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Study of Using Monogenea Parasites on Free – Living Fishes in the Lake of 16 Tishreen Dam as Bio Indicators of Environment Pollution

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Abstract: The aim of the study was to develop a data base about the water quality in the Lake of 16 Tishreen Dam through, study was performed to determine of non biotic factors values of water in Lake and infecting Free- Living fishes with monogenea parasites, and finding a relationship between parasites distribution and environmental conditions changing. 144 Free – living fishes were examined for detecting the infection with parasitic monogenea, and determine their distribution rate. Fishes were collected randomly once a month, during the period from 12/2012 until 11/2013. Collected fishes samples were belonged to the following species: Cyprinus carpio L., Varicorhinus damascinus, Garra rufa, Tilapia zillii, and Liza abu. T. zillii was the most prevalent one. Fishes were infected with monogenea- parasites, 6 species were isolated and classified. they belonged to four Genera, they were: D. lenkorani, D.extensus belong to the Genus (Dactylogyrus SP.), two species of (Gyrodactylus sp.) are:G. Medius and G. Mugilis, and one species of Cichlidogyrus sp. C. sclerosus, and one species of Microcotyle Sp. Isolated parasitic species were recorded for the first time on the free - living freshwater fishes in the lake of 16 Tishreen dam, while by four species of them were recorded for the first time in Syria :(D. lenkorani, G. mugilis, C. sclerosus, Microcotyle.sp) in this study. Monogenic Parasites showed high specificity to the host, and infected organ. General rate of infection with monogenic - parasites was 27.1%. Varicorhinus damascinus was more infected than Tilapia Zillii in rate 35.71%, 28.72%. respectively the highest infection rate with monogenic on free living fishes was recorded in summer (High temperature and low concentration of dissolved oxygen, and slightly higher value of BOD). Study showed that the lake of 16 Tishreen Dam was relatively clean. Cichlidogyrus sclerosus was the most important monogenic parasites used as bio indicator of environmental pollution in the lake.

Keywords: Bio Indicators, Environmental Pollution, Free -Living Fishes, Lake of 16 Tishreen Dam, Parasitic Monogenea

1. Introduction

The people has become increasingly aware in recent years that aquatic ecosystems around the world are deteriorating from deposition of anthropogenic pollutants, which inevitably lead to sharp changes in water quality, which will reflect negatively on the actual use of water, in addition to the harm and damage attached to bio components of aquatic ecosystems (Sures,2004, Sanchez-Ramirez et al., 2007).Early warning systems are being developed in response, and fish- parasites have been proposed as effective bioindicators of environmental pollution (Sures, 2004; Marcogliese, 2005). The logic underlying of the use fish parasites is based on the fact that both parasites and their hosts are exposed and, therefore, may respond to pollution in aquatic environment (Williams and Mackenzie, 2003; Khan and Payne, 2004). Monogenean parasites are recognized as useful bioindicators of environmental quality because of their predictable numerical responses to chemical pollution (Khan and Thulin, 1991; Pietrock and Marcogliese, 2003; Thomas et al., 2005). They tend to increase in number when exposed to low and medium pollutant concentrations, but disappear at high concentrations (Moles and Wade, 2001; Khan and Payne, 2004). This was confirmed by the researchers (Sanchez -Ramirez et al., 2007) in Mexico, where they noticed clear differences in the number of worms Cichlidogyrus. sclerosus on the gills of Tilapia fish of highly polluted lakes compared to those less polluted lakes, where it was found that the sediment in the studied sites were contain polycyclic aromatic hydrocarbons (PAHs), and heavy

metals, but in Syria, there are no comprehensive studies on the parasites of freshwater fish.

The limited information available about the parasitic communities of freshwater fish in Syria have been submitted by many studies (AL- Samman, 1989; Zidan, 2000;Dayoub and Salman,2002; Dayuob et al., 2003;Salman,2004; Dayoub et al., 2007). These studies were limited on the parasites of Common Carp fish Cyprinus carpio L. cultured in freshwater fish farms in Syria.

