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Digentetic Trematodes in *Dicentrarchus labrax* cultured in Egypt: Prevalence, Clinical Features, Body Condition, and Histopathology

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ABSTRACT

The current work aimed to study the types and seasonal occurrence of digenetic trematodes in *D. labrax* cultured in Egypt. A total number of 1800 *D. labrax*, (150/season- 600/farm) of average weight 95.14±10.37 gm were collected from three Egyptian farms during 2019. Four digeneans were isolated from the gastrointestinal tract; *Metadena crassulata Derogenes varicus, Acanthostomum spiniceps,* and *Diplomonorchis sphaerovarium*. The highest infection rate was detected in Farm 2 (30.16 %), specifically at spring (27.76%), where the range of water temperature was 21.12- 24.63 ^{o}C, and salinity 15.85-32.34 ppt. The infected fish were apparently healthy, except in heavy infestation where body weakness and anemia of the internal organs were evident. The infected fingerling recoded the lowest condition factor (0.98) and mean parasitic intensity of 3.9, but higher values were noticed in juvenile, adult, and broodstock. Moreover, weak linear regression was reported between infection intensity and weight (R=0.91), fish length (R=0.964), and condition factor (0.956). Remarkable pathological alterations were observed in the intestine of infected fish, including edema, aggregation of inflammatory cells, mucinous degeneration and sloughed necrotic areas. Therefore, preventive and control measures are recommended to avoid such infections that potentially influence the health status and productivity of infected fishes.

INTRODUCTION

European seabass *Dicentrarchus labrax* (Linnaeus, 1758) is one of the most valuable commercial fish not only in the Mediterranean countries but also worldwide **(Enes *et al.*, 2011; FAO, 2018)**. However, the astonishing increase in the biomass of the mariculture facilities could lead to several risks including stress factors, depressed immune system, pathological problems, and subsequent negative impacts on the water environment **(Sapkota *et al.*, 2008; Aly, 2013; Essa *et al* ., 2018)**. Parasitic infections considered great problems facing aquaculture which may cause morbidities, mortalities, and thus economic losses. Under natural conditions,

they comprise the parasite fauna of total biomass, but they could become pathogens to their hosts, invade various external and internal tissues, reduce the fish quality, and harm the human **(Tansel and Fatih, 2012)**.

Among the common parasitic diseases that reported in various fish species including crustacean Isopoda and Copepoda **(Öktener and Sezgin, 2000; Manera and Dezfuli, 2003)**, ectoparasitic monogenean *Diplectanidae; Diplectanum aequans* and *D. laubieri* were reported in sea bass *D. labrax* **(Whittington and Chisholm, 2008).** Besides, other parasitic species of trematodes, cestodes, acanthocephalans, and nematodes which considered the major parasitic diseases, extremely important in aquatic ecosystems, and may cause tissue damage **(Piasecki, 2004; Noga, 2010; Eissa *et al.*, 2017 a)**.

Digenetic trematodes considered of great significance to marine waters **(Cribb *et al.*, 2002)**. They parasitize a wide range of fish hosts in fresh, marine and brackish water fish, and numerous species are described in various fishes worldwide. Sea bass *Dicentrarchus labrax* were susceptible to various digenetic infections including *Acanthostomum spiniceps* **(Morsy *et al.*, 2013)**, *Derogenes varicus* **(Sterud 2002)**, *Pseudoacanthostomum panamense* **(Carvallho *et al*., 2014)**, *Timoniella imbutiforme* **(Erik 2002; Maather 2007)**, *Metadena crassulata, Pseudoacanthostomum panamense, Transversotrema patialense, Pseudallacanthochasmus grandispinu*s, and *Timoniella praeterita* **(Abou Zaid *et al.*, 2018)**. Digeneans are usually asymptomatic infections, mostly inhabit the gastrointestinal tract, rarely infecting the swim bladder, ovary, peritoneal cavity, urinary bladder, or circulatory system **(Noga, 2010).** In some cases, in heavy infection, adult digenetic trematodes may embed themselves in the lining mucosa of the stomach and intestine resulting in anemia, causing local damage and possibly peritonitis, moreover, proteolytic enzymes discharged from adult worms may be degrading the gastric and intestinal tissues **(Woo, 1995).** These parasites commonly have two reproductive generations, larvae and adults, where their life cycles are complex and need at least two hosts **(Noga, 2010).** Their outbreaks are influenced by some factors include nutrient availability, the density of primary intermediate hosts such as snails and crustaceans, secondary consumers such as fish together with biological and ecological factors **(Zander *et al.*, 2000).**

Thus, this study aimed to investigate the digenetic trematodes among cultured European sea bass *Dicentrarchus labrax* in Egypt and their pathological effect and seasonal occurrence.

