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STUDIES ON SOME PARASITES INFECTING THE NILE FISH *CLARIAS LAZERA* IN ASSIUT LOCALITY

(With 4 Tables and 8 Figures)

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دراسات على بعض الطفيليات التي تصيب السمكة النيلية
كلارياس لازيرا بإقليم أسيوط

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تم وصف نوعين من طفيليات التريبانوسوما من دم السمكة النيلية كلارياس لازيرا وكذلك نوع من طفيلي الميكزوبولس ونوع واحد من طفيلي الهينجوييا ونوع من طفيلي الجلوسديم ونوع من طفيلي الأروتتوكرديم. وقد تم وصف هذه الأنواع وصفاً تفصيلياً وقورنت بالأنواع الأخرى القريبة لها من نفس الجنس.

SUMMARY

Four species related to three genera of Protozoan parasites are collected from the Nile fish *Clarias lazera*. They are *Trypanosoma* sp. indet.; *Trypanosoma alhaussainii* Mohammed, 1978; *Myxobolus* sp. indet. and *Henneguya assiuti* Mandour *et al.*, 1988. Two species related to two genera of trematodes collected from the same fishes, they are *Glossidium aswanensis* Abdel-Maksoud, 1988 and *Orientocreadium batrachoides* Tubangu, 1931.

Key words: *Clarias lazera*, *Trypanosoma* sp., *Trypanosoma alhaussainii*, *Myxobolus* sp., *Henneguya assiuti*, *Glossidium aswanensis* and *Orientocreadium batrachoides*.

INTRODUCTION

Records of parasites from the Nile fish *Clarias lazera* have been observed by many authors such as Fischthal and Kuntz (1963);

Mohamed (1978); Abed (1987); Mandour *et al.* (1988); Abdel-Maksoud (1988) and El-Damarany (1992).

The aim of present survey is to study and identify the parasites observed from fish *C. lazera* in Assiut locality.

MATERIALS and METHODS

A hundred adult fish *Clarias lazera* were captured from River Nile at Assiut. Immediately blood smears were made and stained with Giemsa stain and examined microscopically for blood parasites. The smears of myxosporean spores were carried out by homogenization of each organ of such fish. The spores were examined fresh, and after addition of Lugol's iodine solution to show the iodophilous vacuole. Other slides were treated with saturated urea solution or in 5% potassium hydroxide solution to make expulsion of the polar filaments. Others impression smears from kidney, muscles and spleen were stained and examined microscopically. The helminth parasites were collected from intestine, fixed and stained with acetic acid alum carmine and examined microscopically. The measurements were given and compared with related materials previously described from Egypt.

RESULTS and DISCUSSION

Out of 100 fish examined ,only 30(30%) were found to be infected with trypanosomes. Two species of *Trypanosoma* are recorded in the present study *Trypanosoma* sp.indet. and *T. alhausainii*.

***Trypanosoma* sp.indet.** Morphologically monomorphic, body pyriform and widest in posterior third with slightly rounded anterior and posterior ends (fig. 1).Cytoplasm is granular showing numerous darkly stained granules and numerous vacuoles.Nucleus is spherical to oval in shape,it stains red with Giemsa stain and lies at the middle third of the body.The subterminal kinetoplast is a large spherical structure lying near the posterior end of the body and stains deep violet. Flagellum extends along the undulating membrane which stains faint blue and extends anteriorly to form the free flagellum.Undulating membrane moderately developed with one to four undulations.

Trypanosomes of fishes have been distinguished by the shape ,size, and relative position of organelles in stained specimens (Lom, 1979). Qadri (1962) described a monomorphic *Trypanosoma batrachis* from *Clarias batrachus* from India. Saoud (1976) recorded, without

description, trypanosomes from *Clarias anguillaris* and *Clarias lazera* in the white Nile of Sudan. Mohammed (1978) described *Trypanosoma alhaussainii* from *Clarias lazera* collected from River Nile in Egypt which is the same host. Two morphologically different species of *Trypanosoma* are described during the present study from the fish *Clarias lazera*. Both species were monomorphic. *T. alhaussainii* described by Mohammed (1987) were long and slender, tapering at both ends, whereas those of present species were pyriform with a rounded anterior and posterior ends.

