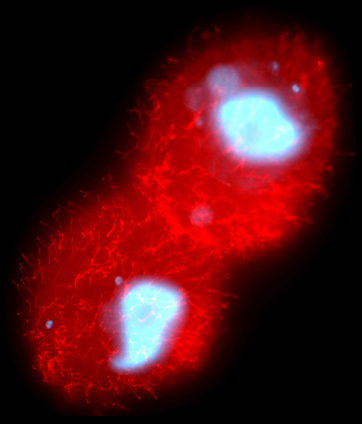
BERGISCHE UNIVERSITÄT WUPPERTAL

The German Society for Protozoology e.V. presents:

The 41st annual meeting of the DGP [summer edition]

12.-15. July, Kardinal Schulte Haus, Bergisch Gladbach



program

abstracts

participants

Organisation:
Martin Simon & Franziska Drews
Molecular Cell Biology and Microbiology
University of Wuppertal

Welcome to the 41st annual meeting of the DGP

BERGISCHE UNIVERSITÄT WUPPERTAL

Invited Speakers

Alexey Potekhin, Pisa

Ben Jenkins, Oxford

Ute Risse-Buhl, Koblenz

Gilles Gasparoni Saarbrücken

Gilles Brocart, Liege

Estienne Swart, Tübungen

Grell Prize lecture: Sabine Schiwitza, Cologne

Honorary membership lecture: Helmut Plattner, Konstanz

Organization:

Martin Simon & Franziska Drews

Division of Molecular Cell Biology and Microbiology

University of Wuppertal

Extended Organization Committee

(Individuals running around happy to help you with any question):

Marcello Pirritano

Katinka Tessier

Melanie Möller

Natalie Fabis

Diana Garza

Welcome to the 41st Annual Meeting of the German Society for Protozoology!

Two years ago in Kaiserslautern at the last conference in person, we decided to organize the 41st meeting in Wuppertal or the surrounding area. Even then, Covid-19 was already an issue, shortly before the first lock down, but even then we assumed that everything would calm down again in 2022.

The opposite is the case, Covid-19 is still present and also leads to restrictions at this conference. In the past two years, science has come more into the focus of society and we all no longer have the same self-image of being able to justify decisions with logic and scientific argumentation. Covid-19 has changed our lives and also affects this conference and its preparation.

We had initially booked in Maria-in-der-Aue in Dabringhausen, a nice hotel which was recommended by the Plant-physiology society. Unfortunately this hotel had to close after the first lockdown. We were really happy when we found a very good alternative here in Bensberg. In Autumn 2021 we then had to realize that it would not have made sense to meet in March due to the high incidence at this time and we decided for this “summer edition” of the DGP. We would like to take this opportunity to thank the current board of directors of the DGP, Hartmut, Thomas, Julia, Anja and especially Renate, who gave us a lot of support with these problems. This concerns the general organization, but also the re-organization of the homepage and the integration of a permanent conference page with registration into our system. Many thanks to Christina Bock for maintaining the website over so many years and to Christian Michelbach from art180, who was so helpful and uncomplicated in managing the changeover and adopting the site for our special needs.

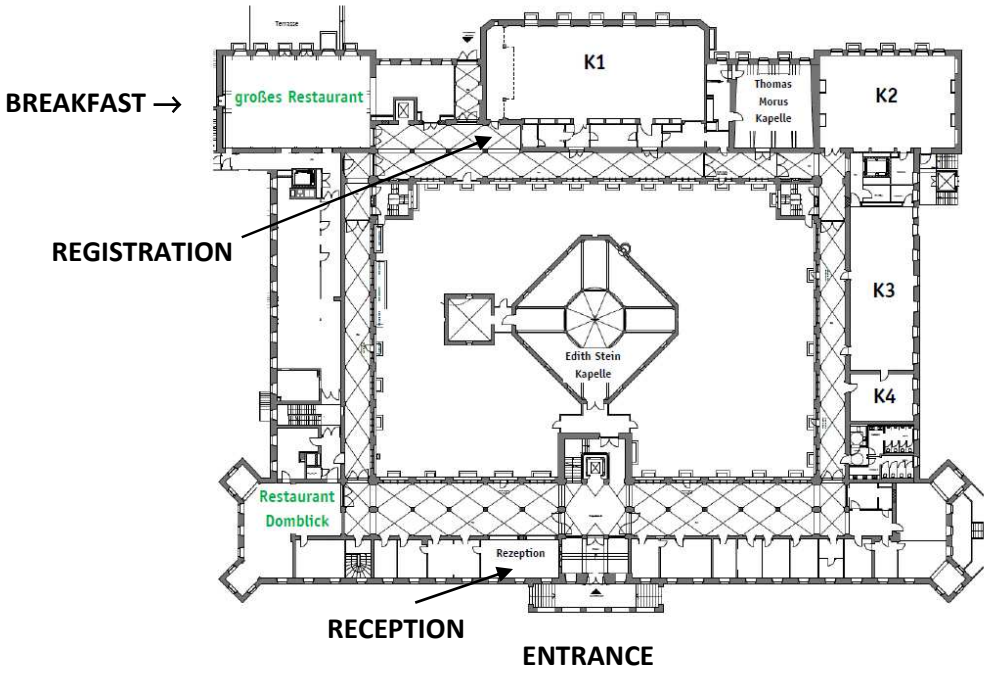
As if there weren't enough problems worldwide, the Russian war of aggression against Ukraine began in February 2022. We are appalled at Russia's attack on Ukraine and stand in solidarity with Ukrainian students and researchers and of course with the entire Ukrainian population who are being tyrannized to the highest degree. We commemorate all those killed, injured and displaced. The entirely unnecessary aggression is now having more and more global repercussions and its consequences will probably keep us away from a normal global situation for a long time to come. We are aware that not all Russian citizens are behind the aggression and we also think of the Russian colleagues who are now unable to attend our conference or, worse still, had to leave their homes and families.

We hope to offer an interesting program, an interesting meeting or simply spoken a fruitful nice meeting.

Martin & Franzi

General Information

WLAN Access	Kennung (Name):	KSH-Gast
	Passwort:	KSH-Online
PARKING	4 € first day, 3 € each following day	
BREAKFAST	from 7.00 to 10.00	
ROOMS/LOCATION	ground floor – conference first level – accommodations	



COVID19

Upon arrival for their first day of attendance, all participants and speakers / trainers must show proof of a current negative result from a supervised / certified test center. The test must have been taken no more than 24 hours before you enter the event (48 hours for PCR tests). The Bensberg testing center is only few meters from the hotel. Your certificate will be checked by members of the committee using the CovPass App.

The hotel requires the wearing of masks in all public areas inside. As we have the meeting in summer, we aim to organize as many lunches, dinners, coffee breaks outside.

We provide one additional Covid-19 antigen test in your conference bag.

We thank all of our sponsors:

Thanks to the DFG for support of invited speakers and ISOP for support of young investigators:

Additional sponsoring comes from

Program

Tuesday 12.07.2022
Room K2

Workshop RNA-seq

13.^{00}-13.^{20}	Isolation of high integrity RNA	Franziska Drews Marcello Pirritano
13.^{20}-14.^{45}	Library preparation methods	Gilles Brocart
14.^{45}-15.^{15}	***Coffee break***	
15.^{15}-16.^{15}	Setting-up, monitoring and evaluating a sequencing run	Gilles Gasparoni
16.^{15}-16.^{35}	Multiomics sequencing - Get a holistic view of biology	Samuel Kroll
16.^{35}-18.^{30}	RNA-seq bioinformatics for “non-model” protists	Estienne Swart
16.^{00}-19.^{00}	**Registration**	
19.^{00}	**Icebreaking dinner**	

Wednesday 13.07.2022

7.00-8.30 Breakfast

Room K1 – main lecture hall

8.45-9.00 Welcome & Short introduction by the organizers — Martin Simon, Franziska Drews

Session 1
Ecology

(Chairperson: Ute Risse-Buhl)

9.00-9.30	Micro-eukaryotic players in biofilms and their effect on biogeochemistry across spatio-temporal scales	Ute Risse-Buhl
9.30-9-50	Impact of tire abrasion proxies on microeukaryotic communities	Guido Sieber*
9-50-10.10	Effect of Stressors on competition between *Ochromonadales* strains belonging to different sub-clades	Lisa Boden*
10.10-10.30	Baltic Sea project: insights in the benthic protist communities in relation to sediment disturbance	Maria Sachs*

10.30-11.00 Coffee break - Coster room K2

11.00-11.20	Chitin and chitin-related factors in microbial protoplast feeders point to new roles of understudied biopolymers in protists	Jannika Moye*
11.20-11.40	Microeukaryotic gut parasites in wastewater treatment plants: Diversity, activity and removal	Jule Freudenthal*
11.40-12.00	Combining modelling and chemostat experiments to analyse non-linear dynamics that drive single-species populations	Johannes Werner*

12.00-14.00 ***Lunch***

* Talk is eligible for student talk prize

Session 2

Genetics, Genomics, Epigenomics

(Chairperson: Franziska Drews)

14.^{00}-14.^{30}	Evidence for ciliates without extensive DNA elimination: the karyorelict *Loxodes magnus*	Brandon WB Seah
14.^{30}-14.^{50}	Genome editing excisase origins illuminated by somatic genome of the ciliate *Blepharisma*	Minakshi Singh*
14.^{50}-15.^{10}	Effects of heavy metal toxicity by Cobalt and Nickel in *Paramecium*	Diana Garza-Amaya*
15.^{10}-15.^{30}	Elucidating the driving forces for genome reduction in bacteria across 44 European lakes	Manan B Shah*
15.^{30}-16.^{00}	***Coffee break - Coster room K2***	
16.^{00}-16.^{20}	Non-templated modifications of small RNA species in *Paramecium*	Marcello Pirritano*
16.^{20}-16.^{40}	Single-cell genomics of vampyrellid amoebae (Vampyrellida, Rhizaria)	Justin TP Bassiaridis*
16.^{40}-17.^{00}	The unusual properties of *Paramecium tetraurelia* macronuclear chromatin	Franziska Drews

Poster - Session A

Coster place K3 (Coffee/poster)

17.^{15}-18.^{45} ***Poster session A***

19.00 ***Dinner, Bar***

Thursday 14.07.2022

Room K1 – main lecture hall

7.^{00}-8.^{30}	Breakfast	

Session 3

Many faces of symbiosis

(Chairperson: Alexey Potekhin)

8.^{30}-9.^{00}	Trouble in *Paramecium* - cooperation and conflict in an emergent endosymbiosis	Ben Jenkins
9.^{00}-9.^{20}	Photosymbiosis in sea slugs	Gregor Christa
9.^{20}-9.^{40}	Impact of a *Nephridiophaga* (*Chytridiomycota*) infection on the fitness of the cockroach *Blattella germanica*	Renate Radek
9.^{40}-10.^{00}	*Leishmania spp.* in Central Europe	Julia Walochnik
10.^{00}-10.^{30}	***Coffee break - Coster room K2***	
10.^{30}-11.^{00}	Elusive bacterial symbionts of *Paramecium*	Alexey Potekhin
11.^{00}-11.^{20}	Breaking *Closterium*: A unique feeding strategy of a novel vampire amoeba (*Vampyrellida, Rhizaria*)	Andreas Suthaus*
11.^{20}-11.^{40}	Natural shifts in the occurrence of bacterial endosymbionts in *Paramecium*	Felicitas Flemming*
11.^{40}-12.^{00}	Transfer of bacterial endosymbionts during sexual reproduction of protists.	Kenneth Dumack
12.^{00}-13^{30}	***Lunch***	
13.^{30}-15.^{00}	***Time out: Rapid Hiking tour or an ice in the sunshine***	

