

AMPHILINA KALRIAI SP.N. (CESTODARIA: AMPHILINIDAE) INFECTING A COMMON EDIBLE FISH *LABEO ROHITA* (HAM.) OF KALRI LAKE, SINDH, PAKISTAN.

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Abstract

A new species *Amphilina kalriai* is described recovered from the small intestine of a common edible, fresh water fish *Labeo rohita* (Ham). caught from Karli Lake, Sindh.

The new species differ from earlier reported species in having ribbon shaped body with greater length, different body shape, size, shape and position and size of male and female gonads, size of eggs and different fish hosts.

Present species is second to be reported from Pakistan.

Introduction

Literature search reveals only eight species of the genus *Amphilina* Wagener, 1858 are mostly reported from freshwater fishes except *A.magna* Southwell,1915 in Coelom of a marine fish *diagramma crassispinum* in Ceylon.

The species reported are:

Amphilina foliacea (Rudolphi, 1819) Wagener (1858) in Acipenseridae in Europe. (Type species) *A. liguloidea* Diesing (1850) from Brazil, *A. neritina* Salensky (1874) from Europe *A. magna* Southwell (1915) from Coast of Ceylon in coelom of a marine fish *Diagramma crassispinum*, *A. paragonopora* Woodland (1923) in body cavity of *Macrones oar* and *M. seenghala* in Allahabad, India, *A. japonica* Goto and Ishii (1936) in the peritoneal cavity of Sturgeon (*Acipenser Mikadoi* Hilgendorf) in Japan, *A. bipunctata* Riser (1948) in Sturgeon in Oregon and *A. tengaria* Rehana & Bilquees (1979) in *Mystus tengara* (Ham.) Bagridae, in Sindh, Pakistan.

Materials and Methods

A total of eighty five fresh water fish *Labeo rohita* were either caught from Kalri Lake, Sindh or bought from nearby fish markets at random intervals.

These were examined in the laboratory for parasitic infections. Besides other helminth parasite infections, one fish was found infected with long ribbon-shaped Cestodarians, blocking the small intestine of the host.

The tape worms were first washed in saline water, later these were flattened by pressing between two slides and tying these slides with white cotton thread by applying adequate pressure and kept for 24 hours in F.A.A. solution (a solution of ethyl alcohol, formalin, acetic acid in a ratio of 92:5:3).

These were later washed with 70% alcohol, stained with Mayer's carmalum, dehydrated in graded alcohol, cleared in clove oil, rinsed with xylene and mounted permanently in Canada balsam. Drawings were prepared with a camera lucida. Measurements are in millimeters. The specimens are deposited in Outreach research activity unit (ORAU) Southern Zone Agricultural Research Center (SARC), Karachi, Sindh.

***Amphilina kalriai* sp. n.**

Figs. (1-4)

Host	<i>Labeo rohita</i>
Site of Infection	Small intestine
Locality	Kalri Lake, Sindh
No. of hosts examined/infected	85/01
No. of specimens recovered	08
Prevalence	1.17%
Intensity	8 worms from one fish

Body ribbon – shaped, body length ranges from 28 – 110 (69.25) anterior end rounded, 1 – 1.02 (1.01) narrower than the posterior end. There is no scolex, the anterior region tapers slightly to a rounded end with weak muscular depression. The posterior extremity of the body is flattened, broader 1.02 – 1.04 (1.03) and bears

two separate openings of the ductus ejaculatorius, terminal excretory duct and vagina. Maximum width is attained at the region of seminal vesicle 1.07 – 1.08 (1.07) in the posterior region. In color the parasite vary from a creamy white to greyish white when alive or when freshly collected.

The anterior end of the worm posses weak muscular depression to which woodland (1923) has designated as proboscis with enormously attached proboscis muscles (or bundle of gland – ducts). The uterine terminal duct proceeds anteriorly to open to the exterior by the side of the weak muscular depression or the so called proboscis. The anterior portion of the uterine duct appears to contain pale yellow, largely oval to rounded ova with smooth walls. Vagina arises from the receptaculum seminis at the base of the ovary, making light – loops, opens at the posterior extremity dorsal to the ductus – ejaculatorius. Vitellaria follicular, the vitelline follicles are arranged in clusters in form of a strip or band on the lateral body margins, starting some distance above the ovary and terminate some distance behind the proboscis. The vitelline bands are external to the testes and the uterine limbs.

Testes are numerous, scattered throughout the body length except at the anterior and posterior regions 0.05 – 0.06 (0.055) by 0.04 – 0.45 (0.042) in size. The sperm duct is the continuation of vasa – efferentia which later transforms into the vas deferens; the terminal portion of the sperm duct is termed as ductus ejaculatorius which opens to the exterior through a median opening in the posterior body extremity.