Accordingly, the present Parasite differ from *Trypanosoma alhaussainii* in size, shape of the body, kinetoplast and number of undulations of undulating membrane (Table, 1). However it is preferable to do further work including life cycle before considering it as a new species.

Trypanosoma alhaussainii Mohamed, 1978, Body is long and cylindrical with pointed anterior and posterior ends (Fig.2). Cytoplasm stains faint violet with Giemsa stain, it is granular showing numerous darkly stained granules and show numerous vacuoles. Nucleus is oval in shape, with the longest diameter lying parallel to the longitudinal axis of the body. It stains red with Giemsa stain and lies posterior to the middle of the body. Kinetoplast is rod-like structure which lies in the posterior end of the body. Flagellum extends along the undulating membrane and is thrown 3-6 folds.

It is clear from the above descriptions that the parasite under discussion is more or less similar to that described by Mohammed (1978) as a monomorphic *Trypanosoma alhaussainii* (Table, 1).

Myxobolus sp.indet.: Only 35(35%) out of 100 fish examined were infected. The spores of the present parasite are collected from muscle, kidney and spleen of *C.lazera*. The spores are oval in shape with narrow pointed anterior and broad rounded posterior ends, each measures $12.95-18.2 \times 8.5 - 11.9\mu$. The two equal polar capsules located anteriorly, each measures $5.5-8.7 \times 2.0 - 3.7\mu$. The polar filament measured 58.9μ , when it is extruded, but when it is resting inside the polar capsule, it is composed of 9.0 coils. The sporoplasm is granulated and contains an iodophilous vacuole measured $2.1-3.9\mu$.

The presence of two polar capsules located at the anterior end and the sporoplasm with an iodophilous vacuole, allocate present parasite under the family Myxobolidae Thelohan, 1892. Moreover, the characters of the spores conform with those of the genus *Myxobolus* Butschli, 1882.

Many species of *Myxobolus* have so far been described from fishes by many authors such as: Fahmy *et al.* (1971, 1975), Abed (1987) and Mandour *et al.* (1993). Myxosporidia are classified largely on the basis of size and morphology of the spores and on their host and organ specificity (Kudo, 1920 and Mitchell, 1987).

When the present material is compared with previously described species, it is clear that the present parasite is not comparable with *Myxobolus niloticus* (Fahmy *et al.*, 1971) and *Myxobolus* sp. (type 1) *Myxobolus* sp. (type 2) *Myxobolus* (type 3), described by Fahmy *et al.* (1975), since the hosts and morphology of the spores differ from the present material.

Accordingly, the parasite under discussion is compared with *Myxobolus clarii* described by Mandour *et al.* (1993) (Table, 2), since these parasites are found in the fish *Clarias lazera* as that recovered in the present work. So, the authors suggest that the present material is a new finding and needs detailed study to erect a new species.

Henneguya assiuti Mandour *et al.*, 1989. The species of the present parasite are collected from gill filaments and respiratory trees of fish *Clarias lazera* in the form of macroscopic cysts.

The parasite was found in 25% of a total of 100 fish examined. The spores are pyriform in shape, the posterior end of the spore is prolonged into more or less extended processes to form two often equal caudal appendages, 9.5-13.84 μ by 4.5 -6.0 μ ; the total length of the spore (including caudal appendages) reached 38.6-51.3 μ . Two equal polar capsules are situated at the anterior end, measuring 4.8-8.1 μ . by 1.5-2.0 μ with a polar filament measuring 40.3-51.9 μ . in length when fully extruded, but when resting inside the polar capsules it consists of 9.0 coils. When lugol's iodine is added a brownish rounded mass is detected within the sporoplasm of the spore commonly known as iodophilous vacuole measuring 1.2-2.6 μ . The sporoplasm is coarsely granulated. Two sporoplasmic nuclei are clearly visible, each measures 1.2-2.0 μ .

The main characters of the present parasite are identical with those of genus *Henneguya* Thelohan, 1892, family Myxobolidae Thelohan, 1892.

Mandour *et al.* (1988) described *H.assiuti* from the same host, habitat and locality. They recorded that the spore measured 41.02-49.98x4.62-5.74 (Total length including caudal appendages x width), while in the present material it measured 38.6-51.3x4.5-6.0 μ (total length x maximum width)

When the present species is compared with previously described species, it is clear that the species under discussion is more or less similar to *Henneguga assiuti* described by Mandour *et al.* (1988).

