

CICHLID SCIENCE 2019

Abstract Book



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Oral presentations

PLENARY LECTURE

Cavefish evolution, from early development to behavior

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We use *Astyanax mexicanus* cavefish to understand developmental, genetic and cellular mechanisms involved in brain and behavioral evolution. Within the *A. mexicanus* species, there are populations of river-dwelling surface fish (“normal fish”) and populations of blind cavefish inhabiting the permanent darkness of caves. Cavefish have undergone some striking losses (eyes, pigmentation) but have also undergone some probably adaptive gains, such as more taste buds, more neuromasts, larger jaws, more teeth, modified hypothalamus and larger olfactory structures. Their physiology and behavior is also very different from their surface conspecifics.

We use a comparative approach including developmental biology (evo-devo), molecular evolution studies, and behavioral analyses to understand the evolutionary forces at work during cavefish brain evolution and its adaptation to cave life.

I will present our work on the evolution of sensory systems in cavefish, focusing on the visual-olfactory trade-off.

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T1.1

Molecular basis of convergent evolution between cichlid fishes of lakes Malawi and Tanganyika

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The great lakes of East Africa each harbour hundreds of species of cichlid fish generated by adaptive radiations with well-established separate origins. Still, some species, like *Petrotilapia* of Lake Malawi and *Petrochromis* of Lake Tanganyika, “resemble each other in an almost uncanny manner, not only in general appearance but in minute details of their anatomy.” (Fryer and Iles, 1972). They offer some of the most iconic and striking examples of convergent and parallel evolution. To investigate the molecular mechanisms underlying convergence between Malawi and Tanganyika cichlids, we collected whole genome DNA data from 20 species of Lake Tanganyika, sequencing 20 individuals from each species. Combining this with a dataset of 725 Lake Malawi cichlids, I search for signatures of selection that distinguish particular species within their lakes and investigate whether the signatures point to selection on the same genes or genomic regions in pairs of species with convergent phenotypes. I am going to present preliminary results of these analyses, together with a discussion of technical challenges concerning combining population genomic datasets across lakes and across different timescales. In addition I am going to present more general insights into allele distribution and allele sharing between the lakes.

T1.2

Molecular mechanisms of color pattern formation and evolution in cichlid fishes

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Coloration phenotypes play many roles in cichlid evolution, covering the entire spectrum of functions including those relevant for sexual selection (e.g. mating preferences) and adaptation (e.g. camouflage). Likewise, cichlid fishes are a famous example for diversification and repeated evolution. Many of their color patterns as vertical bar and horizontal stripe patterns evolved several times and in parallel across the over 1200 species of East African cichlids. Still, the genetic and mechanistic basis determining these traits and what changes cause their diversification and repeated evolution are largely unknown. Here we integrate genomic analyses, genetic mapping, molecular analyses including genome editing, and phylogenetic approaches to uncover the genomic and mechanistic underpinnings of color patterns, and their astonishing evolutionary dynamics.

T1.3

Newly discovered tilapia cichlid fish biodiversity threatened by hybridization with non-native species

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Non-native species introduced into freshwater systems can readily hybridize with indigenous congeneric species. In tilapiine cichlids this hybridization can lead to loss of unique genetic resources, and it is even possible that this irreplaceable biodiversity is being compromised even before its value is recognised. Here we show that newly discovered (2013-2016) evolutionarily significant populations of *Oreochromis korogwe* from southern Tanzania are threatened by hybridization with the larger invasive Nile tilapia (*Oreochromis niloticus*). Using microsatellite DNA we found evidence for *O. korogwe* x *O. niloticus* hybrids in three lakes (Nambawala, Rutamba and Mitupa), and a combination of microsatellite and morphological evidence was consistent with the presence of hybrid swarms. We next show using microsatellite DNA and morphological methods that purebred *O. korogwe* from the southern lakes differ substantially from those in the geographically disjunct populations from northern Tanzania (Zigi River and Mlingano Dam), demonstrating that these southern populations are unique. Finally, we show using whole genome data that the northern and southern *O. korogwe* populations diverged some 370-600 KYA, suggesting that the geographically separated populations are not a result of recent introductions, and instead represent independent evolutionarily significant units. We conclude that newly-discovered phenotypically unique populations of a tilapiine cichlid are already threatened by hybridization with an invasive species, and propose these irreplaceable genetic resources would now benefit from conservation interventions.

T1.4

Behaviour and neural gene expression predict patterns of asymmetric hybridisation in a mouthbrooding cichlid genus (*Ophthalmotilapia*) from Lake Tanganyika

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Cichlid radiations harbour several closely-related species with overlapping niches and distribution ranges. The fact that these species sometimes hybridize in nature raises the question how they coexist without merging into one hybrid species. Earlier studies revealed several instances of introgression among species of the Tanganyika mouthbrooder *Ophthalmotilapia*, although specimens with hybrid morphologies are almost absent in the wild. Additionally, these studies suggested that *O. ventralis* females could be less reluctant to mate with *O. nasuta* males, than the other way around. Therefore, we studied female mate choice in sympatric populations of *O. nasuta* and *O. ventralis*. We staged two sets of experiments to investigate the genetic basis of mate choice behaviour in *Ophthalmotilapia*: (i) females were visually exposed to a conspecific or a heterospecific male, and (ii) physical encounters between conspecific and heterospecific couples were monitored until females picked up fertilized eggs. We made a detailed qualitative and quantitative description of the fishes' behaviour and measured gene expression in five brain parts of 55 female fish. The first experiment revealed that *O. nasuta* females behaved differently when exposed to a con- or heterospecific males. In agreement with our hypothesis, we did not observe this species-dependent behavioural response in *O. ventralis* females. The second experiment only yielded heterospecific matings between *O. ventralis* females and *O. nasuta* males. Remarkably, *O. nasuta* females exposed to a heterospecific male and *O. ventralis* females that mated with a heterospecific male, both exhibited a set of differentially expressed genes linked with hormone production in the diencephalon.

T1.5

Parasite transfer in the Congo Basin: what effect does introduced *Oreochromis niloticus* have on the parasite community of native cichlids?

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Oreochromis niloticus, Nile tilapia, is one of the most popular aquaculture fish species worldwide. This species was introduced into almost every tropical region in the world, including the Congo basin. However, whether its parasites were co-introduced, and if so, whether they also infect local fish species, has hardly been investigated in Africa. We aim to trace parasite transfer through morphological and genetic parasite identification. For this, we compare monogenean community data of native cichlids from recent expeditions from three main parts of the Congo Basin (Upper, Middle and Lower Congo) to those from museum fish specimens from the same areas, pre-dating the introduction of Nile tilapia. Parasites of choice are gill-infecting monogeneans belonging to Dactylogyridae and Gyrodactylidae. These are ideal model parasites, because they have a direct lifecycle, are prevalent, generally host-specific and species-rich, resulting in each species of cichlid generally being infected by a unique community of parasites. We report that nine parasite species were co-introduced with *O. niloticus*, while two others, *Cichlidogyrus rognoni* and *Gyrodactylus cichlidarum* are missing and are possibly not established. We report two putative spillover events, of *Cichlidogyrus sclerosus* and *Cichlidogyrus tilapiae*, to the native *Coptodon tholloni* in the Lower Congo Basin. Additionally, we report the putative spillback of *Cichlidogyrus berradae* and *Cichlidogyrus cubitus* from the native *Coptodon tholloni* to the introduced *Coptodon rendalli* in Lower Congo and the spillback of *Cichlidogyrus flexicolpos* from possibly *Coptodon guineensis* to the introduced *Coptodon rendalli* in Lower Congo.

T1.6

Genetic basis of a key trait for sympatric speciation in cichlids

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One hallmark of the East African cichlid radiations is the rapid evolution of reproductive isolation that is robust to full sympatry of many closely related species. Theory predicts that speciation in sympatry and species persistence in sympatry (with gene flow) are more likely if loci of large effects underlie traits involved in behavioural reproductive isolation. Here we investigate the genetic architecture of a key mating trait, male nuptial coloration, in two pairs of sister species of Lake Victoria cichlid fish of the genus *Pundamilia*. One is a young sympatric species pair, representative of a red-back vs blue male colour polymorphism that is associated with many sympatric species pairs at all stages of genetic differentiation in several genera of Lake Victoria cichlids. The other is a species pair representative of a red-chest vs blue male colour polymorphism that is typically associated with divergence into allopatric sister species of Lake Victoria cichlids, and that is rarely seen in sympatry. We use a QTL mapping approach to map the presence/absence of red in 170 second generation hybrid (F2) males of a cross between the two sympatric species, and 140 F2 males of a cross between the two allopatric species. Our results are indicative of several highly significant QTLs for red colour in the sympatric pair, whereas we find none in the allopatric pair. These findings are consistent with the theoretical prediction that the presence of large effect loci in mating trait differentiation could facilitate speciation and species persistence in sympatry (with gene flow).

PLENARY LECTURE

How do fish see in the dark and what do deep-sea fish and deep-water cichlids have in common?

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Deep sea or the deep freshwater lakes can be challenging environments, especially for the species living in the depth. Extremely low light intensity and narrowed colour spectrum triggered emergence of numerous adaptations in the visual system of deep-water fishes. Recent advances in genomics and transcriptomics enable us to focus on the molecular mechanisms of such adaptations. In dim-light, vertebrates generally rely upon a single rod opsin (RH1) for obtaining visual information. By inspecting over 100 fish genomes, we found that several deep-sea teleost lineages have independently expanded their RH1 gene repertoires. The extreme example among the deep-sea fishes stands out as having the highest number of visual opsins in vertebrates (2 cone and 38 rod opsins). Whether the unique rod-based visual system of the deep-sea fishes is useful for colour vision related to bioluminescence, or rather to boost sensitivity within the entire light spectrum in the depth remains elusive. Several cichlid species are also specialists inhabiting the deep-water habitats. In the deep crater lake Barombi Mbo in Cameroon (West Africa), two species have evolved to live in the depth, even below oxycline. We examine visual system evolution in this small-scale yet phenotypically and ecologically diverse adaptive radiation, and we show that rapid adaptations of the visual system to the novel deep-water habitat primarily occurred at the level of gene expression changes rather than through nucleotide mutations, which is compatible with the young age of the radiation. In my talk I will focus on similar (but different) adaptations to life in the depth, despite of the phylogenetic distance between cichlids and many deep-sea lineages.

