

Ligula intestinalis (Cestoda: Pseudophyllidea) infection of *Engraulicypris sardella* (Pisces: Cyprinidae) in Lake Malawi

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Abstract

Fish parasites have diverse ecological impacts on fish populations and are presently becoming useful as bio-indicators of fish migration and feeding. Studies of fish parasite in Lake Malawi severely lag behind the strides made in eco-evolutionary studies of its ichthyofauna. Over a period of eleven months from April 2003, we examined more than 1300 *Engraulicypris sardella* from Lake Malawi for cestode parasites. About 54 % of the fish had infections of between one and eight *Ligula intestinalis* plerocercoids. The infection occurred throughout the study period (mean prevalence $59 \pm 13\%$) but was most prevalent in December and June and was least prevalent in April. The majority of the hosts (78%) had a light parasite burden (one or two). Parasite prevalence increased with age of the fish. Prevalence and infection intensity oscillated closely with periods of peak abundance of zooplankton, the main food of *E. sardella* and intermediate hosts (copepods) of *L. intestinalis*. Increased exposure to the parasite with age aggravated by an ontogenetic diet shift from phytoplanktivory to zooplanktivory may be interacting with fluctuations in zooplankton production in driving the trophic transmission patterns of *L. intestinalis* in this ecosystem.

Key words: *Engraulicypris sardella*, *Ligula intestinalis*, trophic transmission, Lake Malawi

Introduction

Lake Malawi harbours one of the most diverse, recently evolved and endemic fish faunas in the world; a typical model of rapid adaptive radiation and speciation (Genner & Turner 2005). Many studies have elucidated the complex eco-evolutionary nature and dynamics of this lake's ichthyofauna (e.g. Fryer & Iles 1972, Konings 1990, Thompson & Allison 1997, Genner & Turner 2005) but few (e.g. Taylor et al. 1998) have attempted to unravel the ecological patterns and roles of fish parasites in this textbook model of aquatic diversity. This is surprising, given the ubiquity of freshwater fish parasite infections in Africa (see Fryer & Iles 1972, Barson & Marshall 2003, Dejen et al. 2006, Cowx et al. 2008) and the vital opportunity parasitism in fish presents for understanding host-parasite dynamics (Kennedy 1985). Fish parasites debilitate their hosts, depress their sexual display and reproductive abilities, disrupt predator-avoidance behaviour and may cause severe fish mortalities (Holmes & Bethel 1972, Taylor et al. 1998, Shields 2002, Piasecki et al. 2004). Recently, fish parasites have become useful for fish-stock identification and as bio-indicators of fish feeding and migratory patterns (Moore et al. 2003, Knudsen et al. 2005).

Engraulicypris sardella (Günther, 1868), locally known as Usipa, is an endemic cyprinid species of Lake Malawi (van Lissa 1982, Konings 1990) on which most pelagic piscivorous fishes (mainly *Diplotaxodon* spp. and *Rhamphochromis* spp.) depend for the bulk of their diet (Allison et al. 1996). Although typically zooplanktivorous (Degnbol 1982, Konings 1990), *E. sardella* also feeds on phytoplankton when young (<40mm long) (Degnbol 1982, Allison et al. 1996, Thompson & Irvine 1997). It is therefore a vital ecological link between top predators and lower trophic levels in this ecosystem. The fish spawns throughout the year,

but chiefly in the rainy season)(Thompson 1996, Maguza-Tembo et al, 2009), maturing at about 70-75mm and growing up to 135 mm total length (van Lissa 1982, Thompson & Allison 1997). Larvae and juveniles of up to 30mm long live in the open waters of the lake, after which size they migrate inshore (Konings 1990) and recruit into the artisanal ‘Chirimila’ fishery (Thompson & Allison 1997).

Engraulicypris sardella is an important food fish in Malawi (Konings 1990, Thompson 1996), contributing up to 58% of the nation’s traditional seine net catches in some years (Fisheries Department 1999). The catch is often spread out to dry in the sun and huge numbers of live *Ligula* plerocercoids can be seen emerging from the fish, which can reduce the marketability of the product (Kanyerere, pers.comm). Very little is known about this *Ligula* infection. The objective of this study was to obtain preliminary data on prevalence and intensity of *Ligula* infection in *E. sardella* in southern Lake Malawi.