Ovary is an elongated, tubular structure 3.0 – 3.3 (3.15) by 0.26 – 0.28 (0.27) at the anterior tip and 0.3 – 0.3 (0.30) at the basal region. Uterus arises from the basal or posterior part of the ovary. Receptaculum seminis continues to form convoluted vaginal tube approximately 5.05 – 5.08 (5.06) long and 0.1 – 0.11 (0.10) wide, opens to the exterior, being dorsal to the ductus ejaculatorius.

The uterine limbs form the typical ‘N’ – shape.

Eggs are oval to rounded 0.08 – 0.09 (0.085) by 0.065 – 0.065 (0.065) in size.

Discussion

Available literature reveal very few species of the genus *Amphilina* Wagener (1858) these are:

Amphilina foliacea (Rudolphi, 1819) Wagener (1858) in Acipenseridae in Europe. (Type species)

A. liguloidea Diesing (1850) in *Arapaina gigas* from Brazil.

A. neritina Salensky (1874) in *Acipenser sp.* from Europe.

A. magna Southwell (1915) from Coast of Ceylon in coelom of a marine fish *Diagramma crassispinum*.

A. paragonopora Woodland (1923) in body cavity of *Macrones oar* and *M. seenghala* in Allahabad, India.

A. japonica Goto and Ishii (1936) in the peritoneal cavity of Sturgeon (*Acipenser mikadoi* Hilgendorf) in Japan.

A. bipunctata Riser (1948) in sturgeon in Oregon.

A. tengaria Rehana & Bilquees (1979) in *Mystus tengara* (Ham.) Bagridae, in Sindh, Pakistan.

Woodland 1923 redefined genus *Amphilina* and cited main distinctions between *A. neritina*; *A. foliacea*; *A. liguloidea*; *A. magna*; and *A. paragonopora*. (Table 1).

According to Woodland, 1923: in *A. paragonopora* the body is ribbon-shaped, the posterior end is not pointed the two surfaces are alike in curvature. Body flat varying in outline form an oval to a narrow ribbon. Anterior end pointed or slightly truncated; posterior end pointed, rounded or emarginated. A small evaginable proboscis is present at the anterior end. And connected with this a large boring muscle, the fibers of which end posteriorly in giant anchor cells situated in the parenchyma. The excretory system consists of two main lateral cannels which open posteriorly by a median single pore. Testes numerous. Ovary and openings of vas deferens and vagina are posterior. Uterus is a long convoluted duct consisting of three limbs (N- shaped), each extending nearly up to the entire length of the body, and opening anteriorly at the base of the proboscis.

The present specimens differ from the above named species of the genus in having long, ribbon – shaped body 28 – 110 (69) long, with anterior rounded end, narrower than the broadly flattened posterior end, it however apparently resemble *A. paragonopora* and *A. magna* in having ribbon shaped body which have greater body length, but differ from it in having a rounded anterior end and shape and size of the eggs, and different fish hosts & localities.

The present specimens also differ from *A. tengaria* reported and described from *Mystus tengara* of Kalri Lake, Sindh, Pakistan, in having a greater body length and width, in having a weakly developed muscular proboscis depression anteriorly, position and size of the ovary and length of the male and female terminal ducts and a different fish host.

The present specimens also differ from the type species *A. foliacea* (Rudolphi, 1819) Wagener (1858) and *A. liguloidea* Diesing (1850); *A. neritina* Salensky (1874); *A. japonica* Goto & Ishii (1936) and *A. bipunctata* Riser (1948) in having different body shape and size, shape, position and size of the male and female gonads, sizes of eggs and also the hosts and localities (Table I).

The present specimens are therefore proposed to be new as *A. kalriai* sp. n. This is a second report of the species of the genus *Amphilina* from Pakistan. The species name refers to the locality of the host. *Labeo rohita* is a new host record.

Additionally, Davydov and Kuperman (1993) studied the ultra-structure and histochemistry of the tegument and penetration glands of adult *Amphilina foliacea* from the body cavity and the tissues of the internal organs of *Acipenser ruthenus* and *A. stellatus*. New data was obtained on the localization in the tissues, development and in encapsulation of the adult *A. foliacea* mostly in the liver of *A. ruthenus*.

Results of the above study provided further information about the phylogeny of Amphilinidea and confirm the view of the close phylogenetic relationship of Amphilinidea and Cestodaria.

Biserova *et al.* (2000) studied nervous system of young and adult *A. foliacea* with immunocytochemical, electron microscopical and spectrofluorometrial methods, and described the general neuro anatomy and obtained new data on the structure and development of the brain.

Poddubnaya and Xylander (2010) described the ultra structure of the ovary of *A. japonica* Goto & Ishii (1936) from the body cavity of the American Sturgeon *Acipenser transmontanus*, Richardson using transmission electron microscopy and states that the characters of the ovary of *A. japonica* are different form all other cestodes.

Figs.1-3 *Amphilina kalriai* sp.n.

