STUDIES ON THE DIVERSITY AND STATUS OF HELMINTH PARASITES IN ANURAN AMPHIBIA OF NAGALAND (NORTHEAST INDIA)

by

R. IMKONGWAPANG

DEPARTMENT OF ZOOLOGY

SUBMITTED

in

PARTIAL FULFILMENT OF THE REQUIREMENT OF THE DEGREE OF

DOCTOR OF PHILOSOPHY IN ZOOLOGY

of

NAGALAND UNIVERSITY

HQs: LUMAMI

NAGALAND

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DECLARATION

I, R. Imkongwapang, hereby declare that the subject matter of this thesis is the record of work done by me, that the contents of this thesis did not form basis of the award of any previous degree to me or to the best of my knowledge to anybody else, and that the thesis has not been submitted by me for any research degree in any other University/Institute.

This is being submitted to the Nagaland University for the degree of Doctor of Philosophy in Zoology.

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PREFACE

Of more than 4120 species of anuran Amphibia reported under 248 genera with 17 families, about 180 species under 24 genera and 6 families are known to occur in the Indian sub-continent (Chanda, 1994). With about 62% of the species being endemic, the Indian amphibian fauna is quite distinct. The northeastern region of India is a known area of high endemism. With regard to amphibian faunal diversity, as many as 54 species under 6 families and 18 genera have been reported from the country, out of which 21 species are restricted to the region. Dense tropical rain forests, high precipitation, varying altitudinal gradients, abundance of food species and relatively less eco-disturbances have possibly contributed to the richness and diversity of anuran Amphibia in the northeastern region of India (Chanda, 1994).

The diversity of parasitic fauna in Amphibia in the Indian sub-continent is reflected in various surveys carried out from time to time by many workers. Earlier studies made on similar aspects on Amphibia in Meghalaya revealed the occurrence of some helminth parasite species which were restricted to rhacophorid and/or raniid hosts and have not been reported from anywhere else in India. Interesting information on the biology of the polystomatid monogenea, which appear to be an endemic species of the region, was provided by Dutta (1997). Preliminary studies, on similar aspects, in frog of Nagaland revealed a rich parasite spectrum, with a total of 29 species (Tandon *et al.*, 2001); the study also recorded a high intensity and prevalence of plerocercoid larvae (spargana) from 6 of the host species examined, all of which are used in traditional medicine and local cuisine among the natives of Nagaland (Tandon and Imkongwapang, 1999).

In view of the endemism and species richness of both anuran Amphibia and their sustained parasite fauna of Northeast India, occurrence of isolationist helminth infections is expected in this region (Diengdoh, 1989; Diengdoh and Tandon, 1991; Tandon *et al.*, 2001). Therefore, it was proposed to take up this study with an objective of revealing the diversity of helminth infracommunities through exhaustive exploration by furthering the study area and the host range of anuran Amphibia of Nagaland state.

The proposed study therefore aimed:

1. to study the qualitative and quantitative structural groupings and species composition of parasites and influence, if any, of biocoenotic and phonological factors on their occurrence;
2. to study the morphological characterization and identification of helminth species parasitising the anuran host, occurring in varied climatic zones of Nagaland; and
3. to find out the zoonotic potential, if any, amongst the parasitic forms prevalent the amphibian hosts of the region.

INTRODUCTION:

The anuran Amphibia, except in some pacific islands, deserts and snow clad mountains, are distributed all over the world and form a valuable component in the biosphere. The group Anura, consisting of frogs and toads, comprises the largest living order of Amphibia. Of more than 4000 species of anurans reported under 248 genera with 17 families, about 24 genera under 6 familes are known to occur in the Indian sub-continent (Chanda, 1994). In the mountainous zones of Northeast, India this group of vertebrate has acquired an added value in that the amphibians constitute a consumable and highly palatable food item for a section of the local population, as in Korea and several near- and Far- eastern countries (Heussar, 1974). The value of frogs in food and medicine has been known since ages (Cochran, 1964). Among the native population of Nagaland and Meghalaya, like in many Far East countries, frogs are used for treatment of various ailments (Kiyasetuo, 1986). Many frogs such as the *Rana, Amolops, Euphlyctis, Limnonectes, Rhacophorus, Hyla* and *Polypedates* species are taken as food items.

Many medically and economically important diseases of both man and animals are caused by a group of metazoan parasites, which are mainly representatives of 3 Phyla- Platyhelminthes (Monogenea, Trematoda and Cestoda), Nematoda and Acanthocephala. These metazoan parasites also invade the anuran hosts and utilize them as paratenic or final host. Their involvement in several zoonotic infections has been reported, particularly among the populations in Southeast and Far-eastern countries, where eating of frogs and treatment of wounds with raw flesh of frogs is customary (Suziki *et al.*, 1982; Shen *et al.*, 1988; Bodri, 1994; Arora, 1994; Mastura *et al.*, 1995). Valuable surveys of parasites of amphibians both in the tropical and temperate regions of the world have been made by a number of workers like Johnston (1912), Beauchamp (1912), Ozaki (1935), Mazurmovich (1951), Williams (1961), Lees (1962), Fischthal and Kuntz (1965), Combes (1968, 1976), Crusz and Ching (1975), Combes and Channing (1979), Vjotkova(1989, 1990), Lluch *et al.* (1987), Navarro *et al.* (1988), Shen *et al.*(1988), Fernando(1989), Moravec and Sey (1989), Kohn and Santos (1989), Aho (1990), Oladimegi *et al.*(1990), Lees *et al* (1990), Muzzal (1991, a, b), John (1991), Vincente *et al.* (1991), Kim *et al.*(1992), Sey and Eory (1992), wang and Wang (1992), Kearn (1993), McAllister *et al.* (1993), Tinsely (1993), Al-Alousi (1994), Farooq (1994), Farooq and

Khan (1994), Farooq and Katon (1994), Goldberg *et al.*(1994 a, b, 1995), Baston and Richard (1995), Kim *et al.*(1995), Dyer *et al.*(1995) and Burnomo and Bangs (1996).

The platyhelminth parasites of amphibians were reviewed by Prudhoe and Bray (1982). The studies regarding the helminth parasites of amphibian hosts were carried out by many Indian workers from varied localities of Indian sub-continent. Most noted among them are Bhalerao (1926, 1932), Mehra (1926, 1937), Srivastava (1933 a, b, c; 1934), Pandey (1937), Lal (1942, 1944), Kaw (1943, 1950), Bharadwaj (1962), Dwivedi (1965, 1968)[references in Yamaguti, 1971]; Fotedar (1959), Hasnain (1993), Basit (1993), and Kumari and Verma (1994). However, from the Northeastern states of India, the first studies in this direction, carried out by Diengdoh (1989), pertain to a survey of some amphibian hosts occurring in Meghalaya. Of these parasites, polyopisthocotylean monogenean species revealed restricted geographical distribution and high degree of host specificity (Diengdoh and Tandon, 1991). Dutta (1997) studied the cestode and monogenean components of the helminth parasite spectrum of anuran hosts in Meghalaya. Interesting information on the biology of the polystomatid Monogenea, which appear to be an endemic species of the region, was provided by Dutta (1997).

Although surveys and geographical studies on helminths of Amphibia are numerous, specific studies pertaining to helminth communities of these hosts relating to their species-richness, intensity and diversity of helminth parasites are few (Lees, 1962; Combes, 1968; Plasota, 1969; Hollis, 1972; Fransden, 1974; Dronen, 1977; Vjotkova, 1982, 1989, 1990; Goater *et al.*, 1987; Aho, 1990; Muzzal, 1991 a, b). In the context of Indian Sub-continent such studies are scanty (Devi and Rao, 1990; Kumari and Madhavi, 1996). Highly endemic foci of Amphibia- borne zoonoses, sparganosis in particular are known to occur among populations in the neighboring Southeastern and Far-eastern countries (Mastura *et al.*, 1995 a,).

In view of the richness and endemism of anuran amphibian fauna of Northeast India, a preliminary study was carried out pertaining to the spectrum of helminth parasites of Anura in Nagaland state. This study revealed considerably high diversity and prevalence of helminth parasites from all areas of the region, with newer host and locality record for several helminth species. Altogether 32 (thirty two) species of frogs are reported from Nagaland (Ao Meren *et al.*, 2004). However, the mentioned study covered only 13 species of frogs falling under 3 families and 8 genera from 3 districts of Nagaland, and a

total of 29 helminth types were recovered (Tandon *et al.*, 2001); the study also recorded a high intensity and prevalence of plerocercoid larvae (spargana) from 6 of the host species examined, all of which are used in traditional medicine and local cuisine among the natives of Nagaland (Tandon and Imkongwapang, 1999).

In view of the endemism and species richness of both anuran Amphibia and their sustained parasite fauna of North-East India, occurrence of isolationist helminth infections is expected in this region (Diengdoh, 1989; Diengdoh and Tandon, 1991; Tandon *et al.*, 2001). Therefore, the present study was taken up with an objective of revealing the diversity of helminth infracommunities through exhaustive exploration by furthering the study area and the host range of anuran Amphibia of Nagaland state.

MATERIAL AND METHODS

Study area

The study area comprises five major localities in the state of Nagaland, which offer varied climatic conditions and are expected to have an impact on the prevalence and abundance of both amphibian hosts and their parasitic fauna. Nagaland as a whole lies approximately between 26°6'-27°4'N and 93°20'-95°15' (Figs 1,2), with an altitude ranging from 196-3800 m ASL and the average annual rainfall of 250 cm; the average temperature in summer (June-September) ranges from 13°-19.2°C and 19.6°-34°C mean minimum and mean maximum, respectively and in winter (October-February) 1°-6°C and 8.2°C mean minimum and mean maximum, respectively. The climate of Nagaland is "modified tropical monsoon type" with an average annual rainfall of 250 cm (Husain, 1988); every elevation of the region having its own characteristics has distinctive variations of temperature that differ from altitude to altitude (ranging from 195 to 3800 m ASL).

The study covering localities with varied climatic conditions and rich diversity of both the amphibian hosts and their sustained parasites is expected to bring out more information about the helminths and their hosts. Of the localities under survey, Kohima (which is the state capital and district Headquarters) lies at the foot hill of Japfu peak, has high rainfall (120-250 cm annually) with relative humidity varying between 60 and 78% during the warm and monsoon rain months (March-May, June-October) of summer and between 40 and 60% during the peak winter months, and the temperature in summer ranging from 10-30°C. Mokokchung, situated in the north-east part of Kohima, is another district

Headquarter; with somewhat similar rainfall and humidity conditions as in Kohima, the rainy season starts from the month of May and generally lasts till October, sometimes extending even till December; and the temperature is mild to cold through the year, ranging from 8.6-30°C.Tuensang, situated in eastern part of the state borders Mokokchung and Assam in the west, separated by

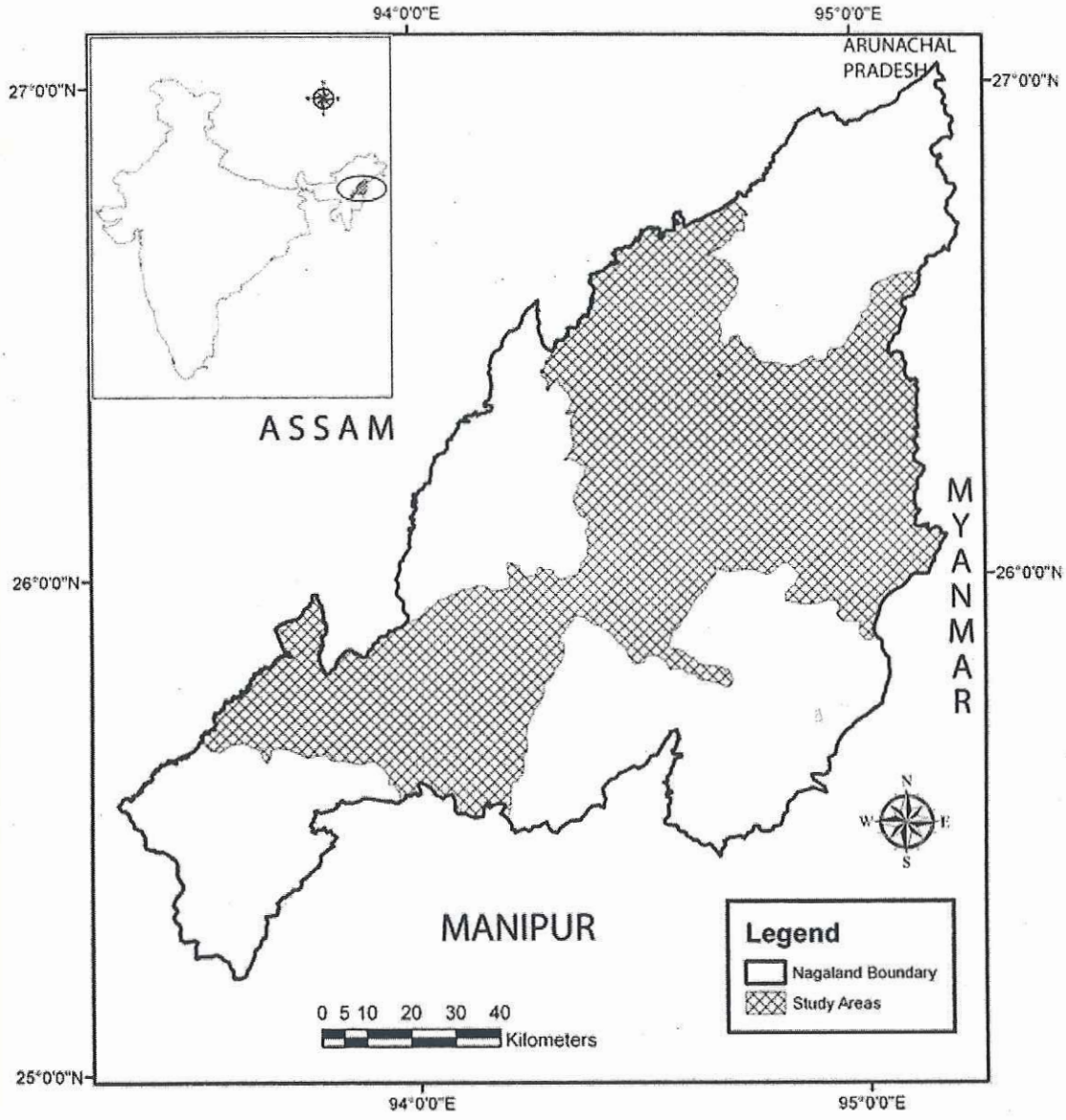


Fig. 1 Map of India and enlarge map of Nagaland with study area shaded

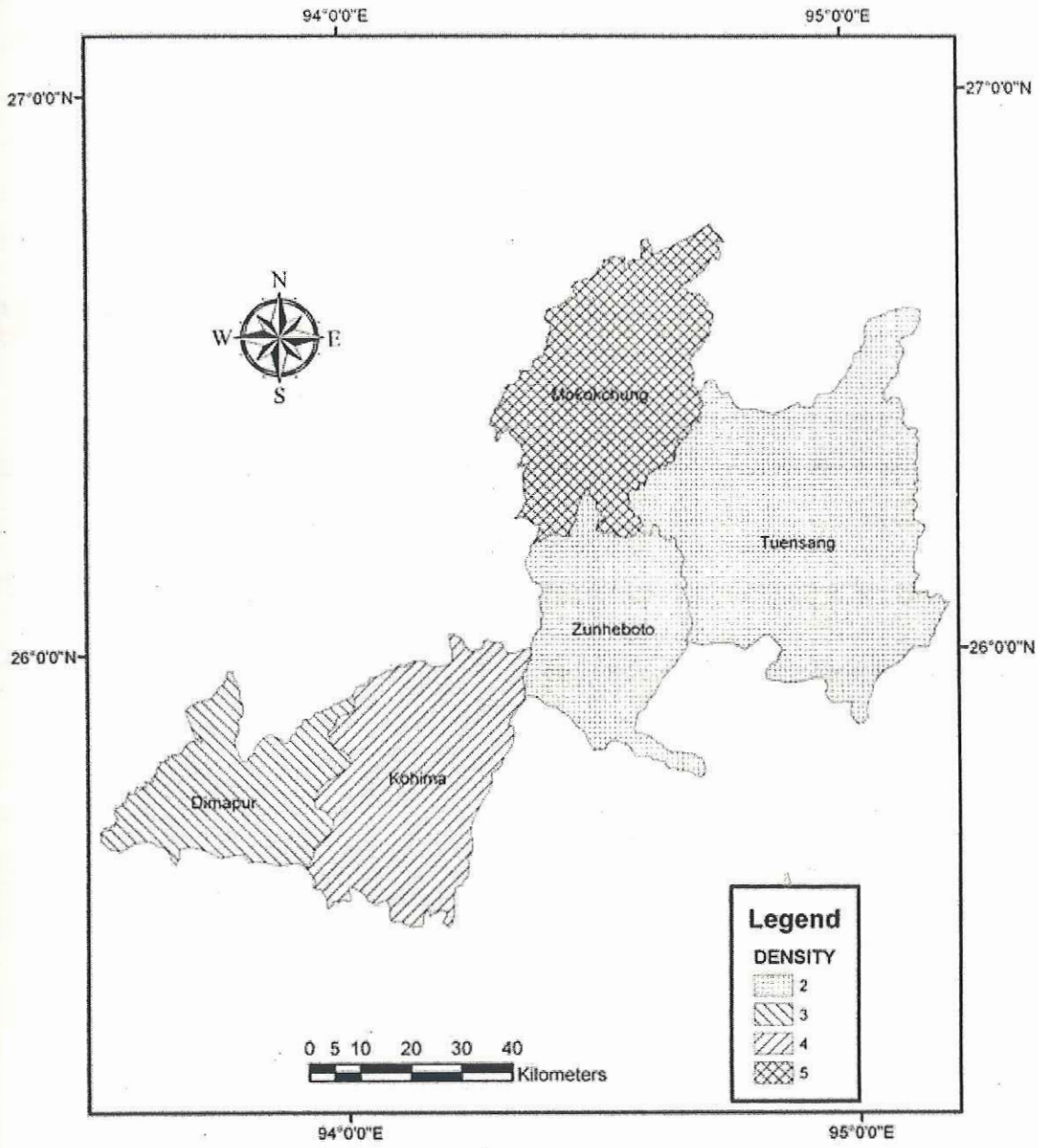


Fig. 2-The districts surveyed and number of frog species collected per district

Mokokchung -	**22**
Kohima -	**10**
Dimapur -	**10**
Zunheboto-	**3**
Tuensang -	**4**

the river Dikhu with the former and Kiliki reserved forest with the latter, respectively; on the eastern part, it borders the Kiphire (District headquarters) and Myanmar. The geo-climatic conditions are somewhat similar with the earlier localities described, but much colder throughout the year. Zunheboto, yet another district is situated in the center of the state, bordered by at least 5 districts on all the sides; the winter is very cold and summer is somewhat similar to Mokokchung. Dimapur a fast growing commercial town, is identical to any low laying tropical plains.

HOST COLLECTED/SURVEYED

The anuran hosts collected and examined for helminth infections during the study include 29 species falling under 15 genera representing 5 families Table 1; the host species, localities surveyed, sites and their habitats are given in Table 2.

Table 1 Host species examined for helminth parasites in Nagaland

FAMILY:	PELOBATIDAE-	*Megophrys glandulosa* (Boulenger, 1890)
		Megophrys wuliangshanensis (Ye and Fei , 1995)
FAMILY:	HYLIDAE-	*Hyla annectans* (Jerdon, 1870)
FAMILY	MICROHYLIDAE-	*Microhyla ornata* (Dumeril and Bibron, 1841)
FAMILY:	RHACOPHORIDAE-	*Rhacophorus maximus* (Frost, 1885)
		Rhacophorus bipunctatus (Ahl, 1927)
		Rhacophorus gongshanensis (Yang et Su, 1984)
		Chirixalus vittatus (Boulenger, 1887)
		Philautus annandalii (Boulenger, 1906)
		Philautus sp.1
		Philautus sp.2
		Philautus sp.3
		Polypedates leucomystax (Graventhorst, 1829)
		P. tarajensis

Theloderma asperum (Boulenger, 1886)

Theloderma sp.

FAMILY: RANIDAE-

Rana khare (Kiyasetuo and Khare, 1986)

Rana danielii (Pillai and Chanda, 1977)

Rana levida (Blyth, 1855)

Rana sp.1

Rana sp.2

Rana sp.3

Amolops marmoratus (Blyth, 1855)

Euphlyctis cyanophlyctis (Schneider, 1799) Dubois, 1922

Limnonectes limnocharis (Graventhorst, 1829)

Limnonectes sp.

Haplobatrachus tigerinus (Daudin, 1803)

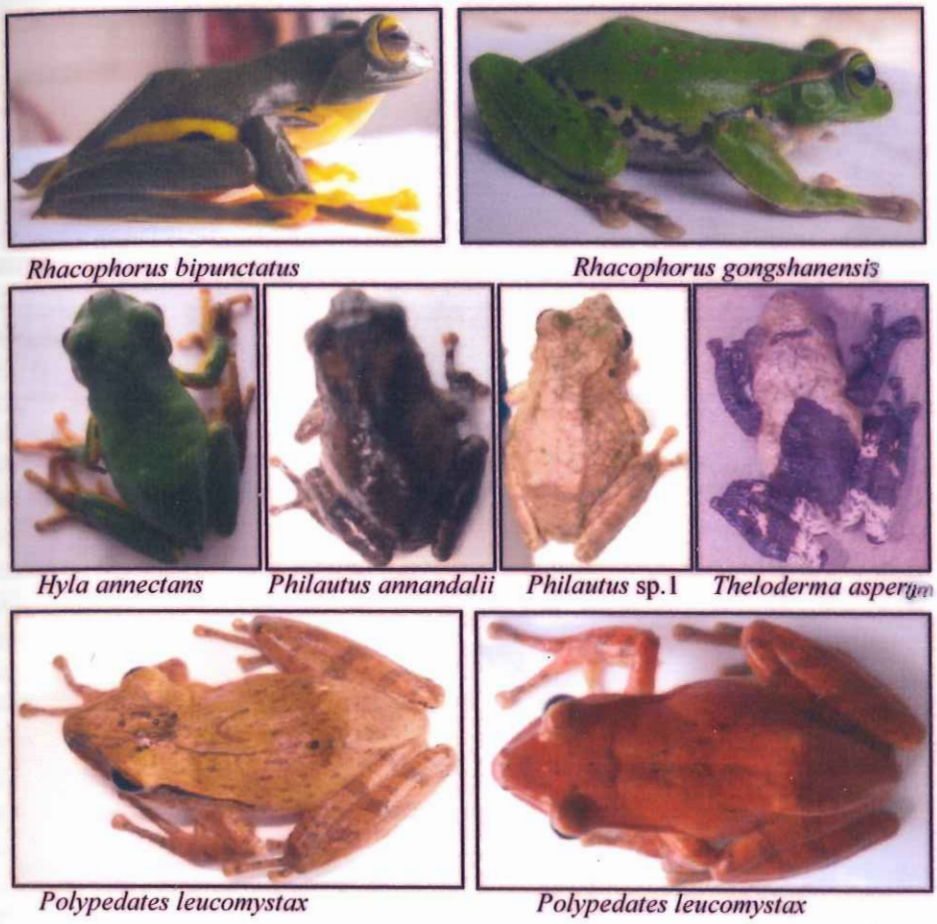
Haplobatrachus crassus (Jerdon, 1853)

Paa mokokchungensis

Since a large number of anuran frog species are edible, they are collected from nature and sold by natives of Nagaland, from early part of March till November; therefore, some were bought from the market and many were caught by making frequent field excursions to different spots of the five localities, throughout the season. Many, but not all, of the earlier surveyed sites and host species of the mentioned study were revisited and reexamined in the present study. Collections were carried out preferably at nightfall either with a net or bare hand by following the croaking sounds of the males or by locating the nest constructed by the female (Cochran, 1961) or by following the native ways of catching frogs.

Those frogs found inhabiting the ravine, cliff, stone, land (forest floor and paddy field) etc., near water bodies were categorized as the **terrestrial-habit species,** and those found in trees, tree branches, leaves, shrubs and bushes, the **arboreal-habit species**.

Plate 1: Some arboreal hosts examined in the study (not to scale)



Rhacophorus bipunctatus *Rhacophorus gongshanensis*

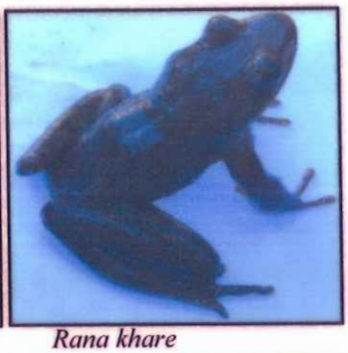
Hyla annectans *Philautus annandalii* *Philautus* sp.1 *Theloderma asperum*

Polypedates leucomystax *Polypedates leucomystax*

Yet again, the frogs were observed exhibiting different mode of adaptation within the habitat itself, in that the terrestrial habit species showed great variation in size, agility and feeding habit. Of this habit species, *Euphlyctis cyanophlyctis* an aquatic species, was the most interesting anuran host collected and examined in the study; they showed the widest range of adaptation and were found to inhabit ponds, lakes, terrace fields, both small and big streams, pot holes etc., spending more time in the water, on banks and in bushes near a water body throughout the seasons, but capable of adaptation to both lentic and lotic systems; they were also very agile in land and in water as was the case with *Rana khare* and *Rana livida* that were very agile and expert swimmers.

Plate: 2-Some of the terrestrial hosts examined in the study (not to scale)

Rana livida



Rana khare

Haplobatrachus tigerinus

Megophrys wuliangshanensis

Microhyla ornata

Megpphrys glandulosus

Rana sp.2

Rana sp.3

Amolops marmoratus also exhibited a wide range in habitat adaptation and was collected from many sites, ranging from large and fast flowing water bodies to a still water bodies near a stream, many a times in the dry land in thick woody forest. The size also varied greatly among the anurans studied in the study; *Haplobatrachus tigerinus* was the largest, weighing150-300 g and the smallest/ tiniest, weighing only 5-9 g were *Microhyla ornata* and the *Philautus* species. The arboreal-habit species were observed to be more sluggish and restricted to the trees, except in the breeding season where some of them such as, *Rhacophorus maximus, R. gongshanensis, Polypedates leucomystax, Megacephalus taraensis, Hyla annectans* and *Philautus* sp.1; were caught/ collected from land (damp and shady area) mostly caught while perched in small branches and leaves or sitting in the banks near a water body. *R. bipunctatus, P. annandalii* and *Philautus* sp.3 were observed to be strictly arboreal in habit, inhabiting the low laying bushes and shrubs, as these species were found mating within their habitat itself and never collected from dry land throughout the study period. The host species-wise sites, sites and localities where collections were made are presented in Table 2.

The anuran hosts were identified following Chanda (1994), Dutta (1997) and Ao Meren *et al.,* (2004). Immediately after the catch the frogs were put in a container and prepared for autopsy. The maximum number of anuran species caught per field trip from one spot was a maximum of 3 species of the terrestrial group and 4 species of the arboreal group, i.e., when caught representing both the groups, the catch never exceeded 7 species from a site on a particular night.

Table 2 Localities and collection sites of the anuran host in Nagaland

Family	Host species	Localities and Collection sites	No. of host collected	Host's habitat	Altitude
Pelobatidae	*Megophrys glandulosa*	**Mokokchung** - Yisemyong and Longkong areas	12	Nanning stream	1200-1450 m ASL
		Tuensang - Chare	2	Dihku tributary	1000 m ASL
	Megophrys wuliangshanensis	**Mokokchung** - Yisemyong and Meink- ong reserved forest			1000-1450 mASL
		Kohima- Jotsoma Sc. College	26	Angetyongpang stream	1450-1500 m ASL
			2	A river bank	1600 m ASL
Hylidae	*Hyla annectans*	**Kohima** -Jotsoma Sc. College, upper Agri. Colony	106	Inundated open ground and a small marshy lakes	1530-1600 m ASL
Microhylidae	*Microhyla ornata*	**Mokokchung** - Mangmetong village	4	Inundated open ground.	1200 m ASL
		Dimapur - Residency colony	6		195 m ASL
Rhacophoridae	*Rhacophorus maximus*	**Mokokchung**-Power house colony, Dilong ward, Mangmetong, Yisemyong	83	Lakes, fishery ponds,river banks, shrubs and trees -,,-	1200-1400 m ASL
	Rhacophorus bipunctatus	**Kohima**- Jotsoma Sc. College area	5	A marshy lake	1600 m ASL
		Zunheboto- Lumami area	6	A marshy lake	900-1100 m ASL
		Mokokchung- Meinkong reserved forest	5	In trees and shrubs near water bodies	1500 m ASL
		Tuensang- Noklak	1	fishery pond	1100 m ASL

Contd.

Family	Host species	Localities and Collection sites	No. of host collected	Host's habitat	Altitude
		Kohima -Jotsoma Sc. College area and upper Agri. Colony	10	In trees and shrubs near Water bodies	1530-1600 m ASL
Rhacophoridae	*Rhacophorus gongshanensis*	**Kohima** - upper agri. Colony	13	Near water bodies on a stone.	1530 m ASL
	Chirixalus vittatus	**Mokokchung** - Yisemyong	15	Bamboo leaves, trees and shrubs near water bodies	1200 m ASL
	Philautus annandalii	**Mokokchung** - Yisemyong,Dilong and power house colony	74	Small short trees and shrubs	1200-1400 m ASL
	Philautus Sp.3	**Mokokchung** - Yisemyong	13	,,	1200 m ASL
	Philautus sp.1	**Mokokchung** - Longkhum village	23	A lake near a thick forest	1560 mASL
	Philautus sp.2	**Mokokhung** - Longkhum village	6	,,	do
	Theloderma asperum	**Mokokchung** - Yisemyong	3	On tree leaves and shrubs	1200 m ASL
	Theloderma sp.	**Mokokchung** - Meingkong reserved forest	4	Tree holes and bamboo stumps where water accumulates	1500 m ASL
	Polypedates leucomystax	**Mokokchung** - Dilong ward,Power house colony and yisemyong	113	Lakes,Fishery ponds and often in river banks,shrubs and trees	1200-1400 m ASL
		Kohima- upper and lower agri. Colony, Jotsoma, SC. College	36	Lakes,Fishery ponds and water bodies	1530-1600 m ASL
		Zunheboto- Lumami area	9	Lake and fishery pond	900-1100 m ASL

Contd.

Family	Host species	Localities and Collection sites	No. of host collected	Host's habitat	Altitude
Rhacophoridae	*P. taraiensis*	**Dimapur** - Island colony and Residency colony	25	Fishery ponds	195 m ASL
		Mokokchung - Yisemyong and Mangmetong	64	Fishery ponds and marshy lakes	1200 m ASL
		Dimapur - Residency colony	3	On trees near water body	195 m ASL
		Tuensang- Chare	2	,,	1250 m ASL
Ranidae	*Rana khare*	**Mokokchung** - Longjang and Aolichen	13	Fishery ponds and tsuten river	1000-1300 mASL
	Rana levida	**Mokokhung**- Longkong,Changtongya, Longchem and Alichen	11	River	1000-1200 m ASL
	Rana sp.3	**Mokokchung** - do	11	River	do
	Rana danielii	**Mokokchung** - Longsa area	12	Dikhu river	1100 m ASL
	Rana sp.1	**Dimapur** - Island colony	43	Fishery pond	195 m ASL
	Rana sp.2	**Dimapur** - Rangapahar	1	Fishery pond	195 m ASL
	Paa mokokchungensis	**Kohima** - Jafu mountain area	1	Tsunaru River	1500-1650 m ASL
	Amolops marmoratus	**Mokokchung**- Yisemyong, Mangmetong, Longkong Milak river tributaries in and around meinkong reserve forest.	151		1250-1500 m ASL
		Tuensang – Chare	3	In Road side near a water body	1000 m AS

Contd.

FAMILY	Host species	Localities and Collection sites	No. of host collected	Host's habitat	Altitude
Ranidae	*Euphlyctis cyanophlyctis*	**Mokokchung-** Power house colony,Mangmetong, Longchem and Yisemyong	118	Fishery ponds,innundated open ground, potholes, etc.	1000-1340 m ASL
	Limnonectes limnocharis	**Kohima-** Agri. Colony, Jotsoma Sc. college	88	,,	1530-1600 m ASL
		Dimapur- Island colony,Rangapahar and neiuland areas	128	,,	195-170 m ASL
		Zunheboto- Lumami area	5	,,	900-1100 m ASL
		Mokokchung- Power house colony,Mangmetong, Longchem and Yisemyong	84	,,	1000-1400 m ASL
		Kohima - Upper Agri. Colony and Jotsoma Sc. college areas	30	,,	1530-1600 m ASL
	Limnocharis sp.	**Dimapur -** Island colony and Residency colony	12	,,	195 m ASL
		Mokokchung - Mangmetong	4	,,	1200 m ASL
		Dimapur - Residency colony	3	,,	195 m ASL
	Haplobatrachus tigerinus *H. crassus*	**Dimapur -** Island colony, Niuland area and Jalukhi	49	Lakes and Fishery ponds	195-900 m ASL
		Dimapur - Niuland area and Jalukhi	27	,,	195-900 m ASL

METHODOLOGY:

Under confinement the anurans do not feed and they tend to get rid of their worm burden (Smyth and Smyth, 1980). Therefore, immediately after the catch the frogs were autopsied and their various organs examined. The recovered parasites on recovery were processed following different techniques for each group:

(i) Monogenea and Trematodes:

After washing in 0.7% saline solution, the live specimens recovered were narcotized with few drops of 70% ethyl alcohol. They were gently flattened between two slides or a slide and a cover glass, depending on the thickness of the specimen, and fixed overnight in70% ethyl alcohol. Whole worm mount were prepared by staining them with Borax carmine or Meyer's carmalum, followed by dehydration through usual dehydration media, clearing in Methyl benzoate and mounting in Canada balsam.

(ii) Cestodes

On recovery, the cestodes were first stretched in warm water, dipping and running along the side wall of the beaker. They were then flattened between a slide and a cover glass and fixed overnight in 70% ethyl alcohol. Further processing for whole mount preparations was done in the same manner as for the trematodes.

(iii) Nematodes:

Nematodes were stretched in hot 70% ethyl alcohol and then transferred to cold 70% ethyl alcohol. For permanent whole mount, the specimens were placed in 50% glycerine which was gradually replaced by 80% and finally pure glycerine in a desiccator's till they cleared and then double mounted in Kaiser's glycerol jelly (50mlwater+80g gelatin+50ml glycerol+0.1g phenol). Some worms were also cleared in lactophenol and their temporary mounts made.

(iv) Acanthocephala:

They were allowed to relax in hot saline and narcotized with the addition of a few drops of 70% ethyl alcohol. Further, usual procedure of flattening and permanent mounting was followed.

Microscopy:

Light microscopy:

All the prepared permanent slides were observed and studied under Wild M5APO stereo microscope, vision analyzer and Leitz Ortholux-2 research microscope. Measurements were taken with the help of ocular and stage micrometers and, unless and otherwise stated, are in mm and based to the extent possible, on 5-10 specimens in each case.

$$\text{STANDARD DEVIATION} = \sqrt{\frac{\sum X^2 - N\bar{x}^2}{N-1}}$$

X= sum of measurements

N= number of observations

Scanning electron microscopy (SEM):

Live specimens recovered were washed thoroughly in 0.7% saline solution and were fixed in 4% cold neutral buffer formalin for 12-18 hrs. Following fixation, the specimens were washed in phosphate buffer and dehydrated with ascending grades of alcohol and ethanol-amyl acetate mixture to pure amyl acetate. After their final treatment in dry amyl acetate, the specimens were critical-point dried using liquid carbon dioxide. In lieu of critical-point drying the specimens after washing in phosphate buffer were dehydrated in ascending grades of acetone and treated with tetramethyl silane [TMS-$(CCH3)_4Si$, boiling point 26.3°C, surface tension 10.3 dynes/cm at 20°C] for 10 minutes and dried off TMS at 25° C following the method of Dey *et al.* (1989). The dried samples were metal coated with gold in a fine coat ion sputter JFC-1100(JEOL). Observations were then made with the scanning electron microscope JSM 35CF (JEOL) under an electron accelerating voltage ranging between 10 and 20 Kv.

For the parasites which were encountered for the first time from North-East India, a brief description precedes the remarks; those earlier reported from the region have not been described herein.

For studying the helminth communities of the various host species, the following parameters were used to analyze the data following Margolis *et al.* (1982) and Muzzal (1991 a, b).

i. Prevalence – Number of individuals of a host species infected with a particular parasite species divided by the number of host individuals examined x 100 (= percentage of infected hosts of each species)

ii. Intensity- number of individuals of a particular parasite species in each infected host in a sample.

iii. Mean intensity- mean number of individuals of a particular parasite species divided by the number of host species examined in a sample.

iv. Species richness- number of helminth species per host species examined.

Identification of the parasites is based on Yamaguti (1958, 1959, 1961, 1963a, b, 1971), Prudhoe and Bray (1982), Agarwal and Pandey (2007) and CIH keys (nos. I-X) of nematode parasites of vertebrates.

Table 3 Helminth parasites recovered from anuran host in the study

HELMINTHS

MONOGENEA

Polystoma indicum Diengdoh *et* Tandon, 1991

Polystoma sp.

Polystoma kohimaensis n. sp.

Polystoma hylai n. sp

Neoriojatrema mokokchungensis n. g., n. sp.

TREMATODA: DIGENEA

Gorgoderina ellipticum Dwivedi, 1968

Gorgoderina sp.

Diplodiscus amphichrus Tubangui, 1933

Diplodiscus mehrai Pande, 1937

Halipegus mehransis Srivastava, 1933

Mesocoelium monas (Rudolphi, 1819) Freitas, 1958

Opisthioparaochis indica Tandon, Imkongwapang et Prasad, 2005

Opisthioparaochis yunnanse Li, 1996

Mehraochis ranarum Srivastava, 1933

Prosotocus infrequentum Srivastava, 1933

Ganeo tigrinum Mehra *et* Negi, 1928

Pleurogenoides gastroporus (Lühe, 1901)Travassos, 1921

Haematoloechus almorai (Pande, 1937) Freitas et Lent, 1939

Batrachotrema nagalandensis Tandon, Imkongwapang et Prasad, 2005

Cathaemasia sp.- Metacercaria

Proalarioides sp.- Metacercaria

CESTODA

Baerietta baeri Hsü, 1935

Nematotaenioides sp.

Proteocephalus tigrinus Gupta and Arora, 1979

Plerocercoid larva

NEMATODA

Aplectana gubernaculum Gupta, 1960

Aplectana sp.2

Oxysomatium macintoshii (Stewart, 1914) Karve, 1927

Oxysomatium sp.

Cosmocercella sp.

Paracosmocerca mucronata Kung et Wu, 1945

Pharyngodon sp.

Rhabdias ranae Walton, 1929

Oswaldocruzia goezei Skrjabin *et* Karokhin, 1952

Ophidascaris sp.

Amplicaecum sp.

Icosiella sp.1

Icosiella sp.2

ACANTHOCEPHALA

Acanthocephalus bufonis (Shipley, 1903) Southwell *et* Macfie, 1925

THE PARASITE SPECTRUM:

Taxonomic studies

MONOGENEA

Superfamily Polystomatoidea Price, 1936

Family Polystomatidae Gamble, 1896

Subfamily Polystomatinae Gamble, 1896

Genus ***Polystoma*** Zeder, 1800

Polystoma indicum Diengdoh *et* Tandon, 1991

(Figs.3-17)

Altogether 115 specimens of ***Polystoma indicum*** Diengdoh and Tandon, 1991 were recovered in the present study; the number of parasites ranged from 1-6 per infected host.

Description (based on 5 matured specimens)

Body elongated or foliate in shape, somewhat tapering towards anterior end from the broadest section of the body, sharply ending in sub-terminal oral sucker; posteriorly terminating bluntly in opisthaptoral region; Surface devoid of spines. Opisthaptor much smaller to body width, three pairs of well developed suckers and a pair of smooth, rounded and entire based macrohooks or hamuli with sharply pointed recurved tips; microhooks between the hamuli were not traceable in the specimen recovered from *R. maximus*, except the presence of a broad based microhook singly in each clamps. The specimen from the host, *T. asper* had 7 between hamuli and single slender shaped microhook in each of sucker. Pharynx prominent, muscular, leading to bifurcate intestine. Intestinal caeca indented at irregular interval on lateral aspects, sending out diverticula directed toward the median line and uniting at irregular interval. Testes numerous, forming insurmountable mass in pharyngeal and post-ovarian inter-caecal zone; vas deferens arising in region of junction of female genital ducts, running anteriad slightly to side of median line; cirrus-sac small, cirrus tipped with genital crown of 11 hooklets, opening in front of female genital pore into common genital sinus latter opening to exterior close behind intestinal bifurcation. Ovary retort shaped, uterus with numerous eggs

The surface fine topography of the fluke revealed numerous tegumental papillae all over the body, with spongy appearance in higher magnification. The outer rim of the oral sucker also revealed domed shaped papillae poked with holes, with many radial folding and infolding in the rim followed by gobble stone-like arrangement of papillae.

Morphometric measurements:

Body: 3.1-3.81x 1.39-1.81 mm.

Egg size: 0.19-0.21x .12-14 mm.

Host: *Rhacophorus maximus*, (=*R. nigropalmatus*), *R. bipunctstus* (=*R. reinwardtii*), *Theloderma asperum*, *Polypedates leucomystax*, *P. taraiensis*

Location: Urinary bladder, ureter, body cavity

Locality: Kohima, Mokokchung, Zunheboto

Remarks:

The present report constitutes 3 new host records from Nagaland, i.e., *Rhacophorus bipunctatus, Theloderma asperum* and *P. taraiensis* for ***P. indicum*** and Zunheboto, as a new locality record. As the first representative of the genus ***Polystoma*** from the Indian sub-continent, ***P. indicum*** was originally described from *Rhacophorus maximus* of Meghalaya, by Diengdoh and Tandon (1991). *R. bipunctatus, P. leucomystax* and *Hyla annectans* are other host species recorded for this species (Dutta, 1995; Tandon *et al.*, 2001).

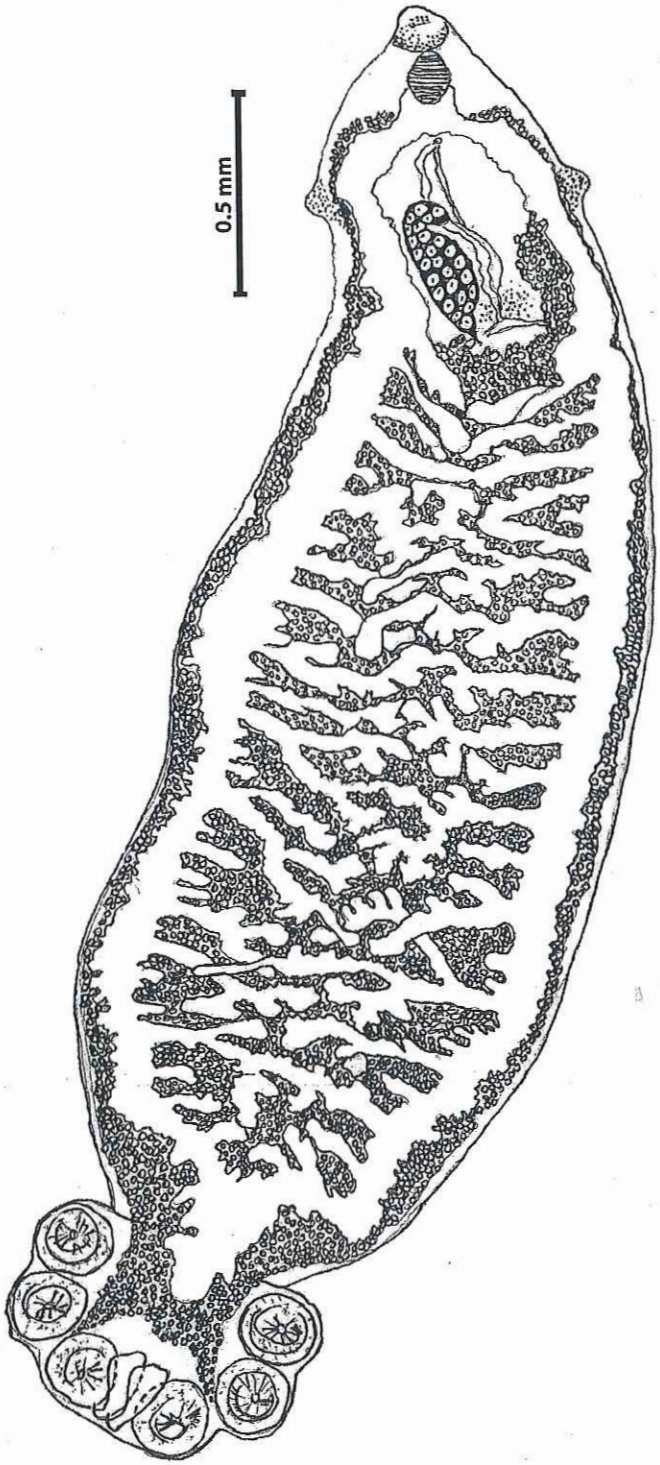


Fig. 3 ***Polystoma indicum*** whole mount, dorsal view.

Figs. 4- 8 *Polystoma indicum* (Host: *R. maximus*)- Photomicrographs.

4. Anterior end of the fluke, ventral view

5. Posterior view of the same fluke, showing the anastomosed and united caeca

6. Magnified view of the hamuli with entire base

7. A microhook between the hamuli

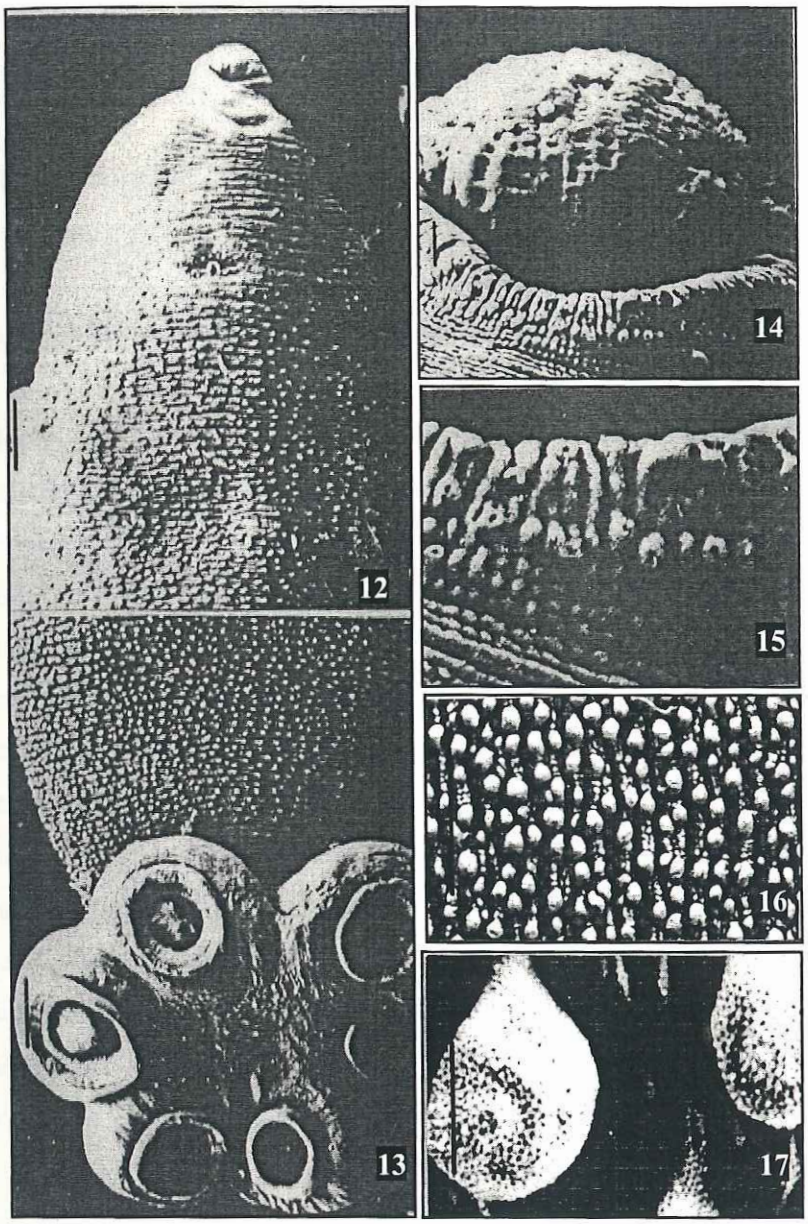
8. Magnified view of the genital crown

Figs. 9-11 *Polystoma indicum* (Host: *T. asper*)

9. A magnified view of the genital crown

10. Microhooklets between the hamuli

11. Microhook in the clamp



Figs. 12-17 ***Polystoma indicum*** (Host: *R. maximus*)- Scanning electron micrographs.

12. Anterior end in ventral view showing the scarcely distributed papillae all over the body surface and genital opening in mid anterior region (Scale bar= 100µm)

13. View of the posterior half, showing the populated papillae opisthaptor and the retracted hamuli (arrow) (Scale bar= 100µm)

14. A magnified view of the oral sucker, showing the domed papillae near the rim indicated by arrow (Scale bar= 10µm)

15 A close view of the oral rim, showing the densely pitted papillae (Scale bar= 10µm)

16. A magnified view, showing dense aggregation of papillae (Scale bar= 100µm)

17. Papillae in higher resolution, showing a spongy surface (Scale bar= 10µm)

Polystoma sp.

(Figs.18-24)

10 specimens of this monogenean under the genus ***Polystoma*** were recovered from 2 of the 23 anuran host examined.

