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Survey On Ectoparasites Infestation Of Nile Tilapia (*Oreochromis niloticus*) Collected From Different Aquatic Habitats In Egypt.

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ABSTRACT

This research aims to detect the prevalence of ectoparasites on Nile tilapia collected from three different aquatic habitats in Egypt with histopathological changes of affected tissues by different ectoparasites. During the period between September 2018 and September 2019; a total of 555 *Oreochromis niloticus* fishes taken from River Nile branch (Al Bahr Al Aazam), fish farm (a farm in Kafr el Sheikh) and drainage canal (Bats Drain at Fayoum governorate). The percentage of infection by monogenean was highest in fish culture (29.5%) than that from drainage (20.4%) and River Nile branch (21.5%). The percentage of infection by protozoa was highest in fish culture (38.0%) and lowest in River Nile (8.5%). In the case of metacercarial infection, fish of the River Nile showed the highest prevalence (31.5%). Ectoparasites recovered from examined fish were; protozoan parasites (*Trichodina heterodentata; Trichodinella epizootica)* and (*Myxobolus tilapiae*); two monogenean parasites (*Dactylogyrus extensus* and *Cichlidogyrus tilapiae*) and *Centrocestus formosanus* with different types of encysted metacercaria (Heterophydiae; Clinostomatidae). Hyperplasia was a common histological finding on the gills of fishes infested by monogenean and protozoan parasites. The infested fishes suffer from gills degeneration, necrotic changes and desquamation of the respiratory epithelium lining the gills secondary lamella.

Keywords: Ectoparasites, monogenea, metacercariae, hyperplasia

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INTRODUCTION

Nile tilapia was considered as one of the main cultured fish in the world (1, 2). This is because Nile tilapia can accommodate a wide variety of environmental conditions (3). Extensive aquaculture with dense fish populations may be affected by many ectoparasites (4) which feed on epidermis, eyes, and gills, damaging or killing their hosts (5). Shalaby and Ibrahim (6) have been demonstrated that ectoparasites are the most dangerous category that causes severe mortalities.

Some of the economically important parasite species affecting fish include *Trichodina spp*, *Gyrodactylus spp* and *Dactylogyrus spp* (7, 8). *Cichlidogyrus* is one of the monogenean parasites that infest tilapia, they anchored the gill filaments feeding on the epithelial cells, mucus and gill blood (9). One important zoonotic disease is the infection of skin, gills, and muscles with *Clinostomum complanatum* (10), which were reported from the African continent (11).

Different ciliated parasites reported being problematic in African aquaculture include (*Ichthyophthirius multifiliis*, *Chilodonella spp*, *Trichodina spp*), monogeneans (*Dactylogyrids* and *Gyrodactylids*), different crustaceans; myxosporidean, larval trematodes (Diplostomatids, Clinostomatids), cestodes, acanthocephalans and nematodes (12).

The warm weather in Egypt enables external parasites to reproduce quickly and cause bad effects on fish. Many scientists in Egypt revealed the highest ectoparasitic infection rate among tilapia at Upper Egypt (13) and in Qena Governorate (14)

Parasitological and histopathological diagnoses are presented as tools to analyze and prevent diseases. Fish tissues may be altered due to stressing agents of the environment (15) or induced by pathogens (16). Gill hyperplasia was observed in fishes infested with trematodes (*Dactylogyrus sp*. and *Gyrodactylus sp*).

So, this study aimed to update the prevalence of ectoparasites in freshwater fish in River Nile, fish culture and drainage in Egypt with histopathological changes associated with the infestation of such parasites.

MATERIALS AND METHODS

Collection of fish

During the period between September 2018 to September 2019; a total of 555 *O*. *niloticus;* 1-year-old, 10-15 cm in length) taken from different three localities in Egypt, 210 fish were taken from Nile river (Al Bahr Al Aazam); 240 fish from drainage canal (Bats Drainage) and 105 fish were from the fish farm (Kafr el Sheikh fish farm). These fish surveyed for ectoparasites. The fishes were brought alive and transported to the laboratory for parasitological; histopathological and biochemical examination. All fishes were kept in several aerated covered glass aquaria of 30-liters capacities (17).

Clinical examination of fish

Collected fish were examined for any clinical abnormalities according to the method described by Amlacher (18).

Parasitological examination of fishes

Each part of the fish was examined carefully under the light microscope (mucous surround the skin; gills and fins); from each examined part prepare a fixed smear with methanol; then these smears stained with prepared Giemsa stain.

For examination of monogenean parasites

Smears mixed by saline 0.9% (NaCl); these smears from each part of the body (the skin; gills and fins). The slides were examined for any monogenean parasites using a dissecting microscope (Olympus 110Al 2X) and a compound microscope (Olympus AX70).

All collected fish were carefully examined for different species of metacercariae using the compression method. The metacercariae were identified based on their morphology (19). All the collected parasites were identified according to Yamaguti (20).

Histopathological studies

Specimens of gills and muscles were taken from each infected fish, fixed in 10% neutral formalin buffer and then processed by paraffin embedding technique; stained using eosin and hematoxylin (H&E) stain. The stained tissue sections were examined by light microscopy and the lesions were photographed using camera Olympus XC30 according to Bancroft et al. (21).

RESULTS AND DISCUSSION

Gross examination of fish

The examined fishes suffered from different non specific lesions as hemorrhage in the skin; sloughing of scales with ulcer; some fishes contain either gill necrosis or fins necrosis or both. Fish were found to have *Clinostomum complanatum* on the skin and gills of fishes which appear as yellow grape- like in gills (yellow grape disease) (Fig.1). In this respect, Mitchell (22) proved that the metacercariae of *C. complanatum* infect the skin and muscle causing harmful pathological effect and changes in fish behavior with subsequent economic losses in fish farms. Dias et al. (23) reported the occurrence of EMC in several fish species across the world.

Prevalence of ectoparasites in Nile tilapia

From 555 examined *O. niloticus* fish species; taken from different localities in Egypt, 210 fishes were examined from River Nile, 240 fishes were examined from drainage canals and 105 fishes were examined from fish farms at the period between September 2018 and September 2019 (Table1).

The rate of infection in cultured fish showed the highest record (72.3%). There was no significant difference between the rate of infection in fishes of River Nile branch (61%) and drainage branch (62%), (Table 2).

The percentage of infection by monogenean (prevalence) was highest in cultured fish (29.5%) than that of fish from drainage (20.4%) and River Nile branch (21.5%).