Room K1 – main lecture hall

Session 4
Taxonomy and Biodiversity

(Chairperson: Jens Boenigk)

15.^{00}-15.^{20}	Integration of morphological and molecular data in determination of ciliate diversity and community structure in Alpine Lake, Mondsee, Austria	Maxwell J Owuor*
15.^{20}-15.^{40}	Patterns of protist diversity from sublittoral to abyssal sediments in the Atlantic Ocean and consequences for speciation processes	Manon Hohlfeld*
15.^{40}-16.^{00}	Ciliates in the southern Baltic	Svenja Huth*

Coster place K3

16.^{00}-17.^{30} ***Poster Session B with coffee break***

Room K2

17.^{30}-18.^{30} ***DGP membership meeting***

Room K1 – main lecture hall

18.^{45}	Banquet: appetizer	
19.^{00}	Grell prize 2022 lecture: **Diversity and ecology of choanoflagellates by integrative taxonomy**	Sabine Schiwitza
19.^{3}0	Ceremonial address: Honorary Membership **Ciliate research was sometimes faster**	Helmut Plattner
20.^{00}	Banquet	
22.^{00}	Dancing Protist, best student talk prize award, best poster prize award	

Friday 15.07.2022

7.^{00}-9.^{00} Breakfast

Room K1 – main lecture hall

Session 5

Biodiversity across habitats

(Chairperson: Alexandra Schoenle)

9.^{00}-9.^{30}	Cafeteria in extreme environments: investigations on *C. burkhardae* and three new species from the Atacama Desert and the deep ocean	Alexandra Schoenle
9.^{30}-9.^{50}	Patterns of distribution and phylogenetic and functional diversity of *Chrysophyceae* in inland waters	Jens Boenigk
9.^{50}-10.^{10}	Life at the dry limit - Gregarines from the Atacama Desert, Chile	Frank Nitsche
10.^{10}-10.^{30}	Biomonitoring of the intertidal zone using *Foraminifera* eDNA metabarcoding	David Singer
10.^{30}-10.^{50}	Molecular phylogeny of unicellular marine coccoid green algae revealed new insights into the systematics of the Ulvophyceae (Chlorophyta)	Thomas Pröschold

10.^{50} Closing remarks

11.^{15} ***Extended Coffee break – pack up your doggy bag/lunch box - Departure***

Abstracts – Talks

in order of appearance….

RNA-seq workshop

RNA Isolation for RNAseq

Marcello **Pirritano, Franziska Drews**[1]

[1] Molecular Cell Biology and Microbiology, Faculty of Mathematics and Natural Sciences, University of Wuppertal, Germany

We will briefly introduce into the different classes of RNA in eukaryotic cells and possible enrichment procedures due to modifications or size. In order to create high quality transcriptomics libraries, the integrity of RNA comes into the focus and this is still the matter of the individual who is doing the cell culture work, even if one is going to send RNA to a commercial company for sequencing.

We will explain the criteria to dissect the quality of a RNA isolate: integrity and purity in terms of contamination with DNA, salts or phenol. We will discuss aspects to improve the RNA quality, especially for RNA isolation from protists, which are difficult to centrifuge by their swimming behavior.

RNA-seq library preparation strategies

Gilles **BROCART**[1]

[1] Diagenode SA - Liège Science Park, Rue Bois Saint-Jean, 3, 4102 Seraing/Liège, Belgium

The library preparation is a key step in the NGS workflow for sequencing RNA. This is when the biological information contained in the sample in the beginning is transformed into a format that can be read by the sequencer. During this transformation, many factors can influence the quality and quantity of the outcome. Furthermore, choosing the appropriate library preparation methodology is crucial in terms of the type of information conveyed and ultimately, the nature of the sequencing results.

In this section of the workshop, it will be presented the main technologies commonly used for RNA-seq library preparation (ligation-based, template switch-based…), important factors to take into account when preparing a library, RNA enrichment techniques and typical QC measurements indicating a good library from a bad one. It will also be presented important up-to-date features figuring in RNA-seq library preparation protocols nowadays and their importance in the downstream analysis of the sequencing data (IVT, UDI, UMI…).

RNA seq workshop: setting-up, monitoring and evaluating a sequencing run

Gilles **GASPARONI**[1]

[1] Genetics department, Saarland University, 66123 Saarbrücken, Germany

In this section of the RNA seq workshop we will go through the essential steps of a next generation sequencing (NGS) run. After successful preparation and quality assessment, NGS libraries are scheduled for sequencing on a next generation sequencing platform. We will discuss how to hybridize libraries onto a sequencing flow cell to build sequencing clusters and what is important to achieve optimal cluster formation and detection. Once the run is initiated, several parameters are monitored for each sequencing cycle such as cluster density, signal to noise ratio, phasing/pre-phasing and base distribution. We will illustrate how indexing of libraries allows to combine multiple samples on the same floe cell and address important points for multiplexing of libraries and downstream demultiplexing of NGS raw data. After the run has finished, we will use tools like fastQC and multiQC to look at (raw) data quality to assess the quality of the run and the sequenced libraries. Also, we will look at potential issues such as over- / underclustering of flow cells or index swapping events and their impact on the run performance.

Multiomics sequencing - Get a holistic view of biology

Samuel **KROLL**[1]

[1] Illumina GmbH Eichhornstraße 3 10785 Berlin

Multiomics is a rapidly emerging approach that incorporates multiple methods and data sets for an integrated understanding of your samples. Enabled by cutting-edge sequencing and array technology, multiomic methods revolutionize research and push the limits of novel scientific insights to make new discoveries possible.

RNA-seq bioinformatics for “non-model” protists

Estienne **SWART**[1]

[1] Max Planck Institute for Biology, Max-Planck- Ring 5, 72076 Tübingen, Germany

Unlike many well-established organisms, with large scientific communities, gold standard reference genomes, gene predictions and pre-processed gene expression analyses readily available in public databases, more work is typically necessary to analyze RNA-seq in protists. I will introduce some of the computational tools and procedures my laboratory uses in manipulating and analyzing mRNA-seq and sRNA-seq. We use such data in predicting genes from scratch for newly sequenced genomes. Gene prediction is a good starting point for considering RNA-seq data, because without accurate genes, downstream analyses may be problematic. Plenty of errors exist in automated gene predictions that users are often unaware of. Many of these can be manually corrected upon close scrutiny of RNA-seq read mapping or fixed using custom software. I will show some of the tools and approaches we use for such inspections, and allow you to inspect the data yourself. Following this, I will introduce some simple, but powerful analyses combining RNA-seq read mapping with other useful data sources. Finally, I will discuss limitations of current popular functional analyses based on RNA-seq, particularly those reliant upon gene ontologies.

Session: Ecology

Micro-eukaryotic players in biofilms and their effect on biogeochemistry across spatio-temporal scales

Ute **RISSE-BUHL**[1]

[1] Faculty of Biology at University of Kaiserslautern & Institute for Environmental Sciences at Campus Landau**,** Erwin-Schrödinger-Straße 52, 67663 Kaiserslautern

Biofilms are complex communities of photoautotrophic and heterotrophic bacteria, protists and metazoans associated by their exopolymeric matrix to surfaces and can thus be considered as hotspots of biodiversity and biogeochemistry in fluvial, e.g., stream and river ecosystems. In general, biofilms are home for all trophic levels of the microbial food web. Protists fulfill different functional roles within biofilms, i.e., setting biofilm autotrophy, top-down control on biofilm microbes and their spatial arrangement, bentho-pelagic coupling or self-purification. Fluvial ecosystems, including their biodiversity and biogeochemistry, are hierarchically organized. Fluvial biogeochemistry is typically determined at the reach scale, the spatial unit at which nutrient spiraling metrics are quantified and also restoration practices are done. While reach-scale patterns are nested to small spatial scales and thus conditioned by them, most predictions of reach-scale biogeochemistry are based on this or larger scale constraints that often fail to explain between and within ecosystem variability. Focusing on emergent properties from smaller spatial scales, i.e., biofilms, can help to better understand larger scale patterns and processes. Thus, an improved understanding is needed to which extent smaller spatial scales, e.g., biofilm architecture and community structure, contribute to reach-scale biodiversity and biogeochemistry in order to predict fluvial ecosystem patterns. (i) At the smallest scale, phagotrophic protists modulate the spatial biofilm architecture in a way that mass transfer of oxygen, carbon and nutrients towards embedded bacterial cells is enhanced. In addition, they accelerate autochthonous and allochthonous carbon flow within biofilm communities. (ii) At the next larger spatial scale, I focus on the impact of dominant environmental factors of fluvial ecosystems related to hydromorphology, e.g., near-bed flow, sediment transport, and intermittency of flow, that vary at different frequencies on biofilm architecture, community structure, and function. Near-bed flow variability and sediment transport challenges the heterogeneous distribution of all trophic levels and their interaction within biofilm communities as well as biogeochemical processes such as nitrogen uptake and metabolism at microhabitat scale. (iii) This small-scale heterogeneity can emerge at larger scales and determine reach-scale biogeochemistry. Sorting of these heterogeneous biofilms at larger spatial scales, e.g., in mesohabitats and reaches, by hydromorphology shapes larger scale biogeochemistry. Thus, there is evidence that the microhabitat-scale spatial architecture, community structure, and function of biofilms at microhabitat scale can influence fluvial biogeochemistry at larger spatial scales.

Impact of tire abrasion proxies on microeukaryotic communities

Guido **SIEBER**[1], Daniele BEISSER[1,2], Jana OLEFELD[1], Manan SHAH[1], Mark SCHUMANN[3], Bernd SURES[2,3] & Jens BOENIGK[1,2]

[1] Biodiversity, University of Duisburg-Essen, Essen, Germany

[2] Centre for Water and Environmental Research, University of Duisburg-Essen, Essen, Germany

[3] Aquatic Ecology, University of Duisburg-Essen, Essen, Germany

Aquatic environments serve as a sink for nearly all anthropogenic discharge. A significant part of the discharge is tire wear, which is increasingly being released into the environment. Main components of tires are plastic and zinc, which can be used as proxies for tire abrasion to study the effect on microbial life. We studied the effects of nanoplastic an zinc on a microeukaryotic community using high-throughput sequencing of the 18S V9 region over a 14-day exposure period. Apart from a generally unchanged diversity upon exposure to zinc and nanoplastics, a change in community structure due to zinc is evident. Apparently, nanoplastic particles do not affect the community, but zinc addition results in functional abundance shifts concerning the trophic mode. In contrast to lasting changes in taxon composition the functional community composition is initially strongly disbalanced after application of zinc, but returns to the original state.

