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Exploring the Relationship Between Water Quality, Parasitic Infestation, and Pathological Alterations in Tilapia Fish

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ABSTRACT

This study investigated the parasitic infestations among 300 tilapia fish (*Oreochromis niloticus* and *O. aureus*) and the water parameters of sixty water samples from Damietta Governorate. Clinically, affected fish displayed emaciation and opacity in the eye with focal external hemorrhages. The detected parasites were protozoa (*Chilodonella hexasticha*), monogenea (*Cichlidogyrus tilapiae*), and digenetic trematode metacercaria (*Heterphyes* spp, *Haplorichis* spp, *Clinostomum tilapiae, Euclinostomum heterostomum, Diplostomum spathacaum, Centrocestus formosanus*, and other unidentified metacercaria). The total infestation rate was 60%, (66.7% in *O. niloticus*, followed by 53.3% in *O. aureus*). Summer was the season with the highest infestation (80% in *O. niloticus* and 64% in *O. aureus*). Water parameters such as visibility, oxygen levels, pH, and total alkalinity exhibited a significantly negative correlation with parasitic infestation. Conversely, water temperature, unionized ammonia, and NO3-N concentrations displayed a positive correlation. The occurrence of parasitic infestation increased in highly polluted water, where zinc exhibited a notable positive correlation with its infestation. Histopathological examinations of tilapia species exhibited various pathological alterations. *C. hexasticha* embedded within the secondary lamellae of the gills that were surrounded by a connective tissue capsule. Additionally, *C. tilapiae* was identified within the musculature, situated between muscle bundles. Parasitic cysts of *Haplorchis* spp. were encapsulated with connective tissue that caused pressure atrophy in adjacent hepatocytes. Furthermore, *E. heterostomum*, was identified in renal tissue. Thus, this study affirmed the connection between parasitic prevalence and changes in the aquatic environment. Understanding these dynamics is essential for managing and mitigating the impact of climate change on the epidemiological maps for parasitic diseases in fish populations, ultimately contributing to disease minimization and the promotion of sustainability in aquaculture.

INTRODUCTION

Worldwide, the Nile tilapia (*Oreochromis niloticus*) is one of the most widely farmed finfish species. In 2020, the inland aquaculture sector produced more than 4.4 million tonnes, ranking it as the third most cultivated fish, trailing only grass and silver carp **(FAO, 2022)**. Tilapia fish species exhibit a remarkable adaptability to various environmental conditions, allowing them to thrive in diverse systems such as ponds, cages, canals, intensive, and super-intensive systems. Their native habitats include lakes, streams, rivers, and brackish waterways **(Prabu *et al*., 2019)**.

Parasitic infestations in fish can be caused by obligatory and opportunistic parasites, affecting host mortality, growth, and reproduction. Some parasites have zoonotic potential, posing risks to public health. Fish parasites include protozoans, helminths, and crustaceans. Protozoans may cause internal or external infections depending on their habitats, while crustaceans and monogeneans act as external parasites, causing significant diseases **(Aly, 2013)**. Additionally, digeneans contribute to internal parasitic disorders, including trematodes, cestodes, nematodes, and acanthocephalans **(Hoffman, 2019)**. Because of the rising economic importance of tilapia, parasitic diseases can lead to substantial economic losses, either through mortality or rendering fish unfit for the market. For instance, when fish experience significant infestations of ectoparasites or endoparasites, their regular growth is hindered or compromised. These parasites, like those of other vertebrates, feed either on the digested contents of the host's gut or the host's own tissue **(Rohde 2005)**. **Reed *et al.* (2012)** exemplified that monogenean infestations cause irritation and excessive mucus production and could create an entrance for bacterial invasion via a hook-like structures that used to attach to the fish. The detrimental effects of monogeneans are heightened when fish experience environmental or behavioral stressors.

The majority of fish health problems are due to environmental factors including poor water quality, overcrowding, nutritional inadequacies, or stress **(Klinger & Floyd, 1998)**. Furthermore, **Marcogliese (2001)** noticed that, parasites within aquatic systems are influenced not only directly by temperature variations but also indirectly by alterations in other abiotic parameters. These indirect effects occur through changes in the distribution and abundance of their hosts. In addition, **Hakalahti *et al*. (2006)** suggested that, the extended durations of elevated summer water temperatures due to climate change can directly impact parasites by prolonging the favorable period for development in snails, the release of cercariae, transmission to fish, and the development of metacercariae. As temperature also controls the population dynamics of the hosts, especially snails, which reproduce continuously when conditions are favorable, increased water temperatures in late summer and autumn may lead to the production of new generations late in the season, which could affect parasite dynamics. Water deterioration, temperature fluctuation, and environmental stressors, combined with increased bacterial load during hot months, in the presence of monogenean parasite lead to significant parasitic infestations that resulted in massive fish mortalities **(Bwoga, 2021)**.

Thus, the present study aimed to identify internal and external parasitic infestations in tilapia species (*O. niloticus* and *O. aureus*) and assess their association with variations in aquatic ecosystem parameters, in addition to investigating the histopathological changes related to these infections.