The percentage of infection by protozoa was highest in cultured fish (38.0%) and lowest in Nile River (8.5%) (Table 3)

In case of metacercarial infection, fishes of River Nile branch showed the highest prevalence (31.5%), it was significantly higher than that of other branches that were (4.5%) and (5.70%) for drainage and fish culture respectively (table 3). In the present study, the index of parasitism was analyzed from samples taken from areas with intensive fish culture, where high levels of infestation in Egypt fish culture were observed, confirmed by the large quantities of Monogenea (prevalence) collected during host examinations. Monogeneans are direct life cycle ectoparasites and their distribution is strongly influenced by stocking densities and environmental factors. So, the decrease of monogenic infections in wild fish can be related to the rare contact fish-to-fish, more frequent in farmed fish. The rate of monogenic infection in fish culture was relatively high (29.50 %) this can be easily explained with the confined conditions of the crowded fish which could enhance the transmission dynamics of these opportunistic ectoparasites. This agreed with the results obtained by Cohen and Kohn (24) that found five species of Dactylogyridae (Monogenea) of characid fishes in fish farms in Brazil. Also, Aragot and Moreno (25) and Aragot et al. (26) studied the epidemiology and pathology of Monogenea from *Colossoma. macropomum*, and reported infections by *L. brinkmanni*.

Mahmoud et al (27) revealed that Monogenetic trematodes recorded an incidence of (33.6%) which is nearly similar to those obtained by Abd El-Maged (28) among examined *O. niloticus* was infected on the other hand higher value (80.76) was recorded by Abd El-Gawad (29) which may be due to difference of sample collection and changes in water quality in different localities.

In the present study, the prevalence was higher in cultured *O. niloticus* than in wild fishes. Obiekezie (30) noted that in wild fishes, monogeneans occurred at low intensities, whereas under cultured conditions (in which fishes are highly crowded) these pathogens build up heavy worm burdens which provoke epizootics. Similarly, Violante-González et al. (31) found higher infection levels of *Diplostomum compactum* in cultured tilapia than wild specimens. They attributed this to higher fish densities in the culture systems. Monogeneans- in cultured fishes- have the highest prevalence followed by protozoans and metacercaria. This may be attributed to variation in physico-chemical factors and that high concentration of animals in cultured site than in the wild. Considering the rate of monogenean infection in wild fishes (Nile River branch and drainage canals) was an average of (20%) that was significantly lower than that of fish culture (30%) probably for the rare contact fish-to-fish requested for the transmission of these monogeneans. Monogeneans have been reported to cause severe mortalities in fish hatcheries in Nigeria (32) and South Africa in catfish, black bass and freshwater ornamental fish. Overcrowding of fish into culture ponds or tanks, together with different environmental and management factors, promotes heavy infestations, which can lead to productivity losses, tissue damages and in some cases mortality (12).

Protozoa

Regarding fish culture in this study, our results (table3) revealed that they contain the highest rate of protozoal infection (38.0%) comparable with monogenea (29.5%) and metacercaria (4.7%). These results confirm the opportunistic behavior of ciliates as trichodinids which are strongly influenced by environmental and zootechnical factors, such as biomass density, organic matter load, etc (33). Furthermore, most protozoans found in tilapias during this survey follow a direct life cycle, with an enhanced propagation in confined and crowded systems such as aquaculture facilities.

Encysted Metacercaria (EMC)

In Egypt, the parasitic larvae of trematode (EMC) are highly prevalent in some freshwater fishes as *T. zilli*. Heterophyid MC is among the most important type of trematodes infecting such fish species (34). With the increasing interest in aquaculture, parasitic infections are becoming threats for fish health management and aquatic crop production throughout the world. Fish born trematode (FBT) diseases studied by many authors (35).

Digeneans show a very complex life cycle and their distribution in fish as metacercariae follow the diffusion of intermediate and definitive hosts. Therefore the differences observed among the prevalence of digenean metacercariae in tilapias from different aquatic systems are to be referred mainly to biotic factors such as presence/absence of snails and piscivorous birds (33). This is in line with our results that showed the highest rate of metacercarial infection in the River Nile branch 31.5%.

Parasitological identification of ectoparasites in *O.niloticus*

Different ectoparasites were recovered from examined fish; two protozoan parasites (*Trichodina heterodentata* (36); *Trichodinella epizootica* (37)) and (*Myxobolus tilapiae*) (38); Table 4 contain the morphological parameters of the collected *Trichodina spp*. in *Oreochromis niloticus* collected from Egypt; in addition, there are *Centrocestus formosanus* with different types of encysted metacercaria (Heterophydiae; Clinostomatidae). And two monogenean parasites (*Cichlidogyrus tilapiae* and *Dactylogyrus extensus*) (39, 40) (Fig.2-7).

Effect of ectoparasites on histopathology of gills and muscles

A- Effect of ectoparasites on the histopathology of gills

Generally, the gill histology of fish is used as an identification tool of diseases; the gill structure is highly influenced by external factors and may serve as an indicator of many problems (41).

According to our results, hyperplasia was a common histological finding on the gills of *O. niloticus* infested by ectoparasites (fig8, A, F)

This research also revealed the gills degeneration, necrotic changes, and desquamation of the respiratory epithelium lining the secondary lamella (fig8, A, B), occlusion of the interlamellar space, congestion in lamellar and branchial blood vessels and telangectasis (fig8, A, F).also, necrotic area and edema had been seen surrounding the parasitic cysts(fig8B).

The examination of gill rackers (fig8C) showed parasitic cysts. In some of the examined cases, we noticed the presence of parasitic cysts embedded in gill cartilages (fig8D, E)

Regarding these results, Fernandes and Mazon (42) described edema as an early defense mechanism of the gills against an external parasite. Also, Nascimento et al (43) stated that necrosis caused by different abuses is irreversible damage occurring on the fish gill. Necrotic cells were observed on the gills of cichlid fish infested with *Gyrodactylus sp*.

Gill damage by protozoan and monogenean parasites may be due to their feeding activity, attachment, and locomotion that caused a great destruction of the respiratory epithelium of the secondary lamellae (44); these agree with the results reported by Abd El- Hady (45) where the histopathological examination of tilapias infested with *Trichodina* showed hemorrhage and congestion. Roberts (46) concluded that hyperplasia is the most common response of the gill to damage by protozoan and metazoan parasites.