MATERIAL AND METHODS

Study location:

The study was conducted on three different farms culturing Sea bass *Dicentrarchus labrax* under different rearing systems. The first farm (Farm1) is located in Borg El-Arab, Alexandria Governorate, 30° 50′ 56″ N, 29° 36′ 42″ E. The second farm (Farm 2) is located in

Edeeba-Triangular, in Shata Damietta Governorate 31° 24′ 32″ N, 31° 52′ 19″ E. The third farm (Farm3) is located at Wadi-El-Rayan, Fayoum Governorate 29° 8′ 52″ N, 30° 23′ 33″ E.

Fish sampling:

The fish sampling procedures and handling were conducted according to the addressed guideline of animal welfare legislation **(Branson, 2008)**. A total number of 1800 *D. labrax*, (150/season- 600/farm) of average weight 95.14±10.37 gm were collected from the three investigated farms during 2019. *D. labrax*, apparently healthy, moribund, and freshly dead were sampled regularly throughout the four seasons from the three farms under the study. They were kept in ice boxes and transported quickly to the Laboratory of Fish Diseases, Aquaculture Division, National Institute of Oceanography and Fisheries, Alexandria Branch, as well as Inland Branch, Shakshouk Fish Research Station, Fayoum Governorate, NIOF, Egypt.

Water sampling:

Physical and chemical water parameters were measured at collection sites such as water temperature, oxygen, pH, and salinity, using specific Electronic probe **(APHA, 1995).** For assurance, another water samples from the same ponds were also collected in glass or plastic bottles, kept in insulated coolers, transported directly to the laboratory, then examined using an environmental testing photometer (Hanna Instruments), within 24 hours.

Parasitological examination:

Gut examination:

Fish samples were anesthetized TMS, (MS-222) at dose 100 mg/L. To expose the alimentary canal, the fish were dissected out to expose the three parts, the proximal part, the middle part, and the distal part **(Pirarat *et al.,* 2011).** Each section was placed separately into dishes containing normal saline, incised, and examined for parasites under a dissecting microscope.

Parasitological identification:

Isolated parasites were counted, and placed in physiological saline overnight in a refrigerator to enable it to stretch and relax. Thereafter, the parasites were fixed in 5% formalin. Parasites were then stained overnight with a weak Erlich's hematoxylin solution, passed through graduated alcohol (30, 50, 70, 90%, and absolute) for 45 min to dehydrate, and cleared in methyl-salicylate and mounted on a slide in Canada balsam. The identification of the isolated parasites was performed according to **Schmidt, (1992) and Woodland (2006).**

Condition factor (Weight–length relationships):

Throughout the study period, fish samples were measured for length and weight. Then, the weight–length relationships (WLRs) of collected *D. labrax* were analyzed at different growth phases **(Froese 2006).**

Histopathological examination:

It was performed following the procedures described by **Bancroft and Gamble, (2007) and Dimitroglou *et al.* (2010)**. Specimens from the gills, liver, kidney, and intestine were carefully removed and preserved in 10% neutral buffered formalin. Following the conventional paraffin embedding technique, they were dehydrated in a graded alcohol series and cleared in xylene then embedded in paraffin**.** Transverse sections of 5 μm thicknesses were fixed in microscopic slides and stained with Mayer's hematoxylin and eosin (H&E). The slides were visualized using a light microscope and photographed with the Nikon digital camera. The processing and analysis of the Photomicrograph were performed using Image J. software version 1.36 (NIH, USA).

Statistical analysis:

Statistical analysis was performed using Package for Social Sciences (SPSS) version 14.0 **(Coakes, and Steed, 2009)**. Where the differences among examined groups were tested using the analysis of variance (ANOVA). Then, the significance of the difference between values was assessed with the LSD posthoc test (least significant difference test) for the detection of the particular differences between groups. The values were represented as the mean ± standard error (mean ± S.E.M). Also, the significance was indicated in figures and tables by an asterisk (*), and was described as (p <0.05).

RESULTS

1. *Parasitological examination:*

Four parasites were isolated from sea bass, *Dicentrarchus labrax* (L.) belonged to Platyhelminthes, digenetic trematodes. They include the following parasites:

1.1. *Metadena crassulata:*

It was isolated from the intestine, mainly the anterior part. The body is oval, or elongated oval has round oral sucker which opens terminally or subterminally. Also, it has short esophagus, and pharynx, blind caeca, intestinal bifurcation dorsal to ventral sucker. Two symmetrical testes anterior to hind-body, and seminal vesicle were seen. Slightly posterior to the testis, deeply lobed ovary and two lateral groups of vitelline follicles were observed **(Figs. 1 a&b)**.

