PRELIMINARY STUDY OF THE PREVALENCE OF HELMINTHS AND THEIR ASSOCIATED PATHOLOGICAL LESIONS IN FOUR FISH SPECIES FROM RIVER TANA


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ETUDE PRELIMINAIRES DE LA PREVALENCE DES HELMINTHES ET DE LEURS LESIONS PATHOLOGIQUES CHEZ QUATRE ESPECES DE POISSON DE LA RIVIÈRE TANA

Résumé

Une étude préliminaire a été menée entre janvier et mai 2006 pour enquêter sur la prévalence des helmintes et leurs lésions pathologiques chez quatre espèces de poisson. Au total, 43 poissons frais de la rivière Tana, vendus au marché de Gikomba, ont été achetés et autopsiés. Ces poissons étaient 15/43 (34.9%) de l'espèce Oreochromis, 11/43 (25.6%) Clarias spp., 10/43 (23.2%) Cyprinus carpio et 7/43 (16.3%) Barbus spp. À l'autopsie, on a trouvé plusieurs vers Contracecum au troisième stade larvaire dans la cavité abdominale, les muscles et derrière les branchies. Au total, 91% des poissons-chats (Clarias spp.) et 20% des tilapias (Oreochromis spp.) avaient une infestation «modérée à grave» de vers Contracecum. L'intensité de l'infestation variait entre 1 et 593 helmintes par poisson, ce qui a provoqué une grave péritonite et l'adhérence sur les organes viscéraux. Au microscope, les helmintes causaient une forte infiltration des hétérophiles, des macrophages, des plasmocytes et la fibrose des organes.

Il y avait une atrophie de la pression de l'épithélium du canal biliaire chez un tilapia, qui a été causée par un ténia migrateur pleurococcidé, un protozoaire Cryptocotyle incrusté sur l'arc branchial, et des lésions parasitaires granulomateuses sur les parois des intestins.

Les autres lésions observées étaient des hémorragies, des ulcères et des blessures sur les nageoires, autour de la bouche et sur la peau. Les résultats de cette étude montrent que les poissons de la Rivière Tana sont infestés par les helmintes qui provoquent de graves lésions pathologiques chez les poissons affectés. Ces poissons peuvent aussi servir de réservoirs de ces parasites pour les poissons d'aquaculture.

Mots clés : Étude préliminaire, helmintes, prévalence, lésions pathologiques, Rivière Tana.

Summary

A preliminary study was undertaken between January and May 2006 to investigate the prevalence of helminths and their pathological lesions in four fish species. A total of 43 fresh fish from River Tana, sold at Gikomba market were bought and subjected to postmortem examination. These fish were 15/43 (34.9%) Oreochromis species, 11/43
(25.6%) Clarias spp., 10/43 (23.2%) Cyprinus carpio and 7/43 (16.3%) Barbus spp. On postmortem examination, numerous third stage Contracecum larval worms were found in the abdominal cavity, muscles and behind the gills. A total of 91% of catfish (Clarias spp.) and 20 percent tilapia fish (Oreochromis spp.) had moderate to severe Contracecum worm infestation. The intensity of infestation ranged from 1 to 593 helminths per fish which provoked severe peritonitis and adhesions on the visceral organs. On microscopy, the helminths caused severe infiltration of heterophils, macrophages, plasma cells and organ fibrosis.

There was pressure atrophy of bile duct epithelium in one tilapia caused by lodged migratory tapeworm pleuroceroid, a Cryptocotyle protozoan parasite embedded on the gill arch and parasitic granulomatous lesions on the wall of the intestines.

Other lesions observed were haemorrhages, ulcers and wounds on the fins, around the mouth and on the skin. Results of the study indicate that riverine fish from River Tana are infested with helminths, which cause severe pathological lesions in affected fish. These fish may also act as reservoirs of these parasites to the farmed fish.

Key words: Preliminary study, helminths, prevalence, pathological lesions, River Tana.