Descriptions (based on measurements and observations of 5 mature specimens):
Body elongated or foliate in shape, somewhat tapering towards anterior from the broadest mid-section of body and sharply ending in sub-terminal oral sucker; toward the posterior, terminating bluntly in opisthaptoral region, with a constriction separating it with the rest of the body; surface devoid of any spine, opisthaptor somewhat same size with broadest section of the body in width, with three pairs of well developed suckers and a pair of macrohooks hamuli, base entire, broad and not smooth but with cracks, and sharply pointed recurved tips; microhooks 5 in number between the hamuli. Pharynx prominent, muscular, leading to bifurcate intestine. Intestinal caeca indented at irregular interval on lateral aspects, sending out diverticula directed toward the median line and uniting to give mesh-like network. Testes numerous, forming insurmountable mass in pharyngeal and post-ovarian inter-caecal zone; vas deferens arising in region of junction of female genital ducts, running anteriad slightly to side of median line; cirrus-sac small, cirrus tipped with genital crown of 11 hooklets, opening in front of female genital pore into common genital sinus latter opening to exterior close behind intestinal bifurcation. 3 genito-intestinal canals directed towards the median from both the caeca; ovary retort shaped, uterus with one roundish egg.

Morphometric measurements of this form are given in Table 4

Host: *Philautus* sp. 1
Locality: Mokokchung (Longkhum)
Location: Urinary bladder

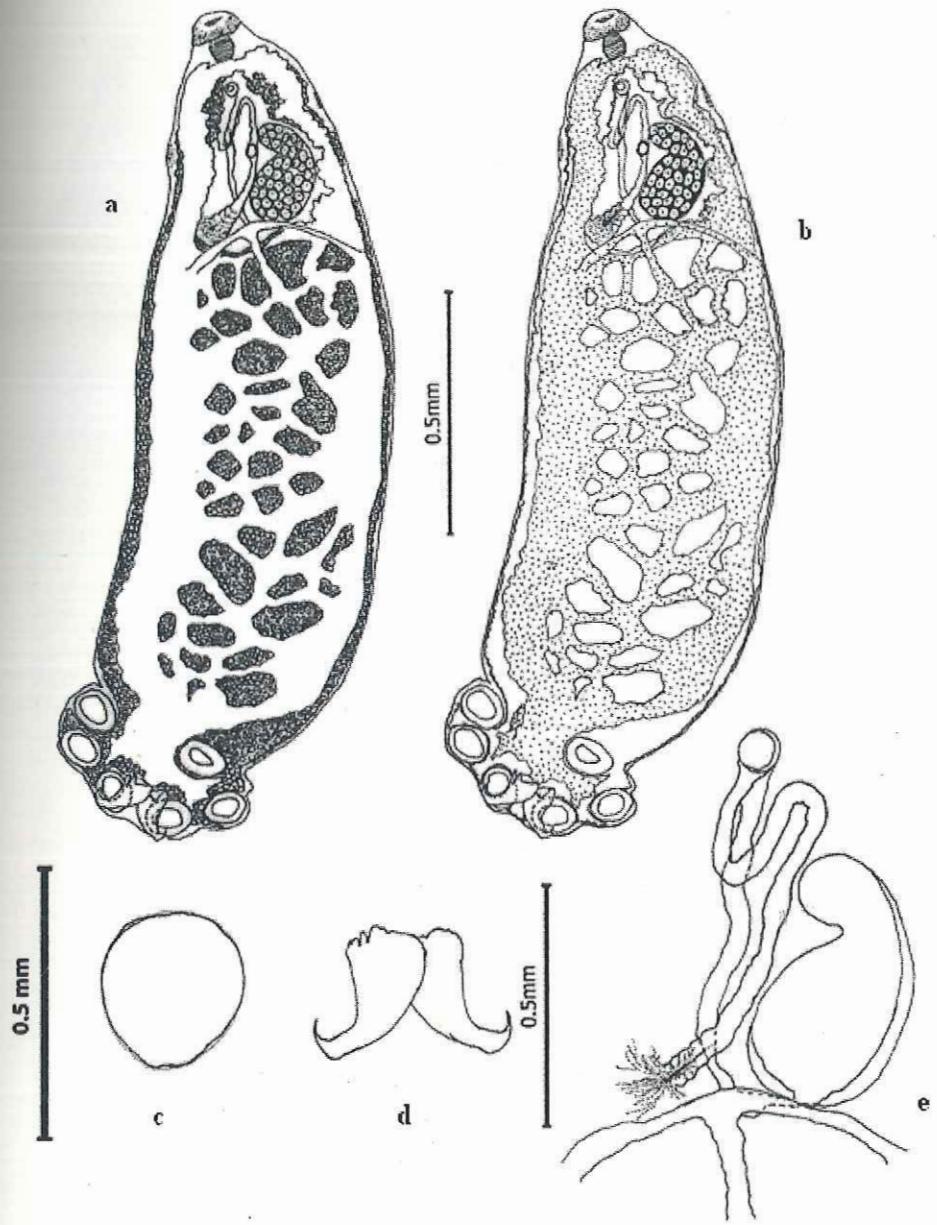
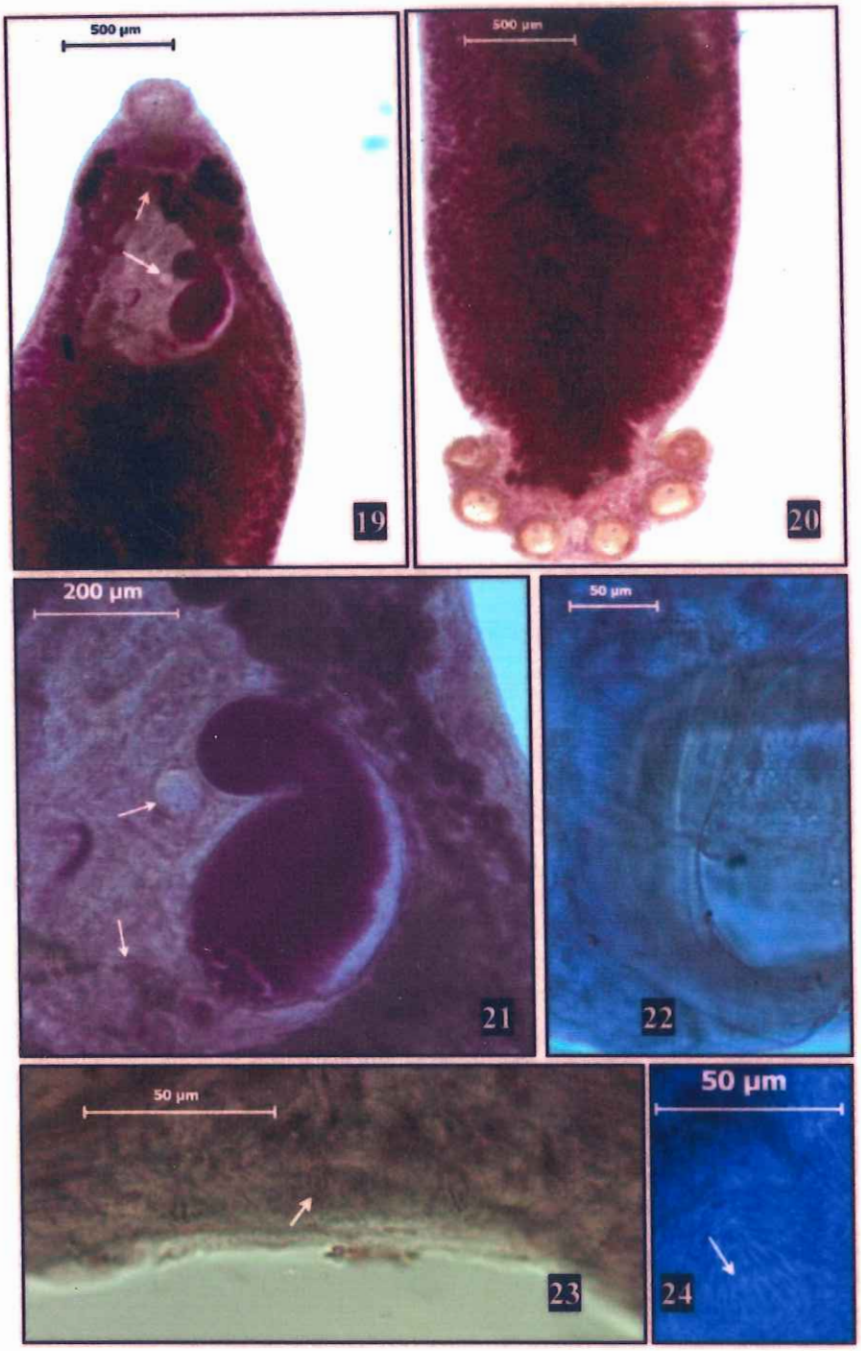


Fig. 18 ***Polystoma*** sp

a. Whole mount, ventral view
b. Same fluke with the intestinal diverticula highlighted.
c. Egg
d. Hamuli
e. Outlines of the fluke reproductive unit.



Figs. 19-24 ***Polystoma*** sp.- Photomicrographs

19. Anterior end (ventral view), showing the extent of testes and vitellaria, the genital pore (short arrow) and the egg (long arrow)

20. Posterior end showing the extend of vitellaria into the opisthaptor

21. Magnified view of anterior region, showing genito-intestinal canal, egg (arrows) and vaginae in top right of ovary

22. A magnified view of a single hamulus

23. Microhooks between the hamuli (arrow)

24. Magnified view of genital crown (arrow)

Table 4 Morphometric measurements of *Polystoma* sp.

Characters		Range	Mean	S.D
Length of the body		2.48-5.38	4.319	±1.0870
Maximum width of body		1.31-1.47	1.398	±0.0595
Width at level of vaginae		0.87-0.94	0.906	±0.0308
Oral sucker:	Length	0.13-0.20	0.174	±0.0262
	Breadth	0.16-0.34	0.271	±0.0752
Opisthaptor:	Length	0.52-0.62	0.579	±0.0411
	Breadth	1.15-1.38	1.251	±0.0838
Haptor length: Body length		3.58-4.85	4.089	±0.4709
Haptor breadth: Maximum somal breadth		0.80-0.96	0.887	±0.0682
Hamulus		0.33-0.38	0.360	±0.0170
Pharynx:	Length	0.11-0.16	0.138	±0.0162
	Breadth	0.11-0.13	0.128	±0.0125
Ovary:	Length	0.51-0.69	0.602	±0.0866
	Breadth	0.25-0.34	0.289	±0.0348
Distance of vaginae from anterior end		0.82-0.96	0.892	±0.0549
Distance of genital pore from anterior end		0.46-0.64	0.532	±0.0682
Egg size:	Length	.0008	.0008	
	Breadth	.0007	.0007	

Remarks:

Though the present form closely resembles ***P. indicum*** Diengdoh and Tandon, 1991, this fluke appears to be a variant of the latter on account of having a few minor deviations, namely, opisthaptor body width ratio somewhat equal, caeca highly anastomosed in pre-opisthaptoral region to give a mesh-like appearance, a single egg and a different anuran host. The overall appearance sets it apart from ***P. indicum***, which has the following characters viz. hamuli with entire base, opisthaptor considerably small compared to body dimensions, intestinal anastomoses few and random and eggs numerous in the uterus.

The species identification of the present form is thus kept in abeyance for want of more specimens and the form is designated herein as ***Polystoma*** sp.

Polystoma kohimaensis n. sp.

(Figs. 25-32)

Twelve specimens of a monogenean belonging to the genus ***Polystoma*** were recovered from 7 of the 13 *Rhacophorus gongshanensis* hosts examined in Kohima.

Description (based on measurements and observations of 5 mature specimens):
Body elongated or foliate in shape, tapering towards anterior end from the broadest mid-section of the body, abruptly ending in sub-terminal oral sucker and in posterior terminating bluntly in opisthaptoral region; surface devoid of any spines. Opisthaptor somewhat half the size of broadest section of body; three pairs of well developed suckers and a pair of macrohooks or hamuli present, hamulus base entire, not broad, with sharply pointed recurved tips; 2 microhooks between hamuli. Pharynx prominent, muscular, leading to bifurcate intestine. Intestinal caeca indented at irregular interval on lateral aspects, sending out diverticula directed toward median line, caeca not uniting throughout length. Testes numerous, forming insurmountable mass in pre and post-ovarian, both in marginal and inter-caecal zones; vas deferens arising in region of junction of female genital ducts, running anteriad slightly to side of median line; cirrus-sac small, cirrus tipped with genital crown of 12 hooklets, opening in front of female genital pore into common genital sinus, latter opening to exterior close behind intestinal bifurcation. Ovary round, bulbous in shape, uterus with a single large oblong or oval egg.

Morphometric measurements of this form are given in Table 5

Host: *Rhacophorus gongshanensis*
Locality: Kohima
Location: Urinary bladder

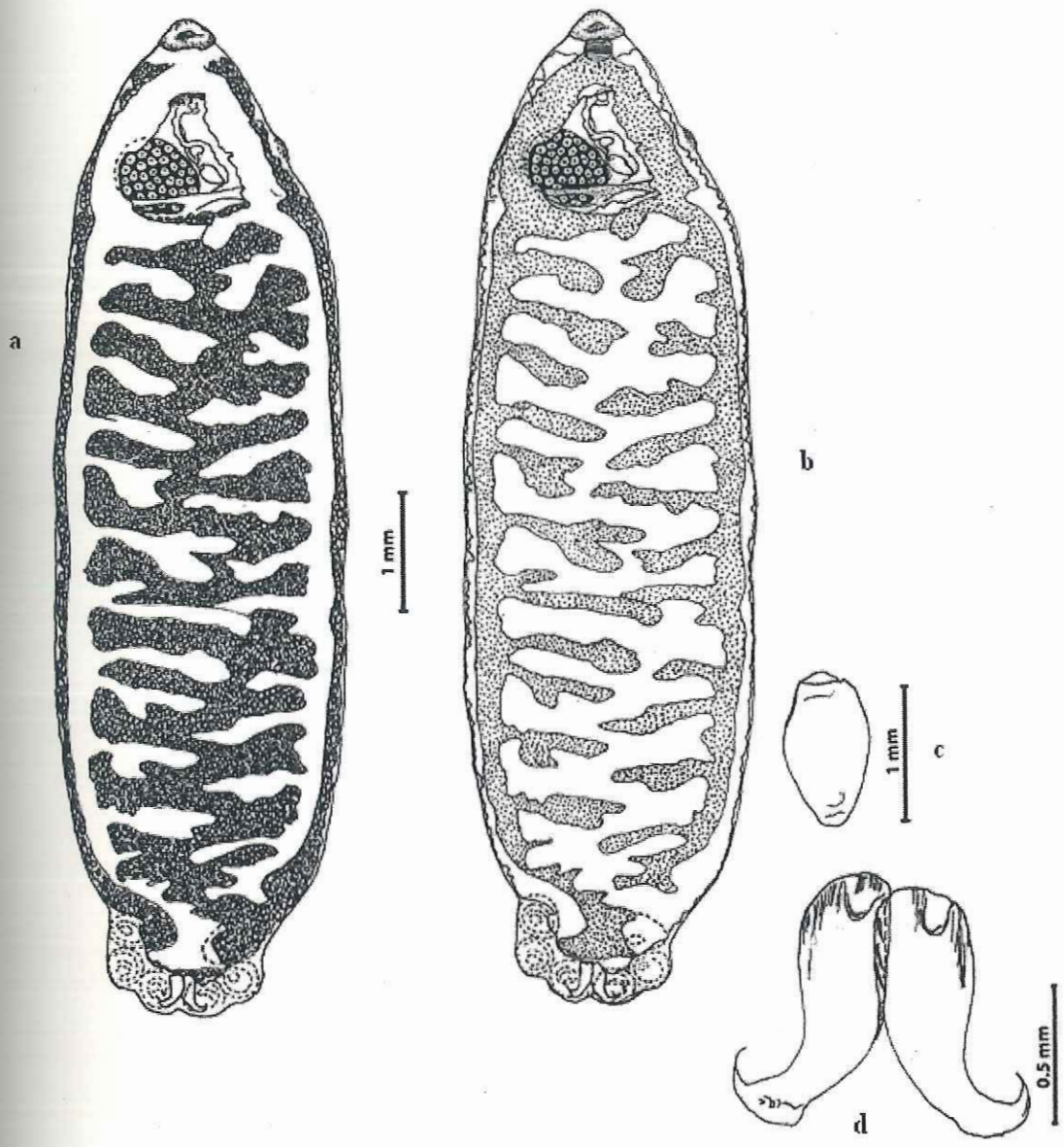
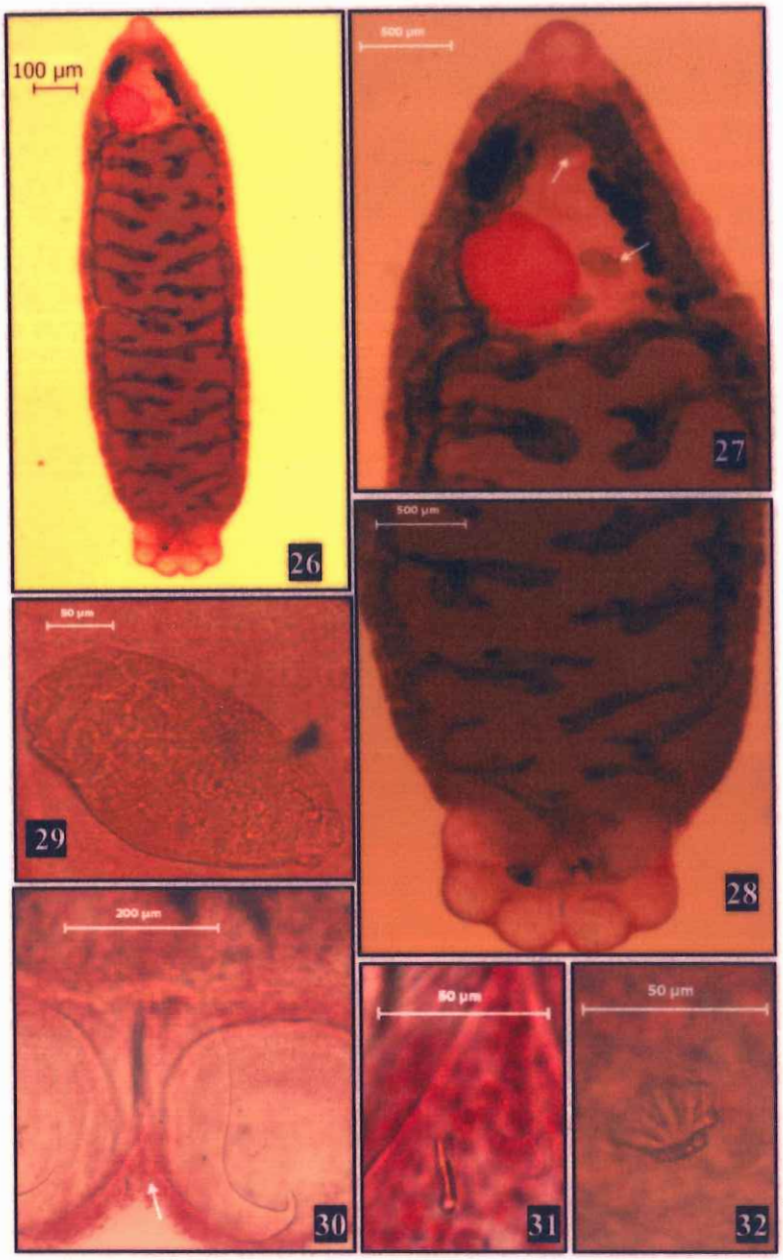


Fig. 25 ***Polystoma kohimaensis*** n. sp.

a. Whole mount, dorsal view
b. Same fluke with intestinal diverticula highlighted
c. Egg
d. Hamuli



Figs. 26-32 ***Polystoma kohimaensis*** n. sp.- Photomicrographs

26. Whole mount, dorsal view

27. A Magnified view of the anterior half of body with the round and bulbous ovary, a single large egg in the uterus (arrow) and the genital opening (small arrow)

28. Posterior half, showing the pattern of the intestinal caeca and their extent into the opisthaptor

29. A magnified view of the single egg

30. The two microhooklets between the hamuli

31. Magnified view of the hamuli

32. A magnified view of the genital crown

Table 5 Morphometric measurements of *Polystoma kohimaensis* n. sp

Characters		Range	Mean	S.D
Length of the body		4.623-8.464	6.4630	±1.7827
Maximum width of body		1.242-1.242	1.8630	±0.4640
Width at level of vaginae		1.173-1.173	1.3708	±0.1505
Oral sucker:	Length	0.138-0.138	0.1978	±0.0621
	Breadth	0.322-0.322	0.4278	±0.0720
Opisthaptor:	Length	0.736-0.736	0.7774	±0.0252
	Breadth	1.288-1.288	1.4306	±0.1449
Haptor length: Body length		2.461-2.461	4.8852	±2.3045
Haptor breadth: Maximum somal breadth		1.035-1.035	1.1500	±0.1508
Hamulus length		0.048-0.048	0.0540	±0.0042
Pharynx:	Length	0.161-0.161	0.1978	±0.0385
	Breadth	0.161-0.161	0.1932	±0.0206
Ovary:	Length	0.345-0.345	0.5336	±0.1670
	Breadth	0.230-0.230	0.4324	±0.1325
Distance of vaginae from anterior end		0.943-0.943	1.1086	±0.0968
Distance of genital pore from anterior end		0.483-0.483	0.6348	±0.0957
Genital crown hooklets length		0.0161	0.0161	±0.0161
Egg size:	Length	0.918-0.918	1.0350	±0.0758
	Breadth	0.486-0.486	0.5418	±0.0419

Discussions:

On account of having the characters viz. a body leaf-like, opisthaptor with a pair of hamuli, testes numerous, post ovarian in intercaecal field, uterus pre ovarian, vaginae present, the present form is relegated to the genus ***Polystoma***.

This monogenean form also closely resembles ***P. indicum*** Diengdoh and Tandon (1991), the hitherto known representatives of the only genus from Indian sub-continent. Hamuli with entire base, opisthaptor relatively small as compared to the body dimensions, intestinal anastomoses few and random, eggs numerous in uterus are the diagnostic characters of ***P. indicum***. However, in the present form the opisthaptor is half the size in width to the broadest section of the body, caeca do not unite and anastomose in pre- and opisthaptoral regions but show finger-like projections of divericula giving off 1 or 2 branches towards the median line; uterus is with only one but very large egg; moreover, in addition to these characters the ovary rounded in shape, genital crown with 12 hooklets as compared to 8 in ***P. indicum***, one of the intestinal caeca extending into the opisthaptoral area giving out branches and a different host are the characters that set the present form completely apart from ***P. indicum***. The present forms also do not tally with ***Polystoma*** sp. n.sp. described herein from the host, *Philautus* sp.1 in many major characters, namely, opisthaptor body width ratio somewhat equal, caeca highly anastomosed in pre-opisthaptoral region to give a mesh-like appearance, a single egg and a different anuran host. Hence, it is considered to be a new species under the genus ***Polystoma*** and named as ***Polystoma kohimaensis*** after the type locality from where it was collected.

Species diagnosis: Polystomatidae: Polystomatinae

Opisthaptor half the size in width to the broadest section of the body, caeca not united throughout the length, uterus with one very large egg, genital crown with 12 hooklets.

Type host: *Rhacophorus gongshanensis* (Yang *et* su, 1984)

Type Locality: Kohima, Nagaland (25°30'13.86"N and 93°55'13.86"E to 26°02'8.44" N and 94°22'0.36" and 94°22'0.36" E, India)

Etymology: Named after the type locality from where it was collected

Polystoma hylai n. sp.

(Figs. 33-46)

Thirty specimens of another polystomid were recovered from 10 of the 106 *Hyla annectans* hosts collected from Kohima.

Description (based on measurements and observations of 5 mature specimens):

Body elongated or foliate in shape, tapering towards anterior end, abruptly ending in sub-terminal oral sucker, posteriorly terminating bluntly in opisthaptoral region; surface devoid of any spines. Opisthaptor somewhat equal to or wider in size with broadest section of body; three pairs of well-developed suckers and a pair of macrohooks (hamuli) with base entire, broad and sharply pointed recurved tips present. Pharynx prominent, muscular, leading to bifurcate intestine. Intestinal caeca indented at irregular intervals on lateral aspects, sending out diverticula directed toward median line, caeca united in pre- and opisthaptoral regions to send out again 2-3 diverticula into opisthaptor, number of prehaptoral anastomoses being more than three. Testes numerous, forming insurmountable mass in pre- and postovarian, both in marginal and intercaecal zones; vas deferens arising in region of junction of female genital ducts, running anteriad slightly from lateral to side of median line; cirrus-sac small, cirrus tipped with genital crown of 10 hooklets, opening in front of female genital pore into common genital sinus, latter opening to exterior close behind intestinal bifurcation. Ovary retort shaped; vaginae on either lateral margin of body. Vitellaria extensive, follicular, commencing from pharyngeal level, posteriorly extending into opisthaptoral zone, intruding in spaces between suckers, overlapping intestinal diverticula. Uterus with a single large, oblong to oval egg.

The surface fine topography of this flukes did not reveal any tegumental papillae, but revealed many transverse folding and infolding of the body. The oral sucker showed many dome shaped papillae in the rim and the clamp also revealed numerous radial folding and infolding.

Morphometric measurements of this form are given in Table 6

Host: *Hyla annectans*

Locality: Kohima

Location: Urinary bladder

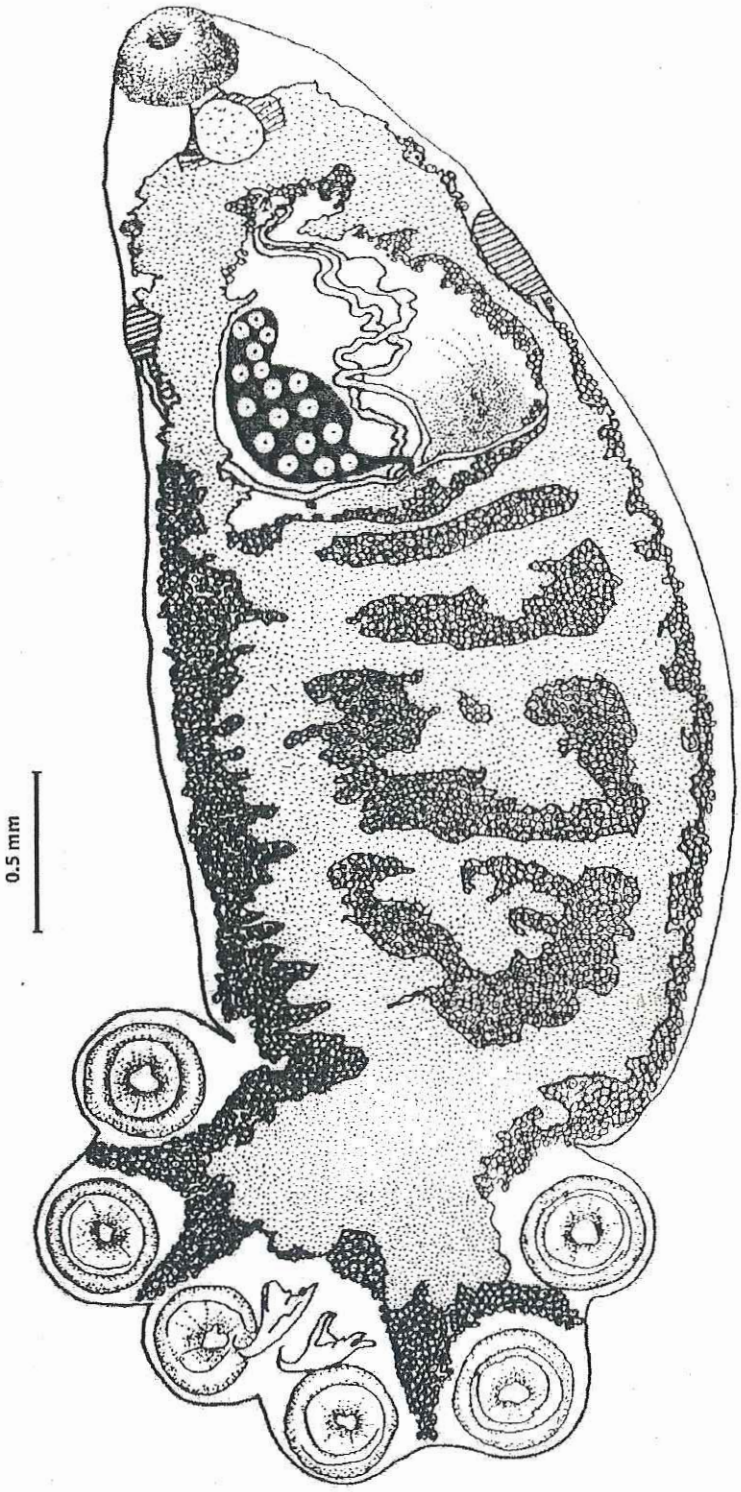


Fig. 33(i) ***Polystoma hylai*** n. sp

a. Whole mount, dorsal view

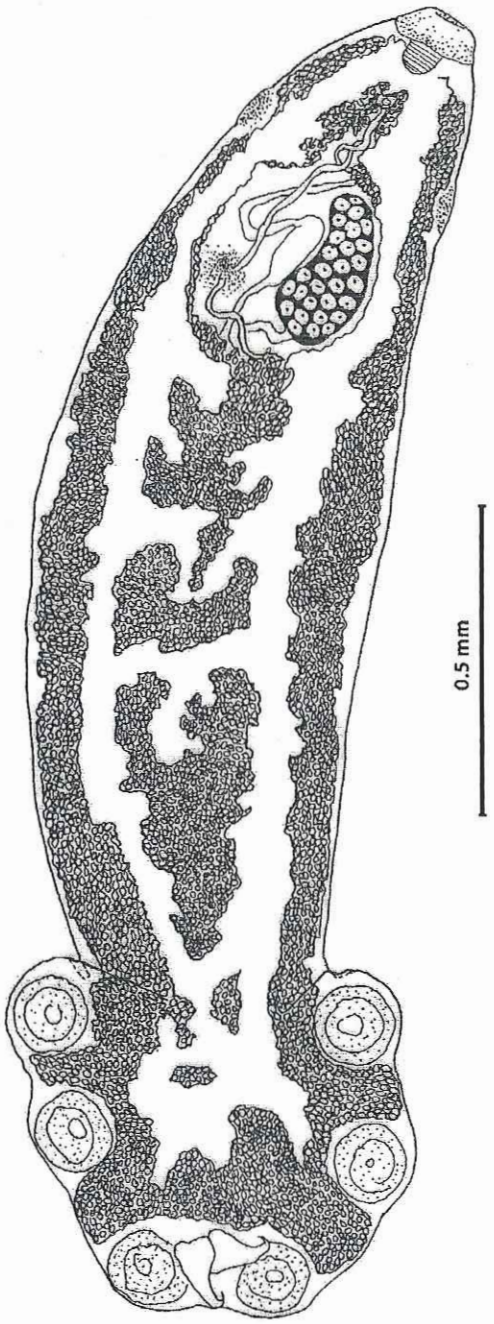
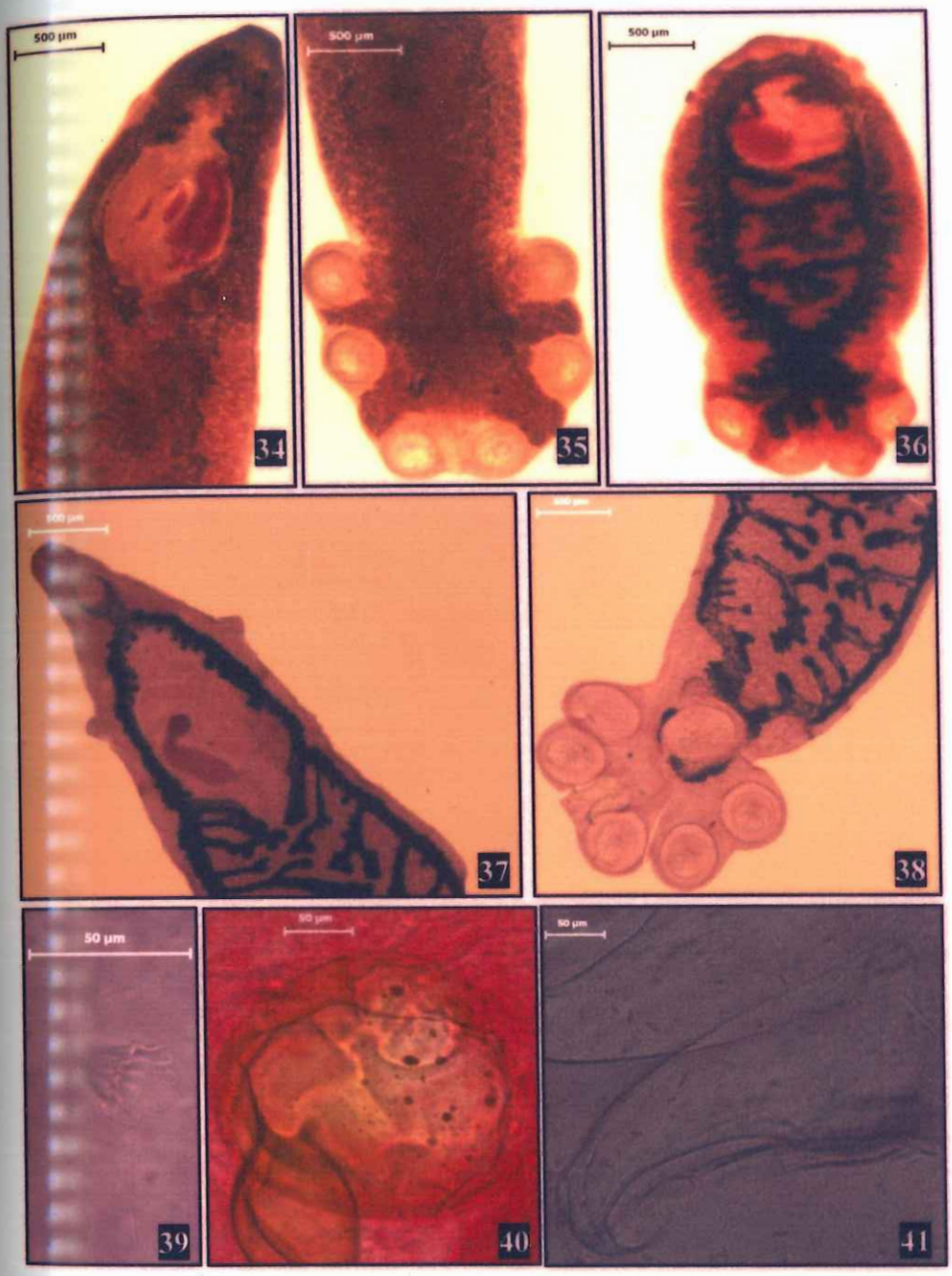


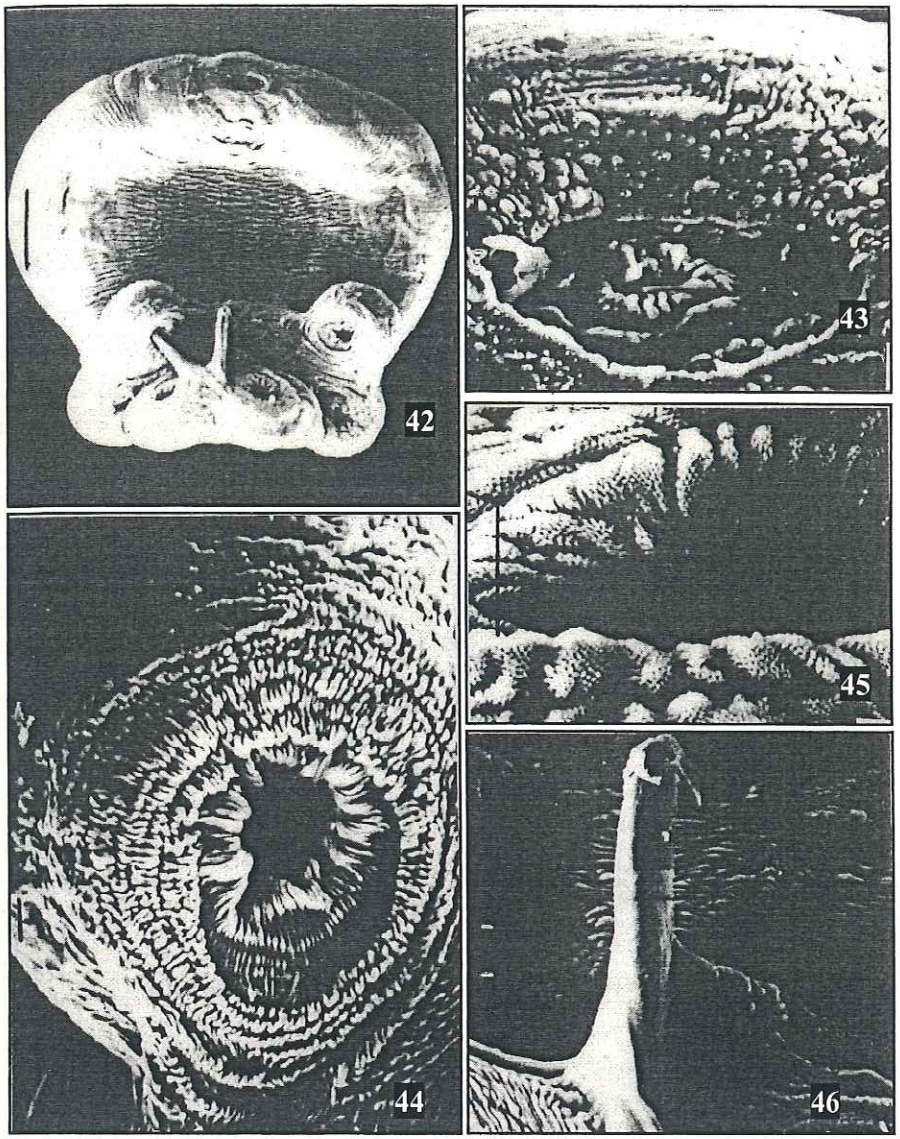
Fig. 33(ii) ***Polystoma hylai*** n. sp

a. Whole mount, ventral view of another fluke



Figs. 34-41 ***Polystoma hylai*** **n. sp.- Photomicrographs**

34. Anterior part of the fluke, ventral view
35. Posterior region showing the opisthaptor and intrusion of vitellaria between clamps
36. A whole mount of juvenile fluke
37. Anterior end of another matured specimen
38. Posterior part of the same specimen
39. A magnified view of the genital crown
40. Egg
41. A magnified single hamuli with whole base of an adult fluke



Figs. 42-46 ***Polystoma hylai*** n. sp.- Scanning electron micrographs.

42. Full worm, ventral view (scale bar= 100μm)

43. A magnified view of the oral sucker, showing domed papillae in circum-oral region (scale bar= 10μm)

44. A magnified view of an opisthaptoral clamp, showing the radial striations and folding, and indicaton of underlying musculature (scale bar= 10μm)

45. A magnified view of the vaginal opening, showing fine pitted texture in the folding (scale bar= 10μm)

46. A magnified view of an hamuli, showing the terminal hook (scale bar=10 μm)

Table 6 Morphometric measurements of *Polystoma hyla* n. sp.

Characters		Range	Mean	S.D
Length of the body		4.554-8.602	5.515	±1.7313
Maximum width of body		1.288-1.817	1.573	±0.2136
Width at level of vaginae		0.851-1.173	1.030	±0.1221
Oral sucker	Length	0.115-0.276	0.193	±0.0701
	Breadth	0.184-0.506	0.377	±0.1404
Opisthaptor:	Length	0.897-1.173	1.053	±0.1059
	Breadth	1.472-1.817	1.633	±0.1445
Haptor length: Body length		2.415-7.291	4.204	±2.0878
Haptor breadth: Maximum somal breadth		1.472-1.748	1.591	±0.1020
Hamulus length		0.037-0.055	0.050	±0.0076
Opisthaptoral suckers				
Pharynx:	Length	0.092-0.207	0.165	±0.0471
	Breadth	0.115-0.207	0.156	±0.0341
Ovary:	Length	0.598-0.667	0.621	±0.0281
	Breadth	0.184-0.276	0.23	±0.0363
Distance of vaginae from anterior end		0.759-1.219	0.910	±0.1948
Distance of genital pore from anterior end		0.368-0.943	0.547	±0.2273
Egg size:	Length	.0023	.0023	
	Breadth	.0019	.0019	

Discussion:

The present form seems to be quite different from ***P. indicum*** Diengdoh and Tandon, 1991, in having a different body contour and surface texture- elongated slender body lacking tegumental papillae, equal or slightly larger width of opisthaptor than that of the body; intestinal caeca with few (4-5) anastomoses, uniting in opisthaptoral zone and sending out 2-3 diverticulation into the opisthaptor; genital crown with 10 hooklets; uterus with a large single egg. In earlier studies, a few specimens of a polystomid form were recovered from *Hyla annectans* from Kohima locality and reported as ***Polystoma*** sp. by Tandon *et al.* (2001). During the present study, many more specimens of the same form were collected. On comparison with the known species of the genus ***P. indicum*** and the new species ***P. kohimaensis*** (described herein), the form collected from hylid anurans was revealed to be different from both.

In view of the foregoing differences, the present fluke is considered to be yet another new form from the region and can undoubtedly be regarded as a new species and is named as ***Polystoma hylai***, after its host.

Specific diagnosis: Polystomatidae: Polystomatinae.

Opisthaptor sequal or more in width than that of the soma or preopisthaptoral body. Intestinal anastomoses few, restricted to 4-5 in number and diverticulate, extending posteriad into the opisthaptor. Egg single, large in size.

Type host: *Hyla annectans* Jerdon, 1870

Type locality: Kohima, Nagaland (Nagaland, 25°30'13.86"N and 93°55'13.86"E to 26°02′8.44" N and 94°22′0.36" and 94°22'0.36" E, India)

Etymology: Named after the anuran host in which it was collected.

Genus *Neoriojatrema* n. g.

Neoriojatrema mokokchungensis n. g., n. sp.

(Figs.47-66)

Altogether, 13 specimens of this form were recovered from the bladder of *Megophrys glandulosa* in Mokokchung and Tuensang localities.

Description (based on measurements of 8 mature flukes and observations of 1 specimen under the SEM):

Description (based on measurements and observations of 5 mature specimens)

Body leaf-like conical in shape, tapering towards anterior end, abruptly ending in sub-terminal oral sucker, posteriorly terminating at the junction of body and opisthaptor; body aspinose. Opisthaptor discoid lacking hamuli or macrohooks. Intestinal caeca highly diverticulate, giving out primary and secondary branches at irregular intervals on both lateral and median aspects, restricted in soma and not uniting in pre-opisthaptoral regions. Testes numerous, forming insurmountable mass in pre- and postovarian, both in marginal and intercaecal zones; vas deferens arising in region of junction of female genital ducts, running anteriad slightly from lateral to side of median line; cirrus-sac small, cirrus tipped with genital crown of 11-12 hooklets, opening in front of female genital pore into common genital sinus, latter opening to exterior close behind intestinal bifurcation. Ovary retort shaped with flowery or lobed distal end; vaginae on either lateral margin of body. Vitellaria extensive, follicular restricted in the soma. Uterus with 7-22 oval eggs.

The surface fine topography of the fluke revealed numerous tegumental papillae mounted in a mesh-like net work; some of the papillae below the oral region were ciliated. The tegumental papillae under higher resolution revealed spongy in appearance and the opisthaptoral region was clearly devoid of any papillae.

Morphometric measurements of this form are given in Table 7

Host: *M. glandulosa*

Location: Bladder

Locality: Mokokchung, Tuensang

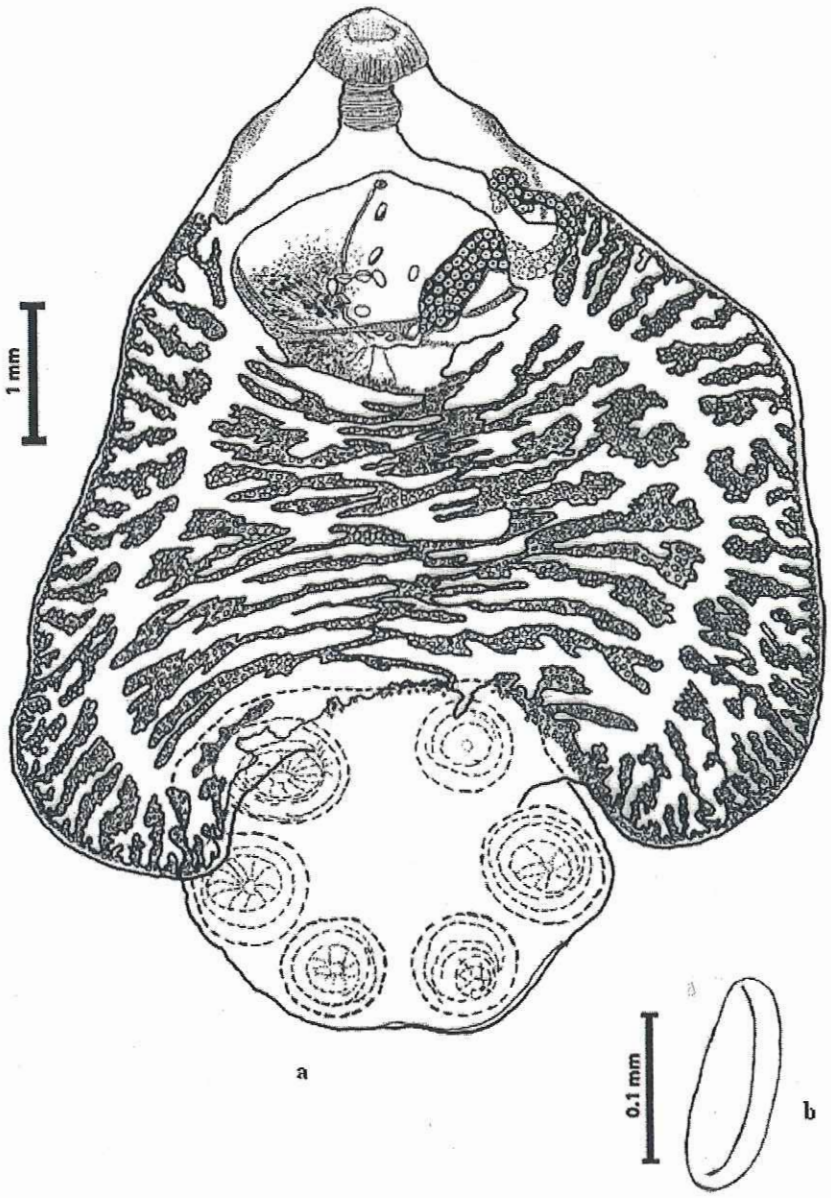


Fig. 47 ***Neoriojatrema mokokchungensis*** n. g., n. sp.