Effect of Stressors on competition between *Ochromonadales* strains belonging to different sub-clades

Lisa **BODEN**[1], Jens BOENIGK[1]

[1] Biodiversity, University of Duisburg-Essen, Universitätsstraße 2, 45141 Essen

The *Ochromonadales,* among other *Chrysophyceae*, are abundant and play an important role in freshwater habitats. Within the *Ochromonadales*, studies found significant differences related to temperature as well as salinity tolerance. Organisms belonging to the C2-clade are adapted to life in cold habitats, while organisms belonging to the clades C1 and C3 prefer temperate or warm habitats, respectively. The sub-clades C1, C2 and C3 consist mainly of unicellular organisms with one or two flagella, which makes it difficult to microscopically identify them based on morphological characteristics. Fluorescence *in situ* hybridization (FISH) is a robust method for identifying and quantifying microorganisms on single-cell level by hybridizing the ribosomal RNA with fluorescently labeled probes. We designed the FISH probes O1C531, O2C613 and O3C723 targeting the C1-, C2-, and C3-clade, respectively. We analyze the individual and combined effects of heat and salinity stress on the competition between the *Ochromonadales* strains JBMS11 (C1-clade), A-R4-A6 (C2-clade) and JBM10 (C3-clade) in mixed cultures by using the newly designed probes to determine the fraction of cells belonging to each of the three clades under different conditions.

Baltic Sea project: insights in the benthic protist communities in relation to sediment disturbance

Maria **SACHS**[1], Manon HOHLFELD[1], Julian WAGENHOFER[1], Svenja HUTH[1], Anja SCHERWASS[1], Hartmut ARNDT[1]

[1] University of Cologne, Institute of Zoology, General Ecology, Zuelpicher Straße 47b, 50674 Cologne

Representing one of the largest brackish environments, the Baltic Sea is surprisingly rather unstudied regarding benthic protist communities. In the context of this economically heavily exploited environment our project aims to understand the impact of bottom fisheries especially on the benthic microbial community. Considering carbon flow, protists are key players between bacterial production and higher trophic levels in the marine food web and it is therefore likely, that sediment disturbances (e.g. through bottom trawling) will have a strong impact on those relationships. In this baseline study we assessed the current state of the benthic nano- and microfauna in three marine protected areas (MPA) of the German Baltic Sea with regard to community composition, abundances and diversity as well as experimentally observe the direct impacts of bottom trawling on this community. We found that on the one hand, community structures as well as abundances highly depend on abiotic factors such as sediment type and oxygen concentration as we found higher protist abundances and e.g. micro-aerophilic species in coarser, more sandy areas. On the other hand, we could experimentally show that the direct effect of bottom trawling can be seen in a) a striking difference of abundances in the different, net induced sediment structures and that b) the net might cause the (temporary) loss of certain taxa in the uppermost sediment layers. This, we assume, can lead to a loss of its natural functions. Furthermore, we were able to establish around 50 mono-cultures, revealing a high number of taxa that had no resemblance in GenBank so far, once again showing how important taxonomic studies are for example for amplicon sequencing studies as they are able to validate results.

Chitin and chitin-related factors in microbial protoplast feeders point to new roles of understudied biopolymers in protists

Jannika **MOYE**[1], Jennifer V. GERBRACHT[1], Stefan CORD-LANDWEHR[2], Bruno MOERSCHBACHER[2], Sebastian HESS[1]

[1] Institute for Zoology, University of Cologne, Zuelpicher Straße 47b, 50674 Cologne

[2] Institute of Plant Biology and Biotechnology, University of Münster (Schlossplatz 7-8, 48143 Münster)

Chitin (a polymer of N-acetylglucosamine) and its deacetylated form chitosan are widely known for their importance in extracellular structures connected to protection (e.g. cell walls of fungi, exoskeletons of invertebrates, cysts and spores of various protists). Transcriptome data of *Orciraptor agilis* (*Viridiraptoridae, Cercozoa*), a protoplast feeder that attacks diverse freshwater green algae, revealed a large set of chitin-related proteins. *O. agilis* was found to express a chitin synthase, putative chitin binding modules (families CBM18 and CBM50), chitinases (families GH18 and GH19) and lytic polysaccharide monooxygenases (LMPOs). Surprisingly, some of these factors were highly upregulated during the attack on the algal cells, although there is no evidence for chitin or related substances in green algae. With the application of fluorescent probes, we demonstrate the presence of chitin/chitosan on algal cells that have been emptied by *O. agilis*, and present mass spectrometry data, which support this observation. It appears that a chitin/chitosan co-polymer produced by *O. agilis* is involved in the feeding event. Specifically, we hypothesize that this substance could enable the protoplast feeder to bind to the algal surface, due to the specific physico-chemical properties of the found biopolymer.

Microeukaryotic gut parasites in wastewater treatment plants: Diversity, activity and removal

Jule **FREUDENTHAL**[1], Feng JU[2], Helmut BÜRGMANN[3], Kenneth DUMACK[1]

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[2] Key Laboratory of Coastal Environment and Resources of Zhejiang Province, School of Engineering, Westlake University, 310024 Hangzhou, China & Institute of Advanced Technology, Westlake Institute for Advanced Study, 310024 Hangzhou, China

[3] Eawag, Swiss Federal Institute of Aquatic Science and Technology, 6047 Kastanienbaum, Switzerland

During wastewater treatment, the wastewater microbiome facilitates the degradation of organic matter, reduction of nutrients, and removal of gut parasites. While the latter function is essential to minimize public health risks, the range of parasites involved and how they are removed is still poorly understood. Using shotgun metagenomic (DNA) and metatranscriptomic (RNA) sequencing data from ten wastewater treatment plants in Switzerland, we were able to assess the entire wastewater microbiome, including the often neglected microeukaryotes (protists). In the latter group, we found a surprising richness and relative abundance of active parasites, particularly in the inflow. Using network analysis, we tracked these taxa across the various treatment compartments and linked their removal to trophic interactions. Our results indicate that the combination of DNA and RNA data is essential for assessing the full spectrum of taxa present in wastewater. In particular, we shed light on an important but poorly understood function of wastewater treatment – parasite removal.

Combining modelling and chemostat experiments to analyse non-linear dynamics that drive single-species populations

Johannes **WERNER**[1], Tobias PIETSCH[2], Frank M. HILKER[2], Hartmut ARNDT[1]

[1] Department of General Ecology, Institute for Zoology, University of Cologne, D-50674 Cologne

[2] Institute of Mathematics and Institute of Environmental Systems Research, School of Mathematics/Computer Science, Osnabrück University, Barbarastraße 12, D-49076 Osnabrück

The importance of oscillations and deterministic chaos in natural biological systems which was originally based on discrete-time models of population growth has been discussed since the 1970s. Later, dynamics considering interspecific and trophic interactions such as predator-prey, parasitism and cannibalism have shown all aspects of non-linear dynamics in more realistic continuous models. However, continuous-time models and experimental systems of single-species without trophic interactions characterized by non-linear dynamics are still missing, even though multiple non-linear processes are already active on the cellular level. Here we show, that dynamics of single-species systems of protists in continuous experimental chemostat systems and corresponding continuous-time models reveal typical characteristics of deterministic chaos. A new automatic video registration enabled a nearly continuous undisturbed observation of abundances with a high measuring resolution allowing for a detailed analysis of the dynamic behavior. Our simple general continuous-time model simulating the cell cycle can exhibit a remarkable spectrum of dynamical behavior and is providing explanations for the experimental results as a principle of proof with estimated parameter values. For the first time, experimental and model data demonstrate the importance of non-linear dynamics already on the level of a single type of cells without any external forcing, showing the necessity of high temporal resolution measuring methods. In future, more detailed single-species population measurements are necessary to determine parameter values exactly for being able to provide reliable models predicting the dynamical behavior in this still sparsely investigated field of research.

Session: Genetics, Genomics, Epigenomics

Evidence for ciliates without extensive DNA elimination: the karyorelict *Loxodes magnus*

Brandon KWEE BOON **SEAH**[1], Aditi SINGH[1], David Emanuel VETTER[1], Christiane EMMERICH[1], Moritz PETERS[2], Volker SOLTYS[2], Bruno HUETTEL[3], Estienne SWART[1]

[1] Max Planck Institute for Biology, Max-Planck-Ring 5, 72076 Tübingen, Germany

[2] Friedrich Miescher Laboratory, Max-Planck-Ring 9, 72076 Tübingen, Germany

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Each ciliate cell has two types of nuclei: the germline micronucleus (MIC), which is usually transcriptionally silent, and somatic macronucleus (MAC), where most gene expression takes place. In most ciliates, MACs develop from MIC precursors following sexual conjugation. Thousands of interspersed DNA segments called internally eliminated sequences (IESs) ranging from 10s to 1000s bp length are eliminated, and chromosomes also experience fragmentation, rearrangement, and amplification, although the extent varies between species. The MAC genome content is hence a subset of the MIC genome.

Here we report an apparent lack of IESs in the ciliate *Loxodes magnus*. We separated MIC and MAC by flow cytometry and confirmed purity by morphology and Western blotting against MAC-specific markers. Unexpectedly, sequence libraries prepared from purified nuclei showed only a small fraction of k-mers specific to the MIC libraries. Indels found by mapping MIC reads against MAC draft genomes appeared to represent allele variants rather than IESs. Both genomes were relatively large: after filtering out low-complexity repetitive sequences (up to 43% of total), assemblies were 450-470 Mbp (MAC) vs. 460-480 Mbp (MIC). There may hence be some MIC-limited genome content, but not in the form of typical abundant interspersed IESs. Nonetheless, histone markers and nucleosome profiling suggest that the MACs are the site of active gene expression, like other ciliates. *Loxodes* belongs to the class Karyorelictea, whose MACs cannot divide vegetatively unlike other ciliates, and so are obliged to undergo MIC-MAC development for every cell division, including vegetative divisions. We hypothesize that the loss or streamlining of genome editing may be a way to avoid costly overheads during cell division.

Genome editing excisase origins illuminated by somatic genome of the ciliate *Blepharisma*

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Massive DNA excision occurs regularly in ciliates, ubiquitous microbial eukaryotes with somatic and germline nuclei in the same cell. Tens of thousands of internally eliminated sequences (IESs) scattered throughout a copy of the ciliate germline genome are deleted during development of the streamlined somatic genome. *Blepharisma* represents one of the two earliest diverging ciliate classes, and, unusually, has dual pathways of somatic nuclear development, making it ideal for investigating the functioning and evolution of these processes. Here, we present the somatic genome assembly of *Blepharisma stoltei* strain ATCC 30299 (41 Mb), arranged as numerous alternative telomere-capped minichromosomes. This genome encodes eight PiggyBac transposase homologs liberated from transposons. All are subject to purifying selection, but just one, the putative IES excisase, has a complete catalytic triad. We propose PiggyBac homologs were ancestral excisases that enabled evolution of extensive, natural genome editing.

Effects of heavy metal toxicity by Cobalt and Nickel in *Paramecium*

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Cobalt (Co) and Nickel (Ni) became important environmental contaminants since they are among others present in lithium batteries which are commonly used and sometimes trashed into the river Rhine by “stupid” people.

We used *Paramecium* to characterize the toxicity of both metals as a chloride salt. We found that Ni shows higher toxicity with a LD50 of 323 µM compared to a LD50 of 1504 µM for Co after 2h exposure. We show by inductively coupled plasma optical emission spectrometry (ICP-OES) that paramecia can handle much higher amounts of cellular Cobalt as compared to cellular Ni.

In the environment, also the co-exposure becomes relevant. Co-exposing Ni and Co seem not to alter the individual uptake, both metals are ingested in relation to the exposed concentration, which let us assume that they are taken up independently from each other.

