, Rhizaria) and a novel family of algivorous nanoflagellates, the Viridiraptoridae. This family represents a morphologically and ecologically distinct lineage of glissomonad flagellates (Cercozoa, Rhizaria), whose ultrastructure has been studied by transmission and scanning electron microscopy. In particular the flagellar transitional region and basal apparatus, both reconstructed from serial sections, revealed ultrastructural traits that agree with the phylogenetic placement of the Viridiraptoridae within Glissomonadida. Furthermore, an acorn/V-shaped filament system was discovered at the proximal end of the flagellar transitional region and used to establish a basal body triplet numbering system for flagellate cells of the Rhizaria.



ABSTRACTS OF TALKS

sorted alphabetically by first author

THE CYTOSKELETON OF VIRIDIRAPTORID AMOEBOFLAGELLATES IN FREE-LIVING AND TROPHIC STATES

Anna Busch, Sebastian Hess, Michael Melkonian

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Viridiraptorid amoebflagellates (Cercozoa, Rhizaria) currently comprise two genera, *Viridiraptor* and *Orciraptor*, which feed on freshwater green algae and occur in motile flagellate states (during dispersal) and amoeboid states (during feeding). As 'protoplast feeders' *Viridiraptor* and *Orciraptor* perforate algal cell walls and feed exclusively on the cell contents by phagocytosis. This involves 1) attachment to the algal cell wall, 2) perforation of the cell wall, 3) invasion of the algal cell (*Viridiraptor invadens*) or extraction of the algal plastid (*Orciraptor agilis*), and 4) phagocytosis. In the light of these elaborate feeding processes some questions concerning the viridiraptorid cytoskeleton arise: What is the structure/distribution of the cytoskeletal elements actin and tubulin in free-living and trophic states? To which extent are these cytoskeletal elements involved in the feeding process? AND What generates the force required for the invasion process of *Viridiraptor invadens*? To address these questions we developed a sequential staining method to visualise F actin and α tubulin in viridiraptorid cells attached to the filamentous food algae in various feeding stages as well as the algal cell wall. Time lapse observations on living *Viridiraptor* cells during invasion and scanning electron microscopy of cell wall perforations produced by *Orciraptor agilis* supplement our data. Viridiraptorid cells in the free living flagellate state (gliding or swimming) displayed a peripheral, cage-like system of microtubules, whereas F-actin was not detectable. In surface-attached, amoeboid cells F-actin was present at the adhesion sites and the microtubular cytoskeleton persisted, but seemed to be less well organised than in the flagellate state. Furthermore, F-actin is apparently involved in the feeding activities of viridiraptorids in a species specific manner: The arrangement of F actin rich cell extensions in feeding *Orciraptor* cells suggests that these pseudopodia are used to bend away a lid like cell wall disc (resulting from a c shaped perforation pattern) and to grab the plastid. In contrast, *Viridiraptor* was observed to send a single, actin rich pseudopodium through a much smaller perforation into the algal cell.



Subsequently, this pseudopodium broadened and formed conspicuous hyaline zones, which apparently generate the force required for the cell to slip from one side of the perforation to the other. These hyaline zones correspond to a prominent F actin rich ring that presumably plays a crucial role during prey-cell invasion.

AN INVENTORY OF MEIOTIC GENES IN ALVEOLATES REVEALS AN INDEPENDENT LOSS IN CILIATES AND DINOFLAGELLATES

Jingyun Chi, Micah Dunthorn

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Two crossover pathways exist in meiosis. Class I crossover pathway uses meiosis-specific proteins, some of which comprise the synaptonemal complex. Class II crossover pathway uses proteins that are also involved in mitosis. Both pathways likely evolved in the ancestor of all eukaryotes; however, their current distributions in protists are unknown. To evaluate the pathways in the alveolates, we inventoried meiotic genes in available genomes. Results are consistent with an independent loss/reduction of class I crossover pathway in ciliates and dinoflagellates. We hypothesize that these losses/reductions are due to ciliates and dinoflagellates having non-canonical genome architectures.



FROM BACTERIA TO PARASITES - PROKARYOTIC ANCESTRY AND GENE FUSION OF A DUAL LOCALIZED PEROXIDASE IN MALARIA PARASITES

Marcel Deponte

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Horizontal gene transfer has emerged as a crucial driving force for the evolution of eukaryotes. This also includes *Plasmodium falciparum* and related economically and clinically relevant apicomplexan parasites, whose rather small genomes have been shaped not only by natural selection in different host populations but also by horizontal gene transfer following endosymbiosis. However, there is rather little reliable data on horizontal gene transfer between animal hosts or bacteria and apicomplexan parasites. Here we show that apicomplexan homologues of the peroxidase peroxiredoxin 5 (Prx5) have a prokaryotic ancestry and therefore represent a special subclass of Prx5 isoforms in eukaryotes. Using two different immunobiochemical approaches, we found that the *P. falciparum* Prx5 homologue is dually localized to the parasite plastid and cytosol. This dual localization is reflected by a modular *Plasmodium*-specific gene architecture consisting of two exons. Despite the plastid localization, our phylogenetic analyses contradict an acquisition by secondary endosymbiosis and support a gene fusion event following a horizontal prokaryote-to-eukaryote gene transfer in early apicomplexans. The results provide unexpected insights into the evolution of apicomplexan parasites as well as the molecular evolution of peroxiredoxins, an important family of ubiquitous, usually highly concentrated thiol-dependent hydroperoxidases that exert functions as detoxifying enzymes, redox sensors and chaperones.

References: Djuika CF, Huerta-Cepas J, Przyborski JM, Deil S, Sanchez CP, Doerks T, Bork P, Lanzer M, and Deponte M. (2015) *Microbial Cell* 2:5-13.



PROMISCUOUS OR CONSERVATIVE SYMBIONT ACQUISITION IN THE GENUS *NUCLEARIA*?

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Intimate associations between organisms have been observed for a long time. One can just intuitively argue that the possibility of interactions/interconnections increases, if spatial distance is reduced. Knowing that such interactions can be manifold, we use the term symbiosis in a very general manner. Thus, we simply call the phenomenon of a close living together of organisms belonging to different species a 'symbiosis'. We focused on members of the amoeboid genus *Nuclearia* (Opisthokonta, Nucleariidae) that often live in symbiosis with ecto- and endosymbiotic bacteria. Here we define ectosymbionts to be bacteria usually nicely arranged in the amoebae's mucous sheath, whereas endosymbionts are located inside cells. We isolated 22 *Nuclearia* strains from five different Swiss lakes, and surprisingly, only about half (13) of them were associated with at least one symbiont. All of the isolated amoebae were first characterized based on the few formerly described morphological traits. Since these features are often not distinctive, we sequenced the 18S rRNA genes for a phylogenetic tree reconstruction. The obtained tree showed four already established monophyletic branches (made up by the six so far sequenced species of the genus) and an additional cluster formed by two new isolates. A very heterogeneous picture emerged by highlighting *Nuclearia* strains with associated symbionts in the tree. Apart from one cluster which included only *Nuclearia* spp. with symbiotic bacteria and two clusters with no symbionts, we also found mixed clusters that were composed of nucleariid amoebae with and without symbionts. By analysing 16S rRNA genes of symbiotic bacteria, the picture got even more 'obscure'. Although we have already identified six different symbiotic bacterial strains, it seems that we still are only scratching the surface of the symbionts' diversity. On the one hand strains of *Nuclearia thermophila* harbour the same endosymbiont even when isolated from different lakes. This points to a rather obligate and persistent interaction. On the other hand we found two isolates of *Nuclearia delicatula* to be associated with different endosymbiotic bacteria. In summary, it seems that we are dealing with a group of opisthokonts that is keenly experimental regarding symbiotic interactions. As far as we know there are no other documented cases of protists belonging to the Opisthokonta with prokaryotic symbionts. This fact is especially remarkable considering the importance of symbiotic interactions for higher opisthokonts. Thus, Nucleariidae represent an ideal model group to study the basic principles of symbioses between opisthokonts and prokaryotes.



SPECIES DESCRIPTION OF THE SOIL DWELLING LECYTHIUM TERRESTRIS, A FUNGI AND ALGAE FEEDING PROTIST BASED ON MORPHOLOGY AND SSU SEQUENCE DATA (CHLAMYDOPHRYIDAE).

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Testate amoebae have been frequently studied by protistologists, but still little information is available on some groups like members of the Chlamydomphryidae. These species are difficult to culture and therefore only quantitative information on their morphological characters are rare. We describe *Lecythium terrestris*, as a new species of testate amoebae, present its phylogenetic position and the life cycle based on an established culture. We could confirm its phylogenetic position among the Cercozoa where it groups closely to Pseudodiffugiidae. Furthermore we investigated the life cycle and ecology of *L. terrestris* and describe it as a rather general grazer of fungi and algae.

SOIL TESTATE AMOEBEA DIVERSITY FOLLOWS A UNIMODAL LATITUDINAL PATTERN IN SOUTHWESTERN SOUTH AMERICA (18°-56° S) CAUSED BY SHORT- AND LONG-TERM PROCESSES.

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Soil testate amoeba diversity follows a unimodal latitudinal pattern in southwestern South America (18°-56° S) caused by short- and long-term processes. While for metazoans there is a growing recognition that the latitudinal diversity gradient (LDG) is the result of a balance between short- and long-term processes, there is a yet unresolved debate on the existence of LDG in free-living microorganisms and the mechanisms governing their diversity patterns. Moreover, most studies on microbial biogeography and macroecology are based on data from the northern hemisphere and/or are biased toward aquatic taxa, while soil microorganisms remain comparatively poorly documented. In this study, we evaluated whether soil testate amoebae from the southwestern South America (18° S- 56° S) exhibit a LDG and the possible ecological (short-term) and historical-evolutionary (long-term) causes for this pattern. The LDG was evaluated using a dataset that includes 234 species and summarizes over 170 years of taxonomic studies in the region. Nine classical



ecological hypotheses invoking a relationship between richness and short-term processes were tested: (a) the Rapoport effect (b) climatic variability; (c) species-energy; (d) species-water; (e) water-energy balance; (f) productivity effect; (g) habitat heterogeneity; (h) species-area; and (i) the mid-domain effect. The influence of long-term processes was indirectly tested by: (a) contrasting the LDG observed with the predicted values generated from a null-model of species richness; (b) analyzing the latitudinal variation in the taxonomic distinctness; and (c) assessing the degree of nestedness at different taxonomic levels. Our results showed that these organisms exhibit a unimodal LDG with a peak of richness at mid-latitudes. A simulation analysis showed that this diversity gradient is robust to latitudinal sampling artifacts. We found support for two of the nine ecological hypotheses tested: Rapoport effect and water-energy balance. This finding suggested that this diversity trend is mainly driven by ecological processes involving a latitudinal trade-off in water-energy dynamics. Long-term processes also seemed important in structuring the LDG. Mid-latitudes exhibited an empirical diversity peak that was higher than the values generated by a null-model and a high taxonomic distinctness. Also, testate amoeba assemblages were nested at different taxonomic levels along the latitudinal gradient studied. This outcome suggested that large-scale environmental changes that experimented southwestern South America in the past affected species richness and composition by promoting different migration, extinction and speciation rates. While the LDG observed for these microorganisms may seem unusual, this pattern matches the distribution of many metazoans in this region of the planet. Our results therefore suggested that the origin and maintenance of the observed pattern lies in the combined influence of short- and long-term processes for both free-living microorganisms and macroscopic organisms. Therefore, we conclude that free-living soil microorganisms meet the patterns documented for metazoans and that diversity patterns of all living entities are governed by similar mechanisms at broad spatial scales.