The changes of number and species of parasites on freshwater fish, can play a vital indicator role of environmental changes, and this leads to deeper changes in the components of the ecosystem. From here came the idea of research on:

The possibility of using parasitic monogenea on free living fishes in the Lake of 16 Tishreen Dam as a vital bio indicators of environmental pollution.

The aims of the study were:

1. determine the values of some abiotic environmental parameters of waters in the Lake of 16 Tishreen Dam.
2. determine the types of parasitic monogenea on free living fish in the lake.
3. discovery of the relationship between the species of registered parasites and abiotic environmental indicators and the species of free- living fishes.

2. Materials and Methods

2.1. Study Field

The study was performed on the Lake of 16 Tishreen Dam, it is one of the artificial lakes in the Syrian Coast. The lake is located in the north-west of Syria, Latakia city, Within the extend of AL-Kabeer Al-Shamali River., About16 km from Latakia city. storage capacity is (210-200) million M3 of water approximately, Lake water used to irrigate agricultural land and fish breeding (figure 1)

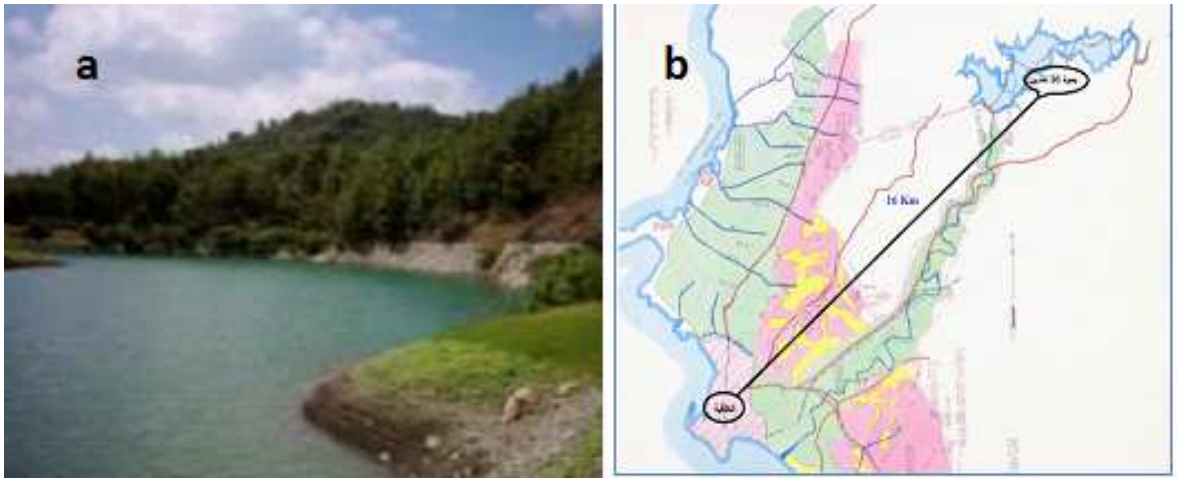

Figure 1. *a: A general view of part of the lake of 16 Tishreen Dam and, b: distance from Latakia city.*

Fish samples were Collected from the Lake of 16 Tishreen Dam,monthly, During the period from 12/2012 till 11/2013, using cages and fishing nets, slots diameter (18 mm) (Figure 2).

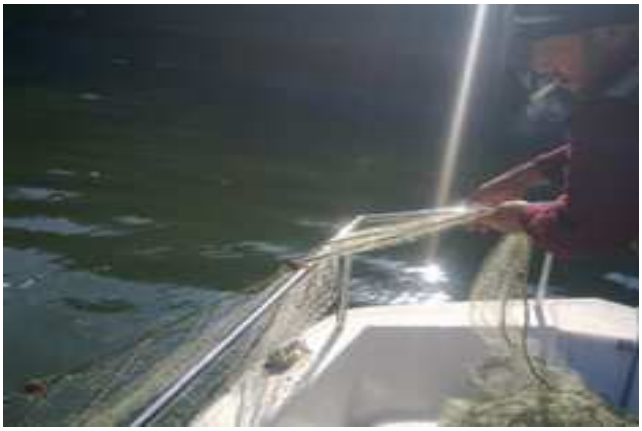

Figure 2. *Collecting fishes samples using fishing nets.*

Fishes were Transferred alive directly to the laboratories of Higher Institute for Environmental Research at the Tishreen University, Placed within the large glass basins filled with fresh water and aeration locked ventilation, water Has been replaced by periodically in order to keep the fish alive during the examination period.