T2.1

Dynamics of sex chromosome evolution in an adaptive radiation

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Sexual reproduction is ancient and almost universal in the eukaryotic kingdom. However, the mechanisms that actually determine male or female sex are highly diverse encompassing a variety of extrinsic and intrinsic drivers. Intrinsic factors, commonly referred to as genetic sex determination, comprise systems as simple as a single base pair difference between the sexes to highly differentiated sex chromosomes. Why sex-determining systems change frequently in some taxa, whereas the same sex chromosome pair persists over long evolutionary time in others, remains an open question. With ~3000 species, cichlid fishes are one of the largest vertebrate families and an extraordinary example of adaptive radiations. If different sex-determining mechanisms contribute to the species richness of this fish lineage is largely unknown. The oldest and most diverse African cichlid assemblage is the one of Lake Tanganyika, which consists of 250 endemic species. Using whole genome sequencing, comparative genomics and expression data, we investigate the genetic basis of sex determination and the evolution of sex chromosomes in this radiation. We identified at least 27 sex chromosome transitions in cichlids with a transition rate of 0.19 per million years. To the best of our knowledge, this is the highest transition rate described so far. We show that sister species can have different sex chromosomal systems indicative of very recent turnover events. On the other hand, we also found cases of convergently evolved sex chromosomes in distantly related cichlid lineages. Chromosomal fusions likely played a role in cichlid sex chromosome evolution. We compare patterns of transitions in sex chromosomes but also heterogamety between cichlids and other ray-finned fishes. Cichlids perfectly exemplify the diversity as well as the constraints of sex chromosome evolution and can reveal the forces driving the evolution of sex-determining systems. Sex chromosomes appear to be one of the ultra-fast evolving traits in this astonishing group of fishes.

T2.2

Ancient DNA and phenotypic data from lake sediment-embedded fish subfossils as tools to study cichlid evolution

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Many evolutionary processes unfold over time periods longer than we can observe directly, with environmental conditions and ecological settings constantly changing. Because of this, inferring the causes and consequences of past diversification, extinction and migration from data of extant populations alone is often difficult. In East African cichlid fishes, where the extant diversity and phylogenetic relationships of adaptive radiations are known, the conditions at the beginning and during these radiations are much less studied. Abundance of competitors or prey, availability of habitats, or extinction events due to environmental change are just some examples of important factors influencing diversification where paleontological data are needed. Such data can be gathered from sediment records through phenotypic analyses and sequencing of ancient DNA from fish fossils. Here, I will present findings from studying sediment records of several East African lakes and highlight the potential and challenges this approach holds for the study of cichlid fish biodiversity and evolution.

T2.3

Bridging the knowledge gap: The challenges of characterizing parasite host specificity in a cichlid model system through a meta-analysis of published literature

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Parasitic species are estimated to account for at least half of all species on the planet. Therefore, understanding parasite ecology and evolution is key to improve knowledge of speciation and ecosystem processes. Host specificity is a defining characteristic of parasite ecology as it describes the ecological niche of parasite species. However, the underlying mechanisms that determine the host range of these organisms are poorly understood. Monogenean gill parasites from cichlid fish belonging to the genera *Cichlidogyrus* and *Scutogyrus* (Monogenea, Dactylogyridae) have been proposed as suitable model species for research on parasite evolution because of the well-known ecology and diversity of the cichlid hosts and a single-host lifecycle. Based on an extensive meta-analysis of previously-published literature and additional data collected for this study, we highlight challenges faced when analysing biased and incomplete data sets, and recommend procedures to improve on data collection and analysis for a host-parasite model system. Furthermore, we summarise the current state of knowledge and propose a standardised method to quantify and analyse host specificity data, identify generalist and specialist parasite species, and find potential predictors of host specificity using phylogenetic, ecological, and morphological data.

T2.4

Cameroon crater lake cichlids: searching for sympatric speciation in the genomic era

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Sympatric speciation illustrates how natural and sexual selection may create new species in isolation without geographic barriers. However, recent genomic reanalyses of classic examples of sympatric speciation reveal complex histories of secondary gene flow from outgroups into the radiation. I will present a detailed genomic, ecological, and behavioral investigation of the speciation process in sympatric radiations of cichlids in Lake Ejagham and Barombi Mbo, Cameroon. We infer that Lake Ejagham was colonized by *Coptodon* cichlids soon after its formation 9,000 years ago, yet speciation occurred only in the last 1,000–2,000 years. We show that secondary gene flow from a nearby riverine species has been ongoing, into ancestral as well as extant lineages, and we identify a cluster of olfactory receptor genes that introgressed near the time of the first speciation event, coinciding with a higher overall rate of admixture. In Barombi Mbo, the situation is more complex and it is difficult to point to a clear sign of adaptive introgression despite ongoing riverine gene flow into all eleven species. We conclude that sympatric divergence occurred in both radiations, but was potentially triggered by adaptive introgression in Ejagham *Coptodon* and this scenario is also difficult to rule out in Barombi Mbo.

T2.5

Genomic changes involving olfactory receptor genes across distinct evolutionary scales

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The sensory abilities of living organisms determine many of their basic behavioural responses. Although vision seems to be at the top of primary senses in most fish, olfaction has been also related to vital activities such as mating, feeding or avoidance of predators. Thus, olfactory perception is a complex trait that might evolve fast as phylogenetic and ecological diversification of a given lineage occurs. At the molecular level, odour detection largely depends on a repertoire of G-protein-coupled receptors expressed in the olfactory epithelium that recognize specific chemical signals in the water. Previous comparative studies in vertebrates revealed great variation in the number of olfactory genes among species reflecting diverse functional capabilities for this sense. However, many open questions remain regarding the macroevolutionary dynamics of olfactory perception as well as its possible role in adaptive divergence during speciation episodes. We investigate the type of genomic changes involving olfactory receptor genes on two distinct phylogenetic scales: the teleost clade and a sympatric radiation of cichlid species in a small crater lake of Cameroon. As the olfactory receptor genes are mostly located within genomic clusters, we focused preferentially on high-quality teleost genome assemblies retrieved from public databases. In addition, we also applied Nanopore sequencing in a few cichlid species. We identified several species of teleosts with particularly enlarged and reduced numbers of olfactory receptors, supporting a birth-and-death model that could be linked to their ecology.

T2.6

Gene expression evolution in Lake Tanganyika cichlid fishes: Novel insights through data integration

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Adaptive radiation is the rapid proliferation of eco-morphological diversity within an organismal lineage on the basis of rapid adaptation to distinct ecological niches and an important evolutionary process shaping the diversity of life. One striking example of an adaptive radiation is found in Lake Tanganyika, where hundreds of species of cichlid fishes have evolved in less than 12 million years. And yet, it is not fully understood how intrinsic factors and external events interplay to promote this exceptional diversity. Gene expression regulation is a prime candidate factor and thought to be responsible for many phenotypic differences observed among the closely related cichlid species. To investigate the genomic landscape of speciation and the transcriptional diversity of Tanganyikan cichlids, we conducted a comprehensive analysis of gene expression evolution. We sequenced the transcriptomes of six tissues from 76 representative cichlid species. By comparing gene expression patterns across the Tanganyikan cichlid radiation, we demonstrate that the rate of gene expression evolution varies among organs but also among the different cichlid lineages, indicating differences in selective pressures. By combining our expression data with existing genomic, morphometric and ecological data, we also identify candidate genes that might underlie key features of the cichlid evolution such as the lower pharyngeal jaw bone. Our study is the so far most thorough examination of the transcriptomal diversity of any adaptive radiation. Taken together, our results provide a unique opportunity to gain insights into cichlid speciation and extend the current knowledge of how selection acts on an entire biological system.

T2.7

The cichlids of the Lake Edward system: diversity and ecology

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Haplochromine cichlids are well known for their extraordinary diversity and their ability to form adaptive radiations. They radiated in many of the East African Great Lakes, giving rise to a.o. the Lake Victoria region superflock, which includes the haplochromines from Lakes Albert, Edward, George, Kivu, and Victoria. The extraordinary species richness coupled with subtle interspecific morphological differences resulted in many East African cichlid species remaining undescribed. This especially holds for those from Lakes Edward and George of which only 32 of the expected 60–100 species have been formally described. We perform a systematic revision of these *Haplochromis* species. As a first step, specimens are grouped based on morphological characteristics that suggest a similar trophic ecology. For each group, a morphometric study is carried out and species are delineated and (re)described. Stable isotope analyses are combined with gut content observations to deduce the trophic positions of each species. Hitherto, we discovered a species richness of 26 species within six trophic groups. Fourteen of which have remained undescribed including three species of oral snail shellers with different oral and pharyngeal dentition and lower jaw shapes, seven species of piscivores with different head morphologies and colour patterns, and four species of generalists with different tooth morphologies. The preliminary results of our stable isotope analyses reveal clear ecological differences between species. Unexpected was the high trophic level of the supposedly insectivorous *H. eduardianus*. This contradiction will be further investigated by means of stomach content observations.

PLENARY LECTURE

The parasite fauna of Middle American cichlids: a case of extreme hidden biodiversity?

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Middle America holds a very diverse cichlid fauna. Cichlids are represented in Middle America with c. 124 species included in one lineage, the heroine cichlids, excepting by two species found in Panamá and Costa Rica. Two areas, the Usumacinta and San Juan hydrological systems are hyper-diverse in cichlids. Their parasite fauna is also diverse, particularly helminths. Middle American cichlids have been studied for helminth to a certain extent. Thus far, at least 120 species of helminths have been recorded as parasites of cichlids in this geographical region. The use of DNA in the description and characterization of the helminth parasite fauna revealed unusual levels of cryptic diversity. The overall diversity patterns of the helminth parasite fauna of Middle American cichlids are explored, and examples of hyper-diverse groups are documented. This information is very useful because parasites have been documented as drivers of the diversification of their hosts; the proper taxonomic identification of the helminth parasite fauna will be very useful for further understanding cases of cospeciation among hosts and parasites.

