

**Diagnosis of some prevailing blood parasites in fresh water fish
Chrysichthys auratus at Al Fath center in Assiut Governorate, Egypt
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Abstract

This study was conducted to identify the blood parasites parasitizing *Chrysichthys auratus* in Assiut Governorate, and studying their effect on the differential leukocytic count. A total of 200 specimens of *Chrysichthys auratus* were captured at Al Fath center in Assiut Governorate and were examined for haemoparasites. Thin blood smears were examined and observed to be infected with haemoparasites with prevalence of 61.5%. The infection by *Trypanosoma*, *Babesiosoma* and *Myxobolus sp.* were inspected with prevalence of 55.5%, 12% and 13% respectively. *Chrysichthys auratus* males were harboring more blood parasites than females. The morphological and morphometric characterizations of these parasites were determined as *Trypanosoma cyanophilum*, *Babesiosoma mariae* and *Myxobolus heterosporus* type I. Their measurements were recorded. Leukogram picture showed the different types of leucocytes. No basophils were detected. There was a significant variation in lymphocytes, monocytes and eosinophils count between infected and non-infected fish while neutrophils count showed non-significant difference.

Keywords: *Trypanosoma sp.*, *Chrysichthys auratus*, Blood parasites, *Babesiosoma sp.*, *Myxobolus sp.*, Leukogram picture.

Introduction

One of the most important problems facing our world nowadays is food deficiency. In Egypt, the continuous increase in human population leading to increase the demand for animal protein. Fishes were considered comparatively low price source of protein to compensate the continuous lack of such element (El-Tantawy and El-Sherbiny, 2010).

Chrysichthys auratus is an African species, which belongs to Siluriformes (Family: Bagridae). It is abundant in the River Nile of Egypt and the interconnecting lakes (Atobatele, 2013). In addition to their economic importance and good sensorial properties of meat, it is considered one of the most popular fish in Upper Egypt, because of its cheap price and palatable for consumer (Bishai and Khalil, 1997). They are omnivorous in nature feeding on varieties of food stuffs which include seeds, insects, bivalves and detritus (Reed et

al., 1967).

In their natural habitats, fish may suffer from various diseases including parasitic infections (Bamidele, 2015). Parasitic diseases of fish are common all over the world and are of particular importance in the tropics (Roberts and Janovy, 2009). Parasites affect fish health, growth and survival causing economic losses (Marzouk et al., 2013). Blood parasites have a damage effect on haemopoiesis (Ismail 2003, and Supamattaya et al. 2005).

Trypanosomes are haemoflagellates, protozoa that transmitted to fish by leech vectors (Woo, 2006 and Shahi et al., 2013). They are living principally extracellularly in the blood and tissue fluids (Overath, et al., 1999). Most species of Trypanosomes infecting fishes cause pathogenic diseases of considerable medical and economic importance (Noga, 1996). A sign of piscine trypanosomiasis depends on the intensity of infection which range from mild

anemia associated with low levels of parasitaemia to severe pathological changes due to a heavy parasite burden (Zintl *et al.*, 1997). The first record of a trypanosome was likely made by **Valentin, 1841**. Since this observation, more than 200 Trypanosome species had been identified in freshwater and marine fish worldwide. Identification usually based on the taxonomic criteria of morphology, geographical and host origin (Ferreira *et al.*, 2013 and Borges *et al.*, 2016). Many Egyptian authors were recorded *Trypanosoma mansouri* and *Trypanosoma cynophilum* in *Chrysichthys spp.* (Mohammed, 1978; Marwan, 1980 and Ahmed, 2001). While *Trypanosoma alhussaini* described in *Clarias lazera* by Ahmed (2001). The dactylosomatid apicomplexans include two genera; *Dactylosoma* Labbe, 1894 and *Babesiosoma* Jakowska and Nigrelli, 1956 (Aragort, *et al.*, 2005). *Babesiosoma* are parasites inhabit both circulating erythrocytes and erythrocytes from reticulo - endothelial tissues. It complete its life cycle in two hosts i.e., a vertebrate host (include both marine and freshwater fish as well as frogs and toads) and an invertebrate host (Leeche) (Woo, 2006). In the vertebrate (intermediate) hosts, the babesiosomes undergo primary and secondary cross-like (cruciform) merogony yielding 4 merozoites on each occasion, within erythrocytes. Gamonts form within the same host cells from secondary or tertiary merozoites these gamonts associate, and gametes fuse following gametogenesis, forming an ookinete (motile zygote) (Aragort *et al.*, 2005 and Shahi *et al.*, 2013). Only 3 valid species of fish babesiosomes may remain currently: *Babesiosoma bettencourti* (Franca 1908) from the European eel, *Babesiosoma mariae* (Hoare, 1930) from many species of freshwater fishes in Africa; and *Babesiosoma tetraogonis* Becker and Katz, 1965 from suckers, *Catostomus spp.*, from California, U.S.A (Aragort, *et al.*, 2005; Woo, 2006). Myxosporidian are obligate parasites of tissue (histozoic forms that reside in intercellular spaces or blood vessels that reside intracellularly) and organ cavities. Each parasite is somewhat species specific as well as organ specific (Markiw, 1989). They are multicellular organisms and highly specialized metazoan parasites of aquatic hosts with a very wide host