2.2. Examination of Fish to Detect on Infection with Monogenea Parasites

Fish samples Were examined directly after killing it by beat on the head, and then recorded the measurements of length (cm) and weight (gr) (Figure 3), and determination of sex. Infection with monogenea was detected by magnifying first, where examined the outer surface of the body (the skin and fins and gill Cover).

Microscopic examination for all parts of the external body (skin, fins, gills) were done using Olympus microscope and different magnifications 10x, 20x, through Wet smears.

Monogenea Worms were isolated from wet smears using fine needles, then placed in a drop of water on a glass slide, Then fixed directly by formalin 4%, colored by hymatoxelen (Noga, 1996).

Permanent preparations were made using Glycerin, Then

the preparations were studied microscopically to determine morphometric character of taxonomically important parts of the body (Ramirez – Sanchez,2007; Abdul – Ameer, 2010;Bichi and Ebrahim, 2012).

Taxonomic identification of monogenea is based upon the morphology of the posterior attachment organ (The shape and dimensions of the Chitinized Parts of opisthohaptor), and The shape and dimensions of Copulatory organ. The presence or absence of eyespots (Bychovskaya and Plavovskaya, 1962; Gussev,1985;pariselle and Euzet, 1996).

Figure 3. *Killing way of fish and take measurements for the total length and weight of the fish.*

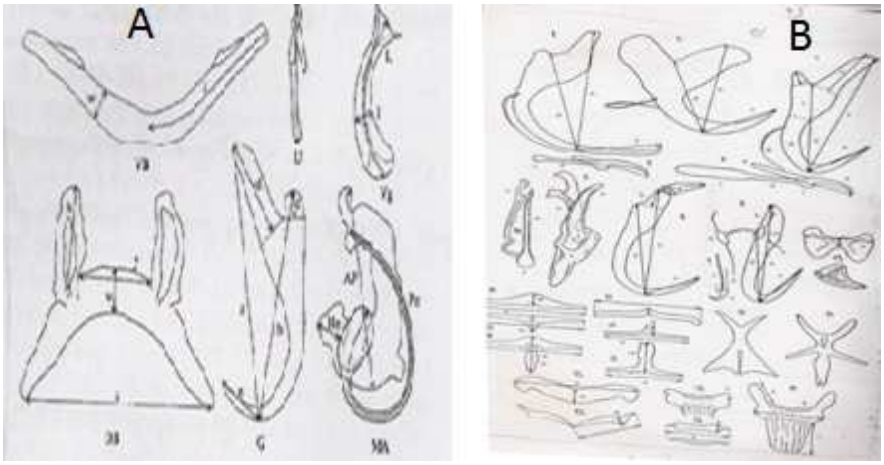

Figure 4. *The way for taking the micrometric measurements of the hard parts of opisthohaptor and Copulatory organ: A: of The Cichlidogyrus sp. B: of the both genera Dactylogyrus and Gyrodactylus*

2.3. Analysis of Water Quality Parameters

Concentration of oxygen dissolved in water and PH value were measured in field using an instrument model orion (0.835 m), and water temperature was measured by using a mercury thermometer runway 0-100 c °

The biological oxygen demand BOD 205 has been done on the special instrument BOD, But The concentration of ammonia and nitrite ions in water have been measured using a spectrometer instrument (UV/ ViS Spectrometer + T80 (Ltd Pg-instruments).

3. Results and Discussion

3.1. Classifection of Isolated Parasitic Monogenea

144 fish samples were examined from the Lake of 16 Tishreen Dam, contain five species,belong to three families :*Cyprinus carpio L.* (8 Fish), *Varicorhinus damascinus* (14 Fish), *Garra rufus* (8 Fish) from Cyprinidae, and *Tilapia zillii* (101 Fish) from Cichlidae,and *Liza abu* (13 Fish) from Mugillidae (Figure 5). Fish samples were classified using the manual of Bechman (1962).