Materials and methods

A total of 1345 *E. sardella* (mean length 93.31 ± 0.65 mm, range: 35 -135mm) were sampled from ‘Chirimila’ catches at fishing beaches from April 2003 to February 2004 at Monkey Bay, Lake Malawi (Fig. 1). The fish were obtained early in the morning immediately upon landing of the previous night’s catch and kept in a freezer. Undissected fish total wet weight (nearest 0.01 g) and length (nearest mm) were determined on thawed tissue-mopped (to remove excess water) samples. After dissection, the number, individual length (nearest mm) and total wet weight (nearest 0.01 g) of *Ligula* plerocercoids per fish were recorded. Identity of the parasites was confirmed using Meyer & Olsen (1975). Parasite index (PI), a measure of the severity of infection (Bagamian et al. 2004) was established for each infected fish according to Barson & Marshall (2003). Fish ages were estimated using the growth formula for *E. sardella* in the same area as derived by Ruffli & van Lissa (1982) and confirmed by Maguza-Tembo et al (2009).

Data analysis was carried out using statistical package Palaeontological Statistics (Hammer et al. 2001). After Shapiro-Wilk tests for data normality, Kruskal-Wallis and Mann-Whitney pairwise comparison tests (with Bonferroni-corrected p values) were used for assessing differences in means. Correlations were done on *Ligula* parasite prevalence against fish size classes. Variations about the mean are expressed as 95% confidence intervals.

Results

The oldest and youngest *E. sardella* sampled were about 76 weeks and two weeks of age, respectively (mean age: 19.5 ± 0.3 weeks). Most of the fish (about 90%) sampled throughout the study were equal to or less than 30 weeks old (Fig. 2). About 54% of *E. sardella* sampled were infected with *Ligula intestinalis* plerocercoids. The smallest and biggest infected fish were 72mm (about 11 weeks old) and 135mm long (about 76 weeks old), respectively. Parasite prevalence ranged from 33% to 83% (mean \pm SE: $59 \pm 13\%$) (Fig. 3A). It was highest in June and December (83%) and lowest in April (33%). Fish size (as age class) was positively correlated with prevalence ($r = 0.87$, $p < 0.01$)(Fig. 4).

Parasite intensity varied significantly across the months (Kuskal-Wallis test, $p < 0.0001$). It was lower in April than in November ($p < 0.0001$), December ($p < 0.01$) and January ($p < 0.00001$), lower in May than in November ($p < 0.00001$), December ($p < 0.001$) and January ($p < 0.000001$), lower in June than in November ($p < 0.01$) and January ($p < 0.001$), lower in July than in November ($p < 0.01$), December ($p < 0.05$) and January ($p < 0.001$), lower in August than in January ($p < 0.05$) and was also lower in October than in November ($p < 0.05$) and January ($p < 0.01$) (Fig. 3B). Only about one fifth (22%) of the infected fish were heavily parasitized with three or more plerocercoids while the majority (78%) had a light worm burden of one or two. The longest plerocercoid (320 mm) was almost thrice the size of its host fish (120mm long). Difference in size between the shortest and longest parasite in the same host ranged from 5 to 275 mm. The parasite index was highest in November (14.05 ± 1.95 , $n = 60$) and lowest in February (0.42 ± 0.13 , $n = 60$) but generally fluctuated between 6.11 ± 1.45 and 9.82 ± 1.90 (Fig. 3C). Infected fish were older (149.6 ± 3.1 days, range: 75.4 – 530.9, $n = 725$) than uninfected ones (120.6 ± 3.2 days, range: 13.0 – 530.9, $n = 617$)

($p < 0.0001$)