Glossidium aswanensis Abdel-Maksoud, 1988. The parasite was found mixed with another trematode parasite, *Orientocreadium batrachoides*. Only 30 (30%) of 100 fish examined were infected. Morphology: (based on 3 adults). All measurements of the worm are shown in table (3). The body is elongate, of fairly uniform width. The cuticle is covered with minute spines which are denser anteriorly and sparser posteriorly. The subterminal oral sucker is crown-shaped. The acetabulum is more or less circular, situated at the level of the anterior third of the body. The pharynx is a strong muscular organ. Oesophagus is short. The bifurcation of the intestine occurs approximately in a region halfway between the two suckers. The caeca are long, extend to the beginning of the last body fifth. The genital pore is situated just in front of the acetabulum. The testes are situated obliquely tandem in the posterior half of the body. The anterior testis is slightly smaller than the posterior one. The elongated claviform cirrus sac has its anterior end slightly swollen, curves around the acetabulum. There is a bipartite seminal vesicle with a large posterior part inside the cirrus sac which is filled with prostatic cells. A short duct leads to an reversible cirrus. The ovary is situated on the right side of the median line, separated from the acetabulum by the posterior end of the cirrus sac. Follicular vitellaria lie in the lateral middle fields of the body, sometimes overlapping the intestinal caeca, extending from the posterior level of the acetabulum to the posterior border of the posterior testis. The uterine coils extend between the testes reaching the posterior end of the body. The numerous, operculate eggs have light yellow thin shells. The excretory vesicle could not be seen (Figs. 5, 6).

The above description places the present parasite in the Family Plagiorchiidae Luhe, 1901; subfamily Styphlodorinae Dollfus, 1937 and the genus *Glossidium* Looss, 1899. In (1899) Looss erected that genus which included two species *Glossidium pedatum* from *Bagrus bayad* and *B. docmac* caught from River Nile. Yamaguti (1958) added *Glossidium geminum*, (Mueller, 1930). Van Cleave and Mueller (1934) transferred the species to *Alloglossidium*. Abdel-Maksoud (1988) transferred the new species. *Pristotrem clarii* which was described by El-Naffar (1970) from *Clarias lazera*, caught from Assiut, to the genus *Glossidium* Looss, 1899. He also erected a new species and suggested the taxonomic name *Glossidium aswanensis* from *Clarias lazera* caught from Aswan. So

genus *Glossidium* now contains only two species which are *G.pedatum* Looss, 1899 and *G. aswanensis* Abdel-Maksoud, 1988.

From table-(3) and the description of the present material it is evident that the species under consideration is more or less similar to *Glossidium aswanensis* described by Abdel-Maksoud (1988). However, Assiut Governorate is a new locality for the parasite.

Orientocreadium batrachoides Tubungui, 1931. The present parasite is collected from the intestine of *C. lazera*. Only 30% of total 100 fish examined were infected. The parasite was encountered in a mixed infection with *Glossidium aswanensis*, the worm burden varied from 2-12 per infected fish. Morphology: (Based on 5 adults). Measurements are shown in table (4). The body is elongated to oval in shape, spinose, widest at testicular region (Figs. 7, 8). The two suckers are more or less circular in out line and nearly equal in size. The oral sucker is subterminal. The acetabulum is located at boundary of first and second thirds of the body. Prepharynx short and leads into a well developed pharynx, oval to round in shape, and followed by a short oesophagus. Cecal bifurcation about half way between oral sucker and acetabulum. The two tubular intestinal caeca are wide, extending laterally and ending near the posterior extremity of the body. The two testes are slightly oblique but may be tandem in well-extended specimens. They are ovoid, located in the second half of the body. There is a conspicuous inter-testicular space. The posterior testis is slightly bigger than the anterior one. The cirrus sac is pear-shaped, thick walled and lies dorso-lateral to the acetabulum. It encloses a well developed seminal vesicle, which end by a cirrus that opens in the genital pore. The ovary is nearly rounded to oval in shape, lies postero-lateral to the acetabulum and may be in contact with it. The small pear-shaped receptaculum seminis lies between the ovary and the anterior testis. Vitellaria in lateral fields, follicles large, extending from level of the ovary to the posterior extremity, lateral, ventral, and dorsal to the caeca. The number of follicles is about 17-28 on each side. The uterus is coiled, extending to posterior extremity, lateral body margins, and acetabulum. Eggs numerous, operculate, oval in shape and yellowish in colour.