A LAKE-DWELLING MICROALGA (BATHYCOCCACEAE) EXHIBITS POPULATION GENETIC STRUCTURE AMONG LACUSTRINE POPULATIONS IN THE SOUTHERN ARGENTINEAN PATAGONIA

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Protists have huge population sizes, small body sizes and many of them can form dormant stages that allow them to survive in adverse environmental conditions. Accordingly, it is assumed that protists have unlimited gene flow and dispersal and thus do not display population genetic differentiation among populations as these would be prevented by free gene flow. We have analyzed the genetic population structure in a lake-dwelling photosynthetic picoeukaryote (Bathycoccaceae) and tested the hypothesis that there is no population genetic structure among lacustrine populations distributed along the southern Argentinean Patagonia (45° S - 55° S, c. 800 km gradient). These lakes are located in three geographic regions clearly separated by geographical barriers (i.e. the Andes Mountains and the Magellan Strait). Clonal strains were retrieved from six of 13 lakes surveyed and were genotyped using a PCR approach to obtain ITS sequences. Our results revealed the existence of 25 different haplotypes distributed along the studied gradient. Populations isolated by potential barriers to gene flow exhibited a high proportion of exclusive haplotypes (c. four to six haplotypes) suggesting that haplotypes are distributed following a nonrandom geographical pattern. The amplitude of the genetic differences between all populations was moderate to high and always significant (F_{ST} ranged from 0.154 to 0.771). Even geographically close populations exhibited a moderate to high genetic divergence, although the highest genetic divergence was found between two populations that are in areas geographically widely apart. An AMOVA analysis revealed the existence of a strong population genetic structure between those three regions that are separated by geographical barriers. The percentage of variation among regions ($F_{ST} = 22.06\%$), among populations within regions ($F_{SC} = 18.94\%$) and within populations ($F_{CT} = 58.99\%$) were significant and progressively strong. Standard and partial Mantel tests confirmed a significant correlation between the matrix of population genetic distances and the matrix of pairwise geographical distances (standard and partial Mantel tests: $r = 0.50$ and $r = 0.49$ respectively), confirming thus that the genetic similarity between populations is inversely proportional to the geographic distance between them (i.e. isolation by distance). We did not find a significant correlation between the matrix of population genetic distances and the ecological characteristic of each lake (standard and partial Mantel tests: $r = 0.23$ and $r = 0.20$, $p > 0.05$ respectively), suggesting that the measured environmental parameters are not important in shaping the population genetic structure observed. Our study rejects the



null hypothesis that there is unlimited gene flow among lacustrine populations of protists. Instead, lentic protist populations exhibit a robust population genetic structure driven by both the geographical barriers and the geographic distance that exist between lakes.

A LAKE-DWELLING MICROALGA (BATHYCOCCACEAE) EXHIBITS POPULATION GENETIC STRUCTURE AMONG LACUSTRINE POPULATIONS IN THE SOUTHERN ARGENTINEAN PATAGONIA

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I studied 80 soil samples from the neotropis, mainly from Venezuela and some from Brazil and the Galapagos Islands, using the non-flooded Petri dish method and classic and modern taxonomic tools. I identified about 400 species of which about 100 were undescribed, representing 30 new genera and some new families. These data were compared with similar studies from Namibia (Africa) and Central Europe, showing about 60% species overlap. This and some "flagships" (*Condylostomides coeruleus* n.sp., *Sleighophrys pustulata* n.g., n.sp., *Luporinophrys micelae* n.g., n.sp., *Lingulothrix galapagensis* n.g., n.sp., *Cataphractes austriacus* n.g., n.sp., *Notodeviata halophila* n.g., n.sp.) emphasize a moderate ciliate endemism globally and a huge number of undescribed ciliates. The new species are described in a forthcoming monograph.

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EXPLORING AND ESTIMATING MICROBIAL EUKARYOTE DIVERSITY IN DISTINCT MARINE COMPARTMENTS ALONG THE EUROPEAN COASTLINE

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In the context of the BioMarkS project we analyzed RNA sequences of the hypervariable SSU-V4 region obtained from unicellular microbial eukaryotes via high-throughput sequencing. Using a strict quality and taxonomic filtering approach we gained an in-depth look into the distribution of the major microbial eukaryote communities inhabiting the benthic compartments and their overlaying planktonic habitats along the European coastline. We revealed distinct differences in microbial communities, depending on the sampled marine compartment they inhabited. Throughout all analyses, which included traditional diversity indices, as well as plotting the genetic diversity and comparing community compositions, our results strongly indicate that the diversity and OTU richness of benthic communities considerably exceeds the diversity and richness of planktonic communities. Our results also point towards a homogenous distribution of diversity in planktonic habitats in contrast to a more heterogeneous distribution in the benthos. At the same time, our estimations show that we still miss the majority of organisms living in marine sediments. Thus, our current data may only reveal the tip of the sediments' microbial diversity, while the true extent still remains hidden in the benthos. We argue that the main reasons for the observed diversity patterns can be attributed to the adaptations of the organisms to their different life styles and encourage researchers looking for novel diversity to dive deeper into the benthic microbiota of the oceans.



EXPLORING AND ESTIMATING MICROBIAL EUKARYOTE DIVERSITY IN DISTINCT MARINE COMPARTMENTS ALONG THE EUROPEAN COASTLINE

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By exploring ciliate communities of different oceanic regions and layers in the photic zone our goals were the identification of biogeographical distribution patterns and the investigation of environmental factors, which are shaping these distribution patterns. In our study we used data from the TARA Oceans expedition, which sampled the world's oceans on a three-year global voyage to study the structure of planktonic ecosystems and to analyze the dynamics of plankton populations and communities in relation to their physico-chemical environments. Using high-throughput sequencing techniques, more than 6 million sequences of the V9 SSU rDNA, which could be assigned to the phylum Ciliophora, were obtained from 47 sampling stations, including samples from two different layers in the photic zone, around the world's oceans. We analyzed the ciliate diversity in the oceans using diversity indices and statistical methods and we revealed that there were not only differences between ciliate communities from different oceanic regions but also between communities from different layers in the oceans.



HUNTING FOR AGILE PREY: TWO NOVEL LEPTOPHRYID AMOEBAE (VAMPHYRELLIDA, CERCOZOA) DEVOURING PLANKTONIC FRESHWATER ALGAE

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Vampyrellid amoebae (Vampyrellida, Cercozoa) are well known for their fascinating ability to perforate algal cell walls and to feed on protoplast material, as seen in several species of the genus *Vampyrella*. Besides these 'protoplast feeders' there are vampyrellids in freshwater and soil ecosystems that engulf whole prey organisms such as desmids, diatoms or even micrometazoa. Some of these voracious forms have been placed in the Family Leptophryidae, which is well separated from the protoplast-feeding Vampyrellidae by molecular phylogenetic analyses. The leptophryid representatives studied so far (*Leptophrys*, *Theratromyxa*, *Platyreta*) display an expanded, surface-attached morphotype and were thought to have a benthic or terrestrial life style. Two novel vampyrellid amoebae feeding on motile, planktonic algae (genera *Euglena* and *Eudorina*) have been isolated from freshwater ponds. According to phylogenetic analyses of the SSU rDNA gene the new isolates are members of the Leptophryidae, which agrees well with their voracious feeding behaviour and the morphology of digestive cysts. A microcontroller-based setup for time-lapse photography and a special preparation technique were used to investigate food capture events and feeding processes. A feeding experiment involving zygmatophycean, volvoclean and euglenophycean algae revealed the food range specificity of the new isolates and enabled an autecological comparison with *Leptophrys vorax*.



HOW TO DEAL WITH CRYPTIC DIVERSITY IN PROTISTOLOGY – A CASE EXAMPLE IN GENUS *PARAMECIUM*

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Cryptic species, in general, show only minute or no visible morphological differences to existing and valid ‘morphospecies’, but are often genetically quite different and consequently represent distinct biological entities. Therefore, species that are morphologically and superficially seen cosmopolitan species with apparently high abundances could in reality correspond to multiple species with smaller geographic ranges, different ecological demands and much smaller population sizes. Due to the advent of comparatively inexpensive DNA sequencing techniques, studies on cryptic species diversity have dramatically increased. Such studies prove the importance of cryptic diversity and its implications for biodiversity estimations and management. Surprisingly, no common convention on how to describe, delimit and formally name new cryptic species exists to date. While morphological and/or anatomical characteristics are usually required as the primary source for species descriptions, DNA-only taxonomy is not generally accepted. Integrative approaches to species delineation incorporating several adequate characteristics including DNA sequence data are therefore representing the best and most accepted method at present. This, however, does not require morphology as the sole element, but rather involves the detection of the most reliable and efficient character set for proper species delineation. Nevertheless, as long as no other obvious character exists besides DNA sequence information to distinguish the candidate species from a valid morphospecies, cryptic speciation will continue to be an important topic in taxonomic and biodiversity studies. In our opinion, this requires a common practice on how to formally name these species and to indicate their cryptic status in order to distinguish them from valid biological species. This cryptic status should, however, only be provisional, since future studies may be able to elucidate their taxonomic status using new techniques or additional characters. We therefore propose and would like to canvass the term “Eucandidatus” used to be as a systematic component preceding the formal taxonomic name. It is intended to make a distinction between valid biological species and the provisional cryptic species status of newly described eukaryote candidate species. I will present data of combined traditional morphological approaches and thorough molecular analyses on the genus *Paramecium* to indicate cryptic speciation and potentially hidden species. Based on these data we suggest not only a higher biodiversity in *Paramecium* than previously thought, but also describe a new morphospecies, *Paramecium buetschlii* sp. nov., found in a slowly running freshwater stream in Norway.



