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Parasite escape through trophic specialization in a species flock

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Running title: Parasite escape through trophic specialization

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Abstract

Adaptive radiation occurs when species diversify rapidly to occupy an array of ecological niches. Since opportunities for parasite infection and transmission may greatly vary among these niches, adaptive radiation is expected to be associated with a turnover of the parasite community. As major agents of natural and sexual selection, parasites may play a central role in host diversification. The study of parasite turnover may thus be of general relevance and could significantly improve our understanding of adaptive radiation. In the present study, we examined the parasite faunas of eleven species belonging to the tribe Tropheini, one of several adaptive radiations of cichlid fishes in Lake Tanganyika. The most parsimonious ancestral foraging strategy among the Tropheini is relatively unselective substrate browsing of aufwuchs. Several lineages evolved more specialized foraging strategies, such as selective combing of microscopic diatoms or picking of macro-invertebrates. We found that representatives of these specialized lineages bear reduced infection with food-web transmitted acanthocephalan helminths, but not with parasites with a direct life cycle. Possibly, the evolution of selective foraging strategies entailed reduced ingestion of intermediate invertebrate hosts of acanthocephalans. We conclude that some species belonging to the Tropheini virtually escape acanthocephalan infection as a by-product effect of trophic specialization.

Keywords: Acanthocephala, adaptive radiation, fish, host-parasite interaction, Lake Tanganyika, Speciation

Introduction

Hosts and parasites can engage in long-lasting, antagonistic co-evolutionary relationships (Poulin, 2006; Decaestecker *et al.*, 2007). While such co-evolutionary battles could be won by either one of the parties, hosts may be able to 'escape' from the battlefield. For instance, hosts may escape from their parasites as a side-effect of range expansion, i.e. when they colonize areas that do not harbour their native parasite communities ('enemy release hypothesis'; Keane & Crawley, 2002). Alternatively, hosts may undergo *in situ* evolutionary parasite escape, e.g. as a side effect of changes in host traits that are directly or indirectly linked with parasite infection (Chew & Courtney, 1991).

However, host evolution may also lead to new parasite infections (Bell & Burt, 1991). In general, host evolution may thus lead to a turnover of the parasite community either through the loss or gain of parasite species (Knudsen *et al.*, 1996; MacColl, 2009a).

The likelihood of parasite infection is, aside from parasite infectivity and host susceptibility, often determined by exposure risk related to habitat use and trophic position. Many parasites have larval stages that infect the prey of their secondary or final host (Williams & Jones, 1994). Food-web transmission is therefore a prime infection route, especially for intestinal helminths. At the micro-evolutionary scale, trophic divergence has been shown to lead to predictable differences in parasite infection among recently diverged species (Karvonen & Seehausen, 2012) or phenotypes (Stutz *et al.*, 2014). Similarly, diet has been widely recognized as an important predictor of parasite infection at the macro-evolutionary scale (e.g. Drobney *et al.*, 1983; Bell & Burt, 1991; Vitone *et al.*, 2004; Valtonen *et al.*, 2010; Hayward *et al.*, *in press*). As such, trophic specialisation may lead to the gain of new parasite species through new prey species or other niche-related features (e.g. interactions with other hosts species) (Bell & Burt, 1991). Alternatively, hosts may escape from certain parasites by avoiding intermediate hosts as prey items, breaking up infection routes.

Diversification along dietary gradients occurs in many adaptive radiations including the iconic examples of lake whitefish (Kahilainen *et al.*, 2011), cichlids (Muschick *et al.*, 2012), Hawaiian honeycreepers (Freed *et al.*, 1987), Darwin's finches (Grant, 1999) or marsupials (Lee & Cockburn, 1985). Since parasites represent major agents of natural and sexual selection, parasite community turnover may have itself major consequences for local adaptation and species diversification (Eizaguirre *et al.*, 2009; MacColl, 2009b; Karvonen & Seehausen, 2012; Vanhove *et al.*, 2016). For instance, by intensifying divergent natural selection among populations, contrasting parasite communities may facilitate the build-up of reproductive isolation by reducing the viability and fecundity of hybrids or immigrants (Karvonen & Seehausen, 2012). In addition, contrasting parasite communities may contribute to reproductive isolation by intensifying sexual selection directly (Hamilton & Zuk, 1982; Taylor *et al.*, 1998; Maan *et al.*, 2008) or indirectly via pleiotropic effects of immunity traits (Eizaguirre *et al.*, 2009). Insights in changes of parasite infection upon host diversification are therefore of general relevance and could significantly improve our understanding of adaptive radiations.