Introduction

The annual fish production in Kenya is approximately 200,000 tonnes that earn the fishermen over Kshs 7 billion (approximately US$90 million), and the country about Kshs 4 billion (approximately US$50 million) in foreign exchange, thus contributing to poverty alleviation in rural Kenya. Wild fish are often infested with many parasites that may cause diseases in them. Monogenean (Gyrodactylus spp.) and digenean (Haemorchis spp.) trematodes have been reported in farmed fish in Mombasa, with heavy mortalities seen after handling while Clinostomum and Diplostomum spp. have been observed in various farmed fish in Kirinyaga. Other species of trematodes Heterophyes Dactylogyrus, Cichlidogyrus spp., adult Gymnarchus niloticus, Sanguinicola and cestodes such as Bothrioccephalidae, Caryophyllidae, Proteocephalidae and Amphiliidae spp. have been reported from various fishes in Lake Victoria. Nematodes Contracecum spp. have been reported in Lakes Victoria, Baringo, Magadi, Nakuru and Naivasha. Eustrongylides have been found in East African lakes, including Lake Tanganyika and Victoria. However, these worms have not yet been documented in riverine fish in Kenya. These parasites are reported to cause pathology in highly dense fish populations.

Research in fish diseases and parasites in the country is limited, and the little that there is, has mostly been undertaken in large water bodies (lakes and reservoirs). The objective of this study was thus to determine and document the occurrence of helminths and their associated pathological lesions in riverine wild fish in the River Tana basin.

Materials and methods

Study design and fish

Four purposive sampling visits were undertaken between January and May 2006.
to Gikomba wholesale fish market to purchase fresh fish. A stratified random sampling, based on species, was used to select fish that were purchased from those sold at the market. For this purpose, the fish were grouped into four species (Oreochromis, Clarias, Cyprinus and Barbus). These were the commonest fish catches landed at various sites on the River Tana basin. A total of 43 randomly selected wild fresh fish from the River Tana basin and sold at the market were studied. These were of various ages, sexes, sizes and comprised of 15/43 (34.9%) Oreochromis (tilapia) species, 11/43 (25.6%) Clarias (catfish) spp., 10/43 (23.3%) Cyprinus carpio (common carp), and 7/43 (16.3%) Barbus spp. They were then transported in coolboxes with ice to the laboratory, at the Department of Veterinary Pathology, Microbiology and Parasitology, Faculty of Veterinary Medicine, University of Nairobi for postmortem examination and other tests.

Postmortem examination

In the laboratory, the fish were subjected to postmortem examination as described below and lesions recorded. Each fish was laid on its side on a paper towel in a dissection tray to prevent slipping, and a midline incision made with a scalpel blade starting at the anterior end of the vent. A lateral incision from the vent side in an arc, on the abdominal wall of the fish up to the upper corner of the operculum, was made to expose the swim bladder and other organs. The body wall was then lifted and the organs observed grossly and in situ. A third incision connecting the two previous incisions (opercular incision) allowed the skin flap to be completely removed. The swim bladder, heart, gills, liver, gastrointestinal tract and spleen were separated and examined. Any lesions and parasites encountered were counted and recorded. A cut was made through the musculature to check for lesions and parasites.

Worm identification

Worms collected from the fish body cavity were preserved in 70% ethyl alcohol, manually counted and recorded. Nematodes were identified using the labial parts, presence and shape of esophagus, ventricle and ceacum and the presence of cuticle and genital organs as given elsewhere. Tapeworms were identified using the size and shape of scolexes, number and modification (armed or unarmed) of suckers, the size of segments, cirrus pouch, number of testis and positioning, the shape and positioning of vitellaria, ovaries and uterus, and the position of uterine diverticula.