ALITUDINAL DISTRIBUTION OF ZOOPLANKTON AND THE OCCURRENCE OF *BATRACHOCHYTRIUM DENDROBATITIS*

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In the dynamic properties of freshwater communities, microorganisms like viruses, bacteria, micro-algae, fungi and protozoa play a key role, eminently in nutrient cycling and energy flows. The members of the microcosm are involved and/or concerned directly or indirectly in all important properties of an ecosystem. The idea that various species can prevent or control the prevalence of a disease through various mechanisms recently translated in the controversial theory that diversity and/or composition of a species community can protect against infectious emerging diseases. Recent work of my team delivered one good example of biotic resistance to a pathogen. Our study contributed to the understanding of the colonization success of one of the most devastating pathogens known today, the amphibian fungal pathogen *Batrachochytrium dendrobatidis* (hereafter Bd), the causal agent of the emerging infectious disease chytridiomycosis (Schmeller et al. 2014). Several environmental factors have been identified that covary with the prevalence of Bd infection and chytridiomycosis (Bosch et al. 2007; Walker et al. 2010). Most prominently, lower temperature regimes at higher altitudes are associated with higher Bd infection probability in the Pyrenees, but the occurrence pattern of Bd is not homogeneous at local and regional scales. It is particularly intriguing that some sites, which apparently are appropriate and in vicinity to long-term infected sites, are not colonized by Bd. Until recently, it was thought that temperature directly acts to modify Bd prevalence by influencing host immunity and pathogen growth rates. However, both prevalence of infection and mortality are more common in the Pyrenees when environmental temperatures are very low, and when laboratory estimates of Bd growth rates and zoospore production indicate that infection should be rare. Therefore, for understanding the differences in the prevalence of Bd in amphibian populations, assessing the environmental limitations of Bd and the comprehension of its dynamics is indispensable. Our recent study sheds new light on this unexplained pattern. Temperature, and more generally abiotic environmental conditions, is likely to modify the growth and activity of microorganisms across infected sites, thereby influencing Bd prevalence indirectly. Abiotic environmental factors may also explain the composition, density, and dynamics of the planktonic communities across seasons. Here, we will present the relationship between altitude, temperature determining the interaction between zooplankton and the fungal pathogen Bd.



CHOANOFAGELLATE SYSTEMATICS AND TAXONOMY

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Choanoflagellates, the sister group of metazoans, play an essential role in the microbial food web. Until today, less than 1/4 of the described species has been investigated regarding their molecular identity. Adding data of marker genes to the morphospecies is an important step to interpret data, for example from next generation sequencing.

SPATIAL COMPETITION BETWEEN MICROALGAE AND STALKED PROTOZOA IN BIOFILMS DEVELOPED UNDER DIFFERENT LIGHT REGIMES

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River biofilms are complex assemblages of various microbial groups including heterotrophic bacteria, photoautotrophic primary producers and protozoan grazers. Biofilms grown under light conditions usually show very high abundances of microalgae and bacteria, which in turn can support mainly vagile protozoa such as heterotrophic flagellates, amoeba and ciliates with food. In dark-grown biofilms, however, microalgae are rare and heterotrophic bacteria and their protozoan grazers are dominant. In addition, dark-grown biofilms can show remarkably high abundances of stalked protozoa such as choanoflagellates and peritrich ciliates, which are somewhat less abundant in light-grown biofilms. We hypothesised that such pattern, i.e. low abundances of stalked protozoa in light-grown biofilms vs. high abundances in dark-grown biofilms, results from spatial competition between microalgae and stalked protozoa rather than from differences in the availability of food. This hypothesis was tested by culturing river biofilms in flow channels continually fed with water from the Elbe River (Magdeburg, Germany). One set of flow channels was permanently kept dark. A second set of flow channels was illuminated for 14h per day. Potential effects of different planktonic particle loads in light and dark treatments were excluded by using one water reservoir for feeding one flume of each light treatment. We found strong significant differences between light- and dark-grown biofilms with regards to both structural and functional parameters. Regarding our hypothesis we found benthic microalgae to affect both the settlement and growth of sessile protozoa.



CALCIUM SIGNALING IN PROTOZOA AND BEYOND

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Mechanisms for extrusion of ever permeating Ca^{2+} - primarily a cell toxin - are prokaryotic. Then, endo-/phagocytotic vesicles have imported Ca^{2+} from the outside medium, thus making it available for intracellular signaling. Therefore, Ca^{2+} -release channels (CRCs) have evolved, making Ca^{2+} available locally, thus not only avoiding toxic effects and reducing energy consumption for Ca^{2+} downregulation/extrusion, but also to account for locally restricted $[\text{Ca}^{2+}]$ increase at activation sites. To meet these requirements, organellar CRCs were formed, together with pumps and $\text{Na}^+, \text{H}^+/\text{Ca}^{2+}$ antiporters. How is the evolutionary context currently seen? Many components relevant for Ca^{2+} handling and signaling are now predicted from choanoflagellate (Monokonta) genomic database mining, but this still requires experimental verification. Both approaches have already been combined in work with Paramecium (Bikonta). We ascertained, for instance, true inositol 1,4,5-trisphosphate receptors and ryanodine receptor-like CRCs in Paramecium, as well as intermediates thereof. Several data argue for a common ancestor of these channels. Beyond this, by a comparison of the two main lineages, either by informatics/database mining (Monokonta) or a combination thereof with experimental work (Bikonta) allows us now to recognize that some molecules are maintained, some are newly “invented”, some are neo-functionalized and some deleted during evolution of the two lineages.

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RESPONSE OF SOIL MICRO-EUKARYOTES TO CADAVER DECOMPOSITION AS ASSESSED BY HIGH THROUGHPUT SEQUENCING

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Cadavers represent a natural perturbation that impacts ecosystems by releasing high amounts of nutrients over a short time period. This perturbation influences soil biodiversity but little is known about the impact on the overall micro-eukaryotic diversity. Therefore we hypothesize that (1) cadavers change biodiversity and community structure and that (2) both “cadaver lovers” and “cadaver haters” can be identified, responding respectively positively or negatively to cadavers. In forensic investigations, postmortem interval (PMI) estimations based on classical methods are only reliable for the first weeks after death. New tools are therefore required, and we suggest that the analysis of eukaryotic soil diversity could reveal new PMI indicators. To test these hypotheses, we conducted a field experiment in a mixed beech and oak forest with three treatments: control, fake cadaver (plastic bags filled with soil and covered with a cotton cloth to simulate microclimatic effect of the cadaver without its fluids) and pig cadavers. The treatments were replicated three times in three blocks. The soil was sampled at defined intervals beneath the cadavers and in the other two treatments. To analyze the response of micro-eukaryotic communities we used high throughput sequencing (Illumina) of V9 region of the rDNA SSU gene. We selected indicator OTUs using the IndVal approach. The treatment significantly affected the micro-eukaryotic communities, confirming their sensitivity and potential as indicators of PMI: We identified 191 indicator OTUs (significant IndVal, $p < 0.05$), 111 of which responded positively to the treatment and 80 responded negatively to the treatment. Our analysis showed that different groups of organisms are more sensitive to the treatment at different time points and therefore an optimal PMI tool may be developed based on a combination of several taxonomic/functional groups of micro-eukaryotes.



THE ROLE OF HYDRODYNAMICS IN SHAPING THE COMPOSITION OF BIOFILMS IN TWO STREAMS WITH CONTRASTING TROPHY

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Biofilms that are surface-associated microbial communities (usually dominated by bacteria, cyanobacteria, algae and protozoans) embedded in a matrix of exopolymeric substances constitute an integral part of aquatic ecosystems and are controlled by many factors such as light, grazing, resource availability, water chemistry and flow conditions. Natural mountainous streams are characterized by a high spatio-temporal variability of the flow field provoking a heterogeneous stream bed. As a consequence, hydrodynamics become a dominant factor shaping biofilm attributes through drag forces and control the supply of nutrient resources through mass transfer processes. Previous studies have been restricted to flume experiments where the highly complex flow field of natural streams cannot be reconstructed to the full extent. In a novel approach we are aiming at linking detailed investigations on stream bed heterogeneity and associated development of flow fields to biofilm attributes, i.e. biofilm community structure, spatial morphology, biomass and quality. We conducted measurements in two mountainous streams (Harz region, Germany) comparable in stream bed morphology but distinctly differing in water chemistry. Compared to the Kalte Bode, the water of the Selke has higher concentration of N, P and Chl *a*. The water of the Selke has a mean N : P ratio of 11 while that of the Kalte Bode has a mean value of 30 indicating P limitation. As observed by confocal laser scanning microscopy, the spatial morphology of the studied stream biofilms range from thin layered films of coccoid cyanobacteria and bacteria to heterogeneous films composed of diatoms, coccoid and filamentous bacteria, cyanobacteria, and algae. Biofilms in both streams are highly variable regarding biofilm biomass, Chl *a*, bacterial and protozoan abundances. First results indicate higher Chl *a* concentrations with increasing shear stresses at higher nutrient concentrations in stream water. However, shear stresses seemed not to be the main factor shaping protozoan communities and pigment composition in stream biofilms.



MICROPREDATORS DICTATE THE DYNAMICS OF A FUNGAL DISEASE

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Research on emerging infectious wildlife diseases has placed particular emphasis on host-derived barriers to infection and disease. This focus neglects important extrinsic determinants of the host/pathogen dynamic, where all barriers to infection should be considered when ascertaining the determinants of infectivity and pathogenicity of wildlife pathogens [1–3]. Those pathogens with free-living stages, such as fungi causing catastrophic wildlife declines on a global scale [4], must confront lengthy exposure to environmental barriers before contact with an uninfected host [5–8]. Hostile environmental conditions therefore have the ability to decrease the density of infectious particles, reducing the force of infection and ameliorating the impact as well as the probability of establishing an infection [9]. Here we show that, in nature, the risk of infection and infectious burden of amphibians infected by the chytrid fungus *Batrachochytrium dendrobatidis* (Bd) have a significant, site-specific component, and that these correlate with the microfauna present at a site. Experimental infections show that aquatic microfauna can rapidly lower the abundance and density of infectious stages by consuming Bd zoospores, resulting in a significantly reduced probability of infection in anuran tadpoles. Our findings offer new perspectives for explaining the divergent impacts of Bd infection in amphibian assemblages and contribute to our understanding of ecosystem resilience to colonization by novel pathogens.