The tribe Tropheini comprises one of several adaptive radiations of cichlid fishes from Lake Tanganyika, one of the Great East African Lakes. It currently includes 23 nominal species which occur mostly in sympatry on rocky outcrops in the littoral zone throughout the lake. Phylogenetic relationships among tropheine species are well resolved (Koblmüller *et al.* 2010; Fig. 1A) and the trophic and behavioural ecology of most species has been studied extensively (Kawanabe, Hori & Nagoshi 1997 and references therein). Tropheini diversified into four trophic-behavioural ecomorphs: pickers (preying on arthropods), suckers (molluscs), combers (diatoms attached to aufwuchs) and browsers (aufwuchs, mostly consisting of filamentous algae; Yamaoka 1997; Muschick *et al.* 2012; Tada *et al.* 2016). Considering the most complete phylogenetic tree of Tropheini (Koblmüller *et al.*, 2010), opportunistic browsing of aufwuchs is the most parsimonious ancestral state for the Tropheini radiation from which more specialized foraging strategies have evolved. With both specialized aufwuchs-feeders as well as more generalist species supplementing their algae-diet with insects, crustaceans, fish and fish eggs (Muschick *et al.*, 2012), browsers form the most heterogeneous group with respect to feeding ecology. Foraging strategies in the Tropheini have direct effects on many axes of diversification including morphology and the feeding apparatus (Kawanabe *et al.*, 1997; Muschick *et al.*, 2012), intestine length (Sturmbauer *et al.*, 1992; Wagner *et al.*, 2009; Tada *et al.*, 2016), territorial behaviour (Kawanabe *et al.*, 1997) and dispersal capacity (Wagner & McCune, 2009; Koblmüller *et al.*, 2010; Grégoir *et al.*, 2015; Vanhove *et al.*, 2015).

Evolutionary versatility of the feeding apparatus and novel ecological opportunities allowed species of the Tropheini to conquer new positions in the food-web. Adaptation to new trophic niches might be associated with gain or loss of particular parasites taxa, especially if they are food-web transmitted, as are many intestinal helminths. We therefore hypothesize that trophic specialization within the Tropheini radiation is accompanied by turnover of the intestinal parasite communities, and that these shifts are predictable based on trophic ecology. Specialization in foraging ecology among tropheine species has repeatedly and strongly been associated with evolutionary changes in intestine length (Sturmbauer *et al.*, 1992; Wagner *et al.*, 2009; Tada *et al.*, 2016). Pickers feed on readily digestible prey and, in line with the costly tissue hypothesis (Tsuboi *et al.*, 2016), evolved short intestines. In contrast, combers and some specialized browsers (both species of *Pseudosimochromis* Nelissen 1977 represented in our sampling) have long intestines that can digest low quality diet such as diatoms or filamentous algae. We test our hypothesis by relating intestine lengths (serving as a univariate proxy for diet) with the abundance of metazoan parasites in eleven species of Tropheini, covering all genera and ecomorphs of this species flock.