T3.1

Parasite infection and immunogenetic adaptation in a cichlid radiation

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In the last two decades, it became clear that evolutionary divergence has a strong ecological component. How ecological factors contribute to evolutionary divergence at historical or contemporary time scales is therefore a key question in evolutionary biology. Parasites represent a strong ecological pressure, which is predominant in all animals. Since parasites influence survival and reproduction, they can also influence adaptation, reproductive isolation and ultimately perhaps even speciation of their host. Whether or not parasites induce local immunogenetic adaptation and hence divergence of their host across landscape, depends on how parasite communities respond to environmental and biogeographic variation, and the strength of host-parasite co-evolution. Interestingly, adaptive divergence causing shifts in diet, habitat use or behaviour might directly relate to shifts in parasite infection. We discuss whether this ecological differentiation significantly contributed to the morphological and genetic diversity found in Lake Tanganyika cichlids today.

T3.2

Parasite contribution to host divergence: insights from African cichlid fish and their flatworms

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Parasites may have strong eco-evolutionary interactions with their hosts. Consequently, they may contribute to host diversification. The radiation of cichlid fish in Lake Victoria provides a good model to study the role of parasites in the early stages of speciation. We analysed the macroparasite fauna of a community of 17 sympatric cichlids, and a species pair from multiple localities, representing different levels of genetic differentiation. Parasite infection profiles were different among host species with (nearly) complete reproductive isolation. This suggests that differences in parasite infection accumulate after the initiation of host differentiation. Infection differences were similar between 2 sampling years, indicating that parasite-mediated selection is stable through time. We separately analysed the community composition of *Cichlidogyrus* morphospecies (Monogenea) infecting host species. *Cichlidogyrus* is considered a good candidate for driving parasite-mediated speciation, because of its high species richness and host specificity. We found that the species composition of *Cichlidogyrus* infection was similar among the most closely related host species (members of the Lake Victoria radiation), but two more distantly related species (belonging to non-radiating sister lineages) showed distinct infection profiles. This is inconsistent with a role for *Cichlidogyrus* in the early stages of divergence. To conclude, we find significant interspecific variation in parasite infection profiles, consistent with parasite-mediated divergent selection. However, we find no evidence that parasites contribute to the initiation of speciation.

T3.3

Diversity of the gut microbiota in African and Central American cichlid fish radiations

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Cichlid fishes, with their repeated colonization of lakes and subsequent radiations at different scales of phylogenetic and ecological diversification, offer an excellent model system to understand the factors shaping the host-gut microbiota association in nature. Here, we characterized the gut microbiota of the *Amphilophus* species complex from Central America (known as the Midas cichlid complex), encompassing 161 wild specimens (13 species) collected from seven Nicaraguan lakes, and combined these data with previously published data from two African lakes (spanning 29 species). Our aim was to comprehensively explore trends in microbiota variation and persistence along the large spatial and temporal scales of cichlid diversification (from the oldest radiation in Lake Tanganyika, 9-12 My old, to young ones in Nicaraguan crater lakes, < 0.5 My old), in allopatry and sympatry (within and across lakes), and across the range of dietary niches (from highly specialized to generalist feeders). Despite their extraordinary diversity, cichlids shared a remarkably conserved microbial taxonomic profile, which argues for a primary role of the host genetics in the assembly and maintenance of these microbial communities. Within this partly constrained microbiota profile, geographic isolation (continent and lake) represented the first level of discrimination. For the Midas cichlid, a partial congruency was found between host microbiota and genetic distances, suggesting that microbial communities have partly diversified along their cichlid phylogeographic history of crater lake colonization. In sympatry (within lakes), the young and poorly ecologically diversified cichlid assemblages of Central American lakes display largely unresolved gut microbiotas (in terms of both alpha and beta diversities), whereas the phylogenetically and ecologically diverse species found in African lakes showed greater microbial interspecific diversity. This pattern largely points to the level of habitat segregation, trophic niche overlap, and reproductive barriers as major modulators of the gut microbiota connectivity among sympatric species.

T3.4

The transcriptomics of parallel trophic adaptation

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The parallel evolution of phenotypes in replicate radiations has fascinated biologists for decades. One such phenotype are the oral and pharyngeal jaws in cichlid fishes that have repeatedly adapted to similar trophic niches in different lakes. The striking trophic diversity in cichlids has been associated with the exploitation of novel resources post-speciation. Using 200 whole-transcriptomes of these two sets of jaws from species adapted to various herbivorous and carnivorous diets, we aim to investigate if the adaptive radiations of cichlid fishes in Lake Tanganyika, Malawi, and Victoria evolved parallel jaw morphologies in response to similar selection pressures via parallel gene regulatory pathways or not. Using riverine outgroups, we will also investigate if this parallelism can be explained by ancestral plasticity in the 'flexible stem' that seeded these sister radiations, which results in the same deterministic outcome to the same problems. Additionally, we will build upon our previous findings and dissect the contribution of gene expression versus alternative splicing as salient sources of rapid diversification during adaptive radiation. Here we present the first findings from this largest cichlid jaw transcriptomic dataset to date.

T3.5

Developmental transcriptomics of jaw morphology in Lake Victoria cichlids

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The cichlid species flock of Lake Victoria is comprised of ~500 species and represents the youngest adaptive radiation compared to those in Lake Malawi and Tanganyika, with a maximum age of ~100,000-400,000 years. Even at such a short evolutionary timescale, they evolved an astounding array of trophic diversity in their oral and pharyngeal jaws. Here we investigate the underlying differences in gene expression between these two sets of jaws in species adapted to herbivorous and carnivorous trophic niches by comparing two key life stages: (1) the postlarval stage, which is marked by the completion of yolk absorption and subsequently the beginning of independent foraging and (2) the onset of adulthood, marked by changes in body coloration and breeding behavior. By comparing these stages, we want to investigate whether or not the observed gene regulatory patterns at the end of larval development are already established and stable or if significant changes towards adulthood can be seen. Building on a previous study from our lab (Singh et al. 2017), which suggested alternative splicing and differential gene expression as main drivers of adaptation in trophic morphology in six Lake Tanganyika species of the tribe Tropheini at the end of postlarval development, we compare eco-morphological divergence in connection to ecological speciation between the three African Great Lakes. This will also enable us to investigate the routes of parallel evolution of similar eco-morphologies across the lakes. Here I will present preliminary results on the developmental transcriptional trajectories of postlarvae and adults of Lake Victoria cichlids.

T3.6

Disparity and the temporal dynamics of eco-morphological adaptation in the cichlid radiation of Lake Tanganyika

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The assemblage of cichlid fishes from East African Lake Tanganyika consists of about 200 described and approximately 40 un-described species and shows an extraordinary degree of morphological and ecological diversity. In an integrative study we investigated multivariate morphological measurements in three ecologically relevant trait complexes, and approximations of the ecology of virtually all of the ~240 species of the endemic cichlid fauna of Lake Tanganyika. We show how the different cichlid species densely filled up an incredibly diverse (trophic) “morpho-space”, whereas the extent of overlap and scattering seems to vary among phylogenetic lineages. As trophic morphology strongly correlates with the ecology of the species, a similar pattern can be observed in niche use. Based on a genome-wide phylogeny we trace back patterns of eco-morphological evolution through the phylogenetic history of the radiation and test predictions of the adaptive radiation theory.

T3.7

The relationships between social structure, behavioural repertoire size, and neuroanatomy in ecologically similar but socially divergent cichlid fishes

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Living in social groups may lead to the development of more advanced cognitive abilities, and potentially more complex brain architectures, as individuals navigate a greater number of differentiated relationships with social partners. Alternatively, processes of distributed intelligence or cognitive division of labour may reduce the cognitive load on socially living animals compared with their solitary counterparts. In this talk I will discuss our ongoing work examining the social network structure, behavioural repertoire size, and neuroanatomical differences among 10 species of cichlid fish that span the spectrum of sociality. Although they share almost identical life histories, morphologies, and ecologies, these species differ greatly in their social structures and behaviour, solving identical ecological tasks either individually or in social groups. Studying wild groups in their natural habitats, we use machine vision and automated tracking to quantify social network structure of wild groups in Lake Tanganyika, creating detailed maps of the number and types of relationships individuals in each species have. We then use behavioural decomposition to map out the behavioural repertoire of each species, asking whether species with more complex social structures have richer behavioural repertoires, or if conserved repertoires take on different functional effects ('meanings') in the different species. Finally, we create neuroanatomical maps of each species, examining whether differences in social structure and behaviour are reflected in changes in the brain nuclei associated with the social decision-making network, paving the way for future analyses of neural activity and gene expression. We ask whether social evolution produces specialised behaviours and brain architectures (the social brain hypothesis), or whether a shared neuroanatomical template can plastically produce diverse social behaviour across species.

T4.1

Biodiversity policy research in Africa: lessons learned and potential for cichlid research?