In another set of experiments, we dissected the impact of the food bacteria to the cellular uptake. We found that bacteria bind a massive amount of Ni and Co. Hypothesizing that food bacteria may be a shuttle for the metals, we pre-exposed bacteria with Ni and/or Co and fed these to paramecia: we can then show the bacterial delivery of Ni/Co to paramecia by ICP-OES. Thus, we conclude that there at least two different uptake mechanisms for Ni and Co in *Paramecium*: uptake of soluble salts likely by pinocytosis and uptake by food vacuoles with bacterial delivery.

However, starving paramecia without food bacteria show a higher sensitivity to both metals (LD50 of 185 µM for Ni and 1347 µM for Co). This is in conflict with a bacterial shuttle for metals. It seems likely that this is the result of either a different uptake mechanism (phagocytosis vs. pinocytosis), due to a bacterial metabolization leading to a reduced bioavailability of the metals or simply due to the fact that free bacteria represent a buffer for the metals thus reducing the free concentration.

To inquire the cellular response to both metals, we analysed the transcriptome of paramecia exposed to sublethal concentrations of Ni and Co. Both toxins induce common responses e.g. the down-regulation of ammonium transporter activity and acyltransferase activity, and the up-regulation of catalytic activity on tRNA, which is a common stress response of eukaryotic cells. Otherwise, we also found several differences in the transcriptomes for Ni and Co exposure. Comparing them, we can identify higher levels of genes associated with cell-redox homeostasis in Ni exposure, which could suggest that Ni causes some oxidative stress to the cells. Our results show that exposure of different metals not only alone but also in different combinations and ratios cause specific and characteristic transcriptomic responses indicating dynamic metabolic adaptations to different combinations of metals..

Elucidating the driving forces for genome reduction in bacteria across 44 European lakes

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Bacteria are in constant state of evolution, and any alteration/adaptation that helps them in survival and/or propagation is advantageous for them. One such evolutionary adaptation is a reduction in their genome sizes. In this study we examine the mechanism of genome reduction and the possible external forces, in the form of nutrient limitation (genome streamlining) or grazing pressure (genome reduction), that can promote or expedite this process. We also study the habitat preference of the most common bacterial species in our survey of European freshwater ecosystems. We employed over 300 good quality Metagenome Assembled Genomes (MAGs) extracted from metagenomes of 44 environmentally diverse lakes spread across the European continent. We observe that genome reduction results in various genomic changes, these include, in addition to an overall reduced number of genes, three other major changes: (1) A distinct change in GC preferences in certain phyla, especially in form of biased codon preference, (2) fewer non-essential genes including non-essential sigma factor genes, certain biosynthesis and motility genes, (3) more compact genomes with very few non-coding regions. It is also possible that nutrient limitation and predation can be working in concert as pressures to cause genome reduction. We demonstrate the benefits of high-throughput genome reconstruction from metagenomes on the one hand to identify and describe understudied but ecologically significant lake bacteria and their habitat preferences and on the other hand to investigate evolutionary mechanisms that manifest themselves in genomic signatures.

Non-templated modifications of small RNA species in *Paramecium*

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Regulation of gene expression is a crucial task an organism has to perform in order to stay alive, hence a lot of different mechanism of gene expression regulation has evolved. While some of those mechanism act on a genomic level and involve repression and activation of gene expression by promoter, enhancer and transcription factors, other mechanism can alter the expression of the gene on a level beyond the genomic “hardware”. One of those mechanism is RNAinterference, a procedure where small RNAs attack either mRNAs or nascent RNA transcripts in a homology dependent manner, which leads to the phenotypical silencing of a specific gene.

While most of the enzymes involved in RNAi mechanism are self-explanatory in their function, e.g. a Dicer usually cuts dsRNA to small RNA duplices, some other enzymes are more miscellaneous regarding their function/targets. An example for those enzymes are Cid-proteins, which are capable of modifying RNAs with nucleotides that were not present during their biogenesis, hence adding so called “untemplated nucleotides” to those RNAs.

In *Paramecium*, at least two different Cid-Proteins, Cid1 and Cid2, are involved in modifying RNAs derived from different RNAi-pathways present in the cell, e.g. “RNAi by Feeding” and transgene induced RNAi.

In this study, we want to characterize the extent to which small RNAs derived from these pathways are modified with untemplated nucleotides and see whether we can use this information to make assumptions about the biogenesis of those smallRNAs and the biological requirements and functions of the untemplated modification of those RNAs.

Single-cell genomics of vampyrellid amoebae (Vampyrellida, Rhizaria)

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The vampyrellid amoebae (Vampyrellida, Rhizaria) are naked, filose amoebae that live in marine, freshwater and soil habitats. Phylogenetic trees based on the SSU rRNA gene indicate that the vampyrellids are genetically diverse organisms, spanning over several family-level clades. The members of these clades exhibit diverse morphologies as well as interesting variations in their feeding strategies and prey range specificities. Vampyrellids are well known to attack algae, fungi and even small animals, suggesting that they have important ecological roles. Currently, we still lack genomic data of vampyrellids, which could help understand the evolution and diversification of these rhizarian amoebae. We established a workflow for “single-cell genomics” based on whole genome amplification and the short-read Illumina sequencing technology, and generated 12 functionally annotated de novo draft genomes of diverse vampyrellids. The assemblies had a BUSCO completeness of 70-80 % and we could identify thousands of protein-coding genes per species with the help of reference transcriptomes. Here, we discuss technical aspects of our workflow and present some preliminary findings from comparative vampyrellid genomics.

The unusual properties of *Paramecium tetraurelia* macronuclear chromatin

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Epigenetic regulation in unicellular ciliates can be as complex as in metazoans and is well described regarding small RNA mediated developmental genome rearrangements. The global chromatin organization in the vegetative macronucleus (MAC) however is poorly understood and unusual characteristics of the somatic nucleus, like high polyploidy, high genome coding density and absence of heterochromatin are ought to call for complex regulation to orchestrate gene expression.

In this study, data on the nucleosome organization in the highly crowed nucleus will be presented, including the description of histone post translational modifications (PTMs) that are well described in terms of gene expression regulation in metazoans. The study includes the first data on PTMs on the vegetative histone H3 N-terminal tails that were detected by mass spectrometry. The repressive mark H3K27me3, usually described as being exclusively linked to developmental gene silencing in metazoans and developmental genome processing in Paramecium anlagen, was indeed detected in the vegetative MAC. From immunoprecipitation experiments and combinatorial patterns of histone marks, we draw a model on gene expression regulation involving repressive and activating marks in broad chromatin domains together with low Polymerase II processivity.

The Paramecium Pol II CTD is highly divergent, lacking the heptameric repeat structure that becomes phosphorylated to regulate stages of transcription in many species. How Pol II elongation is controlled in Paramecium remains obscure, but differential gene expression data from silencing of the associated Paf1 elongation complex offers preliminary insights on Paramecium's divergent gene expression machinery.

Session: Many faces of symbiosis

Trouble in *Paramecium* - cooperation and conflict in an emergent endosymbiosis

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Endosymbiosis – one organism living within another – is a biological process which sparked an explosion of evolutionary innovation. Yet an outstanding question of biology is how unrelated, fundamentally selfish, organisms can form a long-term, stable endosymbiotic interaction. Conflict is inherent within endosymbiosis, and can result in break-down or over-exploitation of the interaction. However, stability is required for the metabolic and genetic integration that drives the formation of obligate endosymbioses and new organelles. Identifying the molecular mechanisms that maintain stability in an emergent endosymbiotic interaction is important for understanding how endosymbioses have evolved.

Paramecium bursaria is a ciliate protist harbouring mutualistic green algae. The endosymbiosis is both heritable and facultative, making *P. bursaria* a valuable model to explore the mechanisms that govern an emergent endosymbiotic interaction. Importantly, *P. bursaria* is genetically tractable via RNA-interference (RNAi) allowing disruption of host genes that maintain the endosymbiosis. Through this approach we have revealed: (i) a mechanism of RNA-RNA inter-actions that punish the host for endosymbiont digestion; (ii) a host 'metabolic switch' of polyamine biosynthesis that controls nitrogen flow to the endosymbiont; and (iii) a non-catalytic host chitinase, resembling an innate immune factor, involved in perception and regulation of endosymbiont breakdown.

These findings support that there is not one recipe for endosymbiosis in *P. bursaria*, but rather a series of distinct regulatory pathways acting simultaneously to maintain the interaction. These mechanisms have functions that vary transiently in response to internal and external stimuli, including conflict between symbiotic partners. In *P. bursaria* these processes have acted to promote cooperation and stability, driving maintenance of the emergent endosymbiotic inter-action across fluctuating ecological conditions. However, the nature of these mechanisms emphasize the blurred distinction between mutualism and antagonism in endosymbiosis. We should therefore think of endosymbiosis not simply as a 'mutualism', but rather as a protracted, context-dependent power struggle.

Photosymbiosis in sea slugs

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Photosynthesis is restricted to certain bacteria, algae, and plants. Animals are heterotrophs and their only option to directly tap the benefits of photosynthesis is the establishment of a symbiosis with a phototrophic organism (photobiont). In animals, photosymbiosis is found in taxonomically divergent taxa and is considered highly beneficial for both partners. Basis of this symbiosis is a nutrient exchange between the host and the photobionts that can even supersede the nutritional demands of the host enabling growth in oligotrophic waters. In most cases, the photobionts are dinoflagellates belonging to the Symbiodiniaceae and photosymbiosis best understood in reef building corals, which has provided a reference system of the molecular fundamentals underpinning photosymbiosis. In other animals, for instance sea slugs, this is poorly understood. Among sea slugs, two different systems of photosymbiosis evolved, once in Nudibranchia and once in Sacoglossa. Nudibranchia obtain their Symbiodiniaceae through feeding on photosymbiotic cnidarians and become the only known secondary host among photosymbiotic animals. Sacogolossa pierce the cell walls of green macroalgae, suck out the cell content, and subsequently only retain the chloroplasts as some sort of photobiont. In both system the photobionts are incorporated intracellularly in cells of the digestive gland system and are surrounded by a phagosomal membrane. Both systems are highly relevant for the understanding of the (molecular) evolution of photosymbiosis in animals. Nudibranchia allow a firsthand comparison with one of the evolutionary oldest photosymbiosis in animals, the cnidarians they feed on, and Sacoglossa allow to understand what impact the evolutionary history of the photobionts has on a photosymbiosis. Hence, sea slugs can deliver a comprehensive and unprecedented insight to understand how to become and be a photosymbiotic animal.

Impact of a Nephridiophaga (Chytridiomycota) infection on the fitness of the cockroach *Blattella germanica*

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Nephridiophagids are unicellular fungi (Chytridiomycota), which infect the Malpighian tubules of insects. While most life cycle features are known, the effects of these endobionts on their hosts remain poorly understood. Here, we present results on the influence of an infection of the cockroach *Blattella germanica* with *Nephridiophaga blattellae* (Ni = *Nephridiophaga*-infected) on physical, physiological, and reproductive fitness parameters. Since the gut nematode *Blatticola blattae* is a further common parasite of *B. germanica*, we included double infected cockroaches (N+Ni = nematode plus Ni) in selected experiments. Ni individuals had lower fat reserves and showed reduced mobility. The lifespan of adult hosts was only slightly affected in these individuals but significantly shortened when both *Nephridiophaga* and nematodes were present. Ni as well as N+Ni females produced considerably less offspring than parasite-free (P-free) females. Immune parameters such as the number of hemocytes and phenoloxidase activity were barely changed by *Nephridiophaga* and/or nematode infections, while the ability to detoxify pesticides decreased. Quantitative proteomics from hemolymph of P-free, Ni, and N+Ni populations revealed clear differences in the expression profiles. For Ni animals, for example, the down-regulation of fatty acid synthases corroborates our finding of reduced fat reserves. Our study clearly shows that an infection with *Nephridiophaga* (and nematodes) leads to an overall reduced host fitness.