Material & Methods

Sampling and parasitological screening

Fish were collected in September 2011 and August 2012 at three locations (Cape Kanwankoko (2012): 8° 27' 8.0'' S, 30° 27' 20.0'' E, Muzumwa (2011): 8° 42' 5.7'' S, 31° 11' 59.8'' E and Toby's Place (2012): 8° 37' 18.9'' S, 31° 11' 59.9'' E) at the Zambian shoreline of Lake Tanganyika at a depth of 0.5 - 3 m (Fig. 1B). Eleven species of Tropheini cichlids encompassing all nine genera were sampled at one, two or three locations (sample sizes in Table 1). At all sites, the collected species occupy the same habitat and are commonly caught in the same net. Between capture and dissection, fish were kept in tanks filled with lake water for at least one night to empty their intestine. This procedure allows for a more reliable parasite count. Keeping fish up to three days in such tanks has little (slight increase for *Gyrodactylus*) or no effect (other parasite taxa) on parasite counts (Raeymaekers et al., 2013). Fish were euthanized with an overdose of MS222 prior to dissection, and measured to the nearest 0.1 mm (standard length). Intestines were screened for metazoan macro-parasites in the field using a stereomicroscope. The dissection of each fish started with the screening of its integument for monogeneans and crustaceans (copepods, branchiurans, isopods) and any kind of helminthic cyst. The mouth cavity was then inspected for parasitic isopods and branchiurans. Fish were inspected for gill parasites including branchiurans, copepods, bivalves, monogeneans, and any kind of helminthic cyst. To do so, the gills were immediately dissected and stored in 100% ethanol for later processing. Regarding endoparasites, fish were screened for monogeneans, digeneans, acanthocephalans, nematodes, and any kind of helminthic cysts. To do so, stomach, intestines, gall and urinary bladder were dissected immediately after euthanization of the host and inspected in a Petri dish with lake water. Processed fish were wrapped in cheese cloth, preserved on formalin and deposited at the Royal Museum for Central Africa (Tervuren, Belgium) as vouchers (samples 2011: collection MRAC B1.23; samples 2012: collection MRAC B2.38). Mean intestine length (as a proportion of host standard length) per species was obtained from Tada *et al.* (2016).

Statistical analyses

We used a generalized linear model (GLM) to describe the effects of host species, sampling site, host size (standard length) and host sex (male, female or immature) on abundance (count of parasite specimens per host individual) of acanthocephalan helminths, and the gill-infecting ectoparasite genera *Cichlidogyrus* Paperna 1960 (Monogenea, Ancyrocephalidae) and *Ergasilus* von Nordmann 1832 (Copepoda, Ergasilidae). Other parasites (Nematoda, Digenea and unidentified helminthic cysts) were found in the intestines in low numbers (overall prevalence < 0.05), preventing the application of statistical models. The effect of sampling year was confounded with the intrinsic effect of the site Muzumwa, and was therefore not included in the model. It has been shown earlier that parasite infection was relatively stable between the two sampling years in one host species (*T. moorii*; Raeymaekers *et al.*, 2013). To test for a site effect without confounding year effect, we ran the main effect GLM again without the data from Muzumwa. Since we were interested in how strong the species effect varies among sites, we ran the model a second time after adding a species x site interaction effect. Abundance was fit using a GLM assuming a Poisson distribution of parasite counts. Analysis of variance was conducted using type II sums of squares.

In order to test the association between parasite infection and intestine length, we first obtained data on mean intestine length (as a proportion of host standard length) from Tada *et al.* (2016). The median abundance of acanthocephalans, *Cichlidogyrus* or *Ergasilus* per host species and per site was then analysed in a second-order polynomial regression model including the linear and quadratic effect of mean intestine length. Sampling site was included as a random effect. Since the distribution of parasite counts was not normal (few host individuals had many parasites) and the relationship between Acanthocephala counts and intestine was right-tailed (Fig. 1D), both response and predictor variables were log-transformed prior to statistical analyses. We repeated the analysis with a reduced dataset from which all host species with less than 10 specimens (*Interochromis lookii* (Poll 1949), *Limnotilapia dardennii* (Boulenger 1899) and *Pseudosimochromis curvifrons* (Poll 1942)) were excluded in order to assess whether our analysis was sensitive for the limited sample size of some of the host species. Co-infection patterns were investigated using Pearsons's product-moment correlations (r) among median abundances of the three parasite groups. All analyses were conducted in R v.3.3.0 (R Development Core Team, 2011).