Discussion

Infection of *E. sardella* by *L. intestinalis* occurred throughout the year, with marked temporal variations. This is in accord with *Ligula* infection patterns of other cyprinid species across Africa (e.g. Barson & Marshall 2003, Cowx et al. 2008). Most helminth parasites utilise the food chain for their transmission (Marcogliese 1995, Piasecki et al. 2004, Bagamian et al. 2004, Knudsen et al., 2005, Dejen et al. 2006). Dubinina (1980) and Loot et al. (2006), among many authorities, describe how this is achieved by *L. intestinalis*. Its coracidia larval stages are free-swimming and get ingested by the first intermediate host, a planktonic copepod. In the copepod the parasite develops into a proceroid form. When infected copepods are eaten by fish the proceroid larvae develop into plerocercoids in the fish's abdominal cavity. The definitive host is a piscivorous bird where the parasites mature and lay eggs that are later shed into the water through bird faeces. The main diet of adult *E. sardella* in Lake Malawi consists of zooplankton *Mesocyclops* sp. and *Cyclops* sp. (Degnbol 1982, Van Lissa 1982, Allison et al. 1996). These copepod genera are intermediate hosts of *L. intestinalis* (Meyer & Olsen 1975, Piasecki et al. 2004). Trophic transmission may be the most likely route of *Ligula* into *E. sardella*.

Zooplankton abundance in Lake Malawi fluctuates seasonally (Twombly 1983, Irvine & waya 1999) in direct response to increased phytoplankton production due to intensified lake mixing and available nutrient supply in June–July (Patterson & Kachinjika, 1993). Maximum and minimum zooplankton abundances (including those of *Mesocyclops aequatorialis aequatorialis* and *Thermocyclops neglectus*) occur in July and early months of the year (December-February), respectively (Twombly 1983, Irvine & waya 1999). The prevalence of cestodes in their copepod hosts fluctuates seasonally as a function of encounter rates determined by zooplankton abundance (Marcogliese 1995, Loot et al. 2006). This variation in the availability of infected copepod hosts that are food for fish is among the most frequent causes of fluctuations in prevalence and abundance of *Ligula* infections (Dejen et al., 2006, Loot et al. 2006). Although the exact nature of correspondence between its copepod intermediate host abundance and *Ligula*'s prevalence in *E. sardella* is not very clear here, the period of high parasite prevalence (October-December) and high parasite index (November) obtained in this study both occur following a period of maximum abundance of zooplankton (July-October) (Twombly 1983, Irvine & waya. 1999). The seasonal variation in *Ligula* prevalence may probably be a combined effect of temporal changes in the availability of their intermediate hosts (copepods) coupled with variation of cestode parasites themselves in their zooplankton intermediate hosts.

The present study revealed ontogenetic differences in *L. intestinalis* infection. As juveniles, *E. sardella* feeds on both phytoplankton and zooplankton but switches to a zooplanktivorous diet from a size of 40mm onwards (Degnbol 1982, Van Lissa 1982, Thompson & Irvine 1997), adopting a diet dominated by *Mesocyclops* sp. and *Cyclops* sp. (Degnbol 1982, Van Lissa 1982, Allison et al. 1996). These copepods are intermediate hosts of *L. intestinalis* (Meyer & Olsen 1975, Piasecki et al. 2004). In host-parasite systems, where transfer of parasites between two intermediate hosts largely depends on ingestion of one host by another (Marcogliese 1995, Piasecki et al. 2004), such a diet shift (to zooplanktivory) may increase the fish's exposure to parasites and thus the propensity for infection (Dogiel 1961, Kennedy 1985, Dejen et al., 2006, Cowx et al. 2008). Size-related exponential increases of *L. intestinalis* prevalence due to diet switching to copepods have also been documented in other cyprinids in Lakes Victoria and Tana (Dejen et al., 2006, Cowx et al. 2008).

Fish parasitism may depress fish's marketability (Piasecki et al. 2004) and, in the case of *L. intestinalis*, the infection may significantly reduce the fecundity of hosts (Marcogliese 1995, Bagamian et al. 2004) and thus

affect their reproductive output (Coxw et al. 2008). *Engraulicypris sardella* is known for its occasionally wild inter-annual recruitment variations (Thompson 1996, Thompson & Allison 1997) and its fast growing short-life history pattern (van Lissa 1982, Thompson & Bulirani, 1993, Thompson & Allison 1997, Maguza-Tembo et al, 2009). None of the *E. sardella* in this study exceeded 76 weeks of age. The potential effect of parasitism on such a life cycle can not therefore be totally ruled out. More, longer and wide-covering studies of *Ligula* infection of *E. sardella* in this lake are necessary to elucidate the potential impacts of this parasitism on the productivity of such an important food fish.