MATERIALS AND METHODS

Samples collection and location

Three hundred wild *O. niloticus* and *O. aureus* fish (150 fish/ each species) were caught with an average weight 50± 2.5g. Fish samples were collected from water drainage canals in Damietta Governorate. Collected fish were transported for parasitological analysis to the fish disease and management Lab, CLAR, Egypt. The fish were kept in well-prepared glass aquaria supplied with water taken from the fish collecting area. Each aquarium was regularly aerated using an air pump.

Clinical and postmortem examination of naturally infested fish

The experimental fish were euthanized in a bath containing a clove oil solution (12.5mg/ L) and then individually subjected to clinical and postmortem examinations to detect any external or internal abnormalities. These examinations were carried out in accordance with the procedures specified by **Amlacher (1970)**.

Parasitological investigations

Macroscopic and microscopic examination

Macroscopic examination was performed following the standard scheme outlined by **Syme (1966)** to identify parasites in various parts of the fish body with notched eyes and hand lenses. A scalpel blade was used to scrape the fish's body from the area just behind the operculum to the tip of the tail fin. Microscopic smear slides were prepared from scales and mucus, branchial cavity, eyes, stomach, intestine, internal organs, and musculature. For an additional identification, permanent slides were made and subjected to staining **(Lucky, 1977)**. Additionally, the metacercaria extracted from each eye were counted as separate lots and placed in a Petri dish containing saline solution before being fixed in 70% alcohol, and then examined with a stereoscopic microscope. All samples were fixed directly in 70% alcohol, without flattening, to prevent errors in morphometric measurement.

Identification of the isolated parasites

All detected parasites were examined microscopically and identified based on morphological and morphometric features, as described by **Witenberg (1929)** for encysted metacercariae, **Thoney and Hargis Jr (1991)** for monogenean, and **Lom and Dyková (1992)** for protozoa.

Water analysis

Sixty water samples were seasonally collected from the investigated area in the Damietta Governorate at the same time together with fish samples. Five hundred mL of water samples were collected in sterile, dark stoppered glass from a depth of of 1.0 meter in the canals. These samples were transferred in an insulated icebox to the Fish Farming and Technology water analysis lab, Suez Canal University for the evaluation of un-ionized ammonia (NH3-N), NO_2, No_3, total hardness, total alkalinity, quantitative determination of Fe, Mn, Zn, Cu, Cd, and Pb. Additionally, on-site measurements were achieved for water visibility, temperature, dissolved oxygen (DO), salinity, and pH **(Boyd, 2020)**.

Histopathological examination

Tissue samples, including gills, musculature, liver, and kidney, obtained from naturally infested fish, were fixed in 10% phosphate-buffered formalin. Following a 48-hour immersion in running water, the tissue specimens underwent dehydration using alcohol gradients of different concentrations and were cleared in xylene. Subsequently, they were embedded in paraffin wax and sectioned into thin pieces with a thickness of 5 microns. The sections were initially stained with H&E and then subjected to microscopic examination **(Suvarna *et al.* 2018**), and photographed by a Zisse prime star research microscope (Carl Zeiss, MicroImaging, 37081, Gmbh, Germany) fitted with an AxioCam digital camera (Zeiss,ERc5s, Germany).

Statistical analysis

Data of water parameters analysis were expressed as mean ± standard error (SE) and analyzed using one-way analysis of variance (ANOVA) for all tested samples **(Snedecor & Cochran, 1989)**. Means separations were assessed by Duncan's multiple range test according to **Duncan (1955)**. The present data were analyzed using SPSS (25) for windows. Results were considered significant at the probability level of 0.05 ($P\leq 0.05$).

RESULTS

Clinical and postmortem examinations of experimental fish

The clinical signs in the naturally infested fish (*O. niloticus* and *O. aureus*) revealed abdominal distension, hemorrhagic areas on gill cover, pectoral, caudal and dorsal fins. In addition to slight unilateral exophthalmos and white cloudiness, characteristic small white dots were observed against the eye pupil. Unilateral atrophy in the eye was also noticed (Fig. 1A, B).

Internally, the postmortem examination of infested fish revealed paleness of gills and congestion in addition to an enlargement of the liver, spleen, and kidneys (Fig. 1B, C).