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T4.2

Cichlids from Morocco: a review of native and introduced populations

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Moroccan freshwater fish diversity is low with approximately 20 native species, mainly Cyprinids, and 16 introduced ones (34%). Cichlids are present in numerous watersheds, the taxonomic status of these different populations was studied based on morphometric and genetic characters. Three native cichlid species were identified: *Oreochromis aureus* (Steindachner, 1864) and *Coptodon zillii* (Gervais, 1848) in the Drâa basin and *Coptodon guineensis* (Günther, 1862) in the Oued Chbeyka and its tributary Oued Aabar. The latter species is also present in Southern Morocco in small pools (160) of the Sebkha Imlili where it should be considered as functioning as a metapopulation (the different populations from the various pools are genetically differentiated). Despite the fact that these individuals are not really genetically distant from other *C. Guineensis* specimens from Morocco (Oued Chbeyka) or other African countries as Senegal, certain phenotypic and behavioural traits are so original that they can be considered as belonging to a different species. For aquaculture purpose, one or two cichlid species (*O. niloticus* and probably *O. aureus*) have been introduced (strains from Egypt and West Africa), and escaped in the wild. They are now present from Oued Bouregreg to Moulay Bouselham lagoon in Northern part of Morocco. Genetic studies are ongoing to better characterize these populations. Finally, a new species of Cichlidae has recently been observed in the Nador Canal (Moulay Bouselham Lagoon) whose identification is currently in progress.

T4.3

Epigenomics, plasticity, and adaptation in haplochromine cichlids

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The past two decades have observed an increase in evidence supporting non-DNA sequence-based inheritance (or epigenetic inheritance) in a variety of organisms, raising the possibility for the heritability of acquired characteristics in evolution. However, despite the rising importance of epigenetic inheritance in the study of heredity, little is known or agreed upon regarding the persistence of vertebrate non-genetic inheritance across multiple generations. The cichlid model presents an ideal opportunity to evaluate the impact of epigenetic inheritance and phenotypic plasticity upon evolution, adaptation, and varying rates of diversification. To characterize the roles of non-genetic inheritance and phenotypic plasticity in facilitating the widespread adaptive radiations characteristic of cichlid fishes, I am performing diet perturbation breeding experiments, reciprocal crosses of riverine subspecies, and comparing the epigenomes of diverse cichlids from throughout the African Great Lakes. Specifically, I am characterizing the epigenomes of *Astatotilapia calliptera*, *Rhamphochromis longiceps*, *Astatotilapia burtoni*, *Oreochromis niloticus*, and several more. Here I will present ChIP-seq data defining enhancer and promoter activation in these species, allowing for the direct comparison of epigenetic states between these fish. In this presentation, I will discuss my analysis of these results, the importance of epigenetics in understanding the details of cichlid diversification, and the implications of my findings. Furthermore, I will review the design of my breeding experiments and their potential to reveal the impact of phenotypic plasticity and epigenetic inheritance upon cichlid evolution.

T4.4

Combination of ancestral and introgressed variants promotes phenotypic divergence of stripe patterns

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There is increasing evidence for the importance of standing genetic variation and introgression in facilitating rapid phenotypic diversification. A prime example of such rapid phenotypic diversification is the evolution of pigmentation patterns in East African cichlid radiations. There, melanic horizontal stripes evolved repeatedly through regulatory evolution of a single gene, the agouti-related peptide 2 (*agrp2*). However, if causal variants have been recruited from standing variation, through introgression or evolved *de novo* remains unclear. To find and characterize the origin of the causal allelic variants, we reconstructed their evolutionary history using target-enrichment and whole-genome data. Different regions of the *agrp2* locus associate with stripes in the cichlid radiations of Lakes Malawi and Victoria. In Lake Victoria cichlids, stripes are largely determined by a single highly associated haplotype. Yet, the evolution of the causal haplotype predates the major adaptive radiation of Lake Victoria cichlids. Our results show that this haplotype evolved via a *de novo* combination of ancestral and introgressed variants. Thereby we provide novel evidence to how admixture can spark rapid phenotypic diversification at the onset of adaptive radiations.

T4.5

Encapsulation of the invasive parasite *Anguillicola crassus* by European eels may disrupt its response to native parasites

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Encapsulation of the parasitic nematode *Anguillicola crassus* is a commonly observed response in its native host, the Japanese eel (*Anguilla japonica*). This is thought to explain the lower infection intensities and better performance compared to the parasite's more recently acquired host, the European eel (*A. anguilla*). Encapsulation has been described in the European eel and there is evidence that the frequency has increased since the introduction of *A. crassus*, suggestive of ongoing adaptation. We explored whether encapsulation of *A. crassus* provides an advantage to the continental stage of its novel host in the natural environment. We provide the first evidence of reduced infection intensity of *A. crassus* in European eels that encapsulated the parasite, particularly of adult parasites. In contrast, prevalence of three native parasites were higher in these eels, although the overall parasite composition did not differ between encapsulating and non-encapsulating eels. The strength of encapsulation was negatively correlated with the strength of expression of two genes key to the adaptive immune response, suggesting that encapsulation reduces the necessity of mounting such a response. In contrast, expression of two metabolic genes was not affected by encapsulation, possibly due to increased costs from a higher prevalence of native parasites and an overall low impact of *A. crassus* on the continental stage of the European eel. Our results indicate that the European eel may be adapting to *A. crassus*, but that it may come at the expense of defending against the native parasite fauna.

T4.6

Hypothalamic cell type diversity following cave adaptation in *Astyanax mexicanus*

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Changes in behaviour are necessary for an animal's ability to adapt to new environmental challenges and ecological niches. The genetic and molecular mechanisms underlying behavioural evolution are poorly understood. Moreover, the relationship between genetic mutations and the composition and identity of neuronal cell types is unknown. To interrogate this relationship, we performed high resolution single-cell RNA-sequencing of hypothalamic cells from the blind Mexican cavefish *Astyanax mexicanus*. This species includes two morphotypes; eyed surface and blind cave dwellers, which evolved independently multiple times over the last 2-5 million years. Cave morphs share morphological, developmental, and behavioural phenotypes, including loss of sleep, reduced schooling, and hyperphagia compared to surface morphotypes. Using single-cell sequencing, we identified multiple cell-type specific differences between cave and surface fish within the hypothalamus. These included at least 20 morph specific cell clusters, and broad changes in the immunological repertoire of cave fish. By integrating analysis of whole genome sequences of *A. mexicanus* with our single cell RNA-seq data, we also identified specific genomic variants associated with cave adaptation and the observed cell type differences. Studying these genomic variants may further our understanding of the mechanisms underlying behavioural adaptation, and provide insight into how cell type definitions are shaped by evolutionary forces. We are continuing these studies using cichlids, which display extensive morphological and behavioural diversity, and will allow us to further interrogate the relationship between cell type diversity and behavioural evolution.

T4.7

Social complexity in Lamprologine cichlids

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The complexity of the social environment has been proposed to be a key driver of the cognitive evolution of animals. Despite recent progress in conceptualising social complexity, identifying a unifying index that allows comparisons within a broader range of taxonomic units still remains a major challenge. Here we propose an agent-based model that employs social network analytical metrics and takes on an information theoretical point of view to provide a quantitative assessment of social complexity. By constructing dynamic social networks of varying size, edge density, interaction density, relationship stability and breadth of behavioural repertoire, we determined the relative contribution of these parameters in driving the systemic uncertainty as perceived from the perspective of individuals embedded within a social group. In order to illustrate a case example of how our model-based approach could be applied to real-world data, we investigated the social behaviour of shell-dwelling Lamprologine cichlid species which exhibit striking ecological similarities, but differ substantially in their social organisation. After reconstructing species-stereotypic social networks, we compared the model predictions to estimate the level of complexity that each species is facing in its natural context. In so doing, we strive to gain insight into the social sources of selection that underlie the evolution of neuroanatomical and socio-cognitive features in a quantitative, comparative framework employing both empirical and theoretical approaches.

T4.8

Cellular and transcriptional analysis of the color dimorphism of *Melanochromis auratus*

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The Lake Malawi cichlid *Melanochromis auratus* has a prominent sexual dimorphism in coloration. While females and juveniles are bright yellow with two dark horizontal stripes (yellow morph), dominant males undergo a drastic change in coloration and become dark with two light blue horizontal stripes (dark morph). Here, we describe the cellular basis and transcriptional differences between the two morphs. Using light microscope and transmission electron microscope imaging, we characterize the pronounced differences on the cellular level including differences in chromatophore distribution, shape, and properties. Our transcriptomic analysis uncovers beside differentially regulated pigmentation genes also several neural genes that are highly expressed in the skin of the dark morph. By performing immunostaining, we further support these results as we show an increased density of neuronal fibers in the dark morph compared with the yellow morph. Our results give therefore novel insights into the cellular, physiological and genetic basis of color change in cichlid fishes.

T4.9

Jellyfish blooms alter activity and behaviour of territorial fish

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Jellyfish blooms pose multiple threats to wild fish. Yet, we still know little about how blooms impact fitness-related behaviours of fish caught within their areas of effect, e.g. for littoral fish that are not able to relocate to avoid incoming blooms of stinging jellyfish. Here, we documented the behavioural impacts of freshwater jellyfish (*Limnocnida tanganjicae*) blooms on a territorial cichlid (*Variabilichromis moorii*) as well as on the wider community of cichlids in a shallow-water rocky habitat of Lake Tanganyika. Compared to non-bloom conditions, *V. moorii* individuals reduced their swimming and territory defence activities during jellyfish blooms (each by ~ 50%), but not their foraging or affiliative behaviours. Despite this reduction in activity, *V. moorii* could not entirely avoid being stung and preferred to remain closer to the rocky substrata as opposed to the more open demersal zone. Many other fishes similarly hid among the benthic substrata, changing the composition of the fish community in the demersal zone during bloom conditions. Reductions in activity could have multiple fitness-related implications for individual fish. Understanding these behavioural consequences is an important topic of research as blooms are predicted to increase in severity in the future especially along coastal habitats.