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References

- Allison E.H., Irvine K., Thompson A.B., Ngatunga B.P. (1996). Diets and food consumption rates of pelagic fish in Lake Malawi, Africa. *Freshwater Biology* 35: 489-515
- Bagamian K.H., Heins D.C., Baker J.A. (2004). Body condition and reproductive capacity of three-spined stickleback infected with the cestode *Schistocephalus solidus*. *Journal of Fish Biology* 64: 1568–1576
- Barson M., Marshall B.E. (2003). The occurrence of the tapeworm *Ligula intestinalis* (L.) in *Barbus paludinosus* from a small dam in Zimbabwe. *African Journal of Aquatic Science* 28(2): 175–178
- Coxw I.G., Rollings D., Tumwebaze R. (2008). Effect of *Ligula intestinalis* on the reproductive capacity of *Rastrineobola argentea* in Lake Victoria. *Journal of Fish Biology* 73: 2249–2260
- Degnbol P. (1982). Food habits of larval *Engraulicypris sardella* in Lake Malawi: Diurnal feeding pattern and comparison of size groups, pp 57-67. In: FAO Fishery Expansion Project, Malawi; Biological studies on the pelagic ecosystem of Lake Malawi. Rome, FI: DP/MLW/75/019, Technical Report 1.
- Dejen E., Vijverberg J., Sibbing F.A. (2006). Spatial and temporal variation of cestode infection and its effects on two small barbs (*Barbus humilis* and *B. tanapelagus*) in Lake Tana, Ethiopia. *Hydrobiologia* 556:109–117
- Dogiel V. A. (1961). Ecology of the parasites of freshwater fishes, pp 1-47. In: Dogiel V.A., Petrushevski G.K., Polyanski Y. I. (eds.). *Parasitology of Fishes*. Oliver & Boyd, Edinburgh & London
- Dubinina M. N. (1980). Tapeworm Cestoda Ligulidae of the fauna of the USSR. Springfield, IL: US Department of Commerce National Technical Information Service.
- Fisheries Department (1999). Fish Stocks and Fisheries of Malawian Waters: Resource Report 1999. Malawi Government Fisheries Bulletin No. 39, Lilongwe, Malawi.
- Fryer G, Iles T.D. (1972). The Cichlid Fishes of the Great Lakes of Africa. Oliver & Boyd, Edinburgh, UK
- Genner M.J., Turner G.F. (2005). The mbuna cichlids of Lake Malawi: a model for rapid speciation and adaptive radiation. *Fish and Fisheries* 6: 1–34
- Hammer Ø., Harper D.A.T., Ryan P.D. (2001). PAST: Paleontological Statistics Software Package for Education and Data Analysis. *Palaeontologia Electronica* 4(1): 9pp.
- Holmes J.C., Bethel W.M. (1972). Modification of intermediate host behaviour by parasites. pp 123-149 In: Canning EU & Wright CA (Eds.). *Behavioural aspects of parasite transmission*, Linnean society of London-Academic Press, London.