1.2. *Derogenes varicus:*

This digenean was isolated from the stomach and intestine. It was a fusiform shape, sized about 0.960-2.810 mm length, and 0.440-0.800 mm width. The anterior part has an oral sucker, open to the elongated pharynx, and longer esophagus. Close to the pharynx, the intestinal bifurcation was situated with vitelline and terminal caeca. The testes were two smooth ovals, the anterior testis smaller than the posterior one. The ovary was also smooth, almost spherical, with two vitelline glands. The ventral sucker was oval lies in the middle of the body **(Figs. 1 c&d)**.

1.3. *Acanthostomum spiniceps:*

It was isolated from the intestine. It has elongated body sized 3.37 x 0.57 mm, and funnel-shaped oral sucker bearing a crown, about 27 spines. The prepharynx was long, with short esophagus which bifurcates in front of the acetabulum, and the cecal opening at the posterior end. A single ovary was located anteriorly to two tandem testes, where the anterior testis was smaller than the posterior one **(Fig. 1 e)**.

1.4. *Diplomonorchis sphaerovarium:*

It was isolated from the intestine. It has an elongated body, the diameter was 1.82 -2.93 mm length, and 0.45- 0.69 mm width. Anteriorly, there was an oral sucker, then prepharynx, pharynx, esophagus, and posteriorly ventral sucker **(Figs. 1 f&g)**.

2. The prevalence of digenetic trematodes in D. labrax:

The seasonal occurrence of the digenean infection in *D. labrax* showed significant to non-significant differences among different seasons and various localities. Spring recorded the highest digenean prevalence of 27.76 %, and the lowest rate 14.44 % in winter. The spatial occurrence reported the highest prevalence of 30.16 % in Farm 2, Edeeba-Triangular, 18.64 % in Farm 1, Borg El-Arab, and the lowest rate of 5.83 % in Farm 3, Wadi-el-Rayan **(Table 1)**.

3. Water physic and chemical basic tests:

The recorded water quality parameters were significantly differed ($p \leq 0.05$) in the three farms throughout various seasons. The lowest water temperature 14.22 ^{o}C was recorded in the winter in Farm 1, Borg El-Arab, and the highest 29.7 ^{o}C in summer at Farm 2, Edeeba-Triangular. The recorded pH was the lowest value of 7.49 in winter at Farm 3, Wadi-El-Rayan, and the highest 8.36 was in summer at Farm 2. Also, the lowest salinity 13.54 ppt was reported in winter at Farm 3, and the highest value 34.36 ppt in summer in Farm2 **(Table 2)**.