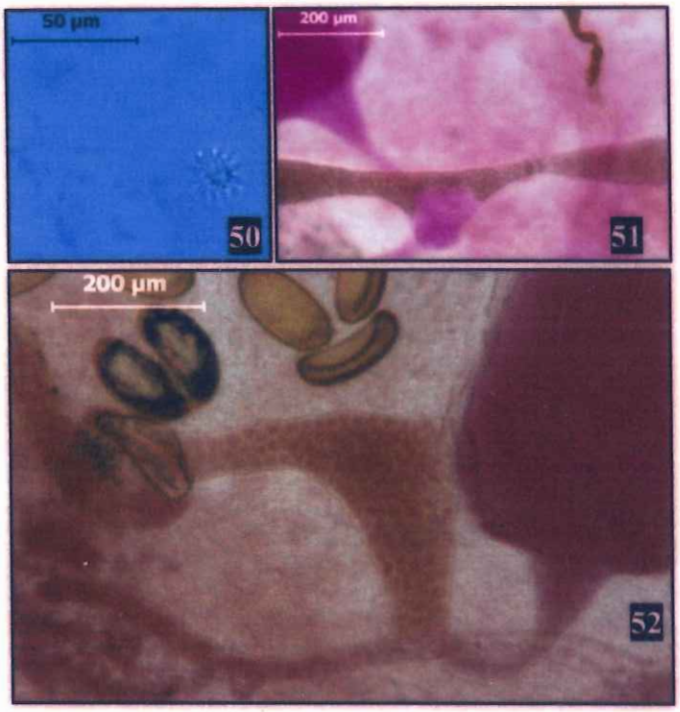
a. Whole mount, ventral view

b. Egg

Figs. 48-49 ***Neoriojatrema mokokchungensis*** n. g., n. sp.- Photomicrographs

48. Anterior end of the worm, dorsal view

49. Posterior end.

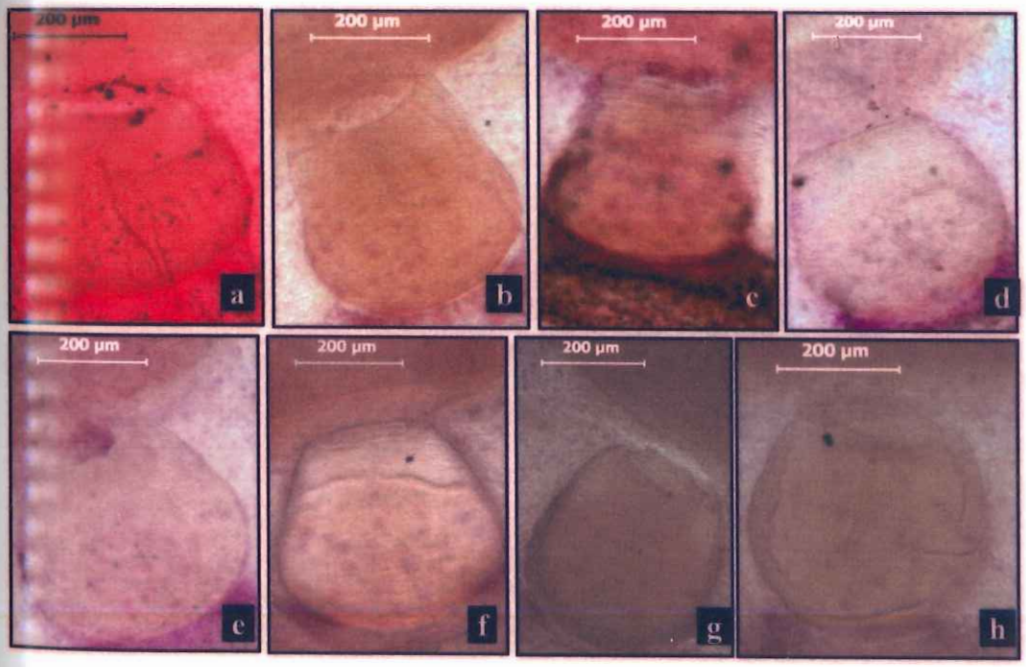


Figs.50-52 ***Neoriojatrema mokokchungensis*** n.g., n sp.- Photomicrographs

50. Genital crown of the fluke in high resolution

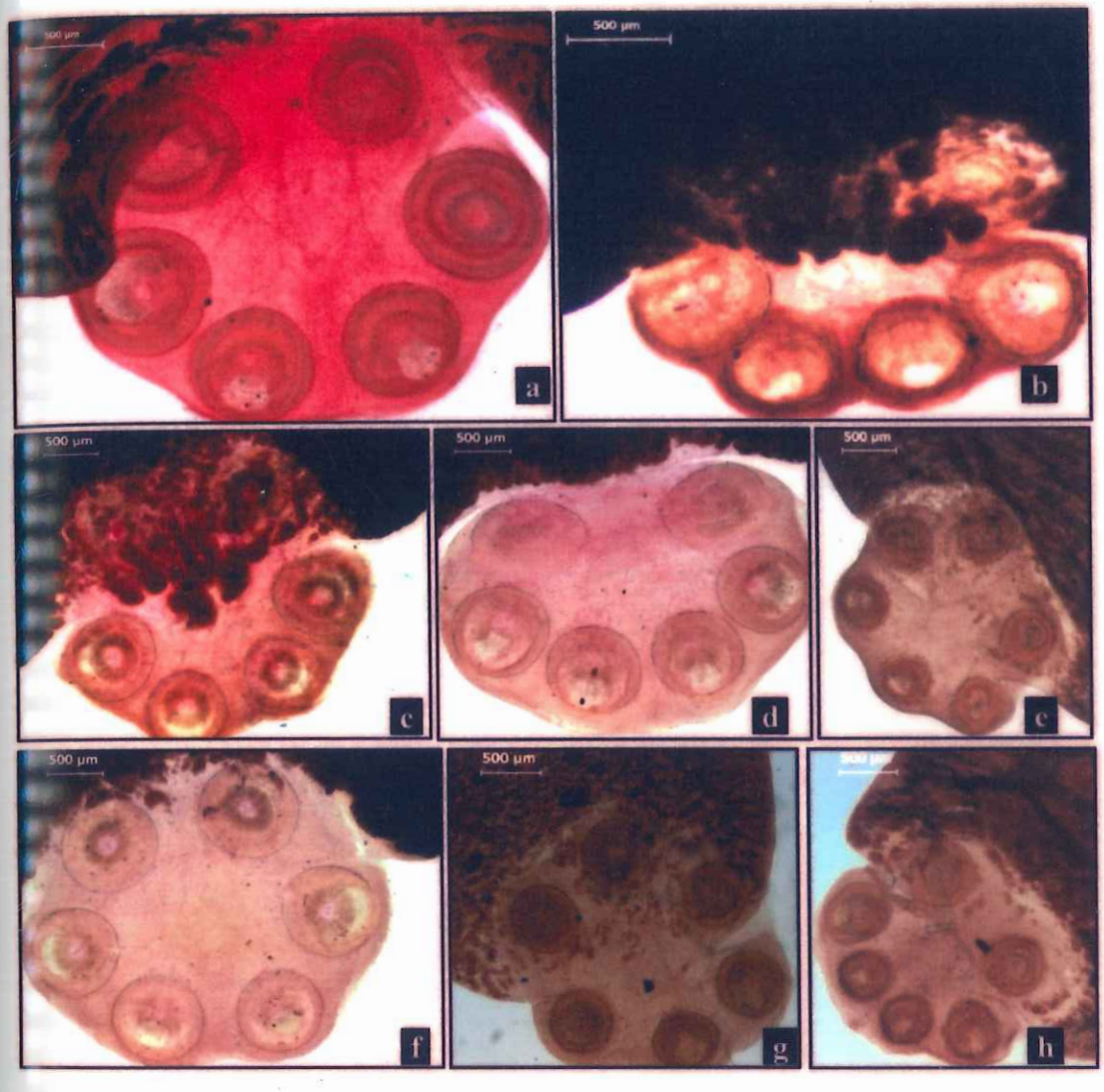
51. A magnified view of Mehlis' complex with ovary on the left side, dorsal view

52. A magnified ventral view of another specimen showing the genito-intestinal canal region and eggs in the uterus

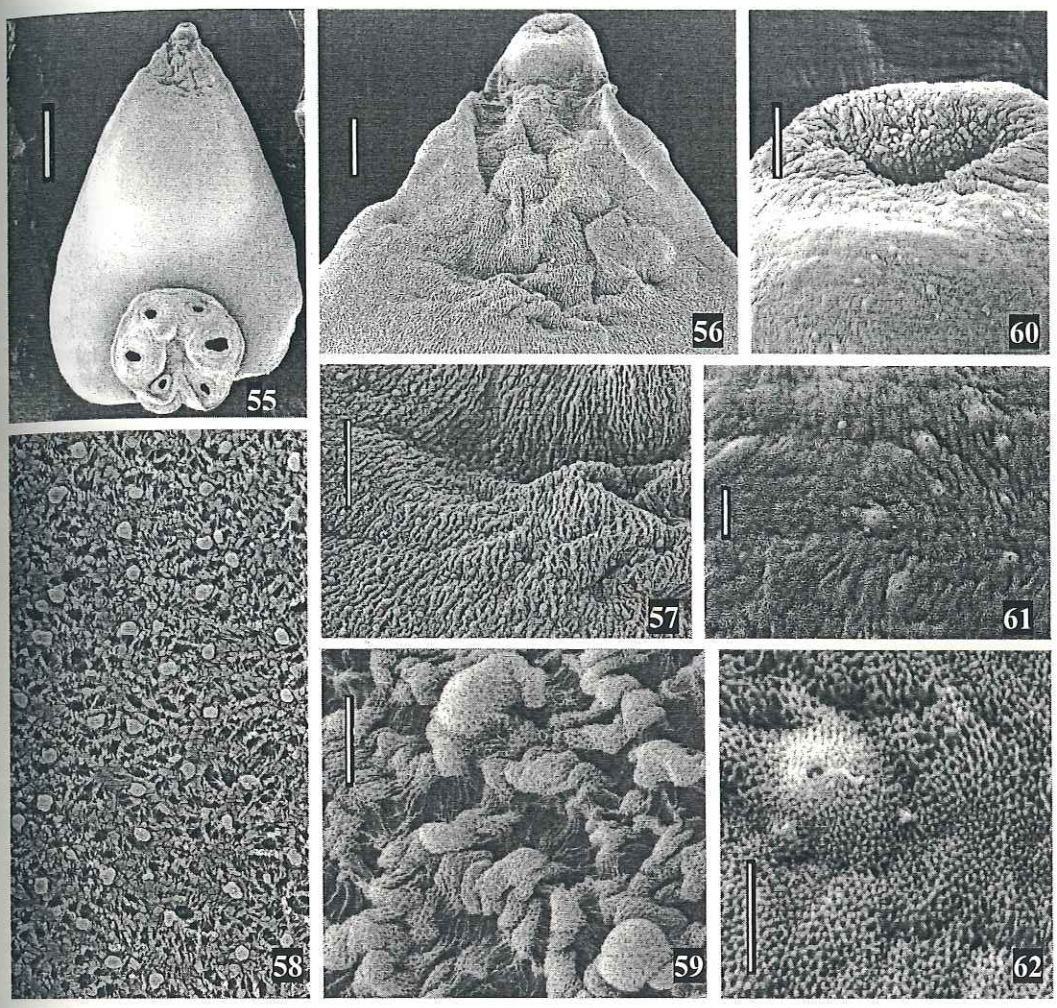


Figs. 53(a-h) ***Neoriojatrema mokokchungensis*** n. g., n. sp.- Photomicrographs.

Bulbous pharynx of 8 mature specimens with lower rim of oral sucker in top. (Borax carmine and Meyer's Carmallum)



Figs.54 (a-h) ***Neoriojatrema mokokchungensis*** n. g., n. sp.- Photomicrographs. Opisthaptors and posterior part of the soma of 8 mature specimens showing the lack of hamuli and variations in the extent of caeca and vitellaria.



Figs. 53-60 Neodiojatrema ***mokokchungensis*** n. g., n. sp- Scanning electron micrographs

55. Whole fluke, ventral side (scale bar= 1mm)

56. Anterior end, showing many foldings around the genital pore, (scale bar=200μm)

57. Body surface below the genital opening covered by numerous dot-like papillae scattered on the mesh-like network, (scale bar=100μm)

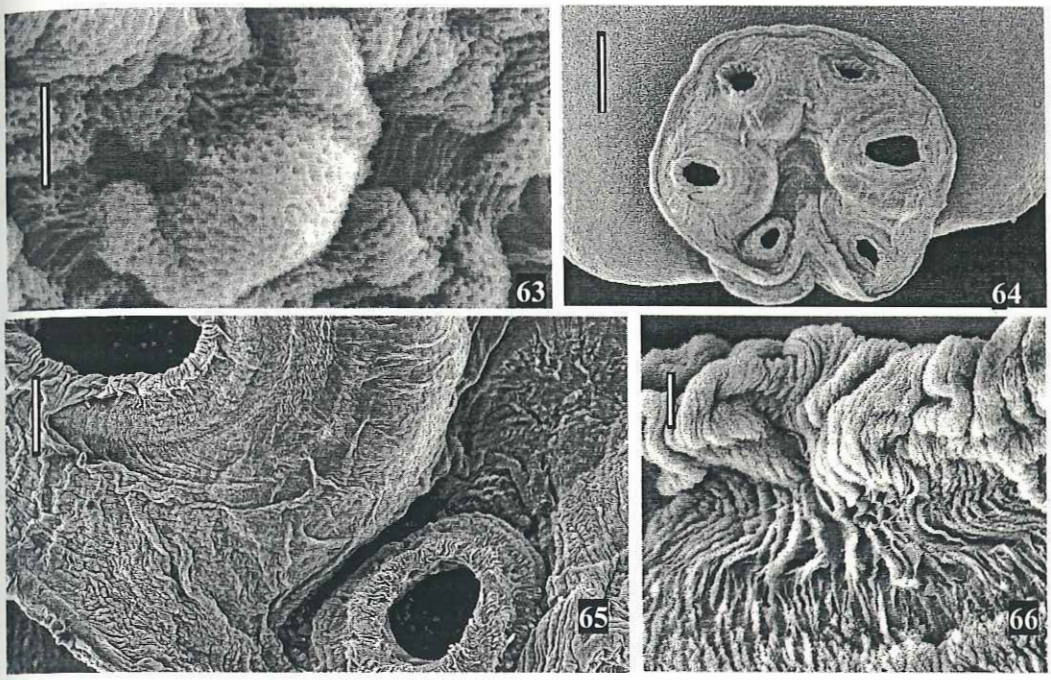
58. A close up view of the general tegument showing the many papillae mounted in a mesh-like network, (scale bar=50μm)

59. A view of the many papillae-like out-growth in higher magnification, (scale bar=10μm)

60. Anterior extremity with many papillae in the circum-oral and below the oral sucker (scale bar= 50μm)

61. A magnified view below the oral sucker with domed-shaped ciliate papillae (scale bar=10μm)

62. Domed-shaped ciliate-papillae in higher resolution (scale bar=5μm)



Figs. 63-66 ***Neoriojatrema mokokchungensis***- Scanning electron micrographs

63. Magnified view of a single papilla-like outgrowth of the general body tegument (scale bar=5μm)

64. A view of the hook less opisthaptor with the six suckers showing absence of any papillae-like out- growth in its surface, (scale bar=500μm)

65. A close up view of the suckers with many folding in the surface and ring-like musculature around the sucker, a clear indication of the powerful underlying musculature, (scale bar=100μm)

66. Rim of the sucker in higher resolution showing the many transverse folding and infolding with fuzzy appearance (scale bar=10μm)

Table 7 Morphometric measurements of *Neoriojatrema mokokchungensis* n. g., n. sp.

Characters		Range	Mean	S.D
Length of the body		6.969-13.34	9.565	±2.5127
Maximum width of body		3.795-7.82	5.911	±1.5940
Width at vaginae		1.61-2.116	1.932	±0.1602
Oral sucker:	Length	0.667-0.805	0.747	±0.0576
	Breadth	0.299-0.506	0.393	±0.0781
Opisthaptor:	Length	2.093-4.554	2.837	±0.8062
	Breadth	2.093-3.358	2.590	±0.4743
Haptor length: Body length		4.876-10.626	7.59	±1.9538
Haptor breadth: Maximum somal breadth		2.392-6.739	5.097	±1.3258
Opisthaptoral suckers:	Length	0.506-0.897	0.727	±0.1092
	Breadth	0.506-1.012	0.807	±0.1315
Pharynx	Length	0.345-0.414	0.382	±0. 0273
	Breadth	0.299-0.437	0.376	±0.0458
Ovary	Length	1.495-2.484	2.236	±0.3060
	Breadth	0.299-0.46	0.391	±0.0521
Distance of genital pore from anterior end		0.897-1.265	1.063	±0.1421
Distance of vaginae from anterior end		1.058-1.173	1.121	±0.0383
Egg size	Length	0.138-0.167	0.146	±0.0095
	Breadth	0.080-0.085	0.082	±0.0017

Discussion:

With leaf-like body shape or pyriform, opisthaptor discoid with six suckers, prohaptor in the form of oral sucker situated terminally or subterminally, followed by a well developed bulbous pharynx, intestinal caeca extending up to the posterior region of the body, genital pore median and on or near intestinal bifurcation, genital crown present, testes reticulate, vitellaria follicular, and vaginae present, the present form can be undoubtedly relegated to the superfamily Polystomatoidea Price, 1936.

In having a compound 6-suckered opisthaptor and testes in the form of follicular mass, the present form belongs to the family Polystomatidae and subfamily Polystomatinae. Of the so far known six genera under the subfamily, the present form comes closer to ***Eupolystoma*** Kaw, 1950; ***Pseudopolystoma*** Yamaguti, 1963 and ***Riojatrema*** Lamothe-Argumedo, 1964, in all of which the opisthaptor lacks the hamuli or microhooks. However, ovary located in the anterior third of the body is the character of the former two genera that does not tally with the present form. For the third genus ***Riojatrema***, the diagnostic characters include the width of opisthaptor greater than body, cylindrical pharynx with a constriction in the middle, intestinal diverticula anastomosing prehaptorally and uniting in the region of the opisthaptor and follicular vitellaria extending from level of genito-intestinal canal to opisthaptor.

The genus ***Riojatrema***, so far includes only two species, namely, ***R. bravoae*** Lamothe-Argumedo, 1964, from Mexico and ***R. ecuadorensis*** Dyer, 1985, from Ecuador. Intestinal caeca uniting posteriorly in the opisthaptoral region, possessing numerous diverticula forming 3-4 anastomoses in median field of body; cirrus with 8-9 spines, ovary pyriform, vitelline follicles widely scattered and into the opisthaptoral region are the characters of ***R. bravoae***. Further, in ***R. ecuadorensis*** the opisthaptoral disc is broader than the main body or the soma, the cylindrical pharynx is constricted in the middle, the intestinal caeca with lateral diverticula anastomose prehaptorally, uniting in the opisthaptoral region, the vitellaria extent from the level of the genito-intestinal canal up to the opisthaptor.

On comparison with the species of ***Riojatrema,*** the present form shares only the characters of opisthaptor (lacking hamuli) and the uterus in level with the ovary. However, it differs from both the species in several characters-opisthaptor relatively smaller than the body, pharynx bulbous and not cylindrical with a constriction, intestinal caeca with lateral diverticula but not uniting or anastomosing prehatoprally, ovary long with a lobed/flowery head end, genital crown spines 11-12, vitellaria not extending into the opisthaptoral area, besides a different host, which set the present form completely apart from them.

In view of the high degree of host specificity demonstrated among the anuran monogeneans, host specificity and geographical distribution have also been assigned significant importance in species differentiation within the genus ***Polystoma*** (Maeder *et al.*, 1970; Combes and Knoepffler, 1977; Dupouy, 1978; Combes and Channing, 1979; Murith, 1981; Kok and Van wyk, 1986; Kok and Seaman, 1987; Diengdoh and Tandon, 1991). However, the extent to which host specificity could be depended upon as a differential character is controversial (Prudhoe and Bray, 1982). And the foregoing differences in generic diagnostic characters, deviating in the present form (i.e, larger width of body than opisthaptor, pharynx not cylindrical and not constricted in the middle but round and bulbous, caeca not extending into the opisthaptor and uniting, vitellaria not extending into the opisthaptioral region) the present form cannot be relegated to the genus ***Riojatrema***. In addition, the characters viz. the genital crown with 11-12 spines, ovary retort shaped with a flowery or lobed distal end and a different anuran host, necessitate the erection of a new genus, so as to accommodate the present monogenean.

In view of the foregoing differences from the hitherto described species and also of the strict host specificity existing among amphibian polystomids, it is therefore, proposed that a new genus be erected for the present form with the name ***Neoriojatrema*** under the subfamily Polystomatinae Gamble, 1896, and a new species ***mokokchungensis*** named after the type locality, Mokokchung in Nagaland, Northeast India.

Diagnostic characters: Polystomatidae

Polystomatidae: Opisthaptor lacking large hooks, not broader than forebody; intestinal caeca highly diverticulate but not anastomosing medially and not uniting posteriorly. Ovary in anterior third of body length, vaginal apertures present. Genital crown with 11-12 spines

Type host: *Megophrys glandulosa* Boulenger, 1890

Type habitat: Urinary bladder

Type Locality: Mokokchung, Nagaland (Nagaland, 26°11'45.21'N and 94°17'33.04"E to-26°45'46.74"N and 94°45'52.49", India)

Etymology: Named as a newer form close to ***Riojatrema*** genus with the species name after the locality from where the host was collected.

DIGENEA

Family Gorgoderidae (Looss, 1899) Looss, 1901

Subfamily Gorgoderinae Looss, 1899

Genus ***Gorgoderina*** Looss, 1902

(Syn. ***Microlecithus*** Ozaki, 1926)

Gorgoderina ellipticum Dwivedi, 1968

(Figs.67-71)

Altogether only 12 specimens were recovered from 4 of the 339 hosts examined, from 4 of the 5 localities surveyed, throughout the study period.

Body size: 2.72-2.92 x 1.28-1.33mm

Egg size: 0.024-0.025 x 0.015-0.017mm

Host: *Euphlyctis cyanophlyctis*

Location: Urinary bladder

Locality: Dimapur

Remarks:

Originally ***G. ellipticum*** was described from *E cyanophlyctis* (=*Rana cyanophlyctis*) in Madhya Pradesh. Diengdoh (1989) reported this species from the same host and *Limnonectes limnocharis* (=*Rana limnocharis*) in Meghalaya. The species was also reported from the same host and locality(*E. cyanophlyctis*, Dimapur) by Tandon *et al.* (2001)

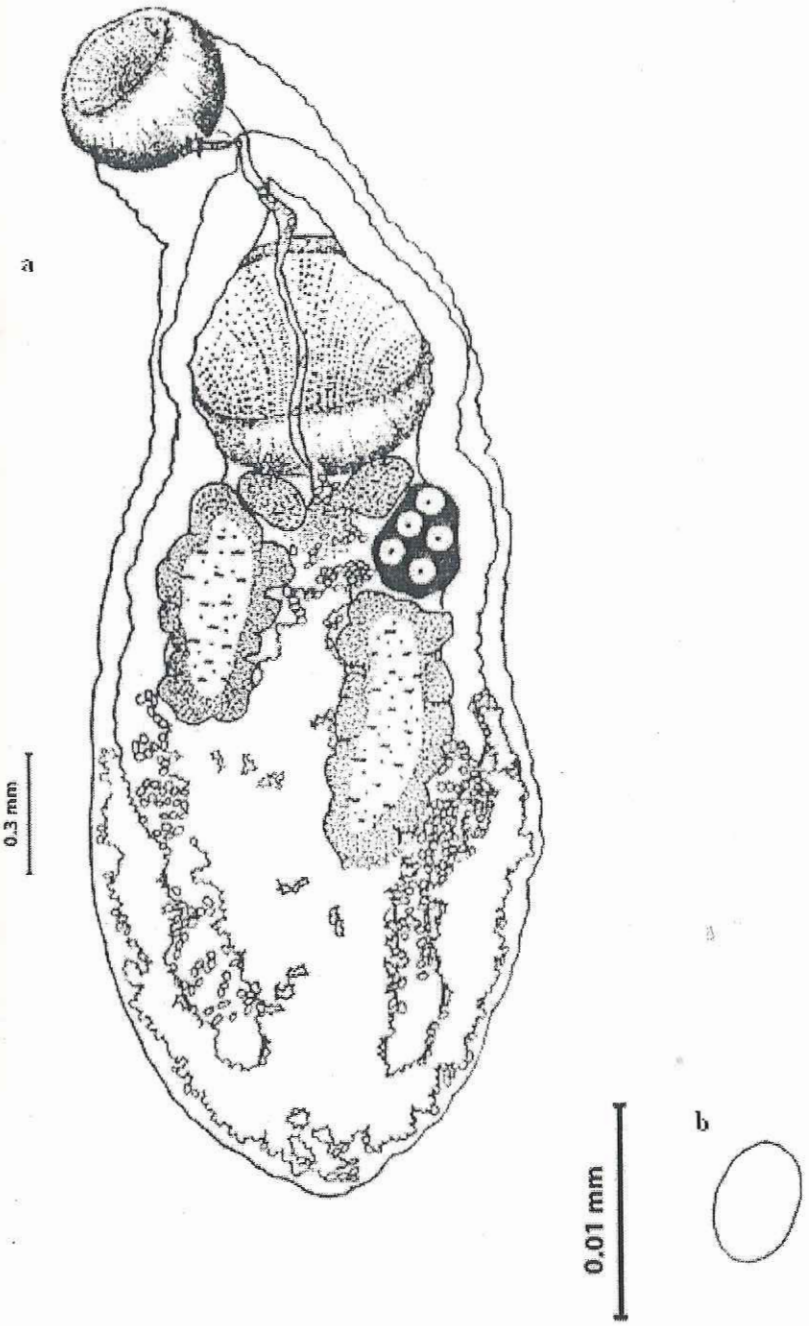
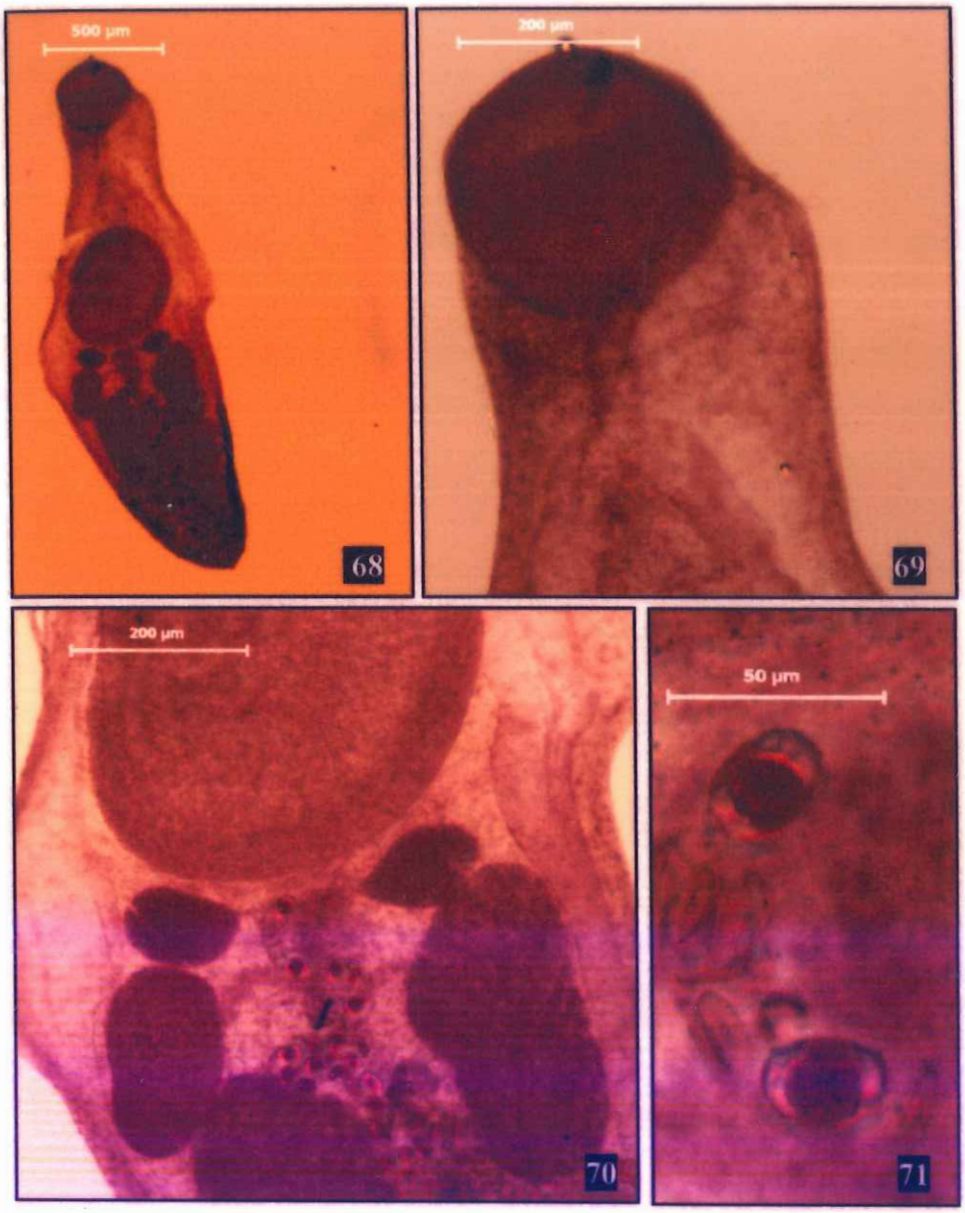


Fig. 67 *Gorgoderina ellipticum*

a. Whole mount, dorsal view

b. Egg



Figs. 68-71 ***Gorgoderin ellipticum***- Photomicrographs

68. Whole mount of the fluke
69. A view of the magnified anterior end
70. Magnified view of the mid section showing the lower rim of Ventral sucker, vitellaria, ovary, testes and the eggs
71. Eggs magnified

Gorgoderina sp.

(Figs. 72-76)

17 specimens of this form were recovered from *Amolops marmoratus* during the study period, the range of infection per infected host being 1-3.

Descriptions (based on measurements and observations of 8 mature specimens):

Body lanceolate, broadest in mid testicular region, tapering and terminating bluntly towards anterior end with much longer leaf-like/lanceolate posterior end from the ventral sucker; surface devoid of any spine, oral sucker terminal; ventral sucker pre-equatorial located in broader part of body, twice the size of oral sucker, covering half of body in breadth, strong and muscular. Pharynx absent and oesophagus very short; intestinal caeca terminating some distance from the posterior end. Testes elongated, laying in tandem in equatorial region, covering the broadest middle of body; posterior testis reaching anterior part of ovary, slightly larger in size than anterior; seminalis vesicula/cirrus sac large and reaching ventral sucker. Ovary rounded, in level with the anterior testis, post acetabular. Genital opening directly below oral sucker and above intestinal bifurcation. Uterus extensive, filling all available post-acetabular space. Vitellaria in two smooth compact masses, unlobed, posterior to ventral sucker, one on either side of median axis. Eggs small, oval, numerous.

Morphometric measurements of this form are given in Tables 8

Host: *Amolops marmoratus*

Location: Urinary bladder

Locality: Mokokchung, Tuensang

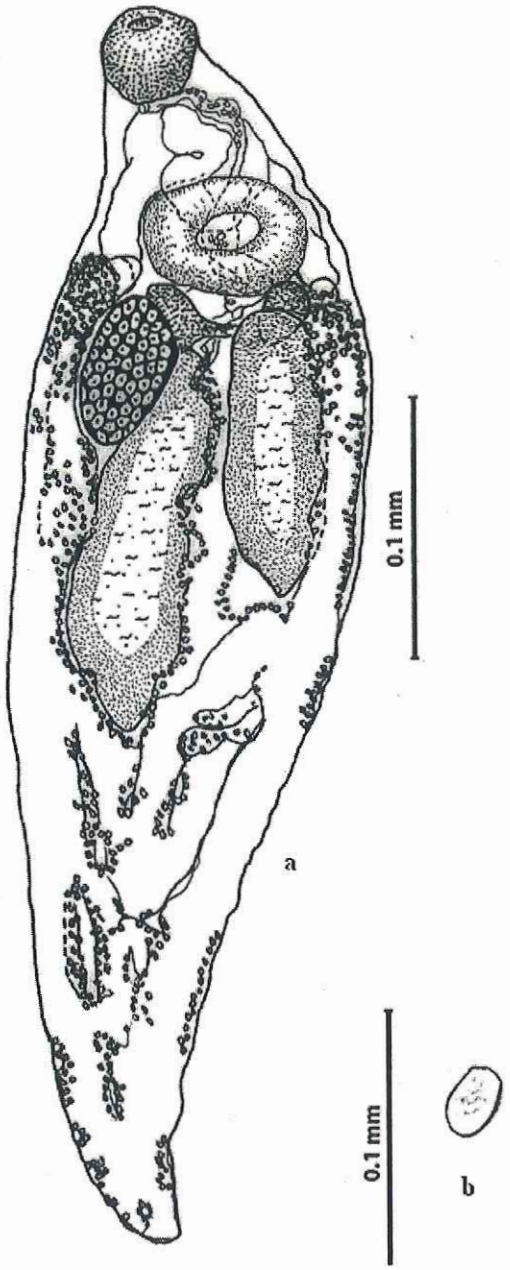
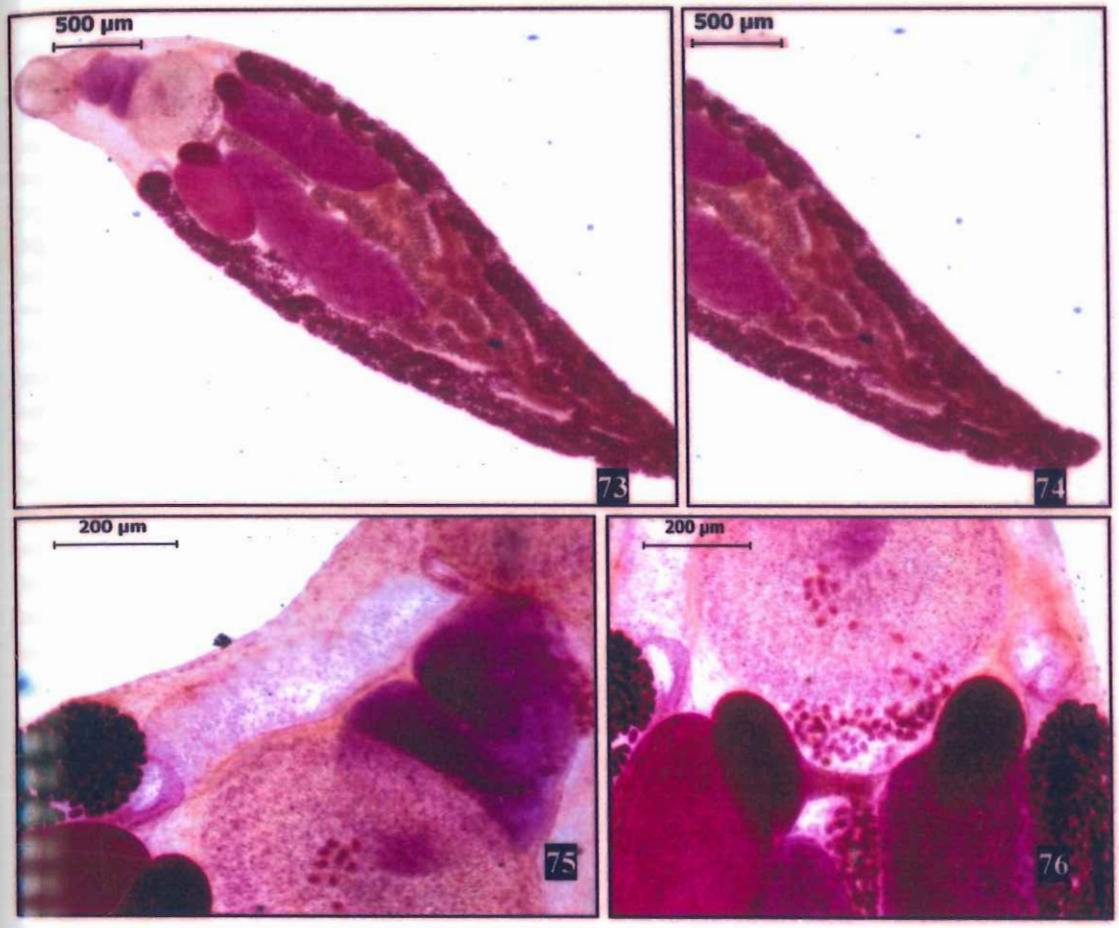


Fig. 72 *Gorgoderina* sp.

a. Whole fluke, ventral view

b. Egg



Figs. 73-76 *Gorgoderina* sp.- Photomicrographs

73. Whole fluke, ventral view

74. Posterior region

75. Magnified view of genital opening below the oral sucker, in top right; vesicula seminalis/cirrus sac reaching the anterior portion of the ventral sucker

76. A view of ovary, testes and vitellaria

Table 8 **Morphometric measurements of *Gorgoderina* sp.**

Characters		Range	Mean	S.D
Length of the body		3.818-4.669	4.2504	±0.3587
Maximum width of body		1.173-1.449	1.2972	±0.1414
Oral sucker:	Length	0.322-0.437	0.3956	±0.0442
	Breadth	0.322-0.437	0.3772	±0.0477
Ventral sucker:	Length	0.437-0.667	0.5244	±0.0897
	Breadth	0.575-0.667	0.6164	±0.0378
Oesophagu length		0.046-0.069	0.0506	±0.0103
Testes 1 (Right):	Length	0.897-1.058	0.9706	±0.0595
	Breadth	0.322-0.437	0.3818	±0.0448
Testes 2 (Left):	Length	0.989-1.426	1.1178	±0.1771
	Breadth	0.276-0.460	0.3910	±0.0780
Ovary:	Length	0.414-0.575	0.4738	±0.0621
	Breadth	0.253-0.368	0.3036	±0.0499
Cirrus sac:	Length	0.004-0.005	0.004	±0.2790
	Breadth	0.002-0.003	0.002	±0.1709
Vitellaria 1:	Length	0.230-0.276	0.2576	±0.0192
	Breadth	0.161-0.184	0.1656	±0.0103
Vitellaria 2:	Length	0.230-0.276	0.2438	±0.0206
	Breadth	0.161-0.184	0.1702	±0.0126
Distance of genital pore from anterior end		0.3220-0.5290	0.4232	±0.0839
Egg size:	Length	0.0276-0.0702	0.0370	±0.0186
	Breadth	0.0138-0.0432	0.0211	±0.0124

Discussions:

Originally the species was described from *E. cyanophlyctis* in Madhya Pradesh; the species was redescribed from Meghalaya Northeast, India by Diengdoh and Tandon, 1991 and Tandon *et al.*, 2001 from Nagaland (i.e., *Limnonectes limnocharis* and *E. cyanophlyctis* of Meghalaya and *E. cyanophlyctis* of Nagaland, respectively). The present form reported herein does not tally with and shows many variations from *E ellipticum*, on account of having a lanceolate shape, bigger size of body and in lacking a conspicuous pharynx and oesophagus. The paired and much larger unequal testes are smooth, the anterior smaller testis being in level with the ovary and the larger posteriorly placed testis are post ovarian in position. Vesicula seminalis/cirrus sac reaches the anterior part of the ventral sucker. Genital pore directly below the oral sucker and sub median. Ovary smooth, compact and roundish; vitellaria unlobed but in two smooth compact mass connected by a duct. In view of the apparent morphological differences observed in the present form and also different anuran host species for the same, it is considered a new species under the genus ***Gorgoderina***, the species identification is kept in abeyance for pending comparison with hitherto known type species.

Family Paramphistomidae Fishchoeder, 1901

Subfamily Diplodiscinae Cohn, 1904

Genus ***Diplodiscus*** Diesing, 1836

Diplodiscus amphichrus Tubangui, 1933

(Syn. ***D. sinicus*** Li, 1937; ***D. amphichrus magnus*** Srivastava, 1934; ***D. mehrai*** Pandey, 1975 nee Pande, 1937)

(Figs.77-85)

52 specimens of this species were recovered from 4 different host species during the study period: 24 from *R. maximus*, (the fluke intensity being 2-7 per infected host); 24 from *P. leucomystax*, the range being 1-5 per infected host, 3 from *Rana* sp.3 and 1 from *Chirixalus vitattus*.

Body size:	4.83-6.34 x 1.46-1.89mm
Egg size:	0.1mm in diameter

The fine topography of the fluke revealed by scanning electron microscopy (SEM) shows fine transverse foldings of the surface tegument. The latter at high resolution presents a finely beaded texture throughout. The oral rim as well as the inner wall of the buccal cavity which have fine radial and longitudinal tegumental foldings, respectively, are studded with button-like elevated papillae.

Host: *R. maximus, P. leucomystax, Rana* sp.3 and *C. vitattus*

Location: Intestine and rectum

Locality: Mokokchung

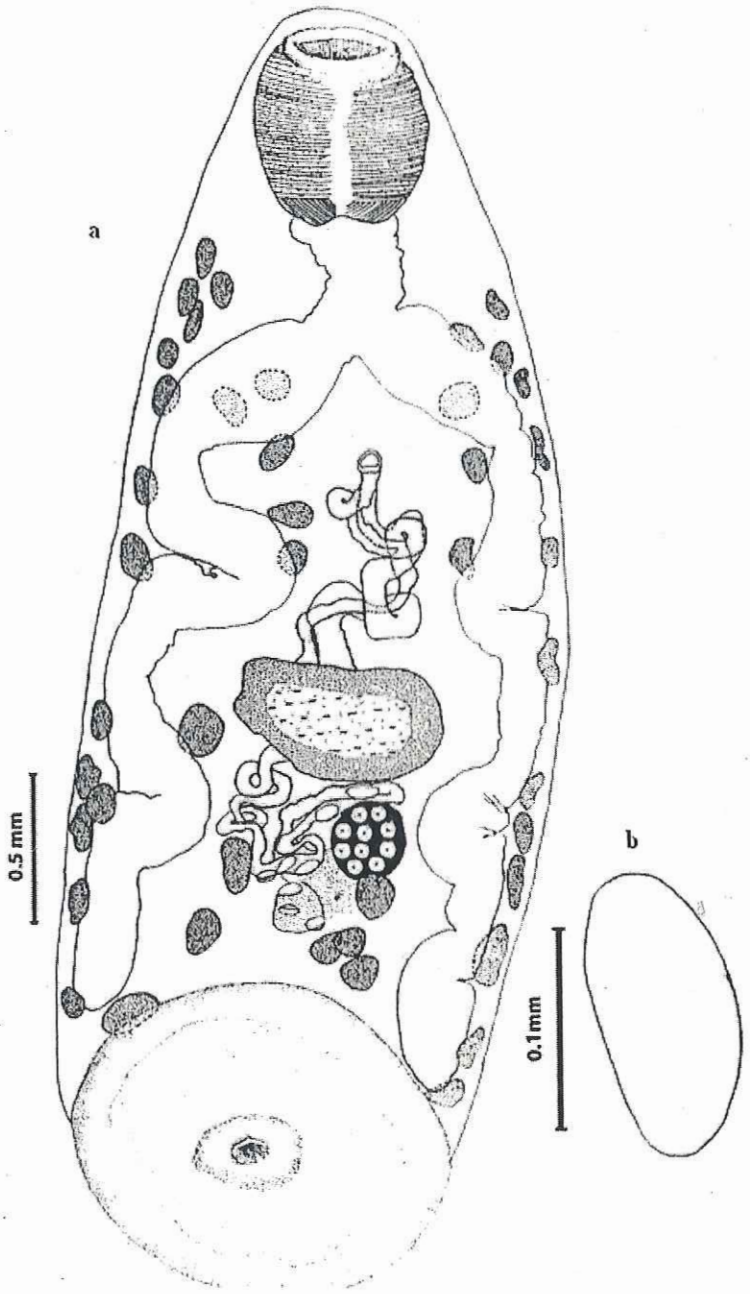
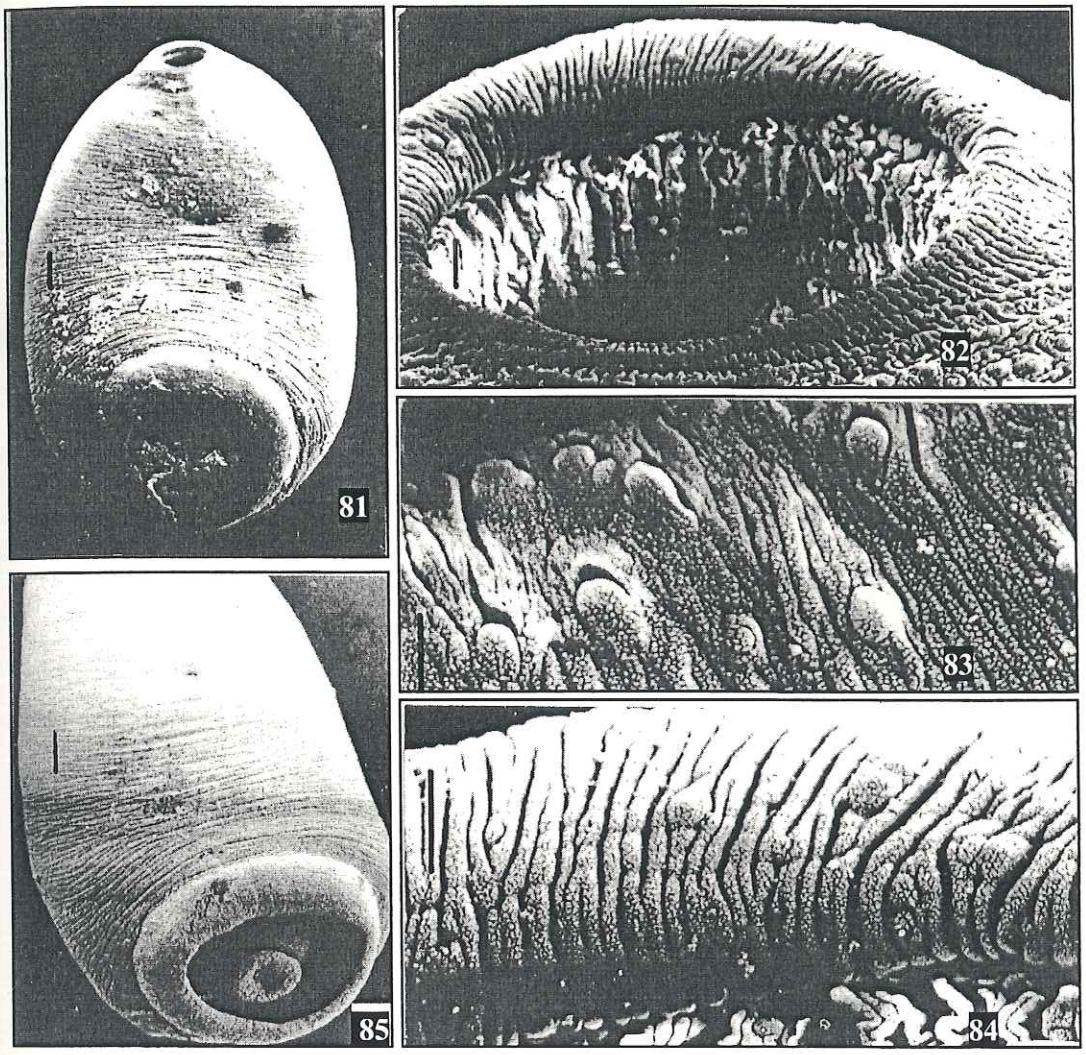


Fig. 77 ***Diplodiscus amphichrus***

a. Whole mount, dorsal view

b. Egg



Figs. 81-85 ***Diplodiscus amphichrus*** - Scanning electron micrographs

81. Full fluke, ventral view (scale bar= 100μm)

82. A magnified view of the oral sucker, showing the oral rim as well as the inner wall studded with button-like elevated papillae (arrows) (scale bar= 10μm)

83. Magnified view of the lower rim of the oral sucker, showing button like papillae (scale bar= 10μm)

84. Anterodorsal rim of the oral sucker under higher resolution, showing the radial foldings studded with button-like papillae and below it a part of inner wall of sucker (scale bar= 10μm)

85. Another specimen, showing the inner protuberance in the floor of the acetabulum (scale bar= 100μm)

Remarks:

D. amphichrus has been reported from many localities all across India such as Uttar Pradesh, West Bengal, Maharastra, Tamil Nadu, Kerala etc (Agarwal, 1966; Mukherjee and Ghosh, 1972; Nama and Khichi, 1973; Pandey, 1969; Singh, 1977). From North-East India, this species was first reported by Diengdoh (1989) in Meghalaya and by Tandon *et al.* (2001) in Nagaland from 2 hosts, *P. leucomystax* and *R. maximus*. The present study constitutes reports *Rana* sp.3 and *C. vitattus* as new host for this amphistomid fluke.

Diplodiscus mehrai Pande, 1937

(Syn. ***D. amphichrus*** (Tubangui, 1933) Singh, 1954

(Figs.86-94)

71 specimens of *D. mehrai* were recovered from *E. cyanophlyctis* from Kohima and Dimapur; *Rana* sp.1, *Haplobatrachus tigerinus* and *H.crassus* from Dimapur and *Limnonecte limnocharis* from Mokokchung in Nagaland.

Body size:	1.66-2.64 x 0.85-1.13mm
Egg size:	0.09-0.11 x 0.05-0.06mm

The general body surface which is aspinose has fine transverse folds of tegument both on dorsal and ventral sides and these folds acquire a concentric arrangement at the oral end. The circum-oral and circum-acetabular tegument, however, has radial folds. At higher resolution, the tegument of the acetabulum exhibits a texture of fine foldings with circular button-shaped papillae abounding near its rim. The tegument of the circum-genital pore region presents a cobble-stone-like texture, with large domed papillae aggregated at the pore rim. The rest of the body tegument appears to be devoid of papillae.

Host:	*E. cyanophlyctis*, *Rana* sp. 1, *H. tigerinus, H.crassus* and *L. limnocharis*
Location:	Rectum
Localities:	Mokokchung, Kohima, Dimapur

Remarks:

D. mehrai was earlier described from Almora (Kumoan hill) and Kashmir from *E. cyanophlyctis*, *Bufo viridis* and *Rana* species. It was reported for the first time from North-East India, by Tandon *et.al.* (2001) in Nagaland from *Rana* sp.1 and *E. cyanophlyctis*. The present study recorded another three hosts for ***D. mehrai***- *Haplobatrachus tigerinus* and *Haplobatrachus crassus* from Dimapur and *Limnonectes* sp. from Mokokchung.