The present species under consideration is identical with *Orientocreadium batrachoides* described by Tubungui (1931) from specimens collected from the intestine of the fish host, *Clarias batrachus* caught from Philippines. It appears that this species has a wide geographical distribution. It has been recorded from the Philippines and India in Asia (Tubungui, 1931 and Kakaji, 1969). In Africa, Khalil

(1961) described *Orientocreadium lazera* as a new species from *Clarias lazera* from Sudan and this species was the first to be recorded of the genus from Africa. He stated that it resembles to some extent *O. batrachoides*. In Egypt, *O. batrachoides* Tubangui, 1931 has been recorded and redescribed by several authors. Table (4) shows a comparison between the present species and some of those previously described from Egypt. From the table it is evident that the present parasite is more or less similar to those described by Fischthal & Kuntz (1963) and El-Damarany (1992). Therefore, the present parasite is identical with *Orientocreadium batrachoides* Tubangui, (1931). Moreover, Assiut Province is a new locality for the parasite.

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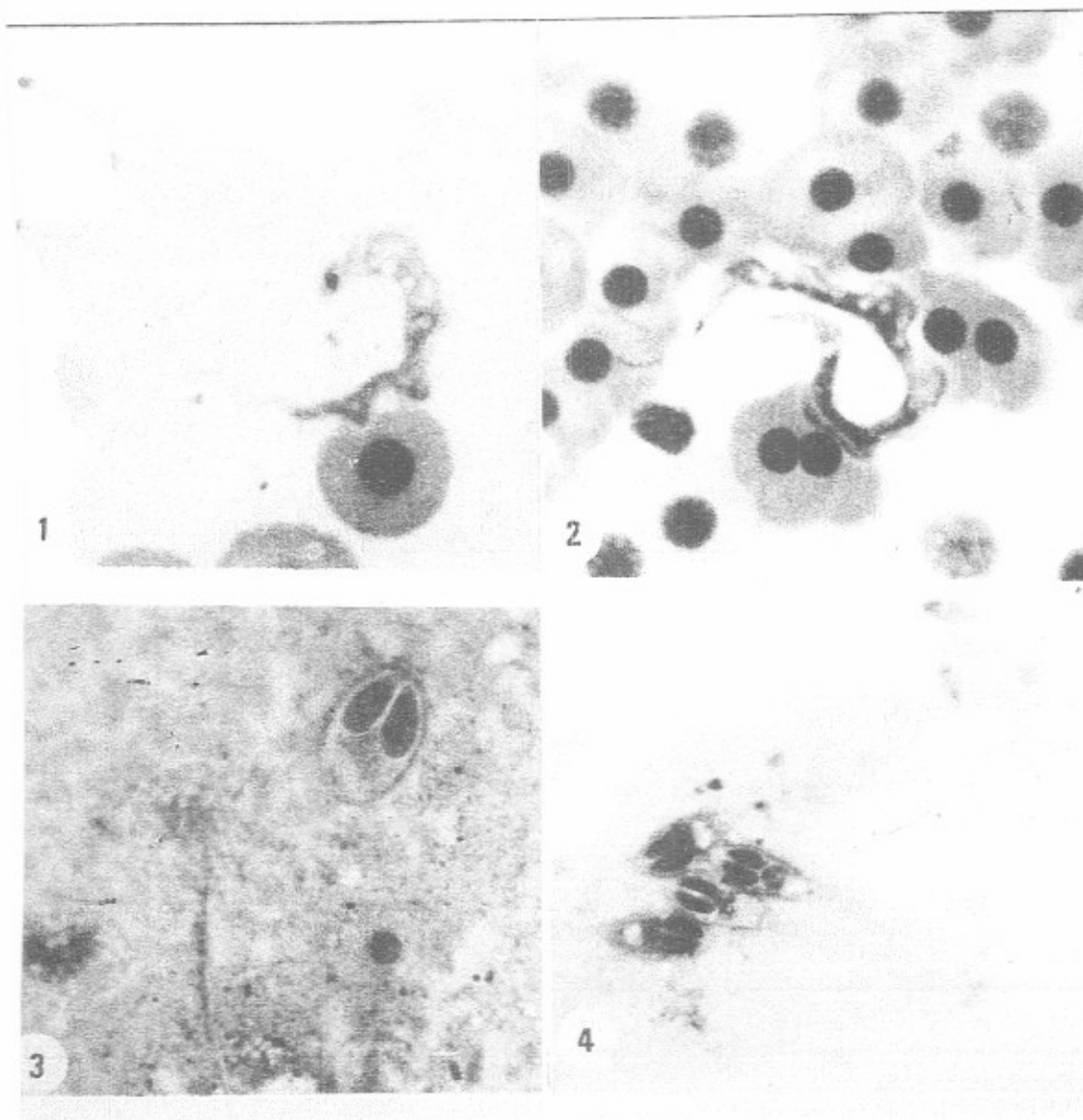
EXPLANATION OF FIGURES