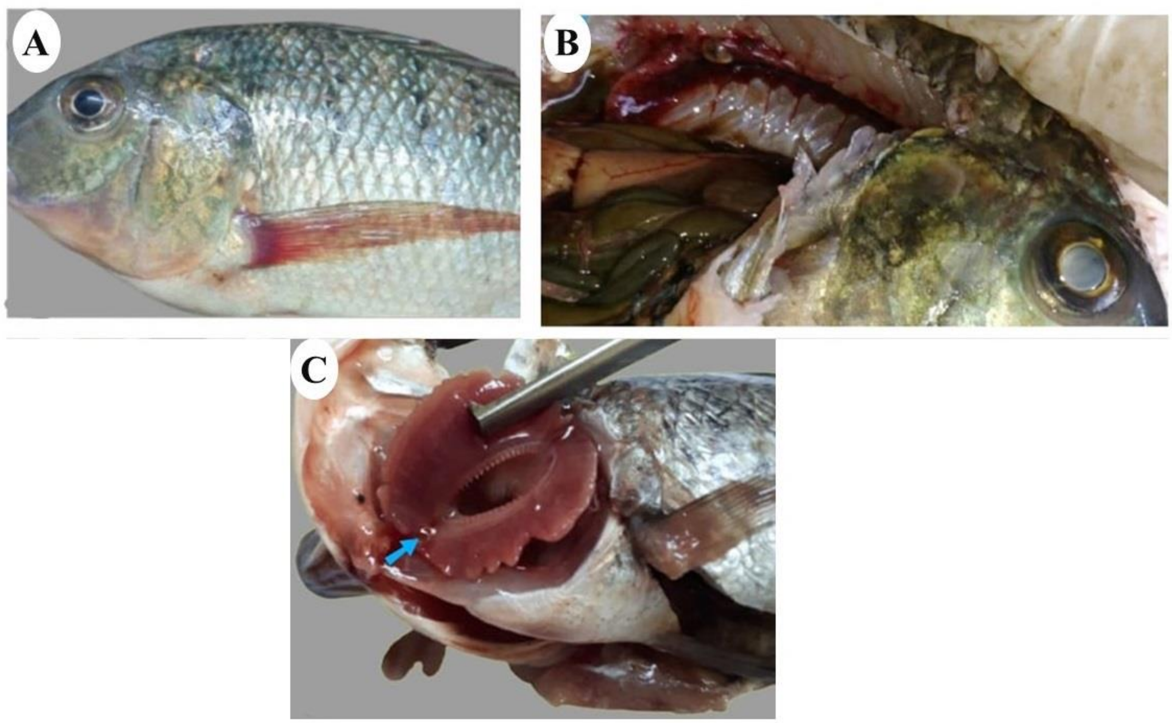

Fig. 1. Postmortem indicators of parasitic infestations among examined Tilapia species including hemorrhages in **(A)** Pectoral fins; **(B)** Pale liver with cloudiness in eye, and **(C)** Paleness in gills

Parasitological investigations

The infested fish were macroscopically and microscopically examined for the presence of parasites, and the isolated parasites were morphologically identified (Fig. 2).

Morphological description of the collected parasites

1. Family: Chilodonellidae, genus: Chilodonella (*C. hexasticha)* (Fig. 2A).

Motile ciliates were isolated from *O. niloticus* gills which are asymmetrically oval bristle-like or heart-shaped and have a notch in the posterior margin. The ventral surface is concave, while the dorsal surface is convex and has a single short row of cilia. A single oval or round macronucleus with a single micronucleus was also observed.

2. Family: Ancyrocephalidae, genus: Cichlidogyrus (*C. tilapiae)* (Fig. 2B).

Elongated gill flukes were isolated from both *O. niloticus* and *O. aureu* with four black eye spots (pigment spots). The anterior end (prohaptor) was mostly divided into four cephalic lobed heads with sticky and adhesive organs (cephalic glands), a mouth near the anterior edge, and a pharynx. The posterior part (opisthaptor) appears discshaped (dome-shaped) with two large pairs of hamuli (central hooks) and seven pairs of small marginal hooklets.

3. Family Clinostomidae, the genus Clinostomum (*C. tilapiae*) (Fig. 2C).

Yellowish encysted metacercariae were isolated from the branchiostegal musculature of *O. niloticus* and *O. aureus* samples. They appeared as small to large yellow to orange pea or pomegranate seed-like objects. The excysted (free) metacercariae are tongue-shaped with rounded ends. They have a smaller oral sucker, a developed ventral acetabulum, a short pharynx with no-esophagus, and a bifurcated digestive tract with long tubular intestines.

4. Family: Clinostomidae, the genus Euclinostomum (*E. heterostomum)* (Fig. 2D).

Grayish-white metacercariae were also collected from the kidneys. The excysted form was leaf-shaped and large. The ventral sucker was very large (about 5 times the size of the oral sucker).

Additionally, from the musculature and liver, various digenetic metacercariae species belonging to the family: Heterophyidae, such as Heterophyes spp., *Haplorchis* spp., and *Centrocestus formosanus* were also detected.

Prevalence of parasitic infection among inspected tilapia species

The results revealed that *O. niloticus* was the most infested species (66.7%) compared to *O. aureus* (53.3). Moreover, among *O. niloticus* samples, the highest prevalence was recorded for encysted metacercaria (80%), monogenea (66.7%), followed by digenetic trematodes (26.7%), and protozoa (2%). Similarly, encysted metacercaria (66.7%) and monogenea (53.3%) were the major parasitic infestations detected in *O. aureus* samples, with the lowest frequencies for digenetic trematodes (20%) and protozoa (0.67%).

Table 1. Total prevalence of parasitic infestation in examined tilapia species

Tilapia species	No. of fish	Total parasitic infestation		Protozoa		Monogenea		Digenetic trematodes		E.M.C*	
		No.	%	No.	%	No.	%	No.	%	No.	%
O. niloticus	150	100	66.7	3	2	100	66.7	40	26.7	120	80
O. aureus	150	80	53.3	1	0.67	80	53.3	30	20	100	66.7
Total	300	180	60	4	2.67	180	60	70	23.3	220	73.3

*E.M.C: encysted metacercaria.