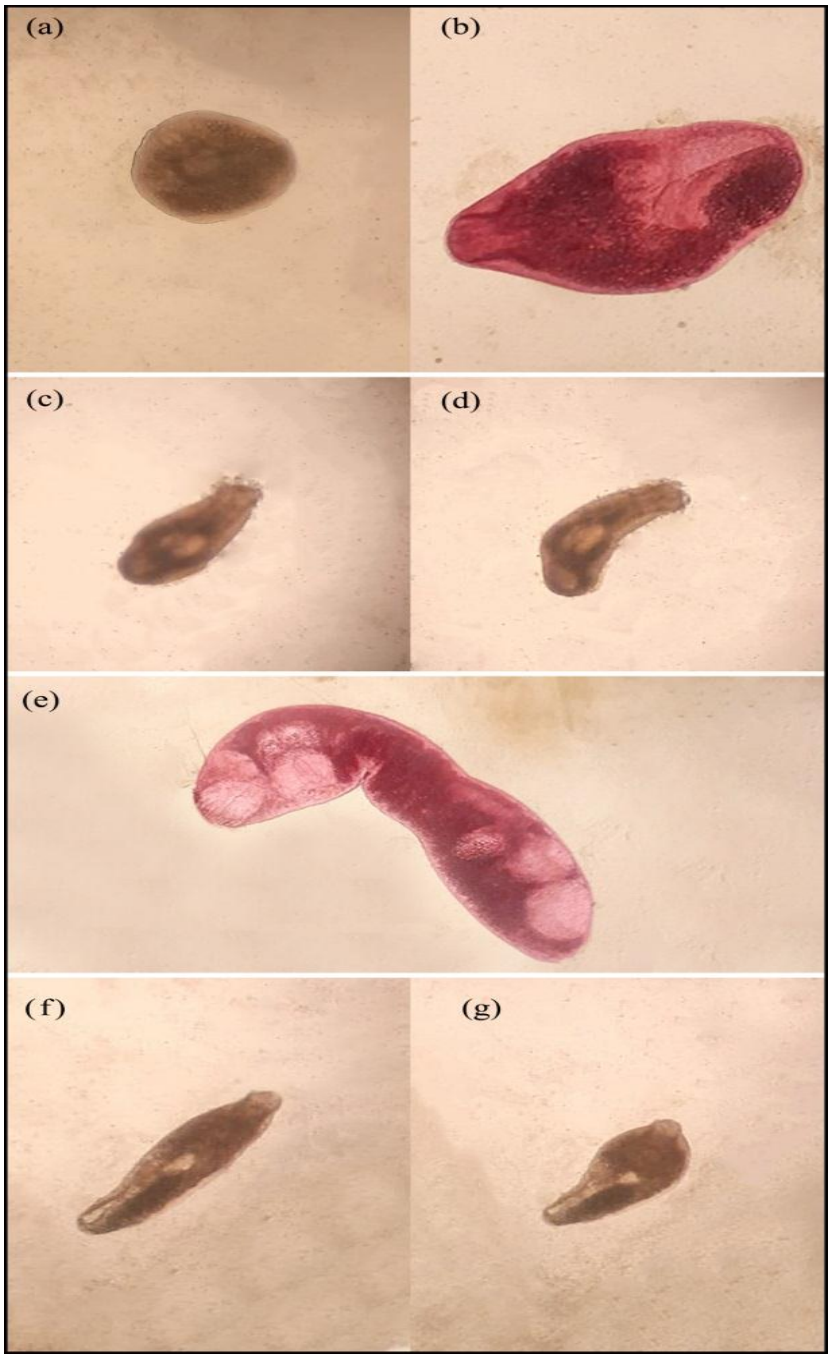

Figure 1: (a&b): digenetic trematode, *Metadena crassulata;* **(a)** non stained intestinal scraping from European seabass containing digenetic trematode, *Metadena crassulata* (X10), **(b)** carmine stained *M. crassulata* (X 200), **(c&d):** non-stained intestinal scraping containing digenetic trematode, *Derogenes varicus* (X10) **(e):** stained digenean, *Acanthostomum spiniceps* with carmine (X 200), **(f&g)**: non-stained intestinal scraping containing digenetic trematode, *Diplomonorchis sphaerovarium* (X 10).

Table 1: The seasonal occurrence of digenetic trematodes in sea bass, *Dicentrarchus labrax* (L.) cultutred in Egyptian farms throughout 2019.

Farm	Winter	Spring	Summer	Autumn	Total
1 (Borg El-Arab) Alexandria Governorate	18.66^{a}	25.30^{b}	19.33^{b}	11.33^{b}	18.64^{**}
2 (Edeeba-Triangular) Damietta Governorate	24.66^{a}	48.66^{a}	19.33^{b}	28.00^{a}	30.16^{***}
3 (Wadi-El-Rayan) Fayoum Governorate	0.003^{b}	9.33^{b}	6.00^{c}	8.00^{b}	5.83^{*}
Total	14.44^{**}	27.76^{***}	14.89^{**}	15.78^{**}	

$^{(a, b, C, d)}$: Average in the same row having different superscripts significantly different at ($P \leq 0.05$)&
*: There is a significant difference at ($P < 0.05$)

Table 2: The physiochemical parameters of examined water throughout different seasons in the three farms* under the study during 2019.

Parameters / **Farms**	**Temperature (ºC)**			**pH**			**Salinity (ppt)**		
	Farm 3	**Farm 2**	**Farm 1**	**Farm 3**	**Farm 2**	**Farm 1**	**Farm 3**	**Farm 2**	**Farm 1**
Winter	15.12^{d}	14.35^{c}	14.22^{d}	7.49	8.10^{b}	7.52^{c}	13.54^{d}	29.66^{c}	13.85^{b}
Spring	21.12^{c}	24.63^{b}	22.73^{c}	7.55	7.98^{b}	$7.74b^{c}$	15.89^{c}	32.34^{b}	15.85^{ab}
Summer	28.59^{a}	29.70^{a}	28.69^{a}	7.87	8.36^{a}	8.16^{a}	18.06^{a}	34.36^{a}	17.69^{a}
Autumn	25.66^{b}	25.77^{b}	25.62^{b}	7.67	7.96^{b}	7.99^{ab}	16.73^{b}	31.78^{b}	16.88^{ab}
LSD	**1.57**	**1.79**	**1.34**	**0.45**	**0.29**	**0.48**	**0.68**	**1.21**	**1.06**

(a, b, C, d): Average in the same column having different superscripts significantly different at ($P \leq 0.05$).
LSD: Least significant difference.
***Farm1**, Borg El-Arab, Alexandria; **Farm2**, Edeeba-Triangular, Damietta; **Farm 3**, Wadi-El-Rayan, Fayoum