Leishmania spp. in Central Europe

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Leishmania spp. are parasitic protists transmitted by the bites of female sand flies (Phlebotominae). In humans, they can cause visceral leishmaniosis and various forms of cutaneous leishmaniosis. Worldwide, around 12 million humans are infected and more than 50,000 deaths are recorded every year, mainly due to the visceral form. In Central Europe, *Leishmania* infections are generally imported infections, mostly associated to travelling and migration. However, occasional autochthonous cases in humans as well as in animals have also been reported and the question arises, if *Leishmania spp.* have arrived in Central Europe? This study aimed to evaluate reported cases of *Leishmania* infections in Central Europe in the past years for the involved *Leishmania* species and review the current state of knowledge on available reservoirs and vectors for *Leishmania spp.* in Central Europe.

Elusive bacterial symbionts of *Paramecium*

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Many bacteria are known only from metagenomic data. This was the case for an orphan group of the 16S rRNA gene sequences which could not be attributed to any family within the order Rickettsiales until the first alive representative of a new family Deianiraeaceae was found as an epibiont of ciliate *Paramecium primaurelia* (Castelli et al., 2019). Now we report the second representative of this family, which developed fast acute infection on the cell surface of ciliates *Paramecium calkinsi*. Infected alive and dead ciliates were contagious; infection, lethal for all recipient cells, was transmitted via medium. The thick layer of small bacteria was formed over the ciliate cell cortex disrupting cilia. The metagenome of the infected ciliates was sequenced on Illumina and Oxford Nanopore platforms. We succeeded to assemble two complete bacterial genomes. One of these belonged to *Flavobacterium sp.*, while the other bacterium represented the new genus of Deianiraeaceae family. Specific probes targeting 16S rRNA were designed for both bacteria, and FISH unequivocally showed that it was the new representative of Deianiraeaceae that resided on the surface of paramecia. Comparative genome analysis of Deianiraeaceae representatives provided insights into the mechanisms of host-symbiont interactions and evolution of this very specific group of extracellular Rickettsiales, which might be isolated from nature only on peak of the ongoing rapid infection process in its protozoan host.

Flavobacterium sp. acted as scavengers feeding on the dead ciliates in the system described above. However, *Flavobacterium* can be commonly found in *Paramecium* microbiomes, and at some occasions it can become an intracellular symbiont. We observed a complex succession of the symbionts in the *Paramecium bursaria* strain. Initially cytoplasm of its cells was shared by symbiotic microalgae and bacterial endobionts which turned to be representatives of a new Holosporaceae genus. When the ciliates were cured of algae with cycloheximide, bacteria also disappeared, and the vacant cytoplasmic compartment was invaded by Flavobacterium sp. However, this association was a subject for further successions of the symbionts.

The Holosporaceae symbionts sometimes are also demanding and fragile. We found another Holosporaceaen bacterium inhabiting the cytoplasm of *Paramecium caudatum*. This symbiont appeared to be psychrophilic, bacteria were able to survive in the host cells at temperature +10-12° C. Moreover, spontaneous loss of bacteria led to increase of thermosensitivity of the host ciliates, which also could not survive at temperature over +16° C.

Thus, probably, many bacterial symbionts of ciliates may be observed only at certain environmental conditions or under “lucky” circumstances. Such elusive bacteria may represent a huge part of biodiversity which is known only from metagenomic sequences.

Breaking *Closterium*: A unique feeding strategy of a novel vampire amoeba (Vampyrellida, Rhizaria)

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The order Vampyrellida comprises predatory amoebae, which form a large and genetically diverse clade of rhizarian protists. We know that these intricate microbes specifically prey on other eukaryotes, most famously by localized perforation of the algal cell wall. So far, four distinct feeding strategies have been reported among the Vampyrellida. As part of the 'Taxon-omics' Programme (German Research Foundation), we examine the Vampyrellida in a more modern light and discover new lineages. Here we will report on a novel vampyrellid belonging to the Leptophyridae which possesses a hitherto unknown feeding strategy. This 'fifth' feeding strategy involves the uptake of whole *Closterium* (Zygnematophyceae) cells, 'breaking' of the cell wall along the isthmus and subsequent discarding of the emptied *Closterium* remnants. We will showcase this vampyrellid by light microscopy, examine its feeding process and prey spectrum, highlight its lifecycle, characterize its phylogenetic position and compare it to other known vampyrellids and their feeding strategies. We will also offer some conceptual insights into the mechanisms behind this strategy. This data highlights the diversity of feeding strategies in the Vampyrellida and shows how much remains to be discovered about these widespread and fascinating microbes.

Natural shifts in the occurrence of bacterial endosymbionts in *Paramecium*

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Symbiotic interactions are complex yet important factors affecting the realized ecological niche of organisms - thereby influencing their occurrence, relative abundance, and determining the fitness of each symbiotic partner. *Paramecium* and its diverse bacterial symbionts are a suitable model system to address molecular and eco-evolutionary consequences and questions regarding these symbiotic associations. In such system, knowledge about the miscellaneous role of bacterial symbionts harbored by heterotrophic *Paramecium* species is still limited. Obligate intracellular bacteria allegedly always inflict costs. However, various experimental studies have shown paramecia carrying bacterial endosymbionts can benefit from their infection.

We address the question which endosymbionts occur in natural paramecia populations isolated from a small lake over 5 years and which factors might explain observed shifts and persistence in the symbionts' occurrence. From over 100 monoclonal strains investigated, approximately two-third harbored intracellular bacteria. While "*Candidatus Megaira polyxenophila*" was observed in the majority of isolates, *Caedimonas varicaedens* was detected only once. After the appearance of *C. varicaedens*, "*Ca. M. polyxenophila*" prevalence dramatically dropped with some delay but recovered to original levels at the end of the surveyed period. Possible mechanisms explaining such observations include differences in infectivity, host range, impact on host fitness and host competitive capacities. Growth experiments revealed fitness advantages for infected paramecia harboring "*Ca. M. polyxenophila*" and *C. varicaedens*. Furthermore, we showed cells carrying *C. varicaedens* gain a competitive advantage from the symbiosis-derived killer trait. Other characteristics like infectivity and overlapping host range were taken into consideration, but the observed temporal persistence of "*Ca. M. polyxenophila*" is most likely explained by the symbiont's positive effect to its host.

This work's experimental approach provides an additional perspective to the observed association's occurrences. Insights into genetic broadness of potentially realizable symbioses allow assessment of additional biotic or abiotic factors' impact shaping the formation and occurrence of symbioses.

Transfer of bacterial endosymbionts during sexual reproduction of protists

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For long endosymbiotic bacteria in protists were overlooked unless being detrimental, especially markable, or leading to changed phenotypes. Nonetheless, inconspicuous endosymbiotic bacteria are present in every other protist culture. Most of these bacteria seem to be exclusively vertically transmitted to daughter cells during mitosis. In endosymbiosis theory, vertically transmitted endosymbionts must adapt to mutualism; parasitic taxa are expected to evolve into mutualists unless being occasionally horizontally transmitted, i.e. entering a novel evolutionary line of hosts. Natural conditions can not be replicated in the laboratory and accordingly it is especially unclear how endosymbiotic parasites are transferred. One may ask: Is sex involved in the dispersal of endosymbiotic bacteria? Here I will present some recent and yet unfinished work on the link between sexual reproduction and the transmission of endosymbiotic bacteria. Many protistan taxa fuse occasionally in their life history and it is still poor-ly understood why. I will present a comparative transcriptomics study that indicates that during the fusion of *Fisculla terrestris* (Cercozoa, Rhizaria) meiosis genes are differentially expressed, potentially indicating sexual reproduction. However, we also challenge common presumptions regarding meiosis in eukaryotes. We hypothesize that even the occasional link between sex and transmission of endosymbiotic bacteria opens up parasitism to vertically transmitted endosymbionts.

Session: Taxonomy and Biodiversity

Integration of morphological and molecular data in exploring ciliate diversity and community structure in the alpine Lake, Mondsee, Austria

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Ciliates are excellent indicator protists for water quality assessment. However, ciliate communities on submersed plants and stones from lake shorelines are relatively unknown. Therefore, we here integrated morphological and molecular datasets of several ciliate species to present an informative and reliable taxonomic approach on studying ciliate diversity and community structure in Lake, Mondsee, Austria. Live observation, morphometric measurements and protargol staining were used for revealing morphological details of the ciliates. Moreover, their molecular sequences (ribosomal operon, ITS) were investigated. The most abundant ciliates belonged to the Oligotrichida, the Hymenostomatia, the Haptorida, the Colpodea, and to the Peritrichia. According to this integrated approach, we will add valuable information on some ciliate species in respect to adding new single cell sequences to publicly available databases and reveal their ecological role in lake shore communities.

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Patterns of protist diversity from sublittoral to abyssal sediments in the Atlantic Ocean and consequences for speciation processes

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Only recently protists inhabiting deep-sea sediments became subject of next-generation-sequencing studies revealing their high and specific diversity. A range of different species has also been cultivated from the deep sea and some have proven able to survive and even grow under the conditions prevailing in this harsh environment. However, little is known about vertical distribution patterns of benthic protists from sublittoral down to abyssal depths, although this knowledge is important to understand evolutionary processes among protists. If islands/seamounts are surrounded by deep-sea areas, they can act as “stepping stones” for species allowing the distribution over large distances or they can act as “trapping stones” stopping the distribution of species and the gene flow between populations.

Using a combination of metabarcoding, live-counting and cultivation techniques, we investigated the taxonomic composition and abundance of benthic protist communities in sediments from sublittoral to abyssal depths (50-3000m) around three islands and two seamounts of the Azores’ archipelago in the North Atlantic Ocean. Protist abundance decreased significantly and community composition changed with increasing depth. While some species were found at all depths, others were only detected in sublittoral or lower bathyal sediments, indicating that some benthic taxa are limited in their distribution to a certain depth, whereas others are also present at the deep-sea floor. The proportion of unidentified specimens increased with depths pointing towards a high number of so far still undetected species in the deep-sea realm.

Ciliates in the southern Baltic (MGF-Ostsee project)

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Although, marine benthic protist communities are assumed to play an essential role in terms of the microbial food web and to have severe influence on the composition and dynamics of the ecosystem, sufficient data on the marine benthos are still rather scarce. One of the most diverse and ecologically important groups of unicellular eukaryotes is the phylum Ciliophora, a group that shows similar morphological characters such as cilia, dimorphic nuclei, and a cell cortex. Here, the benthic ciliate diversity of the Oderbank in the southern Baltic Sea is addressed by characterizing new species we found, members of the genii Euplotes and Aspidisca. Based on SSU rDNA sequences, as well as a description of the affiliated morphology received through different staining techniques (e.g., Protargol, DAPI and dry silver nitrate stain) and scanning electron microscopy, respectively, one Euplotes and one Aspidisca species could be described as new species. The assignment to a genus level based on taxonomic analysis was done for all six isolated strains. In some cases, the obtained ciliate phylogenies revealed close relations to environmental sequences isolated under similar habitat conditions. Interestingly, some ciliate strains showed affiliations to anaerobic genotypes, that should be further investigated during ongoing studies of the MGF-Ostsee project, which currently investigates the impact of trawling fishery on the benthic ecosystem of the Baltic Sea. Furthermore, we will present results of the occurrence of these species under different salinities and classify them within already existing data for the corresponding genus (Syberg-Olsen et al. 2016). The gained knowledge shall help estimating and defining species richness by comparing the results to a metabarcoding analysis of environmental samples obtained during the MGF-Ostsee project. We hope to add valuable information on a small fraction of the Baltic benthic protist community and support future studies on ciliate diversity and function.