Figure 5. *Free living fishes species were studied from the lake of 16 Tishreen Dam.*

Tilapia zillii was the most prevalence 70.13%, following Varicorhinus damascinus 9.7%.

The results of the study shown presence of six different species of parasitic monogenea: D.extensus, D.lenkorani belong to the genus Dactylogyrus sp, and the both species G.

medius,G. mugilis belong to the genus Gyrodactylus sp., and one species of the genus Cichlidogyrus sp. is C.sclerosus, and one species of the genus Microcotyle sp.

Isolated Monogenea were Showed high specificity to the host, This result was agree with many studies (Pariselle and Euzet, 2003; Soylu et al., 2010).

The both species of the genus (D.) Dactylogyrus (D.lenkorani, D. extensus) were recorded for the first time on free living fish in the Lake of 16 Tishreen Dam, while the species D.lenkorani was recorded for the first time in Syria in this study.

D.lenkorani infected the gill of Varicorhinus damascinus, while the second species was infected the gill of Common Carp fish, they are oviparous worms. and have four eyespots in front of body, Attachment disk is equipped with 7 pairs of small marginal hooks, and one pair of large median hooks. This characteristics was confirmed by many researchers (Gussev, 1985, Noga, 1996, Soylu et al., 2010). Worms of D.lenkorani showed high specificity toward the host, where did not record on Varicorhinus damascinus only, it has been characterized as a medium-sized worms. The average length of the worm (120µ.m), and (95µ.m) width, length of marginal hooks about (40.5µ.m), dimensions of the median hooks (a: 48.6, b: 43.2, c: 6.75, d : 18.9), (dorsal) 5.4x 32.4 µ.m, the dimensions of connecting bars (ventral)(3.2x 24.3µ.m), the total length of copulatory organ 35.1 µ.m. (Figure 6). The measurements of this species were resembled with the measurements recorded by the researcher (Abdul - Ameer, 2010).

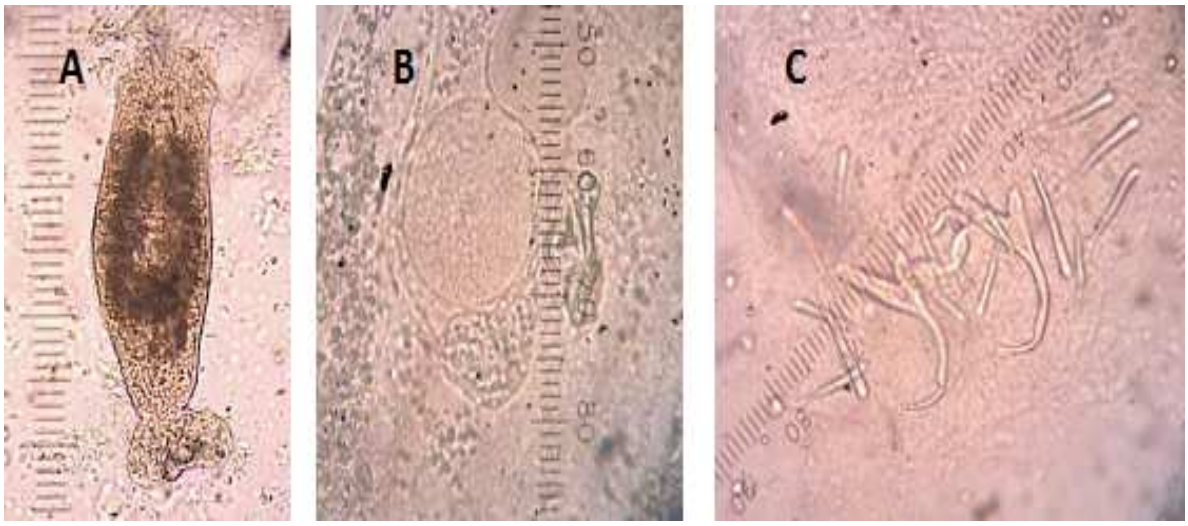

Figure 6. *A: The general shape of D.lenkorani 10×, B: The shape of copulatory organ40×, C: the hard parts of the attachment disk 40×.*

Extensus are relatively large worms, length 744.4µ.m, width 235.2µ. m, it is common on gills of Common carp fish in general, this species was recorded previously by the researchers (Zidane, 2000; Dayoub *et al*., 2003) on the gills of common carp fish in fish farms in Syria.