DEPOSIT OF PROTOZOA AT DSMZ – CONSIDERATIONS CONCERNING ACCESSION AND DISTRIBUTION

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To expand the DSMZ activities and to complete its spectrum of biological material the DSMZ decided to accept deposits of protozoa, cyanobacteria and certain algae. It is also planned to build up public collections of these organisms for distribution. Responsible within the DSMZ will be the plant cell culture department, due to its experience in the organization of a living collection and the cryopreservation of biological material. Different legal modes of deposit will be available at DSMZ: Deposits in the public collection, safe deposits and patent deposits. Apart from private property rights other considerations have to be taken into account. These are:

- Considerations concerning public law (CBD)
- Considerations according to biosafety and biosecurity regulations (safety level)
- Considerations due to practical aspects like economy, costs and complexity of cultivation conditions
- Considerations due to cost and complexity of characterization and identification procedures

Deposits require safe forms of maintenance and propagation of biological material. Although for protozoa the maintenance of living material may be at least temporarily unavoidable, the maintenance of material by long term preservation should be achieved. However a lack of routine methods for long term preservation requires efforts for the development of new cryopreservation strategies. Other specific problems have to be solved for the deposit of protozoa: Often cultivation conditions are more complex than for other organisms. Nutritional requirements of protozoa necessitate the cultivation and preservation of feeding organisms. The frequent lack of axenicity in protozoan cultures entails more complex characterization strategies. In addition feeding organisms have to be considered in the obligatory risk assessment.



ROLE OF PROTOZOANS IN PLANKTON - BIOFILM SYSTEMS: DYNAMICS IN MORPHOLOGY OF BACTERIA AND THEIR PROCESSING WITHIN THE FOOD WEB

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Predator-prey interactions are of major significance in ecology, because they control population dynamics and ecosystem functions. It is well known that prey organisms develop defense mechanisms to avoid predation and reduce mortality. Several examples show that bacteria increasingly form biofilms to defend themselves from planktonic protozoan grazing. Therefore in simple food chains (with one predator and one prey) biofilms are “dead end roads” in that sense that bacterial biomass cannot be processed within the food web. In more complex systems biofilms might be consumed by other predators and thus prey biomass and nutrients become available for the predator guild. This requires a new dynamic view on biofilm-plankton systems. In the context of the DFG Priority program “DynaTrait” we investigate the consequences of variable feedings types in the predator guild on predator-prey dynamics as well as on major ecosystem functions. As model organisms we are using the bacterial prey species *Pseudomonas putida*, which can form contrasting phenotypes, i.e. planktonic cells and biofilms. As predators, we are using a specialized plankton predator species (*Paramecium tetraurelia*) and a specialized biofilm predator species (*Acanthamoeba castellanii*). In our chemostat system, protozoan predators affect the single prey species that can react with a fast phenotypic plasticity in order to defend itself against either one or the other type of predator but not both simultaneously. We postulate that variability of feeding types in the predator guild together with the phenotypic plasticity of prey organisms will lead to ongoing cyclic changes in predator and prey biomasses. In comparison to the one predator one prey food chain, we assume a more effective carbon flux and a higher biomass allocation towards the predator guild. Here we will combine innovative chemostat experiments and mathematical modeling to test these hypotheses. First modeling results support our hypothesis and sensitivity analysis shows that the level of migration between biofilm and plankton morphotypes is essential for the dynamics of the system. We will demonstrate that defended morphotypes within biofilms are no “dead end roads” for biomass and nutrients in food webs. Ongoing cyclic population dynamics cause increased biomass and carbon fluxes towards higher trophic levels.



INGESTION OF BOVINE-SERUM-ALBUMIN-METHACRYLATE MICROPARTICLES IN THE CILIATE *TETRAHYMENA PYRIFORMIS*

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Recognition and, as a consequence, ingestion of food particles, whether natural food as bacteria or synthetic particles, are important prerequisites for all further processes inside the cell, like digestion or even internalisation and escaping digestion. We developed an experimental setup that allows us to study ingestion and digestion processes in *Tetrahymena pyriformis*. Until now, little is known about the recognition mechanisms of food in *T. pyriformis* and also about the structure of the food vacuole membrane. On the other hand, it was shown that *T. pyriformis*, which is dependent on phagocytosis of food particles (Rasmussen and Kludt, 1970), also takes up synthetic microparticles, e. g. latex particles (Ricketts, 1971a). We used bovine-serum-albumin-methacrylate microparticles (BSA particles), approximately 5.5 μm in diameter, as food particles and performed microscopical analysis after feeding. The BSA is embedded in methacrylate for stabilisation of the particles. The BSA particles were stained with different dyes to follow the uptake and digestion within *T. pyriformis*. Staining with neutral red, a pH-indicator, showed that the particles were not only ingested but also digested, whereas latex particles are not digestible and do not induce digestion after uptake (Ricketts 1971a). Growth experiments showed that *T. pyriformis* is able to survive and even divide with BSA particles as sole food source. Ingestion rates were similar to those found in literature (Chapman-Andresen and Nilsson, 1968; Ricketts, 1971a and b; Hoffmann et al., 1974; Rasmussen, 1976; Nilsson, 1977). Co-feeding of BSA particles labelled with fluorescein isothiocyanate (FITC), a fluorescent dye, and a transformant strain of *Escherichia coli* XL1-blue (Bullock et al. 1987), which shows red fluorescence, revealed that there is no preference for either the particles or the bacteria. In conclusion, BSA particles were digested, served as food source and they seemed to behave like normal food. BSA particles reveal advantages. They can be chemically modified by covalent coupling with other substances. We used amino acids, proteins or enzymes, thus modifying the surface properties of the particles. Depending on the coupled substance, ingestion was decreased (e.g. single amino acids) or increased (e. g. additional BSA). This underlines the thesis of Ricketts (1972) that ingestion of food particles is mediated by receptors. Furthermore, it could be revealed by staining with Nile red, a membrane-specific fluorescent dye, that the provided enzymes (pepsin, trypsin and beta-glucuronidase), bound to the BSA particles, had no obvious influence on membrane composition and did not destroy food vacuoles. Another opportunity to reveal membrane traits, in this case the glycosylation pattern of lipids and proteins of the food vacuole



membrane, was followed by staining with the lectins wheat germ agglutinin (WGA) and concanavalin A (conA), which were both coupled to a fluorescent dye. Glycosylated membrane proteins or lipids are often involved in recognition. Since Ricketts (1971a) has shown that digestion is not automatically induced by ingestion, those glycosylated proteins or lipids could possibly play a role in perception of digestible food particles. In vivo staining of *T. pyriformis* with both lectins revealed that food vacuole membranes contain many glycosylated structures. Both lectins stain different glycosides, but both label mainly N-glycosylated sugars. The experimental setup and the behaviour and simple modification of the BSA particles help to reveal traits of the food vacuole membrane which are important in either digestion or for evading digestion of ingested bacteria. It has recently been shown that not only pathogens, but also a non-pathogenic strain of *E. coli* is able to escape digestion and, instead, internalizes in the cytoplasm of *T. pyriformis* (Siegmond et al., 2013). Offering derivatives of BSA particles with defined enzymatic and surface properties together with adequate staining techniques helps to reveal the physiological prerequisites provided by the host.

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MONITORING ACANTHAMOEBA INFECTIONS IN AUSTRIA

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Acanthamoebae are the causative agents of a painful inflammation of the cornea, the so-called *Acanthamoeba keratitis* (AK), on one hand and on the other hand of several disseminating infections potentially resulting in granulomatous amoebic encephalitis (GAE) in the immunocompromised host. The first case of AK was reported in the early seventies and in the mid-eighties the association between AK and contact lens wear was discovered. Today, acanthamoebae, besides pseudomonads and staphylococci, are regarded as among the most important causative agents of keratitis in contact lens wearers, particularly also because of the frequently severe progression of the disease and the lack of specific treatment. At our institution, which is the Austrian reference institute, *Acanthamoeba* diagnostics was established in 1993. Since then 1067 clinical samples have been screened for *Acanthamoeba* spp. by culture and/or PCR and the detected amoebae have been genotyped. For culture, the samples (corneal scrapings, biopsies, contact lenses, solutions) are inoculated centrally onto a non-nutrient 1.5% agar plate covered with 100 µl of a 24 h old culture of *Escherichia coli* in bacterial broth. Plates are sealed with parafilm, incubated at 30°C for 7 days and examined daily for amoebal growth by inverted phase contrast microscopy. For PCR, an approximately 500 bp fragment of the 18S rRNA gene is amplified, that allows for genotyping by subsequent DNA sequencing of the amplicon. Genotypes are assessed with the model assumption of a <5% sequence dissimilarity within one genotype. Altogether, 168 cases of AK and 3 cases of GAE were diagnosed. The AK patients were 8-82 years old (mean 35.6), 56% of them were female and almost 90% were contact lens wearers. The predominant genotype in the AK cases was T4, other genotypes found were T3, T5, T6, T10 and T11. The GAE patients were 2-25 years old (mean 14.7), all HIV-negative, but two were severely immunosuppressed at the time of diagnosis. The three GAE cases involved genotypes T2, T4 and T5. All amoebae isolated by culture were thermotolerant (34°C).