Phylogenetically related host species might have similar parasite communities. To test this assumption, we compared genetic divergence with parasite community differentiation using a Mantel test as implemented in the *mantel* function of the R-package *vegan* (Oksanen *et al.*, 2016). The genetic (Euclidean) distance matrix was based on species-level consensus AFLP (Amplified Fragment Length Polymorphism) genotypes (data from Koblmüller *et al.*, 2010), while the pairwise parasite community differences (Bray-Curtis dissimilarities) were based on median abundances of the different parasite groups.

Results

Parasites infecting every species included intestinal acanthocephalans, the monogenean gill parasite *Cichlidogyrus*, and the ectoparasitic copepod *Ergasilus* (Appendix S1 Fig. S1-S3; see Table 2 for relevant characteristics of these parasites). Parasites which were not present on every single host species included the gyrodactylid monogenean *Gyrodactylus* on skin and fins, intestinal nematodes, the monogenean *Urogyrus* in the urinary bladder, branchiurans in the gill cavity or on the opercula, intestinal digeneans, and a number of unidentified helminthic cysts in skin, fin or gill tissue. Acanthocephalans (11 host species; median abundance: 0-6.5; Fig. 1D) dominated the intestinal parasite fauna while nematodes (7 host species; median abundance: 0), digeneans (2 host species; median abundance: 0) and helminthic cysts (3 host species; median abundance: 0) were observed sporadically (Appendix S1 Table S1). Host species was a highly significant predictor for the abundance of the three main parasite groups (Table 3). Also significant, but of minor importance, were the effects of sampling site, host size, host sex and the interaction between host species and sampling site (except for the effects of host size on acanthocephalan abundance and host sex on *Ergasilus* abundance; Table 3). After excluding the site Muzumwa (visited in 2011), the site difference between Cape Kanwankoko and Toby's Place (visited in 2012) remained significant for Acanthocephala (χ^2=20.113, DF=1, P < 0.001) and *Ergasilus* (χ^2=25.14, DF=1, P < 0.001), but became non-significant for *Cichlidogyrus* (χ^2=1.26, DF=1, P = 0.262; Appendix S1 Table S3). This implies that the observed spatial and temporal variation in *Cichlidogyrus* infection might be confounded.

Intestine length significantly predicted median Acanthocephala abundance (Table 4). The association was curvilinear with species with short or long intestines bearing the lowest number of Acanthocephala (Table 4; Fig. 1D). The polynomial regression term remained significant after removing three host species with low sample sizes (p = 0.004). All pickers had very low numbers of Acanthocephala (median abundance: 0; Appendix S1 Fig. S1). Suckers and three genera with intermediate intestine length (*Limnotilapia* Regan 1920, *Simochromis* Boulenger 1898 and *Tropheus* Boulenger 1898) showed the highest Acanthocephala infection (median abundance: 4-6.5). Finally, the two browser species of the genus *Pseudosimochromis* with relatively long intestines as well as all combers were infected with low numbers of Acanthocephalans (median abundance: 0.5-3). In contrast, intestine length was not significantly correlated with *Cichlidogyrus* and *Ergasilus* counts (Table 4; Appendix S1 Fig. S4), and this lack of association did not change upon removal of three host species with low sample sizes (P = 0.579 and P = 0.184). We further found that there was no significant co-infection across host taxa (Acanthocephala vs. *Cichlidogyrus*: r = -0.143, P = 0.676;

Acanthocephala vs. *Ergasilus*: r = -0.340, P = 0.307; *Cichlidogyrus* vs. *Ergasilus*: r = 0.407, P = 0.214). There was also no correlation between phylogenetic divergence and parasite community differentiation (Mantel r = -0.009, p = 0.554).

