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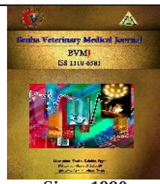

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Review Article

Prevalence of nodular parasitic diseases in freshwater fishes in Egypt: A Review

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ABSTRACT

Aquaculture has significant potential to contribute substantially to food security and enhance export incomes. Diseases have emerged as a significant limitation to aquaculture output on a global scale, hindering the progress of both economic and social development worldwide. Several factors contribute to the incidence of fish diseases, such as intensification, environmental contamination, climatic changes, a lack of effective biosecurity measures, and a lack of aquaculture awareness about diseases and their control. Freshwater fish intensification led to various infectious viral, bacterial, fungal, and parasitic disease outbreaks, resulting in high mortality and significant economic losses. Therefore, infectious diseases, particularly parasitic ones, pose a significant challenge to aquaculture sustainability, accounting for over 80% of fish diseases in Egypt. Parasitic diseases of fish have greatly reduced fish survivability, growth performance, and reproductivity, leading to loss of fish production due to high mortalities and pathological changes in infected organs. The present review deals with parasitic diseases that have severe economic concerns for freshwater fish and have zoonotic importance.

1. INTRODUCTION

Aquatic food forms a significant part of the global food future, encompassing both marine and freshwater modes of production. (Saidon et al., 2024). Egypt is ranked as the top aquaculture producer in Africa and the third largest tilapia producer globally, after China and Indonesia, according to FAO (2022). Nile tilapia is the most cultivated freshwater species due to its characteristic traits (El-Gohary et al., 2020). In addition, African catfish are a common species in the Nile River and have been cultured with other fish species to enhance productivity (Abdelkhalek et al., 2024).
Parasitic diseases are one of the most serious problems, affecting fish production globally and causing huge economic losses, affecting fish health, and having potential risks to human health (Tessema, 2020). In Egypt, parasitic diseases account for about 80% of fish diseases (Eissa, 2002), and many earlier studies have been conducted on parasites affecting freshwater fishes (El-Asely et al., 2015; Shaheen et al., 2017; Eissa et al., 2021). The warm climate and water pollution facilitate the proliferation of parasites through the reproduction of intermediate hosts, resulting in a widespread impact (Younes et al., 2016). Fish may be affected by numerous external and internal parasites that cause harmful effects on the fish's health conditions (Abd-ELrahman et al., 2023). Digenetic trematode metacercariae are considered significant endoparasites, causing substantial economic losses in several cultured and wild fish species. They can also seriously affect human health when consumed raw or improperly cooked fish (Song et al., 2018). In this regard, we focused in our review on the prevalence of parasitic diseases associated with nodular or cyst formation in wild and cultured freshwater fishes in Egypt, such as microsporidiosis, myxosporidiosis, henneguyasis, clinostomiasis, and euclinostomumosis. These diseases have a great impact on aquaculture production and have human health risks.

Microsporidiosis

Microsporidia is a class of obligatory intracellular eukaryotes that are closely related to the fungi and infect a wide range of vertebrate and invertebrate hosts (Larsen et al., 2017). Microsporidia can be transmitted by food and water and are likely zoonotic, as they occur in both immunocompetent and immunodeficient humans (Lv et al., 2024). About 220 genera of microsporidia are known to infect aquatic organisms, with more than 160 species under 22 genera affecting fish (Liu et al., 2019). The most economically important microsporidia that infect fish are Glugea, Loma, Pleistophora, Enterocytozoon, and Nosema, which cause significant injury to organs (Weng et al., 2023). Currently, Anncaliia, Encephalitozoon, Enterocytozoon, Mi crosporidium, Nosema, Pleistophora, Trachipleistophora, T ubulinosema, and Vittaforma are known to be zoonotic microsporidia in fishes (Han et al., 2021). Microsporidiosis has been mainly reported in immunocompromised people and is mainly associated with diarrheal symptoms, encephalitis, keratoconjunctivitis, hepatitis, sinusitis, and myositis (Weiss, 2014).
Microsporidia life cycles are more complex and may involve generating several distinct developmental stages, which include the proliferative phase (merogony), sporogonic phase (sporogony), and mature infective spores (Vávra and

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Lukeš, 2013). These spores have an extreme chitinous layer resistant to environmental stress and lysis, allowing the organism to keep viability in the aquatic environment for prolonged periods and to fight digestive enzymes in the gastrointestinal tract of the hosts (Shaw et al., 2000).

