Infestation and Pathological Alterations by *Ergasilus sarsi* (Copepoda) on the Tanganyika Killifish from Africa

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Abstract

A total of 204 *Ergasilus sarsi*, a copepod, were collected from Tanganyika Killifish *Lamprichthys tanganicanus* in Lake Tanganyika during March 2010. The prevalence was 86.40%, the mean intensity was 7.56, and the mean abundance was 6.38. Only 27 of the fish were infested, and the highest infestation on one fish was 29. Proliferation of mucus cells and lamellar fusion occurred. Haemorrhage due to blood vessel compression was noted. This is the first record of *E. sarsi* from Tanganyika Killifish. This study is also the first to provide a description of the pathological alterations caused by *E. sarsi*.

During an opportunity to survey in Lake Tanganyika in March (summer) 2010, the copepod *Ergasilus sarsi* was collected from Tanganyika Killifish *Lamprichthys tanganicanus*, a benthopelagic, nonmigratory fish that is endemic to the shore regions of Lake Tanganyika (Huber 1996). The fish is characterized by the intense iridescent blue color and adults reach approximately 10 cm TL rendering the species a potential candidate for aquaculture; as such, it has been introduced into various parts of the world.

*Ergasilus* copepods occur widely distributed throughout Africa; they are nonhost-specific fish parasites (Wilson 1911; Oldewage and van As 1987) inhabiting fresh, marine, and brackish water (Boxshall and Defaye 2008). Four *Ergasilus* species occur in Lake Tanganyika *E. flaccidus*, *E. kandti*, *E. megacheir*, and *E. sarsi* Capart, 1944. Only females are parasitic and they attach with large modified second antennae. Although attachment and feeding may cause extensive pathological change to the gills, some *Ergasilus* species simply embrace the gill filament (Oldewage and van As 1987) while others (e.g., *E. colomeus*) insert the third segment of the antennae into the host gill filament (Thatcher and Boeger 1983), causing terminal necrosis of the filament (Thatcher 2006).

Furthermore, *Ergasilus* feed on gill tissue consisting of blood, mucus and gill epithelium (Einzsporn 1965). The parasite feed regularly in order to sustain the energy levels required for the parasitic lifestyle (Oldewage and van As 1987) and feeding involves secretion of proteolytic enzymes that aids in external digestion (Paperna 1996).

Attachment results in compression of gill tissue (Oldewage and van As 1987). Furthermore, lesions may become secondarily infested by bacteria, fungi, and virus growth and an inflammatory response marked by an increase in Rodlet and mucus cells (Dezfuli et al. 2003; Roubal 1989). Compression of the blood vessels causes hypoxia, and high intensities may impair growth (Dezfuli et al. 2011) and cause death (Hoffman 1977; Abdelhalim 1990).

Each of the four *Ergasilus* species from Lake Tanganyika attacks to a different site on the host and the pathological...
lateration varies accordingly. Fryer (1965) reported that *E. kandti* cause considerable damage to the gill tissue and its antennae become overgrown by the gill tissue and that *E. megacheir* causes gill compression and a visible indent in the gill filament. However, Fryer did not describe the pathological effects by *E. sarsi*.

The aim of this study was to comment on attachment of *E. sarsi* and to describe the pathological alterations caused by attachment and feeding.

**METHODS**

In March 2010 a total of 32 Tanganyika Killifish were collected with hand nets at the shore of Lake Tanganyika in the Democratic Republic of the Congo at three study sites: Kisokwe (4°14′31″ S, 29°10′35″ E), Mufazi (7°05′12″ S, 29°54′45″ E), and Mugayo (6°46′51″ S, 29°33′42″ E) on the North Western shore.

The fish were killed immediately after collection by severing the spinal cord, the gills were removed, and parasite specimens were fixed intact on the gills in acetoformaldehyde alcohol (AFA) solution and preserved in 70% ethanol. The gill samples were later examined for ectoparasites using a dissection microscope. The position of attachment was recorded according to the regions suggested by Gelnar (1987), and data were subjected to statistical analysis (Pearson's chi-square test) to compare attachment to the left gill and right gill arches. Differences between the dorsal, ventral, and medial attachment, as well as between distal, proximal, and central regions were also tested. Prevalence, mean intensity and abundance were calculated as suggested by Bush et al. (1997).

*Ergasilus* specimens for scanning electron microscopy were hydrated, freeze dried, and sputter-coated with gold and studied with a JEOL 5600 Scanning Electron Microscope.

Tissue samples for histology were dehydrated, infiltrated with resin, and 5-µm serial sections were made. Sections were stained with a Heidenhains AZAN trichrome stain (Humason 1979) and studied and micrographed.

**RESULTS**

Infestation Statistics

The prevalence at Mugayo was 90%, the mean intensity was 6.9, and the abundance was 6.5; at Kisokwe there was 100% prevalence, mean intensity was 18, and an abundance was 18; at Mufazi (near Momba) prevalence was 70%, mean intensity was 7.8 and abundance was 5.6. The overall mean prevalence was therefore 86.40%, the mean intensity was 7.56, and the mean abundance was 6.38. The 204 parasites were unevenly distributed between the 27 infested hosts. The highest intensity was 29.

Pearson’s chi-square test was used to compare attachment on left or right gill arches, and attachment to dorsal, ventral, and medial areas as well as between distal, proximal, and central regions. Results for preference for either gill arch indicated that *E. sarsi* do not have a preference for either one of the gill archers (*P* = 0.12).

Related to the distribution of *E. sarsi* on the gill arch (dorsal, median, ventral, distal, central and proximal), the Pearson’s chi-square results indicated a significant difference (*P* = 0.0005) between the dorsal, median and ventral; however, no significant difference (*P* = 1.19) occurred for distal, central, and proximal (Figure 1). Furthermore, an equal number of parasites occur on the four gill arches (*P* = 7.88), gill-arch 2 having the most parasites (Figure 2).