Discussion

We hypothesized that diversification in foraging ecology in tropheine cichlids could be accompanied by shifts in parasite communities. Using the adaptive radiation of the Lake Tanganyika cichlid tribe Tropheini as a model, we found that the abundance of trophically transmitted acanthocephalan helminths was predicted by inter-specific variation in intestine length, which itself is strongly correlated with differentiation in foraging strategy and diet (Sturmbauer *et al.*, 1992; Wagner *et al.*, 2009; Tada *et al.*, 2016). In contrast, intestine length did not predict the abundance of ectoparasites with direct life cycles. We discuss to what extent trophic diversification and parasite infection are intertwined, and how this interplay might influence adaptive radiation.

Foraging ecology predicts parasite infection

Pickers and combers showed reduced infection levels with acanthocephalans relative to browsers (the ancestral foraging strategy) and suckers, suggesting that the evolution of specialised feeding strategies may entail parasite escape. Acanthocephalans exhibit a complex life-cycle with arthropods (commonly amphipods, ostracods or copepods (Williams & Jones, 1994)) serving as intermediate hosts (Table 2). Infection occurs upon ingestion of the intermediate host (Williams & Jones, 1994), which remains unidentified to date for Lake Tanganyika. Browsers (the most parsimonious ancestral ecomorph) shear filamentous algae *in toto* from the substrate (Yamaoka, 1997), along with the associated micro-invertebrate fauna which may include the intermediate host of acanthocephalans. We repeatedly observed ostracods in the intestines of several Tropheini species, making them candidate intermediate hosts for these acanthocephalans. However, the abundance of these substrate-bound arthropods was not recorded systematically and as such no association between ostracod ingestion and acanthocephalan infection could be verified. The grazing species *Petrochromis* spp. and *Interochromis loocki*, in contrast, are specialized diatom feeders who comb their food from filamentous algae (Yamaoka, 1997). They are able to selectively ingest tiny particles, thus excluding the putative intermediate host, which measures around 1 mm, while diatoms are in the range of 0.002 to 0.2 mm. The picker-lineage encompassing *'Ctenochromis' horei* (Günther 1894) and *'Gnathochromis' pfefferi* (Boulenger 1898) evolved into selective predators of insect larvae and larger crustaceans (e.g. shrimps (Muschick *et al.*, 2012)) that probably do not carry acanthocephalan larval stages. The sucker *Lobochilotes labiatus* (Boulenger 1898) mainly preys on molluscs (Colombo *et al.*, 2013), which are not known as hosts of Acanthocephala either (Williams & Jones, 1994). However, due to its sucking feeding behaviour, the species is (similar to browser species) relatively indiscriminate about the ingested items (Muschick *et al.*, 2012). Indeed, we found, on average, high infection with acanthocephalan parasites in *L. labiatus*. As such, our findings for browsers and suckers parallel the case of trematod infection in the herbivorous blue sea chub *Kyphosus cinerascens* (Forsskål, 1775), which ingests the gastropod intermediate host along with its plant diet (Huston *et al.*, 2016).

Our observation of a curvilinear relationship between intestine length and parasite abundance at the host species level contrasts with some conclusions of a recent study on parasite infection in 37 species of Lake Tanganyika cichlids (including ten of the eleven Tropheini species in the present study). Hayward *et al.* (*in press*) found that intestine length was correlated with parasite load within, but not between species, and that this correlation was positive and linear (i.e. individuals with longer intestines had more parasites). Our results highlight that strength and direction of relationships between host traits and parasite infection vary across different levels of biological organisation.

Also significant, but of minor importance, was the effect of geography on infection levels. Both intestinal and ectoparasites tended to be more abundant at Toby's Place and less abundant at Cape Kanwankoko. Geographical variation in parasite communities for two species of tropheine cichlids within the same study area has been reported earlier (*Tropheus moorii* Boulenger 1898 and *Simochromis diagramma* (Günther 1894); Raeymaekers *et al.*, 2013; Hablützel *et al.*, 2016). Such variation may reflect local dynamics of spatially independent parasite populations or differences in abundances of intermediate or alternative hosts. At the within-site level, we observed no relevant spatial segregation among host species as they all occur in the very same habitat (littoral zone of rocky outcrops) and were commonly caught in the same net. As such, our results likely reflect foraging ecology rather than spatial segregation at the microhabitat level. Microhabitat segregation in parasite communities is commonly found in sympatric or parapatric species pairs of Holarctic fish, such as in littoral and deep-water arctic char (*Salvelinus alpinus* (Linnaeus 1758); Knudsen *et al.*, 1997), and benthic and limnetic three-spined stickleback (*Gasterosteus aculeatus* Linnaeus 1758; MacColl, 2009a).