CULTURE GROWTH OF TESTATE AMOEBAE UNDER DIFFERENT SILICON CONCENTRATIONS

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Testate amoebae with self-secreted siliceous shell platelets (“idiosomes”) play, with respect to annual biosilicification, an important role in terrestrial silicon cycles. In this context, as a prerequisite for further studies on protozoic silicon fluxes in terrestrial ecosystems, silicon-dependent culture growth dynamics of idiosomic testate amoebae are of interest. The objective of this study was to test if (i) amoebal shell size and (ii) culture growth dynamics are affected by available silicon, followed by a (iii) preliminary quantitative estimation of amoebal silicon consumption. Clonal cultures of idiosomic testate amoebae were analyzed under different silicon concentrations: deficiency (50 $\mu\text{mol L}^{-1}$), moderate/site-specific (150 $\mu\text{mol L}^{-1}$) and excessive silicon supply (500 $\mu\text{mol L}^{-1}$). Food (the yeast *Saccharomyces cerevisiae*) was provided in surplus. (i) Shell size of four different clones of idiosomic testate amoebae (*Trinema galeata*, *Euglypha rotunda*, *E. rotunda* cf., *E. filifera*, all clones provided by Ralf Meisterfeld, Aachen) was not affected by silicon concentration. (ii) Culture growth of idiosomic *Euglypha rotunda* was dependent on silicon concentration. The more silicon available in the culture medium, the earlier the entry into exponential growth phase. Culture growth of idiosomic *Euglypha rotunda* was dependent on origin of inoculum. Amoebae previously cultured under a moderate silicon concentration revealed highest sustainability in consecutive cultures. Amoebae derived from cultures with high silicon concentrations showed rapid culture growth which finished early in consecutive cultures. (iii) Silicon (diluted in the culture medium) was absorbed by amoebae and fixed in the amoeba shells according to the respective growth dynamics. It appeared that idiosomic testate amoebae were able to absorb almost all available silicon. However, due to complex culturing conditions, additional experiments are necessary to enable quantitative studies on silicon consumption and –fluxes by idiosomic testate amoebae.



MICROBIAL FOOD WEB INTERACTIONS WITHIN BIOFILMS

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Environmental biofilms are considered hot spots of microbial life. They contribute significantly to the flux of matter in aquatic ecosystems, particularly in running waters, shallow lakes and groundwater systems. Biofilms potentially harbor all relevant microbial groups including bacteria, algae, protozoans, fungi and small metazoans and their densities usually exceed those in planktonic systems by some orders of magnitude. However, ecological studies on biofilms mostly focus on bacteria or algae and biofilm functions mediated by them. Studies on the occurrence and role of the highly abundant micro-grazers (protozoans and small metazoans) are rare, particularly in comparison to the numerous studies on grazers and food web interactions within planktonic systems. One reason for this underrepresentation might be the common but erroneous view that bacterial biofilms are generally resistant against microbial grazing. It is therefore necessary to develop a conceptual framework on microbial food web interactions within biofilms. Here we will review recent findings on the role of micro-grazers in altering the structure and functions of biofilms. We will develop a conceptual food web model with respect to the origin of resources used (autochthonous vs. allochthonous), food quality for the grazers and food web complexity. We will illustrate that grazers are an essential component of environmental biofilms, altering both structural and functional attributes of the biofilm.

MOLECULAR EVIDENCE FOR PLASTIDS IN *SPUMELLA* (HETEROTROPHIC CHRYSOPHYCEAE)

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Primary production through oxygenic photosynthesis is the primary energetic basis of any food chain. Besides the primarily photosynthetic lineages (Archaeplastida) many eukaryotic lineages acquired – through secondary and tertiary endocytobiosis – plastids and with that the potential for photosynthesis, specifically the euglenids, the chlorarachniophytes and the ‘chromist’ algae including heterokonts, cryptophytes and haptophytes. The loss or at least the reduction of the plastid occurred even more often. However, the plastid is not only the key organelle for photosynthesis but also involved in many other metabolic pathways such as fatty acid and tetrapyrrol metabolism. One key for an understanding of the radiation of eukaryotes and the functional diversity of protists is therefore the regulation and localisation of plastid-associated metabolic pathways in the course of plastid reduction. For addressing this issue specifically the chrysophytes are an outstanding model group as within the chrysophytes the reduction of plastids occurred at least three to five times independently and many heterotrophic, mixotrophic and phototrophic lineages are known. We examined the transcriptomes of different strains of the polyphyletic genus *Spumella*, a heterotrophic organism, for genes involved in the biosynthesis of tetrapyrroles and carotenoids. As expected, transcripts of genes involved exclusively in the formation of chlorophylls were absent, whereas we found transcripts of nearly all the genes necessary for the biosynthesis of hemes and siroheme. Surprisingly, we also detected transcripts of genes involved in the formation of carotenoids up to β -carotene. The results indicate the presence of cryptic plastids in heterotrophic *Spumella*.



ABSTRACTS OF POSTERS

sorted alphabetically by first author

REDESCRIPTION OF THE TINTINNID *FAVELLA PANAMENSIS* KOFOID AND CAMPBELL, 1929 (ALVEOLATA, CILIOPHORA, SPIROTRICHA) BASED ON LIVE OBSERVATION, PROTARGOL IMPREGNATION, AND SCANNING ELECTRON MICROSCOPY

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Favella panamensis was collected at the east coast of the USA. Live observations, protargol impregnation, and scanning electron microscopic studies were performed. The campanulate lorica is 160–260 μm (= 222 μm ; n = 80) long and hyaline and has usually one apical ring. Its opening is 63–103 μm across (= 81 μm ; n = 104) and has a smooth rim. The posterior process is 20–90 μm long (= 57 μm ; n = 80), hollow, and dextrally twisted. The 2–4 μm thick lorica wall has a smooth surface and is monolaminar with alveoli (\sim 1 μm across), except for the posterior process with several layers. The replacement lorica is of the Coxiella type with about 13–15 spirals and a broadly rounded posterior end. The cell is obconical in vivo, while broadly obconical after preservation. The cell proper measures 60–150 \times 60–95 μm in vivo and is attached to the lorica inside the hollow posterior process by a highly contractile peduncle 80–100 μm long. The morphostatic specimens have two macronucleus nodules (17–42 \times 6–14 μm in size) and up to three micronuclei 1–3 μm across. The cytophyge is near the base of the peduncle and the dorsal kineties. The somatic ciliary pattern comprises a right, left, and lateral ciliary field as well as a ventral kinety and a variable number of dorsal kineties (74 % of specimens have two rows, some have three rows, and very few have four). The right ciliary field comprises 39–52 kineties (= 48; n = 11), the left field 26–39 kineties (= 33; n = 5), and the lateral field 6–8 kineties (= 7; n = 18). A unique ciliary tuft 11–14 μm long originates from the anterior third of the lateral field. The adoral zone of membranelles is 60–80 μm across in vivo and composed of one buccal membranelle and 16 or 17 collar membranelles, of which four are elongated into the buccal cavity. The collar membranelles are 30–45 μm long. The endoral membrane, tentaculoids, striae, and accessory combs were not recognizable. It feeds on thecate dinoflagellates (\sim 10–70 \times 7–65 μm in size) and the tintinnid *Tintinnopsis cylindrica* (120 \times 30 μm in size). The specimens match the original description and authoritative redescription in the lorica length (136–232 μm), opening diameter (64–86 μm), process length (12–28 μm), and wall texture.

The study was financially supported by the FWF (project P 20461-B17).



PUTATIVE TINTINNID FOSSILS (ALVEOLATA, SPIROTRICHA, OLIGOTRICHEA) ARE ACTUALLY ACRITARCHS

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Paleontological studies have greatly contributed to our understanding of the evolution of life. Since ciliates (Alveolata, Ciliophora) rarely possess hard parts such as shells that easily fossilise, the scarcity of fossils hampers evolutionary reconstruction in this large and ecologically important clade of microbial eukaryotes. Tintinnids, a group of loricate (house-forming) planktonic ciliates, are the only group that has a significant fossil record from modern sediments to those dating back to the Mesoproterozoic era. The forms older than Jurassic, however, possess characters that cannot be found in extant ciliates (Lipps et al. 2013). This also holds for a recently described fossil *Nassacysta reticulata* (Stemans et al. 2014), which deviates distinctly in several respects from tintinnids and ciliates in general. Until future studies have conclusively demonstrated that these organisms are most likely extant orchid seeds or fossilised plant remnants, they are best considered incertae sedis and are thus regarded as acritarchs (organic-walled microfossils of unknown biological affinity) or remains of some ‘other eukaryotes’. To prevent the publication of uncertain taxa as ciliates that distorts the interpretation of the early ciliate evolution, experts on extant ciliates should always be included in the review process.

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PROTIST COMMUNITY COMPOSITION ALONG AN ELEVATIONAL GRADIENT IN COSTA RICAN BROMELIADS

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Climate warming, habitat fragmentation, environmental pollution and other anthropogenic stressors have been observed to affect ecosystems and their functions. Therefore, the ability to predict these changes will be of great importance for future conservation attempts. Naturally changing gradients of environmental variables are a popular approach to study this topic in the field. One of the best known opportunities is provided by elevational gradients. They are prevalently used in studies to assess changes in community compositions induced by, for example, changes in temperature. We conducted a field survey along three different mountainsides in Costa Rica to assess the community composition of protists (inter alia flagellates, ciliates, amoeba) living in the water-filled tanks of epiphytic bromeliads. Our aim was to investigate if a change in community composition occurred along the elevational gradient and to identify the relevant environmental variables that can cause such a shift in community composition. Within the scope of our field survey, water samples of bromeliads belonging to the genera *Werauhia* and *Guzmania* were taken during the wet season 2014 (May-August) along three different transects on inactive volcanoes in the Área de Conservación Guanacaste, Costa Rica. Altitudinal gradients ranged between 683 - 1906 m above sea level. Parameters measured included water temperature, dissolved oxygen concentration, pH, dry weight of detritus and light availability. Protists were preserved with Lugol solution and identified and counted under the microscope after the end of the field period. We found that temperature is the only environmental variables that significantly changes along the elevational gradient. However, preliminary results show that there is no change in protist community composition along the three studied transects. This indicates that the composition of protist communities is probably regulated by other factors such as predation pressure through insect larvae. Within the scope of our field work another study on aquatic insect community composition in bromeliads along an elevational gradient was carried out. Results from this other study will be used to gain further insight into the factors driving protist community structure.