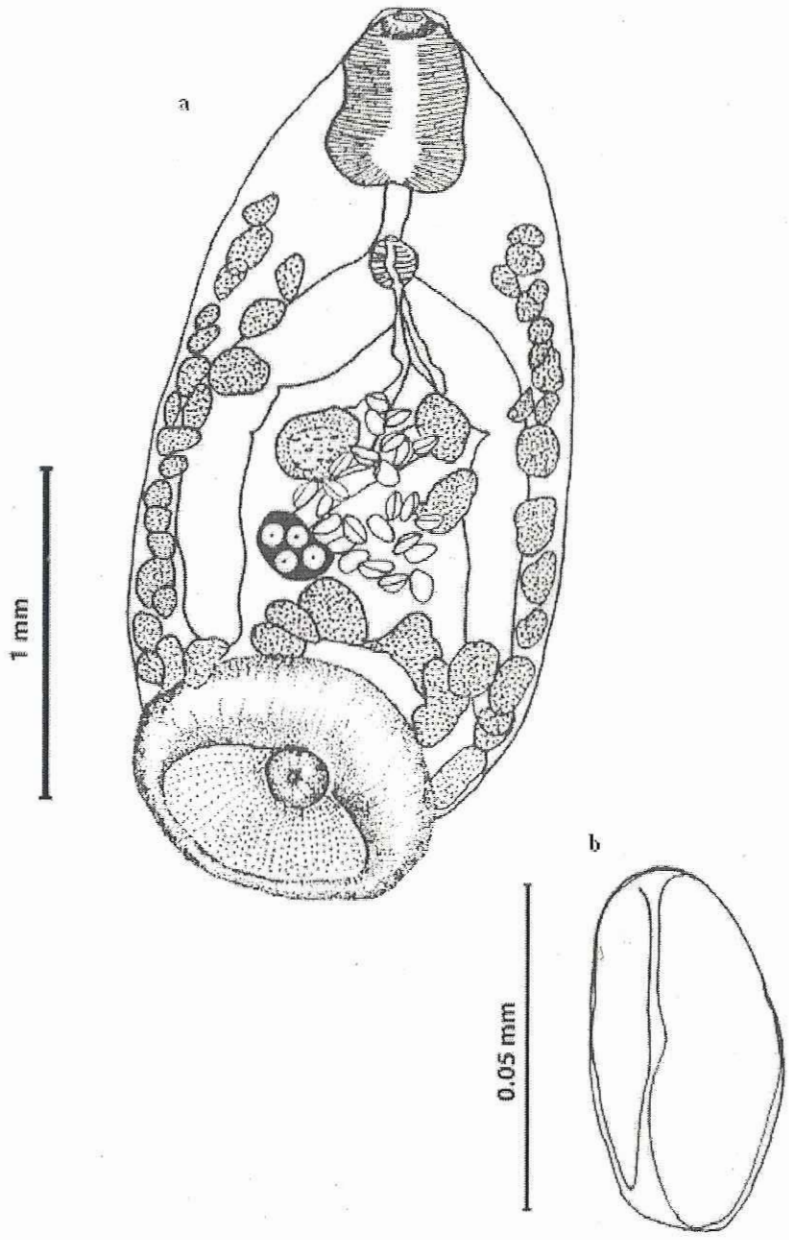


Fig. 86 ***Diplodiscus mehrai***

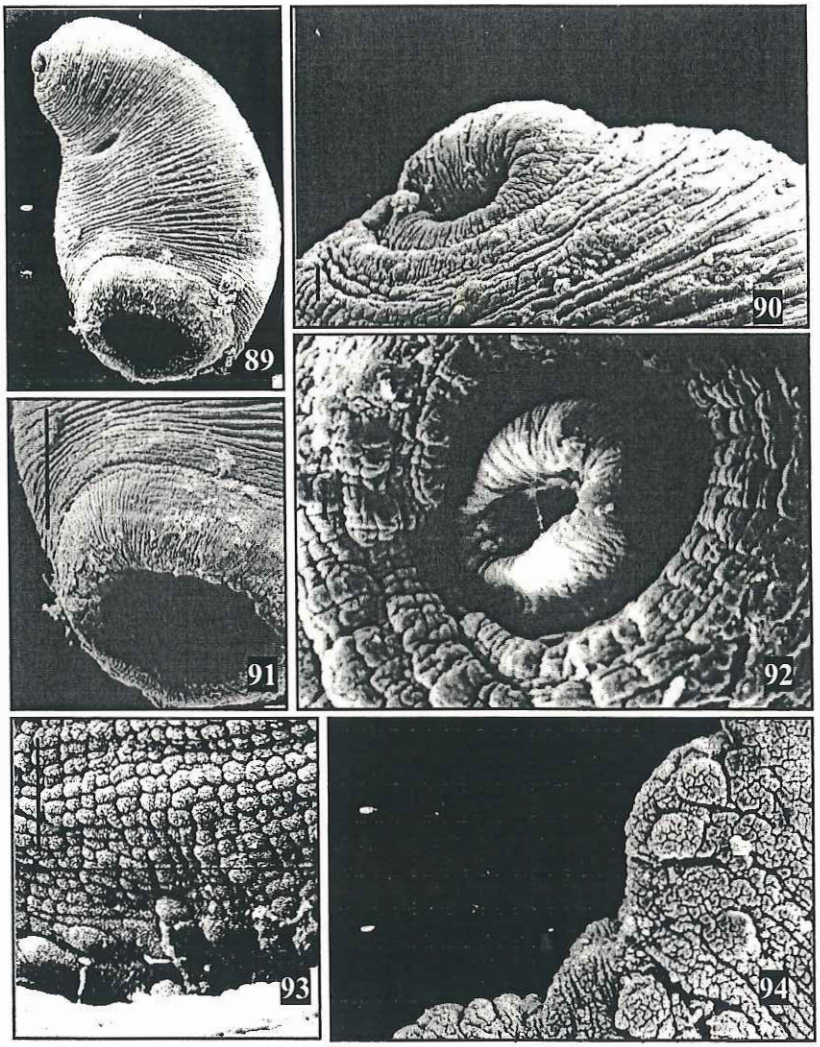
a. Whole fluke, ventral

b. Egg

Figs.87- 88 ***Diplodiscus mehrai***- Photomicrographs

87. A view of whole fluke with anterior rim of the acetabulum

88. Egg



Figs. 89-94 ***Diplodiscus mehrai*-** Scanning electron micrographs

89. Full fluke, ventrolateral view (scale bar= 100μm)

90. A magnified view of the anterior region, showing the oral sucker and foldings in the body tegument (scale bar= 10μm)

91. A magnified view of the posterior region, showing the acetabulum (scale bar= 100μm)

92. An enface view of the oral sucker and anterior end (scale bar= 10μm)

93. A magnified view of the genital pore region, showing the cobble-stone-like texture of the tegument and the domed papillae in the upper inner rim of the pore (scale bar= 10μm)

94. The acetabular rim in a closer view, showing the foldings with circular button-like papillae (arrows) (scale bar= 10μm)

Family Hemiuridae Luhe, 1901

Superfamily Halipeginae Ejsmont, 1931

Genus ***Halipegus*** Looss, 1899

Halipegus mehransis Srivastava, 1933

(Syn. ***H. mehransis minutus*** Srivastava, 1933; ***H. spindale*** Srivastava, 1933)

(Fig. 95)

Only 12 specimens of this species were collected in the study period from 3 hosts from Dimapur

Body size: 3.98-4.24 x 1.17-1.19mm

Egg size: .42-4.25 x 14 mm

Host: *E. cyanophlyctis*

Location: Stomach

Locality: Dimapur

Remarks:

Originally, ***Halipegus mehransis*** described from *E. cyanophlyctis* (=*Rana cyanophlyctis*) by Srivastava (1933), this species was reported from Shillong (Meghalaya) by Diengdoh (1989). Other locality records of this species include Rajasthan (Gupta, 1970), Maharastra (Mukherjee and Ghosh, 1972), Bihar (Sinha *et al.* 1974), Tamil Nadu and Kerala (Sinha, 1977), Meghalaya (Diengdoh, 1989) and Nagaland (Tandon, *et al.* 2001) from *E. cyanophlyctis*/ *H. tigerinus*. Besides these anuran amphibian hosts, the species has also been reported from a reptilian host in Andhra Pradesh (Sinha, 1958). This fluke was not encountered in other frog species in the present study.

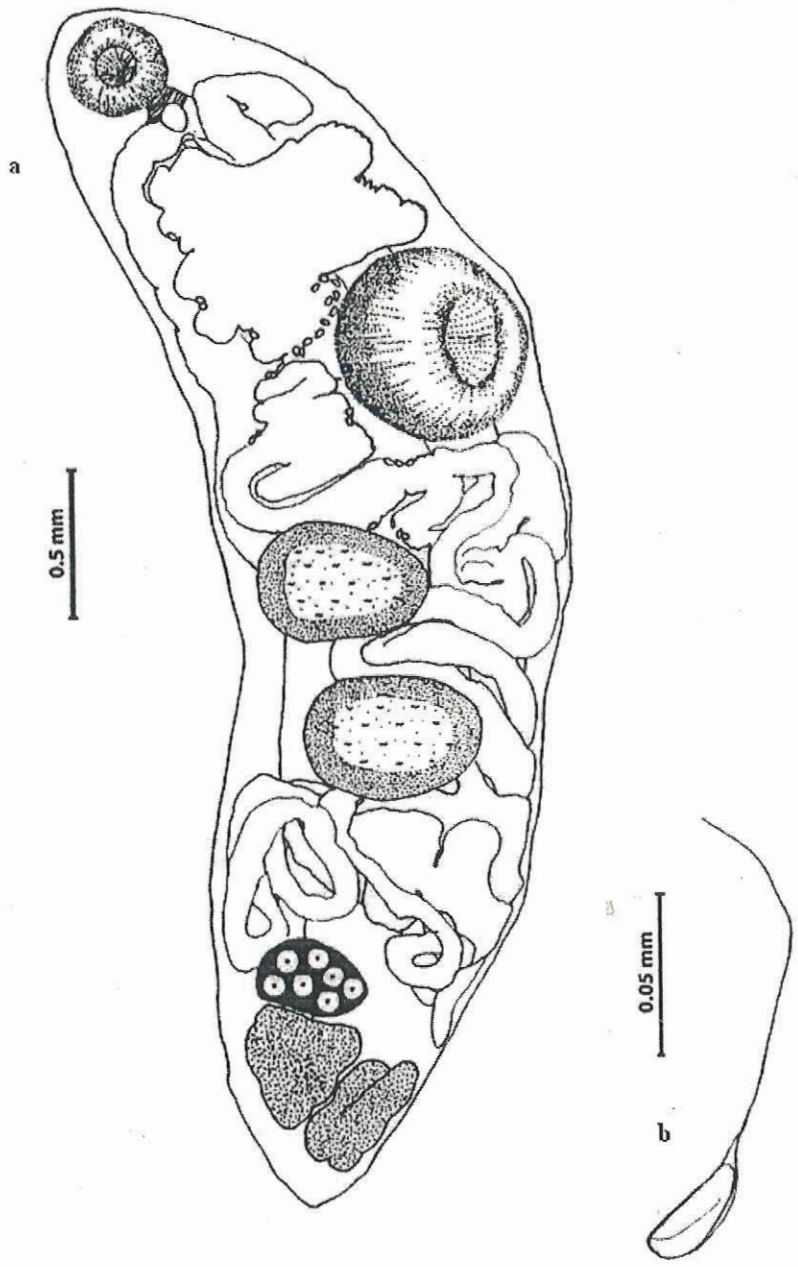


Fig. 95 ***Halipegus mahransis***

a. Whole mount, ventral view
b. Egg

Family Brachycoelidae (Looss, 1899) Johnston, 1912

Subfamily Mesocoeliinae (Faust, 1924) Dollfuss, 1929

Genus ***Mesocoelium*** Odhner, 1910

Mesocoelium monas (Rudolphi, 1819) Freitas, 1958

(Syn. ***Distoma monas*** Rudolphi, 1819; ***D. sociale*** Luhe, 1901; ***M. mesembrinum*** Johnston, 1912; ***M. oligoon***, Johnston, 1912; ***M. micron*** Nicolis, 1914; ***M. carli*** Andre, 1915; ***M. incognitum*** Travassos, 1921; ***M. megittii*** Bhalerao, 1927; ***M. monadi*** Dollfuss, 1929; ***M. americanum*** Harwood, 1932; ***M. burti*** Fernando, 1933; ***M. marral***, Fernando, 1933; ***M. leiperi*** Bhalerao, 1936; ***M. waltoni*** Pereira *et* Cuocolo, 1940; ***M. travassosi*** Pereira *et* Cuocolo, 1940; ***Mesocoelium*** sp. Travassos *et* Freitas, 1942; ***Mesocoelium*** sp. Travassos, 1945; ***M. schwetzi*** Dollfuss, 1950; ***M. marcoccanum*** Dollfuss,1951; ***M. geogesblanci*** Dollfuss, 1945; ***M. macrebense*** Dollfuss, 1954; ***M. brachyenteron*** Dollfuss, 1954; ***Mesocoelium*** sp. Ucros, 1959; ***M. sociale*** Skrjabin, 1964).

(Figs. 96-105)

Altogether 137 specimens were collected during the study period, the range of infection being 1-26.

Body size: 2-2.57 x 0.66-0.84mm

Egg size: 0.035-0.038 x 0.021-0.023mm

Host: *Polypedates leucomystax*, *P. taraiensis*, *Philautus annandalii*, *Hyla annectans*, *Rana livida*, *Euphlyctis cyanophlyctis*, *Haplobatrachus tigerinus* and *Amolops marmoratus*.

Location: Intestine

Locality: Kohima, Mokokchung, Tuensang, Dimapur

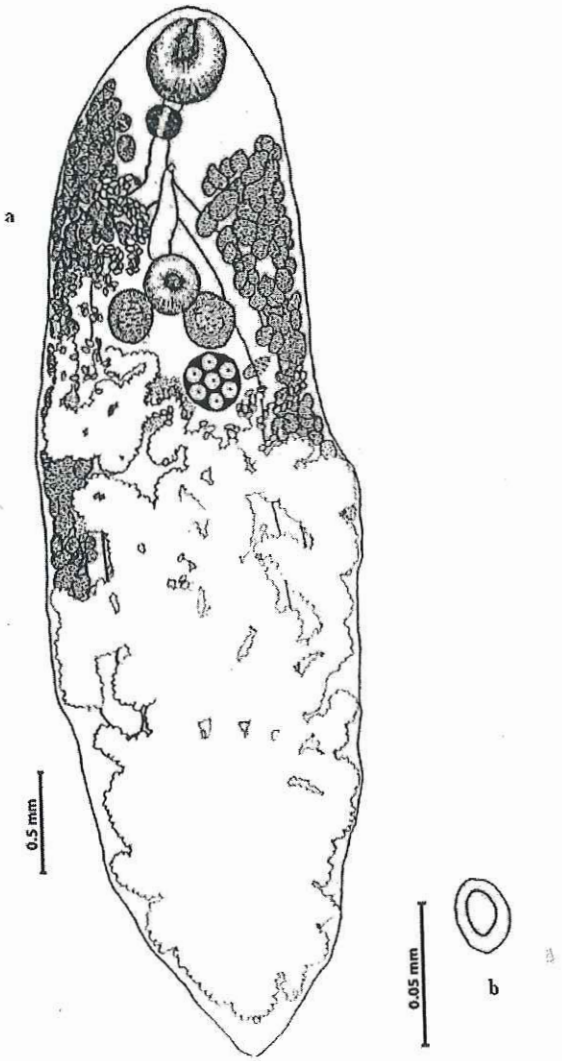
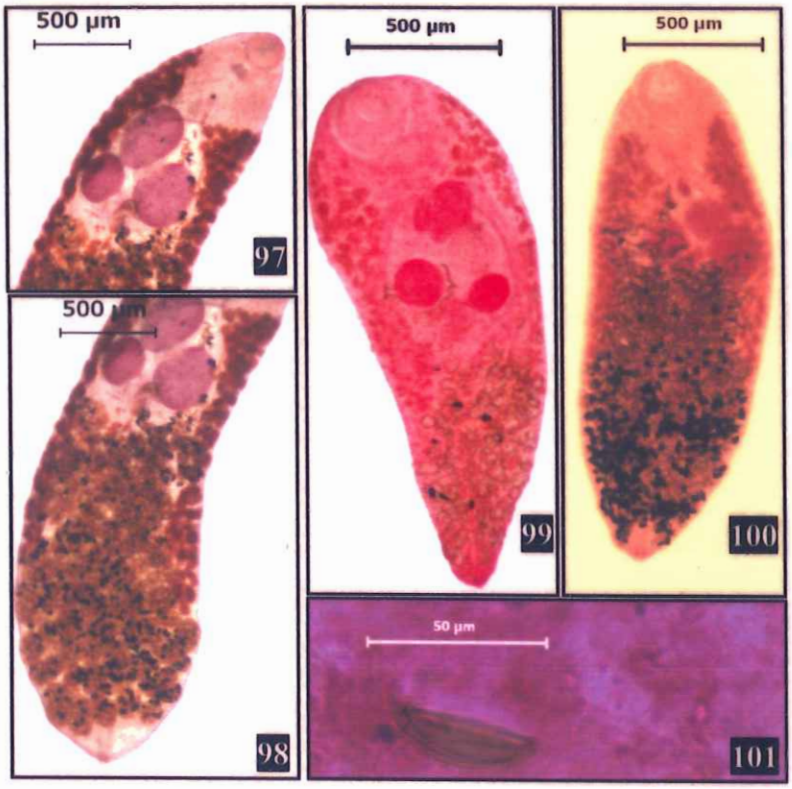


Fig. 96 ***Mesocoelium monas***

a. Whole mount, dorsal view

b. Egg



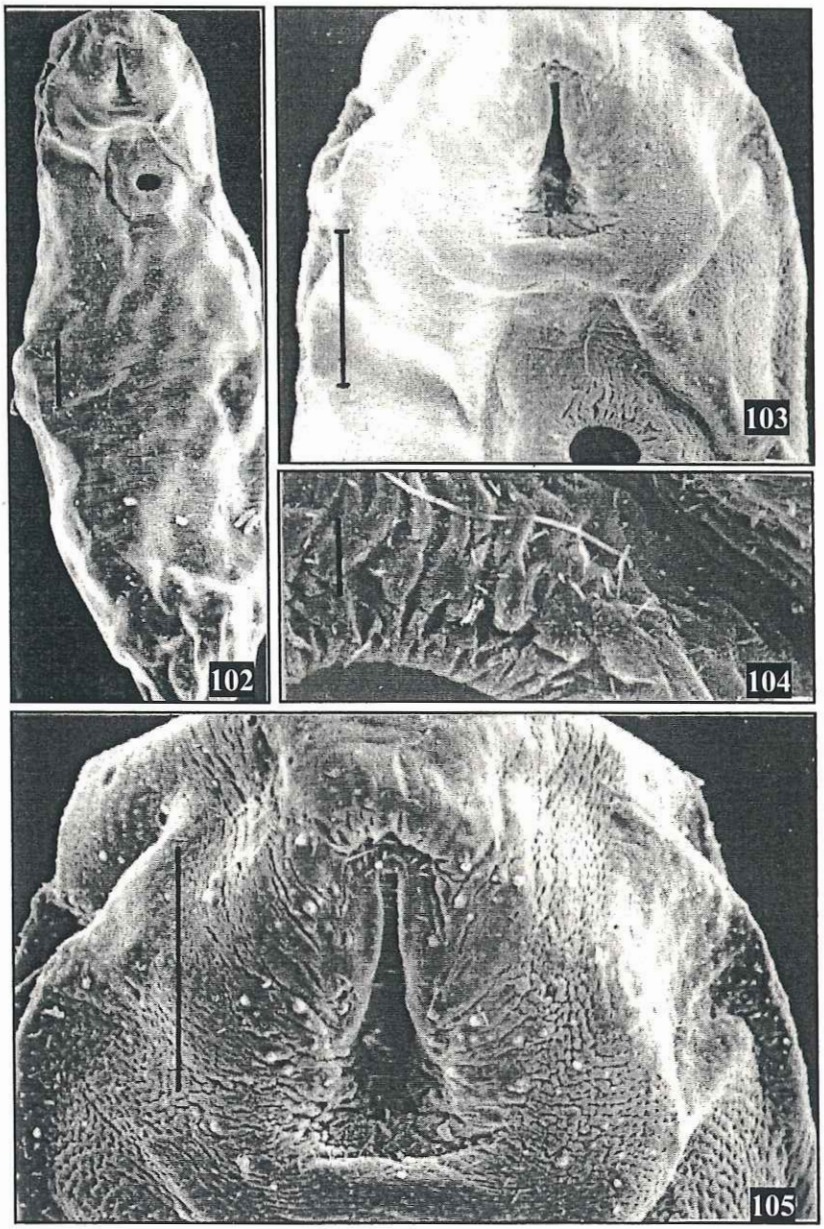
Figs. 97- 101 ***Mesocoelium monas***- Photomicrographs

97. Anterior end, dorsal view

98. Posterior end of the same fluke

99, 100. Ventral and dorsal view, respectively, of different specimens from different host species.

101. Egg enlarged



Figs. 102-105 ***Mesocoelium monas***- Scanning electron micrographs

102. Whole fluke, ventral view (scale bar= 100μm)

103. Anterior region in higher resolution, showing the oral and ventral sucker (scale bar= 100μm)

104. A portion of the rim of the ventral sucker, showing the radial folding (scale bar= 10μm)

105. Oral sucker under higher resolution, showing the papillae in the circum-oral region (scale bar= 100μm)

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Remarks:

Earlier the species ***Mesocoelium*** have been reported from different localities and a variety of amphibian and reptilian hosts in the Indian subcontinent or erstwhile British India. Sewell (1920), Lühe (1901), Odhner (1910) described ***M. sociale*** from *Bufo melanostictus* in Calcutta; in Burdwan, West Bengal (Mukherjee and Ghosh, 1972), Kerala and Tamil Nadu (Singh, 1977). ***M. meggitti,*** which was described by Bhalerao (1927) from lizards from Burma (Myannmar) was also reported to occur in toads and frogs (Meggit, 1972; Bhalerao, 1936 a, b, c). Diengdoh (1989) and Tandon *et.al.* (2001) reported ***M. monas*** from *Polypedates leucomystax* from and Nagaland, respectively. Other species described in the genus ***Mesocoelium*** from India include ***M. varunae*** Baugh, 1956 from *B. melanostictus* and ***M. thapari*** Gupta and Jahan (1976) from *Haplobatrachus tigerinus*.

The anuran species i.e. *P. tardensis, Philautus annandalii, Hyla annectans, Rana livida, Euphlyctis cyanophlyctis*, *Amolops marmoratus* are new hosts record from the Indian subcontinent and Dimapur forms a new locality record for ***Mesocoelium monas***.

Family Lecithodendridae (Lühe, 1901) Odhner, 1910

Subfamily Opisthioparorchinae

Genus *Opisthioparorchis* Wang, 1980

Opisthioparorchis indica Tandon, Imkongwapang, *et* Prasad, 2005

(Figs.106-115)

During a survey of amphibian hosts, 38 specimens of a lecithodendriid fluke were recovered from the intestine of *Amolops marmoratus* (=*Amolops afghanus*) from Mokokchung. On a later occasion, similar specimens 38, in numbers, were again recovered from the same host and locality. All these specimens were found new to science and described herein as a new species of the genus ***Opisthioparorchis***.

Description (based on measurements of 10 mature flukes and SEM observations on 2 specimens):

Body elongate or oval with rounded anterior end, rounded or slightly conical posterior end, spinose. Oral sucker subterminal, spherical. Ventral sucker pre-equatorial or equatorial almost equal to oral sucker in size. Prepharynx indistinct; pharynx muscular; oesophagus of moderate length; intestinal caeca extending in postacetabular region up to level of anterior margin of testes. Testes round or oval, lying symmetrically juxtaposed in posterior region of body; cirrus sac well developed, elongated, somewhat S-shaped, recurved, lying on left side of body, extending from near junction of oesophagus and intestinal caeca up to anterior margin of oral sucker. Ovary rounded, pretesticular, postacetabular; receptaculum seminis conspicuous, lying posterior to ovary; uterus pretesticular, occupying major area between ovary and testes, extending anteriad partially overlapping intestinal caeca. Genital pore marginal on left at level of anterior margin of oral sucker. Vitellaria in varying follicular size, extending in lateral fields from level of pharynx posteriorly up to post-testicular region, becoming confluent medially in intercaecal, pretesticular and post testicular fields. Eggs numerous, ovoid, operculate.

SEM revealed the surface fine topography of the fluke. The body surface is studded throughout with dense spination except for the anterior circum-oral region, which appears devoid of spines. Whereas the spines covering the tegument in most parts of the body are

scale like with rounded tips, those abounding in the posterior region are conical, with broad base and pointedly tapering distal end. The non-spinous tegument of the anterior region presents a spongy texture.

The morphometric measurements of this form are given in Table 9

*Ref. Tandon V., Imkongwapang R. and Prasad P. K. (2005). On two new species of the trematode genera, *Batrachotrema* Dolfus et Williams, 1966 and *Opisthioparorchi* Wang, 1980 (Batrachotrematidae), with a report of a Chinese species of *Opisthioparorchis* from anuran Amphibia hosts in India. **Zoos' Print Journal Vol. 20,** No. 6 pp:1883-1887

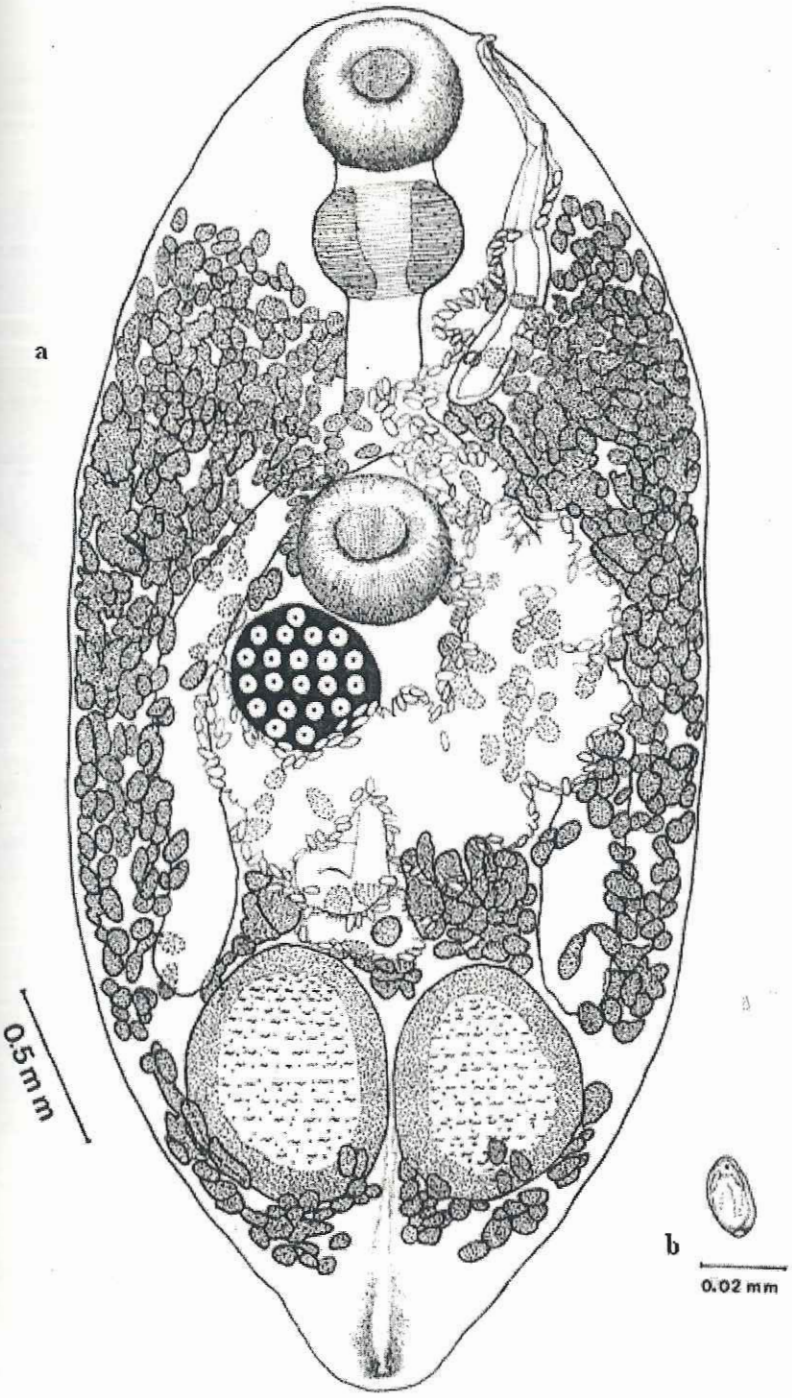
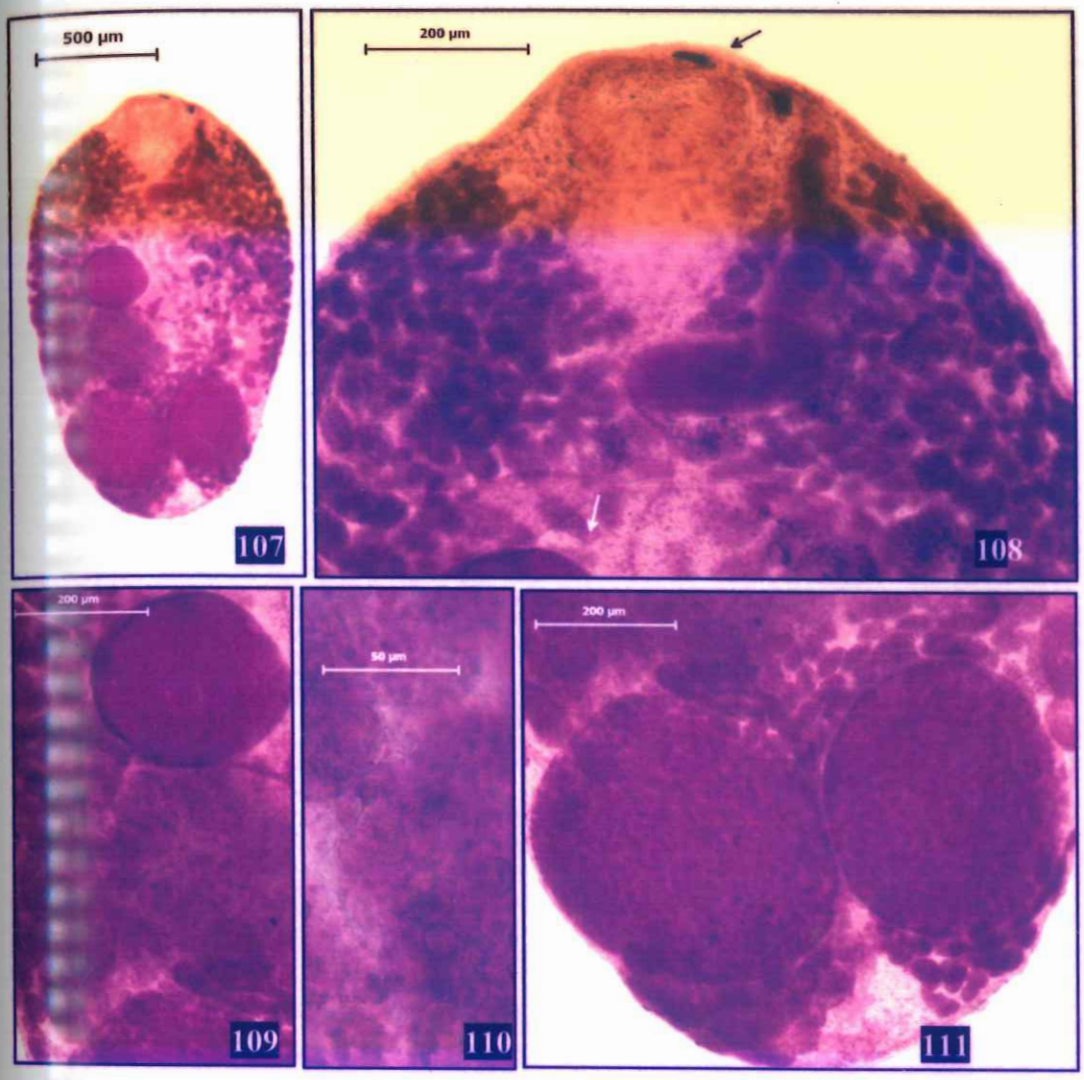


Fig.106a. ***Opisthioparorchis indica*** Tandon, Imkongwapang, *et* Prasad, 2005

a. Whole mount, ventral view

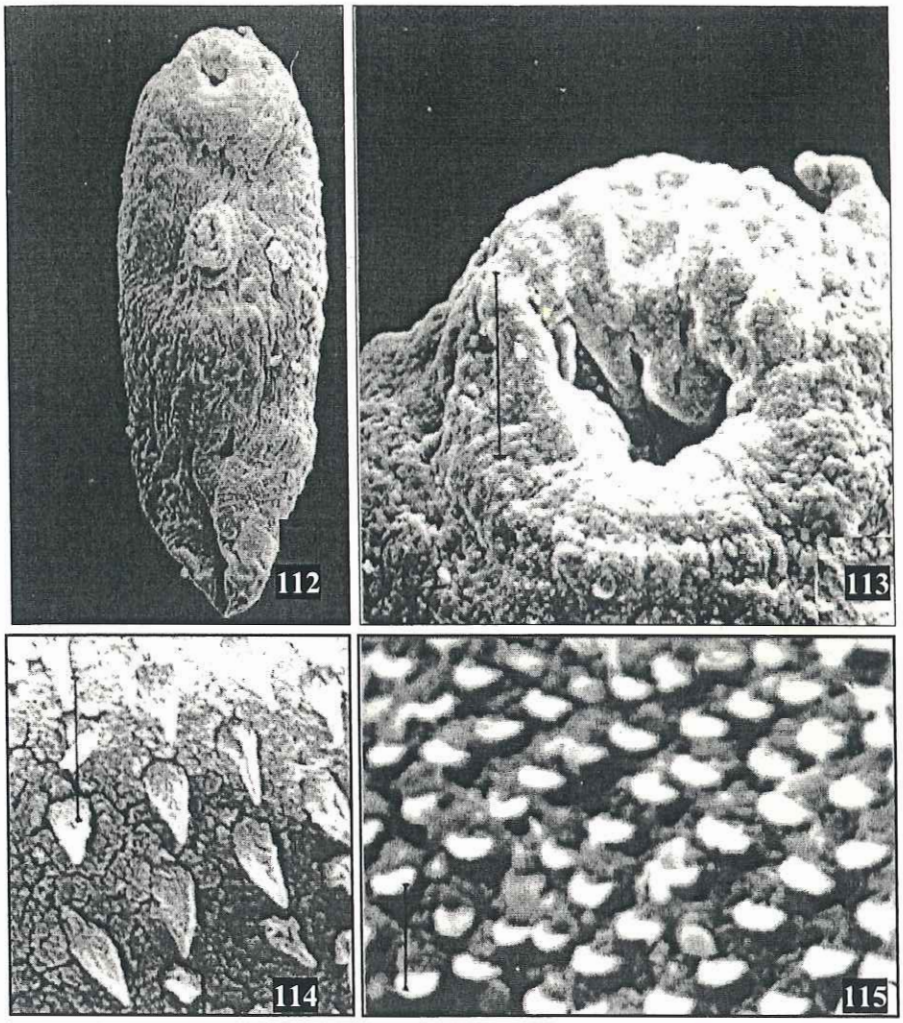
b. Egg



Figs.105-111 ***Opisthioparorchis indica*** Tandon, Imkongwapang, *et* Prasad, 2005

-Photomicrographs

107. Whole worm ventral view
108. Anterior region showing the genital pore (black arrow) and the ovary (white arrow)
109. A magnified view of the ovary and vitellaria
110. Eggs
111. Posterior end showing the testes



Figs. 112-115 ***Opisthioparorchis indica*** Tandon, Imkongwapang, *et* Prasad, 2005

-Scanning electron micrographs

112. Whole worm, ventral view (scale bar = 100 μm).

113. Oral end, magnified view (scale bar =10 μm).

114. A magnified view of the tegumental spination in the mid ventral region (scale bar =10 μm).

115. Tegument of the posterior part of the body depicting conical spines (scale bar =10 μm).

Table 9 **Measurements of *Opisthioparorchis indica*** Tandon, Imkongwapang, *et* Prasad, 2005

Characters		**Range (Mean)**	**± S.D.**
Length of Body		1.80-2.33 (2.05)	±0.18
Maximum width of body (at level of ventral sucker)		0.89-1.08 (0.97)	±0.08
Oral sucker:	Length	0.21-0.26 (0.24)	±0.02
	Breadth	0.23-0.30 (0.26)	±0.026
Ventral Sucker:	Length	0.21-0.25 (0.23)	±0.014
	Breadth	0.21-0.27 (0.24)	±0.018
Pharynx:	Length	0.19-0.235 (0.21)	±0.018
	Breadth	0.2-0.24	±0.016
Length of Oesophagus		0.14-0.23 (0.18)	±0.03
Distance of intestinal caeca from hind end		0.58-0.77 (0.68)	±0.06
Intestinal bifurcation from anterior end		0.53-0.65 (0.61)	±0.04
Testis (right)	Length	0.33-0.46 (0.39)	±0.04
	Breadth	0.26-0.33 (0.29)	±0.023
Testis (left)	Length	0.33-0.41 (0.38)	±0.03
	Breadth	0.25-0.32 (0.28)	±0.029
Cirrus sac	Length	0.52-0.66 (0.57)	±0.05
Ovary	Length	0.17-0.25 (0.22)	±0.027
	Breadth	0.17-0.23 (0.20)	±0.022
Receptaculum seminis	Length	0.36-0.53 (0.44)	±0.07
	Breadth	0.03-0.065 (0.047)	±0.013
Eggs		0.027-0.037 x 0.011-0.018 (0.031 x 0.015)	±0.002

Discussion:

In having a well-developed cirrus sac and marginal or sub-marginal genital pore located in the region of the oral sucker or pharynx, the present form belongs to the family Opecoeliidae/Batrachotrematidae.

The genus ***Opisthioparorchis*** was created by Wang (1980) for an intestinal fluke of *Rana spinosa,* which was characterized by having juxtaposed testes located at the posterior end of the body and intestinal caeca extending up to just near the anterior border of testes. At present the genus includes six species all described from China. *O. ranae* Wang, 1980 (type species) from *Rana spinosa* in Fujian Province South; ***O. pleurogenitus*** Wang, 1980 from the same host and locality as those of the type species; *O. boheansis* Wang, 1980 from *Staurois wuyiensis* also from Fujian; ***O. megaloonis*** Liang et Ke, 1988 and ***O. meixianensis*** Liang et Ke, 1988- both from *R. spinosa* in Changsha (Meixian, Guangdong Province) and ***O. yunnanse*** Li, 1996 also from *R. spinosa* from Yunnan Province.

In having the vitellaria extending in the post testicular region, the present form resembles the type and other species but stands apart from ***O. pleurogenitus*** and ***O. yunnanse,*** in both of which the vitellarial follicles are distributed only in the particular region and the testes are the posterior most structures in location in the fluke body. It also differs from all those species having post testicular vitellaria in several characters; the latter species, have a much smaller body size, the ventral sucker is smaller than the oral and unlike the present form, the vitellaria do not become confluent medially in the pretesticular, inter-intestinal area and posttesticular region.

In view of the conspicuous differences of the fluke with the hitherto known species of ***Opisthioparorchis***, it was consider as a new species and named as ***O. indica.***

Specific Diagnosis:

Intestinal caeca extending much beyond ventral sucker up to anterior level of testes, ventral sucker almost equal to oral sucker in size, genital pore in level with anterior margin of oral sucker, vitellaria extending beyond testes posteriorly, confluent medially in pre-and posttesticular regions.

Type Host: *Amolops marmoratus* Blyth 1855 (=*Amolops afghanus*)

Type Location: Intestine

Type Locality: Mokokchung (Nagaland, 26°11'45.21'N and 94°17'33.04"E to-26°45'46.74"N and 94°45'52.49", India)

Etymology: Named after the country.

Holotype No: W 8341/1; Paratype No: W 8342/1, 8343/1

Deposited in Zoological Survey of India, Kolkata 700 053, India

Opisthioparorchis yunnanse Li, 1996

(Fig. 116)

Description (Whole mount):

Body elongate, somewhat narrow anteriorly, broader posteriorly, with bluntly rounded ends, spinose. Oral sucker subterminal, almost twice as large as ventral sucker, latter equatorial in position. Prepharynx short, pharynx muscular; oesophagus short; intestinal caeca extending posterial up to a little in front of testes. Testes round or oval, juxtaposed near posterior extremity of body; cirrus sac club-shaped, elongated, extending from a little in front of ventral sucker anteriad up to level of oral sucker. Ovary rounded, just by side or slightly posterior to ventral sucker; uterus mainly in near post ovarian region, extending as ascending narrow tube anteriad. Genital pore marginal, on left at level of oral sucker. Vitellaria extending extensively from level of oesophagus posteriad up to just in front of testes. Eggs elliptical, operculate.

Host: *Paa mokokchungensis* (=*Rana liebigii*)

Location: Intestine

Locality: Kohima, Nagaland (25°30'13.86"N and 93°55'13.86"E to 26°02'8.44" N and 94°22'0.36" and 94°22'0.36" E, India)

Remarks:

The whole mount preparation of one specimen collected from *Rana liebigii*, though apparently resembled *Opisthioparorchis* species, turned out to be different from the new species of the genus described herein. In its general morphology (elongated body, oral sucker larger than the ventral sucker(almost double the size), vitellaria limited to only pretesticular zone and long club-shaped cirrus sac), the present specimens stands close to *O. yunnanse* Li, 1996, which was originally described from *Rana spinosa* from Yunnan Province, China. However, the only conspicuous deviation it shows from this species is the length of the oesophagus; in *O. yunnanse* the oesophagus is quite long, though short in the present form. However, considering that just one character i.e. length of the oesophagus, is not enough to erect a new species, the present form is considered representing *O. yunnanse*

Table 10 **Measurements of *Opisthioparorchis yunnanse*** Li, 1996

Characters		**Measurements**
Length of Body		2.5
Maximum width of body (at level of ventral sucker)		1.94
Oral sucker:	Length Breadth	0.28 0.29
Ventral Sucker:	Length Breadth	0.08 0.08
Pharynx:	Length Breadth	0.18 0.2
Length of Oesophagus		0.09
Distance of intestinal caeca from hind end		0.58
Intestinal bifurcation from anterior end		0.6
Testis (right)	Length Breadth	0.37 0.35
Testis (left)	Length Breadth	0.38 0.31
Cirrus sac	Length	0.97
Ovary	Length Breadth	0.235 0.23
Receptaculum seminis	Length Breadth	0.42 0.047
Eggs	Length Breadth	0.041 0.021

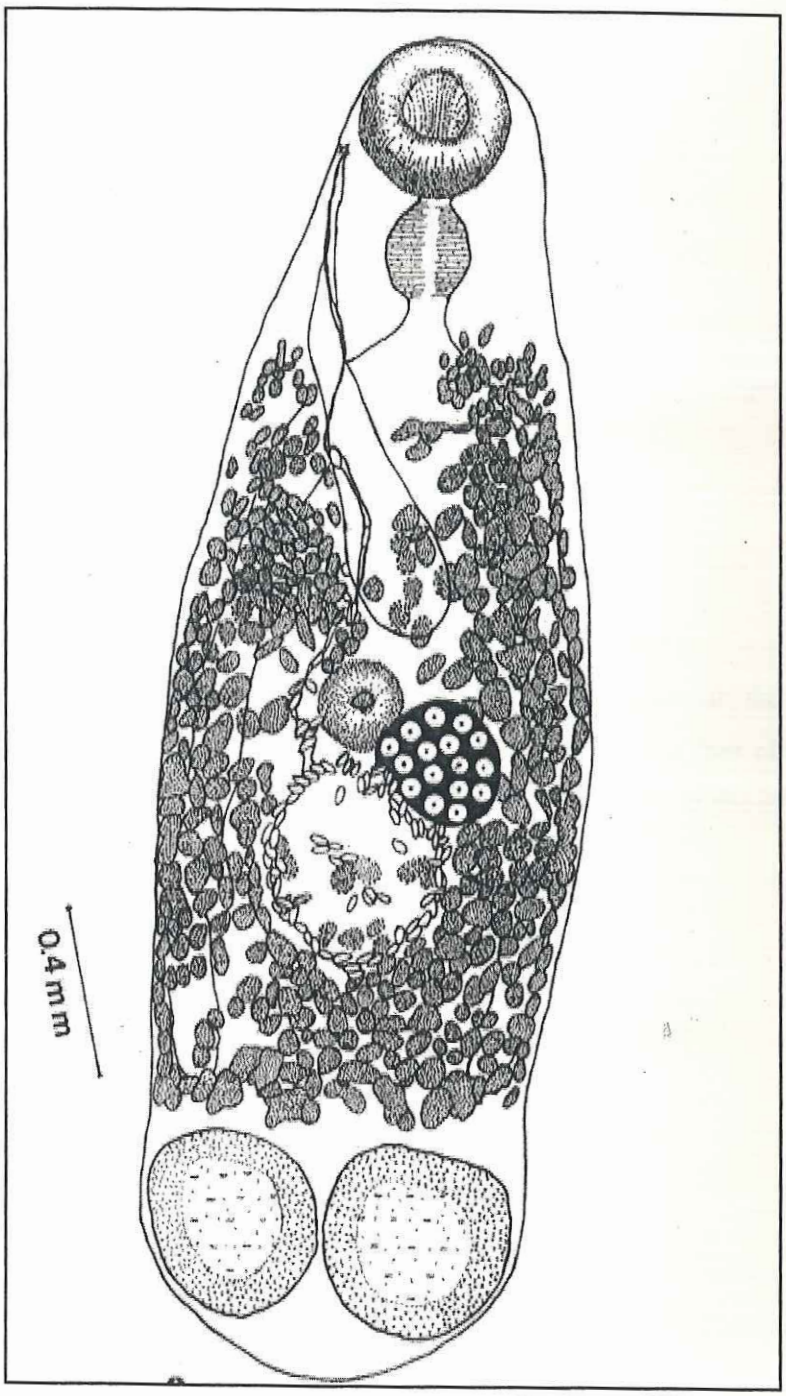


Fig.116 ***Opisthioparorchis yunnanse*** Whole worm, dorsal view

Subfamily Prosotocinae Yamaguti, 1959

Genus ***Mehraorchis*** Srivastava, 1933

Mehraorchis ranarum

(Figs.117-124)

The present form was recovered from the gall bladder of *Haplobatrachus tigerinus* in Dimapur and altogether 44 specimens were collected during the study period

Body size: 2.41-3.10 x 1.77-2.23mm

Eggs size: 0.03 x 0.012-0.016mm

The fine surface topography SEM revealed spination all over the body tegument; the spines, crystalline in appearance limited around the sub-terminal oral sucker leaving free of any, in both the suckers; the posterior extremity also showed lack of spination around the excretory pore.

Host: *H. tigerinus*

Location: Gall and bile duct

Locality: Dimapur

Remarks:

Originally described by Mukherjee and Ghosh, 1970, in West Bengal from the anuran host *H. tigerinus*. It was also reported from Nagaland, a new locality record, Dimapur (Tandon, *et al.*, 2001). In the present study no new anuran host was encountered other than the same anuran species for ***M. ranarum.***

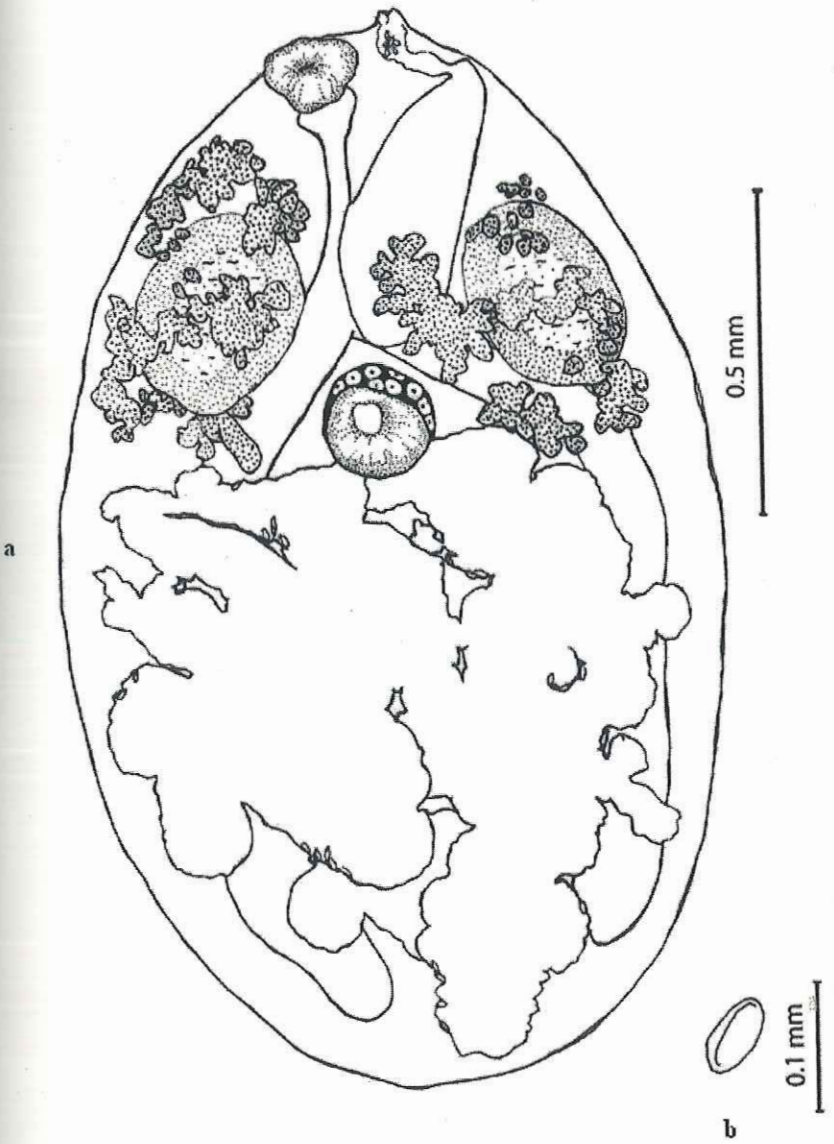
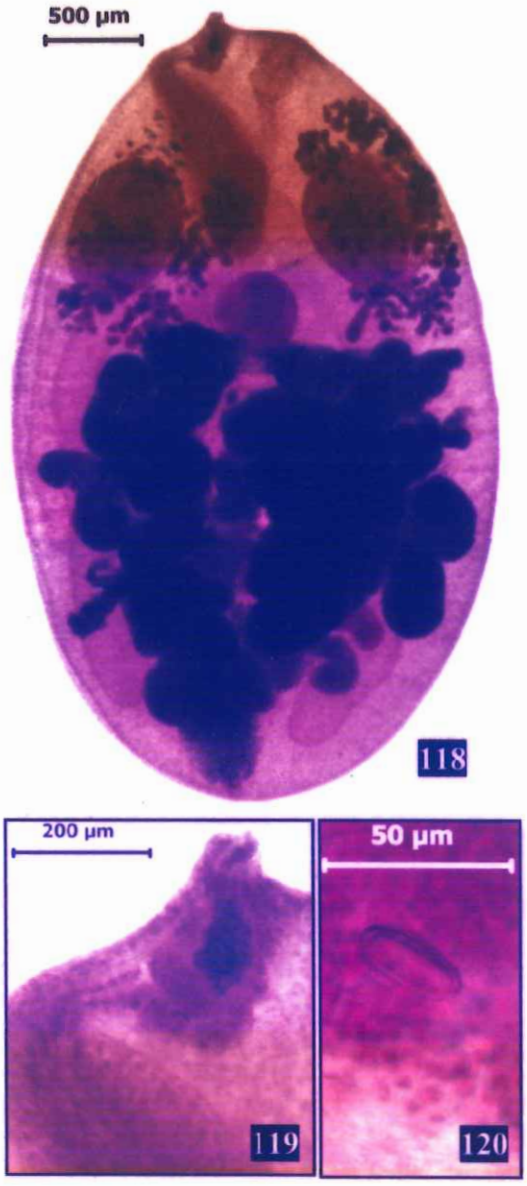


Fig. 117 ***Mehraorchis ranarum***

a. Whole fluke, ventral view

b. Egg

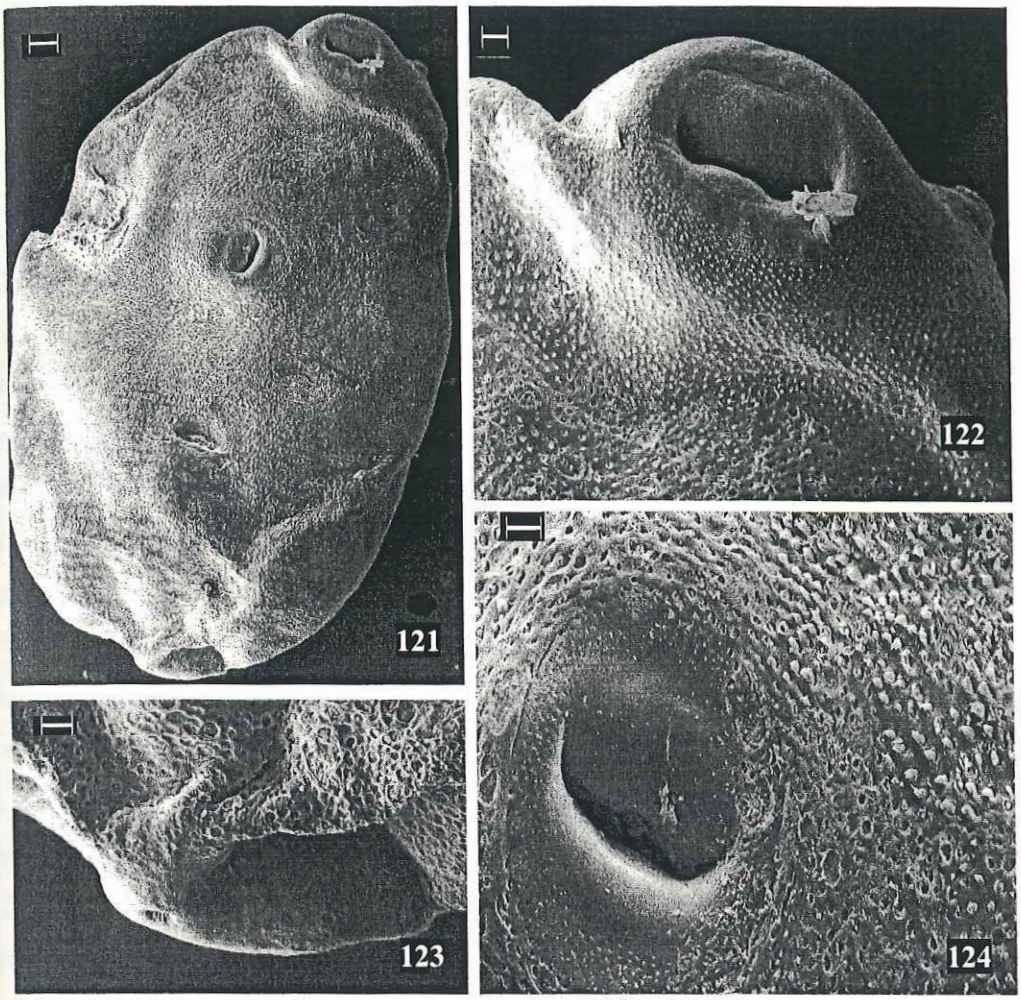


Figs.118-120 ***Mehraorchis ranarum***- Photomicrographs

118. Whole mount dorsal view

119. Enlarged genital pore

120. Egg



Figs. 121-124 ***Mehraorchis ranarum***- Scanning electron micrographs

121. Whole fluke ventral view (Scale bar=100μm)

122. A magnified view of the oral suckers, showing the spination limited around the rim of sub-terminal oral sucker and the genital pore- top right (Scale bar= 30μm)

123. Posterior end showing the excretory pore (Scale bar= 30μm)

124. A view of the ventral sucker (Scale bar= 30μm)

Genus ***Prosotocus*** Looss, 1899

Prosotocus infrequentum Srivastava, 1933

(Figs.125-132)

A total of 17 specimens were recovered from 5 hostS in Dimapur, the range of infection per host being 1-4

Descriptions (based on measurements and observations of 5 mature specimens):

Body rounded or oval, covered with spines. Ventral sucker median and equatorial, as large as oral sucker. Pharynx of moderate to large size, oesophagus short, bifurcating about mid-way between suckers; intestinal caeca short, ending in anterior part of ventral sucker. Genital pore at left of median line, sub-marginal in pharyngo-oesophageal region; cirrus-sac relatively large, lies between suckers; contains coiled seminal vesicle; well developed pars prostatica and protrusible cirrus present. Testes round to oval, disposed side by side directly or obliquely, in region of intestinal bifurcation. Ovary situated posterior to testes, to right of median line, anterior to ventral sucker. Vitelline follicles in pharyngo-oesophageal region. Eggs oval, small, numerous.