Grell prize lecture

Diversity and ecology of choanoflagellates by integrative taxonomy

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Innovative molecular techniques, as high-throughput sequencing, produce massive amounts of reads, which vastly expand our knowledge on protist biodiversity patterns, but only few of these generated sequences can be assigned to any known eukaryotic group. Thus, an urgent need exists in terms of matching known morphospecies with anonymous sequence data in order to refine and augment the future knowledge output of the rapidly developing molecular toolbox.

Within our study, we focus on an integrative taxonomical approach to extend common reference databases with encompassing species descriptions, which is still fundamental regarding the interpretation of any metabarcoding study. These integrative data will allow for a comprehensive analysis regarding protist biodiversity, community structures and their ecological functionality. In this context, we investigated diverse habitats (from freshwater to hypersaline water bodies) to extend our current knowledge on the diversity and ecology of one group of organisms, the choanoflagellates, which we use as a model group for our studies on integrated taxonomy. Based on detailed morphological (light and scanning electron microscopy), molecular (SSU and LSU rDNA sequencing as well as transcriptomic) and autecological (salinity tolerance) data, we were able to describe several new choanoflagellate species. The combination of different methodologies resulted in detailed species descriptions for a comprehensive integrative taxonomy, which can elucidate the ecological and evolutionary role of choanoflagellates. The present thesis emphasizes the urgent need of taxonomy to generate reliable reference data for further molecular meta-analyses investigating the biodiversity of protists.

Ceremonial address: Honorary membership

Ciliate research was sometimes faster

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Ciliate research was several times leading in the elucidation of specific aspects of cell biology. This ranges from work honored by a Nobel Prize, i.e,. identification of telomerases and and ribozymes, to a variety of other fields. Most important were discoveries about ciliary activity. Many proteins were elucidated on a molecular level and localized in ciliate cells by means of widely different methods. Time resolved electron microscopy revealed unexpectedly rapid exocytosis/endocytosis coupling, to give just two examples. Many proteins were identified and their function elucidated on a molecular level and also localized in ciliate cells by means of widely different methods. Currently most important work comes from the study of transgenerational epigenetics with ciliates - a topic rather hotly debated with mammalians.

Session: Biodiversity across habitats

Cafeteria in extreme environments: investigations on *C. burkhardae* and three new species from the Atacama Desert and the deep ocean

Alexandra **SCHOENLE**[1], Manon HOHLFELD[1], Alexandra RYBARSKI[1], Maria SACHS[1], Frank NITSCHE[1], Hartmut ARNDT[1]

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The heterotrophic nanoflagellate genus *Cafeteria* has been found to be ubiquitously distributed in the marine realm. We could isolate and cultivate ten strains morphologically similar to *Cafeteria* from various types of environment, including the deep sea, brackish waters and also meso- to hypersaline inland waters of the Atacama Desert in Chile. Molecular analyses targeting the 18S rDNA, ITS-1 and 28S rDNA and morphological characterizations obtained by high resolution microscopy were conducted for all strains. Besides *Cafeteria burkkhardae* isolated from the North Atlantic Ocean, we described two new species from the Atacama Desert and one new species from brackish waters of the Baltic Sea, while one other strain could not yet be assigned. In addition, we exposed several strains to different salt concentrations (2-150 PSU) to investigate their salinity tolerance and their possibility to inhabit a broader spectrum of habitats including hypersaline environments besides the deep sea with its high hydrostatic pressure.

Patterns of distribution and phylogenetic and functional diversity of Chrysophyceae in inland waters

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While biogeographical patterns are well-known for plants and animals in Europe, we here investigated diversity and distribution patterns of protist freshwater communities on a European scale (256 lakes) in the light of the well-studied post-glacial distribution patterns of macroorganisms. Thus, we compared alpine protist communities of lakes located in the Alps, Carpathians, Pyrenees, and the Sierra Nevada with that of surrounding lowland lakes. High proportions of region-specific alpine specialists indicate an increased occurrence of distinct lineages within each mountain range and thus, suggested either separated glacial refugia or post-glacial diversification within mountain ranges. However, a few alpine specialists were shared between mountain ranges suggesting a post-glacial recolonization from a common lowland pool. Our study demonstrates that Chrysophyceae are one of the most common groups in freshwaters. Therefore we examplarily link OTU diversity of Chrysophyceae with phylogenetic affiliations based on an extensive phylogenetic tree and phylogenetic placement. Ochromonadales and a Chrysosacca-Apoikiida clade (including Apoikiida, Chrysosaccales, Chrysastrella) are the most widespread Chrysophyceae groups and show a high degree of OTU diversity. Most detected and assignable OTUs were affiliated with mixotrophic Chrysophyceae. Niche width differs only slightly between members of different clades and between the different trophic modes.

Life at the dry limit - Gregarines from the Atacama Desert, Chile

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Gregarines are endobiontic apicomplexans (Alveolata, Eukaryota), found within terrestrial and aquatic invertebrates. Most of them are assumed to be host specific and therefore of special interest for host-endobiont co-evolution and population studies. Gregarines inhabiting darkling beetles of the Atacama Desert are particular interesting for these studies, since the molecular clock of their hosts, as well as the geological origins and tectonic formatting of their habitat have been well described. In this study we focused on the establishment of a replicable and universal method for the isolation and amplification of rRNA from gregarines, to study evolutionary processes of gregarines and present first results on gregarines from this extreme environment.

Biomonitoring of the intertidal zone using Foraminifera eDNA metabarcoding

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Environmental biomonitoring is a prerequisite for adapted and efficient actions to remediate ecosystem degradation due to pollution or climate change. Estuaries, particularly intertidal mudflats, are key habitats to monitor these changes because of the interactions of anthropogenic and natural terrestrial, freshwater and marine stressors. Foraminifera (Eukaryote, Rhizaria) are excellent bioindicators of environmental conditions. A large literature based on morphological characterization has demonstrated the sensitivity of this clade to changing environmental parameters like heavy metal pollution, natural stress, salinity... Today, advances in molecular tools have open new and promising perspective in the assessment of this diversity. Nevertheless, the development and the validation of the different sampling protocols, molecular procedures and data analyses remain to be completed before such tools can be routinely applied.

We conducted a large environmental DNA survey of six estuaries of the French Atlantic coast. A metabarcoding approach using specific formainiferal markers was used, and amplicons were sequences using an Illumina MiSeq platform. To analyse the dataset, we developed our customs bioinformatics pipeline as well as a reference database including all existing foraminifera sequences. We obtained 104 taxa belonging to Monothalamea and Globothalamea. We also recovered species that are commonly found in morphology based studies (Ammonia spp., Haynesina sp.). The analysis of the distribution of the foraminiferal communities showed that 40% of the variance was explained by the chemical composition of the sediment and the climatic conditions.

Our study using eDNA metabarcoding presents a promising tool to explore and characterize the diversity of Foraminifera in intertidal mudflats. Our results demonstrate the strength but also the future challenges of eDNA based studies that remain to be assessed for the development of reliable indices on the environmental conditions. Since eDNA metabarcoding gives highly complementary information compared to traditional morphological studies, the simultaneous application of both methods may yield optimal results.

Molecular phylogeny of unicellular marine coccoid green algae revealed new insights into the systematics of the Ulvophyceae (Chlorophyta)

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Most marine coccoid and sarcinoid green algal species have traditionally been placed within genera dominated by species from freshwater or soil habitats. For example, the genera *Chlorocystis* and *Halochlorococcum* contain exclusively marine species; however, their familial and ordinal affinities are unclear. They are characterized by a vegetative cell with lobated or reticulated chloroplast, formation of quadriflagellated zoospores and living epi- or endophytically within benthic macroalgae. They were integrated into the family Chlorochytriaceae which embraces all coccoid green algae with epi- or endophytic life phases. Later, they were excluded from the family of Chlorococcales based on studies of their life histories in culture, and transferred to their newly described order, Chlorocystidales of the Ulvophyceae. Both genera form a "*Codiolum*"-stage that serves as the unicellular sporophyte in their life cycles. Phylogenetic analyses of SSU and ITS rDNA sequences confirmed that these coccoid taxa belong to the Chlorocystidales, together with the sarcinoid genus *Desmochloris*. The biflagellated coccoid strains were members of the genus *Sykidion*, which represented its own order, Sykidiales, among the Ulvophyceae. Considering these results and the usage of the ITS-2/CBC approach revealed three species of *Desmochloris*, six of *Chlorocystis*, and three of *Sykidion*. Three new species and several new combinations were proposed.

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Abstracts – Poster

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P1: Baltic Sea project: Comparison of benthic protist communities of Baltic and North Sea sampling sites

Merle HEILMANN[1], Maria SACHS1, Julian WAGENHOFER[3], Hartmut ARNDT[1]

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Bacterivorous flagellates living in the sediment of the sea play a major role in marine food webs, especially regarding the carbon flow to higher trophic levels. Their diversity is estimated to be huge, but only a small fraction is discovered yet. Samples were taken from different regions of the Baltic, one of the largest brackish water bodies, and from the North Sea to isolate protists and establish long-term cultures. These were analysed regarding their morphology and molecular identity to compare the different species from the North Sea and the Baltic Sea to extract phylogenetic data of 18S rDNA and calculate their taxonomic position. The distribution of protist species at the different sites of the North Sea and the Baltic Sea will be presented and potentially new species indicated.

P2: How to get dsRNA out of food vacuoles? First characterization of Pds1/2 as components of the dsRNA uptake machinery

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"RNAinterference (RNAi) by feeding" is a mechanism which changes the expression of genes within an organism in dependence to an RNAi-trigger (usually a double-stranded RNA, dsRNA) that is present in the food. This dsRNA is processed into small RNAs by different enzymes of the RNAi-machinery, e.g. Dicer, which then mediate the silencing of a gene by attacking mRNA in a homology dependent manner.

For many organisms, the Sid2 channel is responsible for the uptake of dsRNA from food, making it accessible to the cellular RNAi machinery. It was long thought that organisms lacking this channel are not susceptible to dsRNA from food, as it is thought of e.g. humans.

Contrary to that, *Paramecium tetraurelia* is an organism which is capable of "RNAi by feeding" but does not possess a homolog of Sid-proteins within its genome, which raises the question, how *Paramecium* how dsRNA escapes food vacuoles becoming accessible to the RNAi-machinery.

During a first genome-wide screening, one protein of unknown function, Pds1 (**P**aramecium **ds**RNA-induced RNAi-specific protein 1), was identified to be required for the "RNAi by feeding"-mechanism. Later, a second protein, Pds2 (unpublished), was also found to be necessary for this pathway.

Since the function of all other proteins identified in this screening are known, the hypothesis rises that Pds1 and/or Pds2 might be involved in the uptake of dsRNA from food vacuoles.

With this work, we performed a first characterization of the two proteins, had a look for predicted domains and identified their subcellular localization using GFP-fusion proteins.