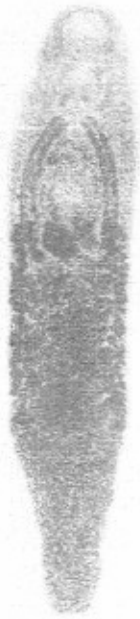
- Fig. 1:** Photomicrograph showing *Trypanosoma* sp. X 1250.
- Fig. 2:** Photomicrograph showing *Trypanosoma alhoussainii* X 1250.
- Fig. 3:** Photomicrograph showing *Myxobolus* sp.X 1250
- Fig. 4:** Photomicrograph showing *Henneguya assiuti*.X 1250
- Fig5:** Photomicrograph showing ventral view of *Glossidium aswanensis* X100.

Fig. 6: Photomicrograph showing ventro-lateral view of *Glossidium aswanensis* X.52.

Fig. 7: Photomicrograph showing the ventral view of *Orintocreadium batrachoides* X.100.

Fig. 8: Photomicrograph showing the ventro-lateral view of *orintocreadium batrachoides* X. 100.





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Table 1: Comparison between trypanosomes of Nile fish *Clarias lazera*.

Measurements	Present work		<i>Trypanosoma alhussainii</i> Mohammed, 1978
	<i>Trypanosoma sp</i>	<i>Trypanosoma alhussainii</i>	
Length of Kinetoplast	0.32-1.30 = 0.81 μ	0.38-1.71 = 1.05 μ	0.4-1.4 (1.12 μ)
Width of Kinetoplast	0.24-0.65 = 0.45 μ	0.23-0.82 = 0.53 μ	0.4-0.8 (0.6 μ)
From posterior end of the body to posterior edge of Kinetoplast	0.07-1.71 = 0.89 μ	0.68-2.02 = 1.35 μ	0.6-3.2 (1.13 μ)
Length of nucleus	1.74-3.42 = 2.58 μ	2.87-3.42 = 3.15 μ	2.8-5.0 (3.71 μ)
Width of nucleus	1.0-2.74 = 1.58 μ	2.05-2.05 = 2.05 μ	1.0-2.8 (1.72 μ)
From anterior end of Kinetoplast to posterior edge of nucleus	10.94-13 = 11.97	13-19.84 = 16.42	10.4-18.8 (16.35 μ)
From anterior end of nucleus to anterior end of the body	10.02-18.84 = 14.43 μ	17.1-21.89 = 19.50 μ	16.8-25.6 (20.27 μ)
Length of free flagellum	5.47-8.89 = 7.18 μ	11.97-13.60 = 12.79 μ	5.6-10.4 (8.17 μ)
Total of length excluding free flagellum	25.11-36.65 = 30.88 μ	33.02-45.72 = 39.37 μ	35.2-48.4 (41.23 μ)
Maximum width of Undulating membrane	0.57-1.71 = 1.14 μ	1.37-2.05 = 1.71 μ	0.6-1.6 (1.2 μ)
Width at level of nucleus	2.74-3.08 = 2.91 μ	3.08-3.48 = 3.28 μ	1.2-4.0 (1.98 μ)

Table 2: Comparison between *Myxobolus sp.* collected from Nile fish *C.lazera* and *Myxobolus clarii* described by Mandour *et al.* (1993).