This species of worms characterized by the tube of copulatory organ which takes the shaped (L) and a supporting part like a straight cylinder (Figure 7).

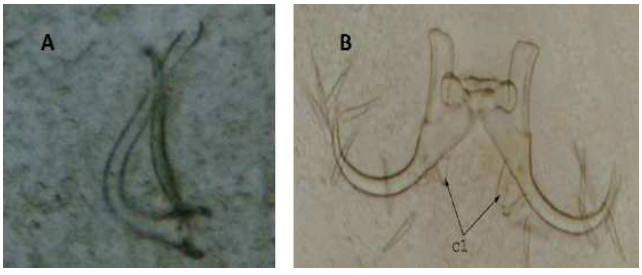

Figure 7. *A: A shape of copulatory organ, B: The hard parts of the attachment disk of D.extensus 40×.*

It also has been isolated and classified two species belong to the genus *Gyrodactylus* sp. On the skin and fins of free living fish in the Lake of 16 Tishreen Dam are: *G.medius*, *G.mugilis*.

The first species *G.medius* Isolated from fins of *Varicorhinus damascinus*, it is small in size, the diameter of attachment disk (81-84 µ.m), Total length of the large median hooks (81-84 µ.m), the ratio of length of the internal process to the body of hook 1:2.3-2.5 (Figure 8).

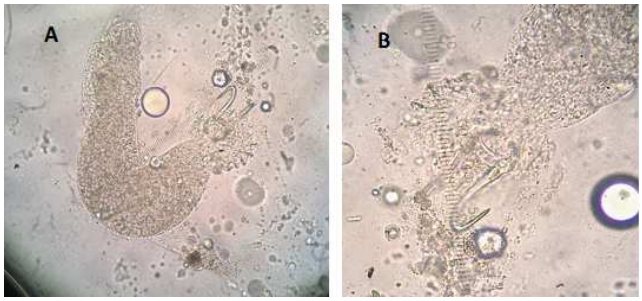

Figure 8. *A: General shape of G.medius 10×, B:Tthe attachment disk 20×.*

While *G.mugilis* was isolated from skin and fins of mullet (*Liza abu*), And characterized (small worms, length372.4 µ. m, width 78.4µ. M, Dimensions of the attachment disk 58.8 × 78m. µ) (figure 9).

Figure 9. *Attachment disk of G.mugilus 40× with large median hook and connecting bars.*

The study showed that *G.medius* worms can infect a wide range of host closely related, Such as different species belong to Cyprinidae, and this was confirmed by many researchers (Bykhovskaya – Pavlovskaya et al., 1962, Gussev, 1985), this species was recorded previously by the researcher (Dayoub et al., 2003) on the skin and fins of Commen carp in fish farm The second *G.mugilis* showed that high specificity to the host, it is recorded for the first time in Syria in this study

Cichlidogyrus SP. (C.) are Different worms from the last both genera Dactylogyrus sp., and Gyrodactylus sp., They have two pairs of median hooks, One of them is posterior with connecting bar like(x), and the other is ventral with connecting bare resemble (V), affects only gills of Tilapia fish, This agree with many researchers (Pariselle and Euzet,2003; Pouyaud et al., 2006), One species of the genus *Cichlidogyrus* was recorded in this study: *Cichlidogyrus* sclerosus, It was recorded for the first time in Syria, on the gills of *Tilapia zillii*. It is a large or medium- sized worms, length 558.7 µ.m, width 109.03 µ.m, haptor rounded, with 2 pairs of hamuli and 7 pairs of hooklets, the dimensions of attachment disk are(72.1 x 58.6 µ.m)figure (10,A), the dimensions of Dorsal gripus are(a:37.8, b: 36,c: 4.05,d:8.1, e:10.8, w:6.75), we could find eggs inside the body of adult worm (figure (10,B), copulatory organ with large serrated plate; copulatory tube thin and arched, with tapered end figure(10,c)