T4.10

Genomic signals of congeneric hybridisation confirmed in the Lake Tanganyikan cichlid genus *Ophthalmotilapia*

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The cichlid genus *Ophthalmotilapia* of Lake Tanganyika, like many other cichlid groups, has been severely impacted by paleoclimate-driven lake level changes that have affected its evolutionary history and species distribution range. Earlier studies using mtDNA and microsatellite data have shown how this has resulted in intra-specific geneflow, which in some cases among cichlids may have led to the origin of new species via hybridisation. Such hybridisation has meant that mtDNA-based trees for *Ophthalmotilapia* deviate from the phylogeny of the genus. To further investigate the extent of hybridisation and admixture within this genus and resolve the *Ophthalmotilapia* phylogeny, we used GBS (genotyping-by-sequencing) data from over 400 specimens collected from 91 localities along Lake Tanganyika's >1500km shoreline. The GBS phylogeny supports the monophyly of *Ophthalmotilapia*, and discrepancies between the mtDNA and GBS datasets confirms instances of natural congeneric hybridisation. Analysis of population genomic structure using the ADMIXTURE software also revealed the presence of hybrid populations in regions of the lake where species occur in sympatry, although these hybrid populations were genetically distinct from F1 hybrids bred in the lab. The extent of population structure found within species was roughly correlated with the distribution size of each species, with *O. nasuta*, the species with the largest distribution, having the most amount of population structure. For smaller K models, which show the groups with the most distinct ancestry, some populations of *O. heterodonta* and *O. ventralis* shared the same ancestral component, suggesting that these two species may actually be better delineated as one species.

PLENARY LECTURE

The cichlids of the north - stickleback as model system in evolutionary biology

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Parasites are ubiquitous. They impact both ecological dynamics as well as adaptive evolution of their hosts. Focusing on a series of experiments using the three-spined stickleback as a model host species, I will report about various molecular bases of resistance whether genetics or epigenetics. Furthermore, I will elaborate using field data and experiments how parasites can reinforce and even promote adaptive divergence of their stickleback hosts. Lastly, I will describe how parasites and the evolution of parasite resistance can mediate eco-evolutionary feedbacks altering prey communities impacting the selection pressure on subsequent host generations. Given the high parasite abundance in natural populations, and the rapid evolution of resistance, I argue parasites are still neglected as agents of natural selection and the stickleback model system is an ideal “supermodel” (along with cichlids!) to tackle fundamental questions in evolutionary biology.

T5.1

Evolution of hemoglobin genes in cichlid fishes

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In evolution, vertebrates underwent two rounds of the whole-genome duplication, which multiplied their genomic content by “one-to-four” (genes) rule and therefore probably facilitated subsequent evolutionary success of early vertebrates. Teleost fishes faced an additional (third) teleost-specific genome duplication around 226-316 mya. Cichlids are prominent group of teleost fishes with remarkable variety in number of species, diet and reproductive strategies, sometime hypothesized to be associated with increased number of paralogous genes that could promote evolutionary success of some lineages.

Hemoglobin protein is responsible for oxygen transport and is composed of two alpha and two beta subunits. Hemoglobin subunit genes passed through quantitative expansion in modern teleost, which have the greatest number of hemoglobin genes among gnathostomes organized in two clusters (LA and MN). In this study we investigated the whole genome data and found that *Oreochromis niloticus* has the highest number of hemoglobin genes among bony fishes (29 in MN cluster and 3 in LA) or even the jawed vertebrates. We have further focused on the syntenic organization of hemoglobin genes in different lineages of bony-fishes with special focus on the cichlids (10 species including four endemic cichlid species from Barombi Mbo lake, West Africa). Furthermore, we reconstructed large scale phylogeny of hemoglobin genes of teleosts and few other groups of vertebrates. We show that multiplicity of hemoglobin genes in cichlids is likely a product of tandem duplications. Moreover, we suggest the evolutionary scenario describing general evolution of the hemoglobin genes in different groups of teleost fishes.

T5.2

Which role does behaviour play in the adaptive radiation of cichlid fishes from Lake Tanganyika?

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Behaviour determines how individuals interact with their environment and is, hence, deeply intertwined with mechanisms considered to play an important role in the build-up of reproductive isolation. For instance, the exploitation of a novel resource or the scouting for new habitats are directly influenced by individual behaviour. However, empirical studies on the link between behaviour and speciation processes are scarce. We apply a standardized approach, using wild-caught animals under semi-natural conditions, to investigate the role of a basic behavioural trait in the context of the massive adaptive radiation of cichlids in Lake Tanganyika. More precisely, we gather quantitative data on novelty seeking/exploration tendency for about 40 cichlid species comprising species-rich and species-poor lineages. The resulting behavioural data will be integrated with available whole-genome sequences and existing eco-morphological information of the respective species in order to get first in-depth insights into how behaviour contributes to the species' propensity to diversify.

T5.3

Contrasting signatures of genomic divergence in rapidly speciating crater lake cichlid fishes

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Historically, speciation was assumed to require geographic barriers preventing gene flow. Still today, empirical examples for sympatric speciation are rare. Theory predicts that the genetic architecture of divergently selected traits can influence whether it can occur. To test this empirically, we studied a young adaptive cichlid radiation that speciated within Nicaraguan crater lakes using 453 re-sequenced genomes. We analyzed genomic divergence along multiple phenotypic axes and investigated their genetic bases. Species that differ in mono- or oligogenic traits affecting ecological performance and/or mate choice, show remarkably localized genomic differentiation. Genome-wide differentiation of species that diverged in polygenic traits is, however, much higher overall. Contrary to expectation, simple trait architectures are not as conducive to speciation-with-gene-flow whereas polygenic trait architectures can promote rapid speciation in sympatry.

T5.4

Ancestral hybridisation facilitated species diversification in the Lake Malawi cichlid fish adaptive radiation

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The cichlid fish adaptive radiation of Lake Malawi encompasses over 500 diverse species that evolved within the last 800 thousand years. It has been proposed that hybridisation between ancestral lineages can provide the genetic and phenotypic variation to fuel such exceptionally high diversification rates, and evidence for this has recently been presented for the Lake Victoria Region cichlid superflock. Here we report that Lake Malawi cichlid genomes also show evidence of hybridisation between two ancestral lineages that split around two million years ago, today represented by Lake Victoria cichlids and the riverine *Astatotilapia* sp. 'ruaha blue'. Hybridisation-derived loci show striking excess differentiation across Malawi cichlid ecological subgroups, and are enriched for genes involved in immune response and vision. Moreover, sorting of parental alleles into Lake Malawi cichlid species is consistent with selection on incompatibilities having contributed to re-divergence of introgressed alleles. However, more work is needed to fully evaluate the role of incompatibilities during species divergence. Our results reinforce mounting evidence for the role of ancestral hybridisation in adaptive divergence, by demonstrating its significance in one of the largest recent vertebrate adaptive radiations.

T5.5

The origin and population structure of introduced Nile tilapia populations in Central Africa

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Nile tilapia (*Oreochromis niloticus*) is one of the most important fish species for African aquaculture. They are affordable, low demanding in terms of food source, tolerant to environmental stress, fast-growing and they have a high reproductive rate. This species has been introduced in many countries outside its native range. The flipside of the coin is that they can become a threat to native fish fauna, and reduce their abundance by e.g. competition, hybridization and predation. The historical introduction of Nile tilapia in many African countries is poorly documented. Given the well-documented negative effects introduced Nile tilapia can have on native fish species in other regions of the world, it is paramount to identify and trace stocking events in Africa. In the present study, we are exploring the possible origin and population structure of introduced populations of *O. niloticus* in Central Africa. We included specimens from wild and farmed populations and used Restriction site Associated DNA (RAD) markers to explore if, and to what extent, fish populations could be differentiated using high-resolution genetic data. Our preliminary population genomic analyses show that fish from the same geographical location tend to be more genetically alike. Even within the Congo Basin, genetic differentiation was visible between populations from the Upper, Middle and Lower Congo Basin. This could indicate that Nile tilapia was introduced here from different origins or fish stocks. As a next step, we will include more material from the Nile tilapia's native range to infer the geographical origins of these introductions.

T5.6

Sardine science: genomics and stakeholder involvement for sustainable management of Lake Tanganyika clupeid fisheries

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Worldwide, fisheries are threatened by the consequences of climate change and overfishing. For people inhabiting a developing region, collapse of fisheries can be insuperable. The Lake Tanganyika fishery feeds millions of people in one of the poorest regions of the world. Lake Tanganyika is known for its endemic cichlid radiations, yet most of its fishery target species are pelagic non-cichlids. About 60% of the catch consists of two endemic clupeid species: *Limnothrissa miodon* and *Stolothrissa tanganyicae*. Formulation of a much needed evidence-based management plan is hampered because of knowledge gaps concerning stakeholder opinions, biology, and geographic and temporal population structure of the target species. To provide this information, we combine molecular techniques with stakeholder interviews. We screened the genome of *S. tanganyicae* to disentangle its population structure, crucial for the delineation of management units. Results of mitochondrial genotyping and RAD sequencing show absence of genetic differentiation over the lake. This finding suggests frequent clupeid migration and thus crossing of national borders, highlighting the need for joint management by all four riparian countries. Interviews with stakeholders in the DR Congo provided insight into perceived problems and suggested solutions. We found a discrepancy in preferred management strategies between fishermen and state employees. Despite reported decrease of fish abundance and size, awareness to stock decline was low. The results will be communicated to policy-makers in different countries, to allow future integration into management plans. We will develop transferrable methods that can support sustainable fisheries management in other East African Great Lakes and beyond.

T5.7

Extreme environmental tolerance and invasive success of Nile tilapia

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Invasive species have led to substantial changes in the biodiversity of freshwater ecosystems around the world. The African cichlid fish Nile tilapia (*Oreochromis niloticus*) is one of most widely distributed freshwater species globally through its use in aquaculture and capture fisheries improvement. Feral populations readily establish, and these have been proposed to influence the diversity of native species through competitive effects. However, direct evidence of competition between Nile tilapia and native species is notably rare and is not clear how environmental influences such as temperature and oxygen may modulate competition. Here we explored interactions between invasive Nile tilapia and the native Mayan cichlid (*Mayaheros urophthalmus*) which co-occur in freshwaters of south-eastern Mexico. Mayan cichlids were placed in experimental mesocosms varying in temperature, oxygen content and Nile tilapia density, and aggressive behaviour and space use was quantified from video footage. We found that Nile tilapia was by the most active and aggressive of the two species, and that environmental influences only weakly influenced activity. By contrast, the activity of the less aggressive Mayan cichlid was strongly predicted by the activity and aggressive behaviour of the Nile tilapia, and Mayan cichlids showed a steep decline in behaviours with increased water temperature and reduced oxygen. These results provide evidence that the broad environmental tolerance of the intrinsically aggressive Nile tilapia enables it to perform more effectively than native species in challenging conditions. This suggests that Nile tilapia may have an advantage over native species during periods of extreme conditions, which may help to exacerbate its invasive success as those conditions become more frequent in a changing world.