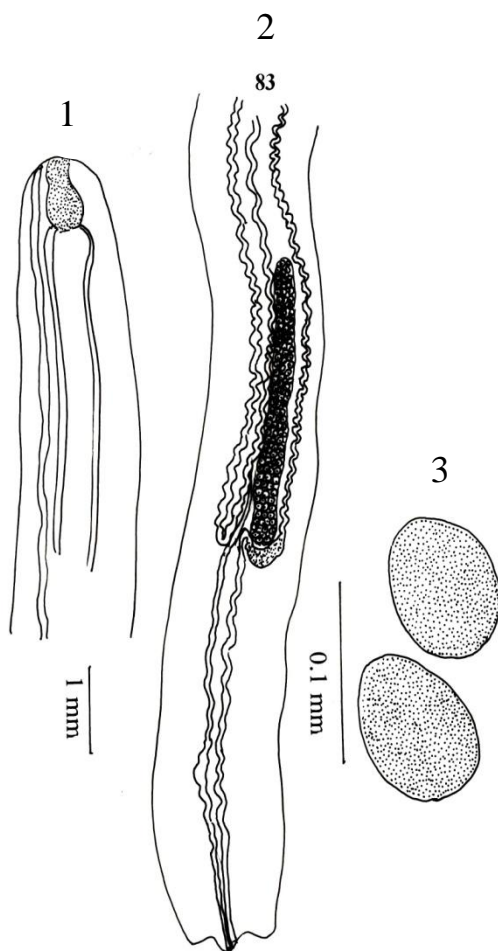


Fig.1 Anterior region of holotype showing muscular depression and uterine opening

Fig.2 Posterior region of the same showing elongated ovary and other reproductive organs

Fig.3 Eggs enlarged.

Fig.4 *Amphilina kalriai* sp.n



Fig.4 *Amphilina kalriai* sp.n Photomicrograph, posterior region showing elongated ovary, uteri, vagina, seminal vesicle and ductus ejaculatrius.

Table 1. Distinctive features in some species of the genus *Amphilina* Wagener, 1858.

Description	<i>A. Nertina</i>	<i>A. foliacea</i>	<i>A. liguloidea</i>	<i>A. magna</i>	<i>A. paragonopora</i>	<i>A. japonica</i>	<i>A. bipunctata</i>	<i>A. tengaria</i>	<i>A. kalriai sp. n.</i>
Colour	Grey-green	Creamy-white	Grey-white	Milky-white	Creamy-yellow to orange	-	-	Greyish white	Greyish-white
Maximum length	18mm	26-60mm	86mm (preserved)	250mm (preserved) 381mm (living)	170mm (preserved) 280mm (living)	44mm (living) 65mm (preserved)	44, 56 & 60mm by 18-24	32.63mm	28-110 (preserved)
Approximate ratio of maximum breadth of body to length	1/2.0	1/1.2-1.30	1/3.5-4.0	1/10.6-12.8	1/20.8-28.3	-	-	1.22-1.90	1/46.6-63.7
Body extremities	Both ends narrow and rounded	Both ends narrow and rounded	Both ends narrow and rounded	Both ends narrow and rounded	Anterior end pointed or rounded, posterior end emarginate	Both ends narrow and rounded	Conically pointed anteriorly, broadly rounded posteriorly	Both ends narrow and rounded	Anterior end rounded posterior end flattened, broader and roughly shaped
Host and localities	<i>Acipenser. sp., Europe</i>	<i>Acipenser. sp., Europe</i>	<i>Arapaina gigas, Brazil</i>	<i>Digrama crassipinum, Coast of Ceylon</i>	<i>Macrons aor et M. Seenghala, rivers of North India</i>	<i>Acipenser/Mikadoi</i>	<i>Sturgeon/Oregon</i>	<i>Mystus tengara, Pakistan</i>	<i>Labeo rohita, Pakistan</i>
Vagina and its aperture	In both species vagina lies posteriorly to left & ductus ejaculatorius and opens on left margin of body about 2mm. away from the median ductus opening		Vagina runs to right of ductus and has dorsal and ventral openings in middle line about 3 mm. from hind extremity. Vagina has anterior diverticulum	Vagina to right of ductus and opens dorsally (?) in middle line 2 mm. from hind extremity.	Vagina to right of ductus and opening at hind extremity just dorsal to ductus opening in middle line.	Vaginal ducts runs straight to posterior end of the body.	Vaginal pore in general body surface, lateral to male papilla	Vagina continues into receptaculum seminis No penial setae	Vagina dorsal to ductus ejaculatorius, open side by side. No penial setae
Penial setae and testes	-	Penial setae said to be scattered	No penial setae, Linear arrangement	No penial setae, Linear arrangement	No penial setae Linear arrangement	In two lateral groups botryoid	350-400cm on each side between vitellaria and uterine coils small compact, with several finger like-lobes	1,194 in number, scattered throughout	Scattered throughout
Ovary	-	Irregular, flower-shape	-	-	Elongated & tubular	Botryoid ovary	Small, compact with several finger-like lobes	Elongated and tubular	Elongated and tubular

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