4. Clinical signs and gross lesions:

The infected fish mostly revealed non-pathognomonic signs or lesions. Sometimes, they displayed debilitation, decreased appetite, and pale gills. In heavy infection cases, abdominal distension, pale liver, kidney, and spleen, and anemia might be evident (Fig. 2)**.**

Figure 2: Infected adult *D. labrax* with digenetic trematode, showing debilitation, and skin erosion. The scale bar is 2 cm

5. Correlation between parasite intensity, fish size, and condition factor:

The recorded mean values of the condition index showed overall significant differences between the different growth stages. The lowest value (0.98) was recorded in *D. labrax* fingerlings, whereas higher values 1.27, 135, and 1.39 were noticed; respectively, in juvenile, adult, and broodstock. Comparing these condition factors values with the corresponding infection intensity, it was generally found to be inversely proportional. The highest infection intensity was recorded in fingerlings, while those values increased at other growth stages. The correlation of digenean infection intensity to *D. labrax* weight, length, and condition factor revealed a general weak linear correlation. Particularly, between infection intensity and fish weight, there was a lower correlation (R=0.91) at different fish stages. Also, the Lower correlation has appeared between the fish length and condition factor with linear regression R=0.964, and 0.956; respectively **(Fig. 3)**.

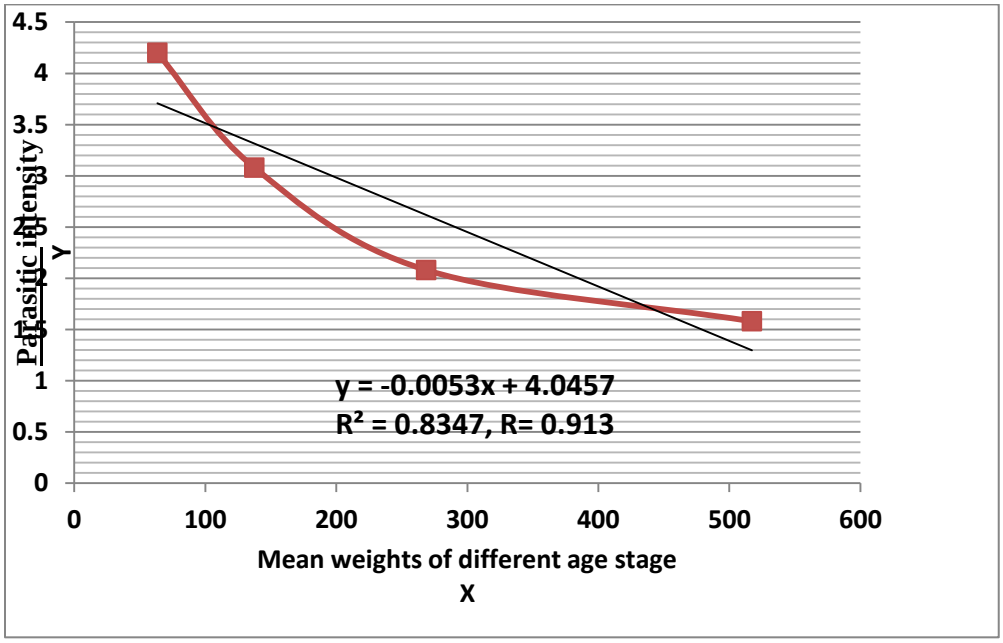

Figure 3: The linear correlation between parasitic intensity and mean weight of infected *D. labrax* by the digenetic trematodes.

6. Histopathological examination:

The gill tissues in the infected *D. labrax* with digenean showed some inflammatory reactions, alternative epithelial hyperplasia, desquamation of secondary lamellae, and hemorrhagic foci **(Figs. 4 a&b).** Also, the Kidney exhibited focal depletion of hematopoietic tissue and tubular nephrosis **(Fig. 4 c)**. The liver tissues showed vacuolar degeneration of hepatocytes **(Fig. 4 d).** Various degenerative changes were detected in the intestinal tissues. The intestine showed hyperplasia of the epithelium lining, mucinous degeneration, sloughed necrotic areas that infiltrated with mononuclear cells, together with focal desquamation and necrosis of the intestinal epithelium. In addition to congestion of the blood vessels, edema, and aggregation of inflammatory cells in the lamina propria and submucosa of the intestine **(Figs . 4 e&f).**