A very recent study (47) indicated that the harmful effect of *Cichlidogyrus sp* on the gills of *Pseudocrenilabrus sp* is due to the attachment of monogenean to its host by alternating the haptor (for temporary attachment or feeding). The sharp anchors are inserted between two adjacent secondary gill lamellae resulted in rupturing of the interlamellar epithelium and hyperplasia. Also, monogenean blades of the anchors penetrate and lift epithelial tissue to secure firm parasite attachment (48). Kaur and Shrivastav (49) reported damage to the gill cartilages as a histopathological effect of monogenean parasites. Also, there is a positive correlation between Trichodina species, protozoans, and hyperplasic epithelium in the secondary lamellae in tilapia gills (50)

Effect of ectoparasites on the histopathology of muscles

The examination of *O. niloticus* muscles infested by parasites revealed that they infested the uppermost epidermal cell layer, with degenerative and zenker's necrosis in the muscular layer in addition to edema (fig 9 A, B); Meanwhile (fig 9C, D) indicated the presence of parasitic cysts. Concerning this aspect, Elgendy et al (51) photographed the presence of parasitic larvae within highly destroyed muscle bundles in *Anguilla anguilla*. In addition, Gado et al (44) revealed the presence of parasitic cysts in the underlying degenerated muscle tissues together with necrosis of some muscle fibers in *C gariepinus.* Another evident of muscle necrosis due to protozoal infection was made by Feist and Longshaw (52).

Table (1): Number of *O. niloticus* examined from different localities

Studied aquatic habitat	number examined of fish			Total number of examined fish
	River Nile branch	fish culture	Drainage branch	
	210	105	204	555

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Table (2): Prevalence of ecto-parasites in examined *O. niloticus* in different localities

Studied aquatic habitat	River Nile branch			Drainage branch			Fish culture			Total		
	EFN	IFN	% of. Infection	EFN	IFN	% of. Infection	EFN	IFN	% of. Infection	EFN	IFN	IFN
	210	129	61.00%	240	148	62.00%	105	76	72.3	555	353	64.00

Where
EFN= Examined Fish Number
IFN = Infected Fish Number

Table (3): Prevalence, intensity and abundance of ectoparasites recovered from *Oreochromis niloticus* in different aquatic habitat

Studied aquatic habitat	EFN	IFN	PT	PN	Pre %	Int	Abn
River Nile	210	45	monogenea	81	21.50%	1.8	0.37
		18	protozoa	88	8.50%	4.8	0.4
		66	Metacercaria	234	31.50%	3.5	1.1
total	210	129		403	61.00%	3.1	1.9
Fish culture	105	31	monogenea	185	29.50%	6	1.7
		40	protozoa	340	38%	8.5	3.2
		5	metacercaria	6	4.7%	1.2	0.05
total	105	76		531	72.3%	7	5
Drinage	240	49	monogenea	115	20.4%	2.3	0.5
		88	protozoa	287	37%	3.2	1.2
		11	metacercaria	40	4.5%	3.6	0.2
total	240	148		440	62%	2.9	1.8

EFN= Examined Fish Number
IFN= Infected Fish Number
PT= Parasite Type
PN= Parasite Number
Pre= Prevalence
Int= Intensity
Abn=Abundance

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Table 4: Morphological parameters of the collected Trichodina spp. in *Oreochromis niloticus* collected from Egypt

Parasite species	*T. heterodentata*	*T.epizootica*
Body diameter	40-60 (57±3.0)	25-30 (27 ±1.0)
Adhesive disc diameter	38-52 (43±2.1)	16-28 (20±2.0)
Border membrane	3-5 (4.3±0.5)	1.8-2.9(1.8 ±0.1)
Denticulate ring diameter	18-32 (23±1.0)	10-17 (13.4 ±0.5)
Number of denticles	20-29 (25± 1.3)	20 –25 (22 ±1.0)
Denticle length	6-10 (9 ±1.5)	1.5–2.5(2.2 ±0.5)
Blade length	3-6 (4.5 ±0.7)	2.3–3.8(2.8 ±0.3)
Ray length	4.5-8 (6.5± 1.0)	0.8-1.7 (1.0±0.5)

Fig.1. gross examination of Nile Tilapia suffered from different lesion as in A: presence of *Clinostomum complanatum*on on the skin of fishes (presence of yellow nodules under the skin, referred by arrow. B: presence of *Clinostomum complanatum* which appear as many yellow nodules resembles grape like (yellow grape disease) in gills. C: fish suffered from sloughing of scales with ulcer, referred by arrows. D: fish had gill necrosis.

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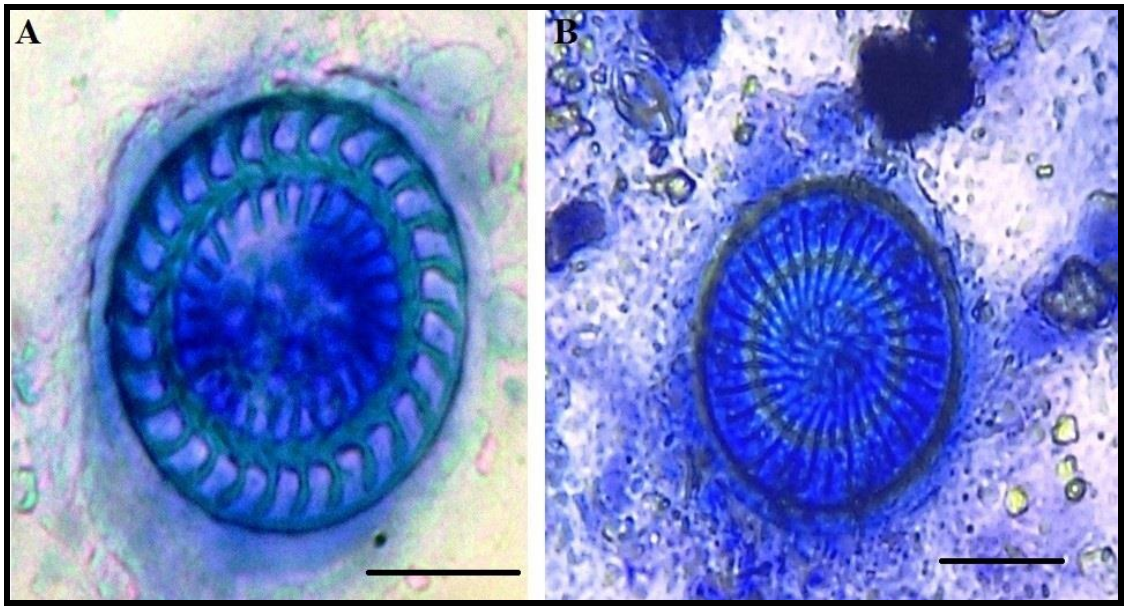

Fig.2: Two species of Trichodina which collected from gills; skin and fins in which A: Trichodina: medium in size (*T. heterodentata*; in A) and B: the other Trichodina spp. is smalls in size (*Trichodinella epizootica*), scale bar in A; B: 100 µm.