range belonging to phylum cnidarians (Zrzavy, 2001). Parasites characterized by spores comprised of several cells, with one or more polar capsules and sporoplasm with one, two or three (rarely more) valves, (Cavalier-Smith, 1993). More than 1350 species of Myxosporidian were described worldwide in freshwater, brackish and marine fish (Sakiti *et al.*, 1999). The method of transmission of myxozoans is unknown; this life cycle may require a vertebrate (fish) and an invertebrate (annelid) host with each life cycle having its own sexual and asexual stages. Severe infestations by these parasites can result in disease and/or death of the host fish (Garden, 1992). They have harmful effects on fish causing mortality (Pote *et al.*, 2000). Besides direct losses, parasites may have considerable impact on production, growth and behavior of fish, their resistance to other stress factors, susceptibility to predation and reduction of marketability (Scholz, 1999). The piscine haemoparasites have not been investigated so far. Thus, the present study is aimed to investigate the prevalence of different haemoparasites in *Chrysichthys auratus* in Assiut Governorate. In addition, were evaluated the dynamic changes in leukocytic count of both infected and uninfected fish.

Materials and Methods

Fish:

Two hundred live freshwater fish (*Chrysichthys auratus*) of different sexes (118 males & 82 females), weights and lengths (57.2 ± 3.7 gm and 13.7 ± 2.2 cm) were randomly collected from the River Nile at Al Fath center in Assiut Governorate. Fish were immediately transported, on tanks, to Animal Health Research Institute, Assiut Lab. to perform the clinical and parasitological examinations. Blood samples were collected from the caudal blood vessels and heart as described by Lucky (1977). Thin blood films were made and air-dried, fixed in absolute methanol for 5 minutes and stained with Giemsa stain in phosphate buffer (pH 7.3) for 30 minutes (Woo, 1981) and examined using 100X objective on a light microscope equipped with a FGDMI HDMI ® digital camera. Morphometry analysis of the different developmental forms was performed using the ToupTek ToupView ® Software,

Version: 3.7, calibrated to a stage micrometer.

Parasitological examination:

Identification of detected parasites based on the morphology and morphometry characters and their measurements and dimensions according to **Lom and Dykova (1992); Fomena and Bouix (1997) and Borges, et al. (2016)**. The measurements of *Trypanosome* and *Babesiosoma* sp were recorded and calculated; while *Myxosporidian* spore dimensions were measured (length, width; length and width of polar capsule).

Leukogram picture:

Differential leukocytic counts (eosinophils, basophils, lymphocytes, neutrophils and monocytes) were determined according to **Stoskopf (1993)** the cells were identified according to morphological features and a mean relative percent calculated. The prevalence was calculated in accordance with **Lucky (1977)**.

Statistical analysis:

Results were analyzed by using SPSS© (ver. 16, IBM, New York, USA) using, T test, χ^2 test with 95% confidence interval, with considering P value ($P < 0.05$) as significant difference (**Sobia et al., 2018**).

Results

Blood smears from examined *Chrysichthys auratus* were observed to be infected with three blood parasites, *Trypanosoma*, *Babesiosoma* and *Myxobolus* sp. (plates 1, 2 and 3).

Infected fish demonstrated various non-specific clinical signs and gross lesions including paler or darker skin than normal, erosions, ulcers, hemorrhagic and/ or black spots or fin rot with emaciation, splenomegaly, mild ascitis and pale internal organs in some infected fish. Percentage of infections in examined fish were summarized in Table 1: out of 200 fish examined, 123(61.5%) were found infected with haemoparasites. Single infection was seen in 43% (86 out of 200) of examined fish, while mixed infection was seen in 18.5% (37 out of 200) of examined fish by more than one of blood parasites. High significant statistical variation were recorded between single and mixed infection ($\chi^2 = 28.19$, $P < 0.0001$), (Table 1). *Chrysichthys auratus* males were harboring more blood parasites 64.4% (76 out of 118) than female ones 57.3% (47 out of 82).