SYNTHESIS OF BOVINE-SERUM-ALBUMIN-METHACRYLATE PARTICLES WITH DEFINED SURFACE PROPERTIES FOR FEEDING TETRAHYMENA PYRIFORMIS

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Bovine-serum-albumin-methacrylate microparticles (BSA particles) can be used as food particles for *Tetrahymena pyriformis*, since these ciliates ingest and also digest them. The BSA is crosslinked by treatment with glutardialdehyde and embedded in methacrylate polymer for mechanical stabilisation. By treatment with carbodiimide, we linked additional compounds covalently to the surface of these particles. By this simple technique we are able to provide food particles with defined surface properties. Especially, we synthesized particles with different hydrophobicity, with basic or acidic groups and, by linking with additional proteins, with enzymatic properties. In a second step, these modified microparticles can be labelled with fluorescein-isothiocyanate or, alternatively, with dyes that allow monitoring the pH directly around the particles during the digestion process. Food bacteria are known sometimes to avoid digestion by escaping from food vacuoles, followed by internalisation in the cytoplasm. Recently, we have shown this for a genetically modified strain of *Escherichia coli*, rendering *Tetrahymena pyriformis* resistant against the aminoglycoside antibiotic paromomycin (Siegmund et al., 2013). By feeding modified food particles and following the fate of these particles in the cell, we intend to gain insight in the nature of such surface properties that decide on interaction with the vacuolar membrane and consequently on the possibility of internalisation. Already the ingestion process itself is remarkably influenced by surface properties of BSA particles. Coupling with additional proteins directly to the surface improved the ingestion process in comparison to those particles that carried only single amino acids. We did not observe differences in ingestion between pure BSA particles and those that were decorated with additional proteolytic enzymes.

References: Siegmund L., Burmester A., Fischer M.S., Wöstemeyer J., 2013. A model for endosymbiosis: Interaction between *Tetrahymena pyriformis* and *Escherichia coli*. Eur. J. Protistol. 49, 552-563



PROTISTS AS MODEL ORGANISMS IN BIOLOGY CLASS – A THEORY-BASED DEVELOPMENT OF EDUCATIONAL CONCEPTS AND EXPERIMENTS WITH UNICELLULAR ORGANISMS FOR HIGHSCHOOLS

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Protists are excellent model organisms and can be used to support the explanation of general biological concepts to pupils. They can help to gain an understanding about complex macroscopic live forms and hence about human beings and their environment. However, in present-day biology classes the application of protists is mostly limited to the demonstration of microscopy techniques. Consequently, education material for the use of protists as model organisms in schools is rare. The aim of this master thesis is to develop instructions for specific experiments which can be used by teachers in their biology classes. In addition, teaching material that is attractive in form and content as well as age-appropriate for the pupils is designed. The motivation for the teachers to use protists and in a further step the selection of appropriate organisms for fascinating experiments depend mostly on an inexpensive way to obtain and cultivate the protists, especially given the restricted financial and infrastructural possibilities of most schools. The organisms should be able to grow under uncontrolled conditions at room temperature, they should require little maintenance and they should be of an adequate size for simple observation under the microscope. The experiments chosen comply with the curriculum requirements and support the knowledge transfer of relevant biological topics. In the context of this study, an experiment was chosen that gives information about the different structures and mechanisms of organisms for locomotion. Another experiment shows the metabolic processes of feeding and digestion. In addition, an experiment that deals with predation-pressure and predator-prey-interactions in food webs, and one experiment that illustrates the impact of different environmental conditions (such as temperature, light and food conditions) on organisms were developed. For the conduction of these experiments, the ciliates *Paramecium caudatum*, *Blepharisma japonicum*, the amoeba *Amoeba proteus*, some crustaeen metazooplankton and unicellular algae were chosen. Selected concepts will be tested with pupils in respect to their quality and practicability and will then be improved, if necessary. Pupils' perceptions about protists will be identified prior to conducting the experiment with them and, for comparison, afterwards. Finally, the learning effect in respect to specific biological topics as a result of the practical examination will be determined. This study should encourage biology teachers to use protists at different levels of education at school in order to demonstrate a wide range of biological questions. Teachers should be supported through the supply of material,



information, tips and suggestions for handling and experimenting with protists. Finally the up-to-date concept of practical and problem-oriented education should help to motivate and inspire pupils for biology and other natural sciences.

NATURAL SELECTION SHAPES THE EVOLUTION OF DNA SPLICING SIGNALS IN *PARAMECIUM*

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DNA splicing plays a central role during the development of several organisms. In ciliated protozoa, it contributes to the formation of a functional somatic macronucleus (MAC), which is regenerated from the germline micronuclear genome (MIC) after events of sexual reproduction. This developmental process involves the removal of micronuclear DNA regions known as internal eliminated sequences or IESs. Previously, we found that hundreds of IESs are inaccurately spliced from the developing MAC of three closely related *Paramecium* species. A fraction of these IESs appear to have integrated into the MAC of each of these species. In addition, our in-silico study identified a set of IES-flanking sites that may have a role in the process of IES recognition/excision (Catania et al., 2013). Here, we examine the role that these putative cis-regulatory sequences have in the process of DNA splicing in the species *P. tetraurelia*. We characterize the association between the quality of the aforementioned putative signals - measured in terms of degree of sequence conservation - and the extent to which IESs are inaccurately excised. Furthermore, we identify patterns of these signals' association with characteristics such as IES size, IES genomic position, and the level of expression of the host gene. Our results provide original insights into the molecular mechanisms of IES excision. They provide further support to the regulatory role of the surveyed set of IES-flanking sites and suggest that natural selection affects the level of accuracy of DNA splicing in *Paramecium*.



EFFECTS OF FUNCTIONAL AND PHYLOGENETIC DIFFERENT CERCOMONAD STRAINS ON BACTERIAL DIVERSITY

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It is well known that heterotrophic protists feed highly selective and exert a strong top-down control on bacterial communities. However, it is unclear whether all protists exhibit similar or largely different feeding preferences. We investigated the grazing-induced changes in a diverse bacterial community from phyllosphere of plants in a feeding experiment with different Cercozoa species. We estimated the genetic relatedness of the protist strains based on the 18S rRNA gene and investigated their grazing impact on the composition and abundance of the bacterial community by using metagenomic profiles obtained by Illumina MiSeq sequencing.

LIGHT CONTROL OF MICROBIAL BIOFILMS IN THE NUTRIENT-RICH ELBE RIVER (MAGDEBURG, GERMANY)

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Microbial biofilms account for a large proportion of matter fluxes in aquatic environmental and also form the basal food source for many aquatic consumers. Their distinct assemblage structures, however, are regulated by a complex network of environmental factors. While in running waters most abiotic factors do not show local differences at a given time, especially the light intensity can show steep gradients between surface and bottom regions. This can lead to significant changes among photoautotrophic biofilm assemblages, which can subsequently alter the structure of microbial consumer assemblages and affect the ecological functioning of the biofilms. Here we addressed the question in how far light-independent microbial consumers (ciliates, amoeba, heterotrophic flagellates) are affected by different light intensities. Biofilms were cultivated in flow channels exposed to two different light intensities (bright light vs. no light) but otherwise standardized experimental conditions using water from the highly eutrophic Elbe River (Magdeburg, Germany). After each 4 and 6 weeks of cultivation, biofilms were analyzed microscopically and selected functional parameters were assessed. We found strong significant effects of light on both the production of microbial biofilms and on the assemblage structure of microbial



consumers, both of which showed significant dependencies of the autotrophy of biofilms cultured at the respective light intensity. Regarding the assemblage structure of microbial consumer assemblages, sessile ciliates and flagellates were dominant in dark treatments but were displaced by dominance-forming diatoms and green algae in light treatments, thereby promoting very high abundances of mobile and algivorous protists.

PROTISTAN COMMUNITY ANALYSIS – A LARGE SCALE MOLECULAR SAMPLING

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To analyze overarching patterns of protistan diversity, huge datasets covering many samples of different habitat types or geographic regions are needed. With the use of standardized protocols, high throughput sequencing technologies (with their huge amounts of deep sequencing data) hold the potential of providing such datasets. So far, however, deep sequencing studies mostly focus on merely one or few sampling sites and are restricted to single habitat types. We here provide a dataset of 295 samples from soil as well as fresh, brackish and marine waters sequenced by a standardized 454 protocol. Our analyses of this dataset cover protistan community structure as well as protistan distribution patterns and reveal distinct habitat specificity. Most interestingly, the distribution of generalist and specialist taxa shows differences in both higher taxonomic groups and habitat types.



PROTOZOON OF THE YEAR 2015: *VAMPYRELLA*

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The genus *Vampyrella* has been chosen to be the 'protozoon of the year 2015' by the German Society for Protozoology. The poster informs about the taxonomic history, diversity, feeding behaviour and phylogenetic position of the genus. Furthermore, participants of the meeting can obtain a batch of printed flyers about *Vampyrella* at the poster.

CELL TYPE VARIETY IN THE LIFE CYCLE OF DIFFERENT CHOANOFLAGELLATE SPECIES

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Choanoflagellates are eukaryotic protists that live in marine and freshwater environments. The cell morphology of choanoflagellates is unique, and elsewhere only found in the choanocytes of sponges. The bacterivore choanoflagellates have an apical flagellum, which is surrounded by a collar of microvilli. Choanoflagellates are the closest known sister taxa to all Metazoa and are able to cell differentiation and colony formation as a primitive form of multicellularity. Among all choanoflagellate clades colony formation occurred several times. Thus, it is supposed, that the last common ancestor of choanoflagellates and metazoans was – similar to choanoflagellates – able to colony formation and cell differentiation. The life cycles of choanoflagellate species are often quite complex. The presented studies deal with the impact of various factors (including the presence of prey bacteria and predatory flagellates) on the formation of life cycle stages in different choanoflagellate species. Furthermore, the colony structure of a species from Mono Lake was investigated by fluorescence microscopy.



DESCRIPTION OF AN ITALIAN POPULATION OF *STYLONYCHIA HARBINENSIS* (CILIOPHORA, OXYTRICHIDAE) BASED ON MORPHOLOGY AND SSU RDNA

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The genus *Stylonychia* Ehrenberg, 1830 is largely confined to freshwater with species of *S. mytilus* complex and *S. pustulata* being the most common ones. Berger (1999) in his detailed revision of the oxytrichids included eleven species within the genus however, only four of them were described in detail (Wirnsberger et al., 1986). The *Stylonychia mytilus* complex includes four species: *S. mytilus*, *S. lemnae*, and the last two species added *Stylonychia ammermanni* (Gupta et al., 2001) from Delhi region of Yamuna River, India, and *Stylonychia harbinensis* (Shi et al., 2004) from Harbin (Heilongjiang Province, China). The latter being previously included in the revision of Berger (1999) although considered as a nomen nudum since the species had been poorly described at that time. As with other genera including cryptic and sibling species, the morphological identification of species within the genus *Stylonychia* is known to be difficult due to the lack of conspicuous and unambiguous differences, and to the fact that most of the descriptions (including *Stylonychia harbinensis*) are only morphological but do not provide any molecular data. In the present study we provide a comprehensive redescription of *Stylonychia harbinensis*, isolated from soil samples of paddy fields collected from the area of Parona (Lombardia, Italy), combining morphology, morphogenesis, and molecular phylogeny.