Ultimately, parasite load will not only be influenced by ecological (exposure to propagules) but also evolutionary (parasite virulence and host susceptibility) factors. We therefore tested for co-evolutionary legacies by comparing phylogenetic relationships with parasite community differentiation. This analysis revealed that closely related hosts did not harbour more similar parasite communities than distant hosts. Acanthocephalans are known to invoke immune reactions, although pathological effects are typically only observed upon mass infection (Paperna, 1996). As such they might engage in co-evolution with their hosts. Laboratory experiments provided empirical evidence for heritable variation in susceptibility to infection by acanthocephalans in three-spined sticklebacks (Mazzi & Bakker, 2003). This suggests that resistance might explain some of the variation in infection intensities among host species. Resistance to parasite infection may come at an immunological cost (Råberg *et al.*, 2009) that trades off against the parasite burden. Therefore, evolution may favour host tolerance over resistance if virulence is low. In at least one species of tropheine cichlids (*T. moorii*), Acanthocephala infection had little or no effect on host body condition, suggesting some degree of tolerance evolution (Hablützel *et al.*, 2014). However, the selective pressure for tolerance or resistance evolution might be expected to be itself related to exposure risk. Species suffering from high exposure should therefore experience the strongest selection pressure to become tolerant or resistant.

Parasite infection: an understudied dimension of adaptive radiation

Specialization in foraging ecology is one of the most prominent processes in adaptive radiations. Species divergence in this context is often considered a consequence of character displacement due to resource competition (Schluter, 1994). The observation that parasite infection is inherently intertwined with trophic diversification adds an understudied dimension to this process. Speciation models and field studies suggest that niche partitioning might be plastic at first and becomes heritable upon genetic divergence of the incipient species (Pfennig *et al.*, 2010). Parasite community turnover upon the hosts' ecological divergence might accelerate and stabilize this process in two ways. First, the cost of adaptation to new food sources might be compensated by parasite escape. Second, immunity gene pools might diverge among incipient host species (Eizaguirre *et al.*, 2012). Dietary versatility through phenotypic plasticity might become costly upon immunogenetic divergence, since neither of the diverging host lineages will be immunogenetically adapted to the parasite community that is associated with the alternative foraging strategy. As such, parasite community turnover may not only be a consequence, but may also play an active role in the diversification process among hosts.

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Table 1: Sampling sizes and median Acanthocephala abundance for eleven Tropheini species. A sampling size of zero for a fish species at Cape Kanwankoko and Muzumwa indicate that the species was very rare (but not necessarily absent) at the respective sampling site. A sampling size of zero for a fish at Toby's place reflect limited sampling efforts due to different initial study aims. Full names of species can be found in Fig. 1A.

	Sample size				**Median Acanthocephala abundance**			
Species	Cape Kanwankoko	Muzumwa	Toby's Place	all sites	Cape Kanwankoko	Muzumwa	Toby's Place	all sites
Ctho	5	6	10	21	0	0	0	0
Gnpf	5	5	10	20	0	0	0	0
Lida	0	6	0	6	–	6.5	–	6.5
Lola	5	6	10	21	0	7	9	4
Inlo	0	6	0	6	–	1	–	1
Pefam	5	7	0	12	0	1	–	0.5
Pefas	5	6	0	11	1	1.5	–	1
Psba	5	5	0	10	4	1	–	3
Pscu	6	0	0	6	0.5	–	–	0.5
Sidi	5	31	35	71	3	4	4	4
Trmo	5	89	86	180	7	5	4	4
Total	46	167	151	364				

Table 2: Relevant characteristics of the three most abundant parasite taxa in this study, and variation in abundance among host species and in response to the hosts' foraging strategies. References: 1) Williams & Jones (1994), 2) Paperna (1996), 3) Kilian (2012), 4) Vanhove *et al.* (2016), 5) Vanhove *et al.* (2015).