The prevalence of infections with *Trypanosoma* sp. were 55.5 % (111 out of 200), males were more infected than females (Table 2).

In blood films, the body of *Trypanosome* stained deep blue, with poorly stained free flagellum arising from the pointed anterior end of the body. The kinetoplast was prominent, lying close to the blunt posterior end of the body (Fig. 1). The nucleus stained pink with Giemsa stain and was closer to anterior end of *Trypanosome* than to its posterior extremity.

Measurements and morphometrical description of *Trypanosomes* demonstrated in Table (3). It is a polymorphic *Trypanosome* species that had been found in three forms; small, medium and large forms (Plate 1), while no division stages were seen in the blood smears.

Small forms (Plate 1 A-D): The thin and elongate body measured 29 - 31 μm in length and 1.8 - 2.4 μm in width. The body has pointed anterior end and slightly rounded posterior end. The free part flagellum length is 11.9 - 12.9 μm . The cytoplasm contains fine granules and many vacuoles. The oval shaped nucleus measured 2.1-3.0 μm in length and 1.1- 1.4 μm in width. The nucleus was situated in the anterior half of the body and the nuclear index (distance between middle of nucleus and posterior end divided by distance between middle of nucleus and anterior end) was 1.2-1.4 μm . The rod-like kinetoplast measured 1.1-1.4 μm in length and 0.7- 0.8 μm in width. It has terminal or slightly sub-terminal position.

Medium forms (Plate 1 E-G): The body is longer (34.3-43.3 μm in length) more wide (2.8 -3.5 μm) comparing with the small forms. The body has a pointed anterior end. The cytoplasm stained very deep blue with Giemsa stain and contains fewer vacuoles. It contains a large nucleus measured 2.3-3.4 μm in length and 2.0-2.2 μm in width. The nuclear index was 1.3-1.4 μm . It contains a terminal or slightly sub-terminal oval shaped kinetoplast. The free part flagellum length is 6.5 - 14.1 μm .

Large forms (Plate 1 H-J): The large forms of *T. cyanophilum* were not common. The body is large and wide; it measured 37.2-42.8 μm in length and 4.1-5.4 μm in width. The free part of the flagellum measured 5.2-8.5 μm . The body has a pointed anterior and posterior ends. It had deeply blue stained cytoplasm and con-

tains a large size pink stained nucleus with Giemsa stain, that measured 3.5-3.9 μm in length and 2.4-3.6 μm in width. The nuclear index is 1.3-1.6 μm . The kinetoplast was strongly shifted to the posterior tip with a measurement of 0.7-0.8 μm in length and 0.7-0.9 μm in width.

According to morphological and morphometric examinations the isolated *Trypanosoma* parasites were belonged to *Trypanosoma cyanophilum*.

Babesiosoma sp. was observed in 24 of the examined 200 *Ch. auratus* with a prevalence of 12% (Table 2). Fish Males were more sensitive to *Babesiosoma* sp. than females (Table 2).

In Giemsa-stained blood films, various stages of *Babesiosoma* parasite were found intra-erythrocytic (Plates 2 A-J). These stages were identified as merozoites, meronts (undivided meronts, meronts yielding merozoites) and gamonts. Displacement of the nucleus in the infected erythrocytes was detected in all of these stages.

Meronts in early division (undivided meronts) (Plate 2 A) were broadly rectangular, with chromatin distributed to the four corners of the parasite body, forming a tetranucleate structure. These stages were largely non-stained except for the nuclear material that stained deep purple it measured 4.8 -5.5 μm .

Cruciform meronts (late meronts) were characteristically cruciform, producing four merozoites (Plate 2 B, C). Meronts measured 4.5-4.9 μm long by 4.2-4.5 μm wide; chromatin was most noticeable at the four extremities.

Merozoites, arising from cruciform meronts (Plate 2 D - G), occurred in pairs or singly and were elongate or oval. They were largely non-stained except peripherally, where they stained blue and measured 2.8-4.0 μm long by 2.2-2.6 μm wide.

Two forms of gamonts were recognized. Immature gamonts (Plate 2 H, I) were broadly elongate with one end slightly narrower than the other end. Cytoplasm was stained pale blue and the nucleus was difficult to discern, but appeared to lie nearer the narrower end of the parasite body, this stage measured 4.7-5.7 x 2.0-2.2 μm . More mature gamonts (Plate 2 J) was curved, with a blunt and a more pointed end, the nucleus was prominent measured 4.6 x

2.4 μm , lay nearer the blunt end. This form measured 7.2 x 2.8 μm . Based on the morphological and morphometric examinations, the detected *Babesiosoma* sp. were belonged to *Babesiosoma mariae*.