T5.8

Sympatric speciation in cichlids: how does it get started?

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An increasing number of studies are indicating that at least part of the process of speciation in cichlids and other fish may take place without complete geographic isolation. If sympatric speciation takes place, how might it get started? I consider some speculative scenarios based on known study systems in the hope of helping us to get an idea of what to look for in our research programmes.

T5.9

Mechanical modularity and functional feeding convergence in Lake Malawi cichlids

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Complexity in how mechanistic variation translates into ecological novelty could be critical to organismal diversification. For instance, when multiple distinct morphologies can generate the same mechanical or functional phenotype this could mitigate tradeoffs and/or provide alternative ways to meet the same ecological challenge. To investigate how this type of complexity shapes diversity in a classic adaptive radiation, we tested several evolutionary consequences of the anterior jaw four-bar linkage for Lake Malawi cichlid trophic diversification. Using a novel phylogenetic framework, we demonstrated that different mechanical outputs of the same four jaw elements are evolutionarily associated with both jaw protrusion distance and jaw protrusion angle. However, these two functional aspects of jaw protrusion have evolved independently. Additionally, although four-bar morphology showed little evidence for attraction to optima, there was substantial evidence of adaptive peaks for emergent four-bar linkage mechanics and jaw protrusion abilities among Malawi feeding guilds. Finally, we highlighted a clear case of two cichlid species that have independently evolved to graze algae in less than two million years and have converged on similar jaw protrusion abilities as well as four-bar linkage mechanics, but have evolved these similarities via non-convergent four-bar morphologies.

T5.10

High-resolution introgression scans reveal adaptive loci in sympatric species of rock-dwelling cichlids from Lake Malawi

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Species introductions provide an excellent setting for studying how hybridization with native species and evolutionary forces like selection and drift operate on genetic variation to promote rapid adaptation. The haplochromine cichlid species diversity of Lake Malawi in East Africa represents one of the most impressive examples of how complex population histories, often involving admixture, can lead to explosive speciation. The propensity of cichlid fishes to diversify makes investigating how introduced populations respond to new environments an ideal context in which to search for adaptive phenotypes and the genes underlying them, and to understand the evolutionary mechanisms that allow these introduced populations to persist. In Lake Malawi National Park, the rock-dwelling cichlid species *Cynotilapia afra* have established wild populations within the last 60 years following escape from fish exporters, and have subsequently hybridized with the native *Maylandia zebra* species. Using 146 high-quality whole genomes we characterized the genetic relationships among *C. afra* and *M. zebra* within and outside of the National Park. We have investigated a genome-wide signal of *M. zebra* introgression into *C. afra* using high-resolution genome scans based on a new statistic that allows us to efficiently identify genomic regions with high similarity exclusive to two populations. This has highlighted five genomic regions showing evidence of selective sweeps, where only the introduced *C. afra* show extensive haplotype sharing with *M. zebra*. Together this points to adaptive introgression of *M. zebra* into *C. afra* at regions spanning genes with functions known to be important in biological invasions such as morphogenesis.

T5.11

Monogenean parasites of sardines in Lake Tanganyika: potential tags for host history and population structure

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Lake Tanganyika is the oldest and deepest of the African Great Lakes and harbours one of the most diverse fish assemblages on earth. Of the entire fisheries concentrated mainly in the lake's pelagic zone, two species of sardines (*Limnothrissa miodon* and *Stolothrissa tanganyicae*) constitute the majority within the total of catches. To provide an additional view of their lake-wide population structure, we examined the monogenean parasites (Platyhelminthes) of the abovementioned species in order to explore the parasites' potential as tags for their hosts' population structure or history. Samples originated from several localities including all three subbasins of the lake. Intraspecific morphological variation was analysed using morphometrics and geomorphometrics of the monogeneans' sclerotized structures. Molecular characterisation was conducted using a range of nuclear and mitochondrial markers. Our results indicate a lake-wide distribution of two monogenean species assigned to a new genus, *Kapentagyryus*, infecting Lake Tanganyika's sardines. Unlike *Cichlidogyryus casuarinus*, a monogenean reported from members of the pelagic cichlid tribe Bathybatini with no host preference, *K. tanganicanus* includes two morphotypes specific to the respective sardine species. Moreover, incipient speciation related to host species identity was reported in *K. tanganicanus*. Based on our molecular markers, we find a near panmictic population of *Kapentagyryus* spp. with an indication of restricted gene flow on a lake-wide scale. Morphometric and shape variation data of both parasite species revealed significant differences in some of the parameters, potentially an indication of limited host migration. Moreover, both parasites species underwent recent demographic expansion that can be linked to paleogeographic events.

T5.12

Division of attention to competing ecological tasks is mediated by social evolution

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Fighting may increase risk of predation due to trade-offs with anti-predator vigilance. In a laboratory study, we focus on the behavioural response to predator presentation of three different shell-breeding cichlids, *Neolamprologus multifasciatus*, *Neolamprologus brevis*, and *Lamprologus ocellatus*, ranging from highly social to highly solitary species, respectively. We altered the presentation of a predator to manipulate perceived predation risk, and examined subsequent aggression towards a conspecific intruder. We hypothesised that males from more social species (*N. multifasciatus*) would show lower overall aggression, and would rapidly shift their attention to the predator, displaying lowest overall aggression in the predator treatment. In contrast, we hypothesised less social species (*N. brevis* and *L. ocellatus*) would show high levels of aggression, and would not be able to divide attention between predator vigilance and aggression towards an intruder. In order to compare the aggression level between these species, we needed a common ethogram, which can be difficult to construct based on objective human comparison. We therefore applied supervised machine learning to deconstruct behaviour into constituent kinematic and interaction elements, populating an objective and quantitative ethogram for each species in each context. These ethograms allow comparison of key features of behaviour as well as sequences of behaviour, and serve as a basis for comparison between social and solitary species. Aggression was significantly reduced in the presence of predators in all three focal species, and the most social species, *N. multifasciatus*, showed lowest aggression overall. This species also showed a longer and more transitional elevation in aggression, developing from display, to mild fighting, to extreme fighting slowly, whereas the less social species moved immediately to overt aggression. Our study reveals how attentional allocation to competing ecological tasks may be mediated by the process of social evolution.

T5.13

High-resolution characterization of the genetic relationships among species of the rock-dwelling *Maylandia* cichlids from Lake Malawi using whole genomes

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The haplochromine cichlids of the East African Great Lakes comprise a recent, extensive, adaptive radiation, making them a model for speciation research. Using over 300 whole-genome sequences, we characterized the genetic relationship among eight species of the rock-dwelling *Maylandia* genus from Lake Malawi. While being mostly consistent with current species classifications, this molecular view has revealed some discrepancies; the most striking of which is that *M. emmiltos* sampled from Luwino Reef is evolutionarily more similar to the *M. fainzilberi* species than to another population of *M. emmiltos* from a nearby location. Demographic inference with MSMC2 and MOMI2 suggests that the Luwino Reef *M. emmiltos* represents a cryptic sister species to the other *M. emmiltos* and that this lineage is more directly ancestral to *M. fainzilberi*. Using D-statistic tests for admixture at the genome-wide level, we also find evidence of potential admixture between the Luwino Reef *M. emmiltos* lineage and sympatric *M. fainzilberi*. D-statistic tests also point to low levels of gene flow between populations of *M. fainzilberi*. For another species, *M. lanisticola* north, we find a transition to transversion ratio that departs substantially from the rest of the genus, suggesting a potentially different mutational profile for this species. These results set the stage for future, high-resolution scans for localized admixture across the genomes of the *Maylandia* species, and for determining whether this admixture could be adaptive.

T5.14

Motion patterns, predictability and predation responses within a wild, freely moving fish school

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The collective motion of fish schools is well understood on a mechanistic level, however, only few studies can link their findings to individual fitness instead of group properties. Here, a cichlid fish species, *Lamprologus callipterus*, was used as a model system to analyse collective motion patterns in natural environments with respect to individual behaviour and interspecific interactions of potential risk. A combination of Structure-from-Motion scene reconstruction and high-resolution animal tracking via deep neural networks allowed detailed trajectory analyses using behavioural decomposition. A time-frequency representation of the animals' motion kinematics via continuous wavelet transformation resulted in a high-dimensional description of continuous behaviour, including temporal information. On this basis, a stochastic neighbour embedding was used to reduce the dimensionality of motion data, allowing subsequent clustering. Hence, unsupervised labelling of group states and individual behaviour was possible and enabled further behavioural analyses. The key findings suggest that the motion behaviours of individuals within schools of *L. callipterus* are synchronised, even under precarious environmental scenarios such as predation. In contrast, the predictability of individual behaviour transitions was found to be reduced from a high baseline under these risky circumstances. This indicates a behavioural adaptation to predation risk on the individual level that is potentially linked to central hypotheses concerning the evolution of collective systems such as the predator confusion hypothesis.

T5.15

Comparative morphometric analysis of trophic morphologies in cichlid fish larvae and adults of the three Great East Africa Lakes

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Cichlids represent an excellent model system for analysing evolutionary processes, especially in terms of adaptation to different trophic niches over the course of their evolutionary history. We use micro-computer tomography images of the viscerocranial area to analyse trophic jaw morphologies between the three lakes at two key life stages. A crucial time point in the development of cichlids is the point at which the feeding apparatus is fully functional. We know from preliminary research that the articular bone and the lower pharyngeal jaw are fully ossified and functional before reaching the end of the larval period, but further differentiation and the adaptation to specific diets occurs at later life stages. In this study we analyse these changes in the craniofacial structures between the postlarval stage, which is marked by the complete absorption of the yolk sac and the onset of independent foraging, and the young adult stage when sexual maturity is reached. By comparing a set of species adapted to distinct trophic niches between the three Great East African Lakes we want to address the degrees of parallel evolution of trophic niche morphologies. As I have just started my master's degree, I will present an outlook of my thesis and methodical aspects of my work.