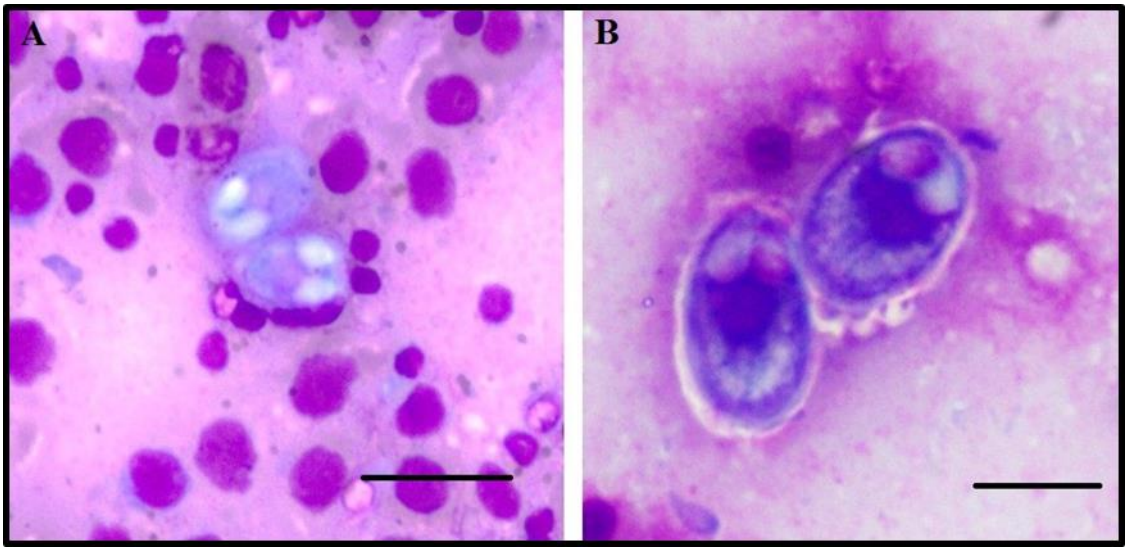

Fig.3: A and B: *Myxobolus tilapiae* spores which are medium sizes, ovoid in shape with 2 smalls polar capsules. Sporoplasm contains one rounded iodinophilous vacuole, scale bar: 100 µm.

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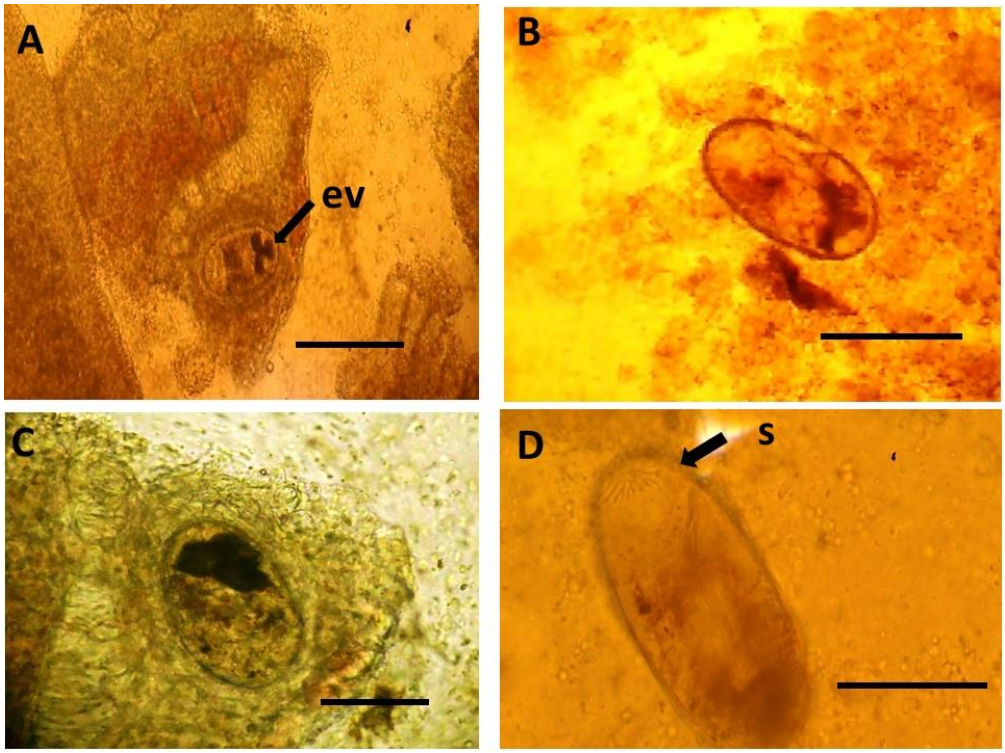

Fig.4: *Centrocestus formosanus* encysted metacercaria from gills (gill trematoda). The oral sucker armed with two rows of circumoral triangular spines referred by arrows; which is prominent located at the most anterior part of the metacercarial body. The execratory vesicle (ev) is characteristic X shape, S (Spines); scale bar: 100 µm.