P3: A citizen science project about ciliate diversity

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We report a citizen science project started in 2021, which brings together the work of amateur microscopists and academic research. First, we selected specific genera to be collected in nature and to be subjected to morphological, ecological and genetic analysis. The sampling sites include various ephemeral to permanent habitats such as puddles, swamps and ponds. Samples are collected from nature, isolated strains are cultivated, where possible. The morphological details of the ciliates found are documented by light microscopy, including diverse staining techniques and silver impregnation techniques. A new fluorescence double-staining technique using Ho342 and acridine orange was established. The double-staining technique can replace the traditional methyl green-pyronin staining in ciliate research. The technique enables live-cell imaging for longer time using a fluorescence microscope, as it does not harm the cells. Further fluorochromes were tested and applied to document morphology and inner cell organisation of the studied ciliates, e.g. the presence and distribution of phagosomes, acidic granules, acidosomes or mitochondria, and ciliary pattern. Genomic analysis will include short genomic sequence analysis for species determination as well as whole genomic analysis for selected isolated ciliate species.

P4: Baltic Sea project: Comparison of benthic protist communities of Baltic and North Sea sampling sites

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[1] University of Cologne, Zuelpicher Straße 47b, D-50674 Cologne

Bacterivorous flagellates living in the sediment of the sea play a major role in marine food webs, especially regarding the carbon flow to higher trophic levels. Their diversity is estimated to be huge, but only a small fraction is discovered yet. Samples were taken from different regions of the Baltic, one of the largest brackish water bodies, and from the North Sea to isolate protists and establish long-term cultures. These were analysed regarding their morphology and molecular identity to compare the different species from the North Sea and the Baltic Sea to extract phylogenetic data of 18S rDNA and calculate their taxonomic position. The distribution of protist species at the different sites of the North Sea and the Baltic Sea will be presented and potentially new species indicated.

P5: Molecular characterization of *Klossiella muris* (Adeleorina, Coccidia, Apicomplexa) detected during field survey of small mammals from Afghanistan

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Small mammals are an important reservoir for causative agents of numerous infectious diseases, including vector-borne diseases, and might therefore represent a threat for humans and animals. In our study, small mammals collected in military camps in Afghanistan were investigated for the presence of apicomplexans using histopathology and molecular methods. For this purpose, well-established and newly developed real-time PCR assays were applied. A high prevalence of apicomplexans was detected in house mice (*Mus musculus*), but also in shrews (*Crocidura* cf *suaveolens*) and grey dwarf hamsters (*Cricetulus migratorius*). The further molecular characterization based on partial 18S rRNA gene sequences revealed a close relationship to a cluster of *Hepatozoon* sp. detected in voles of the genus *Microtus. Hepatozoon canis* DNA was detected in one house mouse as well as in two *Rhipicephalus* ticks from a dog puppy. In addition, a few house mice were found to be infected with far related adeleorinids showing the highest sequence identity of 91.5% to *Klossiella equi*, the only published *Klossiella* sequence at present. For their better phylogenetic characterization, we did additional metagenomics sequencing of two selected samples. The resulting 18S rRNA gene sequences have a length of 2.4 kb including an insertion of about 0.5 kb and are 100% identical to each other. Histopathology together with organ tropism and detection rates verified this sequence as of *Klossiella muris*. In conclusion, in our study we were able to document naturally occurring protozoan life stages and the additional taxonomic characterization of a well-known commensal in mice.

P6: New amoeboid and heteroflagellate culturable *diversity found in association to the cacti Browningia candelaris and Eulychnia* sp. from the Atacama Desert.

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Microbial habitats in the Atacama Desert can be found in arid and hyperarid soils, many harsh enough to be considered analogous to the surface of the neighbor planet Mars. Here, we present the results of irrigation experiments focused on the isolation of protists in the rhizosphere and phyllosphere of *Browningia candelaris* and *Eulychnia sp.*, two endemic cacti found across this ancient desert. According to the result of phylogenetic analyses, complemented with morphological studies, six new species are presented as part of five cercozoan families (Allapsidae, Sandonidae, Cercomonadidae, Spongomonadidae and Rhogostomidae) and one heterolobose soil amoeba of Acrasidae. Phylogenetic analyses based on the complete 18S rRNA gene allowed the description of one new species of the genus Allovahlkampfia (HFCC979), one new species of testate amoeba (HFCC860) highly similar to Rhogostoma. tahiri (p-distance = 0,36). Additionally, four representatives of different branches of Sarcomonadida were isolated from spines of the cacti and included in this study. Our results extend the knowledge on the habitat ranges of these protists and support the idea of endemic microbial populations inhabiting this extreme environment. Follow-up metabarcoding investigations on these habitats verify the distribution of these and other taxa in this desert and benefit from data bank enlargement due to the new sequences.

P7: Baltic Sea project: Morphological and molecular diversity of estuarine heterotrophic flagellates

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The Baltic Sea is one of the largest estuaries and brackish water bodies globally. This unique ecosystem is exposed to changing biotic and abiotic conditions, often anthropogenic-caused. This includes high nutrient influxes as well as bottom trawling fishery. The DAM MGF-Ostsee project aims to investigate the sediment composition, fauna and biodiversity, microbial food web, and biochemistry in the western Baltic, with a special focus on the benthos. It is carried out in the Natura 2000 marine protected areas (MPA) in the German exclusive economic zone (EEZ) and explores the impacts of bottom trawling fishery on marine ecosystems, especially the effects of the otter boards on the marine benthic community. Within this overall framework, the diversity of benthic heterotrophic flagellates from three distinct cruises in the years 2020 and 2021 was investigated and 32 monoclonal cultures could be established from samples that were taken with a multicorer (MUC) system. Phylogenetic 18S rDNA data revealed at least six potentially new species (pedinellid, several new bodonids and chrysomonads), which originate from sediment depths between 0-20 cm. The isolation of strains from anoxic sediment layers raise further questions on how they ended up in anoxic and micro-oxic environments and which mechanisms allow those strains to survive in these layers. Furthermore, the taxonomic analysis revealed several strains that cluster in clades that are either dominated by marine or freshwater strains as well as genotypes that show a wide biogeography.

P8: Formation pathway and bioaccumulation of intracellular metal-containing particles in ciliates

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Metallic compounds and their toxic ions are increasingly released into our environment due to anthropogenic influences - such as medical diagnostics or novel technology applications. In order to explore new, alternative ways to decontaminate and recover metals, research into how microorganisms deal with metals and metal ions in the environment and the associated detoxification mechanisms is a very promising path. An investigation of ciliates, which exhibit processes of detoxification through intracellular bioaccumulation and excretion of metal-containing particles and therefore are able to actively reduce the concentration of dissolved elements in their immediate environment, are the focus of this research project.

With the aim of investigating possible pathways of heavy metal bioaccumulation, initial treatments of the common model organism ciliate Tetrahymena with soluble heavy metal salts were performed, resulting in the intracellular formation of solid metal-containing particles that are processed intracellularly and subsequently excreted into the surrounding medium. Using (electron-)microscopic (TEM, SEM, LM), spectroscopic (EDS) and analytical methods (ICP-OES), the dynamic process of intracellular particle formation can be followed over time and investigated in detail. The excreted particles can be isolated, which allows characterization of their properties, composition and stability.

P9: The possibility of molecular identification of microorganisms after SEM visualizing

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In microbiological practice, an object of studies is sometimes a small unique sample which cannot be replicated. In such cases it would be highly advantageous to perform both morphological study and subsequent metagenomic or metatranscriptomic analysis on one and the same sample aliquot. The sample conservation, fixation, and fast staining with lanthanoids that provides maximal molecular nativity has been proposed and described earlier (1) as an alternative to “severe” classic protocols of SEM samples preparation. We supposed that even after SEM observations DNA and RNA in the sample might remain sufficiently intact for further molecular analyses.

To test this presumption, we performed the complex study of the model bacterial community. The samples were fixed and stained for SEM with lanthanoids, and after SEM observations the quality of DNA in that the same sample was evaluated by metabarcoding and metagenomic sequencing. The protein spectrum in the sample was evaluated with MALDI-TOF which is also used for taxonomic attribution of bacteria. The results of the taxonomic characterization of the community and the set of proteins present after fixation appeared to be non-deviant from the control.

SEM observations showed that the specific morphological features of the bacterial cells abide after lanthanoid staining and sample storage in dry conditions at room temperature for at least a month. Fixation and all further SEM procedures did not alter significantly the quality of the bacterial DNA, as evidenced by a high coverage and specific GC ratio in the shotgun metagenome sequencing data. Lanthanoid staining did not influence the pool of bacterial proteins detected by MALDI-TOF, also confirming good long-term molecular preservation of the stained samples. Thus, lanthanoid stain provides a fundamental possibility to perform at least two kinds of studies on a single sample: SEM visualization and molecular genetic studies. We suggest that this method can be successfully applied also to unicellular eukaryotes. This may be especially important for transcriptomic studies, because the fixation arrests transcription immediately (2), and further manipulations with cells would not change the expression profile.

1. Chebotar et al., Lanthanoid Staining as a Fast Technology of Preparing Microbiological Specimens for Scanning Electron Microscopy. 2017. https://doi.org/10.17691/stm2017.9.3.03

2. Subbot et al., Life-On-Hold: Lanthanoids Rapidly Induce a Reversible Ametabolic State in Mammalian Cells. 2021. https://doi.org/10.3390/biology10070607

P10: A Long Term Ecological Research project to investigate the Rhine Eco-Evolutionary System (REES)

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What is the impact of changing environmental conditions on freshwater eco-evolutionary processes and how do these effects directly feed back on the community or ecosystem?

REES is a larger collaborative effort of multiple ecological working groups of the University of Cologne. With complementary work and focusing on different research aspects, we aim to generate a holistic perspective on the eco-evolutionary dynamics of the Rhine system.

The project focuses on an area at the Lower Rhine in North Rhine-Westphalia (district Rees), which can be described as landscape of ecological succession including several gravel pit lakes, Rhine oxbows, as well as the main river channel.

This wider system of standing and flowing freshwater bodies offers great opportunities to study dynamic fluctuations in the composition of biodiversity at all levels, from species diversity of communities to genomic diversity at the molecular level of individuals and populations.

Whilst population genetics/genomics as well as quantitative genetics approaches have already elucidated many processes where evolution is shaping ecology, the opposite direction from ecology to evolution, however, has scarcely been investigated empirically and even to a lesser extent using genomic data. Using population genomics approaches, both evolutionary as well as ecological processes can be inferred from genome data (e.g. signatures of selection, genotype-environment associations, demographic history).

Along a selected trophic cascade (from protists to fish), representative species are matter of long-term ecological and population genomic assessment. The incorporation of ecological genomics is the core aspect of this LTER project, which is intended to capture the feedback of evolutionary changes on the ecological system.

This project will be most powerful if many collaborators focusing on different target aspects of the ecosystem join forces to build the overall picture.

P11: Vertical distribution of benthic protists on islands of the Atlantic Ocean using fluorescently stained fixed samples

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Much is already known about the abundance of nano- and microfauna organisms in certain terrestrial and limnic systems, as well as in the sunlit part of the oceans, but what is about benthic protists and bacteria in the deep sea? Only little is known about the distribution of nano- and microfauna organisms from the sublittoral to the deep abyssal. How microbial organisms cope with the extreme conditions in the deep sea, caused by high pressures and the limited food availability are questions of current research and could help to understand the functioning of deep-sea microbial food webs.