T5.16

Sex-specific costs of ecological resource manipulation in group-living fish *Neolamprologus multifasciatus*

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When resources become limited in animal groups, costs may be disproportionately felt by individuals in different social positions. As such, conflict between or within particular social classes may increase under conditions of resource limitation but be unaffected in other classes. Here we explore how inter- and intrasexual conflict is mediated by reduction in the availability of breeding sites in a communally breeding Cichlid fish. Small groups of *Neolamprologus multifasciatus* were presented with a foreign male or female conspecific in Lake Tanganyika while the amount of shells (which act as breeding shelters) in the nest was manipulated. Using machine learning assisted tracking and behavioural analysis, we then measured all interactions among group members and towards introduced conspecifics. Aggression among females was predicted to be higher when resources were limited due to higher relative costs than for males, for whom aggression was expected to be unaffected by resource reduction. Inter-sexual interactions were more complex, with males expected to respond more aggressively to foreign males and resident females, but show more courtship to foreign females, when resources were increased. In contrast, female responses to males were predicted to be unaffected in all conditions. Understanding how social group interactions are mediated by fluctuations in resource availability offers insight into the mechanisms of selection on social behaviour generally.

Posters

P1.1

TEs landscape in cichlid species part of the Lake Tanganyika adaptive radiation

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Transposable Elements (TEs) are selfish DNA elements able to move and spread within a genome and therefore considered to be drivers of the evolution of host genomes. TEs are part of the repeat content and usually grouped into two classes: retrotransposons and (DNA) transposons. The structural alterations caused by TEs in addition to mutations generated within TEs vary within and among species. Besides the overall effect of TEs on the sequence, size and structure of the host genome, transposable activity can alter gene expression, function and coding ability. For example, TE insertions can modify or create new exons, regulatory sequences, and RNA or protein-coding genes, potentially leading to novel phenotypes. It has therefore been hypothesized that TEs play a role in evolutionary processes such as adaptation, diversification and speciation. The species flock of cichlid fishes in East African Lake Tanganyika (LT) constitutes one of the most species-rich and ecologically and morphologically diverse extant adaptive radiation. Adaptive radiation – that is the macroevolutionary process by which an ancestral species rapidly diversifies into an array of descendants by evolving adaptations to exploit various ecological niches – is an ideal set-up to examine the role of TEs in organismal diversification. In this project, I identified TEs in more than 220 cichlid species part of the LT adaptive radiation. Then, I looked into the TEs landscape of cichlid species according to their phylogenetic relationship. Particularly, I investigated whether the proportion of TEs can be associated with the species richness of the different tribes.

P1.2

The role of the lateral line system in adaptive radiation of fishes

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The mechanosensory lateral line system is used by fishes to sense the movement of water in their environment. The system provides information about hydrodynamic stimuli, both abiotic and biotic in nature, which informs vital behaviours, including schooling, prey detection, predator avoidance, and alignment to water flow. As such, we would expect significant morphological diversity among African lake cichlid radiations. Cichlids are known to exhibit a degree of diversity in their lateral line system morphology, but variation had never been quantified across a major vertebrate radiation. We aimed to describe variation in the system's morphology for the first time, using the cranial canal system of the Lake Malawi flock. We found high morphological diversity, and strong associations with both gross skeletal morphology and diet, suggesting a role for the head canals in trophic niche segregation during the radiation process (Edgley & Genner, 2019). Building upon this, we aim to determine the role of the lateral line system in adaptive radiation of fishes, and in the evolutionary process more broadly. To do this, we will: a) Characterise phenotypic variation among the Lake Malawi, Tanganyika, and Victoria cichlid radiations using microCT scanning with geometric morphometrics, and fluorescence microscopy. b) Determine the genetic basis of this variation using QTL mapping. c) Use the Lake Massoko *Astatotilapia calliptera* model system to determine the role of the lateral line system in the early stages of ecological speciation, giving insights into whether the system can promote adaptive evolution.

P1.3

Convergent evolution of Amazonian and East African cichlids: a morphological approach

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The richness and the proportion of endemism on the African Great Lakes are unparalleled in cichlid fish. However, also South American – and, in particular, Amazonian – cichlids show a great deal of morphological and ecological diversity. The cichlids offer in abundance two of the characteristics that have facilitated analysis of adaptive traits in other taxa: there are many closely related species that show highly divergent morphology, and there is repeated evolution of similar traits in parallel. Thus, to understand the phenomenon of convergent evolution in cichlids, it is important to investigate diverse cichlid assemblages that are unrelated and isolated from the cichlid assemblages in East Africa. Cichlid fishes from the Neotropics offer such a comparative system. In this study we used geometric morphometric analyses to compare body shape within neotropical cichlids and between South American and African cichlids. In a dataset of 3887 individuals, comprising all the tribes from Lake Malawi, Lake Tanganyika and South America, we found that the South American tribes *Geophagini*, *Retroculini* and *Cichlini* converge in overall body shape within the groups from the African lakes, and that extreme body shapes like those of the genera *Pterophylum*, *Uaru* and *Symphisodom* are located distantly of the main cloud scores in morphospace. Representatives of *Geophagini*, *Retroculini* and *Cichlini* probably share similar habitats and foraging habits with main representative Tanganyika and Malawi genera; *Tropheus*, *Petrochromis*, *Neolamprologus* and *Haplochromis*, sharing the same environmental pressure which can lead to body shape convergence through selection in the pre-existent variation.

P1.4

Red and yellow body coloration in haplochromine cichlids: Characterizing carotenoid diversity

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In cichlids, body coloration plays an important role in species recognition, mate choice and social competition. Variation in carotenoid-based coloration is commonly found between closely related cichlids. Like most animals, cichlids cannot synthesize carotenoids but rely on ingestion and metabolic conversion of dietary carotenoids, and hues of integumentary coloration are determined both by concentrations and types of carotenoids. To compare carotenoid composition between white, yellow and red colored skin, we fed different color morphs of *Tropheus* and *Aulonocara* with identical diets and then analyzed skin extracts with reversed phase Ultra High Performance Liquid Chromatography (UHPLC) and orbitrap-based mass spectrometry. While some yellow carotenoids like lutein and zeaxanthin were present in nearly all skin samples, the red carotenoids astacene (perhaps as a saponification derivative of astaxanthin), rhodoxanthin and canthaxanthin were identified mainly in red skin samples. Interestingly, a white-colored skin region of *Tropheus duboisi* also contained low amounts of carotenoids. Our study identified population-specific carotenoid profiles, indicative of diversification in the underlying metabolic processes (and hence, associated genetic mechanisms) even among closely related cichlid taxa.

P1.5

A technical summary of Tanganyikan Cichlid RNA Extractions

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Since the beginning of gene expression assays leading to today's RNAseq techniques, the quality of total extracted RNA has always been considered as a primary requirement for successful and unbiased gene expression comparisons. Obtaining good quality RNA from organisms that cannot easily be raised in the laboratory is challenging. RNA is particularly sensitive to degradation by nucleases, which can distort the global picture of transcript expression. Within the framework of an integrative transcriptomics project dealing with representatives from all Lake Tanganyika cichlid tribes, we extracted total RNA from five different tissues (brain, gonads, liver, gills and lower pharyngeal jaw) to be subject to Illumina RNA sequencing. Here, we examine several quality criteria obtained from spectrophotometry, fluorometry by hybridisation and fluorescent microfluidic electrophoresis by comparing how they influenced various preliminary sequencing metrics such as the number of raw reads, the percentage of reads that passed the filtering as well as the percentage of reads mapped to a reference. Our data show that RNA fragmentation only influenced the percentage of mapping reads with notable disparities between tribes, species and the type of sampled tissue. In particular, total RNA from large sized species tends to be more fragmented. These findings provide a first overview of RNA extraction performance in Tanganyikan cichlids and will be useful to future studies involving RNAseq from field collection.

P1.6

Classifying colors: resolving the relationships among color variants of the Lake Tanganyika cichlid genus *Tropheus*

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With >120 mostly allopatrically distributed color morphs, the stenotopic rock-dwelling Lake Tanganyika cichlid genus *Tropheus* is undoubtedly one of the most spectacular examples of geographic color variation in vertebrates. Yet, to date, *Tropheus* comprises only four valid species, mainly because overall morphology appears to be quite conserved across most populations and color morphs. However, genetic (and to some extent also morphological) analyses indicated the presence of additional diversity that is not reflected by currently recognized species. Here, we present by far the most comprehensive attempt to disentangle the phylogenetic relationships among *Tropheus* species using a genome-scale dataset obtained by anchored hybrid enrichment, AFLPs, mtDNA sequences, and morphological data. We find evidence for i) the presence of several additional *Tropheus* species, some of which appear to be confined to fairly narrow stretches of shoreline, ii) complex patterns of haplotype sharing/gene flow among (sometimes geographically-distant) populations/species, and iii) signatures of hybridization contributing to the origin of novel phenotypes. Moreover, this study provides a robust phylogenetic framework for analyzing the factors and processes underlying the evolution of new color morphs in this rapid radiation.