Histopathological examination

Tissue samples were collected from various organs and preserved in 10% formalin, processed for histology by dehydration through various alcohol concentrations and cleared using amylacetate and xylene. They were then impregnated in molten wax, sectioned, mounted on slides and dewaxed. The dewaxed tissues were rehydrated using descending alcohol concentrations, stained with haematoxylin and eosin, dehydrated using ascending alcohol grades, cleared using two changes in xylene and mounted using DPX mountant. They were then examined under the light microscope (X4, X10 and X40 magnification) for any cellular changes.
Data analysis

Data was entered in Ms excel, exported to Instat© Statistical package for descriptive statistics17. The prevalence was defined as the percent of the total number of fish species infected with helminths divided by the total number of that fish species examined16.

Results

A total of 43 fish comprising of 11 catfish, 15 tilapia, 10 common carp and 7 barbus were examined for lesions and worms. The main worms observed in the fish were 3rd stage Contracaeum larval spp., tapeworms (adult and pleurocercoids) and trematodes (Cryptocotyle). Parasitic granulomas in intestinal walls were also observed. Of the 43 fish examined, 10/11 (91%) catfish and 3/15 (20%) tilapia had 3rd stage Contracaeum larval worms in both abdominal and branchial region. Three out of ten (30%) catfishes and 1/3 (33.3%) tilapia had the Contracaeum larvae in the branchial region only, while 7/10 (70%) catfish and 2/3 (66.7%) tilapia had the Contracaeum larvae in the abdominal cavity encased in fibrinous material. Catfish had a mean worm count of 169 ± 163 (range 0-593), whereas, the tilapia had a mean of 1± 2 (range 0 – 5) (Fig. 1 and 2). Severe peritonitis characterized by blood stained ascitis, fibrin and adhesions around the larvae and organs in the peritoneal cavity was observed in 10/11 (91%) of the catfish and one tilapia fish.

One (1/11; 9.1%) catfish had in addition an adult Proteocephalidae tapeworm in the intestinal tract (Fig. 3). Grossly, the common carp (Cyprinus carpio) and Barbus sop. did not have worms although they had worm lesions. A total of 5/43 (2/43 tilapia, 2/43 barbus and 1/43 common carp) had swellings around the mouth, while 2 tilapia had grayish lesions on the intestines. Other lesions observed grossly were heamorrhages on the skin, base of fins and eyes in 25/43 (7/43 catfish, 7/15 tilapia, 5/7 common carp and 8/10 Barbus) fishes. Muscular haemorrhages in 15/43 (8/11 catfish, 3/15 tilapia, 3/10 barbus and 1/7 common carp) fishes and exophthalmia in 1 tilapia. Lesions due to handling were not considered.

Figure 1: Many Contracaeum larvae:

A - Enclosed in fibrin strands: (x2)  B - Two worm separated from the fibrin mesh: (x4)
Plate 1. Contracecum larvae

Figure 2: Contracecum 3rd stage larvae showing
A- Head region and
B- Tail region (x10)

Figure 3: Head region of an adult Proteocephalus tapeworm with obvious suckers (arrows) (x40).

Figure 4: Tapeworm pleurocercoid (arrow) in the bile duct of a tilapia causing pressure atrophy of bile, necrosis of pancreatic acinar (x40).
On histological examination of the tissues, four (4/15 or 26.7%) more tilapia fish had parasites. One of these had a pleurocercoid tapeworm stage in the bile duct in the liver. This parasite caused bile duct dilatation, pressure atrophy of liver parenchyma and necrosis of adjacent pancreatic acinar cells (Fig. 4). Another one had a Cryptocotyle-like parasite in the gill arches that was surrounded by intensive inflammation (Fig. 5), while parasitic granulomas in the intestines were observed in the other two tilapia fish (Fig. 6).

All _Contracecum_ infested catfish and tilapia showed severe infiltration of mononuclear and polymorphonuclear cells as well as fibroblasts into the mesenteries, intestinal and stomach serosal surfaces. The fish with the swollen mouth areas showed inflammation of the buccopharyngeal region with pyogranulomatous lesions, characterized by heterophils, plasma and other mononuclear cells. One _barbus_ species had a purulent peritonitis characterized by heterophils and mononuclear cells.