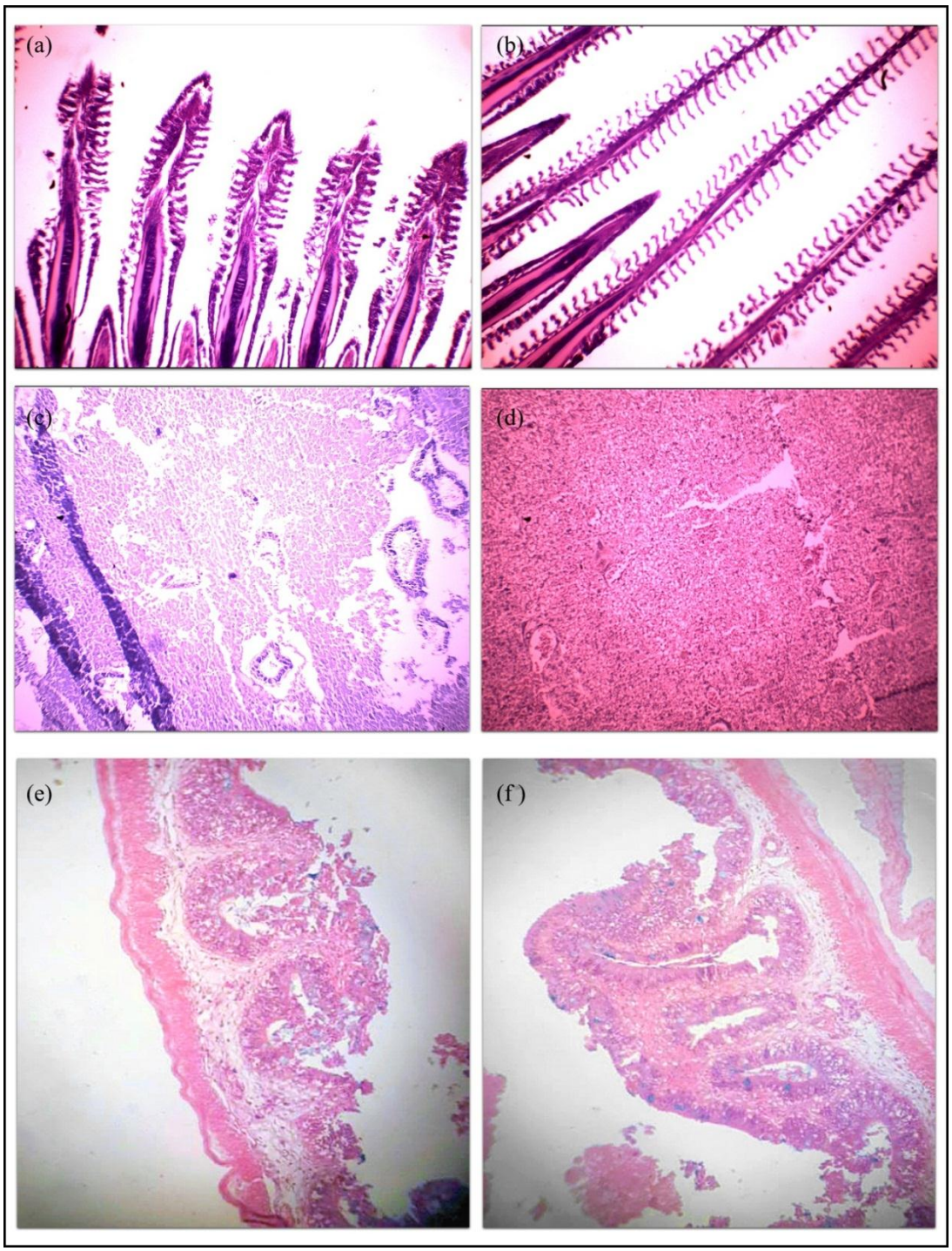

Figure 4: (a&b); Gill tissues in the infected *D. labrax* with digenean showing inflammatory reactions, hyperplasia & desquamation, of secondary gill lamellae **(c);** focal depletion of hematopoietic tissue and tubular nephrosis of the kidney **(d);** Liver tissues showing vacuolar degeneration of hepatocytes **(e&f);** the intestine showed focal epithelial hyperplasia and sloughed necrotic areas that infiltrated with mononuclear cells. (H&E stain ×20).

DISCUSSION

Marine fishes, including sea bass *D. labrax*, are exposed to many parasitic diseases, the most common are crustaceans **(Öktener and Sezgin, 2000, Manera and Dezfuli, 2003)**, monogenetic trematodes **(Whittington and Chisholm, 2008)**, besides, other parasitic species of protozoa, trematodes, cestodes, acanthocephalans, and nematodes **(Piasecki, 2004, Noga, 2010, Bessat and Fadel, 2018)**.