- Irvine K., Waya R. (1999). Spatial and temporal patterns of zooplankton standing biomass and production in Lake Malawi. *Hydrobiologia* 407: 191–205, 1999.
- Kennedy C.R. (1985). Interactions of fish and parasite populations: to perpetuate or pioneer? pp. 1-20 In: Rollinson D., Anderson R.M. (eds.). *Ecology and genetics of host-parasite interactions*. The Linnean Society Symposium Series 11, Academic Press, London.
- Knudsen R., Rikardsen A.H., Dempson J.B., Bjørn P.A., Finstad B., Holm M., Amundsen P.A. (2005). Trophically transmitted parasites in wild Atlantic salmon post-smolts from Norwegian fjords. *Journal of Fish Biology* 66: 758–772
- Konings A. (1990). *Cichlids and all the other fishes of Lake Malawi*. T.F.H. Publications, Neptune, New Jersey
- Loot G., Young-seuk P., Lek S., Brosse S. (2006). Encounter rate between local populations shapes host selection in complex parasite life cycle. *Biological Journal of the Linnean Society* 89: 99–106.
- Maguza-Tembo F., Palsson O.K. Msiska O.V. (2009). Growth and exploitation of *Engraulicypris sardella* in the light attraction fishery of Southern Lake Malawi. *Malawi Journal of Aquaculture and Fisheries* 1: 6-12
- Marcogliese D.J. (1995). The role of zooplankton in the transmission of helminth parasites to fish. *Reviews in Fish Biology and Fisheries* 5: 336-371
- Meyer M.C, Olsen O.W. (1975). *Essentials of Parasitology* 2nd Edition. Wm. C. Brown Company Publishers, Dubuque, Iowa, USA.
- Moore B.R., Buckworth R.C., Moss H., Lester R.J.G. (2003). Stock discrimination and movements of narrow-barred Spanish mackerel across northern Australia as indicated by parasites. *Journal of Fish Biology* 63: 765–779
- Piasecki W., Goodwin A.E., Eiras J.C., Nowak B.F. (2004) Importance of Copepoda in freshwater aquaculture. *Zoological Studies* 43: 193–205.
- Patterson, G., Kachinjika O. (1993). Effect of wind-induced mixing on the vertical distribution of nutrient and phytoplankton in Lake Malawi. *Verh. int. Ver. Limnol.* 25: 872–876.
- Rufli H. van Lissa J.(1982). Age and growth of *Engraulicypris sardella* in Lake Malawi. pp 85-97. In: *Biological studies on the pelagic ecosystem of Lake Malawi*. FAO Technical Report 1. FI:DP/MLW/75/019.
- Shileds B.A., Groves K.L., Rombaugh C., Bellmore R. (2002). Ligulosis associated with mortality in largescale suckers. *Journal of Fish Biology* 61: 448 – 455
- Taylor M.I., Turner G.F., Robinson R.L., Stauffer J.R. Jnr. (1998). Sexual selection, parasites and bower height skew in a bower-building cichlid fish. *Animal Behaviour* 56: 379–384
- Thompson, A.B. (1996). Early life-history of *Engraulicypris sardella* (Cyprinidae) in Lake Malawi/Niassa. *J. Plankton Res.*, 18,1349-1368.
- Thompson A.B., Allison E.H. (1997). Potential Yield estimates of unexploited pelagic fish stocks in lake Malawi. *Fisheries Management and Ecology* 4:31-48
- Thompson A.B., Bulirani A. (1993). Growth of usipa (*Engraulicypris sardella*) in Lake Malawi/Niassa. In Marshall,B.E., Mubamba,R. (eds), *Symposium on Biology, Stock Assessment and Exploitation of Small Pelagic Fish Species in the African Great Lakes Region*. FAO, Rome. C1FA Occ Pap., 19,87-99.
- Thompson A.B., Irvine K. (1997). Diet-shifts and food-dependent survival in *Engraulicypris sardella* (Cyprinidae) larvae from Lake Malawi, *Africa Journal of Plankton Research* 19 (3): 287-301 Twombly S. (1983).
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Van Lissa J. (1982). Food habits of *Engraulicypris sardella* from along the shoreline of Lake Malawi, pp 98-108. In: FAO. Fishery Expansion Project, Malawi. Biological studies on the pelagic ecosystem of Lake Malawi. Rome, FI:DP/MLW/75/019, Technical Report 1.

Figures

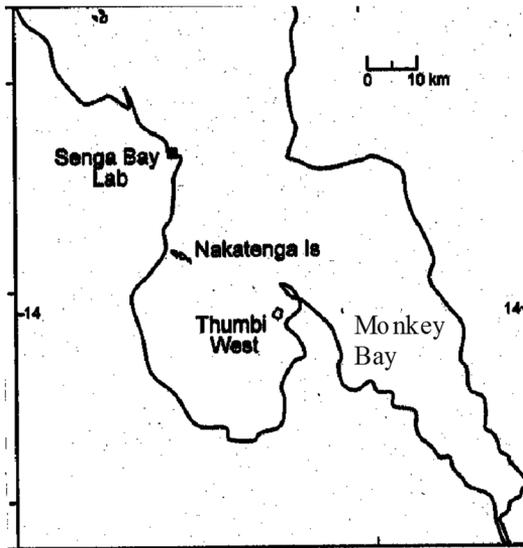


Figure 1: Map of southern Lake Malawi, showing the location of Monkey Bay, the sampling site of *E. sardella* in this study