Myxosporidian parasites appeared in thin blood films as *Myxobolus heterosporus* type I (Plate 3), spores are ovoid or ellipsoidal with round anterior and posterior ends. Length of spores 10.5-11.5 μm with width 6.5- 7.4 μm ; polar capsules were equal in size and pyriform in shape, located at anterior end of spore containing extrudable polar filaments, length of polar capsules 3.6-4 μm with width 2.2 μm with presence of round to oval iodophilus vacuole (glycogen vacuole) deeply stained blue. Only 26 (13%) out of 200 *Ch. auratus* were infected (Table 2). Examined males fish had higher *Myxosporidian* infection (17.8 %) than the female ones (6.1%), (Table 2).

Leukogram, in our study showed different types of leucocytes. No basophils were detected. There was significant difference in lymphocytes count of infected and non-infected fish ($t=7.189$ $P < 0.0001$), significant difference in monocytes count of infected and non-infected fish ($t=3.483$ $P < 0.0006$), significant difference in eosinophils count of infected and non-infected fish ($t=7.658$ $P < 0.0001$) while there were no significantly neutrophils in infected and non-infected fish. Moreover, only *Trypanosoma* sp infected fish had shown significant variation between them and non-infected fish in lymphocytes, monocytes and eosinophils counts, (Table 4).

Discussion

Blood parasites are common in both wild and cultured fishes. They are considered a great problem; they reduced the fish metabolism, decreased growth rate and weight loss (**Shahi et al, 2013**). The results revealed that haemoparasites in *Chrysichthys auratus* reaching prevalence of 61.5%. This may be due to feeding habitats, where fish feed on food from animal origin, plant materials, bivalves, mollusks, small fish, Copepods, crustacean and detritus planktonic organisms (**Ubong and Edidiong, 2015**). Besides, present in the bottom where waste products and organic matters accumulated which may be favorable environment for

intermediate hosts for these parasites (**Reed et al., 1967**).

Some infected fish demonstrated various gross lesions including emaciation, splenomegaly, mild ascitis and pale internal organs. Such results nearly agreed with that produced by **Mariam (2001)**; **Essam and El-Khatib (2004)** and **Eissa et al., (2008)**. The mild ascitis may be produced as a result to *Trypanosomes* and *Babesiosoma* sp. and cause dysfunction of kidneys. Pale internal organs of infected fishes may be due to anaemia and haemodilution in which *Trypanosome* sp. produce haemolysins that lysing the RBCs (**Eissa et al., 2008**).

Single infection in examined fish was more than mixed one, it may be due to the availability of the intermediate host that affect by the environmental factors, aquatic pollution, high level of organic matter and low oxygen (**Vicki et al., 2003**).

This study has shown that male *Chrysichthys auratus* fish established higher prevalence of *Trypanosoma*, *Babesiosoma* and *Myxobolus* sp. than females, such results agreed with **Woo (2006)** and **Biu and Akorede (2013)**. Only *Myxobolus* sp infection had shown significant statistical variation regarding the sex of fish. Higher infection rates may be due to differential feeding habits of males either by quantity or quality of feed (**Emere, 2000**), in addition to immunological differences among host sexes as was suggested by **Tombi and Bilong (2004)**. Variations in parasitic infection among the sexes of studied fish were not significant in *Trypanosoma* and *Babesiosoma* sp implying that higher infection rates in either male or female were simply by chance (**Biu and Akorede, 2013**).

In our work, the prevalence of *Trypanosoma* sp was 55.5%. It is known that these parasites are transmitted by the bite of leeches. Therefore, the facility of infection by leeches promotes the parasitization by *Trypanosomes*, and the behavior of the fish may contribute to a higher or lesser probability of leech infection. *Chrysichthys auratus* have a benthic behavior that facilitates the infection by leeches (**Reed et al., 1967**). **Ahmed (2001)** and **Abed (2005)** listed that prevalence of *Trypanosoma* sp was

(42.3%) and (34%), respectively in *chrysichthys auratus* in Assiut, Egypt. However, high prevalence of *Trypanosoma* sp. that reached 86.7% was reported by **Konaş et al. (2010)** in the River Asi, Turkey.