TRANSMISSION ELECTRON MICROSCOPY SAMPLE PREPARATION PROTOCOLS FOR THE ULTRASTRUCTURAL STUDY OF CYSTS OF FREE-LIVING PROTOZOA

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Detection and localization of intracystic bacteria and examination of the en- and excystment dynamics is a major challenge, as no suitable protocols for ultrastructural analysis of cysts are available yet. While Transmission Electron Microscopy (TEM) is ideally suited for these analyses, conventional TEM protocols tend to result in low cyst yield and images of poor quality, making them not optimal for further cell biological analysis. Cysts of free-living Protozoa (FLP) have received increasing attention as they can act as a vector and shelter for bacteria. This identifies a hitherto little known role of FLP cysts in the ecology and epidemiology of pathogenic bacteria. As such, there is a need for a suitable TEM sample protocol for protozoan cysts. In this study, different protocols for TEM sample preparation of cysts were designed and tested. Two protocols, one based on chemical fixation in coated well plates and one on High Pressure freezing with Automatic Freeze substitution, were selected as most effective for TEM-based ultrastructural studies of cysts. These protocols will allow a better analysis of the cyst structures and a better understanding of bacterial survival mechanisms in cysts. We suggest that the proposed TEM protocols can also be used for weakly adherent cells and fragile cells.



CYSTS OF FREE-LIVING PROTOZOA: A POTENTIAL VECTOR AND SHELTER FOR FOODBORNE PATHOGENIC BACTERIA.

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The production of resistant, dormant cysts forms an integral part of the life cycle of many free-living protozoa, allowing these organisms to survive adverse environmental conditions. There is increasing evidence that some bacteria escape digestion by bacterivorous protozoa and can survive and even multiply inside active protozoan cells. In some cases, intracystic survival has also been demonstrated which is especially relevant as cysts may confer high resistance to unfavorable environments. Given the prevalence of free-living protozoa in food-related environments, it has been hypothesized that these organisms play an important yet currently underinvestigated role in the transmission and epidemiology of foodborne pathogenic bacteria. The present study investigated the survival capacities of foodborne pathogens inside cysts of the model protozoon *Acanthamoeba castellanii*. Invasion assays, encystment monitoring assays and intracystic (stress-)survival assays were performed using the following pathogen strains: *Salmonella enterica* serotypes Typhimurium and Enteritidis, *Listeria monocytogenes* serotypes 1/2a and 4b, enterohaemorrhagic (EHEC) *Escherichia coli* serotypes O:26 and O:157, *Yersinia enterocolitica* bioserotypes 4/O:3 and 2/O:9 and two *Campylobacter jejuni* strains. Results indicate that important foodborne bacteria (i.e. *Salmonella enterica*, *Yersinia enterocolitica*, *Escherichia coli* and *Listeria monocytogenes*) can survive inside cysts of the ubiquitous amoeba *Acanthamoeba castellanii* and resume active growth after excystment, even when they have been exposed to e.g. antibiotic treatment oxidative stress, osmotic stress and highly acidic conditions. Strain- and species-specific differences in survival period were observed, with *Salmonella enterica* surviving up to three weeks inside the amoebal cysts. These differences were not related to variation in trophozoite invasion/uptake efficiency. Transmission electron microscopy revealed that up to 53% of the cysts were infected with pathogenic bacteria, which were located in the cyst cytosol. Apparently intact cells of another common bacterial pathogen, *Campylobacter jejuni*, were observed inside *A. castellanii* cysts, but no cells were observed after excystment. The present study indicates that long-term survival of foodborne pathogens in protozoan cysts is possible. This has an impact on the ecology and epidemiology of pathogenic bacteria, as cysts may act as a vector and shelter against harsh environmental conditions. Moreover, intracystic bacteria may not be detected by the standardized biochemical protocols used to detect foodborne pathogens in food and food-related environments.



SWARM: ROBUST AND FAST CLUSTERING METHOD FOR AMPLICON-BASED STUDIES

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Popular de novo amplicon clustering methods suffer from two fundamental flaws: arbitrary global clustering thresholds, and input-order dependency induced by centroid selection. Swarm was developed to address these issues by first clustering nearly identical amplicons iteratively using a local threshold, and then by using clusters' internal structure and amplicon abundances to refine its results. This fast, scalable, and input-order independent approach reduces the influence of clustering parameters and produces robust operational taxonomic units.

WHAT IS IN A SPECIES? A TRANSCRIPTOMIC APPROACH FOR ANALYZING MOLECULAR VARIATION WITHIN PROTIST SPECIES

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We aim at providing new insights into an old problem - the variability within species and with that the problem of species delimitation in protists. We address this issue exemplarily for the intraspecific and interspecific molecular variation of Chrysophytes. We analyzed functional diversity, with focus on metabolic pathways, based on transcriptomes of three strains affiliated with *Poteroispumella lacustris* that originate from different habitats (New Zealand, China, Austria). The intraspecific molecular variation within this species is compared to the closely related *Poteroiochromonas malhamensis*.



MICROBIAL COMMUNITIES UNDER INVESTIGATION – UNDERSTANDING THE BIASES OF DEEP SEQUENCING DATA

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Deep sequencing is a modern tool providing unknown insights into microbial communities. So far, such sequencing data were often taken for granted as mirroring community compositions for comparative diversity studies. Little is known, however, about the biases modifying sequencing results of microbial communities. To explore such biases, we compared environmental samples of three ecologically differing lakes in two high throughput sequencing runs conducted with the illumina technology. These illumina Mi-Seq runs differed in sequencing length as well as subsequent bioinformatical processing. We hypothesize that the biases we find cause dramatic changes in rare organism counts and even effect the relative abundances of common major groups within a sample. With our results, we intend to improve quantitative molecular community analysis innovatively and to show how deep sequencing techniques have to be adjusted to achieve a fuller picture of true microbial diversity.



MORPHOLOGICAL AND MOLECULAR BASED ANALYSES OF PLANKTONIC CILIATES IN LAKE ZURICH

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Here we present data from a high frequency sampling campaign aimed at characterizing the dynamics of ciliate species in a freshwater lake. We applied both, a quantification of ciliate morphospecies by quantitative Protargol staining (QPS) and a molecular characterization via next generation sequencing of phylogenetic marker genes. Although both, morphology and molecular based approaches showed similar values of total ciliate diversity and assemblage composition (in terms of classes), analyses on the level of genera and species differed markedly. There is an obvious discrepancy between the considerable knowledge on the diversity of freshwater ciliate morphotypes and the scarce information on their molecular phylogeny. Our study emphasizes that: (i) Sequence information is still missing for many well-known and described freshwater ciliate morphotypes. (ii) It seems that some ciliate sequences deposited in databases were attributed to wrong or inadequate species names. (iii) Due to the high copy number of 18S rRNA genes in single ciliate cells, a reliable quantification of ciliate abundance is not yet possible solely by molecular techniques. Thus, QPS is still a powerful tool to analyze the qualitative and quantitative composition of ciliate assemblages. (iv) There is still need to recognize and isolate protistan species for molecular analyses but also for characterization of their autecology. Thus, synergistic teamwork between morphologists, ecologists, and molecular biologists is crucial for studying ciliate assemblages.



SENSE AND NONSENSE OF NETWORK ANALYSES FOR STUDYING CILIATE - PHYTOPLANKTON INTERACTIONS

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In freshwater lakes, ciliates (Ciliophora) are the very first consumers of recurrent spring phytoplankton blooms and their diversity seems to be linked to prey species diversity. Here we present the rapid succession of 21 clearly definable planktonic ciliate morphotypes during spring time in Lake Zurich and relate their abundance to 19 phytoplankton genera, abiotic parameters and to heterotrophic bacteria. For the detection of species-specific significant correlations, we conducted classical statistical analyses and a co-occurrence based network analysis resulting in intuitive graphical presentations. Although networks are commonly used for evaluating environmental sequence data (often 'unattributed' operational taxonomic units (OTUs)), our analysis was based on abundances of protistan morphotypes. Therefore we could partly assess the meaning of co-occurrences, as the autecology of many species was known. On the one hand, the network analysis emphasized the pivotal role of distinct ciliate species as first consumers of primary production and revealed a clear clustering of mixotrophic / omnivorous species. On the other hand, several co-occurrences (positive and negative correlations) were found to be delusive, when the autecology of involved species was regarded. This sounds trivial, but it reflects the major problem of network analyses based solely on unattributed OTUs (which is still the case for many protistan sequences). We emphasize that network analyses are a fascinating tool to visualize microbial food webs and formulate testable hypotheses about the interactions, but there is still need to recognize and isolate protistan species and their potential prey organisms for laboratory experiments.



ULTRASTRUCTURAL CHARACTERIZATION OF A NEW MICROSPORIDIUM (OPISTHOKONTA: CHYTRIDIOPSIDA) FROM THE PIGEON FEATHER MITE *FALCULIFER ROSTRATUS* (ASTIGMATA: PTEROLICHOIDEA)

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Only about 20 species of microsporidia have been described from mites. All except one species produce typical spores with a long polar filament and a polaroplast. We present the first study of an atypical microsporidium infection in a feather mite (*Falculifer rostratus*). The infection is restricted to the colon epithelium where it leads to hypertrophy of the concerned cells. During sporogony multinucleate plasmodial aggregates are formed within a sporont. The sporonts are in direct contact to the host cell cytoplasm. Merogonial stages were not present. Spores are tiny ($3.6 \times 2.6 \mu\text{m}$), broad ovoid in form and monokaryotic. The spore wall of mature spores has a thickness of about 240 nm and consists of a three-layered endospore and a thin, electron-dense exospore. The polar filament is anisofilar and arranged in 3–4 coils. In cross-sections it has a star-like appearance since the electron-dense core forms rounded compartments for lucent material at its surface. In grazing sections this results in a honeycomb-like pattern. A polaroplast is missing. The life cycle features and atypical spore structures clearly classify the species from the feather mite as a member of the order Chytridiopsida. Its affiliation to one of the known genera is discussed.