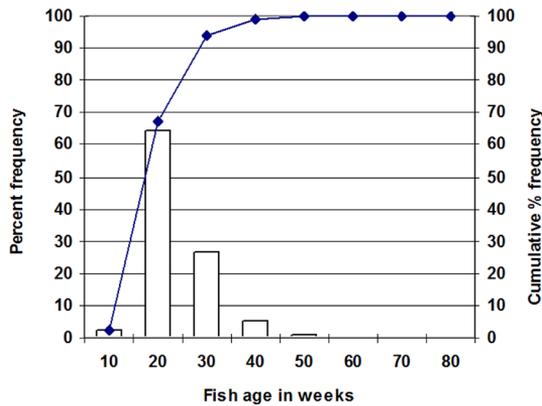


Figure 2: Age frequency distribution of *E. sardella* in southern Lake Malawi (n=1343)

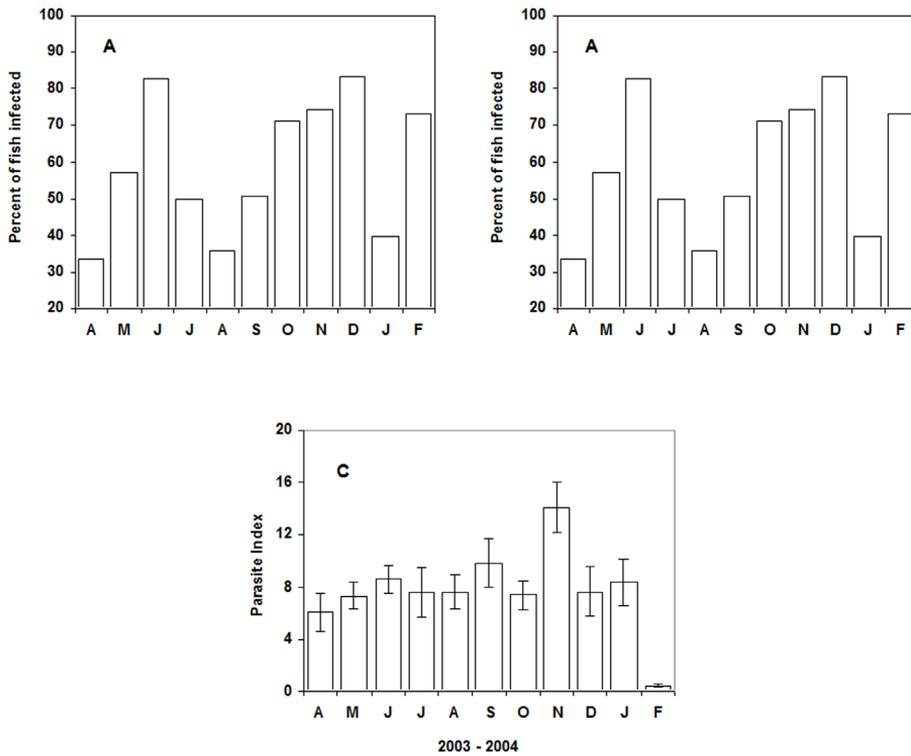


Figure 3: Monthly variation in prevalence (A), intensity (B) and parasite index (C) of a *Ligula* infection in *E. sardella* from southern Lake Malawi.

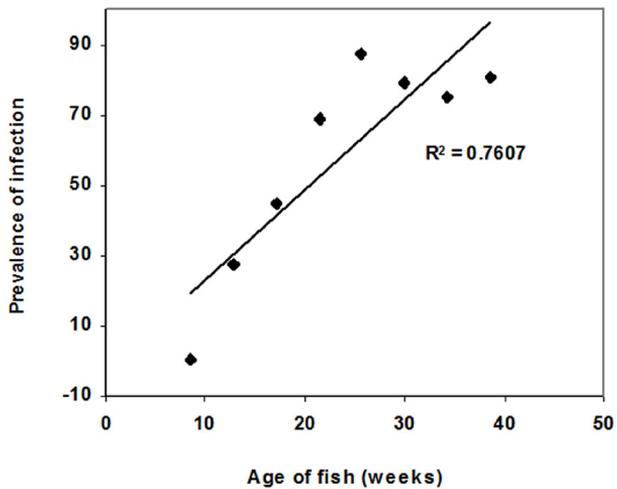


Figure 4: The relationship between *Ligula* infection prevalence and fish age in *E. sardella* in southern Lake Malawi