

***ASTIOTREMA BIRMANII* N.SP. (DIGENEA: PLAGIORCHIIDAE) IN FROG (*RANA TIGRINA* DAUD.) FROM KARACHI, PAKISTAN**

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خلاصہ

Astiotrema جنس reptiles، amphibian اور مچھلیوں سے رپورٹ ہو چکا ہے۔ اس جنس کی ایک نئی نوع *Astiotrema birmanii* کراچی کے مختلف علاقوں میں پائے جانے والے مینڈک سے دریافت کی گئی ہے۔ اس نئی نوع کی خصوصیات یہ ہیں۔ اس کا جسم لمبوتر، پتلا اور بیضوی شکل کا ہے۔ جسم پر چھوٹے چھوٹے خار موجود ہیں۔ oral sucker کے ventral sucker سے چھوٹا ہے۔ Pharynx نمایاں جبکہ Esophagus کی لمبائی اوسط اور acetabulum جسم کے وسط میں ہے۔ اس کا cirrus pouch لمبا اور اس کی ovary تین حصوں پر مشتمل ہے۔ انڈوں کا سائز 0.027-0.036mm by 0.046-0.054 ہے۔

Abstract

The genus *Astiotrema* Looss, has been reported from amphibians, reptiles and fish. *Astiotrema birmanii* n.sp. is described from the frog *Rana tigrina* Daud. collected from different localities of Karachi, Sindh, Pakistan. A total fourteen frogs were examined for helminth parasites, thirty one trematodes were collected from the intestine of four hosts. The new species is characterized by possessing the following characters: Body elongate, slim, cylindrical, slightly curved with minute spines. Oral sucker smaller than ventral sucker. Pharynx prominent, esophagus medium in length, acetabulum in mid body; Cirrus pouch elongate reaching mid esophagus with prostrate complex and a large seminal vesicle. Ovary trilobed; genital pore immediately anterior to caecal bifurcation, Vitellaria located anterior to acetabulum. Testis oval to balloon shaped, posterior testis slightly larger than anterior testis. Uterus extracaecal except near the caecal endings. Excretory pore terminal, eggs numerous, small, oval measuring 0.046-0.054 by 0.027-0.036 mm. The present species are relatively closest to *A. sudanensis* (Khalil, 1959) Bhutta and Khan, 1974 originally reported from fresh water turtle (*Trionyx triunguis*) and later reported in turtle (*Lissemys punctata*) from Punjab.

Key Words: *Astiotrema birmanii* n.sp., *Rana tigrina*, trematode, intestine, Karachi.

Introduction

The genus *Astiotrema* Looss, 1900 includes gastrointestinal parasites of amphibians, reptiles and fishes. It is cosmopolitan and has been reported from Pakistan as well (Farooq and Qazi 1963; Bilquees, 1973; Bilquees 1978). The data concerning the species composition of this genus at present and the validity of its representatives are contradictory (Bray *et al.*, 2006). Yamaguti (1971) reported a total of 29 species in the genus. Depending on the assessment and significance of various criteria for the delineation of species in the genus *Astiotrema* many species were reduced to synonyms. Thus, no consensus has been established for either the species composition of *Astiotrema* belonging to the family Plagiorchiidae Luhe, 1901. Subsequently over the years three genera have been synonymised with *Astiotrema* and in some cases the synonym is not considered valid. Yeh and Fotedar (1958), Fotedar (1971) and Kumari *et al.*, 1973 considered *Gauhatiiana* Gupta, 1955; *Tremiorchis* Mehra and Negi, 1925 and *Pseudoparamacroderoides* Gupta and Agarwal, 1968 as synonyms of the genus *Astiotrema*. The present authors do not agree with Yeh and Fotedar (1958) that size of oral sucker and length of caeca are features of grave importance as they can vary but agree with Agarwal (1966) who reported that these characters may differ. It is therefore suggested that with adult morphology of trematodes which provide important information regarding generic differentiation which is oppressed by the life cycle patterns, but since life histories of large number of groups are entirely unknown it is impossible to clarify all genera of digenea on basis of biotic relationship alone. Looss (1900) attributed cirrus pouch large, reaching to ovary, genital pore preacetabular, vitellaria terminating at level of anterior or posterior testis as the main characters of the genus. Thus we should rely on adult morphological characters and ecology with some more study on those

groups whose life history is unknown. Here we describe a new species of *Astiotrema* (Digenea: Plagiorchiidae) in frog (*Rana tigrina*) from Karachi, Sindh, Pakistan.