The surface fine topography of the fluke revealed wrinkles and spinations all over the tegument. Under higher resolution, entire body surface has fine bead-like texture arranged transversely around the body and the fine tegumental spines appear crystalline with pointed end, surrounded by 4-5 bead-like body in the base.

Morphometric measurements of this form are given in Table 11

Host: *Haplobatrachus tigerinus*

Location: Intestine

Locality: Dimapur

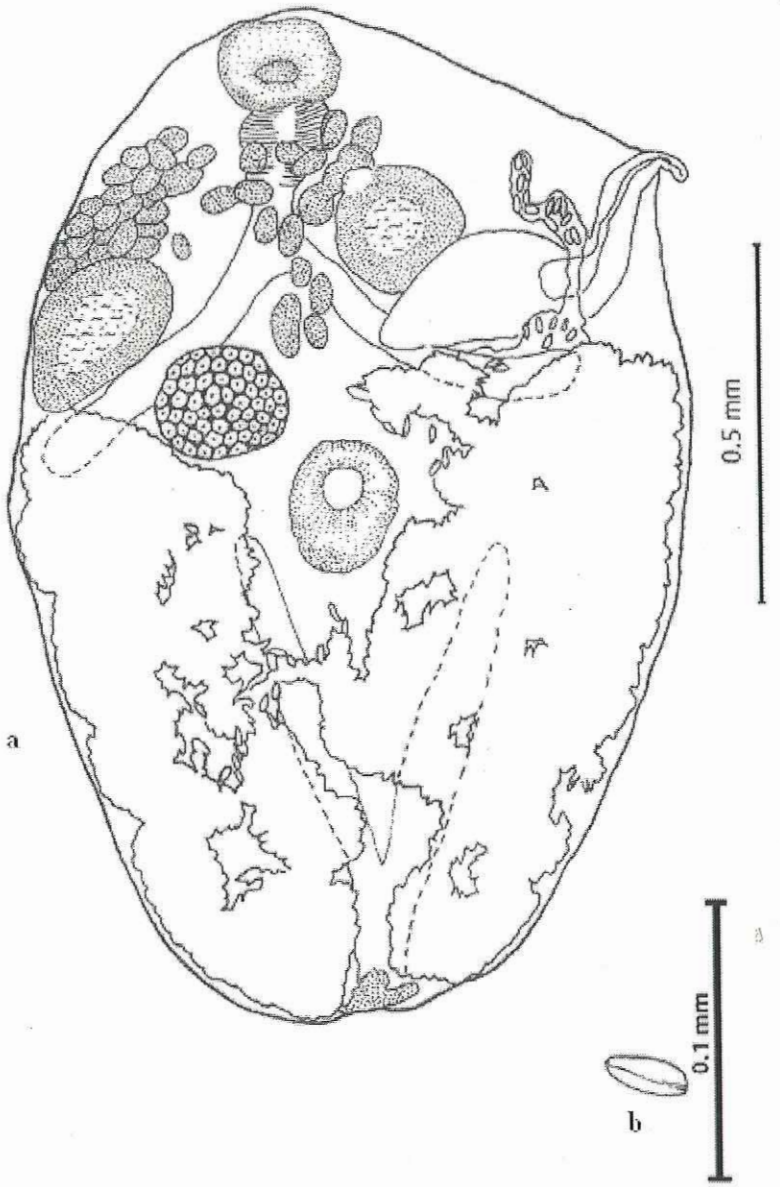
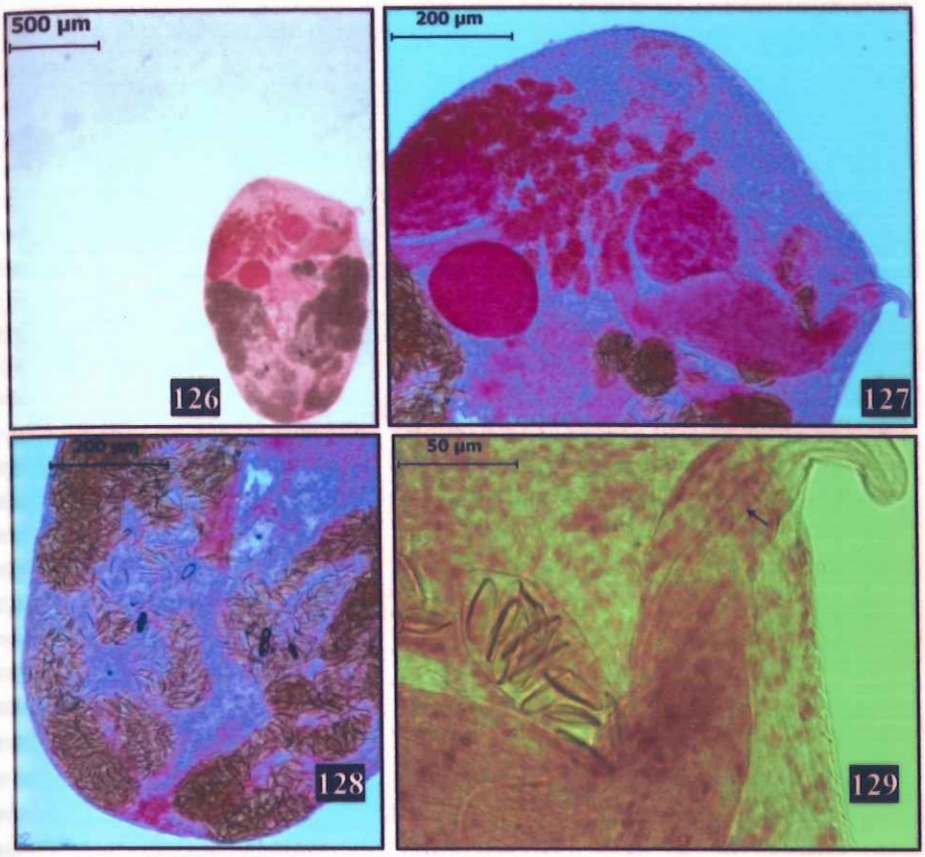


Fig. 125 ***Prosotocus infrequentum***

a. Whole mount, ventral view

b. Egg



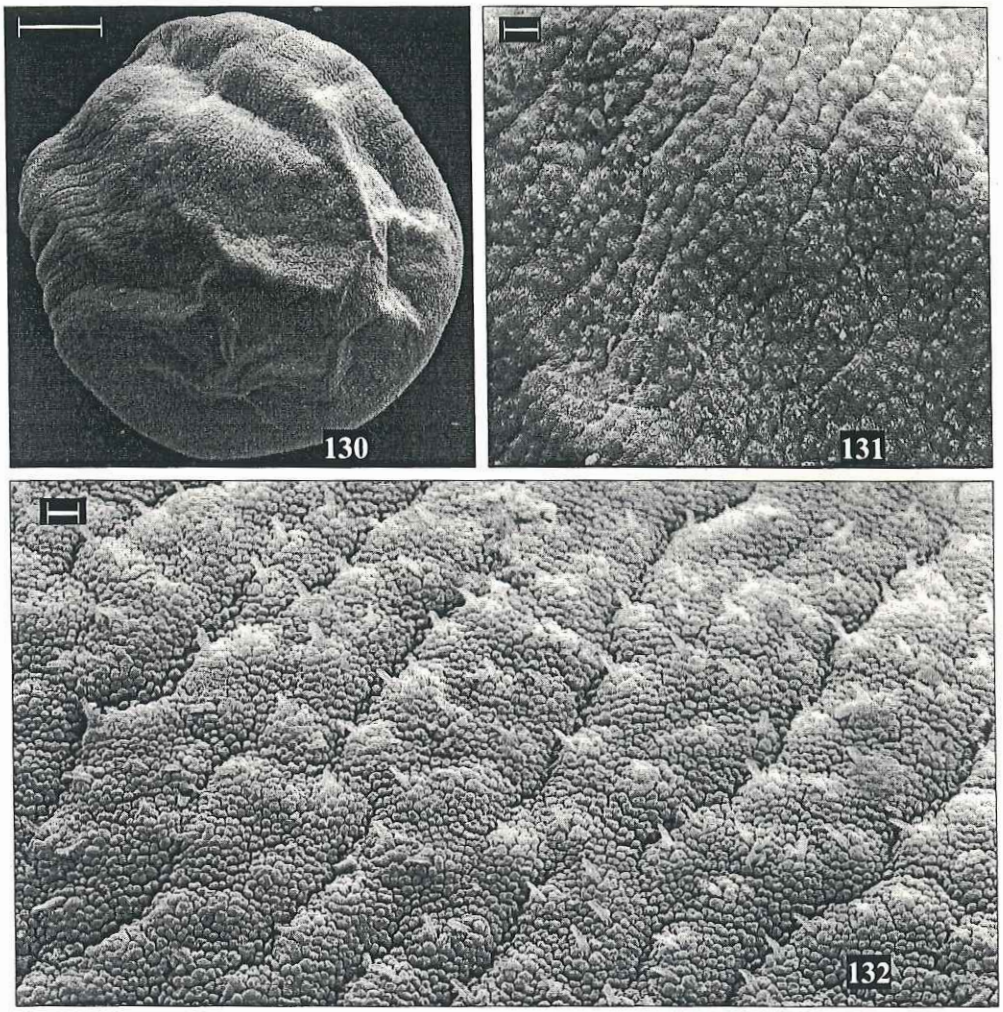
Figs. 126-129 ***Prosotocus infrequentum***- Photomicrographs

126. Whole worm, ventral view

127. Anterior end in higher magnification

128. Posterior end

129. A view of the cirrus sac containing the coiled seminal vesicle (arrow), with egg containing uterus in the forefront.



Figs. 130-132 ***Prosotocus infrequentum***- Scanning electron micrographs

130. Whole fluke, dorsal view (Scale bar= 100μm)

131. The general body tegument, showing spinations and wrinkles (Scale bar= 10μm)

132. Under higher magnification the tegument revealed numerous beaded texture poked with many fine crystalline spines (Scale bar= 2μm)

Table 11 **Morphometric measurements of *Prosotocus infrequentum*** Srivastava, 1933

Characters		Range	Mean	S.D
Length of the body		0.93-1.34	1.2222	±0.1735
Maximum width of body		0.73-0.89	0.7902	±0.0741
Oral sucker:	Length	0.01-0.14	0.1152	±0.0148
	Breadth	0.13-0.16	0.1386	±0.0151
Ventral sucker:	Length	0.13-0.19	0.1602	±0.0280
	Breadth	0.12-0.16	0.1386	±0.0163
Pharynx:	Length	0.04-0.05	0.0450	±0.0064
	Breadth	0.06-0.07	0.0648	±0.0040
Oesophagus:	Length	0.05-0.13	0.0738	±0.0321
Testes 1:	Length	0.13-0.18	0.1584	±0.0207
	Breadth	0.14-0.20	0.1674	±0.0267
Testes 2:	Length	0.21-0.24	0.2196	±0.0151
	Breadth	0.14-0.19	0.1566	±0.0197
Ovary:	Length	0.15-0.16	0.1548	±0.0040
	Breadth	0.14-0.18	0.1602	±0.0195
Cirrus sac:	Length	0.45-0.50	0.4716	±0.0235
	Breadth	0.10-0.14	0.1278	±0.0195
Distance genital from anterior end		0.27-0.52	0.3474	±0.0995
Egg size:	Length	0.03-0.030	0.0271	±0.0019
	Breadth	0.012-0.013	0.0119	±0.0008

Discussions:

Originally ***Prosotocus infrequentum*** was decribed by Srivastava, 1933, in Sitapur (Oudh) from the host *E. cyanophlyctis*. The description of the parasites in the present collection tallies with the original description of the species, with a few minor variations in the size and shape of the body and its organs. Dimapur (Nagaland) Northeast India is a new locality record and *H. tigerinus* as a new host for ***Prosotocus infrequentum***.

Genus ***Ganeo*** Klein, 1905

Ganeo tigrinum Mehra *et* Negi, 1928

(Syn. ***G. attenuatum*** *Srivatava*, 1933; ***G. gastricus*** *Srivastava*, 1933; ***G. Kumaonensis*** Pande, 1937; ***G. linguanensis*** Li, 1938; ***G. srinagarensis*** Kaw, 1950; ***G. govinda*** Dayal et Gupta, 1953; ***G. punjabensis*** Gupta, 1954; ***G. bufonis*** Fotedar, 1959; ***G. lucknowensis*** Gupta et Jahan, 1976)

(Figs.133-137)

481 specimens of this species were recovered during the study period, the range of infections per infected host being 1-19.

Body size: 2.74-3.45 x 1.09-1.65mm

Egg size: 0.01-0.027 x 0.013-0.021mm

Host: *Euphlyctis cyanophlyctis, Haplobatrachus tigerinus, H.crassus, Limnonectes limnocharis, Rana* sp. 1, *Polypedates leucomystax* and *Limnonectes* sp.

Location: Intestine

Locality: Kohima, Mokokchung, Dimapur

Remarks:

Originally described from *H. tigerinus* at Allahabad by Mehra and Negi (1928) ***G. tigrinum*** has been reported from several other parts of the country, mainly represented in *Rana* and *Bufo* spp (Gupta, 1954; Mukherjee and Ghosh, 1970, 1972; Aggarwal and Singh, 1977), though, its occurrence is also reported from reptilian and piscine hosts (Sinha, 1958; Hafeezullah and Dutta, 1985). Two more new hosts i.e., *H. crassus* and a *Limnonectes* sp. were encountered in the present study for ***G. tigrinum*** from Nagaland, North-East India. Earlier it was reported from Meghalaya and Nagaland, by Diengdoh (1989) and Tandon *et al.* (2001), respectively.

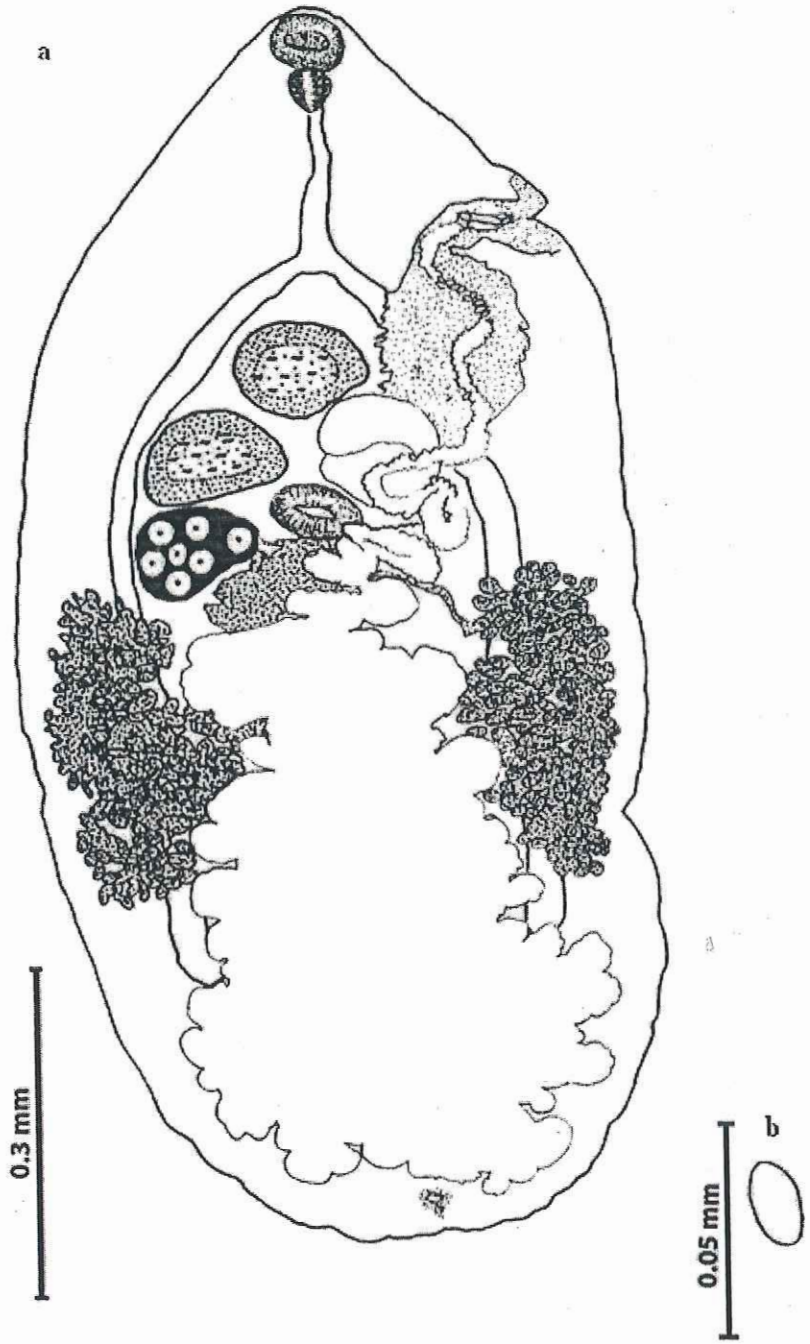
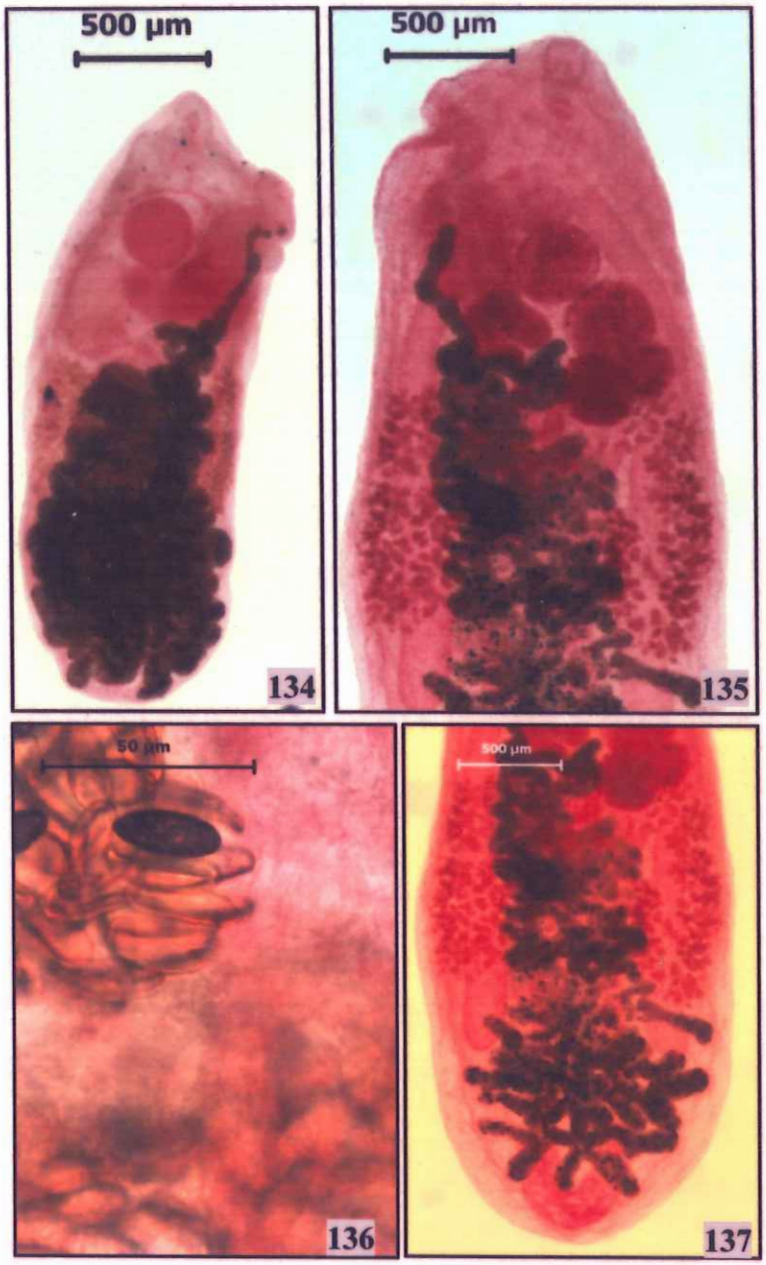


Fig. 133 ***Ganeo tigrinum***

a. Whole mount, ventral view

b. Egg



Figs. 134-137 ***Ganeo tigrinum***- Photomicrographs

134. Whole fluke, ventral view

135. Anterior region of another fluke, dorsal view

136. A fertilized egg (dark) with other unfertilized eggs

137. Posterior end to show the extend of intestinal caeca, the uterus and the U shaped excretory canal

Superfamily Pleurogeninae (Travassos, 1921)

Genus ***Pleurogenoides*** (Travassos, 1921)

Pleurogenoides gastroporus (Lühe, 1901) Travassos, 1921

(Syn. ***Pleurogenes gastroporus*** Lühe, 1901; ***P. (Pleurogenes) gastroporus*** (Lühe, 1901); Mehra et Negi, 1928; ***P. (Pleurogenes) gastroporus* var *equalis*** Mehra et Negi, 1928; ***Pleurogenes orientalis*** Srivastava, 1934; ***Pleurogenes sawanensis*** Gupta, 1954)

(Figs.138-141)

105 specimens of ***P. gastroporus*** were recovered during the study period, the range of infection per infected host being 1-23.

Body size: 1.21-1.33 x 0.95-0.99mm

Egg size: 0.027 x 0.011mm

Host: *Amolops marmoratus* (=*A. afghanus*), *Haplobatrachus tigerinus*, *H. crassus*, *Rana khare*, *Megophrys wuliangshanensis*, *Philautus* sp.1 and *Polypedates leucomystax*.

Location: Intestine

Locality: Mokokchung, Dimapur

Remarks:

Originally described from *H. tigerinus* in Uttar Pradesh by Mehra and Negi (1928) ***Pleurogenoides gastroporus*** has been recorded from *H. tigerinus* and *E cyanophlyctis* from several localities in India such as Rajasthan, Uttar Pradesh, Maharastra, Kerala and Meghalaya (Gupta, 1954; Mukherjee and Ghosh, 1970, 1972; Diengdoh, 1989). Earlier from Northeast region the species was reported from Nagaland with *R. khare* and *A. marmoratus* as new hosts for ***Pleurogenoides gastroporus*** by Tandon *et al.* (2001). In the present study 3 more hosts, *H. crassus*, *Philautus* sp.1 and *P. leucomystax* were encountered, which constitute new hosts record from the region for the same species.

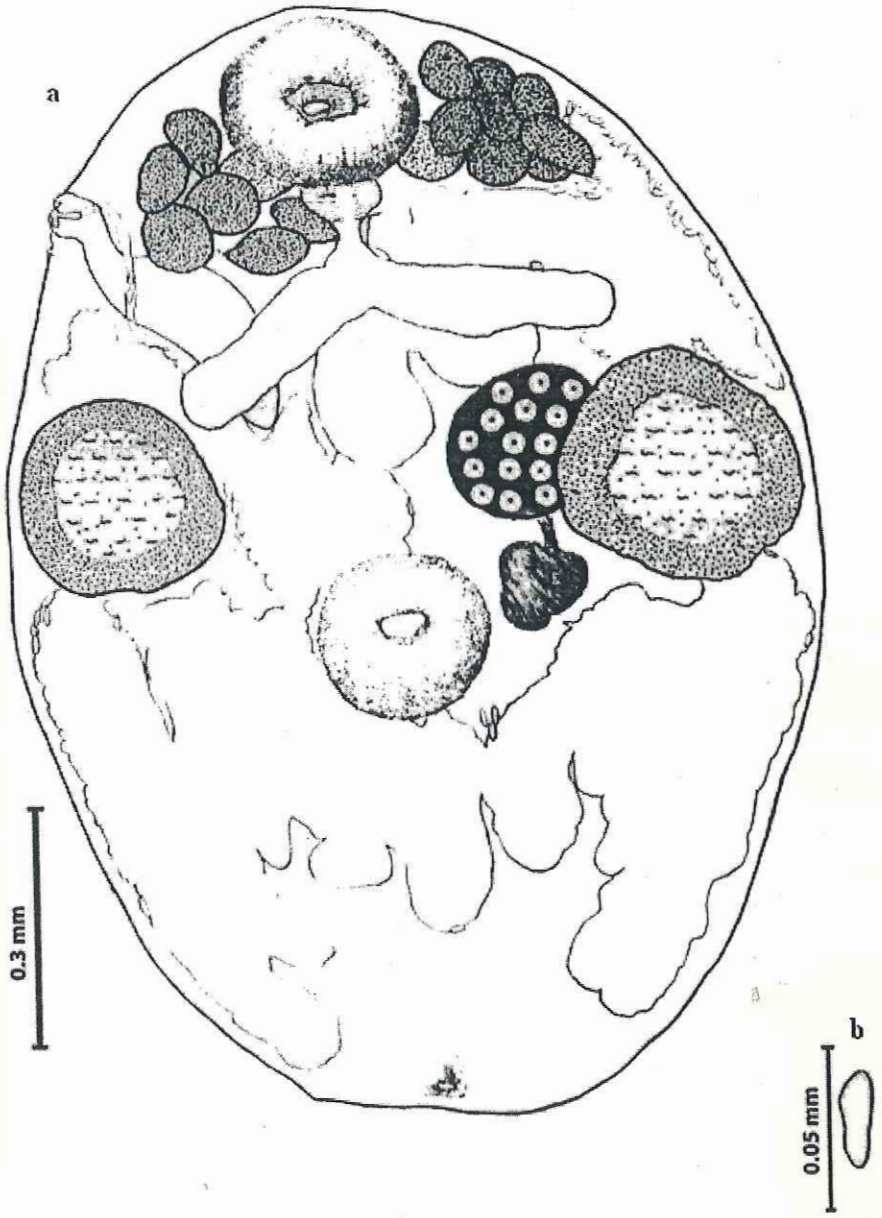
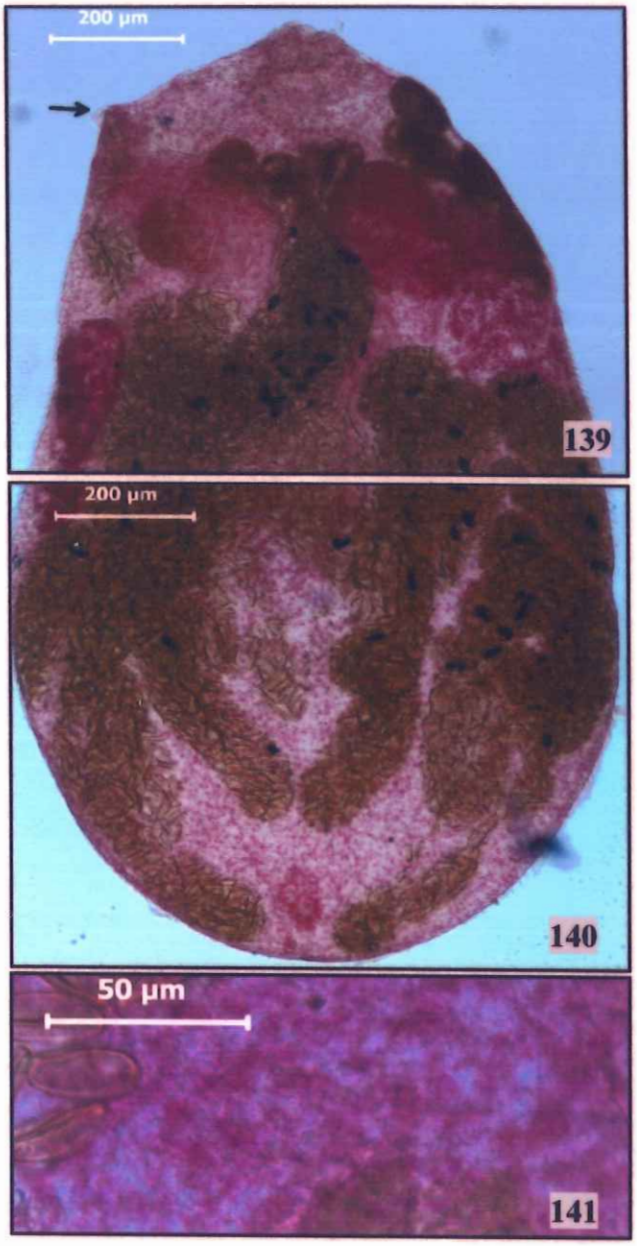


Fig. 138 *Pleurogenoides gastroporus*

a. Whole fluke, ventral view

b. Egg



Figs.139-141 ***Pleurogenoides gastroporus***- Photomicrographs

139. Anterior end, showing the genital pore (arrow), vitellaria and the eggs

140. Posterior end

141. Egg

Family Plagiorchidae (Lühe, 1901)

Subfamily Haematoloechinae (Frietas *et* Lent, 1939)

Genus ***Haematoloechus*** (Looss, 1899)

Haematoloechus almorai (Pande, 1937) Frietas *et* Lent, 1939

(Syn. ***Pneunenoces almorai*** Pande, 1937)

(Figs.142-151)

332 specimens of ***Haematoloechus almorai*** were recovered during the study period, the range of parasite per infected host being 1-22.

Body size: 6.92-9.45 x 1.38-1.74mm

Egg size: 0.31-0.32 x 0.17mm

Host: *E. cyanophlyctis*, *H. tigerinus* and *Rana* sp.1

Location: Lung

Locality: Mokokchung, Kohima and Dimapur

Remarks:

Originally described from *E. cyanophlyctis* in Kumoan hills by Pande (1937), this species has been recorded from Meghalaya and Nagaland by Diengdoh (1989) and Tandon et al., (2001) from the hosts, *E. cyanophlyctis* and *H. tigerinus*, respectively. Together with these hosts, *Rana* species (designated herein as *Rana* sp. 1) was also recorded from the latter locality. ***H. almorai*** is well represented in many localities of India such as Kashmir (Kaw, 1950), Andhra Pradesh (Khan and Mohiuddin, 1968), North India (Tickoo, 1970; Mukherjee and Ghosh, 1972), West Bengal, Maharastra (Mukherjee and Ghosh, 1970, 1972), Tamil Nadu and Kerala (Singh, 1977) from *H. tigerinus*.

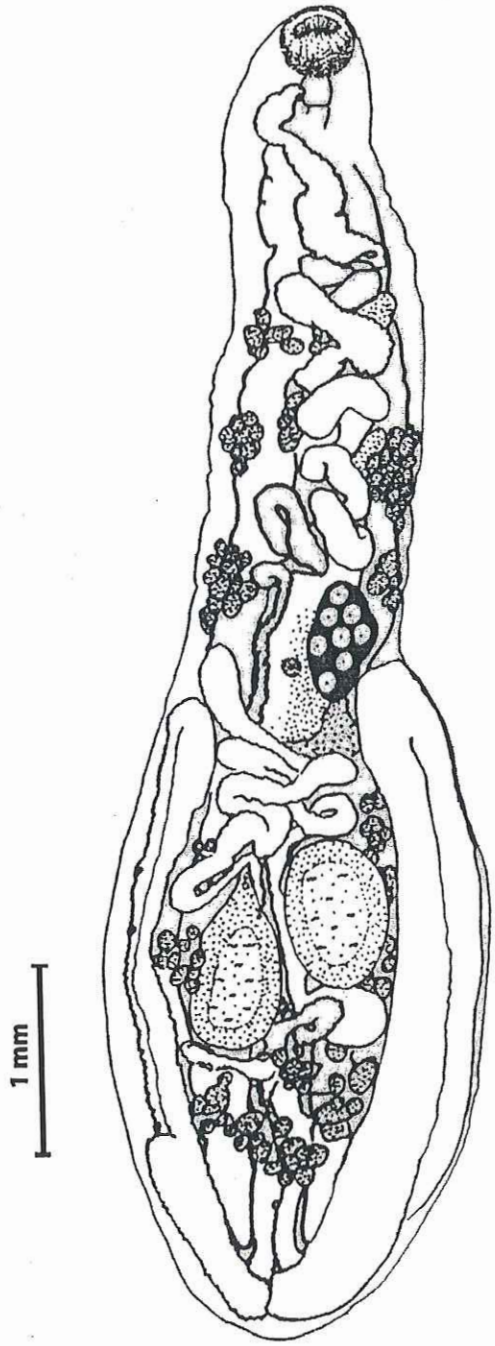
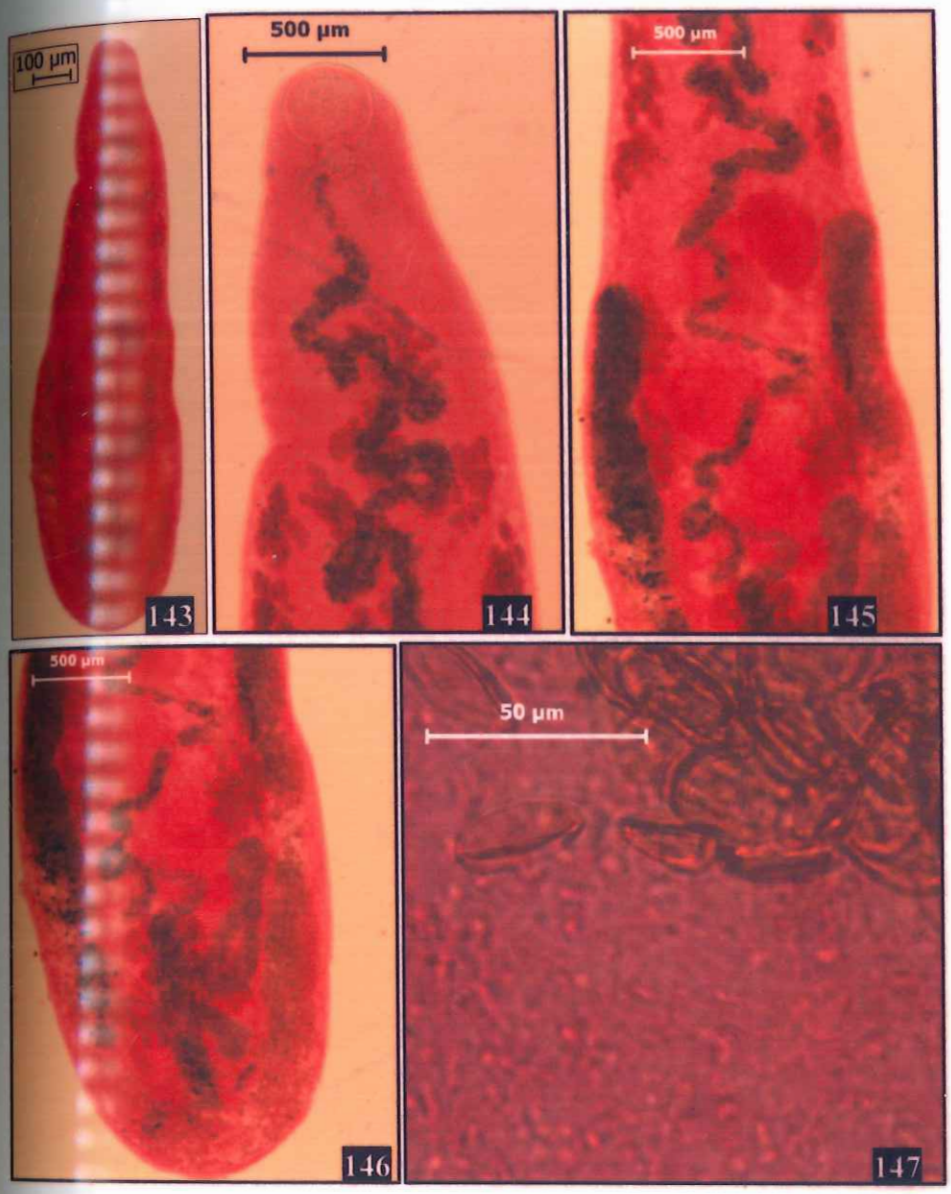


Fig. 142 ***Haemotoloechus almorai*** Whole fluke, ventral view



Figs.143-147- ***Haematoloechus almorai***- Photomicrographs

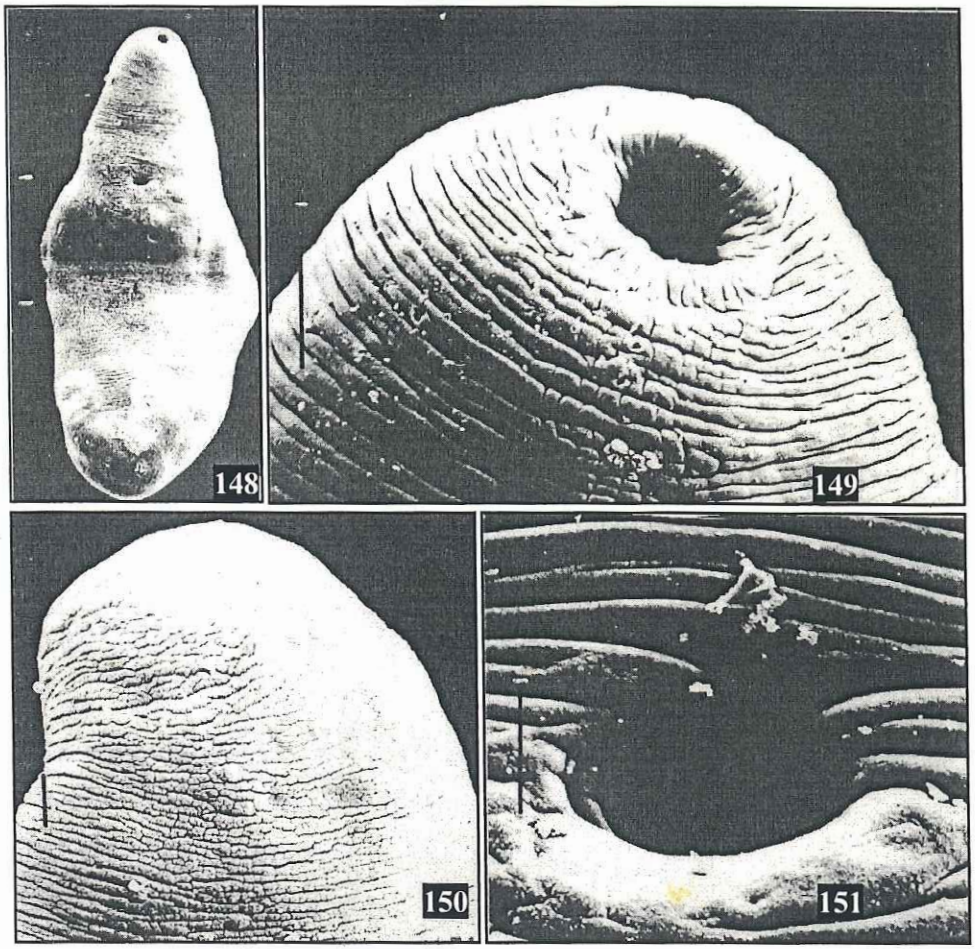
143. Whole fluke, ventral view

144. Magnified anterior end, showing the vitellaria and the egg filled uterus running anteriorly toward the pharynx

145. Mid-section, showing the ventral sucker, the ovary and the testes

146. Posterior end, showing the extent of egg-filled uterus

147. Eggs in utero



Figs.148- 151 ***Haematoloechus almorai***- Scanning electron micrographs

148. Full worm, ventral view (Scale bar=100μm)

149. A magnified view of the anterior end, showing oral sucker, the fine transverse foldings across the body tegument and the radial foldings in the oral sucker rim, an indicatIon of underlying musculature (Scale bar=10μm)

150. A magnified anterior end viewed dorsally showing the continuation of the folding from the ventral side (Scale bar=10μm)

151. Ventral sucker in a closer view. Finely beaded texture of the tegument is seen (Scale bar=10μm)

Family Opecoelidae **(Ozaki, 1925)**

Genus ***Batrachotrema*** (Dollfus et Williams, 1966)

Batrachotrema nagalandensis *Tandon, Imkongwapang *et Prasad.* 2005

(Figs.152-161)

16 specimens were recovered from *Amolops marmoratus* (= *A. afghanus*) and only 1 from *Rana khare* at Mokokchung.

Description (based on measurements of 5 mature specimens and SEM observations on 1 specimen):

Body elongated or somewhat spindle shaped, broadest at shoulder, tapering and terminating bluntly towards anterior and posterior regions; surface devoid of spines. Oral sucker subterminal.Ventral sucker pre-equatorial, located in broader part of body, 3 times larger than oral sucker, covering half of body in width, strongly muscular. Prepharynx indistinct; pharynx muscular; oesophagus of moderate length or short; intestinal caeca reaching near posterior end. Testes rounded or oval, lying in tandem in middle third of body; anterior testis at level of ovary, slightly smaller than posterior; cirrus sac long, extending posteriorly beyond ventral sucker, lying on its left side, extending anteriorly up to and ending in level with pharynx. Ovary rounded, pretesticular, postacetabular; oviduct, Mehlis' gland complex in region anterior to ovary; uterine coils mainly limited in ovarian and postacetabular area, overlapping intestinal caeca. Genital pore at left side of body, marginal or sub marginal, male and female pores opening into genital atrium. Vitellaria follicular, extending from level of intestinal bifurcation up to posterior most part of body, overlapping intestinal caeca, also scattered in intercaecal fields. Eggs numerous, large, oval.