In Egypt, there is very limited information available on the prevalence of microsporidia infection in freshwater fishes. Microsporidia spp. spores have been recorded from African catfish (AbdEl-Lateif and Torra, 2020; Abd Rabo, 2017). African catfish infected with *Glugea anomala* (*G. anomala*) exhibited the highest prevalence rate of 36% in spring, while the lowest infection rate, 18%, was recorded in winter (Yassen et al., 2024). Moreover, Nile tilapia showed whitish nodules of *G. anomala* in the liver and kidneys, at a prevalence rate of 19.75%, with a higher infection rate of 32% in autumn (Yassen et al., 2024). The infection rate with microsporidians was highest in spring in African catfish and was not recorded in the other seasons (Abd Rabo, 2017).

Histopathology is the tedious method by which the developmental stages (spores, sporoblast, presporoblast stages, and sporonts) are found rarely in small aggregates dispersed randomly throughout xenomas. Numerous aggregations of pink to magenta spherical or oval spores in the cytoplasm of hepatocytes and pancreatic acini, as well as the renal tubular epithelium, with severe hepatorenal damage, were recorded in Nile tilapia and African catfish infected with *G. anomala* (Yassen et al., 2024). PCR-based techniques of small subunit ribosomal DNA (SSU rDNA) sequence analysis with the help of phylogenetic analysis provide more reliable identification of Microsporidia spp. (Morsy et al., 2012). Yassen et al. (2024) carried out molecular analysis for Microsporidia spp. isolated from Nile tilapia and African catfish using 18S rDNA gene sequences, which revealed that the isolated species were like the G. anomala sequence, and their sequences were deposited in GenBank under the accession numbers OR766288 and OR766287, respectively.

Myxosporidiosis

Myxozoa is an obligatory parasite belonging to the phylum Cnidaria, affecting marine and freshwater fish and other aquatic animals (Atkinson et al., 2018). Parasitic myxosporidians pose a significant threat to commercially valuable fish species, particularly in aquaculture, by causing extensive damage and mortalities (Abdel Ghaffar et al., 2015).

The complex life cycles of myxozoans involve annelids and bryozoans as definitive invertebrate hosts and mostly fish as intermediate vertebrate animals (Yokoyama et al., 2012). The absence of an effective therapy for myxozoan infections in fish is a contributing factor to the significant economic losses experienced in fisheries and aquaculture (Yokoyama et al. 2012). This parasite has been associated with ovarian disruption, corneal opacity, reduced growth rates, and respiratory distress (Eissa et al., 2020). For instance, *Tetracapsuloides bryosalmonae*, which causes proliferative kidney disorders (PKD) in salmonids, and *Myxosoma cerebralis*, which causes whirling disease, are the most economically damaging fish diseases (Lom and Dyková, 2006).

In Egypt, the early record of described Myxosporea from freshwater fishes was conducted by Fahmy et al. (1975). Different Myxobolus species infecting wild and cultured fishes have been recorded, such as Myxospora sp. (Matter et al., 2013), *M. agolus*, *M. heterosporus* type 2, *M. clarri*, *M. heterosporus* (type 3) (Mohammed et al., 2012, Soror et al., 2012), *M. dermatobius* (El-Sayed, 2020), *M. cerebralis* (Eissa et al., 2006), *M. brachysporus* (Abdel Ghaffar et al., 2015; Abdel-Baki et al., 2015a), *M. israelensis* (Eissa et al., 2020), and *M. tilapiae* (Abdel-Baki et al., 2015b and Eissa et al., 2021). Additionally, M. dermatobia was isolated by Mohamed et al. (2004) from *Tilapia zillii* in Giza province. Abdel-Ghaffar et al. (2008) described *Zschokkella nilei*, *Ortholinea africanus*, *Triangula egyptica*, *M. fomenai*, and *M. branchiophilus* from the Nile tilapia in the River Nile. The infections by these parasites have been reported in various organs such as gills, eyes, fins, intestines, hearts, kidneys, livers, brains, and ovaries (Abdel-Ghaffar et al., 2008; Abdel-Baki et al., 2015a; Eissa et al., 2021).