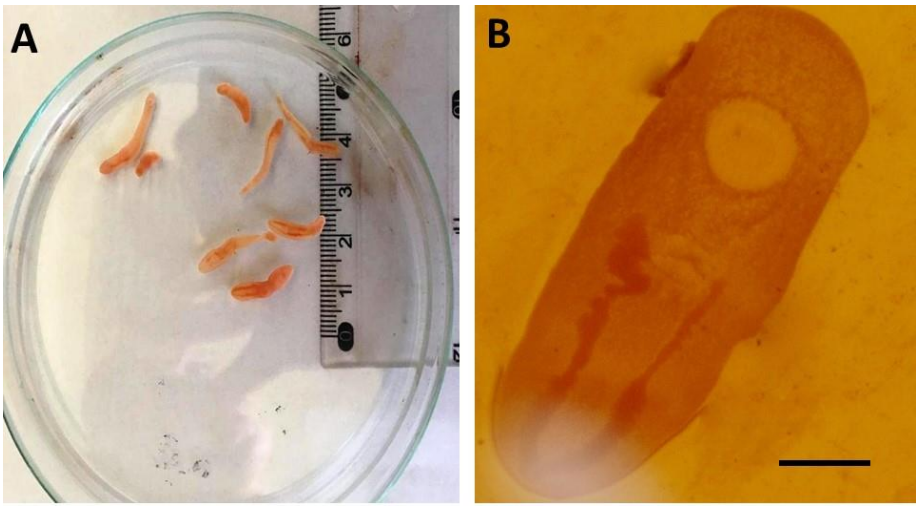

Fig.5: Encysted metacercaria of *Clinostomum. complanatum* was encystment on the fish body. A: *C. Complanatum* excystment which is yellow lingual in shape, flat, and non-segmented. B: *C. Complanatum* showing smooth cuticle unarmed without spines or tubercles. The oral sucker was terminal. The ventral sucker was large and occupied at the first third of the body. The testes were tandem in position in the middle third of the metacercaria, Scale bar: 100 µm.

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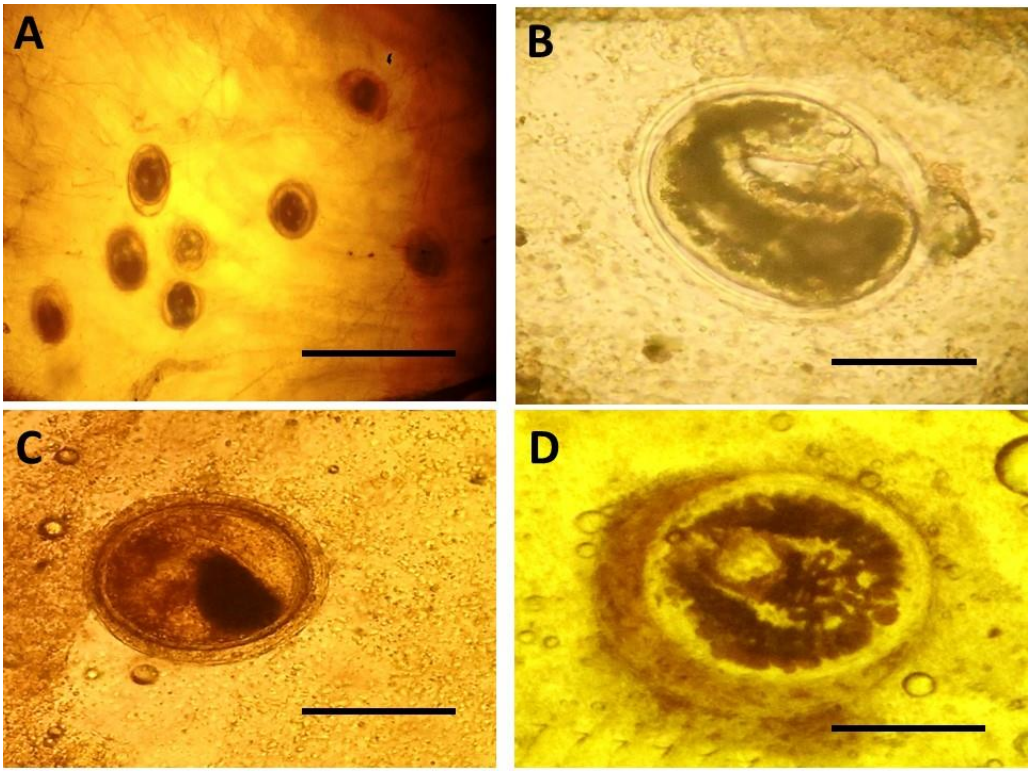

Fig.6: Encysted metacercaria of family heterophyidae. Scale bar: 100 µm. A: heavy infection with Emc in muscles. *B:* Metacercaria of *Haplorchis spp .C: Heterophyes spp* Emc. D: Metacercaria of *Prohemistomum spp*

Fig.7. Monogenean parasitic fluke recovered from gills of infected *O. niloticus*. *A: Cichlidogyrus tilapiae* note the two eyes spots. B. *Dactylogyrus extensus* which have four eyes spots and two pairs of anchors and 14 marginal hooks (7 pairs), one to two connective bars and two needle-like structures.