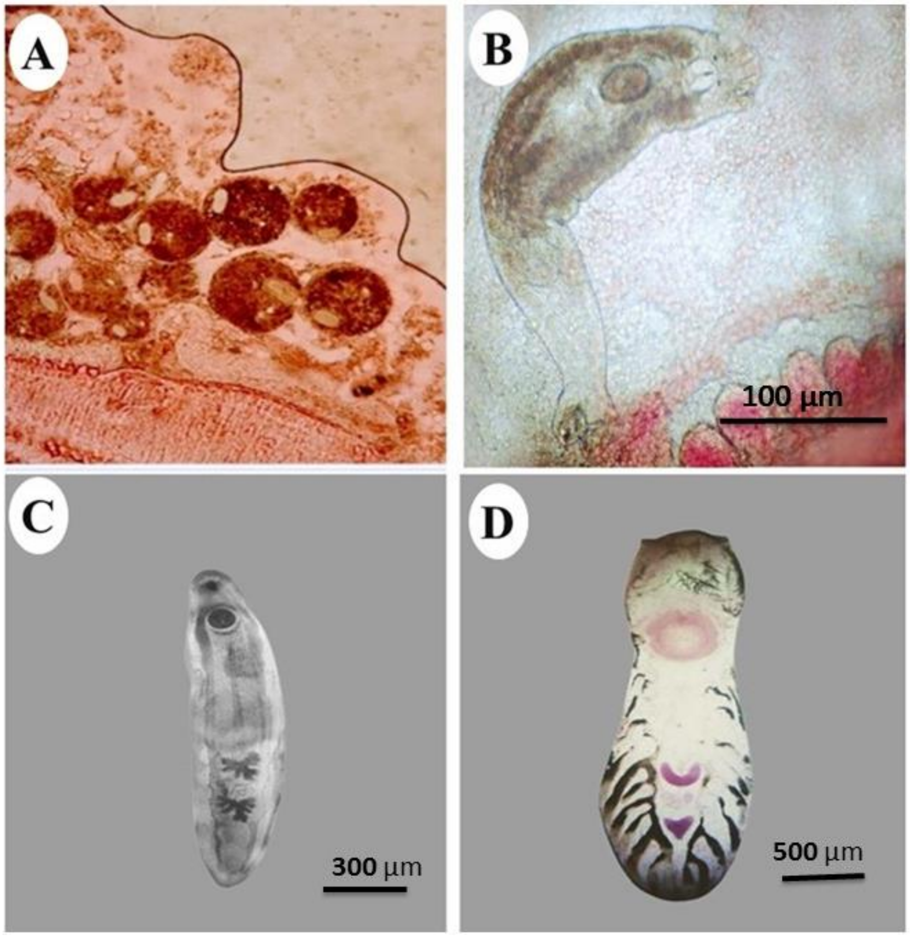

Fig. 2. Macroscopic and microscopic photos of recovered parasites from different tissues of examined tilapia species. **(A)** Gills of *O. niloticus* contain *Chilodonella hexasticha*. **(B)** Gills of *O. niloticus* showing *Cichlidogyrus tilapiae*. **(C)** *Clinostomum tilapiae* metacercaria isolated from branchiostegal musculature of *O. aureus*, **(D)** *Euclinostomum heterostomum* metacercaria isolated from kidneys of *O. niloticus*

Seasonal prevalence of parasitic infection among inspected tilapia species and physicochemical water parameters

The seasonal prevalence of parasitic infestation and analysis of water physicochemical parameters are illustrated in

Fig. 3, Fig. 4) and

Table 2). Both fish species exhibited the highest infestation in summer, and spring followed by autumn, while the lowest incidence was in winter. During winter, the prevalence was 37.5% in *O. niloticus* and 31.4% in *O. aureus*. The most prevalent parasites were *C. tilapiae*, and EMC. Mean physicochemical water parameters were: SD 21cm, Temp. 14.9^{o}C, DO 8.47mg/ l, salinity 0.8ppm, pH 9.43, UIA 0.15, NO2 0.03, NO3 0.15, T.H 308, T.A 321.7, Fe 0.17, Mn 0.19, Zn 0.23, and Cu 0.023mg/ L. In spring, the prevalence was 78.04% in *O. niloticus* and 62.9% in *O. aureus*. The most prevalent parasites were *C. tilapiae*, and EMC. Moreover, the water parameters were: SD 17cm, Temp. 23.87^{o}C, DO 7.1mg/ l, salinity 0.63ppm, pH 8.43, UIA 0.19, NO2 0.02, NO3 0.33, T.H 232.33, T.A 248.3, Fe 0.14, Mn 0.16, Zn 0.31, and Cu 0.023mg/ L.

While, the prevalence in summer was 80% in *O. niloticus* and 64% in *O. aureus*. The most prevalent parasites were *C. tilapiae*, and EMC. The water parameters were: SD 11.3cm, Temp. 27.6^{o}C, DO 6.23mg/ l, salinity 0.33ppm, pH 8.5, UIA 0.29, NO2 0.03, NO3 0.55, T.H 240, T.A 271.7, Fe 0.13, Mn 0.15, Zn 0.38, and Cu 0.022mg/ L. Along autumn, the prevalence was 62.5% in *O. niloticus* and 50% in *O. aureus*. The most prevalent parasites were *C. tilapiae*, and EMC. While protozoa and Haplorichis spp. were not detected. Water parameters were: SD 13.37cm, Temp. 24.8^{o}C, DO 6.97mg/ l, salinity 0.5ppm, pH 8.47, UIA 0.17, NO2 0.01, NO3 0.35, T.H 241.7, T.A 328.3, Fe 0.13, Mn 0.13, Zn 0.39 and Cu 0.021mg/ L, while Cd and Pb are not detectable during the investigation period.