Characters	<i>Myxobolus clarii</i> Mandour <i>et al.</i> 1993	The present materials
Host	<i>Clarias lazera</i>	<i>Clarias lazera</i>
Habitat	Testis	Muscles, kidney and spleen
Locality	River Nile at Assiut	River Nile at Assiut
Size of spore	9-12.21 (10.61 μ) \times 7.50-9.90 (8.7 μ)	12.95-18.20 \times 8.5-11.9 (15.5 \times 10.2 μ)
Size of polar capsule	3.50-4.78 (4.14 μ) 2.15-2.74 (2.45 μ)	5.5-8.7 \times 2.0-3.7 (7.1 \times 2.8 μ)
Polar filament	22.25-35.57 (29.07 μ)	58.9 μ
No. of coils of filament	5 coils	9.0 coils
Shape of the iodophilous vacuole	Rounded mass measured 1.37-2.74 μ	Elliptical to rounded in shape measured 2.1-3.9 μ
Sporoplasm	Finely granulated	Granulated

Table 3: Comparison between the present trematode of *Glossidium aswanensis* and Abdel-Maksoud's (1988). All measurements are in millimeters.

Characters	Abdel - Maksoud (1988)'s specimen	The present material
Tot. body length	2.65 - 2.76	2.14 - 2.71 mm
Tot. body width	0.450 - 0.500	0.44 - 0.48
Oral sucker	0.193 - 0.229 in length	0.184 - 0.191 x 0.224 - 0.227
Acetabulum	0.194 - 0.200 in diameter	0.192 - 0.196 x 0.210 - 0.214
Pharynx	0.07 - 0.08 x 0.09 - 0.1	0.08 - 0.084 x 0.095 - 0.1
Ant. Testis	0.223 - 0.229 in diameter	0.173 - 0.211 x 0.176 - 0.224
Post. Testis	0.259 - 0.267 in diameter	0.210 - 0.251 x 0.203 - 0.268
Ovary	0.158 - 0.168 x 0.190 - 0.200	0.098 - 0.156 x 0.154 - 0.210
Egg	0.043 - 0.051 x 0.027 - 0.032	0.034 - 0.050 x 0.017 - 0.029
The cuticle	Spinose	Spinose
Host	<i>Clarias lazera</i>	<i>Clarias lazera</i>
Locality	Lake Naser, Aswan	River Nile, Assiut.

Table 4: Comparison between *Orientocreadium batrachodes* Tubanguï, 1931 and previously described related species from Egypt.

Characters	Fischthal & Kuntz (1963)'s specimen	El-Damarany (1992)'s specimen	The present specimen
- Body length	1.045-1.770 (1.298)	0.75-2.26(1.46mm)	1.031-2.11(1.52mm)
- Body width	0.245-0.505 (0.37)	0.22-0.56 (0.41)	0.31-0.67 (0.49)
- Oral sucker	0.105-0.145x0.100-0.140	0.08-0.21x0.10-0.17	0.11-0.18x0.13-0.20
- Acetabulum	0.11-0.15x0.11-0.16	0.10-0.21x0.09-0.24	0.12-0.20x0.10-0.20
- Prepharynx	0.025-0.055 (0.043)	0.029-0.070 (0.047)	0.075-0.091 (0.077)
- Pharynx	0.06-0.09x0.05-0.10	0.079- 0.145x0.099-0.16	0.080-0.138x0.085-0.156
- Anterior testis	0.125-0.20x0.13-0.20	0.11-0.25x0.13-0.24	0.13-0.25x0.16-0.31
- Posterior testis	0.13-0.23x0.10-0.22	0.13-0.27x0.13-0.24	0.14-0.25x0.16-0.31
- Internal seminal vesicle	0.03-0.05x0.02-0.04	0.05-0.10x0.03-0.10	0.07-0.17x0.04-0.13
- Ovary	0.10-0.18x0.09-0.10	0.08-0.21x0.10-0.34	0.11-0.22x0.10-0.20
- Excretory bladder shape	Elongate, tubular or saccular	Tubular or saccular	Tubular
- Cirrus	Short, muscular & unspined	Short, muscular & unspined	Short, muscular & unspined
-Distance from acetabulum to testes	--	--	0.281-0.348
-Post-testicular body length	--	--	0.411-0.602
- Egg.	28-33x17-20 Um	31 - 39x15-20 Um	29-34x16-19 Um
- Host.	<i>Clarias lazera</i>	<i>Clarias lazera</i>	<i>Clarias lazera</i>
- locality.	Giza Province.	Sohag Governorate	Assiut Province