The identification of *Trypanosoma* sp. based on morphological and morphometric features revealed that closely to *T. cyanophilum*, which is polymorphic *Trypanosomes* that characterized by its deeply blue stained cytoplasm, the nucleus was situated in the anterior half of the body. This species originally described by **Mohamed (1978)** as dimorphic *Trypanosomes* from *Chrysichthys auratus* and *Ch. reupPELLI*. **Abu El- Wafa (1988)** identified this species as *T. tilapiae* from different species of fishes; later **Negm El-Din (1991)** synonymized this species with *T. cyanophilum*. **Hussein, et al. (2010)** described three forms of *Trypanosoma* from *Chrysichthys* species in Qena. A morphometric comparison among *Trypanosoma* (of the present study) and other previously detected by **Mohamed (1978)** in Egypt is presented in Table 3.

Our results identified the *Babesiosoma* as *Babesiosoma mariae* according to **Shahi et al., (2013)** and **Biu and Akorede (2013)**, with prevalence of 12%. This result is nearly similar to those recorded by **Shahi et al. (2013)** in Kashmir, India (16.6%). Moreover, **Eissa et al., (2008)** declared that *Babesiosoma* sp. showed a prevalence of 24.8% in *C. carpio* while were 46.4% and 36.2% in wild and cultured *Oreochromis niloticus* in Dakahlia governorate, Egypt. On the other hand, reports of high prevalence had been mentioned by **Eli, et al, (2012)** in Lake Victoria where it reached 46% of *O. esculenta* and 70% of *O. variabilis*, respectively. This variation in percentage of infection may be due to difference in localities from which the fish were collected, degree of water pollution or other climatic conditions (**Negm El-Din and Davies, 1999**).

The prevalence of infection with Myxosporidian in examined fish was 13%. This result was higher than that reported by **Hassan et al, (2007)** (1.09%). This alteration may be due to difference availability of invertebrate hosts and environment conditions (**Overath et al, 1999**).

The present species identified as *Myxobolus heterosporus* type 1 according reported myxosporidian key of **Fomena and Bouix (1997)**, while **Hussein (2009)** in Qena identified *Myxobolus* sp. I. from blood with spore length 11.2-12.4 μm with width 6-7.2 μm ; polar capsules were length of polar capsules 4-8 μm with width 2.4-3 μm .

The leukocytes reaction to parasites is shift of the leukogram towards granular forms of cells (**Shahi et al., 2013; Correa et al., 2016; Maqbool and Ahmed, 2016**), which was also found in our study. Number of lymphocytes in infected fish with *Trypanosoma* and *Babesiosoma* sp. was decreased than non-infected one, this line with **Rodrigo et al (2013)** and **Lapirova and Zabotkina (2018)** and opposite to **Shahi et al (2013)**. The products of the metabolism of these parasites can be highly toxic and cause serious, and in many cases lethal diseases to their hosts (**Bienek et al., 2002**). while increased in case of infection by *Myxobolus* sp. Lymphocytes activating phagocytic cells to produce enzymes capable of destroying pathogens. Moreover, they can increase the production of chemical messengers (interferon, interleukins and complement proteins) that stimulate other aspects of the immune system and increase the activity of T and B lymphocytes (**Raa, 1996**).

Number of monocytes increased in *Trypanosoma* sp. infection these agree with **Lapirova and Zabotkina, (2018)**. Some authors mention that the reaction of phagocytes to parasitic infection depends on the intensity of the invasion, parasitic species, stage and fish (**Forlenza et al., 2008**). Moreover, transferrins which penetrate the blood during lysis of erythrocytes significantly activate the killer ability of macrophages (**Stafford and Belosevic, 2003**).

Number of eosinophil was increased in *Trypanosoma* sp and *Babesiosoma* sp. infection these results are also corroborating with the finding by **Aragort et al, (2005); Ahmed et al, (2011)** and **Lapirova and Zabotkina (2018)**. The significant increase which we found in the share of eosinophils in the infected fish coincides

with the assumption that eosinophils are cells "responsible for the organism's reaction to parasites" (**Clauss et al., 2008**).

On the whole, the data we obtained indicate that "parasite-host" is a complicated system of interdependence, which is determined by many factors, of which the most important are immune physiological status and conditions of the fish habitat on the one hand and mechanisms of interaction of parasite and immune system of host on the other hand (**Khan, 2012**). Inflammatory reactions involving cellular reactions, as phagocytosis and phagocyte activity (including oxidative mechanisms), as well as complement activity, are modulated by many fish parasites, including mainly ciliates, flagellates and myxozoans (**Alvarez-Pellitero, 2008**).