The measurements of this form are given in Table 12

*Ref. Tandon V., Imkongwapang R. and Prasad P. K. (2005). On two new species of the trematode genera, *Batrachotrema* Dolfus et Williams, 1966 and *Opisthioparorchi* Wang, 1980 (Batrachotrematidae), with a report of a Chinese species of *Opisthioparorchis* from anuran Amphibia hosts in India. **Zoos' Print Journal Vol. 20,** No. 6 pp:1883-1887

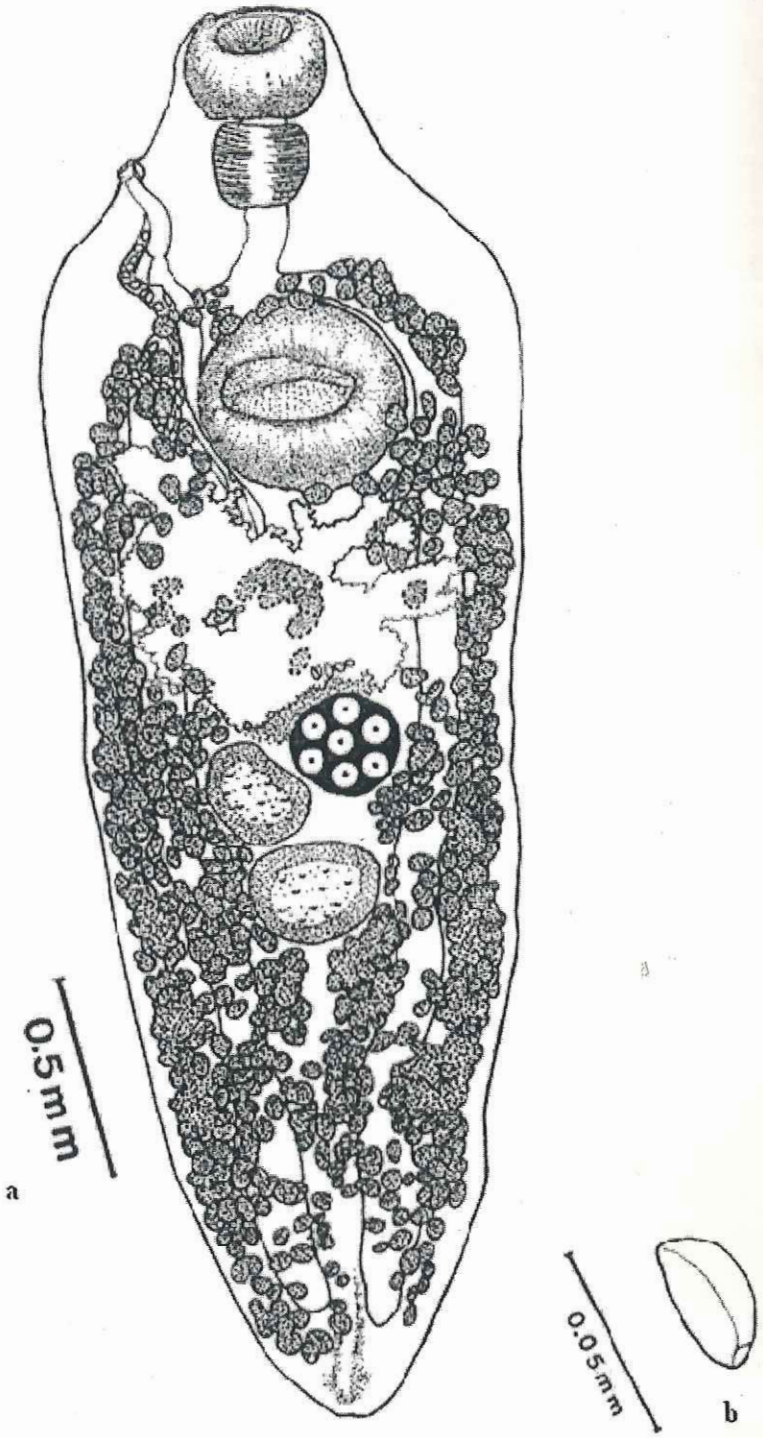
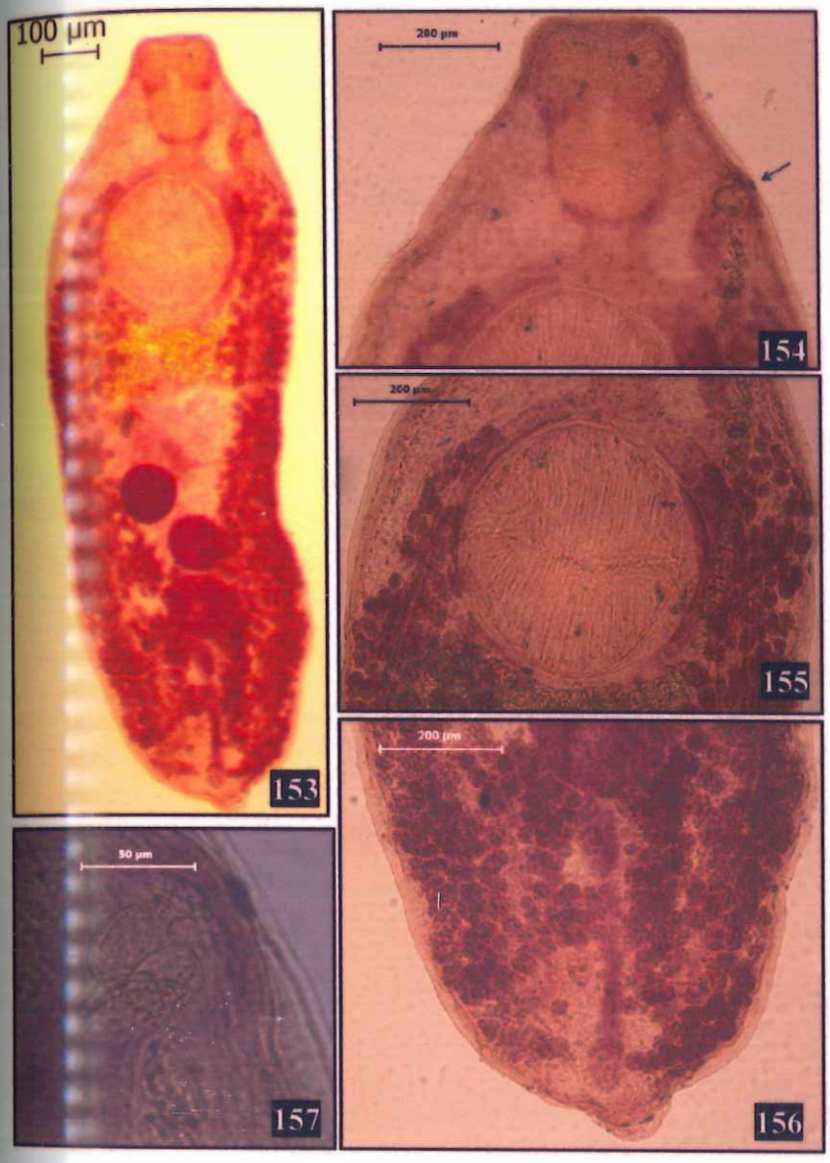


Fig 152 ***Batrachotrema nagalandensis*** Tandon, Imkongwapang et Prasad, 2005

a. Whole mount, dorsal view

b. Egg



Figs.153-157 ***Batrachotrema nagalandensis*** Tandon, Imkongwapang et Prasad, 2005

-Photomicrographs

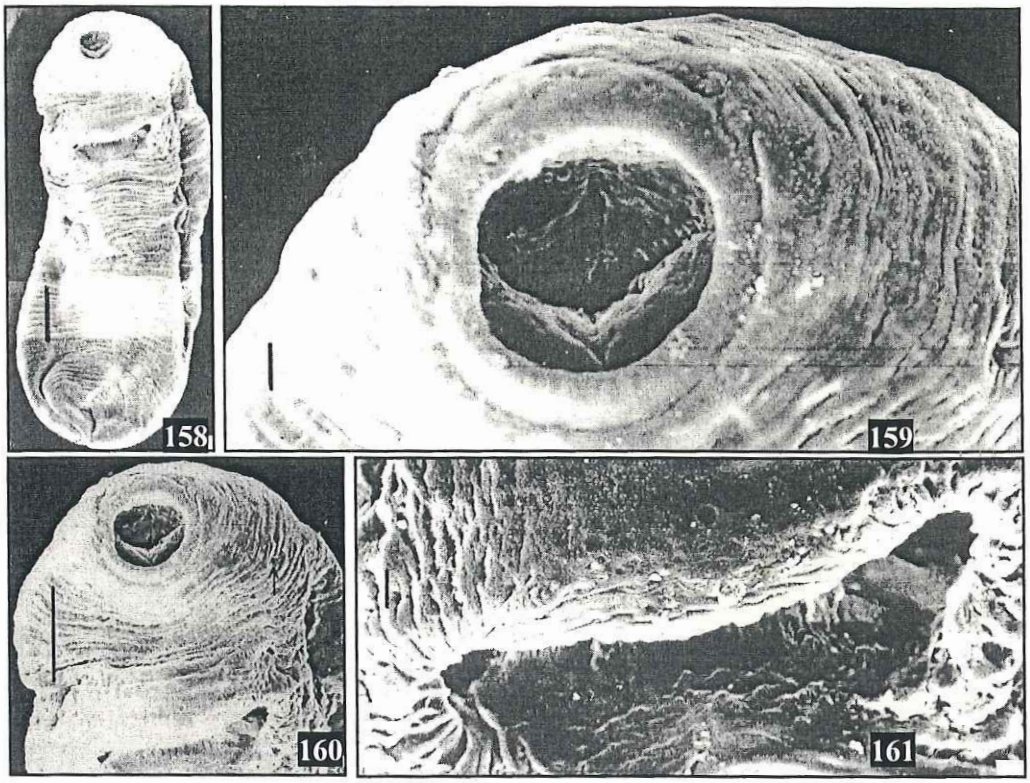
153. Whole fluke, ventral view

154. Anterior end, showing the genital pore (arrow)

155. A view of the mid region, showing the muscular ventral sucker, eggs and the vitellaria

156. Posterior end, showing the extend of vitellaria and on top the testes-2 (posterior testes)

157. Eggs



Figs.158-161 ***Batrachotrema nagalandensis*** Tandon, Imkongwapang et Prasad, 2005- Scanning electron micrographs

158. Full fluke, ventral view (scale bar= 100μm)

159. A magnified view of anterior end, showing the oral and ventral sucker, the genital pore is also seen (arrow) (scale bar=100μm)

160. Surface of the circum-oral sucker under higher magnification (scale bar=100μm)

161. A closer view of the ventral sucker region in higher magnification (scale bar= 10μm)

Table 12 **Measurements of *Batrachotrema nagalandensis*** Tandon, Imkongwapang et Prasad, 2005

Characters		Range (Mean)	± S.D.
Length of Body		2.38-3.89 (2.93)	±0.49
Maximum width of body (at level of ventral sucker)		0.80-1.27 (1.01)	±0.202
Oral sucker:	Length	0.17-0.27 (0.21)	±0.043
	Breadth	0.26-0.37 (0.33)	±0.034
Ventral Sucker:	Length	0.30-0.49 (0.42)	±0.07
	Breadth	0.39-0.56 (0.47)	±0.07
Pharynx:	Length	0.17-0.25 (0.22)	±0.032
	Breadth	0.17-0.31 (0.20)	±0.036
Length of Oesophagus		0.24-0.46 (0.41)	±0.023
Distance of intestinal caeca from hind end		0.07-0.25 (0.22)	±0.037
Intestinal bifurcation from anterior end		0.46-0.75 (0.64)	±0.01
Testis I	Length	0.20-0.22 (0.22)	±0.017
	Breadth	0.22-0.34 (0.27)	±0.035
Testis II	Length	0.24-0.46 (0.31)	±0.077
	Breadth	0.19-0.36 (0.31)	±0.058
Ovary	Length	0.22-0.29 (0.26)	±0.012
	Breadth	0.19-0.28 (0.24)	±0.022
Extent of vitellaria		1.89-3.05 (2.37)	±0.419
Eggs		(0.036 x 0.022)	±0.002

Discussion:

In having a pretesticular ovary, which is nearer to the anterior testis than the ventral sucker and the genital pore lying laterally to the median line in level with the pharynx, the present form bears a close morphological resemblance to the genus ***Batrachotrema*** Dollfus and Williams (1966) proposed a new family Batrachotrematidae for the genus. Yamaguti (1971) retained the family as proposed by Dollfus and Williams and Wang (1980) also accepted its validity. However, Prudhoe and Bray (1982), on the basis of its close morphological relationship with the members of Opecoeliidae, placed the genus under the latter family.

The genus ***Batrachotrema***, Dollfus et Williams, 1966 so far includes 5 species: ***B. petropedetis*** Dollfus et Williams, 1966 from *Petropedetes natator* in Sierra Leone, Africa; ***B. pseudobagri*** Wang, 1981(from *Pseudobagrus fulvidraconis* and *Pseudogastromyzon zebroidus* in Fujian Province, China); ***B. yaanensis*** Zhang et Sha, 1985 (from *Rana phrynoides* in Yaan, Sichuan, China); ***B. opistosacca*** Liang et Ke, 1988 (from *Rana spinosa* in Meixian, Guangdong, China); ***B. vietnamensis*** Moravec and Sey, 1989 (from *Rana kuhlii* in Hanoi, Vietnam). A comparison of the present form with the type species reveals several differences between the two. In the present form the testes lie in the middle third of the body and the cirrus sac is long extending posteriorly beyond the ventral sucker. In the type species the testes are located much posteriorly and the claviform cirrus sac is quite small extending posteriorly only up to the anterior margin of the ventral sucker. Both ***B. petropedetis*** and ***B. opistosacca*** have an elongated oval body shape and have intestinal caeca extending posteriad a little beyond the testes. In having a fusiform body, the present species resembles ***B. pseudobagri***, a species described from piscine hosts, but distinctly differs from it in having large body size, a rounded ovary (lobate in ***B. pseudobagri***), and long cirrus sac that extends posteriad much beyond ventral sucker. In possessing an unspined body, testes situated in the middle third of the body and immediately pretesticular ovary that is not separated from testes by uterine coils, the present form comes close to ***B. petropedetis***, ***B. pseudobagri*** and ***B. opistosacca*** and stands apart from ***B. yaanensis*** and ***B. vietnamensis***. In view of the apparent morphological differences and also different amphibian host species the present form is considered a new species under the genus ***Batrachotrema***. It is for the first time that a representative of the genus is being reported from the Indian Subcontinent.

Specific diagnosis: Body unspined, fusiform, testes located in middle third of body; cirrus sac long, extending beyond ventral sucker posteriorly, ovary rounded.

Etymology: Named after Nagaland, a northeastern state in India, from where the host animals were collected.

Type Host: *Amolops marmoratus* Blyth, 1855(=*Amolops afghanus*,Gunther), *Rana khare* (Kiyasetuo et Khare)

Type Location: Intestine

Type Locality: Mokokchung (Nagaland, 26°11'45.21'N and 94°17'33.04"E to-26°45'46.74"N and 94°45'52.49", India)

Holotype No: W 8338/1; Paratype No.: W 8339/1, W 8340/1

Deposited in Zoological Survey of India, Kolkata 700 053, India

Family Cathaemasiidae (Fuhrmann, 1928, emended Baer, 1932)

Genus ***Cathaemasia*** (Looss, 1899)

Cathaemasia sp.-Metacercaria

(Fig.162)

6 encysted metacercariae of ***Cathaemasia*** sp. were collected from 2 of the 339 hosts examined during the study period, infection rate being 2 and 4 in the infected hosts.

Body size: 1.64 x 0.76mm

Host: *E. cyanophlyctis*

Location: Liver, thigh muscle

Locality: Kohima

Remarks:

Cathaemasia sp. is known to be parasites of birds and *E. cyanophlyctis* represents the second intermediate host for the fluke. The presence of this species was recorded by Tandon *et al.* (2001), from Nagaland in Kohima region. In the present study, collection from another site of the same locality did not carry any infection.

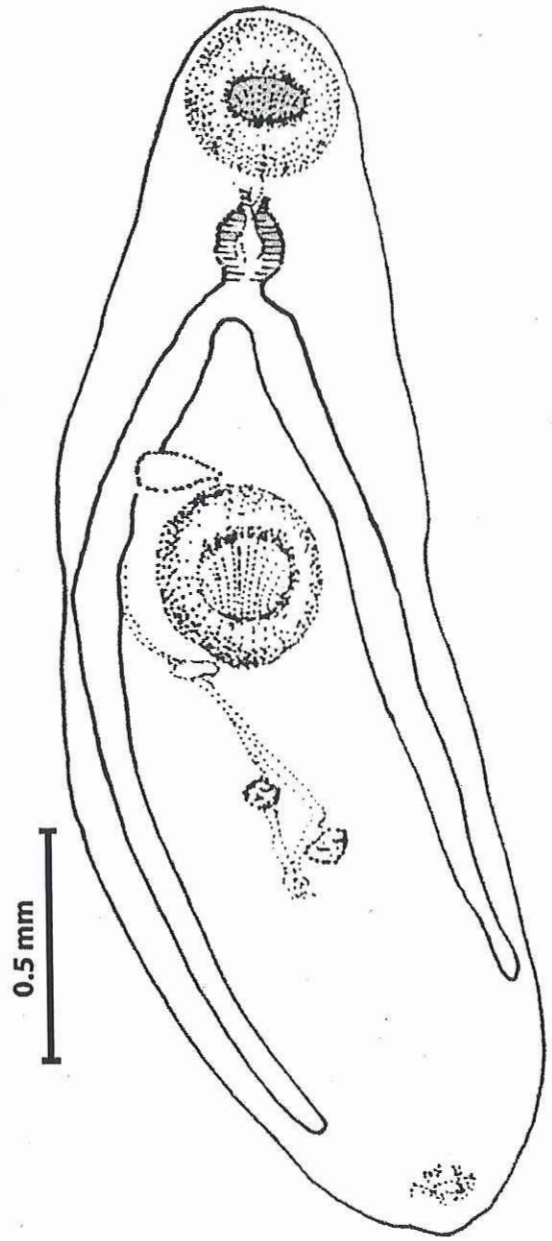


Fig. 162 *Cathaemasia* sp.

Family Proderodiplostomidae (Dubois, 1936)

Genus ***Proalarioides*** Yamaguti, 1933

(Syn. ***Travassosstomum*** Bhalerao, 1938)

Proalarioides sp.-Metacercaria

(Fig.163)

73 encysted specimens were recovered as yellow oval/ elongated cyst from the liver and body muscles of *E. cyanophlyctis* and *H. tigerinus* in Dimapur, Nagaland.

Body size: 2.55 x 1.33mm

Host: *E cyanophlyctis, H. tigerinus*

Location: Liver, body muscles

Locality: Kohima, Dimapur

Remarks:

A metacercaria representing the genus *Proalarioides* was reported for the first time from North- East India by Tandon, *et al.* (2001) in the host, *E. cyanophlyctis*. In the present study, the parasite was recovered from *H. tigerinus* at Dimapur, and thus constitutes a new host and locality record for the species from the region. This metacercaria has earlier been reported from frog host in India from *Bufo melanostictus*, *H. tigerinus* and *E. cyanophlyctis* from several localities (Srivastava and Ghosh, 1964; Karyakarte, 1967; Mukherjee and Ghosh, 1970)

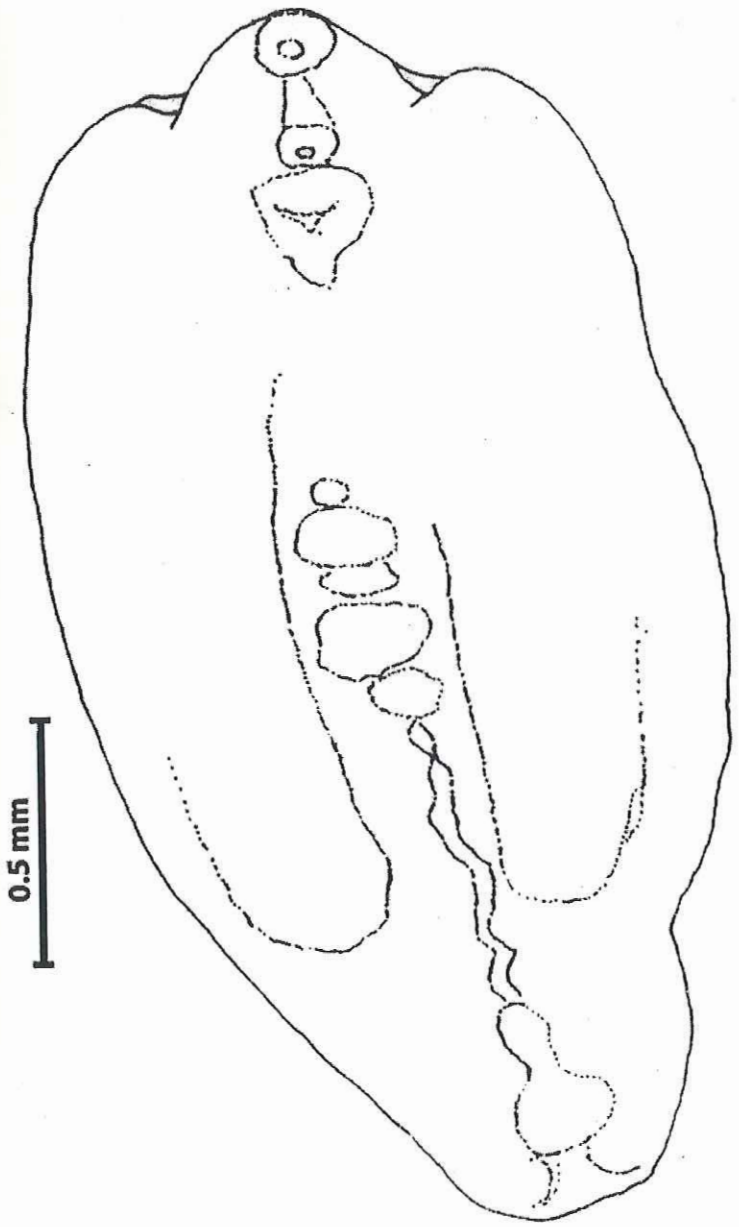


Fig. 163 *Proalarioides* sp.

CESTODA

Order Cyclophyllidea

Family Nematotaniidae Lühe, 1910

Genus ***Baerietta*** Hsü, 1935

Baerietta baeri Hsü, 1935

(Figs.164-169)

Altogether 169 specimens were collected during the study period, the range of parasites per infected host being 1-21. Most of the specimens recovered were immature.

Body size: 0.176-0.243 x 9.5-19.5mm

The surface fine topography reveals an unarmed, non rostellate scolex with four simple Suckersbearing fine microtriches. The whole body tegument has a dense covering of microtriches. However, the latter are much longer than the microtriches that cover the gravid proglottids.

Host: *Hyla annectans*, *Rhacophorus bipunctatus*, *Philautus annandalii*, *Philautus* sp., *P. tardensis*, *Euphlyctis cyanophlyctis*, *Limnonectes limnocharis*

Location: Intestine

Locality: Kohima, Mokokchung

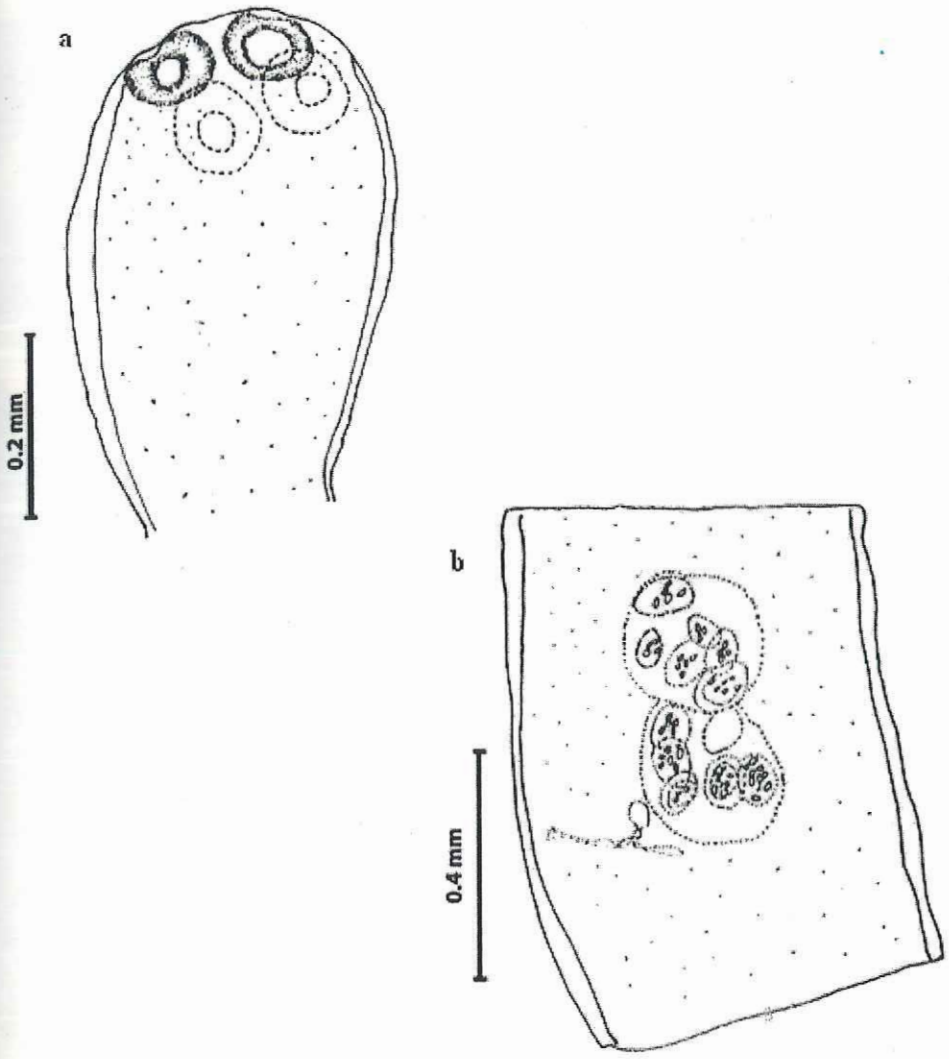
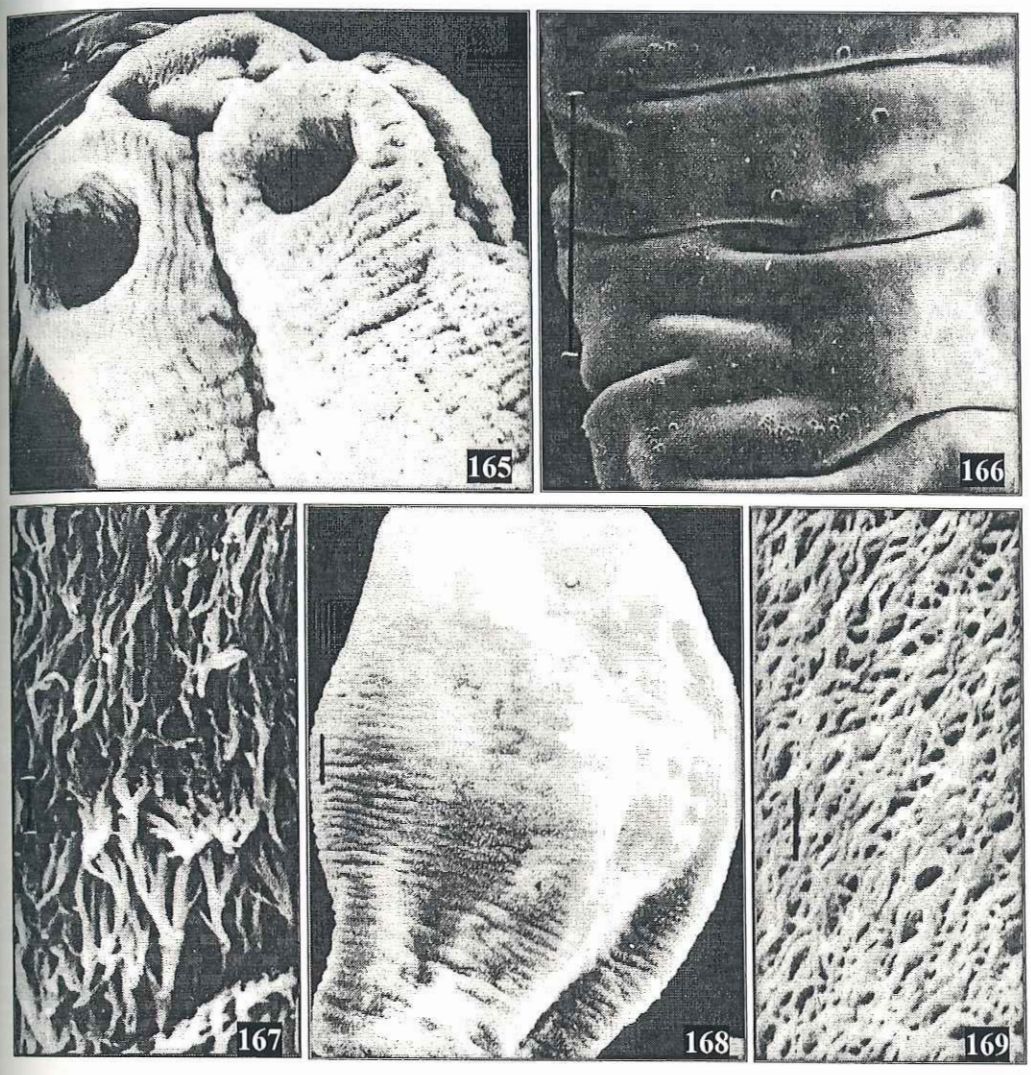


Fig. 164 ***Baerietta baeri***

a. Anterior end (scolex) lateral view

b. A matured proglottid



Figs.165-169 ***Baerrietta baeri***- Scanning electron mirographs

165. A magnified view of the anterior region, showing the non-rostellate scolex (Scale bar= 10μm)

166. A region of the immature segments (Scale bar= 100μm)

167. A view of the neck region in higher resolution, showing longer microvilli (Scale bar= 1μm)

168. A single gravid proglottid (Scale bar= 10μm)

169. Tegument in higher resolution (Scale bar= 1μm)

Remarks:

The occurrence of ***Baerietta*** sp. in the anuran Amphibia for the first time was reported from Meghalaya by Diengdoh (1989) in the Indian subcontinent. Dutta (1991) recovered similar form and identified it to be ***Baerietta baeri*** Hsü 1935. Originally ***B. baeri*** was described from *Bufo asiaticus* in China. Tandon *et al.* (2001) also reported from Kohima (Nagaland), a new locality record with a new host *H. annectans* for the species. The present study recorded its presence in yet another new locality, Mokokchung (Nagaland) with the addition of 5 more new anuran hosts, viz. *R. bipunctatus*, *P. annandalii*, *Philautus* sp., *P. tarajensis*, *L. limnocharis*.

Family Nematotaeniidae Lühe, 1910

Genus ***Nematotaenioides***

Nematotaenioides sp.

(Figs.170-175)

Only 2 adult specimens of this cestode were recovered from *Haplobatrachus tigerinus* in Dimapur and Mokokchung locality.

Descriptions (based on measurements and observations of 1 matured worm)

Body long, cylindrical, filiform, indistinctly segmented except posteriorly. Scolex provided with four suckers, without rostellum. Single set of genital organs in each segment. Genital pore marginal, irregularly alternating between left and right margins; cirrus-sac well developed. Eleven pairs of testes located below female genital unit. Ovary bilobed, vitellaria glands compact, the latter situated dorsally to former lying in ventral medulla.

Host: *Haplobatrachus tigerinus*, *P. tardensis*

Location: Intestine

Locality: Dimapur

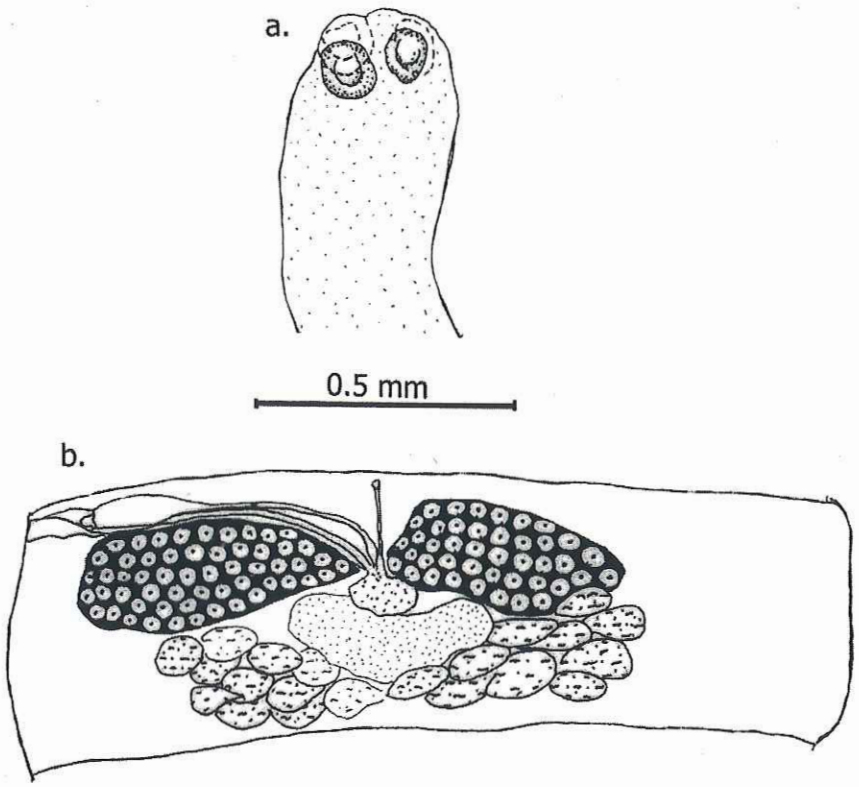
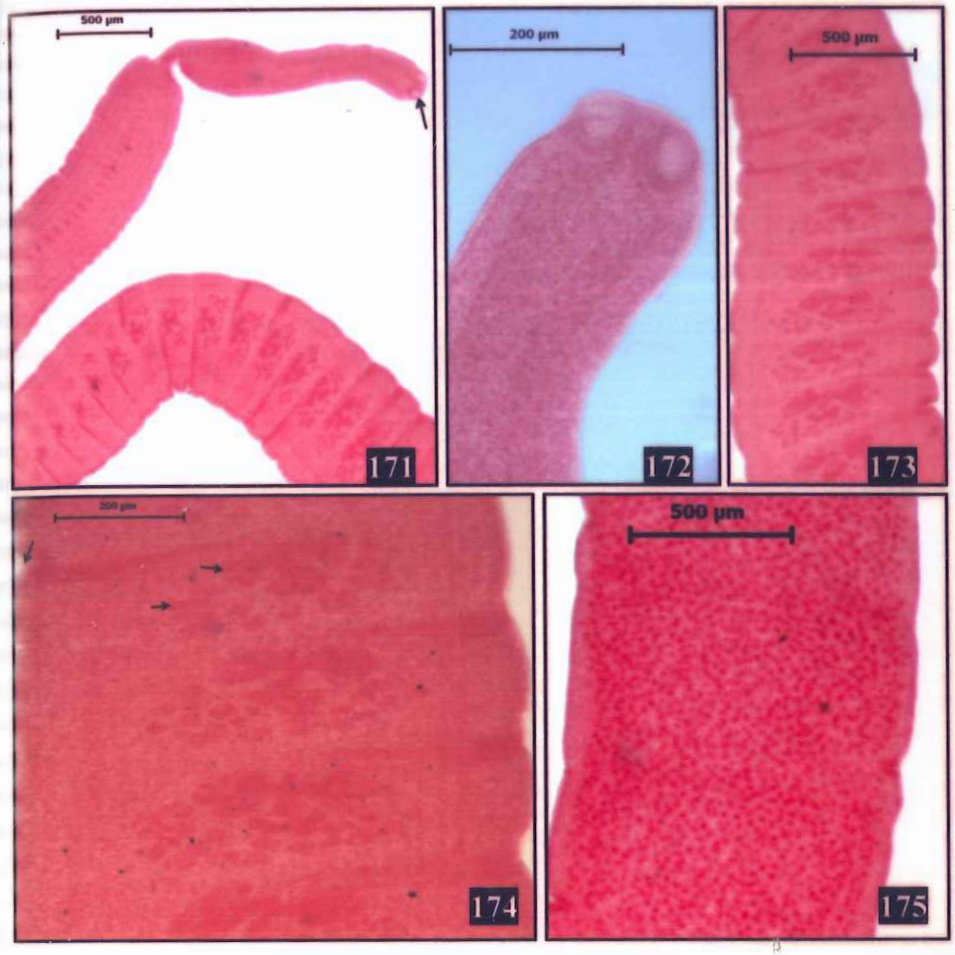


Fig. 170 *Nematotaenioides* sp.

a. Anterior end, lateral view
b. A matured proglottid



Figs.171-175 *Nematotaenioides* sp.- Photomicrographs

171. A view of the anterior portion with the unarmed scolex, 4 suckers (arrow) and strobila

172. A close up of the unarmed scolex with suckers.

173. Mature segment with the reproductive units visible as dark spots in the cortex and the excretory canals seen as longitudinal line on either lateral aspect of body.

174. A magnified view of the mature segment, showing testes (short arrow), bilobed ovary (longer top arrow) and the marginal genital pore (arrow)

175. Gravid proglottids

Discussion:

The present cestode with the following characters, body filiform, indistinctly segmented except in posterior. Scolex provided with four suckers, without rostellum. Single set of genital organs in each segment. Genital pore marginal, irregularly alternating between left and right margins; cirrus-sac well developed. Eleven pairs of testes located below female genital unit. Ovary bilobed, vitellaria gland compact is undoubtedly identified as a representative of the genus ***Nematotaenioides***. However, the specimens recovered were fragile and disintegrated on handling and processing, good whole mounts of these specimens could not be prepared. Therefore, for want of more material to be available for study, the species identification is being kept in abeyance. Nagaland forms a new locality record for the species and *H. tigerinus* and *P. tarqiensis* new host records for the genus from the region.

Family Proteocphalidae La Rue, 1911

Genus ***Proteocephalus*** Weinland, 1858

Proteocephalus tigrinus Gupta N. K. and Arora S., 1979

(Figs.176-179)

Only 1 specimen was recovered from *Haplobatrachus tigerinus*, in Dimapur locality.

Description (based on measurements and observations of 1 specimen)

Worms elongated, body cylindrical, becoming gradually narrow towards posterior end; strobila with unsegmented neck; early segments slightly longer than broad and segmented, later segments becoming longer and narrower with protrusible cirrus. Scolex without rostellum, provided with four suckers, terminal sucker or an apical organ present. Genital rudiments start differentiating behind neck and bilobed ovary clearly vesible as two dark dots segmentally arranged. Genital pores marginally, irregularly arranged. Testes disposed in medulla, between excretory canals, in two lateral fields, may or may not be confluent in median line dorsally to uterus. Cirrus pouch flask-shaped, with narrow neck; cirrus protrusible. Ovary bilobed, transversely elongated, situated near posterior border of segment. Follicular vitellaria arranged laterally to excretory canals. Uterus, a simple tube extending in median field, between ovary and anterior margin of segment, distally bearing numerous lateral outgrowths, which may occupy medullary space between excretory canals.

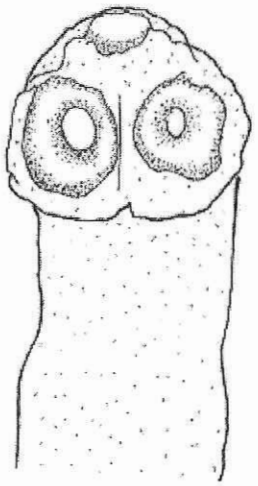
Host: *H. tigerinus*, *P. tarainsis*

Location: Intestine

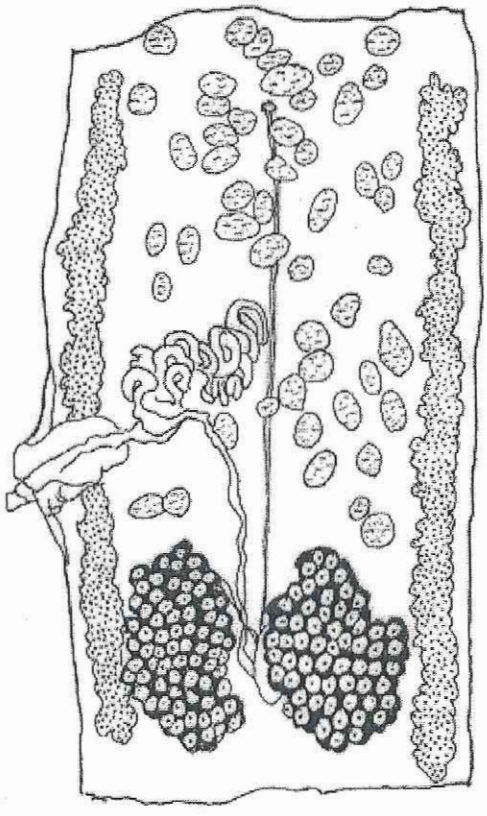
Locality: Mokokchung, Dimapur

Remarks:

Originally described by Woodland (1925) from *Haplobatrachus tigerinus* (=*Rana tigrina*) from Allahabad, Uttar Pradesh, the species ***Proteocephalus tigrinus*** was redescribed by Gupta and Arora (1979) from the same host from Punjab. The description of the parasites in the present collection tallies with the original description of the species, with a few minor variations in the size and shape of the body and its organs. Dimapur (Nagaland) and *P. tarainsis* constitute a new locality and a new host record, respectively for *P. tigerinus*.



a

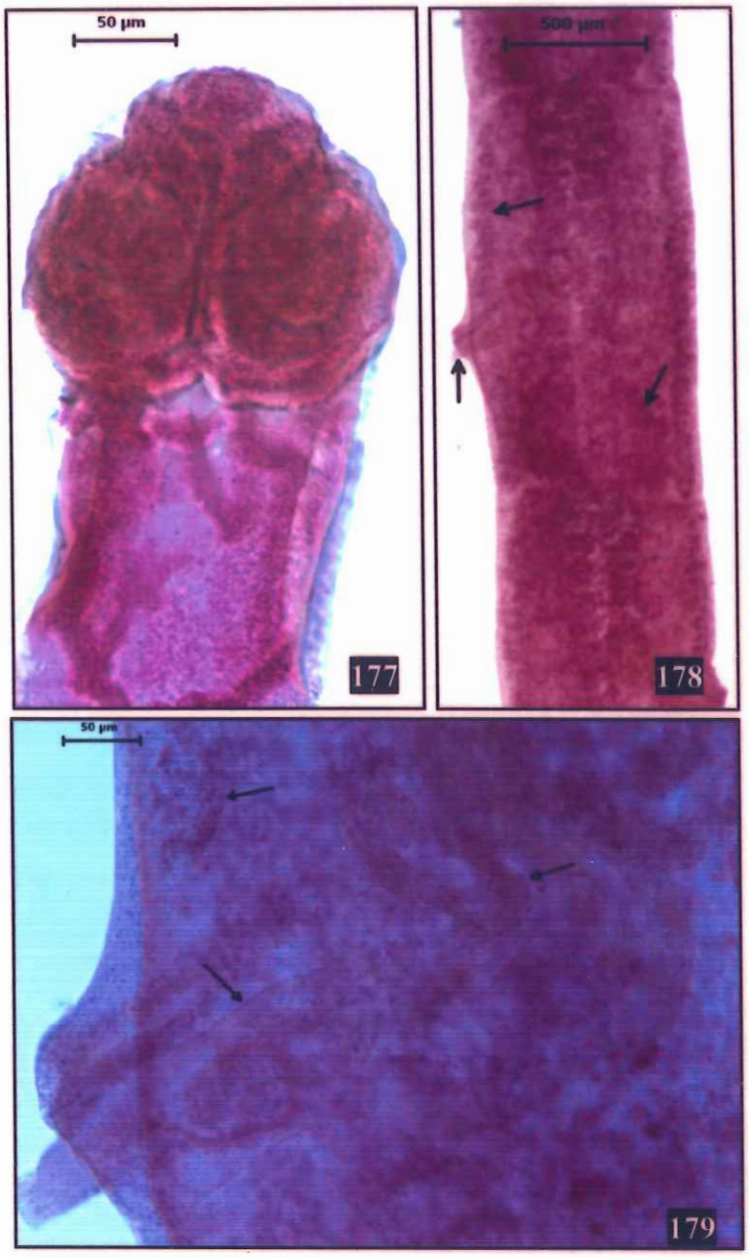


b

Fig. 176 ***Proteocephalus tigrinus***

a. Anterior end (scolex), lateral view

b. Matured proglottid



Figs. 177-179 ***Proteocephalus tigrinus***- Photomicrographs

177. Unarmed scolex, with only two suckers in view.

178. A matured proglottid with laterally arranged vitellaria (top arrow), bilobed ovary (bottom arrow) and the protruded cirrus (arrow)

179. A close up of the mature proglottid, showing the cirrus sac and the protruded cirrus (bottom)

Order Pseudophyllidea

Plerocercoid larva

(Figs.180-184)

Numerous specimens of a plerocercoid larval form were recovered during the study period from 4 of the 5 locality surveyed. The maximum number of parasites in an infected host was 18.

Body length: 50.2- 75.1mm

The surface fine topography of the larva revealed to have numerous transverse folds or wrinkles with slight suggestions of the beginning of proglottization. Under higher resolution the whole tegumental surface is shown to have dense covering of microvilli. The latter did not show any differentiation of their shape or density.

Host: *E. cyanophlyctis*, *H. tigerinus*, *H. crassus*, *L. limnocharis*, *Limnonectes* sp., *Rana* sp. 1, *R. maximus*, *R. bipunctatus*, *P. leucomystax*.

Location: Thigh and body muscle

Locality: Mokokchung, Dimapur, Kohima, Zunheboto

Remarks:

Plerocercoid larvae have been reported earlier from Northeast India by Diengdoh (1989) in Meghalaya and by Tandon and Imkongwapang in Nagaland (1999) from several hosts, namely, E. *cyanophlyctis*, *H. tigerinus*, *L. limnocharis*, *R. bipunctatus* of Meghalaya and *H. tigerinus*, *Rana* sp. 1, *R. bipunctatus* and *P. leucomystax* of Nagaland, respectively. In the present study, Zunheboto forms a new locality record from the region and *H.crassus*, *L. limnocharis* and *Limnonectes* sp. are new hosts for these Plerocercoid larvae.

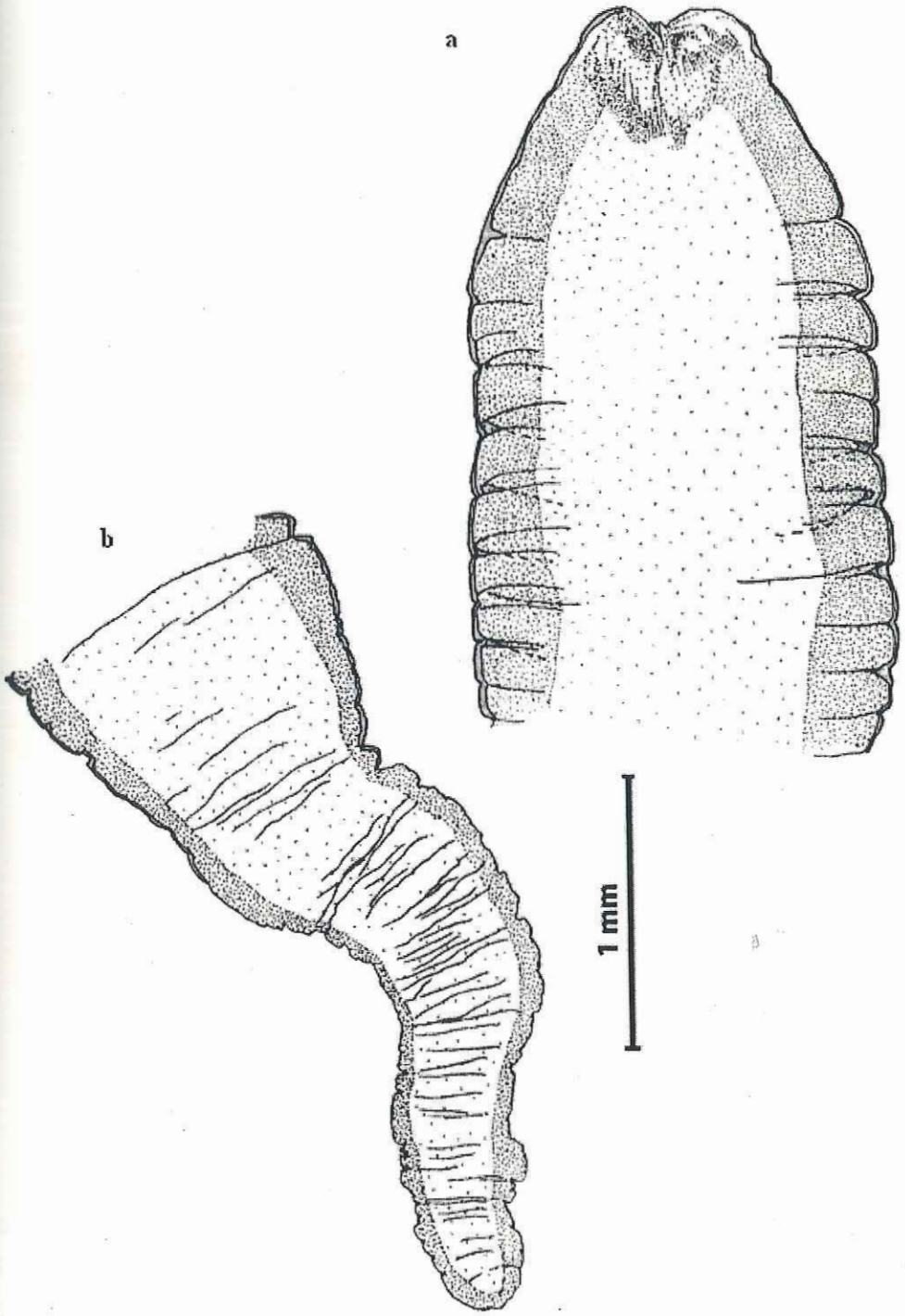
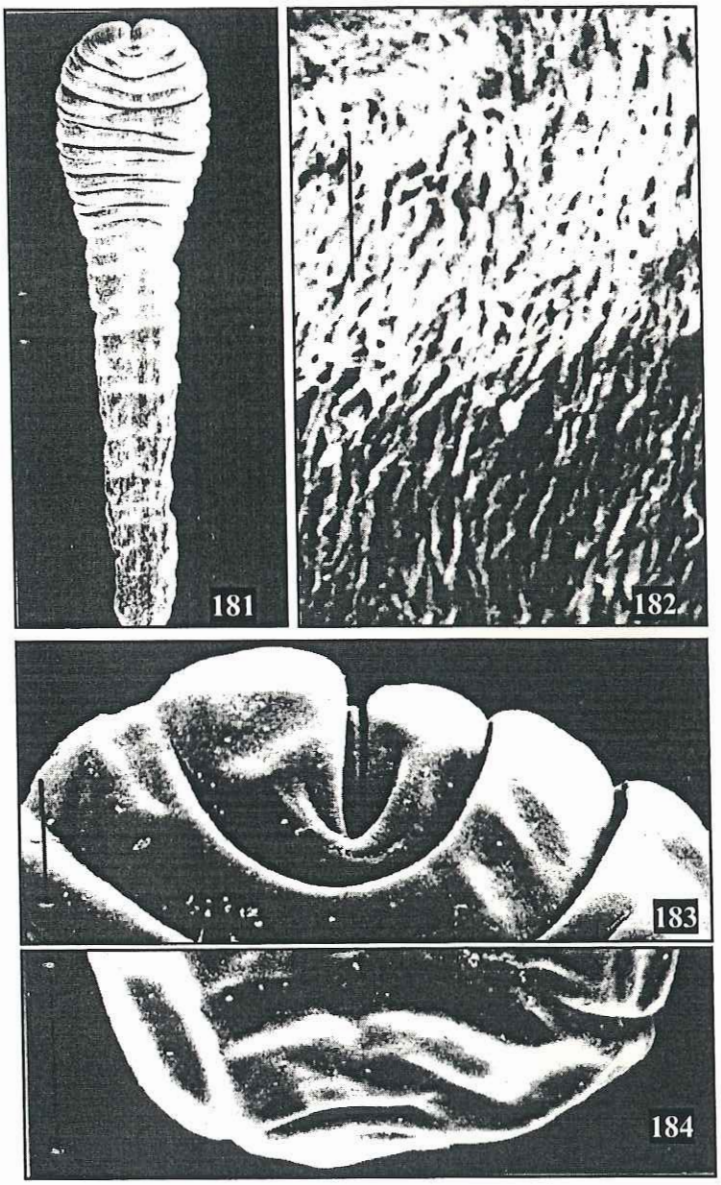


Fig. 180 Plerocercoid larvae
a. Anterior end, lateral view
b. Posterior end, lateral



Figs. 181-184 Plerocercoid larva- Scanning electron micrographs

181. Whole worm (Scale bar=100μm)

182. A view of the body tegument under higher resolution, showing the dense microtriches (Scale bar=10μm)

183. A magnified view of the anterior end, showing the apical tegumental folding (Scale bar=100μm)

184. A magnified view of the posterior end, showing the blunt end (Scale bar=100μm)

NEMATODA

Family Oxyuridae Cobbold, 1864

Subfamily Oxyurinae Hall, 1916

Genus ***Pharyngodon*** Diesing, 1861

Pharyngodon sp.

(Figs.185-192)

Only 6 female specimens were recovered during the study period from 2 species of the anuran host, *P. annandalii* and *Theloderma* sp.

Description (based on measurements and observations of 4 mature female specimens) Small worms, 3 lips with one cephalic papilla in each; esophagusus and pharynx ending in a bulb; vulva postoesophageal, directly below the excretory pore. Excretory pore in level with pharynx. Eggs numerous, with thin membranous shell, elongated.

Morphometric measurements of this form are given in Table 13

Host: *P. annandalii*, *Theloderma* sp.