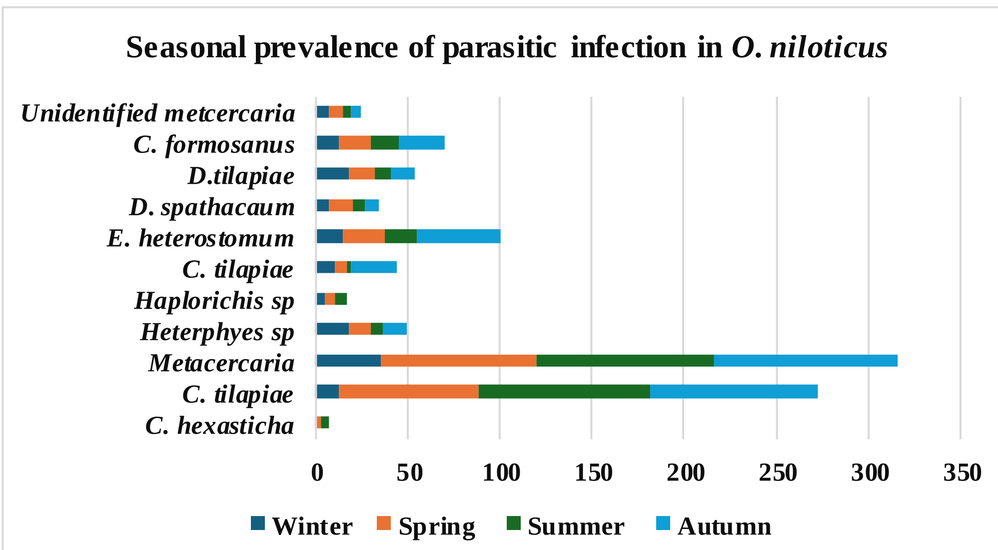

Fig. 3. Seasonal prevalence of parasitic species in *O. niloticus*

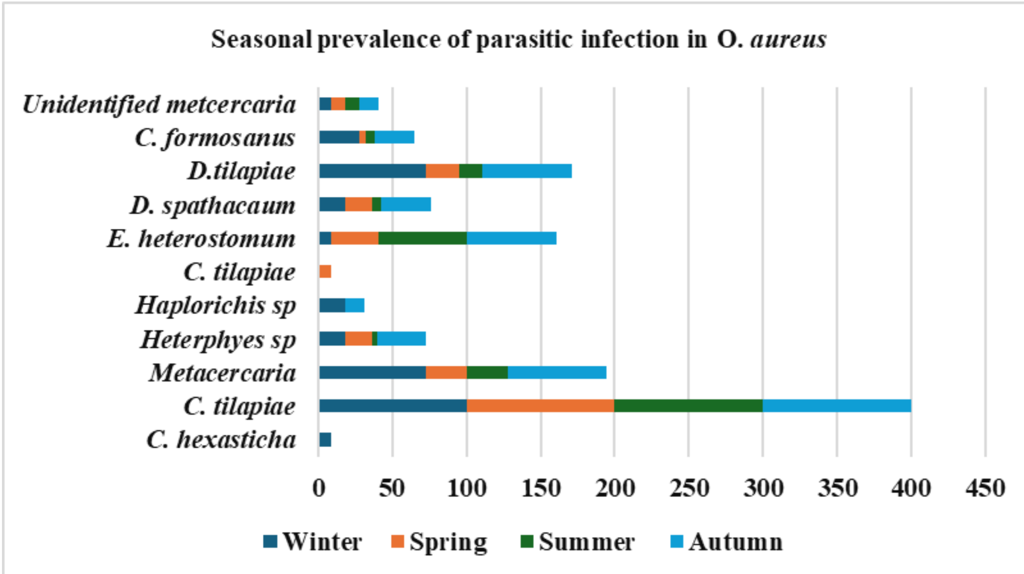

Fig. 4. Seasonal prevalence of parasitic species in *O. aureus*

Table 2. Seasonal prevalence of parasitic infection among tilapia species and their associated physicochemical water parameters