Conclusion

It must be to maintain the aquatic environment away from pollution, which leads to appearance of many parasites due to the presence and richness of intermediate hosts affecting the health of fish. The infection rate of different haemoparasites was high and *Trypanosoma* infection was the highest one.

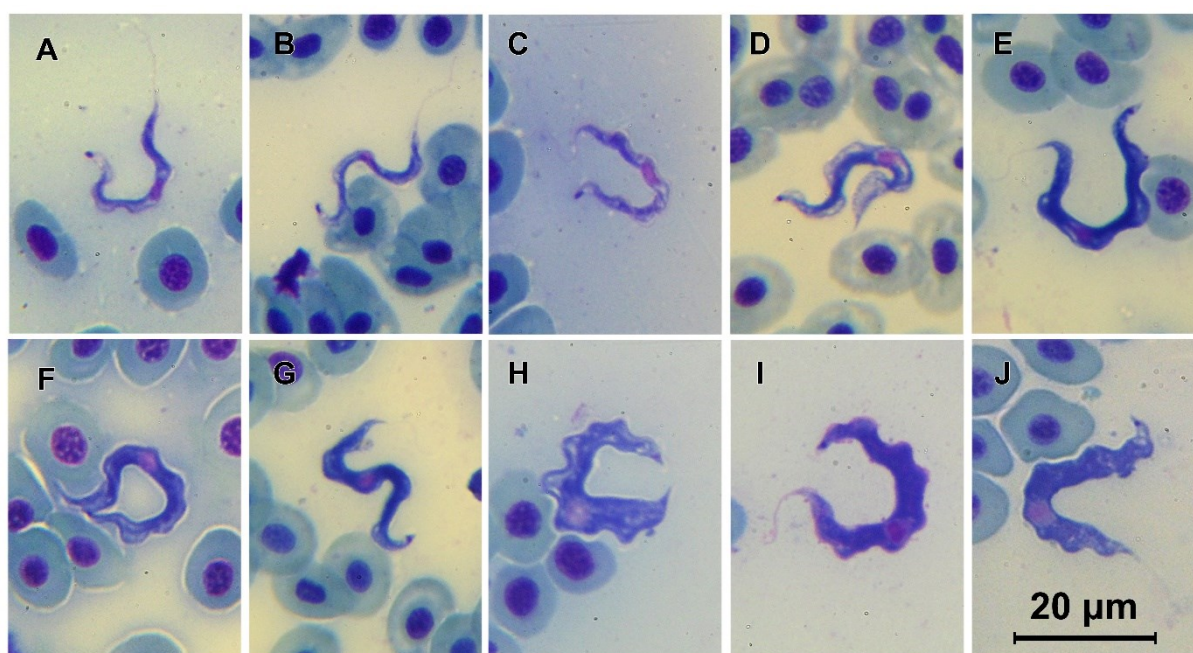


Plate (1): Giemsa stained blood films from *Ch. auratus*, showing three different morphological forms of *T. cyanoophilum*. (A - D) small forms; (E - G) medium forms and (H - J) large form.