Digenetic trematodes are metazoa parasites, several outbreaks of them were reported worldwide, however, the knowledge of their pathogenic effects are still little and questionable. Based upon, we tried to fulfill some of the epidemiological features related to the infectious agent, seasonal occurrence, clinical symptoms, and physio-chemical related factors, as well as the body condition as a reflection of the health status. Digenetic trematodes are uncommon in cultured fish, with the availability of under optimum environmental or biological conditions, the life cycle of the parasite was initiated, and the parasitic infection of fish become potential **(Noga, 2010)**.

Some of the developmental stages of the parasite have been identified, mostly are the adult stages, sometimes accompanied by other nearby stages. The recorded parasites were isolated from the intestinal portion, but, some of them often isolated from the stomach. First, adult digeneans inhabit the gastrointestinal tract of the final host such as fish, bird, or reptiles, produce eggs that pass out, hatches into a miracidium that usually infects the intermediate host (the mollusk intermediate host a snail). Then, the miracidium inside the mollusk host develops to a cercaria, which released and penetrates the fish, penetrated the target tissue, it develops to a metacercaria, which subsequently produces the cyst. When the metacercaria-infected fish is eaten by another final host, the metacercaria develops into the adult, thus the cycle is completed **(Noga, 2010).**

The parasitological identification revealed four common digeneans, isolated throughout study periods in the three farms. Firstly, *Metadena crassulata* **(Yamaguti 1971 after Linton, 1910r),** it was isolated from the anterior intestine, its morphological description corresponded to **Zander and Reimer, (2002)**. The second digenean was *Derogenes varicus* **(Müller, 1784),** it was isolated also from the different intestinal portions which were previously characterized in seabass **(Sterud 2002)** and *Mullus surmuletus* **(Amel *et al.*, 2009; Al-Zubaidy and Mhaisen, 2011; Abou Zaid *et al.*, 2018)**. The third digenean was *Acanthostomum spiniceps* **(Looss 1899 after Brooks 1980),** it isolated from the different gastrointestinal portions. Originally *A. spiniceps* was described by **Moravec (1976)** from *Bagrus docmac* that identified worldwide and also in Egypt among various fish. *Acanthostomum spiniceps* displayed a great similarity to those identified in South America from *Astroscopus sexspinosus* by **Fernandes *et al.* (2002)**. In Egypt, this digenean was isolated from *Bagrus bayad* **(Imam *et al.*, 1991; Soliman and El-**

Damarany, 1995), *Bagrus filamentosus* **(Tadros *et al.*, 1978)**, *Morone labrax* **(El-Shahawi and Al-Bassel 1992)**, African snook, *Lates niloticus* **(Morsy *et al*., 2013),** and Common seabream, *Pagrus pagrus* **(Morsy *et al*., 2013; Eissa *et al*., 2017b).** The fourth isolated digenean was *Diplomonorchis sphaerovarium.* It was originally isolated from *Spheroides testudineus* from Jamaica. Its morphology was largely similar to that isolated from *Ophichthus gomesi* **(Fernandes *et al*., 2002)**. Other species of *Diplomonorchis Hopkins,* was characterized among various fish species, *D. floridensis* was isolated from *Symphorus sp* **(Wallet & Kohn, 1987),** *D. catarinensis* from *Micropogonias furnieri* **(Amato, 1982),** and *D. leiostomi* Hopkins, 1941 from *Boridia grossidens* **(Fernandes *et al*., 1985)** and *Haemulon sciurus* **(Kohn *et al*., 1982).** Naturally, the location of the parasite in the body varies according to the parasite species.

Concerning seasonal occurrence, we recorded differences in the occurrence of the digenetic trematodes among the three farms under the study. Spring recorded the highest digenean prevalence of 27.76 %, followed by autumn 15.78 %, summer 14.89 %, and the lowest 14.44 in winter. The spatial occurrence reported the highest prevalence of 30.16 % in Farm 2, Edeeba-Triangular, 18.64 % in Farm 1, Borg El-Arab, and the lowest rate of 5.83 % in Farm 3, Wadi-el-Rayan. Close lower infection rates, but in winter, were recorded among various fish, 26% - 28.4% prevalence rate from *Sardinella species.* **(Youssef and Derwa, 2005a)** and 24% in *Morone labrax* **(El-Lamie, 2007),** 29.41% in Lane snapper (*Lutjanus synagris*) from the Southern Gulf of Mexico **(Mendoza *et al*., 2016).** Lower prevalence rates (4.6%) were recorded in Siganus fish **(Abdel-Mawla and Abo-Esa, 2011),** and (3%) in wild European seabass, **(Abou Zaid *et al*., 2018).**