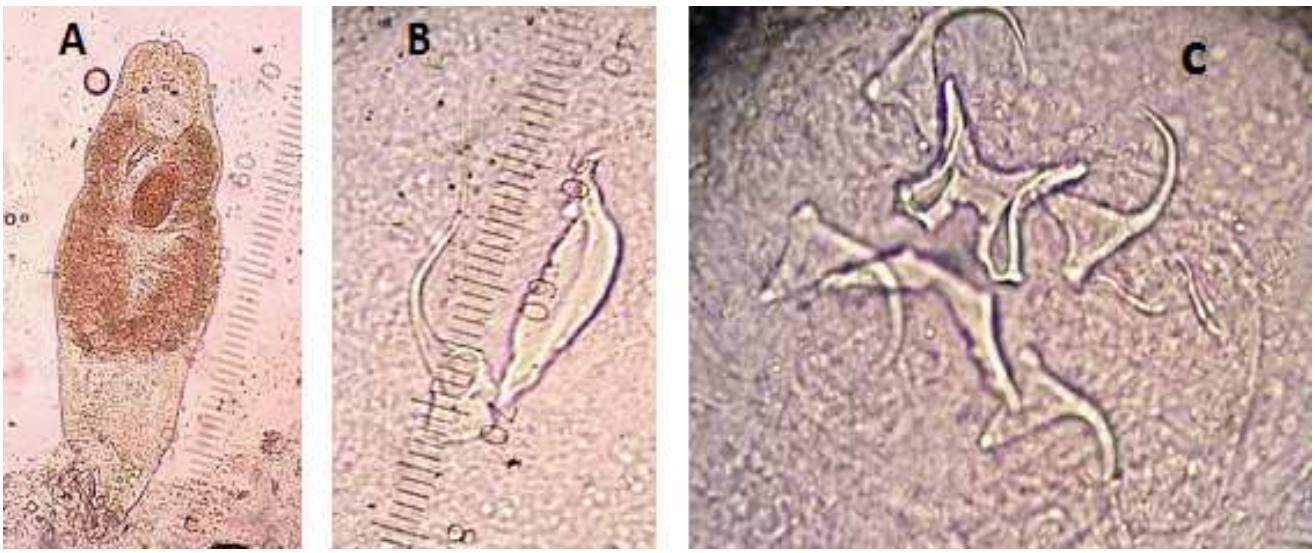

Figure 10. *Cichlidogyrus sclerosus, Paperna & Thurston, 1969, A: Whole worm with egg 10x,B: Copulatory organ40x, C: Haptoral armature40x.*

The finding of *Cichlidogyrus sclerosus* in this study represents the first record in Syria.

Microcotyle (M.) worms were easily distinguished through their haptor having a lot of tiny clamps on the lateral margins. located at the anterior part of the worm, it is the funnel-shaped mouth,this worms haven't eyespots, This is inagreement with the observations of many researchers (Gussev, 1985; Noga, 1996).

One species of the genus *Microcotyle* sp. was classified for the first time in Syria, and Isolated from the gills of Liza abu only. They are larg worms, length 1794.8µ.m, width 382.2µ.m, Could be easily seen with the naked eye, Seemed under the microscope oversize, red color inside the body, They feed on the blood of fish figure (11,A,B). So they are very dangerous worms, This was confirmed by most researchers(Naga, 1996,Gussev,1985), haptor having a lot of tiny clamps, located in two rows lateral Figure(11,D), In addition to the presence of two muscular suckers in the front end of the body figure(11,C). They are oviparous worms, where we were able to see the egg inside the body under microscop easily, Characterized by oval form they with posterior extending as long filamentous final figure(11,C,E), We cannot determine this species accurately because this species affects the mullet in freshwater(*Liza abu*) and this

species marine fish is origin.