Materials and Methods

Fourteen frogs (*Rana tigrina* Daud.) were collected from different localities of Karachi, Sindh, Pakistan and examined for the helminth parasites. During examination of intestinal content, thirty one trematodes were collected from four hosts with the help of forceps, needle and fine brushes in Petri dishes containing normal saline. The trematodes recovered were fixed in 70% ethanol and pressed for 24 hours. Stained with Mayer's carmalum and dehydrated in alcohol series, cleared in clove oil and xylol. They were permanently mounted in Canada balsam. Diagrams were prepared using camera Lucida. Measurements of the worms minimum and maximum are presented in millimeters. Photomicrographs were prepared using automatic photographic camera mounted on a research microscope Nikon Optiphot-2. Holotypes and paratypes are in collection of the second author. The identification of trematodes was done with the help of specific keys and literature available of Yamaguti(1937, 1971), Loss, A. (1099) and Luhe, M.(1901, 1909).

Result and Discussion

Astiotrema birmanii n.sp.

(Fig: 1-2)

Host:	Frog (<i>Rana tigrina</i> Daud.)
Site of infection:	Intestine
No. of host:	Examined/Infected: 14/4
No. of specimen recovered:	31

Description

Body elongate, slim, cylindrical, slightly curved anterior region narrower with minute spines measuring 1.65-1.82 by 0.31-0.42. Oral sucker smaller than acetabulum, muscular, round, terminal measuring 0.11-0.14 by 0.12-0.14. Pharynx prominent measuring 0.036-0.047 by 0.040-0.049. Esophagus medium in length measuring 0.08-1.12 by 0.012-0.016. Caeca terminating 1.30-1.41 from posterior end. Acetabulum in mid-body, larger than oral sucker measuring 0.15-0.18 by 0.14-0.17 at a distance of 0.76-0.84 from oral sucker. Cirrus pouch elongate reaching to mid esophagus measuring 0.12-0.18 containing large seminal duct. Genital pore immediately anterior to caecal bifurcation. Ovary trilobes in posterior half of the body, sub-median, 0.060-0.058 by 0.050-0.063. Vitellaria located only anterior to acetabulum. Testes in the second half of the body, oval to balloon shaped, posterior testes slightly larger than anterior, the anterior measures 0.13-0.23 by 0.13-0.16, the posterior 0.25-0.27 by 0.15-0.19. Excretory pore terminal. Uterus extra-caecal, except near the caecal ending where it becomes inter-caecal. Eggs small, numerous, oval measuring 0.046-0.054 by 0.027-0.036.

As compared to type species *A. reniformis* (Looss, 1898) Stossich, 1904 the present specimens differ in position of vitellaria in *A. reniformis* they reach almost upto posterior testes while in the present species they end anterior to acetabulum, in *A. reniformis* the cirrus pouch does not reach esophagus while in the present specimens it reaches mid length of the esophagus, moreover the shape of ovary, testes, suckers are different in present species from *A. reniformis*.