P1.7

Sex determining loci and genetic divergence between two ecomorphs of *Astatotilapia calliptera* in Lake Massoko, Tanzania

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The cichlid fishes of East Africa constitute some of the most extensive vertebrate evolutionary radiations. In Lake Massoko, a small crater lake in Tanzania, the haplochromine cichlid *Astatotilapia calliptera* has diverged genetically and phenotypically into two distinct ecomorphs, providing a system to investigate the genetic architecture underlying phenotypic diversification and sympatric speciation. Here we characterize the genetic variation among over 300 Massoko *A. calliptera* using high-coverage whole genome sequences mapped to our new, high-quality *A. calliptera* reference genome. We find clear genome-wide separation between benthic and littoral ecomorphs while the littoral morph appears behaviourally more generalist. We also find specific regions with relatively high allele frequency divergence. We also performed a genome-wide association study (GWAS) between males (n=282) and females (n=54) to identify loci involved in sex determination which is poorly characterised in cichlids. We find a highly significant association between sex and gonadal somatic cell derived factor (*gsdf*) SNPs; this gene has previously been implicated in sex determination in other teleost fish and specifically in the closely related Nile tilapia (*Oreochromis niloticus*). Sequencing depth in this region suggests an XX female / XY male system with a duplication of the Y locus. The GWAS also reveals evidence for a possible secondary sex-modifying locus downstream of *gsdf*. The positioning of *A. calliptera* as a putative immediate outgroup of the Lake Malawi cichlid radiation indicates that our findings may also be informative to understanding sex determination in these species.

P1.8

FISHing for opsins: photoreceptor distribution in the retina of the Cameroonian crater lake cichlids

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Cichlid visual system is highly adaptive to the environment. For better understanding of the performance and evolution of cichlid visual perception, we applied fluorescent in-situ hybridisation (FISH) to investigate spatial photoreceptor distribution in the retina of cichlids from the crater lake Barombi Mbo (Cameroon) inhabiting different environments, namely shallow-water species *Konia eisentrauti* and *Stomatepia mariae*, seasonally deep-water *Myaka myaka*, and deep-water *Konia dikume*. The shallow-water cichlids express mainly three types of opsin genes: LWS (red wavelengths), RH2A (green; two copies, RH2A α , RH2A β) and SWS2A (blue). Contrarily, both deep-water species lack expression of the red photoreceptor (LWS) in their retinæ. Instead, the expression of RH2A α (i.e. the copy with longer-wavelength maximum) is increased, suggesting the functional replacement of LWS by RH2A α . This finding corresponds to the light conditions in deep water, where only green/blue part of the light spectrum is present. These two unrelated deep-water species probably developed such adaptation to the environment independently. The adaptation to the environment is also evident in the retinal mosaics. *S. mariae* embodies common cichlid retinal mosaic: single cones expressing SWS2A surrounded by four double cones with LWS in one and RH2A in second part. *K. eisentrauti* follows a similar pattern; however, some double cones express LWS in both parts. In *M. myaka*, RH2A is found in both parts of double cones, i.e. replacing missing LWS. The retinal mosaic of *K. dikume* is less organised with dominance of double cones and virtual lack of single cones; the spatial distribution of opsin genes remains unclear.

P1.9

Modification of colour vision in African riverine cichlids

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Cichlid fishes have as many as seven different cone opsin genes (SWS1, SWS2A, SWS2B, RH2A α , RH2A β , RH2B and LWS) in their genome responsible for colour vision. Each opsin protein is sensitive to different wavelengths (= colours); however, only a subset of them are usually expressed as previously shown in lacustrine cichlids from lakes Victoria, Tanganyika and Malawi. Variation in expression is to certain extent associated with environmental conditions. In our study, we focused on eleven species of African riverine cichlids from eight different tribes spanning the cichlid phylogeny including the basal African lineages (Heterochromini, Tylochromini, Chromidotilapiini, Hemichromini, Etiini, Coptodonini, Gobiocichlini, Pelmatolapiini). Using the transcriptomic data from retina, we show that, in general, cichlids actively use up to five cone opsins in their retina and their vision is dominated by the red-sensitive photoreceptors. Interestingly, we found that in Tylochromini, the dominant red-sensitive photoreceptor (LWS) is lost and is replaced by green-sensitive opsins (RH2). We further identified that riverine cichlids have higher proportional expression of the red-sensitive gene than their lacustrine relatives, which is probably associated with the light environment in the lakes (as compared to rivers). Some of riverine species also lost UV-sensitive opsin expression. In this study, we further sequenced cone opsin genes, focusing on amino-acid mutations in the known key tuning sites of the protein, which can putatively shift photosensitivity of the protein to different colour. Such mutations possibly alter the opsin protein function in response to the environmental conditions or feeding or reproductive strategy.

P1.10

Comparative scale morphology in the Lake Tanganyika cichlid species flock

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The Lake Tanganyika cichlid adaptive radiation consists of about 250 species and shows an exceptional degree of morphological as well as ecological diversity, thus providing an ideal model system to study speciation and adaptation. Here, we report a comparative morphological approach on the basis of scale measurements from virtually all Tanganyika cichlid species, which provides novel insights into the patterns and processes of diversification on the level of species, genera and tribes. We find that different aspects of scale morphology – such as scale type, size and shape – reflect adaptations to different ecological niches and that scale variation is correlated with body morphology in Lake Tanganyika cichlids. We further show that scale size is associated with body depth and that there is a strong phylogenetic signal in standard-length-corrected scale sizes across the Tanganyikan cichlid species.

P1.11

Cichlids from Costa Rica, Central America: diversity and conservation

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Despite of the relatively small continental size of Costa Rica, the country has a rich freshwater fish fauna, with cichlids comprising the richest family with 25 native species (two endemic) and several introduced species (*Oreochromis* spp.). Based on literature review and recent field sampling of the authors, the present work comments on cichlids species from Costa Rica: its taxonomy, systematics, changes in geographic distribution and conservation status. The genus and relationships of most of the species have changed in relation to the seminal book of freshwater fishes of Costa Rica by Bussing (1998). At least one species of *Amphilophus*, *Hypsophrys*, *Neetroplus* and the three species of *Parachromis* have been moved to basins different than its original distribution in the country, which comprise the introduction of species from the Atlantic versant to the Pacific one. The effect on freshwater communities of these translocated cichlids species is still unknown. About conservation status, despite several field trips of the authors to find the species *Cribroheros rhytisma* and *Amphilophus lyonsi*, the species have not been found in places where they were already collected in the past, suggesting a local extirpation. Finally, promising areas of research include mating behaviour, ecomorphology, feeding ecology and response to environmental anthropic alterations.

P1.12

Ecological divergence of *Astyanax* morphs (Teleostei: Characidae) and association to their genetic structure

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The association between ecological segregation and morphological divergence could be considered a signal of divergent selection, which could promote ecological speciation when reproductive isolation occurs. Within the *Astyanax* genus we found two divergent morphs in sympatry (i.e. elongate and deep-body), giving an opportunity to study the correlation between ecology (trophic level) and reproductive isolation. We collected both morphs of *Astyanax* from Río San Juan basin in Lake Managua, Lake Nicaragua and River Sarapiquí and analyzed: 1) their trophic level and niche width by stable isotopic ratios of nitrogen ($\delta^{15}\text{N}$) and carbon ($\delta^{13}\text{C}$), 2) the relation between trophic level and detailed morphology by geometric morphometrics of body and the premaxillary bone and 3) the genetic structure between morphs using the gene fragment mt-cytb and 11 microsatellites. For lacustrine morphs, the trophic niche width for the deep-body morph was greater than for the elongate-body morph which suggests trophic specialization in the elongate-body morph. This morph also showed a higher trophic level: a slender body and a premaxilla with acute angle and a narrow ascending process was positively correlated with $\delta^{15}\text{N}$ ratio. Despite the ecological divergence, there was a weak genetic differentiation between morphs, recovered only by the nuclear markers, however, we observed isolation by distance associated to the hydrography. In conclusion, recent divergence, gene flow or an ecomorphological polymorphic nature of this characin could explain the lack of genetic differentiation between morphs. Further studies including genomic data could shed light on the genetic basis of morphological divergence in this model.

P1.13

Parasite assemblages of the Midas Cichlid in the Nicaraguan lakes

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Host-parasite interactions impose selective pressures capable of inducing evolutionary changes in both hosts and parasites. On the frame of ecological speciation theory, alternative environments hold different parasite assemblages which might promote divergence of host populations. The Midas Cichlid species complex is a recent adaptive radiation, where parallel ecomorphotypes have evolved independently in several crater lakes. In this study, we have characterized the macroparasite communities of the Midas cichlid populations in several lakes in Nicaragua to test the hypothesis that parasites might induce host divergence. We found that the Midas cichlids are infected by multiple species of parasites, composed by a common core macroparasite fauna repeated in all lakes, and lake-specific and species within lake-specific parasite assemblages. This contrasting parasite community could be one of the drivers maintaining and/or promoting differentiation between host species, and the presence of lake-specific parasite communities suggest that each lake might follow to some extent its own evolutionary trajectory.

P1.14

Morphological divergence in neotropical crater lake cichlids

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The Nicaraguan Midas cichlid (*Amphilophus* sp.) is a polymorphic species complex with variable trophic apparatus (pharyngeal jaws) and body shape. The pharyngeal jaw morphotype of the cichlids has been linked to diet preferences, while body shape is related to habitat preference. The Midas cichlid is distributed in the two Great Nicaraguan lakes (Managua and Nicaragua) and in several lagoons of volcanic origin. The Midas cichlid has diverged in sympatry within some crater lakes as a result of ecological speciation mediated by the environment. Here we focus on the radiation of Midas cichlid in Crater Lake Asososca León, one of the most isolated volcanic lagoons in the region. We find evidence of incipient speciation in this depauperated crater lake due to relaxed interspecific competition. To test this hypothesis we linked morphology with fish physiological responses. We used geometric morphometric tools to analyze body and pharyngeal jaw shape of fish and found evidence of morphological divergence within the lake, with individuals with longer bodies having more robust jaws, and fish with taller bodies having jaws with more delicate teeth. In addition, we found differences in the relative size of the spleen associated with jaw and body morphotype. The size of the spleen is related with differences in resistance against infections. Differences in spleen size suggest immunological differences between fish morphotypes. Our results show signs of speciation based on morphology and suggest an involvement of the immune system in this diversification.

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