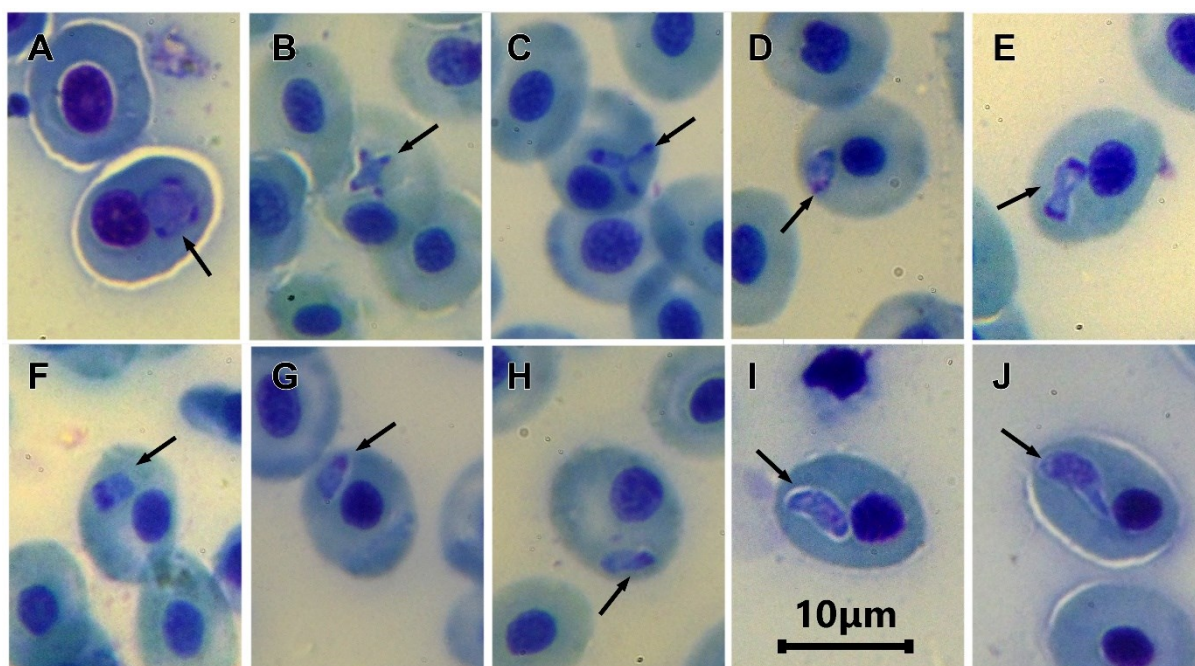


Plate (2): Blood films from *Ch. auratus* stained with Giemsa stain showing *Babesiosoma mariae*. (Arrows). (A) Undivided meronts; (B-C) Cruciform meronts; (D-G) Merozoites; (H-I) Immature gamonts and (J) Mature gamont.

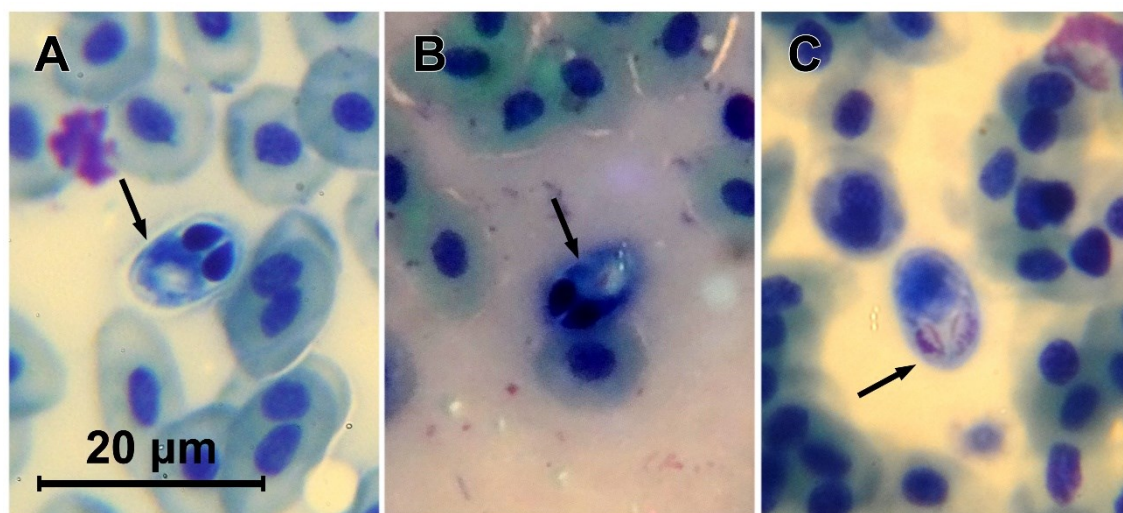


Plate (3): *Myxobolus heterosporus* type I (Arrows) in Giemsa stained blood films from *Ch. auratus*.

Table (1). Showing distribution of single and mixed infection of haemoparasites in examined *Chrysichthys auratus*.

No. of examined	Single		Mixed		Total	
200	No.	%	No.	%	No.	%
	86***	43.0	37	18.5	123	61.5

*** High significant statistical variation between single and mixed infection ($\chi^2 = 28.19$, $P < 0.0001$).

Table (2). Sex distribution of haemoparasites in examined *Chrysichthys auratus*.

	<i>Trypanosoma sp.</i>		<i>Babesiosoma sp.</i>		<i>Myxobolus sp.</i>	
Sex	Male	Female	Male	Female	Male	Female
Examined	118	82	118	82	118	82
Infected	66	45	16	8	21 [*]	5 [*]
%	55.93	54.88	13.56	9.76	17.80	6.10
Total infected	111 ^{***}		24 ^{***}		26 ^{***}	
%	55.5		12.0		13.0	

* Significant statistical variation between sexes in *Myxobolus* infection ($\chi^2 = 5.85$, $P < 0.05$).