Location: Intestine

Locality: Mokokchung

Remarks:

On account of the characters evident in the female worms viz. Vulva post-esophageal directly below the excretory pore. Eggs numerous, thin membrane and elongated, the present form seems to belong to the genus ***Pharyngodon***. However, species identification is difficult if not based on the characters such as number of spicules, presence or absence of gubernaculums etc. in the male worm. Since, the present collection comprised only female specimen, the species identification is being kept in abeyance pending collection of male specimens from the same hosts and locality. Nevertheless, the genus ***Pharyngodon*** is being reported herein for the first time from amphibian hosts and from the country.

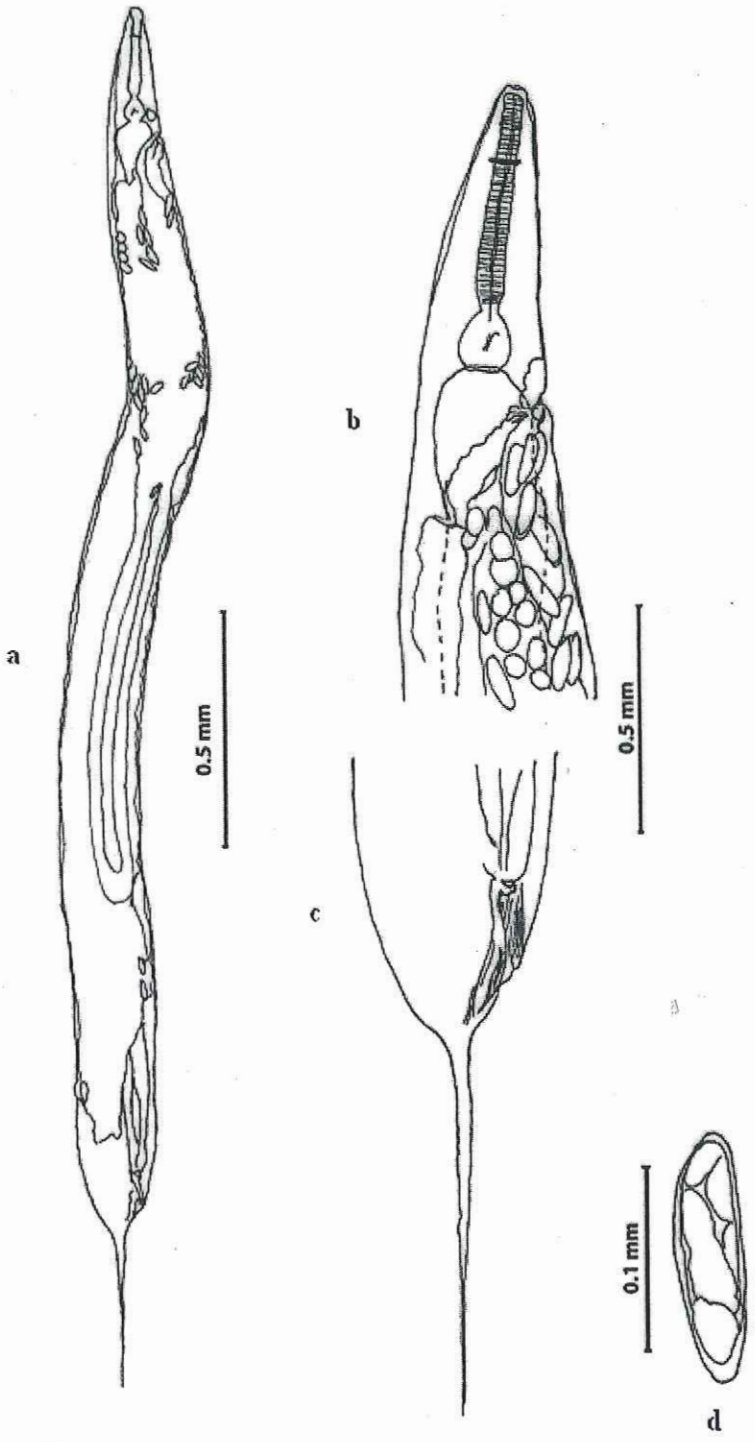


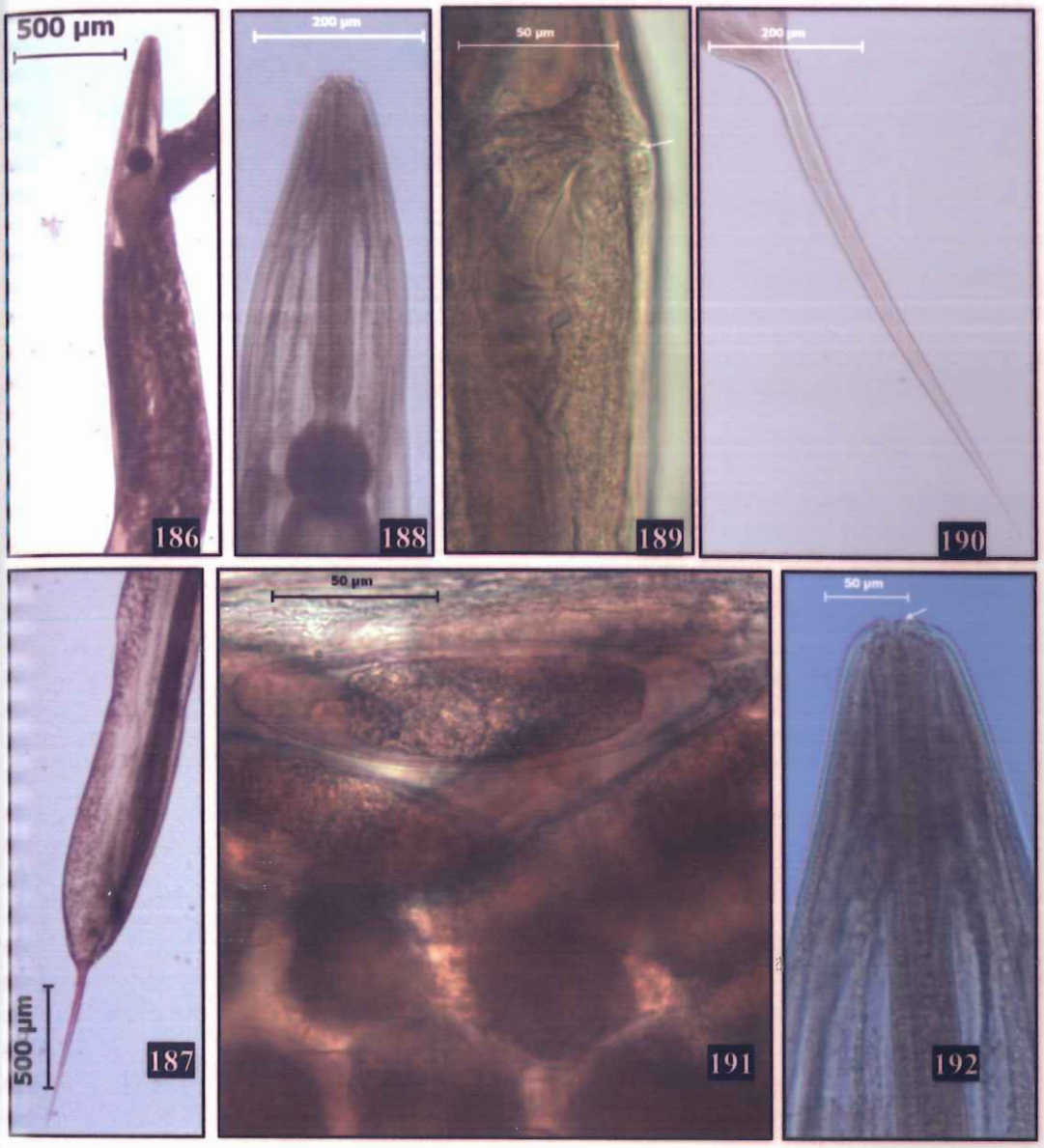
Fig. 185 ***Pharygodon*** sp.

a. Whole female worm

b. Anterior end

c. Posterior end

d. Egg



Figs. 186-192 ***Pharyngodon*** sp.- Photomicrographs

186. Anterior end of a female specimen
187. Posterior end
188. View of anterior end enlarge showing the bulb and excretory pore on the side
189. Vulva (arrow) and the posteriorly directed vagina
190. Posterior end of female with a long spike
191. A single elongated egg
192. A magnified view of the anterior end, showing the cephalic papillae (arrow)

Table 13 **Morphometric measurements of *Pharyngodon* sp. (female)**

Characters		Range	Mean	SD
Length of the body		4.163-7.521	5.359	±1.8759
Maximum width of body		0.230-0.414	0.2999	±0.1003
Pharynx:	Length	0.483-0.598	0.529	±0.0609
Tail:	Length	0.046-0.138	0.084	±0.0479
Spike:	Length	0.660-0.805	0.744	±0.0703
Distance of vulva from anterior end		0.483-0.667	0.544	±0.1062
Egg size:	Length	0.131-0.133	0.132	±0.0013
	Breath	0.032-0.037	0.035	±0.0027

Subfamily Cosmocercinae (Railliet, 1916)

Genus ***Cosmocercella*** (Steiner, 1924)

***Cosmocercella* sp.**

(Figs.193)

A total of 16 specimens of this form were collected comprising 1 male and 15 females during the study period from two of the 94 host examined.

Body size: 3.61 x 0.161mm

Egg size: 3.31 x 0.13mm

Host: *R. maximus*, *P. leucomystax*

Location: Intestine

Locality: Mokokchung

Remarks:

On account of the male having rosette papillae in the caudal region raised on surface of clear vesicle, eggs few in number, large (more than 100µm) without filaments.The present form is relegated to the genus ***Cosmocercella.***

For want of more specimens, the identification of the present form up to the species level is being kept in abeyance. The occurrence of ***Cosmocercella*** sp. from Amphibia in Nagaland forms a new representation of the genus in India and the host species *R. maximus* (=*R. nigropalmatus*) and *P. leucomystax*, a new host records.

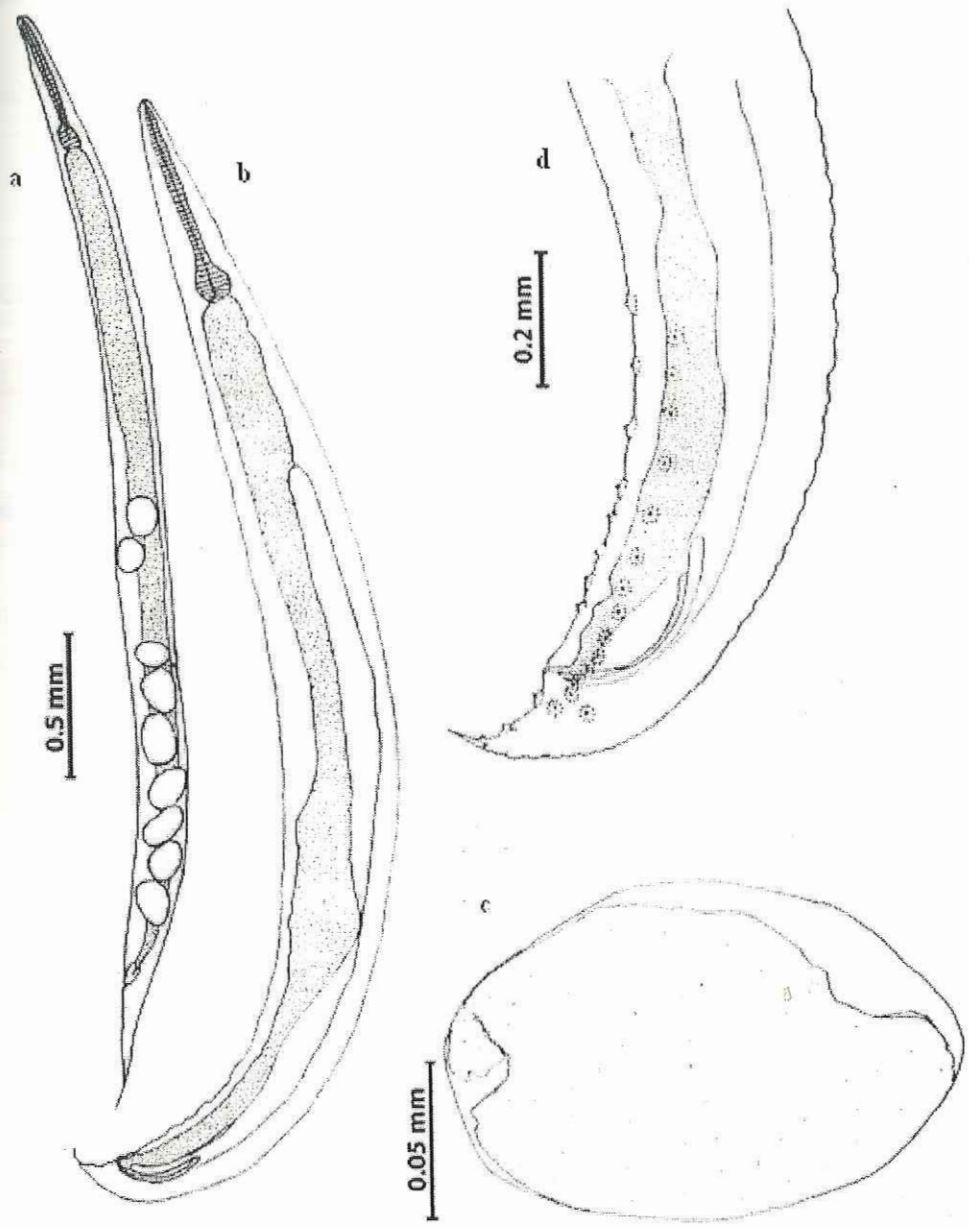


Fig. 193 ***Cosmocercella*** sp.

a. Whole female worm

b. Whole male worm

c. Egg

d. Posterior end of male, showing the rosette papillae, curve tail and the spicule

Superfamily Cosmocercoidea

Family Cosmocercidae (Railliet, 1916) Travassos, 1925

Subfamily Cosmocercinae Railliet, 1916

Genus ***Aplectana*** Railliet *et* Henry, 1916

Aplectana gubernaculum Gupta, 1960

(Syn. ***Neorielletnema*** Ballesteros – Marquez, 1945; ***Nyeraplectana*** Ballesteros – Marquez, 1945; ***Neoxysomatoides*** Yamaguti, 1961)

(Figs.194-208)

A total of 909 specimens, both male and female, of the present form were collected during the study period. The number of worms recovered per infected host ranged between, 1-272.

Description (based on 5 mature female and 5 mature male worms)

Female: Small stout worm, tapering on body both ends, abruptly ending tail. Cuticular annulations or ridges well marked. Mouth with 3 lips; buccal cavity followed by bulbous oesophagus having sclerotised apparatus; excretory pore anterior to hind region of oesophageal bulb. Vulva in equatorial region of body, uterus prodelphic, oviparous. Tail long, tapering to a pointed end.

Male: Smaller, half the length of female, large papillae in postanal region, tail long, tapering to a pointed end, curved inward. Gubernaculum absen.t

Morphometric measurements are given in Table 14

Host: *Rhacophorus maximus*, *Polypedates leucomystax*, *Philautus* sp. 1, *Rana khare*, *Rana* sp. 1, *R. livida*, *R. danielii*, *Euphlyctis cyanophlyctis*, *Amolops marmoratus*,

Location: Intestine

Locality: Mokokchung, Kohima, Dimapur

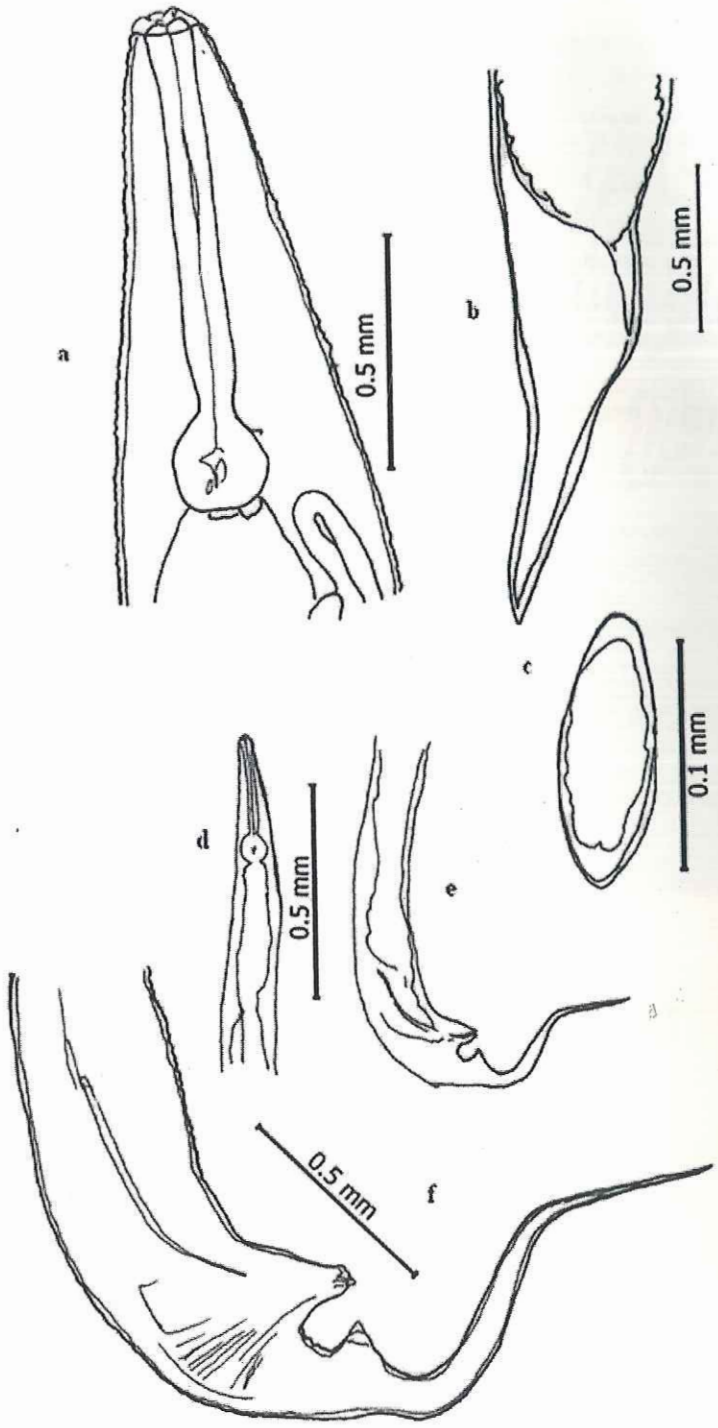
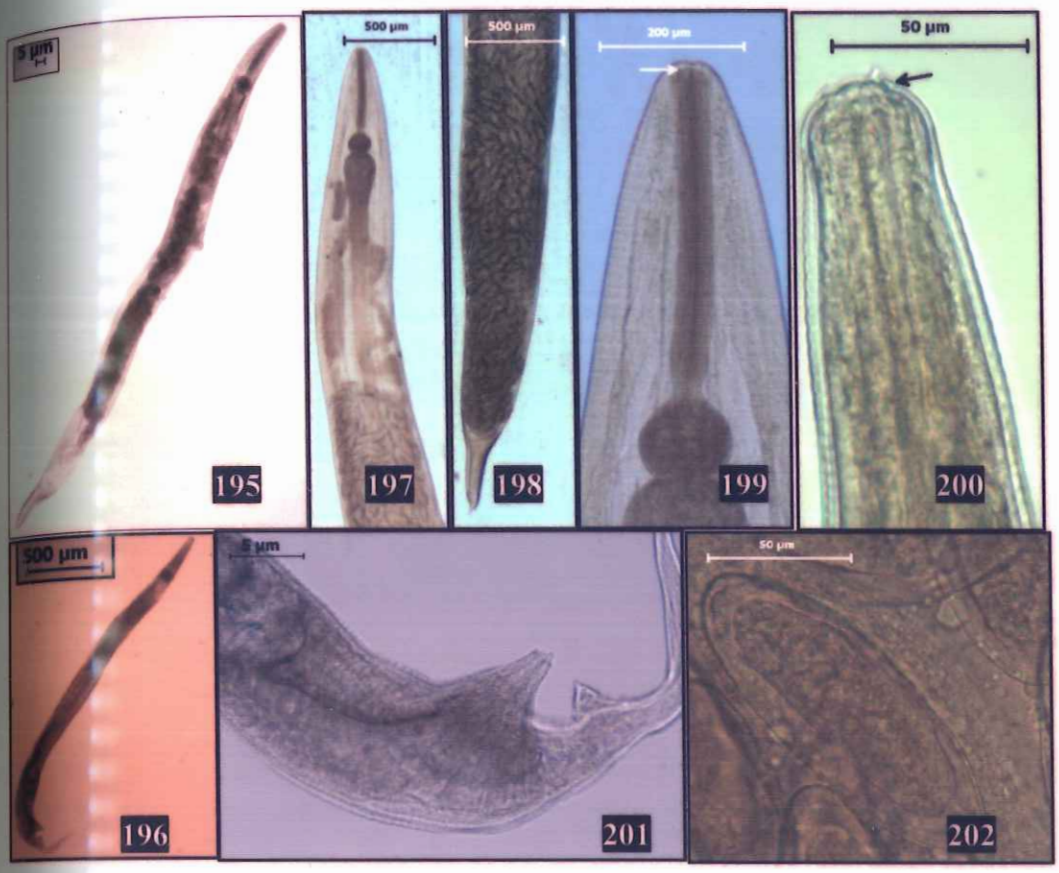


Fig. 194 ***Aplectana gubernaculum***

a. Anterior end of female
b. Posterior end of female
c. Egg
d. Anterior end of male
e. Posterior end of male
f. Enlarged posterior end of male, showing the long tail and the spicule



Figs. 195-202 ***Aplectana gubernaculum***- Photomicrographs

195. Whole worm, female.

196. Whole worm, male.

197. Anterior end of a female

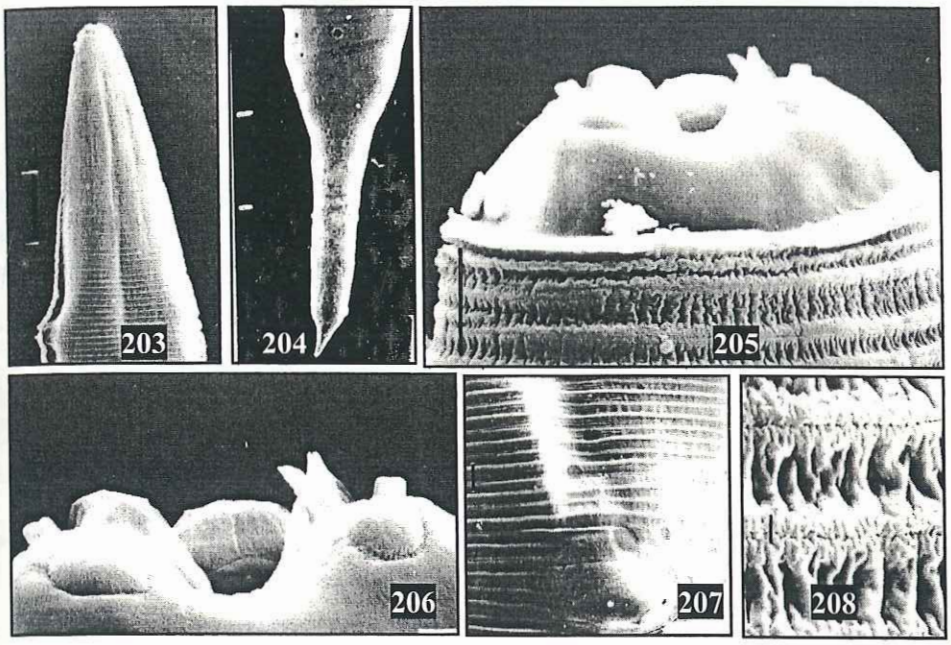
198. Posterior end of female, showing the posterior extent of uterus with eggs

199. A magnified view of the anterior end, showing the papillae (arrow).

200. Another view of the anterior end, showing the papillae (arrow).

201. Posterior end of male, showing the spicule, anal opening and the postanal papilla.

202. Egg, a magnified view.



Figs. 203-208 *Aplectana gubernaculum*- Scanning electron micrographs

203. Anterior end of the worm in low magnification showing fine transverse striation of cuticle (Scale bar= 100μm)

204. Posterior end, showing the tail end and the smooth surface in the posterior part (Scale bar= 100μm)

205. Anterior tip magnified, showing a smooth surface cephalic labial region delimited posteriorly by regular transverse striations of the cuticle (Scale bar= 10μm)

206. A close up of the buccal region, showing the tips, papillae and amphids (arrow) (Scale bar=10μm)

207. A portion of the mid-region showing genital opening (Scale bar= 1μm)

208. Cuticular pattern under higher resolution (Scale bar= 1μm)

Table 14 Morphometric measurements of *Apectana gubernaculum* Gupta, 1960

Characters	Male			Female		
	Range	Mean	SD	Range	Mean	S.D
Length of the body	1.61-3.0	2.5	±0.528	6.46-7.06	6.83	±0.22
Maximum width of body	0.13-0.30	0.19	±0.046	0.47-0.49	0.47	±0.009
Pharynx: Length	0.30-0.42	0.40	±0.051	0.47-0.49	0.48	±0.009
Breadth	0.04-0.04	0.01	±0.003	0.04-0.04	0.01	±0.003
Oesophageal bulb: Length	0.09-0.11	0.10	±0.006	0.12-0.13	0.12	±0.001
Breadth	0.09-0.11	0.10	±0.010	0.09-0.11	0.10	±0.010
Spike length	0.40-0.74	0.49	±0.145	-	-	-
Tail length	0.13-0.19	0.16	±0.023	0.45-0.80	0.61	±0.108
Distance of vulva from anterior end				2.60-2.74	2.66	±0.045
Egg size: Length				0.13-0.14	0.12	±0.003
Breadth				0.04-0.04	0.04	±0.0004

Remarks:

Diengdoh (1989) reported the occurrence of ***Aplectana*** sp. in *R maximus* (=*R. nigropalmatus*) in Northeast India. Tandon *et al.* (2001) also reported the same form from Nagaland, constituting a new locality record and added 5 new hosts-*R. levida, R. khare, E. cyanophlyctis, P. leucomystax* and *A. marmoratus*. However, in both the cases the collection comprised female specimen only and species identification of the nematode could not be done.

In the present study both male and female forms were collected species from the earlier reported hosts and also from *Philautus* sp. 1and *Rana danielii.* Species of ***Aplectana*** are known to occur and are well distributed all over the world among the amphibians (Yamaguti, 1961). Gupta (1960) described 2 species, ***A. gubernaculum*** and ***A. asiatica*** from *H. tigerinus* and *B. melanostictus*. In having a bulbous oesophagus, female with prodelphic condition of uterus and vulva near middle of body; and absence of gubernaculum in the male the nematode in the present collection tally with the species described by Gupta (1960). The present form is therefore identified as *Aplectana gubernaculum* and the new hosts for ***A. gubernaculum*** are *R. livida, R. khare, E. cyanophlyctis, P. leucomystax* and *A. marmoratus*.

Genus ***Aplectana*** Railliet *et* Henry, 1916

Aplectana sp.

(Figs.209-211)

A total of 15 specimens, all females, of the present form were collected during the study period. The number of worms recovered per infected host ranges from 1-11

Body size: 6.30-7.42 x 0.37-0.47mm

Egg size: 0.12-0.12 x 0.039-0.048mm

The surface fine topographical study revealed the general body tegument with fine annular transverse striations all through the length of the worm devoid of any papillae.

Host: *R. maximus*, *P. leucomystax*

Location: Intestine

Locality: Mokokchung

Remarks:

The form was identified as ***Aplectana*** sp. in having a bulbous oesophagus, prodelphic condition of uterus and vulva near middle of body but in having different egg size, tail length and tegumental ornamentation, it was assigned as ***Aplectana*** sp.2 (Tandon *et al.*, 2001). In the present study no male specimens were encountered, therefore, the specimen identification is kept in abeyance.

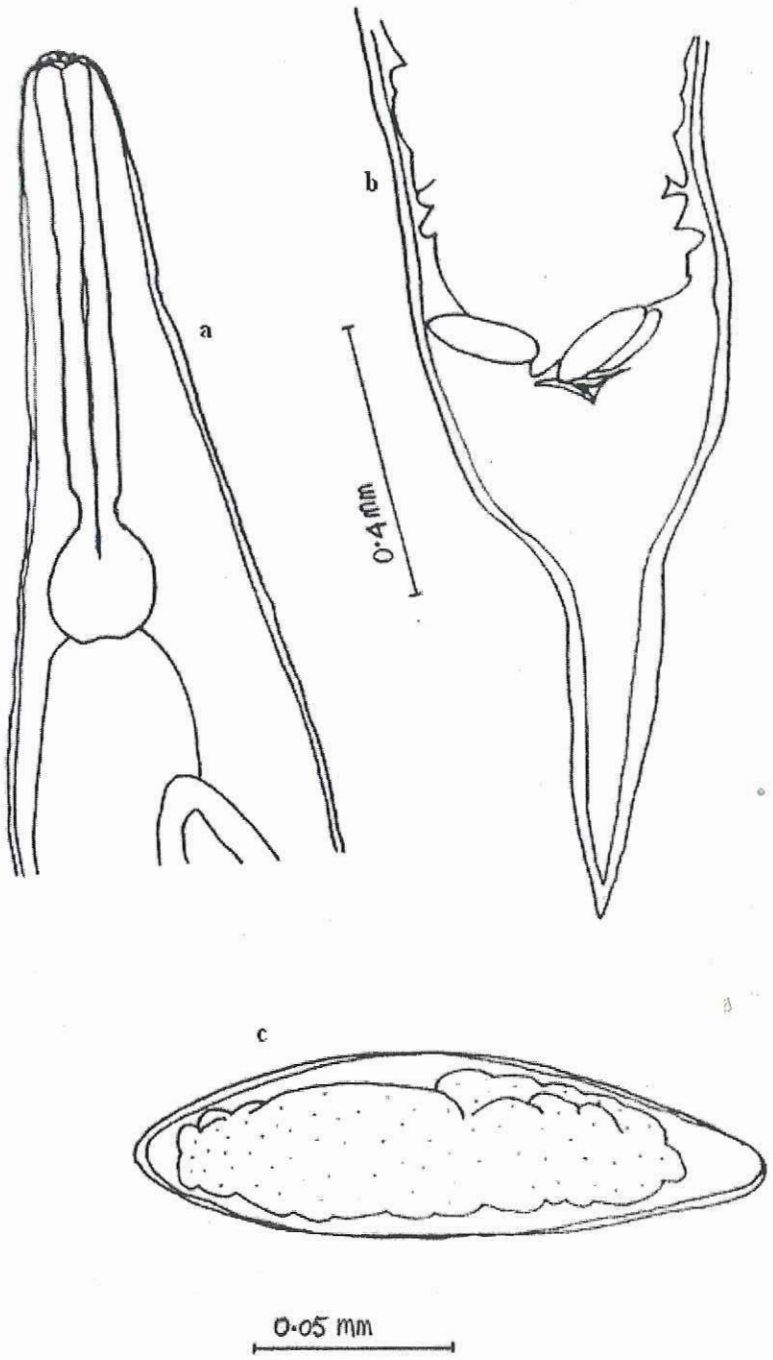
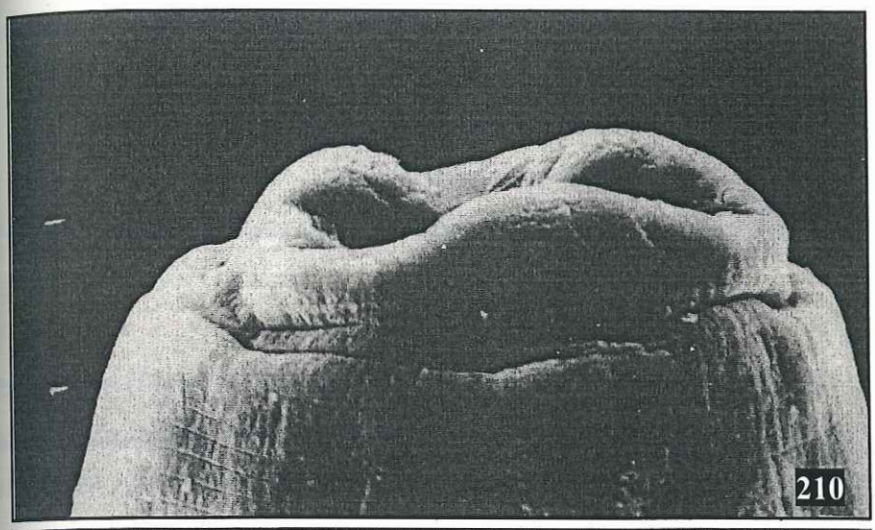


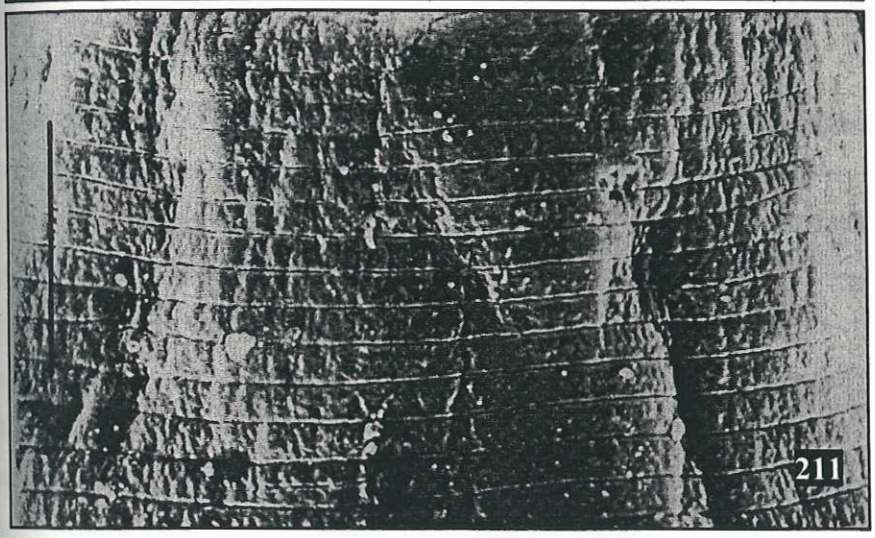
Fig 209 *Aplectana* sp.

a. Anterior end of female

b. Posterior end

c. Egg





Figs. 210-211 ***Aplectana* sp.** - Scanning electron micrographs

210. Anterior end of a female worm, showing the lack of any papilla

211. A view of the mid region showing the transverse striation

Genus ***Paracosmocerca*** Kung et Wu, 1945

(Syn. ***Nematoxys*** Schnider, 1866, partim)

(***Ananconus*** Railliet *et* Henry, 1916)

Paracosmocerca mucronata Kung et Wu, 1945

(Figs.212-219)

22 specimens of this form was recovered during the study period from *P. leucomystax*, *M. wuliangshanensis*, *R. khare*

Description (based on measurements and observations of 5 males and 5 females)

Small medium-sized worms. Mouth with three lips, oesophagus with a small pharynx and a very slight prebulbous swelling and posterior bulb; excretory pore level with oesophageal bulb.

Male:

Lateral flanges narrow, extending entire length of body, Tail tapering to a mucronate tip, 10 longitudinal rows of papillae and 2 preanal rows 5 Plectanes each. A few adanal and several postanal papillae also present. Each plectanes inverted V-shaped in profile, without any internal skeleton. Spicule single, very wide, anterior end broad, posterior end pointed, with lateral borders thickened and strongly chitinized and median portion membranous . Gubernaculum absent. Tail curved inward terminating abruptly and ending in a short spike.

Female:

Tail elongated and pointed at tip. Vulva anterior to middle of body, vagina directed backward from vulva, viviparous; Eggs large.

Morphometric measurements of this form are given in Table 15

Host: *P. leucomystax*, *M. wuliangshanensis*, *R. khare*

Location: Intestine

Location: Mokokchung, Kohima, Zunheboto

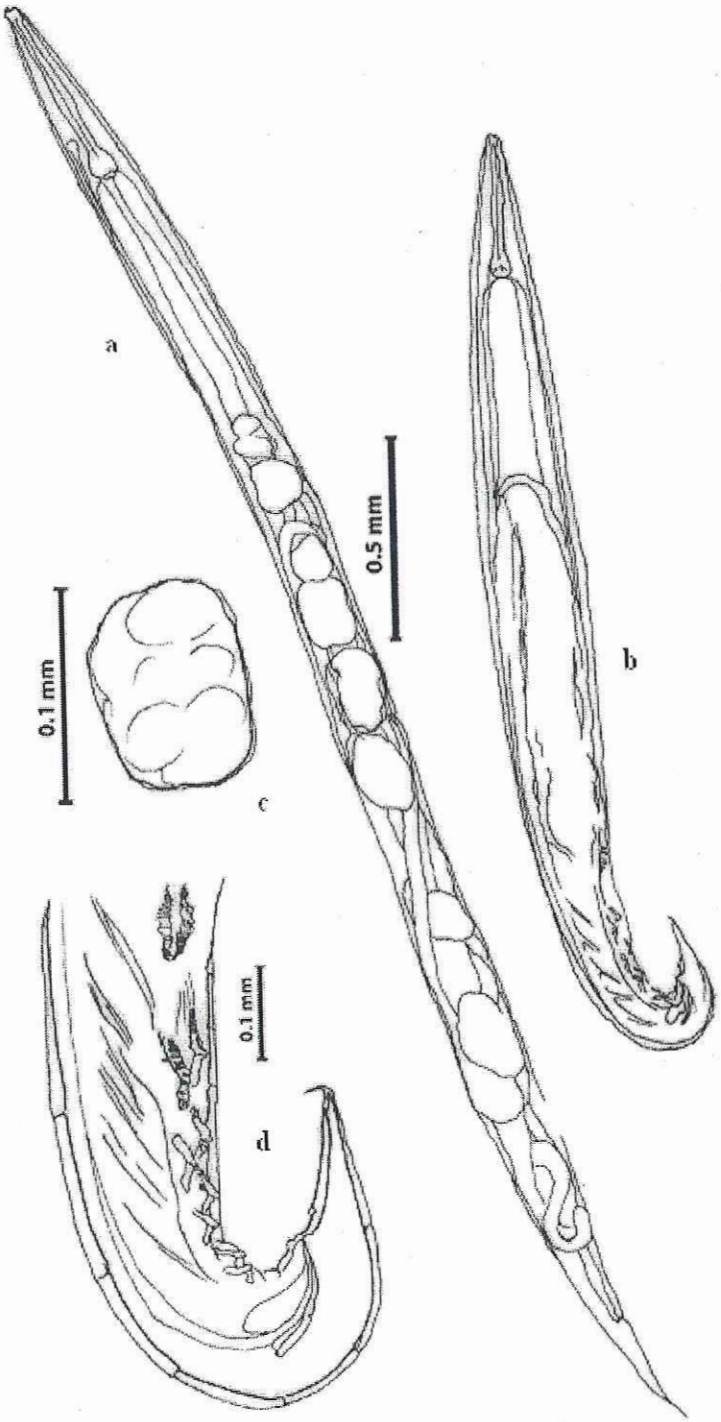
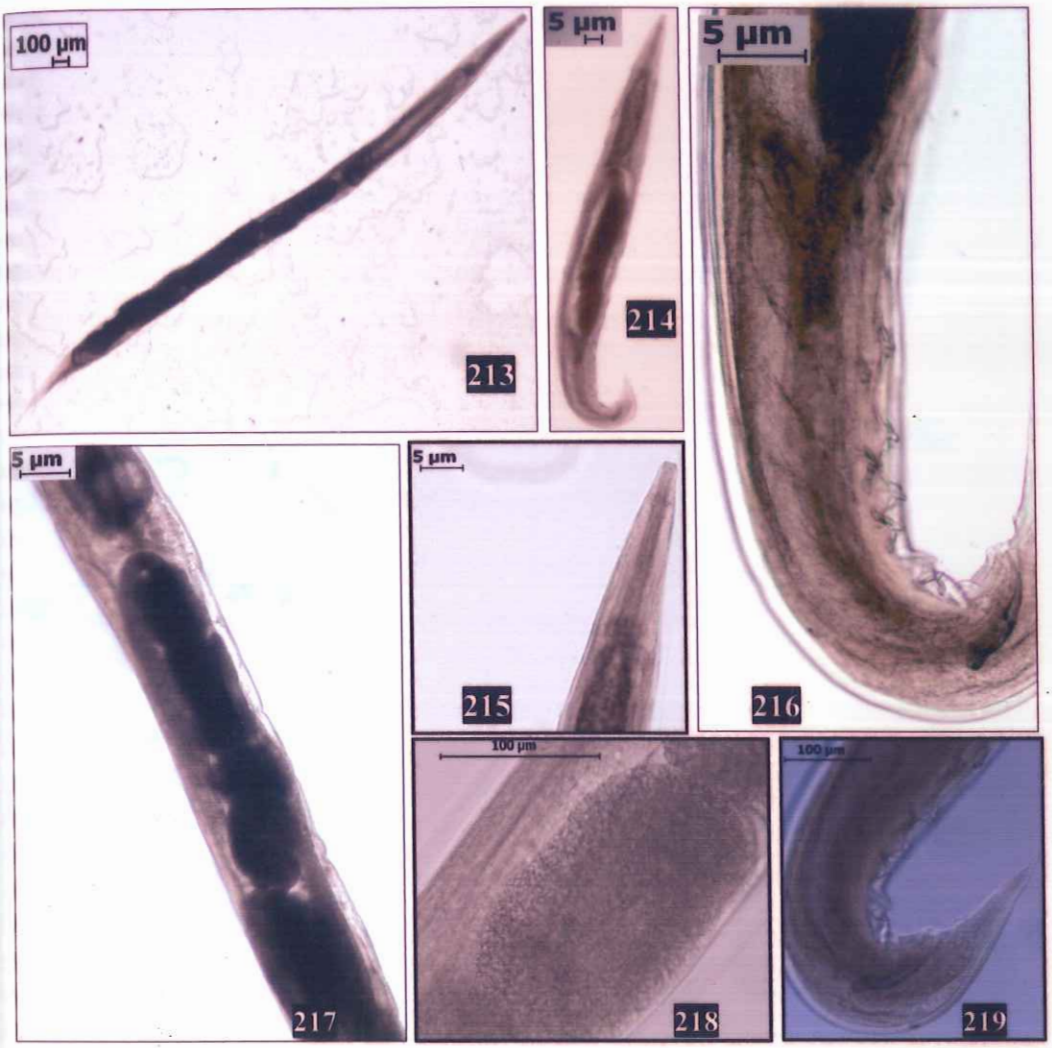


Fig. 212 ***Paracosmocerca mucronata***

a. Whole worm (female)
b. Whole worm (male)
c. Egg
d. Curve posterior end of a male, showing the five pairs of plectanes, spicule and the tip of tail



Figs.213-219 ***Paracosmocerca mucronata***- PhotomicrographS

213. Whole female worm

214. Whole male worm

215. Anterior end

216. Posterior end of male worm, showing the 5 pairs of plectanes and gubernaculum.

217. Mid region of female, showing eggs.

218. Egg

219. A magnified view of another male, showing the tip of tail end

Table 15 **Morphometric measurements of *Paracosmocerca mucronata*** Kung et Wu, 1945

Characters	Male			Female		
	Range	Mean	SD	Range	Mean	S.D
Length of the body	2.72-2.98	2.501	±0.189	3.23-4.12	3.651	±0.417
Maximum width of body	0.23-0.26	0.192	±0.027	0.16-0.17	0.164	±0.005
Pharynx:: Length	0.36-0.38	0.352	±0.010	0.38-0.46	0.421	±0.033
Breadth(base)	0.02-0.02	0.019	±0.002	0.02-0.03	0.022	±0.003
Breadth(Mid. Region)	0.03-0.03	0.022	±0.003	0.04-0.04	0.039	±0.003
Oesophageal bulb: Length	0.06-0.06	0.052	±0.003	0.09-0.11	0.150	±0.003
Breadth	0.05-0.05	0.044	±0.004	0.06-0.08	0.070	±0.004
Spicule: length	0.13-0.13	0.118	±0.006			
Tail: length	0.15-0.15	0.141	±0.006	0.27-0.34	0.293	±0.026
Spike: Length				0.27-0.34	0.038	±0.002
Distance of vulva from anterior end				0.27-0.34	1.637	±0.078
Egg size: Length				0.27-0.34	0.154	±0.016
Breadth				0.27-0.34	0.097	±0.007

Remarks:

P. mucronata was originally described from *Rana nigromaculata*, *R. günther*, *R. limnocharis*, *Bufo bufo*, *Microhyla ornata*, from China by Kung and Wu (1945). Gupta and Duggal (1980) added a new species, ***P. indica*** from *Rana* sp. from Chandigarh, describing it as the first representative of the genus from India and differentiating it from the type species in having 3 pairs of plectanes as against 5 pairs of plectanes in the latter. The present form, in having 5 pairs of plectanes definitely represents ***P. mucronata*** that is being recorded herein from a new locality (Northeast, India) and three new host species.

Subfamily Oxysomatiinae Railliet, 1916

Genus ***Oxysomatium*** Railliet et Henry, 1913

Oxysomatium macintoshii

(Figs.220-226)

159 specimens of ***O. macintoshii*** were recovered during the study period. The range per infected host being 1-28 (57 were male and 102 were female)

Body size

Male:	1.788-2.124 x 0.071-0.08 mm
Female:	2.21-3.4.5 x 0.122-0.198 mm
Egg size:	0.122-0.0203 x 0.066-0.152 mm

Host: *P. leucomystax*, *E. cyanophlyctis*, *H. annectans*

Location: Intestine

Locality: Zunheboto

Remarks:

This nematode species was earlier reported by Diengdoh (1989) from Meghalaya, Northeast India. In the present study Nagaland (Zunheboto, Mokokchung and Kohima) constitutes a new Locality record for ***O. macintoshii*** and *P. leucomystax*, *H. annectans P. annandalii*, *R. gongshanensis* and *E cyanophlyctis* are all new host records from the region for the species.

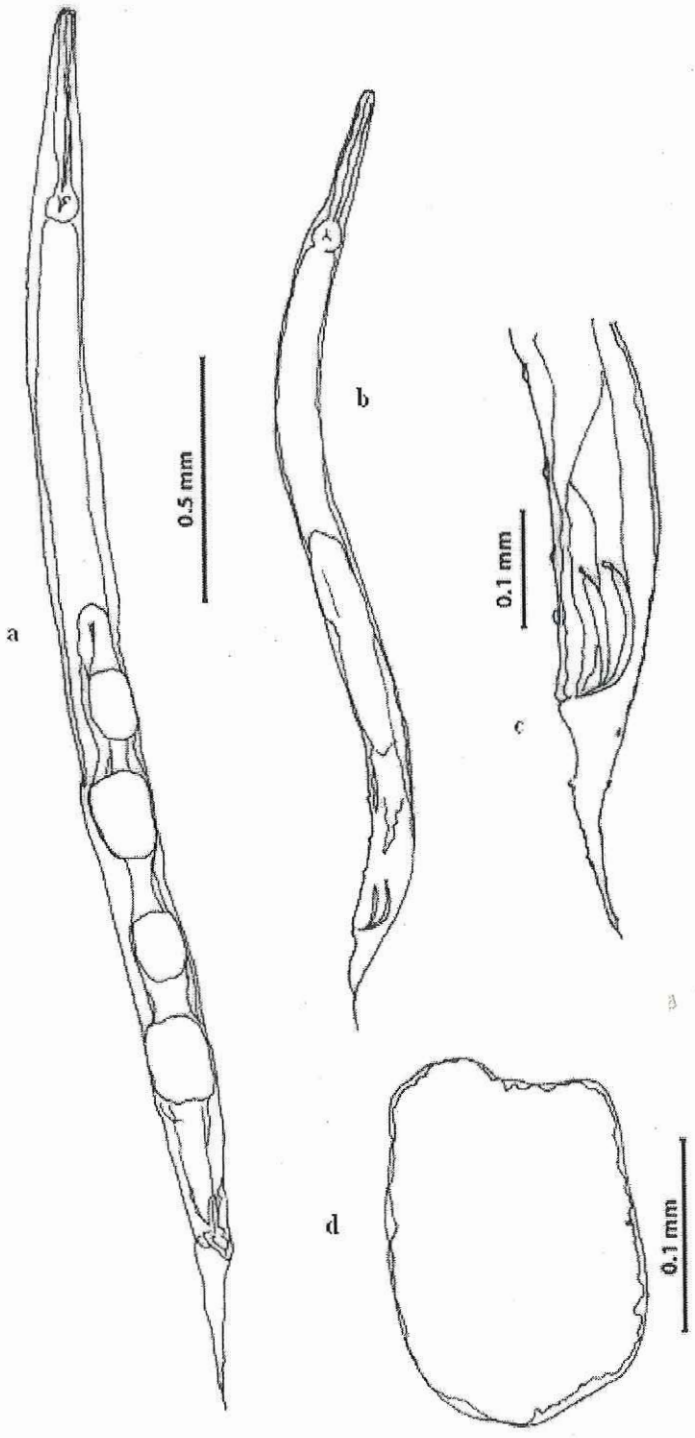
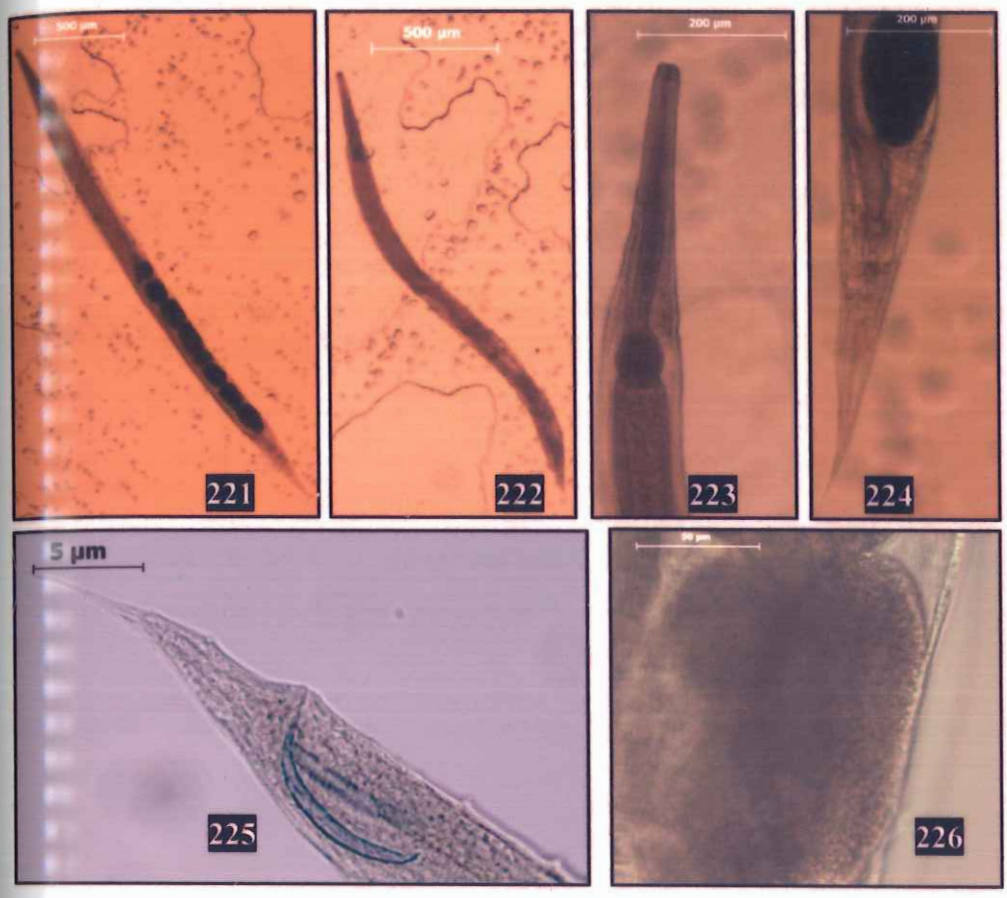


Fig. 220 ***Oxysomatium macintoshii***

a. Whole worm (female)
b. Whole male worm
c. Posterior end of male, showing the papillae and spicule
d. Egg



Figs. 221-226 ***Oxysomatium macintoshii***- Photomicrographs

221. Whole worm, female.

222. Whole worm, male.

223. Anterior end.

224. Posterior end of female, with egg (dark spot)

225. Posterior end of male showing the pre- and post anal papillae and spicule

226. A magnified egg, containing larva

Genus ***Oxysomatium*** (Railliet et Henry, 1916)

Oxysomatium sp.