Seasonal prevalence of myxozoan infection in African catfish was highest in winter (15.5%) as recorded by Shehab El-Din (2008). Also, Matter et al. (2013) reported that the highest seasonal prevalence of myxosporidiosis was in winter, which reached 56.1% in the examined African catfish, 71.4% in *Mugil cephalus*, 85% in *Lates niloticus*, 66.7% in *Ctenopharyngodon Idella*, and 43.4% in Nile tilapia. Histopathological alterations induced by *M. heterosporous* were localized inflammatory changes in corneal tissues with mononuclear cellular aggregation associated with a large number of the myxosporean plasmodial stages (Eissa et al., 2006). Moreover, the infected kidneys with *M. tilapiae* in Nile tilapia collected from the River Nile at Beni-Suef governorate, Egypt, revealed the presence of spores within the Bowman capsule and renal glomeruli, and in more advanced cases, a capsule composed of multilayered fibrous connective tissue developed around the plasmodium (Abdel-Azeem et al., 2015). In the identification of myxosporean parasites, molecular analyses in combination with spore morphology, host specificity, and tissue affinity supplied sufficient tools for the detection of these parasites and revealed the phylogenetic position of myxosporeans within metazoans (Liu et al. 2013). Abdel-Gaber et al. (2017) performed partial 18 SSU rDNA gene sequences for the recovered myxosporean parasites isolated from the naturally infected Nile tilapia that enabled them to be identified as *M. agolus*, *M. fomenai*, *M. brachysporus*, *M. tilapiae*, and *Triangula egyptica*. The phylogenetic tree revealed that myxozoan clades are separated into two main branches: the marine branch, which is represented by the order Multivalvulida (Kudoa species), and the freshwater branch, which is represented by the order Bivalvulida (Henneguya, Myxobolus, Zschokkella, Myxidium, and Sphaeromyxa) (Kent et al., 2000). Meanwhile, 18S rDNA gene sequencing did not advocate the phylogenetic split of the genera Henneguya and Myxobolus (Andree et al., 1999).

Henneguyasis

Henneguya Thélohan, 1892, are fish-parasitic myxospores that cause considerable pathological changes and severe diseases in wild and cultured fish species worldwide (Rangel et al., 2023). Several species were reported to cause severe fish henneguyasis, including *H. ictaluri* and *H. exilis* for proliferative gill diseases and verrucous dermal henneguyosis (Rosser et al., 2016). Henneguyosis mainly infects catfishes, and the respiratory form causes congestion of the gills and accessory respiratory organs, hence causing high economic losses because of the presence of large visible cysts on the gills and the dendritic organs that make infected catfish unmarketable (Eissa, 2002). There is no treatment for the disease, but the prevention depends upon proper feeding, biosecurity measures, quarantine measures, good water qualities, and disinfection by using calcium hydroxide. The parasitic spores can survive for years in the soil at the bottom of ponds and are extremely resilient to harsh environmental conditions (Banu and Rathinam, 2023).

In Egypt, it was detected in the dendritic organ of African catfish (Shehab El-Din, 2008; Soror et al., 2012; Abd Rabo et al., 2017). The first record of Henneguya fusiformis in the ovaries of *Clarias gariepinus* was recorded by Abou Zaid et al. (2021). Henneguya nilotica and *H. suprabranchiae* from the secondary respiratory organ of *Clarias lazera* were observed by Rabie et al. (2009) and El-Mansy and Bashtar (2002), respectively. Microscopic examination of Henneguya infection is the most common step of diagnosis, as its spores appeared elongated with two rounded polar capsules and long tails (spermatozoa-like) (Morsy et al., 2012). Molecular identification is also becoming increasingly available in diagnostic laboratories (Emeish et al., 2022). Henneguya displayed significant differences in seasonal prevalence, with a maximum prevalence in winter followed by spring and autumn and a minimum prevalence throughout summer (Younes et al. 2019; Abou Zaid et al., 2021). Abd Rabo et al. (2017) revealed that the maximum seasonal prevalence of Henneguyasis was in winter in African catfish, and they attributed that to the high level of organic matter and ammonia in the water during winter with a low level of dissolved oxygen. The histopathological alteration that occurred in henneguyosis was focal parasitic nodules and mononuclear infiltration in dendritic organs (Shehab El-Din, 2008). In addition, Abd Rabo et al. (2017) revealed that the infected African catfish had cysts in hyaline cartilage surrounded by connective tissue and compressed of the endothelial layer of them due to the presence of plasmodia and causing atrophy of them. Blood vessels appeared dilated, damaged, and sloughing with necrosis of whole bronchioles in the organ.

Clinostomiasis (Yellow grub disease)

Clinostomiasis is a freshwater disease caused by digenetic trematodes in the genus Clinostomum and known as yellow grubs caused by *C. complanatum* that appeared as yellowish-white nodules or orange pea-like cysts in the musculature of buccal cavities, skin, and base of fins (Hamouda and Younis, 2021; Mahdy et al., 2022). The digenetic trematodes of the genus Clinostomum are endoparasites with a complex life cycle involving two intermediate hosts and one definitive host (Daly et al., 2002). The adult trematodes typically parasitize the oral cavity, pharynx, or esophagus of piscivorous birds; their feces contain Clinostomum eggs, which hatch in the water. Clinostomum spp are considered one of the most important fish trematodes of public health concern detected in several countries, causing laryngo-pharyngitis and halzoun-like disease in people who ingest raw or undercooked fish (Menconi et al., 2020). Furthermore, encysted metacercariae of Clinstomum sp. cause low weight gain, high mortality, and fish unmarketability (Salem et al., 2021).