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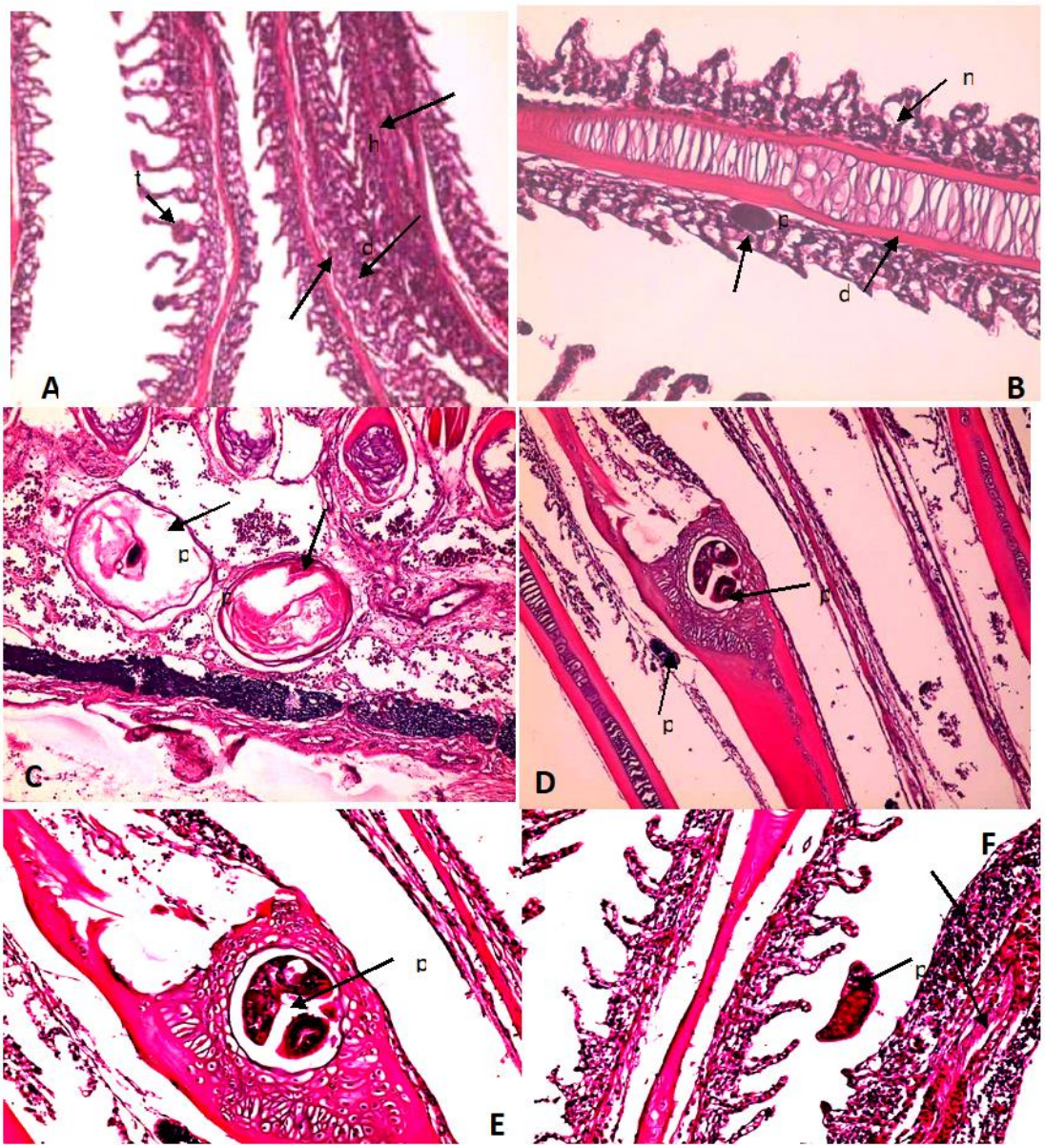

Fig 8: A) Gills showing severe hyperplasia, degenerative and necrotic changes in the respiratory epithelium lining the secondary lamella, occlusion of the interlammeller space, congestion and telangiectasis in lamellar and branchial blood vessels (H&E,X200).

B) gills showing degenerative, necrotic changes and desquamation of the respiratory epithelium lining the secondary lamella, parasitic section in between the respiratory epithelium (H&E,X400).

C(Gill rackers showing parasitic cysts (H&E,X400).

D(Gills showing parasitic cysts Embedded in the gill cartilage (H&E,X200).

E(Gills showing parasitic cysts Embedded in the gill cartilage (H&E,X400).

F) gills showing severe hyperplasia , degenerative changes and necrosis in the respiratory epithelium lining the secondary lamella, congestion, telangiectasis in lamellar and branchial blood vessels and parasitic section in between the secondary lamellae (H&E,X200).

P: parasitic cyst n: zenkers necrosis h: hyperplasia c: congestion t: telangiectasis

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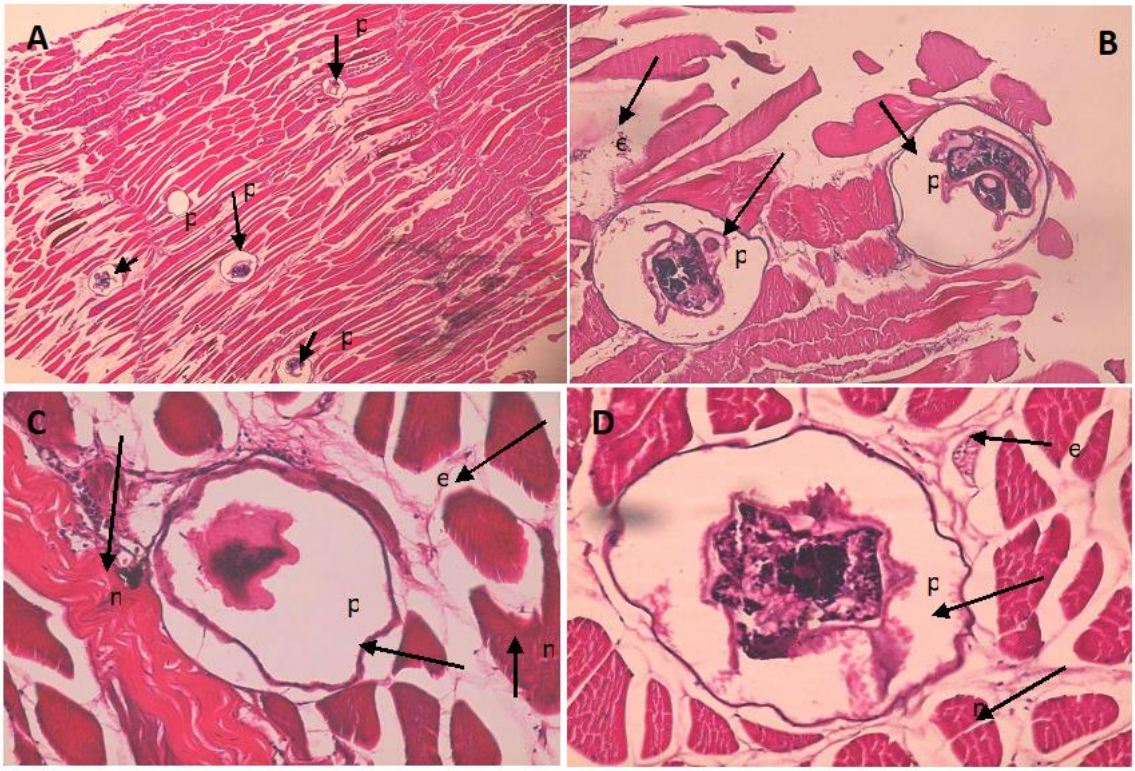

Fig 9:

A) Muscle showing severe parasitic infestation, degenerative and zenkers necrosis in the muscular layer and edema (H&E, X100).

B) Muscle showing parasitic cysts degenerative and zenkers necrosis in the muscular layer and edema (H&E, X200).