Season	*O. niloticus*			*O. aureus*			Physicochemical water parameter													
	No. of examined.	No. of infected	%	No. of examined.	No. of infected	%	SD (cm)	Temp. (°C)	DO (mg/L)	Salinity (ppm)	pH	UIA (mg/L)	NO_2 (mg/L)	NO_3 (mg/L)	T.H (mg/L)	T.A (mg/L)	Fe	Mn	Zn	Cu
Winter	40	15	37.5	35	11	31.4	21 ± 0.02^{a}	14.9 ± 0.12^{d}	8.47 ± 0.17^{a}	0.08 ±0.06	9.43 ± 0.23^{a}	0.15 ± 0.01^{b}	0.03 ± 0.00^{a}	0.15 ± 0.04^{c}	308 ± 3.05^{a}	321.67 ± 2.73^{a}	0.17 ± 0.01^{a}	0.19 ± 0.02^{a}	0.23 ± 0.00^{c}	0.023 ±0.00
Spring	41	32	78.04	35	22	62.9	17 ± 0.29^{b}	23.87 ± 0.22^{c}	7.1 ± 0.4^{b}	0.63 ±0.08	8.43 ± 0.24^{b}	0.19 ± 0.01^{b}	0.02 ± 0.00^{ab}	0.33 ± 0.01^{b}	232.33 ± 24.13^{b}	248.33 ± 8.99^{b}	0.14 ± 0.00^{b}	0.16 ± 0.00^{b}	0.31 ± 0.01^{b}	0.023 ±0.00
Summer	45	36	80	50	32	64	11.3 ± 0.12^{d}	27.57 ± 0.18^{a}	6.23 ± 0.03^{c}	0.33 ±0.03	8.53 ± 0.05^{b}	0.29 ± 0.02^{a}	0.03 ± 0.01^{a}	0.55 ± 0.00^{a}	240 ± 5.77^{b}	271.67 ± 11.67^{b}	0.13 ± 0.00^{b}	0.15 ± 0.00^{c}	0.38 ± 0.00^{a}	0.022 ±0.00
Autumn	24	15	62.5	30	15	50	13.37 ± 0.07^{c}	24.77 ± 0.07^{b}	6.97 ± 0.24^{bc}	0.5 ±0.01	8.47 ± 0.35^{b}	0.17 ± 0.01^{b}	0.01 ± 0.00^{b}	0.35 ± 0.02^{b}	241.67 ± 22.05^{b}	328.33 ± 11.67^{a}	0.13 ± 0.00^{b}	0.13 ± 0.00^{d}	0.39 ± 0.00^{a}	0.021 ±0.00

Data were represented as mean ± S.E. Means carrying different superscripts in the same column are significantly different at ($P \leq 0.05$). SD: Water visibility, Temp: Temperature, DO: Dissolved oxygen, UIA: Un-ionized ammonia, T.H: Total hardness, and T.A: Total alkalinity.

Histopathological examination

The histopathological examination of different tissue specimens from naturally infested tilapia species is illustrated in Fig**. 5**. The gills infected by *C. hexasticha* showed fusion of the gill lamellae and lamellar telangiectasia. Moreover, there is an extensive degeneration and necrosis of the lamellar epithelium, leading to the sloughing of the secondary lamellae. Epithelial hyperplasia was also evidenced, while the degenerative changes subsequently led to erosion of this layer (Fig. 5A). The musculature of *O. aureus* displayed obvious intermuscular non inflammatory edema in addition to diffused hyaline degeneration in the muscle bundles (Fig. 5B). Liver of tilapia species showed parasitic cyst encapsulated with connective tissue and infiltrated by inflammatory cells. Pressure atrophy with marked focal necrosis affected the adjacent hepatocytes with congestion in hepatopancreatic blood vessels. Kidney of tilapia infested by *EMC*, showing focal activation of melanomacrophages nearby the parasite.

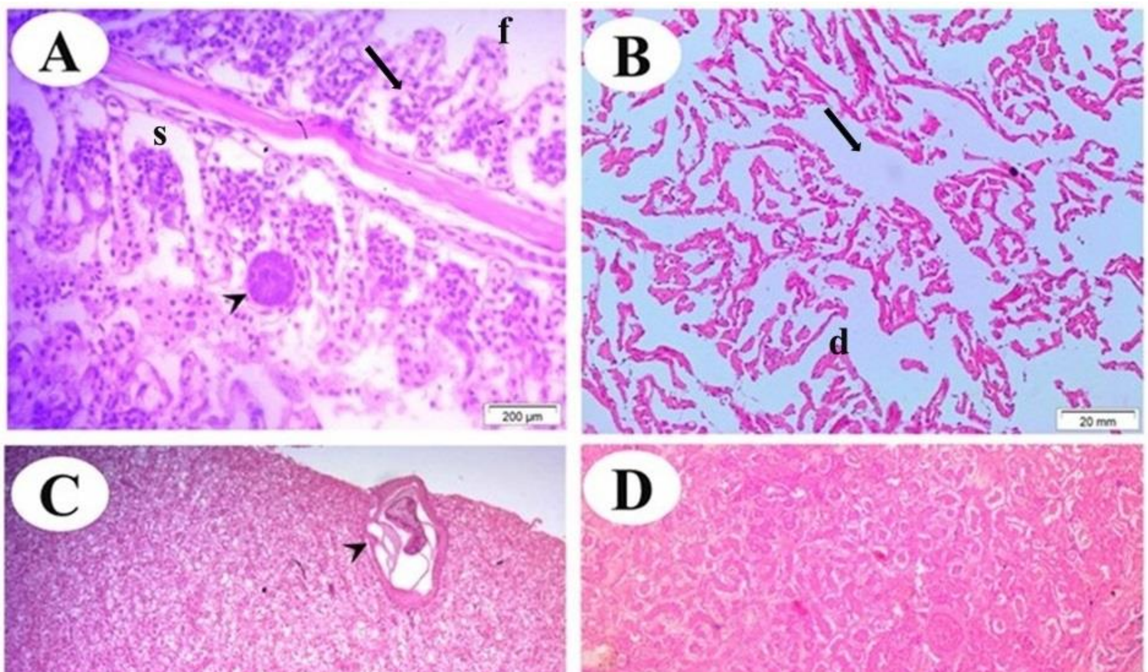

Fig. 5. Histopathology of different tissues from examined tilapia species, H & E stain. **(A)** Gills infested by *C. hexasticha* (arrowhead) with progressing degradation of the lamellar epithelium with lamellar telangiectasia (long arrow), f: fusion, s: sloughing, **(B)** Musculature showing intermuscular edema (arrowed). **(C)** Liver showing large encysted metacercaria (EMC) embedded in the hepatic parenchyma that contained remnant of the larvae (arrow), c: congestion, and **(D)** Kidney infested by *E. heterostomum* with melanomacrophages infiltrations