Hence, these differences about both seasonal and spatial occurrence could be related to certain factors. Potential environmental factors include water temperature, salinity, light intensity, food resources, and predators could influence the abundance and distribution of the parasites **(Martins *et al*., 2015)**. Moreover, digenetic trematodes have indirect life cycles and their outbreaks need at least two hosts, primary hosts such as snails and crustaceans, and secondary consumers including fish and biscivorous **(Zander *et al*., 2000)**. Throughout different seasons, water temperature was ranged from 14.22 - 29.7 ^{o}C, 7.49 - 8.36, and salinity 13.54 ppt-34.36 ppt. Particularly in spring, the water temperature was between 21.12- 24.63 ^{o}C, and salinity 15.85-32.34 ppt. Accordingly, the highest prevalence specifically in Farm 2, indicate the availability of the recorded ranges of water parameter and salinity. Besides, other environmental, and biological factors that favor the growth of primary host and the definitive, all assist the survival and spread of trematodes in the three farms under study **(Syobodova and Kolarova, 2004)**. For example, the snail is a major intermediate host, its growth needs water temperature over 17 ° C **(Noga, 2010)**.

In most cases, the parasites have been isolated from infected *D. labrax* with no characterized symptoms. But in severe cases, heavy infestation stages, some of the obvious symptoms have been recorded, including weakness, pale external and internal organs, and anemia. These lesions may be attributed to the infestation of the digenean to the mucosa layer of

the gastrointestinal tract, causing irritation, inflammation, and local tissue damage. Also, adult worms secrete proteolytic enzymes which could destruct intestinal tissues **(Woo, 1995)**. Moreover, in heavy digenean infestation, the parasites cause mechanical blockage, produce toxic by-products which could occlude stomach, or blood vessels, resulting in degenerative changes **(Abdel-Mawla and Abo-Esa, 2011)**.

It was important to know whether the digenean affected the body condition of the infected *D. labrax*. By correlating the condition factors values, 1.27, 135, and 1.39 of infected juveniles, adults, and breeders; respectively, with the corresponding mean infection intensity 3.05, 2.08, and 1.55 parasites/microscopical field, they were generally weak. However, general weak linear and correlation a regression correlation between the infection intensity to fish weight, length, and condition factor (R=0.91, 0.964, and 0.956); respectively were seen across different fish stages. Thus, the parasitic intensity has been found to have a relatively weak influence on the body condition indices. Except for fingerling, they recoded a mean infection intensity of 3.9 with the lower condition of mean value 0.98. **Lagrue and Poulin, (2015)** found no relationship between total parasite load and fish body condition using the residual index. **Jones *et al.* (1999), and Santoro *et al.* (2013)** reported that, the heavily infected fishes have a lower body condition than those infected individuals. Even at low infection levels, a positive correlation between parasite intensity and fish body condition suggested those parasites can benefit from hosts with high energy reserves **(Maceda-Veiga *et al.*, 2016)**. Other conservative studies stated that, helminth infections have pernicious effects on fish hosts especially at the highest levels of parasite load **(Kelly *et al.*, 2010, Santoro *et al.*, 2013)**. Some factors could be attributed to the poor condition factors at the fingerlings stage, all collectively altering the animal weight such as poorer feeding quality than healthy conspecifics **(Peig and Green, 2009)**.

Further pathological alterations in digenean-infected *D. labrax* have occurred especially the intestines. During the parasite migration, some destructive changes have been induced. Wherein, the intestine showed hyperplasia of epithelium lining, mucinous degeneration, sloughed necrotic areas, desquamation, and necrosis of the intestinal epithelium. These pathological lesions could be attributed to the mechanical infestation of digenean to the mucosal layers of the intestine, causing inflammation, irritation, and local tissue damage. Besides, adult worms secrete proteolytic enzymes that could destruct intestinal tissues **(Woo, 1995)**. Along the migration path, more violent changes may appear during the movement of the parasite to different tissues, including tissue rupture, hemorrhage, and necrosis **(Overstreet *et al.*, 2002).**

CONCLUSION

The current study revealed isolation of four digeneans from the gastrointestinal tract of sea bass *D. labrax* cultured in Egypt namely; *Metadena crassulata Derogenes varicus, Acanthostomum spiniceps*, and *Diplomonorchis sphaerovarium.* The highest values of infection rates were detected in spring and the lowest in winter. Potential environmental factors include

water temperature, salinity, and pH could influence the abundance and distribution of the parasites. Heavy infection stages only reported obvious symptoms including weakness, paleness, and anemia. Generally weak linear correlation and a regression correlation between the infection intensity and fish weight, length, and condition factor were seen. Histopathological alterations in digenean-infected *D. labrax* were reported especially in the intestines. Therefore, preventive and control measures are recommended to avoid such infections that potentially influence the health status and productivity of infected fishes.

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