The body of the present specimens is smaller as compared to *A. amydae* Ogate, 1938 (4.2 by 1.1); *A. fochowense* Tang, 1941 (5 by 1.21); *A. fukuii* Ogata, 1938 (9 by 1.6); *A. gangeticum* Harshe, 1932 (4.0-6.7 by 1.74-2.08); *A. giganticum* Tiwari, 1958 (10-12.5 by 2.32-2.96); *A. hoshiaurpurium* Gupta, 1954 (3.24-4.88 by 1.32-1.4); *A. indicum* Thapar, 1933 (10-11 by 2.15-2.30); *A. lobiorchis* Tiwari 1958 (6.6 by 2.3); *A. loosi* Mehra, 1931 (2.7 by 1.0); *A. Magniovum* Fiocthall and Kuntz, 1965 (6.6 by 2.3); *A. mehrai* Tiwari, 1958 (5.7-6.9 by 1.19-1.49); *A. monticellii* Stossich, 1904 (2.2-2.4 by 0.33-0.58); *A. nathi* Gupta, 1954 (4.08-5.30 by 0.85); *A. odherni* Bhalerao, 1936 (4.6 by 1.0); *A. odherni* Besprozvannykh *et al*, 2015 (3.20-3.54); *A. orientale* Yamaguti, 1937 (4.2-4.0 by 0.80-0.85); *A. rami* Bhalerao, 1936 (1.96-3.67 by 0.6-1.1); *A. spinosum* Chatterjee, 1935 (6.9 by 1.2); *A. sudanense* Khalil, 1959 (2.5-2.90 by 0.6-0.9); *A. thapari* Gupta, 1954 (9.0 by 2.28). *Astiotrema lissemysi* Farooq and Qazi, 1963 differs from the present species in the size of the oral sucker; *A. emydis* Ejsmont, 1930 differs in having vitellaria confluent anterior to ventral sucker. *A. trituri* Grabda, 1959 being the first species of the genus recorded from amphibian differ from the present specimens in having testes posterior to caecal endings; the present specimens differ from *A. turneri* Bray *et al.*, 2006 in which vitellaria start anterior to intestinal bifurcation, ovary round and acetabulum in anterior half of the body and the position of cirrus pouch from *A. mehrai* Tiwari, 1958 which have equal size of oral sucker and acetabulum; from *A. giganticum* Tiwari, 1958 which have more deeply lobed testis at the anterior face. *A. elongatum* Mehra, 1931 which has anterior testis with irregular margins and posterior has entire margin, cirrus sac reaching ovary; from *A. loossii*

Mehra, 1931 which have lobed testes, cuticle devoid of spine; ovary reaches up till the cirrus sac; in *A.nathi* Gupta, 1954 the oral sucker is larger than the ventral sucker; *A.maithaii* Gupta, 1954 which has short esophagus, oral sucker larger than acetabulum; ovary transversely elongated, vitellaria run from level of the acetabulum to middle of posterior testis, from *A. srivastavi* Gupta, 1954 in which vitellaria extend up to hinder margin of anterior testis from *A. thapari* Gupta, 1954 which have testes irregular in shape; from *A.siddiqii* Lal and Prasad, 1980 which lacks esophagus and vitellaria not extending beyond caecal bifurcation; from *A.cirricurvatus* Simha and Chattopadhyaya, 1970 in having ejaculatory duct around posterior and right lateral border of the acetabulum. *E.emydis* Ejsmond, 1930 differ in having vitellaria confluent anterior to ventral sucker. *A. impletum* Hamada, 2002 from *Oreochromis n. niloticus* her specimen had a ventral sucker larger than the oral sucker. *A. impletum*, Abd el kareem and Ibraheem, 2021 from *Tetraodon lineatus* having ventral sucker with sensory papillae and tend to be in four clusters; from *A. reniferum* Zhokhov, A.E *et al.*, 2017 in having smaller body length and the testis are located one above the other.