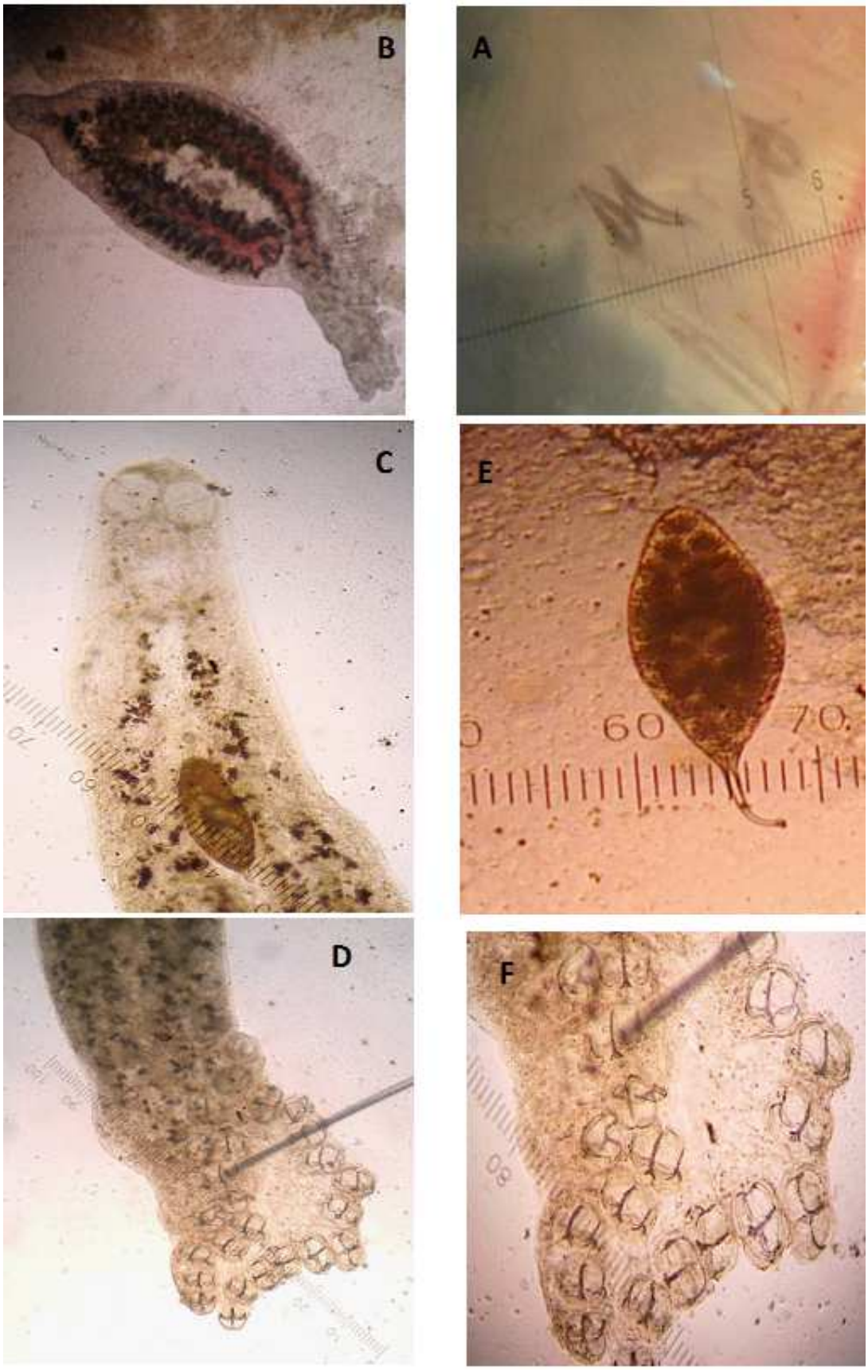

Figure 11. *A,B: general shape of Microcotyle SP. 10 ×, D: Clamps at the end of the body 20 ×, c:Suckers In front of body and the egg inside the body 20 ×, E: the general shape of an egg 40x.*

3.2. The Relationship Between Occurrence of Monogenea and Studied Abiotic Environmental Indicators

The results showed that the infection with monogenea parasites was low or medium relatively by rate 27.1% and was most prevalent on *Varicorhinus damascinus*, followed by *Tilapia zillii* respectively 35.71%, 28.72%, and this may be due a little spread of *Varicorhinus damascinus* in the lake of 16 Tishreen Dam. in addition to high specificity of these parasites to the host (Table 1).