	Acanthocephala	***Cichlidogyrus***	***Ergasilus***
Life cycle	arthropod intermediate host[1]	simple[2]	simple[3]
Transmission	food-web[1]	direct[2]	direct[3]
Infected organ	intestine	gills	gills
Host-specificity	low[4]	high[5]	low[3]
Species diversity	low[4]	high[5]	low[3]

Table 3: Generalized linear model for the abundance of Acanthocephala, *Cichlidogyrus* and *Ergasilus* by host species, accounting for confounding effects of sampling site, host standard length (SL) and host sex.

Acanthocephala

Effect	χ^2	Num DF	Den DF	P
Species	308.89	10	149	< 0.001
Site	37.44	2	149	< 0.001
SL	1.54	1	149	0.214
Sex	16.89	2	149	< 0.001

Effect	χ^2	Num DF	Den DF	P
Species	308.89	10	149	< 0.001
Site	37.44	2	149	< 0.001
SL	0.57	1	149	0.449
Sex	29.93	2	149	< 0.001
Species x Site	81.76	11	149	< 0.001

Cichlidogyrus

Effect	χ^2	Num DF	Den DF	P
Species	5841.70	10	134	< 0.001
Site	394.91	2	134	< 0.001
SL	944.42	1	134	< 0.001
Sex	67.00	2	134	< 0.001

Effect	χ^2	Num DF	Den DF	P
Species	5841.70	10	134	< 0.001
Site	394.91	2	134	< 0.001
SL	592.77	1	134	< 0.001
Sex	30.55	2	134	< 0.001
Species x Site	356.72	11	134	< 0.001

Ergasilus

Effect	χ^2	Num DF	Den DF	P
Species	612.24	10	133	< 0.001
Site	108.71	2	133	< 0.001
SL	164.02	1	133	< 0.001
Sex	2.20	2	133	0.333

Effect	χ^2	Num DF	Den DF	P
Species	612.24	10	133	< 0.001
Site	108.71	2	133	< 0.001
SL	110.1	1	133	< 0.001
Sex	1.22	2	133	0.543
Species x Site	90.91	11	133	< 0.001

Table 4: Results of the general linear mixed-effect model for the relationship between intestine length (predictor) and median parasite count (response). Sampling site was included as a random effect. Wald χ^2-tests were used to assess the statistical significance of the linear model fit or of the improvement of the application of a second-order polynomial function, respectively.

Parasite	**Model**	**DF**	**AIC**	**BIC**	**logLik**	**Deviance**	**Test**	**χ^2**	**DF**	**P**
Acanthocephala	linear	6	128.0	135.0	-58.0	116.0	linear	0.25	1	0.615
	polynomial	10	117.6	129.4	-48.8	97.6	linear vs. polynomial	18.3	4	0.001
Cichlidogyrus	linear	6	225.8	232.8	-106.9	213.8	linear	0.74	1	0.390
	polynomial	10	231.0	242.8	-105.5	211.0	linear vs. polynomial	2.7	4	0.602
Ergasilus	linear	6	152.8	159.8	-70.4	140.8	linear	0.53	1	0.467
	polynomial	10	154.0	165.8	-67.0	134.0	linear vs. polynomial	6.7	4	0.150