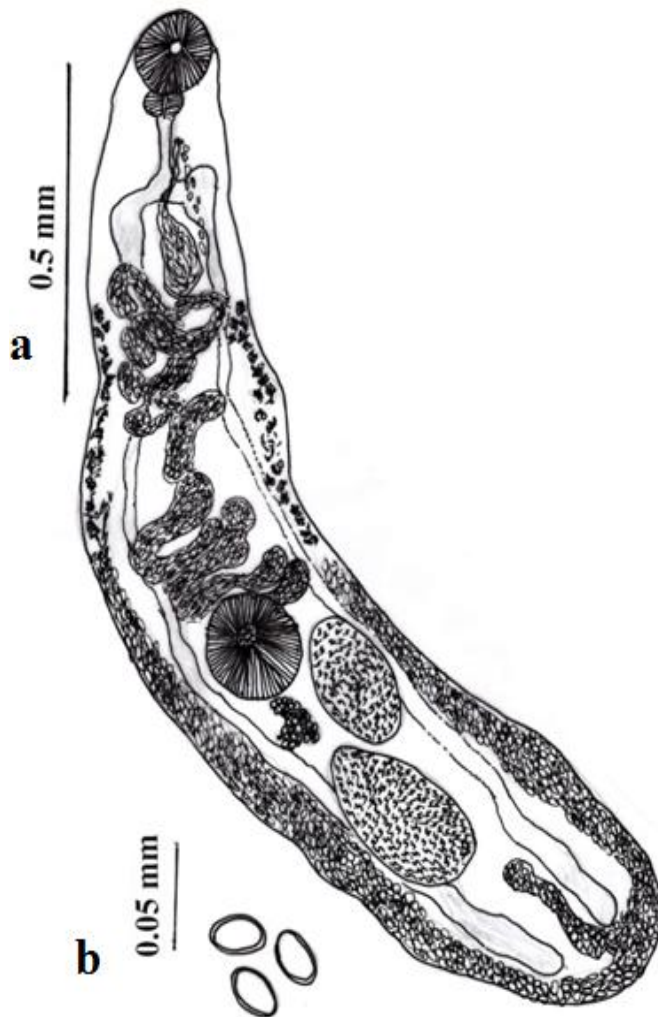


Fig. 1: *Astiotriotrema birmanii* n.sp.
a. Holotype, Entire.
b. Eggs of the same.



Fig. 2: Photomicrograph of *Astiotrema birmanii* n.sp.

The present specimens resemble *A.sudanensis* (Khalil,1959). Bhutta and Khan, 1974 reported from freshwater turtle, in having oral sucker and pharynx smaller than *A.sudanensis*. In present specimens the testes are balloon shaped or oval while in *A.sudanensis* they are rounded. Position of ovary in present specimens is at a distance from cirrus sac. Vitellaria extending upto posterior testis while in present specimens they do not extend beyond acetabulum. The eggs in *A.sudanensis* are smaller than in present specimens.(Table. 1).All measurements are in millimeters as mentioned in materials and methods.

Table 1. Comparison of *A. birmanii* n.sp.with the most similar species of the genus.

Species	<i>A.sudanensis</i> (Khalil, 1959) (Bhutta and Khan, 1974)	<i>A.birmanii</i> n.sp.
Host	Turtle	Frog
Source	Punjab	Sindh
Body length and width	1.24-2.12 by 0.33-0.42	1.65-1.82 by 0.31-0.42
Oral sucker	0.127-0.176 by 0.147-0.196	0.11 -0.14 by 0.12-0.14
Pharynx	0.058-0.081 by 0.068-0.081	0.036-0.047 by 0.040-0.049
Esophagus	0.196-0.303 by ?	0.08-1.12 by 0.012-0.016
Ventral sucker	0.091-0.127 by 0.096-0.119	0.15-0.18 by 0.14-0.17
Ovary	0.088-0.127 by 0.068-0.117	0.060-0.058 by 0.050-0.063
Anterior testis	0.117-0.196 by 0.107-0.166	0.13-0.23 by 0.13-0.16
Posterior testis	0.125-0.225 by 0.107-0.166	0.25-0.27 by 0.15-0.19
Cirrus sac	0.343-0.490	0.12-0.18
Egg	0.015-0.030 by 0.012-0.015	0.046-0.054 by 0.027-0.036

Accordingly, the present specimens are considered new to science and named *A. birmanii* n. sp. in honour of Dr. Nadir Ali Birmani, Dept. of Zoology, University of Sindh, Jamshoro, Sindh, Pakistan for his significant contribution in the field of Helminthology from Sindh.

Conclusion

Based on these characteristics, the present work describes a new species of the genus *Astiotrema* (Looss, 1900) found in frog from Karachi, Pakistan.

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