**Discussion and Conclusions**

These investigations have documented the occurrence of _Contracecum_ larvae, _Proteocephala_ spp., Cryptocotyle-like parasites and other parasites in the riverine catfish, tilapia and other fishes. _Contracecum_ in fish in the River Tana basin in Kenya, were more prevalent in omnivorous catfish (91%). Prevalence in tilapia was 20% and the parasites were mainly found encysted in the peritoneal cavity and buccopharyngeal cavities unlike in the major lakes, where they were reported to occur in the pericardial cavity with very high prevalence rates. Prevalence rates in catfish (91%) were higher than those reported in the lakes, such as Naivasha (85%), Baringo (70%), Magadi (30%) and George (30%) . The high prevalence and mean load of _Contracecum_ larvae worms in catfish could be attributed to their feeding habits. Catfish are voracious omnivorous fish that feed on smaller fishes. They also feed on copepods (intermediate hosts of the _Contracecum_ worms) and aquatic plants, some of which may have had

**Figure 5:** Cryptocotyle-like parasite (A) on the gill arch and a accompanying cellular inflammation (B). (x40)

**Figure 6:** Numerous parasitic granulomas in the intestinal wall of tilapia fish (arrows). (x40)
free-living third stage *Contraceacum* larvae attached. The catfish can therefore accumulate *Contraceacum* larvae more than the other fish. The bigger catfish were found to have very high worm load of up to 590 worms unlike in other studies. The smaller catfish had fewer worms compared to the larger ones, this may be attributed to the shorter worm accumulation period due to their young age and which may therefore explain the high standard deviation observed in the study. *Contraceacum* larvae caused severe pathological lesions in the peritoneal and bucco-pharyngeal cavities. Large number of these worms in the peritoneal cavity may cause pressure atrophy of the visceral organs while those in the bucco-pharyngeal cavity caused swelling and irritation around the mouth region. The pyogranulomatous inflammation, proliferation of mucus goblet cells and impingement of the buccal cavity, could affect the feeding of the fish and thus growth.

Tapeworms are common in major waters of Africa and demonstrate a high degree of host specificity. *Psychobothridae* tapeworm (*Polychobothrium clarias*) was found in *Clarias gariepinus* from Lake Victoria whereas a *Proteocephalid*, *Proteocephalus bivittellatus* was recorded in African cichlid fishes. The catfish in this study was most probably a definitive host.

The pleurocercoid in tilapia bile duct caused damage to the bile ducts, hepatopancreatic acinar and the liver cells. In severe infections, this may cause reduced production due to disturbed metabolism and even mortalities. Locations of pancreatic tissue in fish vary within and between species. The tilapia in this study had hepatopancreas with the pancreatic acinar cells surrounding the portal veins in the liver. Inflammation was reported around *P. clarias* bothria attached to gut mucosa in infected Lake Victoria catfish, where bothridial penetration into gall bladder mucosa caused granulomas. Other parasites were also observed to cause severe pyogranulomatous reaction in affected fish. This may affect their production. Haemorrhages on the skin, fins and eyes and severe purulent peritonitis could be due to parasitic infections or a combination of other causative agents.

Prevention of *Contraceacum* larval infection in riverine natural habitat by control of definitive hosts (piscivorous birds – Pelicans, Cormorants and Herons) or treatment of water or feed is impractical, but may be of value in farmed and aquaria fish, where use of mechanical restraint (meshnets, cages, and electrical wiring) and deterrents (shotguns and scarecrows) may keep them off. Treatment with helminthicides (levamisole, mebendazole or ivermectin) in feed or as a bath combined with the control of intermediate copepod hosts by use of ectoparasiticides (Nemexon or Bromex) has been effective in farmed and aquaria fish. General tapeworm control (of adult and migrating pleurocercoid stages) by use of Di-n-butyl tin oxide and Dibutyl tin dilurate 25 and Yomesan coupled with copepod control is recommended in farmed and aquaria fish.

Results from this study indicate that wild riverine fish from Tana River are infested with helminths, which cause severe pathological lesions in the affected fish. These fishes may also act as reservoirs of these parasites to the farmed fish.

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