C&D) muscle showing parasitic cyst, degenerative and zenkers necrosis in the muscular layer and edema (H&E, X400).

p: parasitic cyst e: edema n: necrosis

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[1] El-Sayed AF. Tilapia culture in salt water: environmental requirements, nutritional implications and economic potentials. Avances en Nutricion Acuicola. 2006

[2] Wang M, Lu M. Tilapia polyculture: a global review. Aquaculture research. 2016 Aug;47(8):2363-74

[3] Philippart JC, Ruwet JC. Ecology and distribution of tilapias. InThe Biology and Culture of Tilapias. ICLARM Conference Proceedings 7, 432 pages 1982 (pp. 15-59). ICLARM-International Center for Living Aquatic Resources Management

[4] Blaylock RB, Bullard SA. Counter-insurgents of the blue revolution? Parasites and diseases affecting aquaculture and science. Journal of Parasitology. 2014 Dec;100(6):743-55.

[5] Reichenbach-Klinke H, Elkan E. The principal diseases of lower vertebrates. Elsevier; 2013 Oct 2

[6] Shalaby SI, Ibrahim MM. The relationship between the monogenetic trematode, Cichlidogyrus tubicirrus magnus (first record in Egypt) and morphological lesions of gills among Tilapia nilotica. Egyptian Journal of Comparative Pathology and Clinical Pathology. 1988;1(1):116-26

[7] Kayis S, Balta F, Serezli R, Er A. Parasites on different ornamental fish species in Turkey. Journal of FisheriesSciences. com. 2013 Apr 1;7(2):114

[8] Syihab M. Irfan MT., Dwi S, Zulham AH, Orbita RD. "BAKTERI PADA TUBUH IKAN GURAMI (Osphronemus gouramy) AKIBAT INFESTASI EKTOPARASIT Argulus sp.(Bacterial Isolate in Gourami (Osphronemus gouramy) as the Result of the Infestation of Ectoparasites Argulus sp.)." AQUACOASTMARINE 8, no. 3 (2015): 14.

[9] Kamil MR, Prayitno SB. STUDI KASUS INFESTASI Cichlidogyrus PADA IKAN NILA (Oreochromis niloticus) DARI SATKER BPBIAT DAN LUAR SATKER BPBIAT JANTI, KLATEN, JAWA TENGAH. Journal of Aquaculture Management and Technology. 2017 Nov 27;6(4):120-9.

[10] Sutili FJ, Gressler LT, de Pelegrini LF. Clinostomum complanatum (Trematoda, Digenea): a parasite of birds and fishes with zoonotic potential in southern Brazil. A Review. Revista Brasileira de Higiene e Sanidade Animal: RBHSA. 2014;8(1):99-114

[11] Britz J, Van As JG, Saayman JE. Occurrence and distribution of Clinostomum tilapiae Ukoli, 1966 and Euclinostomum heterostomum (Rudolphi, 1809) metacercarial infections of freshwater fish in Venda and Lebowa, Southern Africa. Journal of Fish Biology. 1985 Jan;26(1):21-8

[12] Hecht T, Endemann F. The impact of parasites, infections and diseases on the development of aquaculture in sub-Saharan Africa. Journal of Applied Ichthyology. 1998 Dec;14(3-4):213-21.

[13] El-Seify MA, Zaki MS, Desouky AR, Abbas HH, Hady OK, Zaid AA. Seasonal Variations and Prevalence of Some External Parasites Affecting Freshwater Fishes Reared at Upper Egypt. InPhytobiont and Ecosystem Restitution 2018 (pp. 175-183). Springer, Singapore

[14] Hussein AN, Ammar KN, Mansour A, Morsy K. Morphological and Morphometric Characterization of Four Monogenean Parasites from Fishes of the River Nile, Qena Governorate, Egypt. Egyptian Academic Journal of Biological Sciences, B. Zoology. 2019 Jun 1;11(1):31-45.

[15] Brum A, Dotta G, Roumbedakis K, Gonçalves EL, Garcia LP, Garcia P, Scussel VM, Martins ML. Hematological and histopathological changes in silver catfish Rhamdia quelen (Siluriformes) exposed to clomazone herbicide in the Madre River, Santa Catarina State, Southern Brazil. Journal of Environmental Science and Health, Part B. 2014 Mar 4;49(3):169-75

[16] Fernández-Álvarez C, Torres-Corral Y, Saltos-Rosero N, Santos Y. MALDI-TOF mass spectrometry for rapid differentiation of Tenacibaculum species pathogenic for fish. Applied microbiology and biotechnology. 2017 Jul 1;101(13):5377-90.

[17] Saha M, Bandyopadhyay PK. Parasitological and histological analysis of a new species of the genus Thalohanellus and description of a myxozoan parasite (Myxosporea: Bivalvulida) from cultured ornamental goldfish, Carassius auratus L. Aquaculture Reports. 2017 Nov 1;8:8-15.

[18] Amlacher E. "Text book of fish diseases". T. E. S. publication, 1970, Jercy, USA. pp: 135 – 137

[19] Krailas D, Veeravechsukij N, Chuanprasit C, Boonmekam D, Namchote S. Prevalence of fish-borne trematodes of the family Heterophyidae at Pasak Cholasid Reservoir, Thailand. Acta tropica. 2016 Apr 1;156:79-86.

[20] Yamaguti S. Systema helminthum. Vol. IV. Monogenea and Aspidocotylea. Systema Helminthum. Vol. IV. Monogenea and Aspidocotylea.. 1963;4.

[21] Bancroft JD., Stevens A, Turner DR. Theory and practice of histological technique. 4th ed. (1996) Churchill, Livingstone and New York.

[22] Mitchell F. Book Reviews, Accounting Education, (1995) 4:1, 93, DOI: 10.1080/09639289500000010

[23] Dias ML, Eiras JC, Machado MH, Souza GT, Pavanelli GC. The life cycle of Clinostomum complanatum Rudolphi, 1814 (Digenea, Clinostomidae) on the floodplain of the high Paraná River, Brazil. Parasitology Research. 2003 Apr 1;89(6):506-8.

[24] Cohen SC, Kohn A. On Dactylogyridae (Monogenea) of four species of characid fishes from Brazil. Check List. 2009 Jun 1;5(2):351-6.

[25] Aragort W, Moreno LG. Indices epidemiológicos de trematodos monogenésicos en branquias de colossoma macropomum bajo cultivo. Acta biol. venez. 1997:1-8.

[26] Aragort W, Morales G, León E, Pino LA, Guillén A, Silva M. Patologías asociadas a monogeneos branquiales en cachama bajo cultivo. Veterinaria Tropical. 2002;27(2):75-85.