ENLARGING THE HALOTOLERANT GROUP OF PLACIDIDEA (STRAMENOPILA): NEW HETEROTROPHIC FLAGELLATES FROM CHILEAN INLAND WATERS

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Studies of different hypersaline inland waters from Chile revealed a new morphotype of stramenopiles. The 18S rRNA was sequenced and phylogenetic analysis showed its association to the class of Placididea. Only two species are yet described for this recently erected class. The new species *Allegra atacamiensis* sp. nov. and related environmental sequences from the working group of Alastair Simpson will be described in the near future. Recent studies suggested that the diversity of microbial organisms is widely underestimated and that the majority of species still need to be discovered. Athalassohaline lakes like those of the Atacama seem to contain an interesting endemic protistan fauna which is currently under investigation as part of studies on the evolution of protists in our research group.

GETTING ACCESS TO CILIATES: A SIMPLE DETERMINATION KEY FOR CILIATES IN RIVERS

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The determination of protist species and ciliates in particular is challenging for novices to work with these organisms. The determination keys of Foissner and colleagues offer a well-established instrument to get into the tools for the determination of ciliate species in either benthic or pelagic habitats. Moreover, they comprise abundant information about the taxonomy, morphology and ecology of each single species. However, to get started with these keys is challenging for beginners with limited a priori knowledge. Furthermore, in our group we are focusing on protozoans found in the pelagial of rivers, which comprise pelagic as well as benthic species. Therefore, we here present a simple determination key for protozoans in rivers based on Foissner's keys. This simplified key is referenced to the Foissner keys and refers to both, pelagic and benthic ciliates. The poster will give first impressions of this key and its potential applications.



"CANDIDATUS FOKINIA SOLITARIA" (MIDICHLORICEAE, RICKETTSIALES), A NOVEL BACTERIAL ENDOSYMBIONT INHABITING AN UNKNOWN *PARAMECIUM* SPECIES

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The recently described family Midichloriaceae (Rickettsiales, Alphaproteobacteria) consists of bacterial endosymbionts found in diverse eukaryotic hosts like Protozoa, Placozoa, Cnidaria, Arthropoda and Vertebrata. In the present work, we provide a description of the new bacterial endosymbiont "*Candidatus Fokinia solitaria*" inhabiting a *Paramecium* species (Oligohymenophorea, Ciliophora) isolated from a wastewater treatment plant in Rio de Janeiro (Brazil). In order to characterize the symbiont and its host species, prokaryotic and eukaryotic SSU rRNA genes were sequenced. Based on the bacterial SSU rRNA gene sequence, phylogenetic analyses were performed and three species specific oligonucleotide probes were designed and tested for their binding ability and specificity. Transmission Electron Microscopy was performed for ultrastructural analyses. The tiny rod-shaped endosymbionts (1.2 x 0.25-0.35µm in size) are located in the host cytoplasm, stratified in a narrow layer in the host cortex squeezed between the host trichocysts and mitochondria. Bacterial cells show a typical Gram-negative cell wall. The bacteria belong to a separate branch within the recently described family Midichloriaceae. In contrast to widespread and oligo- or euryxenic clades like "*Candidatus Midichloria*" and "*Candidatus Lariskella*", so far "*Candidatus Fokinia solitaria*" is the only representative in its branch. Like other members of the Midichloriaceae it seems to have a rather restricted than ubiquitous distribution, which indicates a narrow host range, similar to *Lyticum* spp., "*Candidatus Cyrtobacter* spp." or "*Candidatus Defluviella procrastinata*". In most of these lineages, the evolutionary rate of the bacterial SSU rRNA gene looks higher than in the average of Midichloriaceae.



PROTISTAN NANOFAUNA DISTRIBUTION: INVESTIGATING SPECIES RICHNESS IN THE MESOSCALE

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Next-Generation-Sequencing (NGS) delivers thousands of sequence reads in a single sequencing run, which should enable detailed studies of community structures and detection of even rare organisms. Sequence reads can differ in length and quality. Several steps of quality filtering are included in every sequence analysis, to exclude low quality sequences and increase the reliability of estimates. We present the largest dataset available for flagellated protists to date.

ION IMAGING AS A TOOL TO UNVEIL THE INTERNAL CELL ENVIRONMENT OF HALOPHILIC PROTISTS

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High salt environments pose major challenges to their inhabitants. Because of the hypertonic surroundings, halotolerant and halophilic protists are exposed to a high osmotic pressure. This triggers a rapid flux of cellular water along the osmotic gradient out of the cell resulting in a highly concentrated cytoplasm. As a consequence internal macromolecules, e.g. proteins and nucleic acids denature and lose their functions. To cope with osmotic stress halophiles evolved different strategies, which so far are mainly studied in prokaryotes. One of the adaptation strategies in halophilic bacteria is the “high-salt-in-strategy”, which intracellularly accumulates inorganic ions (e.g. potassium, sodium and chloride) to provide osmotic balance with the environment. However, while bacterial adaptations are quite well explored, eukaryotes were largely ignored in this respect. This is mainly due to methodological reasons. To study the internal environment of halophilic ciliates we established an approach that relies on ion-specific fluorescent dyes. With this approach we are able to analyze the intracellular ion concentrations in living cells. Our first results with the salt-loving ciliate *Schmidingerothrix salinarum* Foissner, Filker and Stoeck 2014 isolated from the Ses Salines solar salterns (Spain) indicate that ion imaging is a potential reliable method to investigate intracellular ion changes in protists exposed to different salinities.



FLOWCAM ANALYSIS OF PROTIST ABUNDANCE AND DIVERSITY

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The Flow Cytometer And Microscope (FlowCAM®; Fluid Imaging Technologies, Inc., Yarmouth, Maine, USA) represents a combination of an automated microscope (image analysis) for detailed morphological analysis (size, shape, etc.) of protist cells and other particles with the detection of fluorescence values to further discriminate cell types similar to a flow cytometer. A statistical pattern recognition capability ('VisualSpreadsheet') is used to automatically classify the different types of microorganisms found in the sample, based upon the images taken during the passage of the flow cell and the measured characteristics of each particle. Particles ranging in size from ~4 to 200 µm can be measured in fresh and fixed samples. We use the FlowCAM to characterize protist abundance and diversity in freshwater lakes and laboratory experiments. An unequivocal identification at the species level is only possible for taxa with a highly characteristic shape and size, e.g. the dinoflagellates *Ceratium hirundinella* and *Gymnodinium helveticum* and the ciliates *Balanion planctonicum*, *Histiobalantium bodamicum*, and *Askenasia chlorelligera*. To obtain a higher taxonomic resolution, we complement our investigations with classical identification techniques such as protargol staining. We show several examples illustrating the potentially wide application of the FlowCAM technology for protist ecology and ecophysiology.



WHEN IS IT ADVANTAGEOUS TO BE MIXOTROPHIC? THE ROLE OF FOOD QUALITY AND LIGHT

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Mixotrophy is a common form of metabolism among protozoans. Currently it is still unclear under which circumstances this strategy is advantageous compared to heterotrophy. One conceivable explanation is an advantage of mixotrophy when food quality is poor. Building upon work by Wimmer (2013), which investigated only the role of food quality, we additionally studied the influence of different lighting conditions. Therefore, we compared growth rates of one mixotrophic and one heterotrophic species of the two ciliate genera *Paramecium* and *Euplotes*. Each pair of species was kept together under conditions with good, mixed and poor quality of prey, as well as in three different light:dark cycles (6h:18h, 12h:12h and 18h:6h). The cyanobacteria *Synechococcus* sp. was used as prey of poor nutritional quality, because it contains only traces of essential HUFAs (highly unsaturated fatty acids). Good food quality was given by the algae *Cryptomonas* sp. (SAG 26.80), which in turn offers a broad spectrum of HUFAs. A third treatment was a 50:50 mixture of both prey organisms. In the above-mentioned study, both mixotrophic ciliate species always had growth rates less than or equal to those of the heterotrophic species, regardless of the food quality. This is why we also expected faster growth of the heterotrophic species under short and moderate lighting durations. However we expected mixotrophs to outcompete heterotrophs when they are offered poor food quality but are also exposed to a long period of light. Under these conditions the endosymbiotic production of oxygen and carbohydrates should be sufficient to compensate the lack of food quality to the extent to which a clear advantage of mixotrophy compared to a simple phagotrophic feeding method should be observable. When the food quality is raised under the same good lighting conditions, it was expected that this advantage will decline.



25 YEARS AFTER ITS DESCRIPTION *MASSISTERIA MARINA* LARSEN & PATTERSON, 1990 GETS A SISTER: *MASSISTERIA VOERSI* SP. NOV. (CERCOZOA, LEUCODICTYIDA, MASSISTERIIDAE), A RARE SPECIES ISOLATED FROM COASTAL WATERS OF THE BALTIC SEA

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The genus *Massisteria* belongs to the family Massisteriidae within the Cercozoa (Rhizaria). For long time the genus comprised only one species, *M. marina*, based on morphological investigations and confirmed by molecular analysis of cultured strains. This small species with a biphasic life cycle feeds by filose radiating pseudopodia and has further a distinct swimming form. Regularly associations with detritus aggregates were detected for this taxon. Environmental sequences closely related to this species indicate larger species richness than hitherto described for the genus *Massisteria*. Here, *Massisteria voersi* sp. nov. (type strain IOW137) is described and ultrastructurally investigated. Several strains were isolated from brackish water at a Baltic Sea coastal monitoring station. The new species shows the typical characteristics of the genus such as alternating flagellate and amoeboid stages during its life cycle, branched thin pseudopodia with tiny extrusomes, two naked heterodynamic flagella, immobile amoeboid cells and mitochondria with tubular cristae. *Massisteria voersi* differs from *M. marina* by smaller cell size (2.3-3 μm in contrast to 2.5-9 μm) and the absence of fused motile cells. Additionally, in contrast to *M. marina* the new species does not possess a paranuclear body and shows a parallel arrangement of kinetosomes. Both species are quite distantly related regarding their 18S rRNA gene sequences. The sparse availability of environmental sequences closely related to the new species as well as first results from fluorescence in situ hybridization emphasize *M. voersi* to be a representative of low-abundance populations, the so-called “rare biosphere”.



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