In Egypt, larval stages of *C. complanatum*, *C. phalacrocoracis*, *C. tilapiae*, and Clinostomum spp. have been reported from both wild and cultured fish (Shaheen et al., 2014; Soliman and Yacout, 2016; Ahmed et al., 2018; Salem et al., 2021). Clinostomum cutaneum was first detected by Hamouda and Younis (2021) in Aswan Governorate at a prevalence rate of 25% and 12.5% from cultured and wild Nile tilapia, respectively. The excysted metacercaria of Clinostomum spp. was whitish yellow, elongated larvae with two very long intestinal caeca and two large suckers (Shaheen et al., 2014). C. complanatum was observed in buccal cavities, gill chambers, and the skin of Nile tilapia at a prevalence rate of 33.8%, 5.25%, and 1.0%, respectively (Mahdy et al., 2024a).

Diagnosis of clinostomumasis depends mainly on morphological characteristics of the excysted metacercaria; however, the morphological similarities between Clinostomum species are facing challenges. Therefore, molecular approaches are vital for rapid and accurate diagnosis of these prevalent digenean parasites (Younis et al., 2023; Moema et al., 2013). Histopathological examinations of infected Nile tilapia with C. cutaneum revealed atrophy and sloughing of the epidermis and hyalinization and necrosis of muscle fibers as well as the disappearance of portions of the dermis layer (Hamouda and Younis, 2021). Chitosan nanoparticles at a concentration of 12.5 μg/ml exhibited a hopeful antiparasitic effect against clinostomum larvae in vitro (Mahdy et al., 2024b). Praziquantel is an efficient and safe treatment for controlling clinostomid infections affecting cultured Nile tilapia (Mahdy et al., 2024c).

Euclinostomumosis

Several previous studies detected Euclinostomum sp. in Nile tilapia (Hamouda and Younis, 2021, and Abd-ELrahman et al., 2023; Yassen et al., 2023). Euclinostomum sp. appeared as round to oval grayish-black cysts and gave the area around it a faint black color (Mansour, 2019). Younis et al. (2022) recorded an infection rate with *Euclinostomum sp*. in Nile tilapia reaching 25.25%. In Africa, the well-known type species is *E. heterostomum* (Attia et al., 2021), as well as several dubious metacercaria (Hamouda and Younis, 2021). Pathological alterations in the kidneys infected with EMC of *E. heterostomum* exhibited intact cuticle and normal internal structures of parasites surrounded by glomerular congestion, vacuolization, degenerative and necrotic changes with inflammatory cell infiltration, and activation of melanomacrophage centers (Mahdy et al., 2021). There is little molecular investigation to describe *Euclinostomum spp*. in Egyptian fish. PCR amplification and sequencing of mtDNA CO1 gene regions of E. heterostomum were carried out by Mahdy et al. (2021) and the rRNA-28S marker (Younis et al., 2022).

Euclinostomum metacercariae infect the kidneys of Tilapia zillii, causing severe pathological changes (Mahdy et al., 2017). In Aswan, Egypt, the EMC of *E. heterostomum* and *E. ardeolae* was recorded in farmed and wild tilapia spp. that lowered their market value and caused economic losses. They are identified both morphologically and molecularly. They have negative effects on the fish, which have been confirmed histopathologically. Tilapia spp. musculature is free of infections and suitable for human consumption. The seasonal prevalence increases in summer and spring while decreasing in winter (Matter et al., 2013). The total prevalence rate of *Euclinostomum spp*. in naturally infected Nile tilapia was 5.75%, with the highest seasonal prevalence rate of 11% in summer (Yassen et al., 2023). Additionally, Shaheen et al. (2014) reported a total prevalence rate of Euclinostomum metacercariae in Nile tilapia of 18.3%, with the highest seasonal prevalence in summer (27.3%).

2. CONCLUSIONS

Fish parasites may potentially harm fisheries and aquaculture. The prevalence of several parasitic diseases in Egyptian fish species, either cultured or wild, significantly impacts the health and quality of fish. Furthermore, clinostomatid has zoonotic importance; thus, tilapia must be cut back and gutted after harvesting and sufficiently cooked. More investigation into the ecology and epidemiology of parasites is essential to reduce mortalities and economic losses.

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