*** High significant statistical variation between different prevalences of infection ($\chi^2 = 125.6$, $P < 0.0001$).

Table (3). Morphometric comparison of *Trypanosoma cyanophilum* (of the present study) and those previously detected by (Mohamed 1978).

	<i>T. cyanophilum</i> (present study)			<i>T. cyanophilum</i> (Mohamed 1978)	
	Small form	Medium form	Large form	Small form	Large form
TL	42.0 (40.9-42.3)	49.1 (46.6-52.4)	46.3 (42.4-51.3)	May reach 43	May reach 54
BL	30.0 (29.0-31.5)	39.2 (34.3-43.3)	39.5 (37.2-42.8)	25.9 (17.8-30.2)	
BW	2.0 (1.8-2.4)	3.2 (2.8-3.5)	4.7 (4.1-5.4)	1.4 (0.8-2.8)	
FF	12.5 (11.9-12.9)	9.9 (6.5-14.1)	6.8 (5.2-8.5)	12.8	11.6
NL	2.5 (2.1-3.0)	2.7 (2.3-3.4)	3.7 (3.5-3.9)	2.8 (1.2-3.8)	3.6 (2.4-4.8)
NW	1.27 (1.1-1.4)	2.0 (1.9-2.2)	3.2 (2.9-3.6)	1.3 (0.6-2.0)	3.2 (1.4-5.6)
KN	16.17 (14.9-17.3)	20.9 (18.0-23.3)	22.1 (20.7-23.3)	14.2 (9.4-17.9)	18.5 (13.2- 26)
PK	0.7 (0.6-0.9)	1.3 (1.0-1.9)	1.2 (0.8-1.4)	0.5 (0.0 - 0.8)	0.5 (0.0-1.8)
PN	16.9 (15.6-18.2)	22.2 (19.0-25.2)	23.3 (22.1-24.1)	14.7 (9.4-18.7)	19 (13.2-28.4)
NA	13.1 (12.8-13.3)	17.0 (15.2-18.3)	16.0 (14.8-18.3)	12.3 (4.6-14.7)	15.8 (12.4-19.6)
KW	0.7 (0.7-0.8)	0.7 (0.5-0.9)	0.8 (0.7-0.9)	0.53 (0.2 -1.5)	0.56 (0.2-0.8)
KL	1.3 (1.1-1.4)	0.7 (0.6-0.8)	0.7 (0.7-0.8)	0.94 (0 4-1.6)	0.66 (0.2-1.)
NI	1.3 (1.2-1.4)	1.3 (1.3-1.4)	1.5 (1.3-1.6)	1.9 (1.27-2.4)	1.2 (1.06- 1.45)
UW	1.3 (1.2-1.4)	2.2 (1.9-2.3)	2.9 (2.6-3.2)	1.43 (0.8 - 2.8)	3.7 (2.2-6.2)

TL: total body length with flagellum , **BL :** body length along the cell midline, **BW :** body width at the center of the nucleus, **FF:** length of the free flagellum , **NL:** Nucleus Length , **NW:** nucleus width at the center of the nucleus, **KN:** distance from the center of the kinetoplast to the center of the nucleus , **PK:** distance from the center of the kinetoplast to the posterior of the cell, **PN:** distance from the center of the nucleus to the posterior of the cell, **NA:** distance from the center of the nucleus to the anterior of the cell , **KW:** Kinetoplast Width , **KL:** Kinetoplast Length , **NI:** the nuclear index (= NP/ NA) and **UW:** undulating membrane width.

Table (4). Leukogram Picture (Mean \pm SD) in *Chrysichthys auratus* infected with haemoparasites.

Cells Parasites	Lymphocytes		Monocytes		Esinophils		Neutrophils		Basophils	
	Mean	SD	Mean	SD	Mean	SD	Mean	SD	Me an	SD
Tyrpanos.	87.05***	5.088	5.697***	2.838	7.039***	3.862	0.2105	0.5493	0*	
Babsios	91.43	3.047	3.571	1.988	4.857	1.952	0.1429	0.378	0	
Myxoblus	93.33	1.528	3.667	1.155	3	1	0	0	0	
Non infected fish	92.13	4.06	4.26	2.643	3.494	2.18	0.1558	0.4884	0	

* Undetectable

*** High significant statistical variation between *Trypanosoma* sp infected and non-infected fish ($t=7.2$, $P < 0.0001$); ($t=3.5$, $P < 0.0001$) and ($t=7.7$, $P < 0.0001$) among Lymphocyte; Monocyte and Eosinophil, respectively.

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