Table 1. *The prevalence of monogenea parasites on free living fishes in the Lake of 16 Tishreen Dam.*

Fish species	Number of fish studied	Number of infected fish with monogenea parasites	Infection rate %
Tilapia zillii	101	29	28.72
Varicorhinus damascinus	14	5	35.71
Liza abu	13	3	23.08
Cyprinus carpio	8	2	25
Garra rufus	8	-	-
∑	144	39	27.1

For the seasonality changes in the prevalence of parasitic monogenea. We found the maximally prevalence of infection in summer, and minimally in winter (Figure 12).

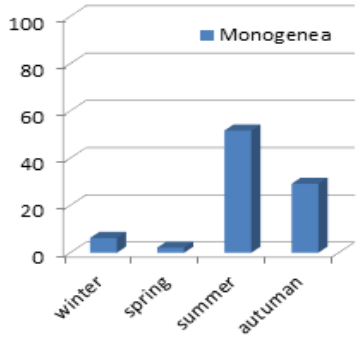

Figure 12. *A Diagram of seasonal changes of infection with parasitic monogenea on free living fish in the Lake of 16 Tishreen Dam.*

This was associated with a significant increasing of concentration of BOD (organic pollution is relatively simple), and decrease of oxygen concentration in the water, These results were similar to the findings of many researchers (Noga, 1996; Sanchez –Ramirez,2007; El- seify et al., 2011) indicated that the intensity of monogenea parasites refers to the bad quality of the water such as organic pollution and low of dissolved oxygen in water.

Table 2. *Seasonal changes of the infection with monogenea parasites on free living fishes in the Lake of 16 Tishreen Dam.*

General infection%	Infection rate of % Monogenea				NO2- m.g/l	NH4 m.g/l	BOD m.g/l	DO m.g/l	PH	TC	NO. of collected fish	Season
	M.SP.	C.SP	G.SP	D.SP								
6.25	6.25	-	-	-	0.032	-	1	10.42	7.8	14	16	Winter
2.17	-	-	2.17	-	0.0055	0.091	2	9.91	7.4	19	46	Spring
51.72	-	43.16	-	6.89	0.009	0.052	4	7.8	8.6	30	58	summer
29.16	8.33	12.5	4.16	4.16	0.025	-	3	9.32	7.8	23	24	autumn
27.1	2.1	20.13	1.38	3.47	-	-	-	-	-	-	144	∑

As for the species of parasitic monogenea isolated from free living fish in the the lake of 16 Tishreen Dam, and changes in infection during the seasons, and the possibility of using these species as bio indicators of environmental pollutions in the Lake, found that the species *Cichlidogyrus sclerosus* was the most prevalent of monogenea on free living fish in the lake of 16 Tishreen Dam in infection rate 20.13%. The highest prevalence was recorded in the summer Table (2), however, the infection of this parasite have been found on Tilapia fish only, These results agreed with the findings of researchers (Sanchez- Ramires et al., 2007)who have demonstrated that the parasite *Cichlidogyrus Sclerosus* and its host of Tilapia fish useful as bio indicators for the aquatic environment pollution.

As for the quality of water in the Lake of 16 Tishreen Dam,, has remained the values of abiotic environmental indicators within the Syrian Standards for irrigation water and this result reflects a clean environment, and reflected relatively the health status of the fish in the lake

From this study, it might be concluded that the parasite *Cichlidogyrus sclerosus* is the most important species of monogenea that can be used as bio indicators of the environmental pollution in the Lake of 16 Tishreen Dam, due to larg spread throughout the year, and infected Tilapia fish which is one of the most distributed fishes species in the Lake of 16 Tishreen Dam.

Acknowledgements

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