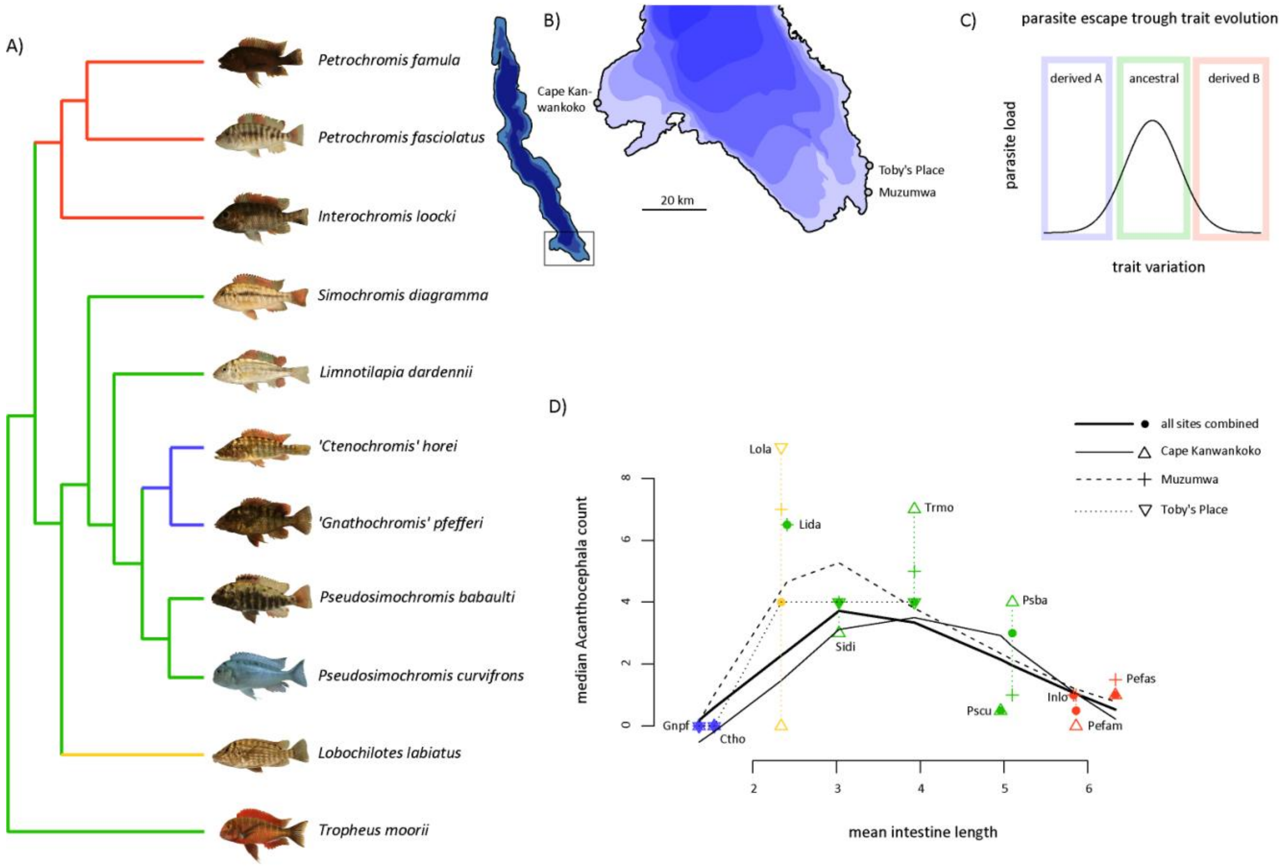

Fig. 1: **A)** Cladogram reflecting phylogenetic relationships following Koblmüller *et al.* (2010). Host ecomorph is coded as follows: green = browsers (the most parsimonious ancestral state), yellow = suckers, blue = pickers, red = combers. **B)** Sampling sites at the southern shore of Lake Tanganyika in Zambia. **C)** Illustration of theoretical framework of parasite escape through specialization. Hosts with ancestral trait state suffer from high parasite infection. Upon specialization, hosts are less exposed to parasite infection. In the present example, the trait is foraging behaviour with browsing as ancestral and sucking, picking or combing as derived states. **D)** The relative length of intestine (which directly relates to diet and hence foraging ecology) is highly predictive for the abundance of acanthocephalan helminths. Lines are lowess-functions. Species abbreviations include the first two (three) letters of the genus names and species epithets, respectively.