[27] Mahmoud AE, Mona SZ, Abdel RY, Hossam HA, Osman KA, Attia AA. Seasonal variations and prevalence of some external parasites affecting freshwater fishes reared at upper Egypt. Life Science Journal. 2011;8(3):397-400.

[28] Abd El-Maged RR. Studies on ectoparasites of freshwater fishes in Dakahlia governorate (Doctoral dissertation, Ph. D. Thesis, Fac. Vet. Med., Kafr El-sheikh Univ).2009.

[29] Abd El-Gawad RA. Studies on ectoparasites of freshwater fish. MV Sc. Thesis. Fac. Vet. Med., Zagazig. Univ. 2004.

[30] Obiekezie AI. The principal pathogens and diseases of cultured fishes in Nigeria. orkshop on research in aquaculture 1991 Jan 23 (pp. 23-27).
[31] Violante-González J, Aguirre-Macedo ML, Rojas-Herrera A, Guerrero SG. Metazoan parasite community of blue sea catfish, Sciades guatemalensis (Ariidae), from Tres Palos Lagoon, Guerrero, Mexico. Parasitology research. 2009 Oct 1;105(4):997-1005.
[32] Obiekezie AI, Taege M. Mortalities in hatchery-reared fry of the African catfish, Clarias gariepinus (Burchell) caused by Gyrodactylus groschafti, Ergens, 1973. Bulletin of the European Association of Fish Pathologists. 1991;11(2):82-5.
[33] Florio D, Gustinelli A, Caffara M, Turci F, Quaglio F, Konecny R, Nikowitz T, Wathuta EM, Magana A, Otachi EO, Matolla GK. Veterinary and public health aspects in tilapia (Oreochromis niloticus niloticus) aquaculture in Kenya, Uganda and Ethiopia. Ittiopatologia. 2009;6(12):51-93.
[34] Ibrahim MM, Soliman MF. Prevalence and site preferences of heterophyid metacercariae in Tilapia zilli from Ismalia fresh water canal, Egypt. Parasite. 2010 Sep 1;17(3):233-9.35) Marzouk MS, Mahdy OA, Mohamed LA, El-Hady AO, Ibrahim TB. Biological studies on some Metacercariae affecting freshwater fishes with special reference to protein characterization. Vet. Med. Assoc. 2007;67:151-70.
[35] Valladao GM, Gallani SU, De Padua SB, LATERÇA M, Pilarski F. Trichodina heterodentata (Ciliophora) infestation on Prochilodus lineatus larvae: a host–parasite relationship study. Parasitology. 2014 Apr;141(5):662-9.
[36] Saha M, Bandyopadhyay PK. First report of some known Trichodinella epizootica Šramek-Hušek, 1953, and Tripartiella Lom, 1959 from the cultured Oranda Gold Fish (Carassius auratus auratus) in India. Zootaxa. 2016 Nov;4184(1):130-40.
[37] Mohammed NI, Rabie SA, Hussein AN, Hussein NM. Infestation of Oreochromis niloticus and Tilapia zilli fresh-water fishes with myxosporean parasites, Qena Province, Egypt. Egyptian Academic Journal of Biological Sciences, B. Zoology. 2012 Dec 1;4(1):235-46.
[38] Paperna I. Studies on monogenetic trematodes in Israel. 2. Monogenetic trematodes of cichlides. Israel. J. aquacult_ Bamidgeh, 1960 12: 20-33.
[39] Maneepitaksanti W, Nagasawa K. Monogeneans of Cichlidogyrus paperna, 1960 (Dactylogyridae), gill parasites of tilapias, from Okinawa Prefecture, Japan. Biogeography. 2012 Sep;14:111-9.
[40] Yandi İ, Kayiş Ş, Er A. Histopathological effects of etiological and nonetiological agents in some fish gills. Fresenius Environmental Bulletin. 2017 Jan 1;26:995-1000.
[41] Fernandez MN, Mazon AF. Environmental pollution and fish gill morphology, Fish Adaptations, edited by AL Val and BG Kapoor." (2003): 203-31.
[42] Nascimento AA, Araújo FG, Gomes ID, Mendes RM, Sales A. Fish Gills Alterations as Potential Biomarkers of Environmental Quality in a Eutrophized Tropical River in South-Eastern B razil. Anatomia, histologia, embryologia. 2012 Jun;41(3):209-16.
[43] Gado MS, Mahfouz NB, Moustafa EM, Lolo EE. Prevalence of some ectoparasitic diseases in African catfish (Clarias gariepinus) at Kafr El-Sheikh governorate. International Journal of Fisheries and Aquatic Studies. 2017;5(3):576-83.
[44] Abdel-Hadi OK "Comparative studies on some parasitic infection of fishes in fresh and polluted water sources." (1998)
[45] Roberts, Ronald J. Fish pathology. John Wiley & Sons, 2012
[46] Igeh PC, Avenant-Oldewage A. Pathological effects of Cichlidogyrus philander Douëllou, 1993 (Monogenea, Ancyrocephalidae) on the gills of Pseudocrenilabrus philander (Weber, 1897)(Cichlidae). Journal of fish diseases. 2020 Feb;43(2):177-84
[47] Madanire-Moyo GN, Avenant-Oldewage A. The histopathology of Enterogyrus coronatus Pariselle, Lambert & Euzet, 1991 (Monogenoidea) in the stomach of the southern mouthbrooder Pseudocrenilabrus philander (Weber, 1897)(Cichlidae). African Zoology. 2015 Apr 3;50(2):175-80.
[48] Kaur P, Shrivastav R. Histological effect of monogenean parasites on gills of fresh water carps. European Journal of Biotechnology and Bioscience. 2014;2(2):50-3.
[49] Steckert LD, Cardoso L, Jerônimo GT, de Pádua SB, Martins ML. Investigation of farmed Nile tilapia health through histopathology. Aquaculture. 2018 Feb 3;486:161-9.
[50] Elgendy MY, Kenawy AM, El-Deen AE. Gyrodactylus anguillae and Vibrio vulnificus infections affecting cultured eel, Anguilla anguilla. Comunicata Scientiae. 2016 May 10;7(1):1-1.
[51] Feist SW, Longshaw M. Histopathology of fish parasite infections–importance for populations. Journal of Fish Biology. 2008 Dec;73(9):2143-60