The combination of live-counting and counting of fluorescently stained and fixed samples enabled us to investigate the abundance of benthic protist as well as bacteria communities in the sediment from sublittoral to abyssal depths around the Macaronesian islands, especially the Azores and Madeira. The sediment samples from the Azores were collected from different depths (50-3000m) using different sampling gears during an expedition with the research vessel Meteor. Whereas the sampling on Madeira was done in 1000m depth, where different organic substrates, with different C/N ratios (*Sargassum sp., Scomberomorus sp. and Synecchococcus sp.*), were incubated on the deep-sea floor for 16 weeks in order to investigate the influence of different carbon sources on the microbial community in the deep sea.

P12: Resolving the composition of a chromatin remodelling complex participating in *Paramecium* genome rearrangement

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Nuclear dimorphism is a unique feature to ciliates that allows the separation of germ and somatic lineage on a nuclear level. The well-established model organism *Paramecium tetraurelia* harbors two diploid and transcriptionally silent micronuclei (MICs), the germline nuclei, and one highly polyploid and transcriptionally active macronucleus (MAC), the somatic nucleus. Each sexual division the MAC is lost and a new MAC arises from a copy of the MIC. This process requires massive genome rearrangement including genome amplification, chromosome fragmentation and excision of germline-specific sequences. Over 45,000 unique Internally Eliminated Sequences (IESs) are located throughout non-coding and coding regions, necessitating precise excision to guarantee a functional new MAC genome. Though several key players involved in IES elimination have already been identified, many steps in this process remain poorly understood. A chromatin remodeller has been shown to assist the excision of a subset of IESs. Here, two complex partners of this remodeller were identified that participate in IES excision. Their contribution to the complex was verified by localization, co-immunoprecipitation experiments and knockdown studies. Elucidating the composition of this complex will further our understanding of how chromatin remodelling contributes to IES excision.

P13: A metatranscriptomic approach to study the effect of multiple stressors on the microbial community involved in CPOM degradation in freshwater habitats

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Coarse particulate organic matter degradation (CPOM) in streams largely depends on microbes i.e fungi and bacteria however, the effect of environmental factors like temperature and salinity on metabolic pathways and enzymatic reactions of these degraders are undetermined. We aim at elucidating the role of different fungal and bacterial groups and their particular role in the enzymatic decomposition of CPOM in Emscher/Boye and Kinzig catchments, with and without stressors. We are using DNA stable isotope probing (DNA-SIP), amplicon and metatrancriptomic sequencing to study the taxonomical and functional diversity involved in leaf litter degradation. In order to determine the active taxa involved in degradation we have used 13C labelled Alder leaves. DNA of the active degraders of 13C leaves will be extracted by DNA-SIP. These targeted organisms will be further analyzed for metabolic pathways and enzymatic reactions. To analyze the metatranscriptomic sequences we built a pipeline for preprocessing the eukaryotic and prokaryotic mRNA and to map it to databases like Mycocosm, CAZy and NCBI for taxonomic and functional information. While studying the effect of multiple stressors i.e. temperature, salinity, and flow velocity we are testing the hypothesis that function recovers faster than community due to functional redundancy. According to our hypothesis we expect that fungi and bacteria share CPOM degrading functions and thus after the release of stressors, function will recover faster than the microbial community composition.

P14: Potential and advantages of cryo-FIB-SEM 3D reconstruction on the biomineralization in *Coleps hirtus*

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Recording 3D data of high-pressure frozen samples through cryo-FIB-SEM allows for the reconstruction of different cell features with a high degree of detail and unprecedented flexibility in its analysis and presentation. Here we used the advantages of this method to improve our understanding of the biomineralization process of alveolar plates in the ciliate *Coleps hirtus* and the role that different types of vesicles play in it. The reconstruction offers a detailed three-dimensional look at the spatial distribution of the biomineralized alveolar plates and the different compartments of the cell, including distinctly different types of vesicles from variable viewpoints.

P15: Resilience of ciliate assemblages facing climate warming in Lake Zurich

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In recent years, climate-induced warming of surface water and associated increased density differences to deep water caused a reduced mixing depth in Lake Zurich. This resulted in a reduced transport of nutrients from the near bottom to the euphotic surface zone. The following nutrient depletion negatively affected the development of vernal phytoplankton blooms and seemingly their main predators, ciliates. In our study, we compared data from two high-frequency spring sampling campaigns in Lake Zurich from 2014 and 2019. Besides environmental parameters, ciliates were assessed qualitatively and quantitatively using Protargol staining to examine whether altered abiotic conditions had an influence on their assemblage composition and abundance. The two years varied markedly in terms of physical conditions, having a cascading effect down to the investigated organisms. In 2014, a low mixing depth resulted in minor epilimnetic nutrient (orthophosphate) concentrations and consequently, no pronounced phytoplankton bloom was observed. In contrast, two mixis events took place in 2019, resulting in increasing surface concentrations of orthophosphate and in the development of two distinct algal blooms. The comparison between the two years showed differences in dynamics of total ciliate numbers. A hierarchical cluster analysis of ciliate species composition revealed a split into early and late spring bloom assemblages in both years. This shift coincided with a decline in ciliate abundances and went in parallel with a rapid increase of water temperature. Although conditions were quite different in the two years, a core group of ciliate species showed equal dynamics, indicating certain resilience to environmental changes.

P16: Plant diversity and drought effects on community composition of Cercozoa and Oomycota

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Due to climate change, European grasslands are increasingly affected by heavy droughts during summer. Drought is known to negatively affect ecosystem functioning. On the other hand, plant biodiversity is associated with increased ecosystem functioning.

The “Jena Experiment” is a long-term field experiment consisting of experimental plots with varying plant communities ranging from 1 to 60 species. In 2008, a sub-experiment was established that emulated repeated summer drought. On all plots, transparent roofs were installed for six weeks in summer from 2008 to 2016. Soil was sampled one year after the last drought period. We investigated by amplicon sequencing how plant species richness and drought treatment influenced the diversity of two protistan taxa, Cercozoa (Rhizaria) and Oomycota (Stramenopila), and whether increased plant diversity would lead to resistance against the drought effects. These two protistan groups are functionally diverse and include a wide range of plant parasites.

Alpha diversity indices of Cercozoa decreased with increasing plant species richness and increased in the drought treatment. Diversity indices of Oomycota also decreased with increasing plant species richness but were unaffected by drought. These changes were mostly explained by changes in evenness. Both protistan community compositions were strongly influenced by plant species richness, but only the cercozoan community was significantly affected by drought. Overall, no buffering effect of plant diversity against drought was observed.

Our results indicate that ecosystems will be increasingly challenged by plant pathogens due to more frequent summer droughts and biodiversity loss. Economically used areas, especially agricultural monocultures, may be particularly affected.

P17: How climate induced changes in water turnover affect microbial food web dynamics in Lake Zurich during spring

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Complete water turnover (holomixis) is an important process in many deep lakes such as Lake Zurich, enabling redistribution of nutrients and oxygen throughout the water column. Typically occurring in early spring, it promotes characteristic phytoplankton blooms and reinvigorates the microbial food web. These so-called spring blooms represent the first peak of primary production and set the course for annual food web succession.
Several studies suggested that lake warming, mainly caused by rising air temperatures, stabilizes thermal stratification of the water column and thus hinders proper holomixis. Such partial mixis events impede upwelling of nutrient- rich deep water. In the case of Lake Zurich, partial mixis may have several pronounced effects, such as facilitation of harmful cyanobacteria blooms and further reduction of primary production in the already oligo-mesotrophic lake. Investigating varying mixis events and their consequences build the foundation for the assessment of microbial food webs and furthers our understanding of lake ecosystems.

In this study, we examined the impacts of different mixis events on environmental parameters and important members of the microbial food web (bacteria, hetero- and autotrophic protists) during spring 2020, 2021 and 2022. The three years differed strikingly in mixis depth, climate conditions and nutrient availability. This had a strong effect on the microbial spring bloom community. Through highlighting key differences and similarities between these years, we showcase the consequences of climate induced changes in mixis depths and pave the way for future studies.

P18: Phylogeny and taxonomy of new genus for Radiococcaceae (Sphaeropleales, Chlorophyceae) based on morphology and 18S rDNA phylogeny.

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The use of molecular tools is disclosing the phylogeny of coccoid green algae members and unveiling new genera and species. Here, one coccoid green microalgae was studied and led to the description of a new genus for Radiococcaceae, isolated from a reservoir in São Paulo State, Brazil. The strain was characterized using a combination of morphotaxonomic and 18S rDNA data, revealing green spherical cells, planktonic, with autosporic reproduction (2-4 autospores per sporangium), cup-shaped chloroplast and a mucilaginous envelope. The 18S rDNA phylogenetic analyses, employing all the published sequences for the related genera and species, showed that the new taxon is a sister clade of *Pharao desertorum* Saber et al., also a genus recently described for Radiococcaeae. The 18S rDNA support, both for Bayesian Posterior Probability as for Maximum-Likelihood bootstrap, the placement of this new lineage in Radiococcaceae. Thus, this study contributes to clarify the phylogeny and taxonomy of this family of green microalgae.

P19: Ecophysiological resilience to temperature and salt stress in the mixotrophic genus Dinobryon (Chrysophyceae, Stramenopiles)

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Chrysophytes are among the most important mixotrophic organisms in aquatic systems. They are crucial primary producers and, at the same time, among the most important grazers of bacteria-sized microorganisms. Members of the mixotrophic genus Dinobryon are widespread in freshwaters and occur under different environmental conditions. This study investigated the ecophysiology of several isolates with distinct geographical origins (e.g. alpine isolates and lowland isolates). Especially the effects of varying temperatures were tested in a series of laboratory experiments in combination with other stressors (e.g. varying NaCl concentrations).

The results showed that temperatures of 15°C to 19°C led to positive growth rates for all tested strains. Higher temperatures like 23°C and 27°C led to mortality in all strains except the alpine isolates of Dinobryon divergens which grew even under 23°C. Under optimal temperature conditions, three strains still showed positive growth up to a chloride concentration of 3 g/l but the combination of temperature stress with different salt concentrations led to a higher mortality rate in most strains.

P20: Integrative approach on key freshwater ciliates of the genus *Urotricha* (Alveolata, Ciliophora, Prostomatida).

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Species of the ciliate genus *Urotricha* are key players in freshwater plankton communities. In the pelagial of lakes, about 20 urotrich species occur throughout an annual cycle, some of which play a pivotal role in aquatic food webs. In ecological studies, urotrich ciliates are usually merely identified to genus rank and grouped into size classes. This is unsatisfying considering the distinct autecological properties of individual species and their specific spatial and temporal distribution patterns. As a basis for future research, we characterized in detail several common urotrich morphotypes using state-of-the-art methods. We applied an integrative approach, in which morphological studies (in vivo observation, silver staining methods, scanning electron microscopy) were linked with a molecular approach (SSU rDNA, ITS-1, ITS-2, hypervariable V4 and V9 regions of the SSU rDNA). We shed light on the diversity of urotrich ciliates as well as on their global distribution patterns, and annual cycles. Additionally, we coupled individual species occurrences and environmental parameters, and subsequently modeled the distribution and occurrence, using logistic regressions. Furthermore, for one strain, we ascertained the optimal cultivation media and food preferences. Thereby, our comprehensive view on these important freshwater ciliates that frequently occur in environmental high throughput sequencing datasets worldwide will allow future studies to better exploit protistan plankton data from lakes.

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