DISCUSSION

In this study, a strong relationship was identified between the existence of parasites and water pollution, both of which had detrimental effects on the histopathological state of the affected fish tissues.

The parasitic infestation among affected fish showed similar clinical signs to those previously recorded **(Aly *et al.*, 2020)** in addition to symptoms of respiratory distress. These signs might be attributed to the tissue damage brought on by external parasites and gill parasites that interfere with breathing and respiration. Internally, in some fish samples, the presence of different encysted metacercariae in various visceral organs was detected, which was also reported in previous studies **(Eissa, 2006; El Deen *et al.*, 2011)**. The mucus hypersecretion that was noticed in the anemic gills is a step in the innate immune protection against parasite infestation, as it contains bioactive substances, such as lysozyme, complement, C-reactive protein, hemolysins, and lectins that affect the establishment and proliferation of many ectoparasitic copepods, ciliates, and monogenea **(Jones, 2001)**.

The parasitological exploration of the gill tissues from both tilapia species exhibited infestation of the ectoparasitic ciliate, *Chilodonella hexasticha*, and with monogenean gill fluke, *Cichlidogyrus tilapiae*. Similar parasites were previously isolated **(Rowland *et al.*, 2006; Pádua *et al.*, 2013)**. Moreover, several yellowish EMC and grayish-white EMC were detected from the branchiostegal and renal tissues. These findings are constant with that detected by **Bichi and Ibrahim (2009)** from *Tilapia zillii* and **Noor El-Deen *et al.* (2015)** from *O. niloticus*. Furthermore, some musculature specimens from both fish species harboured elongated, leaf-like metacercariae, moving in a leech like manner that was described by **Khan *et al.* (2017)** as *Diplostomum tilapiae*. Additionally, metacercariae were detected from eyes and defined as *Diplostomum spathaceum*, previously found in tilapia **(Ndeda *et al.*, 2013; Mavuti *et al.*, 2017)**.

High stocking densities as well as water parameters deterioration favor increased parasite populations **(Costa *et al.*, 2019)**. The water's visibility varied significantly throughout the year. Winter recorded the highest (21± 0.02cm) values, while the summer-time reading recorded the lowest (11.3± 0.12cm) values. Similar findings were detected by **Radwan *et al.* (2021)**. **Snieszko (1974)** hypothesized that due to the turbidity of the water caused by the high load of organic waste and improper management of fish culturing, fish become more vulnerable to infections and disease outbreaks. **Ojwala *et al.* (2018)** stated that several parasite incidences were strongly correlated (positively) with specific water quality elements. For instance, cestoda and acanthocephalan were correlated with turbidity, whereas monogeneans, crustaceans, and protozoans were positively associated with dissolved oxygen, pH, and temperature.

Global warming has profound effects on infectious diseases eruption especially in the two major groups that are anticipated to be most impacted are vector- and water-borne infectious diseases **(Kurane, 2010)**. The dynamic of the parasites and their associated vectors, as well as host species were directly and indirectly affected with climate change **(Short *et al.*, 2017)**. Thus, temperature is the abiotic parameter of water quality that most affects the parasitic assembly in farmed fish since it affects the dynamics of disease development, the release of infectious forms, and their motor activity in the fish host, in addition to acting on parasitological metrics (prevalence, percentage severity, and richness) **(Cable *et al.*, 2017)**.

Water temperature varied during different seasons as mean temperature values ranged between 14.9± 0.12°C in winter to 27.57± 0.18°C in summer. The detected water temperature in summer is similar to that (28.02± 0.08°C) measured by **Reda *et al.* (2022)**. Additionally, the variation in water temperature also altered the dissolved oxygen, DO, values in water, thus there were significant variations in its concentrations during different seasons. The lowest concentration was 6.23± 0.03mg/ l during summer, while the highest concentration was 8.47± 0.17mg/ l during winter, a similar finding was previously detected **(Radwan *et al.*, 2021)**. Heat stress, higher water temperatures than optimal temperatures, as well as low DO not only can cause a gradual loss of equilibrium, and abnormal swimming but also, disrupt the fish's homeostatic mechanisms, which makes fish susceptible to parasitic infections **(Logue *et al.*, 1995)**. Additionally, significant differences in heavy metals concentrations (Fe, Mn, Z, and Cu) in different seasons, varied from data detected by **Abdo and El-Nasharty (2002)**, who found the constant levels of Fe, Mn, Zn, Cu, and Pb along the year. These findings may be changed according to locations and industrial activities in the study sites. The degree of contamination in fish rearing systems may be determined using the species diversity and population patterns of parasites, such as protozoa, monogenea, and digenea. It may thus be assumed that all parasite groups have the ability to respond to changes in the aquatic environment **(Shehata *et al.*, 2018)**.