The results indicate an unequal distribution of parasites between all the attachment sites. However, the distribution between the left and right sides of the host is equal. The sample size was small.

Pathological Alterations

Histological examinations of Tanganyika Killifish gill tissue infested with *E. sarsi* showed that mature females attach only to the tips of the primary lamellae, confirming Fryer’s (1965) observation. The second antennae are modified into claw-like structures, and these are inserted into the gill tissue (Figures 3A, B, 4A).

![Figure 1](image1.png)  
**FIGURE 1.** Bar graphs illustrating (A) the number (*n*) of *E. sarsi* parasites on Tanganyika Killifish along the long- axis of the gill arch, and (B) the distribution of parasites along the short-axis attachment sites.
Following attachment and feeding erosion of the gill superficial epidermal tissue occurred adjacent to the second antennae, maxillipeds, and swimming legs (Figure 4B), and eventually the tip of the filament is lost (compare Figures 4B and 5A). Increased mucus secretion occurs and the parasite and epithelium becomes covered by it. An increase in the number of Mast and Rodlet cells occurs in the vicinity of the parasite (Figure 5B). The maxillipeds and legs scrape pieces of tissue off to expose and break the underlying blood vessels, thereby causing bleeding (Figure 5B). Gill epithelium, mucus strands, and extra vesicular blood cells occur in the area surrounding the maxillipeds (Figure 4A). These tissues are also present in the lumen of the buccal cavity and in the intestine of the parasite (Figure 6A). Epithelial hyperplasia occurs along the length of the gill filament and results in fusion of gill lamellae (Figure 6B). The head region of the parasite becomes embedded in the proliferation.

Scanning electron micrographs corroborated the observations from the histological sections. The anterior end of the parasite becomes embedded in the gill filament (Figure 7A). Copious amounts of mucus are excreted onto the cell surface adjacent to the parasite’s attachment site as well as the gill filament in close proximity. Figure 7B shows the swimming legs of the parasite that is responsible for some of the pathological alterations. The scraping action of the swimming legs removes gill tissue off the gill filament and pushes it towards the mouth.

**DISCUSSION**

Although *E. sarsi* was previously reported from Lake Tanganyika (Sars 1909; Cunnington 1920; Capart 1944; Fryer 1965), this is the first report for this host, the Tanganyika Killifish, and of the pathological alterations associated with it. Fryer (1965) observed that a correlation exists between the attachment site of various *Ergasilus* species and pathological alterations elicited; he also noted that different species attach to different regions, even on the same host. *E. sarsi* bears long antennae that lack spines enabling it to wrap around a gill filament and predominantly attach to the tip of the gill filament where the filament is narrower. This leads to severe pathological alteration of the gill filaments. Changes were noticed on the gill surface along the entire length of the parasite’s body even to the position where the swimming legs and egg sacks occur. Gill fusion and lamellar lifting occurred and this impacts the function of the gill in osmoregulation, as well as respiratory gas exchange. Furthermore, host tissue was scraped of in the vicinity of the appendages indicating that traumatic alterations are not limited to the action of the mouth parts but that all appendages participate. Host cells were observed in the intestine of the parasite, indicating that the parasite actively feed on the host, consuming gill tissue as well as blood.

The presence of Rodlet cells indicate that an immune response was elicited, confirming an observation by Dezfuli et al. (2003, 2011) in *Ergasilus sieboldi* on Bream *Abramis brama*. The inflammatory response occurred along the length of the
FIGURE 4. Photomicrographs of gill tissue of Tanganyika Killifish in close proximity to *E. sarsi* to show the histology (stained with AZAN). Micrographs showing (A) the pathological alterations caused by attachment; where the parasite maxiliped is inserted into the gill tissue (white arrows), and (B) a ruptured blood vessel (white arrow), haemorrhage (black arrow), tissue erosion (striped arrow), and mucus strand and blood cells (colorless arrow). Scale bars represent 100 µm. [Figure available online in color.]

FIGURE 5. Photomicrograph of (A) noneroded gill epithelium of Tanganyika Killifish, the circle shows the tissue that becomes eroded (compare with B; white arrow points to epithelium lifting), and (B) the inflammatory response of the host; arrows show Rodlet cells and mast cells. Scale bars represent 100 µm. [Figure available online in color.]

FIGURE 6. (A) A cross section through *E. sarsi* (white arrow) in the vicinity of the buccal cavity. Red blood cells are present in the oesophagus and gill tissue consisting of blood cells and tissue are present in the circle adjacent to the buccal cavity. (B) Micrograph showing secondary lamellar fusion (circle) and parasite egg sacks (white arrow). Scale bars represent 100 µm. [Figure available online in color.]
parasite. Furthermore, increased mucus secretion was also observed. This will cause hypoxia due to obstruction of gas exchange. This effect is enhanced by high temperature in the tropics, which causes a reduction of the oxygen binding ability of water, creating additional stress to the host fish.

The parasites occur mostly on the second gill arch, which is also the arch that receives the strongest water current, an advantageous locality for distribution of the parasite’s eggs in the surrounding water column.

According to Butler (2013) Tanganyika Killifish require very specific breeding conditions and therefore it is challenging to breed in captivity, but Butler mentions, nevertheless, that the fish provides great enjoyment in a tropical tank because of its beauty. Unconfirmed reports indicate that this fish is exported commercially. *E. sarsi* is not a species-specific parasite (Oldewage and Avenant-Oldewage 1993) and indigenous fishes in importing countries may therefore become infested unintentionally. Future studies should consider the effect of quarantine measures and treatment prior to export.

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