Fish parasites are considered bioindicators of environmental quality **(Marcogliese, 2005)** as parasites diverge affected by seasonal variation. **Hu and Li (2016)** reported that the most critical times for the fish are during the highest infestation levels, hence it is vital to understand their seasonal dynamics in order to decide what interventions are recommended to avoid massive losses, especially in culturing circumstances. In Egypt, the incidence of protozoa among *O. niloticus* aquaculture was temperature-dependent, with higher parasitic infestations in hot months **(Aly *et al.*, 2020)**.

According to the current study, parasite infection was 60% overall. This outcome is higher than what was attained by **Bichi and Dawaki (2010)**. This variation in prevalence could be related to the availability of IMH (snails) and the susceptibility of host to infection together with water pollutants and the suitable water temperature. The results demonstrated that the total prevalence of protozoa infection among examined tilapia species was 2.67%. It was 2% in *O. niloticus* and 0.67 % in *O. aureus*. However, **Bichi and Dawaki (2010)** reported the prevalence of *Chilodonella hexasticha* among *O. niloticus, O. aureus* was 28 and 26%, respectively. Furthermore, **Reda (2011)** recovered *C. hexasticha* from *S. galilaeus* that is considered to be a new fish host. The incidence of protozoa on the gills of immune-stressed fish may be influenced by pollution, which may help these parasites colonize the fish's bodies **(Hashimoto *et al.*, 2016)**. Additionally, there was an increased intensity of protozoa infestation among *O. niloticus* with an accumulation of the nitrogen compounds in the fish raring system **(Ashmawy *et al.*, 2018)**.

The monogenean infection, in the current study, showed 60% infestation in both fish species with 66.7% in *O. niloticus* and 53.3% in *O. aureus*. Similar results were reported by **Shehata *et al.* (2018)**. Likewise, the richness of monogeneans *Cichlidogyrus sclerosus, C. thurstonae* and *Scutogyrus longicornis* in *O. niloticus* and their prevalence upsurge positively with rising the organic matter and rising water temperature in breeding tanks **(Cavalcanti *et al.*, 2020)**. Moreover, the abundance of parasites may be impacted by organic pollution in the aquatic environment as monogenea were the most sensitive to water's total ammonia nitrogen, faecal coliforms, and oxygen content **(Lacerda *et al.*, 2018)**.

In the present work, the total prevalence of E.M.C. among examined tilapia species concurs with that recorded by **Nahla *et al.* (2014)**, who elucidated that the prevalence was 77.37%; these variations might be caused by the fish's immunological condition and the variables influencing cercarial penetrations seasonally as well as the intermediate host (snail). Likewise the EMC's total prevalence among the Nile tilapia was 95% (285 of 300), while infection with macroscopic EMC had a prevalence of 37% **(Abd-Elrahman *et al.*, 2023)**. Moreover, **Bahaa (2012)** pointed out that the highest prevalence of E.M.C. among *O. niloticus* was in autumn, while the lowest rates were during the winter, proving that the temperature has no bearing on the parasite invasion.

The infected tilapia gills with Chilodnella spp. revealed extensive degenerative changes and hyperplasia with fusion in their lamellae. These alterations are similar to those recorded in the study of **Roberts (2012)**. Similarly, an early study elucidated that the gills epithelial cell's common response to damage caused by protozoan parasites is hyperplasia and hypertrophy **(Shinn *et al.*, 2023)**. Additionally, alterations caused by the presence of EMC within hepatic tissue besides the congestion of pancreatic blood vessels. These findings were also detected by **Younis *et al.* (2020)**. Moreover, similar changes were detected in the renal tissue and the EMC causing tubular nephritis accompanied with chronic inflammatory signs with fibrous tissue formation surrounding the cyst. All detected histopathological changes coincide with previous studies that demonstrated the damage caused by parasitic invasion of the viscera, including the gills, liver, musculature and kidney **(Younis *et al.*, 2020; Younis *et al.*, 2022; Shinn *et al.*, 2023)**.

CONCLUSION

Fish parasites serving as indicators in aquatic ecology introduce vulnerabilities. In fish farming, parasites act as bioindicators, demonstrating correlations with water quality parameters and contaminants. Protozoa and *EMC* were the predominant parasites in the examined tilapia species, followed by monogenean parasites. Endoparasites in natural environments proved valuable for detecting heavy metal accumulation. Consequentially, histoccursological alterations in fish visceral tissue occur when these parasites invade, disrupting physiological functions and potentially causing outbreaks.

Ethics approval

All experimental fish in the present study were handled in accordance with the guidelines of the local Administrative Panel on Laboratory Animal Care and Committee of Faculty of Veterinary Medicine, Suez Canal University, Egypt (Code: 2023033). The authors confirm that all methods were performed in accordance with the relevant guidelines and regulations.

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Competing interests

The authors declare no competing interests.

Authors' contributions

All authors have significantly contributed to the study design, methodology, data analysis, drafting, and manuscript editing. All authors have read and approved the final version of the submitted manuscript.

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