(Figs. 227-231)

Numerous specimens of this form were collected during the study period from most of the host examined, the rate of infection being 1-120 per infected host. No male host was recovered throughout the study.

Body size: 3.95 x 0.24mm

Egg size: 0.04 x 0.08mm

Host: *E. cyanophlyctis*, *L. limnocharis*, *H. tigerinus*, *H. crassus*, *R. livida*, *Rana sp. 1*, *Rana* sp.3, *Rana danielii*, *M. microhyla*, *R. khare*, *R. maximus*, *R. bipunctatus*, *P. leucomystax*, *H. annectans*, *P. annandalii*, *Philautus sp.1*, *P. tarajensis*.

Location: Intestine

Locality: Dimapur, Kohima, Mokokchung, Zunheboto

Remarks:

Earlier it was reported by Diengdoh (1989) from North East India in Meghalaya and in Nagaland by Tandon *et al.* (2001) from a variety of host species. In the present study, *H. crassus*, *R. danieli*, *M. microhyla*, *H. annectans*, *P. annandalii*, *Philautus* sp. 1, *P. tarajensis* have been added as yet new hosts for ***Oxysomatium*** sp.

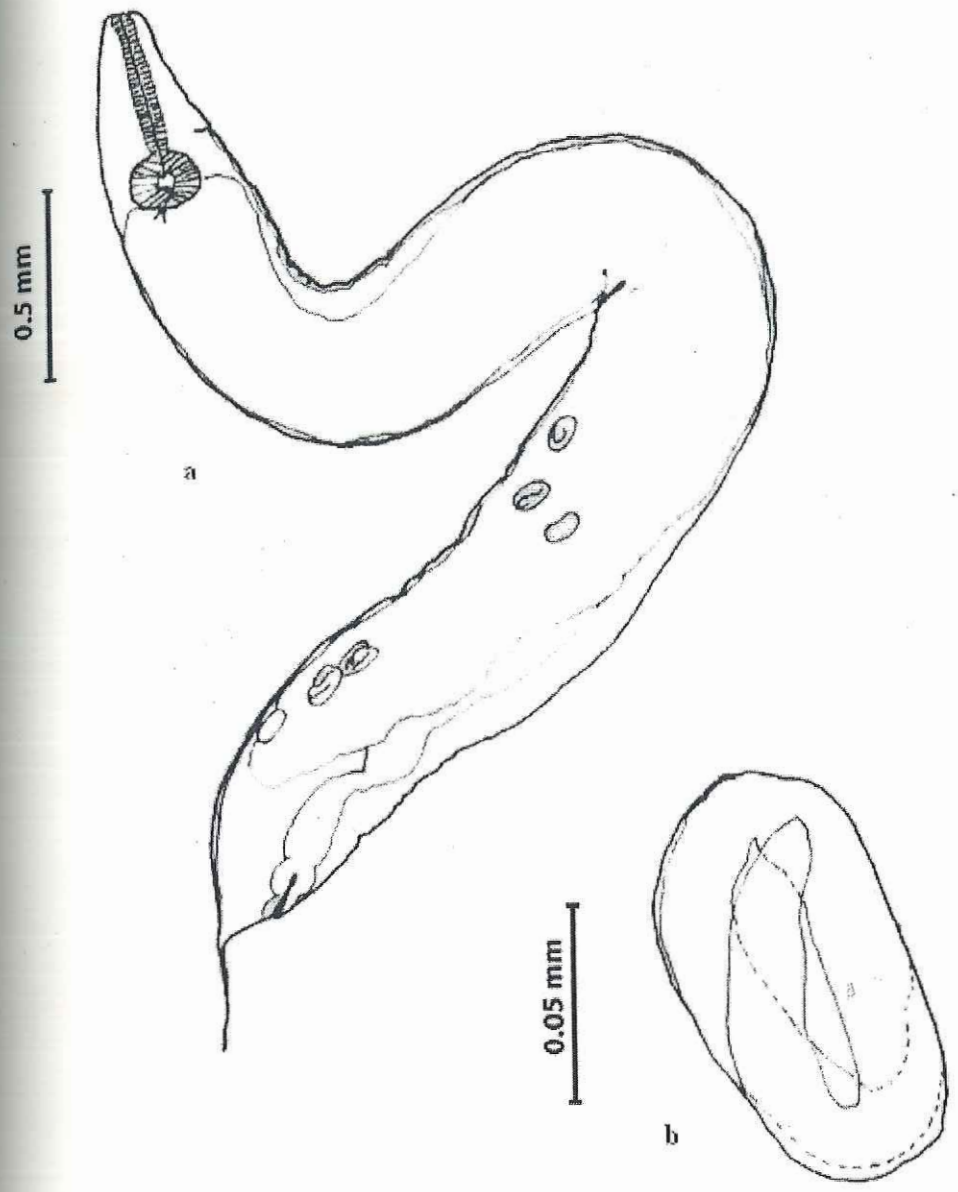
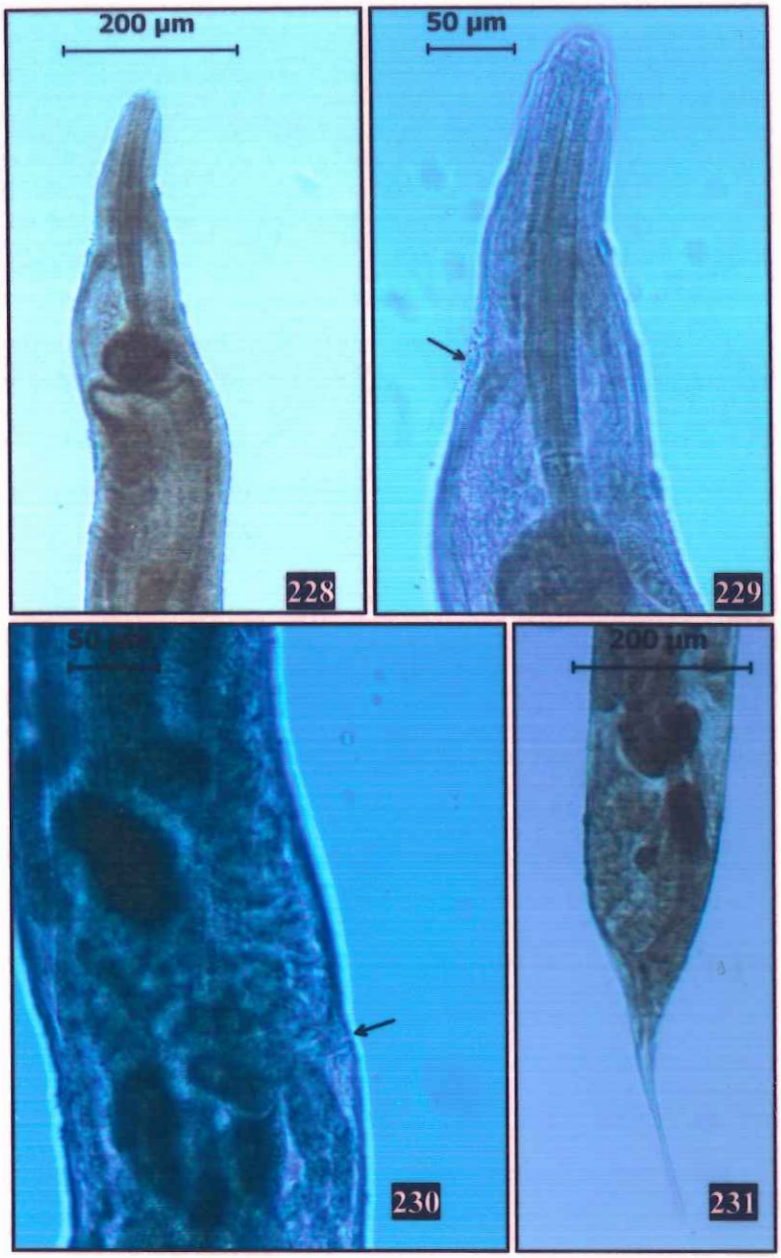


Fig. 227 *Oxysomatium* sp.

a. Whole worm (female)

b. Egg



Figs.228-231 *Oxysomatium* sp.- Photomicrographs

228. Anterior end of a female worm.

229. A view of the enlarged anterior end, showing the folding of cuticle (arrow).

230. Enlarge view of mid-region of the worm showing the genital pore (arrow).

231. Posterior end.

Superfamily Rhabditoidea

Family Rhabdiasidae (Railliet, 1916)

Genus ***Rhabdias*** (Stiles et Hassel, 1905)

(Syn. ***Ophiorhabdias*** Yamaguti, 1943; Singh et Ratnamala, 1967)

Rhabdias ranae Walton, 1929

(Figs.232-237)

Altogether 1151 specimens of this form were collected at several times from varied number of hosts during the study period; the collection always comprised female specimen only. The intensity of infection ranged from 1-16 per infected host. However, in some host species, abnormally high intensity of this species was encountered; especially in *R. maximus*, *H. tigerinus* and *Amolops marmoratus*, the number of parasites recovered were 203, 73 and 24 per host, respectively.

Body size: 10.88-11.27 x 0.29-0.31mm

Egg size: 0.09-0.1 x 0.051-0.052mm

Study under SEM revealed the surface fine topography of the worm with flabby inflated cuticle covering almost two third of the body length. Under higher magnification the mid-body region showed button-like papillae and in posterior bulb-like papillate structure.

Host: *Rhacophorus maximus*, *R. gongshanensis*, *R. bipunctatus*, *H. annectans*, *Theloderma* sp., *P. annandalii*, *Philautus* spp (1, 2 and 3), *P. leucomystax*, *P. taraensis*, *R. khare*, *Rana* sp.3, *R. danielii*, *H. tigerinus*, *E. cyanophlyctis*, *L. limnocharis*, *A. marmoratus*, *M. glandulosa* and *M. wuliangshanensis*.

Location: Lungs

Locality: Mokokchung, Kohima, Dimapur, Tuensang, Zunheboto

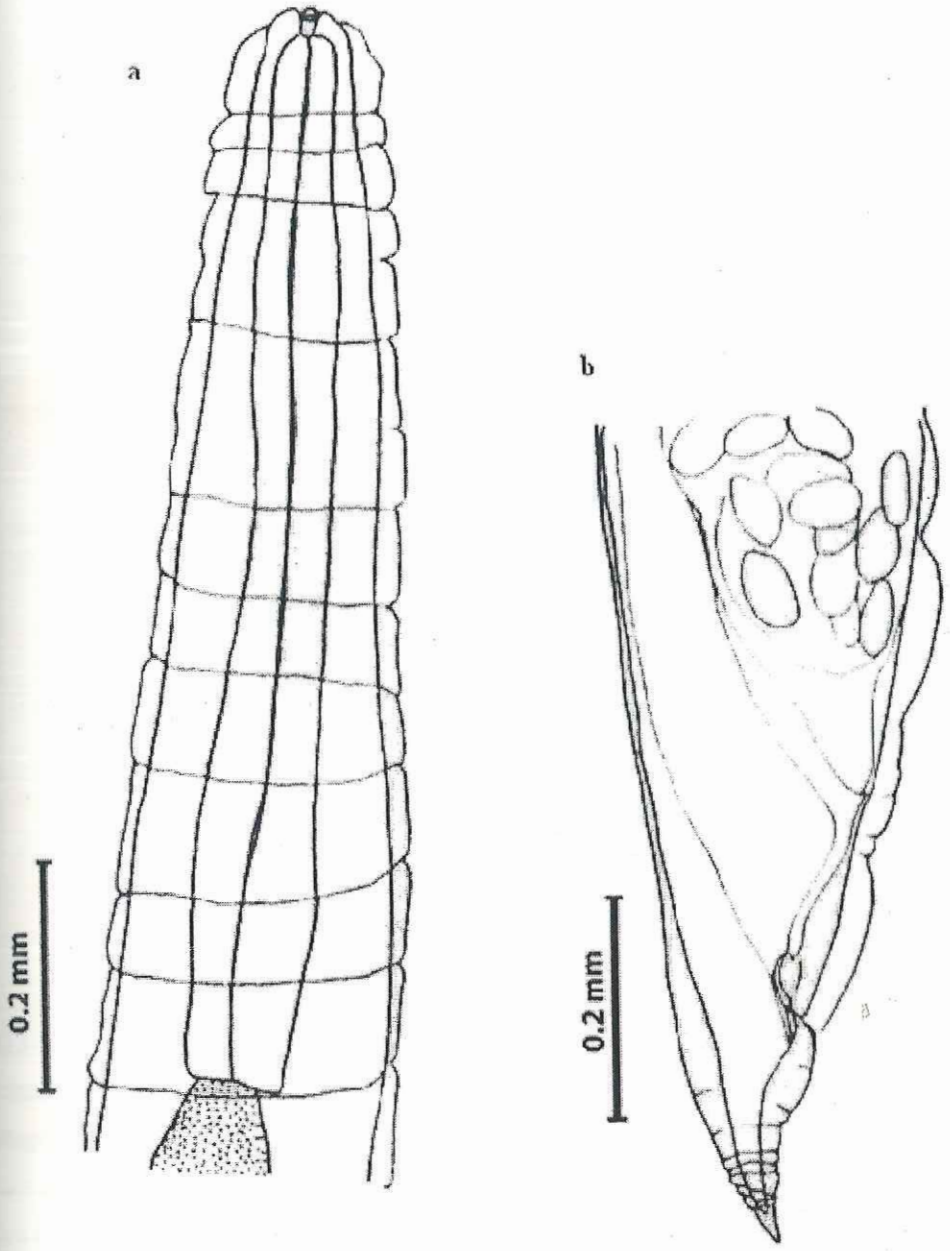
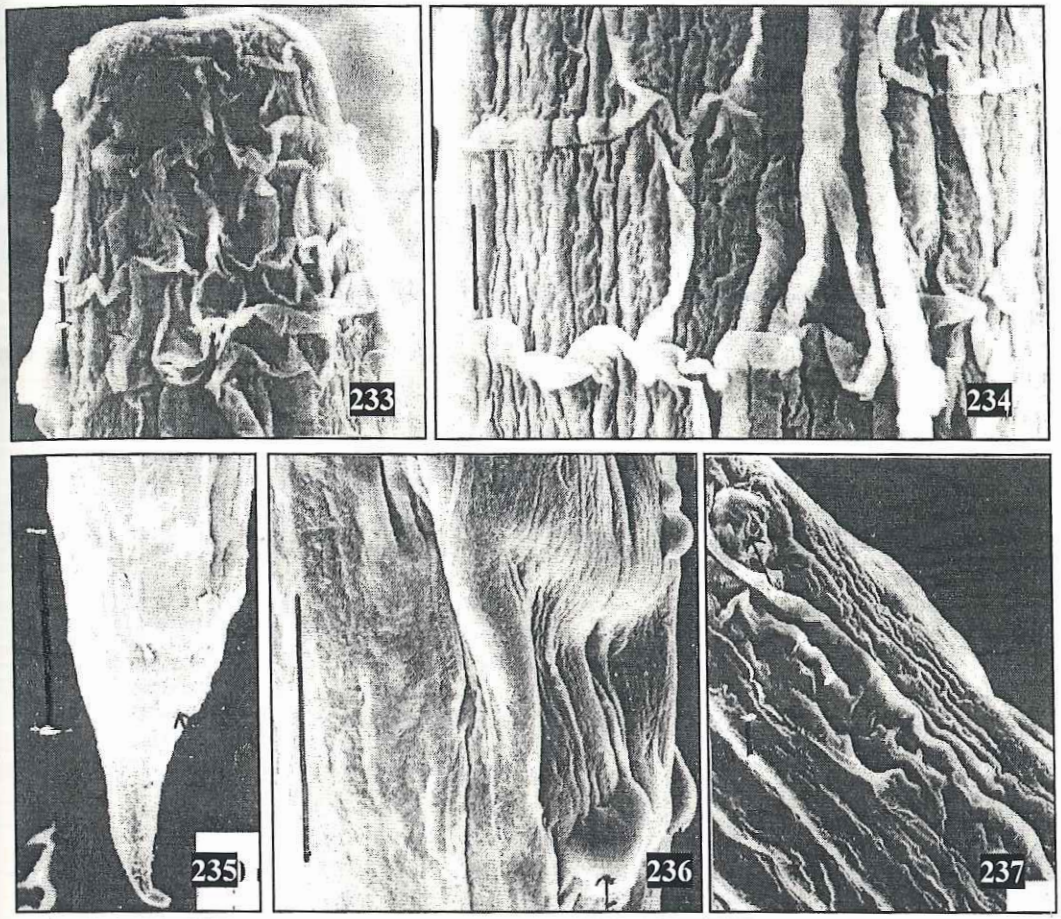


Fig. 232 ***Rhabdias ranae***

a. Anterior end of female
b. Posterior end with eggs in the utero



Figs. 233-237 ***Rhabdias ranae***- Scanning electron micrographs

232. A magnified view of anterior end, showing the flabby inflated cuticle (Scale bar=10μm)

234. Magnified view of mid anterior region, showing the extension of the inflated cuticle (Scale bar=10μm)

235. The tail end, showing the anal opening (arrow)(Scale bar=100μm)

236. A portion of the preanal region, revealing bulb-like papillate structures (arrow) (Scale bar=100μm)

237. A portion of mid-body region, showing cuticular inflation and button-like papillae (arrow) (Scale bar=10μm)

Remarks:

The first reporting of ***Rhabdias ranae*** from North-East India was done by Diengdoh (1989) from various localities of Meghalaya. Tandon *et al.* (2001) also reported from Nagaland, with the addition of some new hosts such as *H. annectans, R. bipunctatus, R. khare* and *E. cyanophlyctis* from the region. In the present study numerous specimens of this form were collected from many hosts, *R. gongshanensis, Theloderma* sp., *P. annandalii, Philautus* spp (1, 2, 3)1, *P. tarajensis, Rana* sp.3, *R. danielii, L. limnocharis, M. glandulosa* and *M. wuliangshanensis* all of which are recorded as new hosts for ***R. ranae.***

Superfamily Trichostrongyloidea

Family Molineidae (Skrjabin et Schultz, 1937) Durette-Desset et Chabaud, 1977

Subfamily Molineinae (Skrjabin et Schultz, 1973)

Genus ***Oswaldocruzia*** Travassos, 1917

(Syn. ***Oswaldocruzia*** (Bialata) Morishita, 1926)

Oswaldocrizia goezei Skrjabin et Schultz, 1952

(Syn. ***Ascaris filiformis*** Goeze, 1782 Partially; ***O. filiformis*** (Goeze, 1782), Travassos, 1917; ***O. indica*** Lal, 1949; ***O. melanosticti*** Gupta, 1960)

(Figs.238)

23 specimens, 7 males and 16 females, were collected during the study period.

Body size

Male: 5.33 x 0.11mm

Female: 11.5 x 0.21mm

Egg size: 0.06-0.07 x 0.02-0.03mm

Host: *E. cyanophlyctis*

Location: Intestine

Locality: Kohima, Dimapur

Remarks:

Earlier ***O. goezei*** was reported from Meghalaya, Northeast India, by Diengdoh (1989) and also from Kohima, Nagaland by Tandon *et al.* (2001). The present study reports Dimapur a new locality from Nagaland for the species.

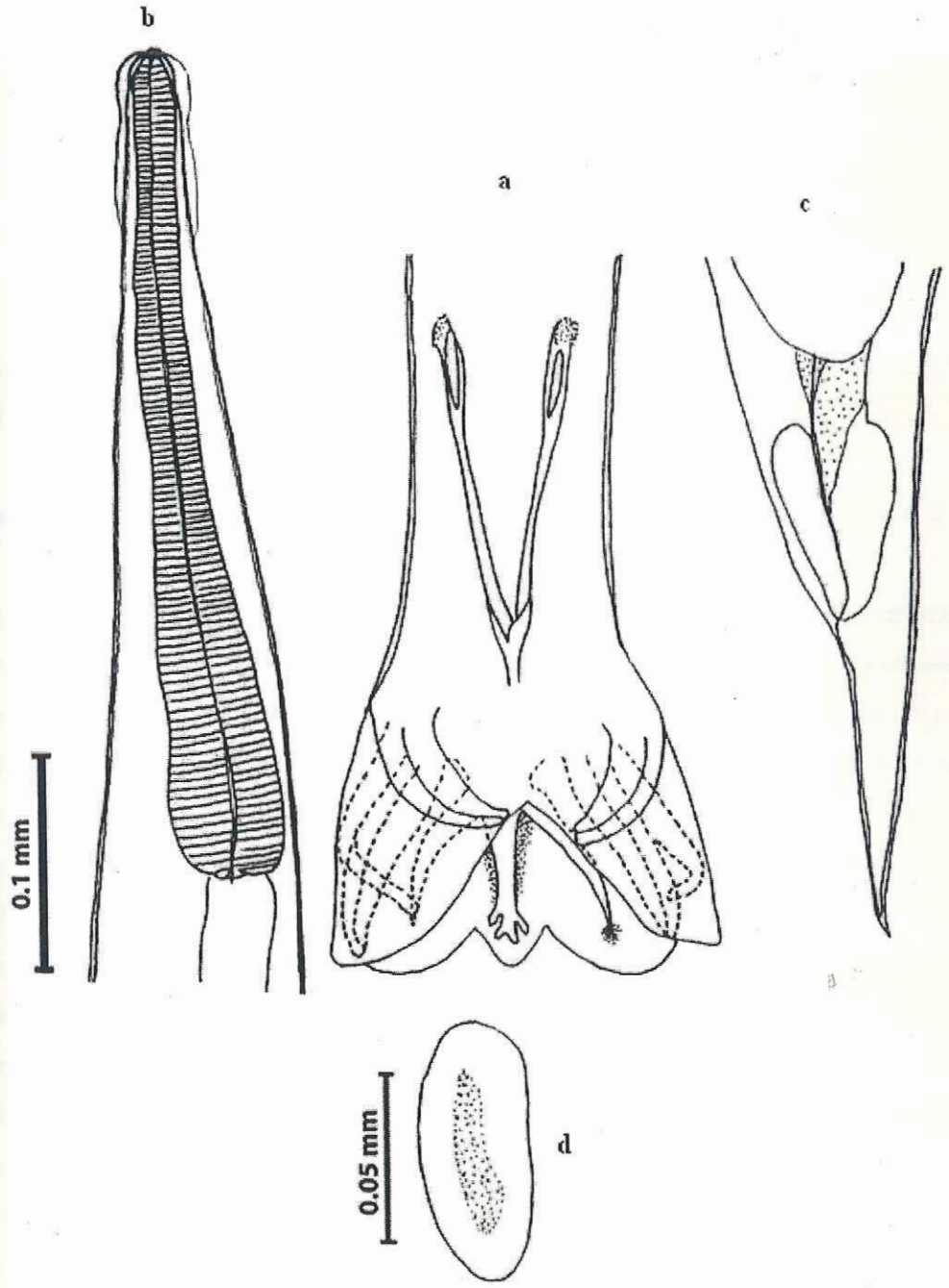


Fig. 238 ***Oswaldocruzia goezei***

- **a.** Posterior end of male, showing bursa with 6 pairs of rays and a centrally placed ray
- **b.** Anterior end
- **c.** Posterior end of female
- **d.** Egg

Family Ascarididae Baird, 1855

Subfamily Angusticaecinae Skrjabin et Karokhin, 1945

Genus ***Ophidascaris*** Baylis, 1920

***Ophidascaris* sp.**

(Figs. 239-252)

Altogether 203 specimens, 30 males and 173 females, were collected during the study, the infection rate being 1-8 per infected host.

Description (based on 5 male and 5 female specimens and SEM observations of 2 males and 1 female specimen):

Long, large stout worm, with fine transverse striation on the cuticle. Cervical alae prominent, lateral alae absent. Mouth surrounded by 3 small lips; interlabia elevated by 3 pairs of cartilaginous rod-like extensions from base of lips. Deep transverse groove running round base of lips. Oesophagus elongated, cylindrical, entirely muscular with slightly dilated posterior region; oesophageal bulb absent. 5 pairs of papillae in anterior region, covering entire length of oesophagus. Intestinal caecum absent.

Male

Spicules 2, subequal, of moderate length. Tail tapering to bluntly pointed end; caudal alae absent. Fairly numerous sessile pre- and post-cloacal papillae present; of postcloacal papillae 5 pairs ventrolateral, 2 lateral, a single papilla near tip of tail, 1 pair in line of cloacal opening; precloacal papillae 2 pairs ventrolatertal, a single large papilla near cloacal opening, anterior most papillae positioned ventrally.

Female

Ovaries 2, short. Vulva in posterior half of body; vagina running anteriorly for a short distance, branching into 2 uterine branches (amphidelphic). Eggs few, moderately large; ovoviviparous condition. Tail blunt with sharp point.

Under SEM the body tegument reveals fine transverse striations, the male showing numerous pre- and postcloacal papillae; in the female the round posterior extremity shows a short spike- like tail and wrinkled cuticular surface.

Morphometric measurements of this form are given in Table 16

Host: *A. marmoratus*

Location: Intestine

Locality: Mokokchung

Remarks:

The form was assigned to the genus ***Ophidascaris*** by Tandon *et al.* (2001) in having the following characteristic features – Interlabia present, oesophagus without posterior bulb or ventriculus; male with fair number of papillae; spicule, gubernaculums absent. Members of the genus ***Ophidascaris*** are mainly parasites of snakes and lizards, occasionally of amphibians. Two species, namely, ***O. gestri*** Parona, 1890 and *O.ajaris* Khera, 1956 have been reported from reptiles of India. Only ***O. labiadopapillosa*** Walton, 1927, has been recorded to be naturally occurring in *Rana* species in USA (Ash and Beaver, 1962).

In the present study numerous specimens of this form, both male and female, were collected from the same host, *A. marmoratus* Blyth, 1855, but never from any of the host species collected from the same spots, the worm seem to maintain a specific host. , The species identification is being kept in abeyance, due to pending examination of and comparison with the hitherto known species of the genus.

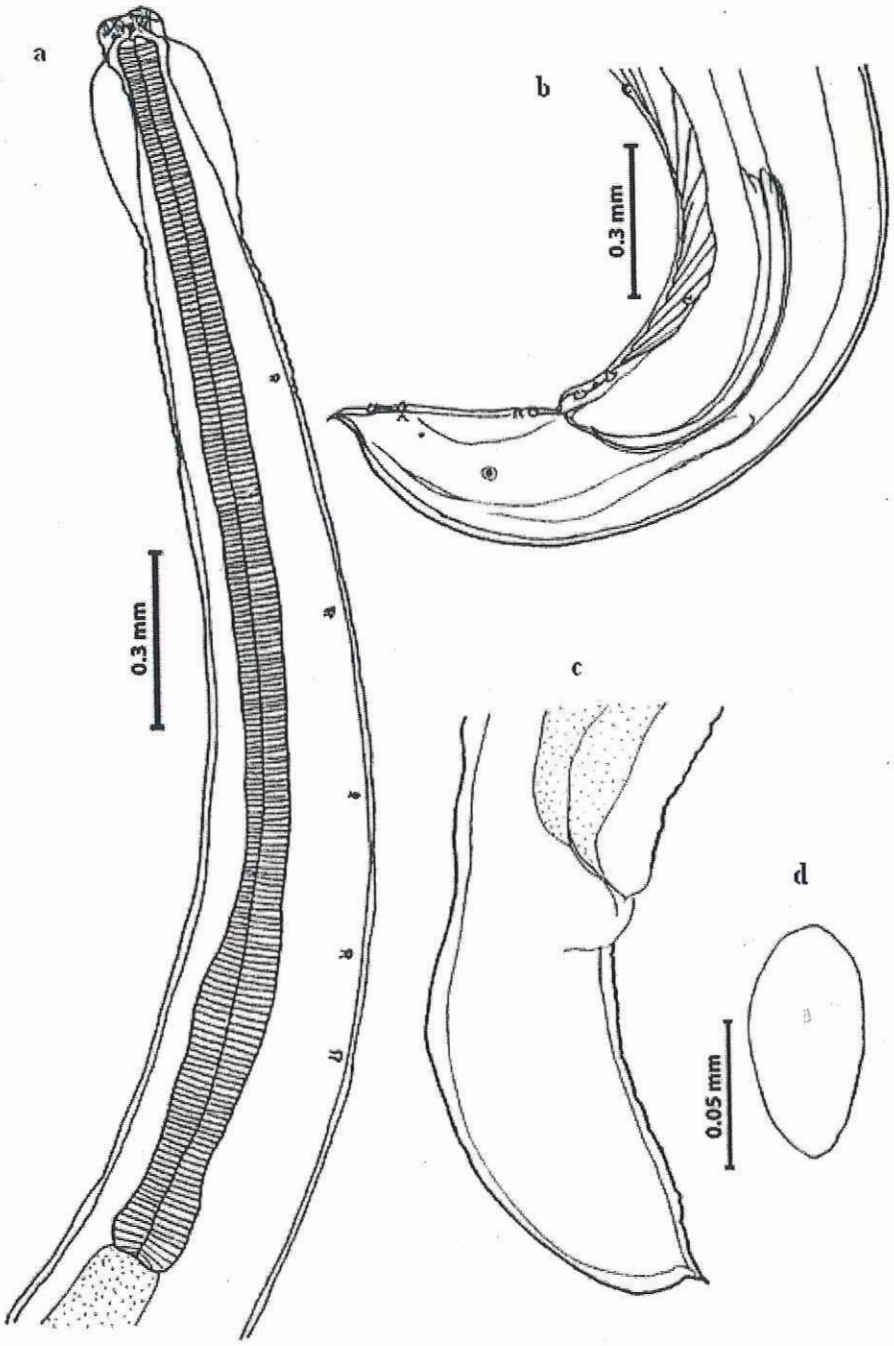


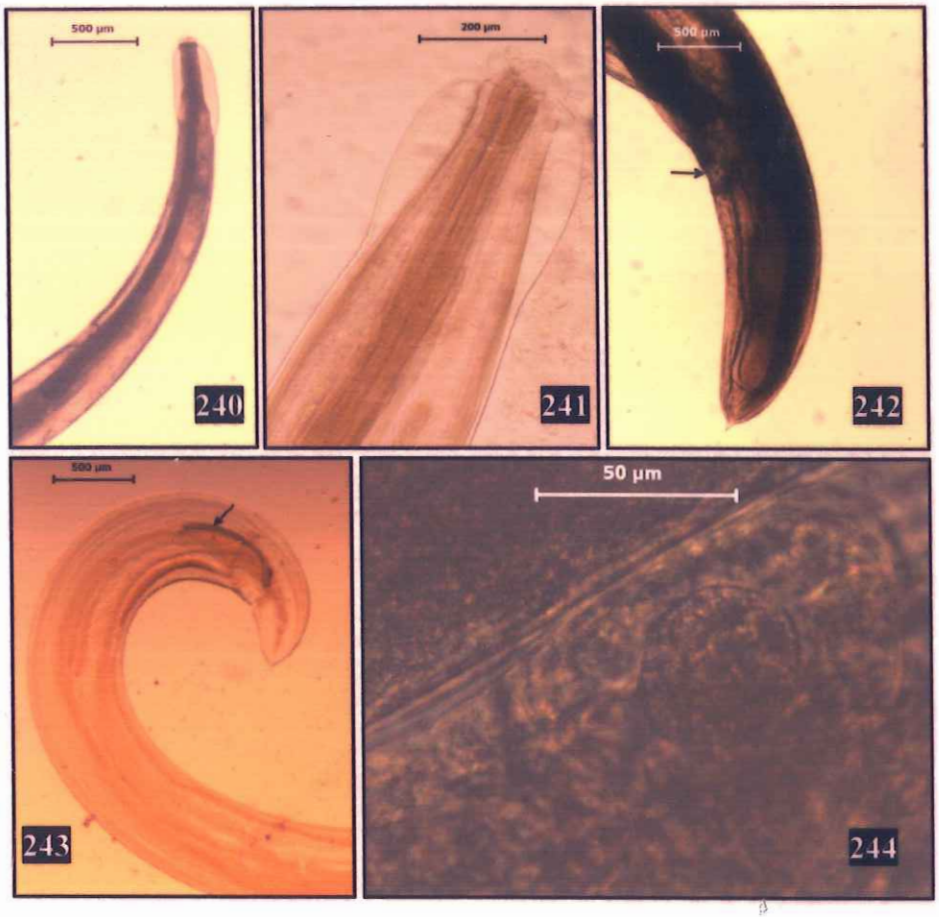
Fig. 239 ***Ophidascaris*** sp.

a. Anterior end of male worm

b. Posterior end of male, showing the spicules and papillae

c. Posterior end of female worm

d. Egg



Figs. 240-244 *Ophidascaris* sp.- Photomicrographs

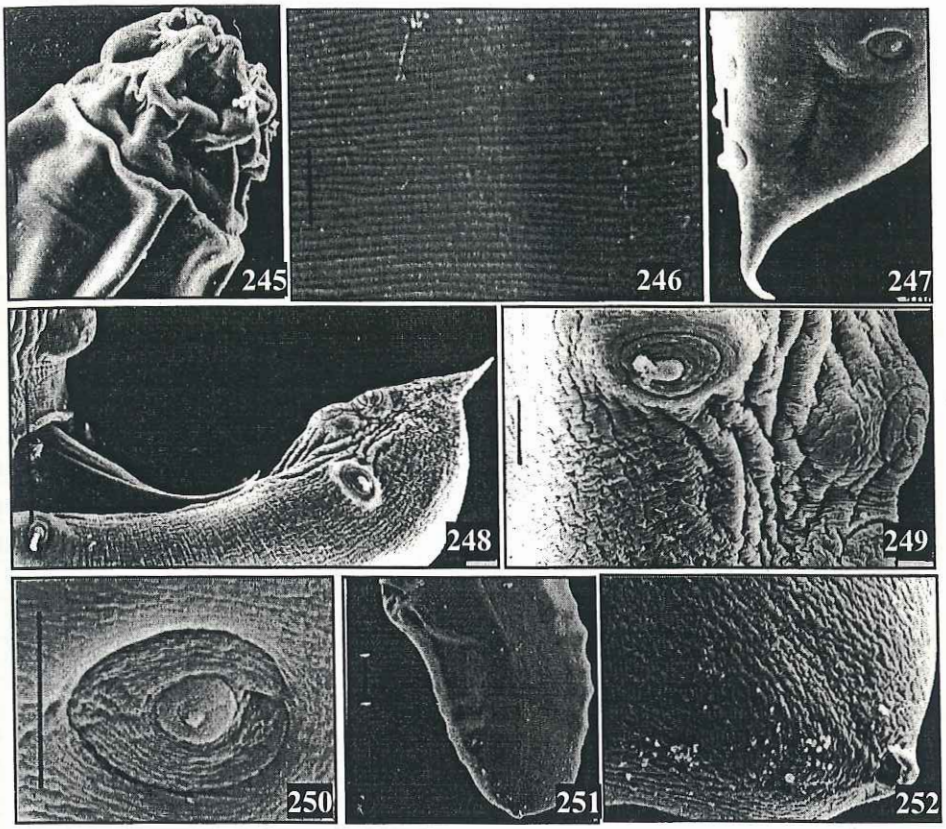
240. Anterior end of male worm with the long oesophagus

241. Anterior end of female, showing the cervical alae

242. Tail end of female

243. Posterior end of male, showing the spicule (arrow)

244. Eggs



Figs. 245-252 ***Ophidascaris*** **sp.-** Photomicrographs

245. Anterior end of male specimen, showing lips and a cervicle alae (Scale bar=10μm)

246. A magnified view of the body, showing fine striations of the cuticle (Scale bar=10μm)

247. Posterior end of a male, showing the post cloacal papillae and tip of the tail (Scale bar=10μm)

248. Posterior end of another male, showing the pre- and postcloacal papillae and the spicule (Scale bar=10μ)

249. Post cloacal papillae in a closer view (Scale bar=10μm)

250. A magnified post cloacal single papilla (Scale bar=10μm)

251. Posterior end of female, showing the anus and tail (Scale bar=10μm)

252. A magnified view of the tail end of female, showing spike-like terminal end and wrinkled cuticular surface (Scale bar=10μm)

Table 16 Morphometric measurements of *Ophidascaris* sp.

	Male			Female		
Characters	**Range**	**Mean**	**SD**	**Range**	**Mean**	SD
Length of the body	50.8-52.79	51.7680	±0.996830	51.73-76.2	66.01200	±13.063165
Maximum width of body	0.01-0.013	0.0106	±0.001342	0.01-0.02	0.01600	±0.005477
Oesophagus: Length	0.022-0.025	0.0234	±0.001517	2.94-3.04	3.00000	±0.054772
Mid region Breadth	0.002-0.011	0.0042	±0.003834	0.002-0.005	0.00380	±0.001095
Base Breadth	0.002-0.004	0.0030	±0.001000	0.002-0.006	0.00380	±0.001789
Spicule: Length	0.002-0.003	0.0022	±0.000447			
Tail: Length	0.004-0.007	0.0056	±0.001342	0.011-0.014	0.01280	±0.001643
Distance of vulva from anterior end				39.19-40.34	39.88000	±0.629881
Egg Length				0.0004-0.0005	0.00046	±0.000055
Breadth				0.0003-0.0004	0.00036	±0.000055

Superfamily Ascaroidea Railliet et Henry, 1915

Family Heterocheilidae Railliet et Henry, 1915

Genus ***Amplicaecum*** (Baylis, 1920)

***Amplicaecum* sp.**

(Fig.253)

Only 13 specimens (only male) were recovered during the study period from 3 species of ranid host, L. limnocharis*H. tigerinus* and *H.crassus*.

Body size: 10.11-11.81 x 0.48.2-0.55 mm

Host: *H. tigerinus*, *H.crassus*

Location: Intestine

Locality: Dimapur

Remarks:

The presence of lips with dentigerous ridges, small interlabia, absence of posterior bulb in the oesophagus and ventriculus, presence of intestinal caecum and male with fairly numerous papillae are the characters which conform to the genus ***Amplicaecum***. Hence, the present form is assigned to this genus. Tandon *et al.* (2001) reported the genus for the first time from Nagaland, North-Eastern region of the Indian subcontinent.

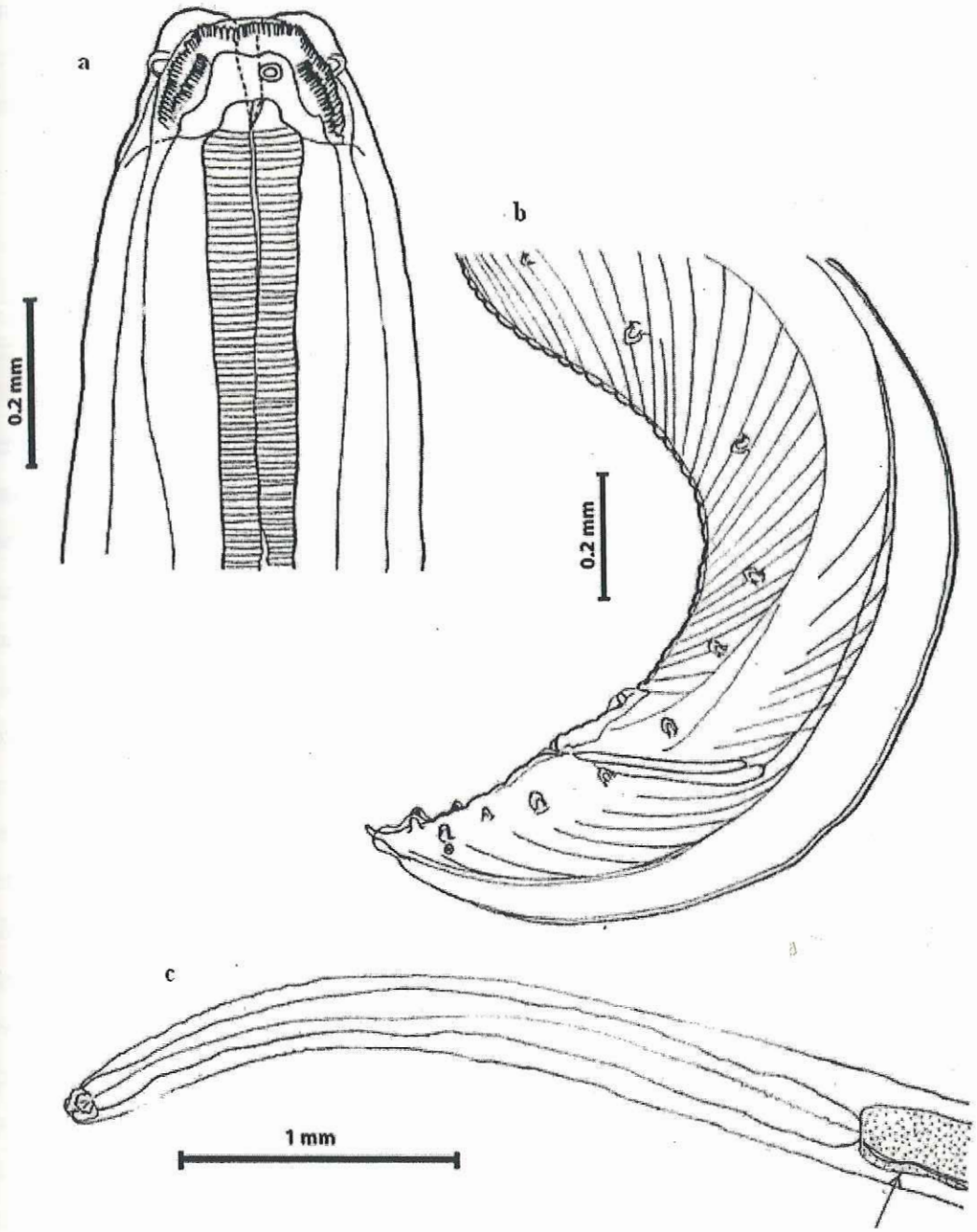


Fig.253 ***Amplicaecum*** **sp.**

- **a.** Anterior end of male
- **b.** Curve posterior end of male, showing the papillae and spicule
- **c.** Anterior end, showing the intestinal caecum (arrow)

Family Onchocercidae (Leiper, 1911)

Subfamily Icosiellinae (Anderson, 1958)

Genus ***Icosiella*** (Seurat, 1917)

Icosiella sp.2 of R. Imkongwapang, 1997

(Figs.254-255)

25 specimens of this very fragile and slender worm, 7 males and 18 females, were recovered from the peritoneal wall and peritoneal cavity of the host in the study period.

Description (base on measurements and observations of 3 mature males and 5 mature females)

Body filiform, cuticle thin and transparent, lateral flanges absent. Mouth surrounded by 4 rounded papillae. Oesophagus remarkably long, clearly divided into short anterior muscular region and a long glandular posterior region.

Male

Spicule subequal, moderate length. Tail short, tapering to bluntly pointed end, caudal alae absent, papillae present, 1 ventrally located near cloacal opening, 6 postcloacal papillae elevated in vesicle, located ventrolaterally, 1 singly positioned papilla near tip of tail. Posterior region having numerous small tubercles on ventral side that extends up to middle third of posterior end.

Female

Ovaries 2, tubular, entangled. Uterous running short distance anteriorly from posterior side, opening into ovijector. Vulva in anterior part of posterior half of glandular oesophagus. Larvae scattered in most parts of pseudocoelome, ovoviviparous. Eggs oval, tail short, and blunt/rounded.

Body size

Male: 16.5x0.5mm

Female: 18.4-28.2 x 0.98mm

Host: *A. marmoratus*

Location: Peritoneal wall, peritoneal cavity

Locality: Mokokchung

***R. Imkongwapang, 1997; M.Phil. Thesis, A study on the helminths parasite spectrum in anuran Amphibia in Nagaland.** School of life sciences, Department of Zoology, North Eastern Hill University, Shillong, Meghalaya.

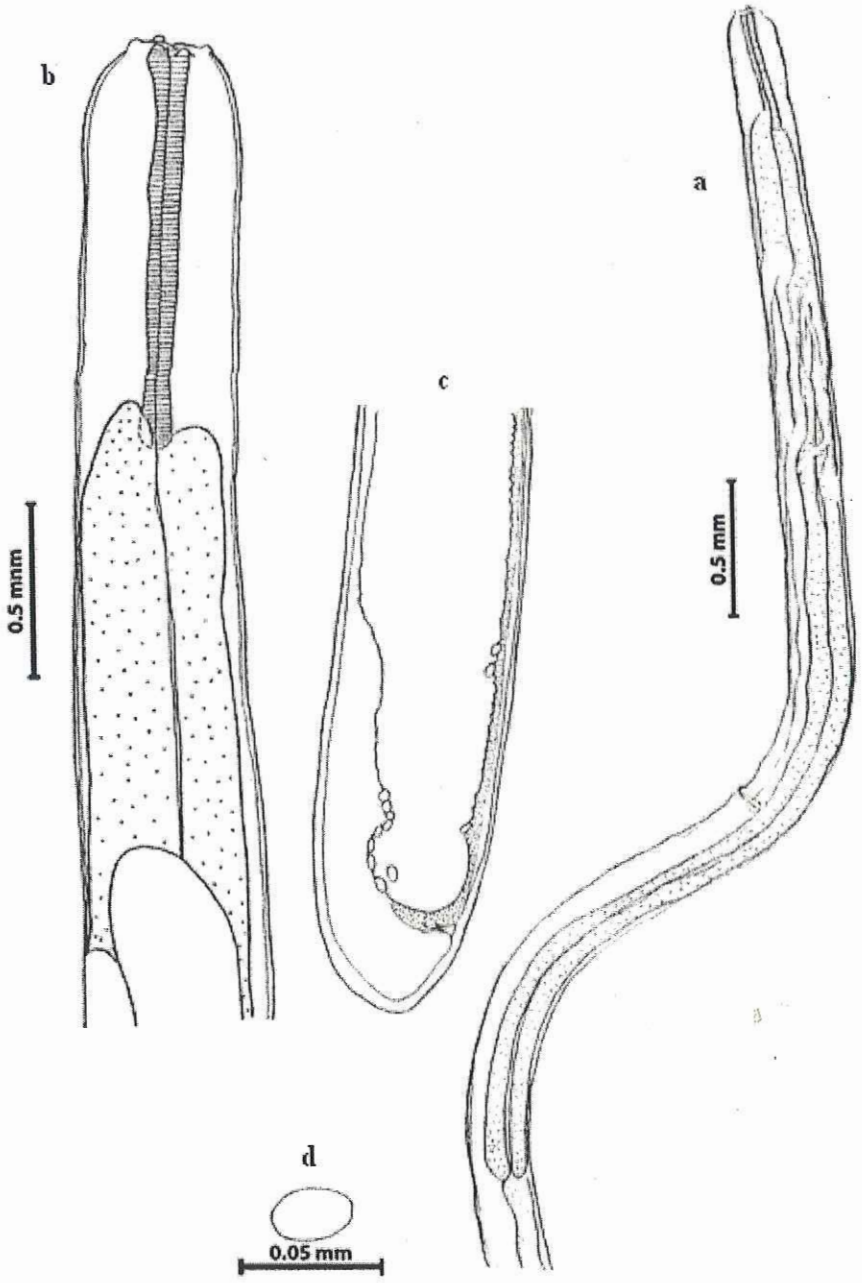


Fig. 254 ***Icosiella*** sp. (female)

a. Anterior end of female worm, showing the vaginal opening and the long glandular oesophagus

b. Enlarged anterior end, showing the muscular pharynx

c. Tail end

d. Egg

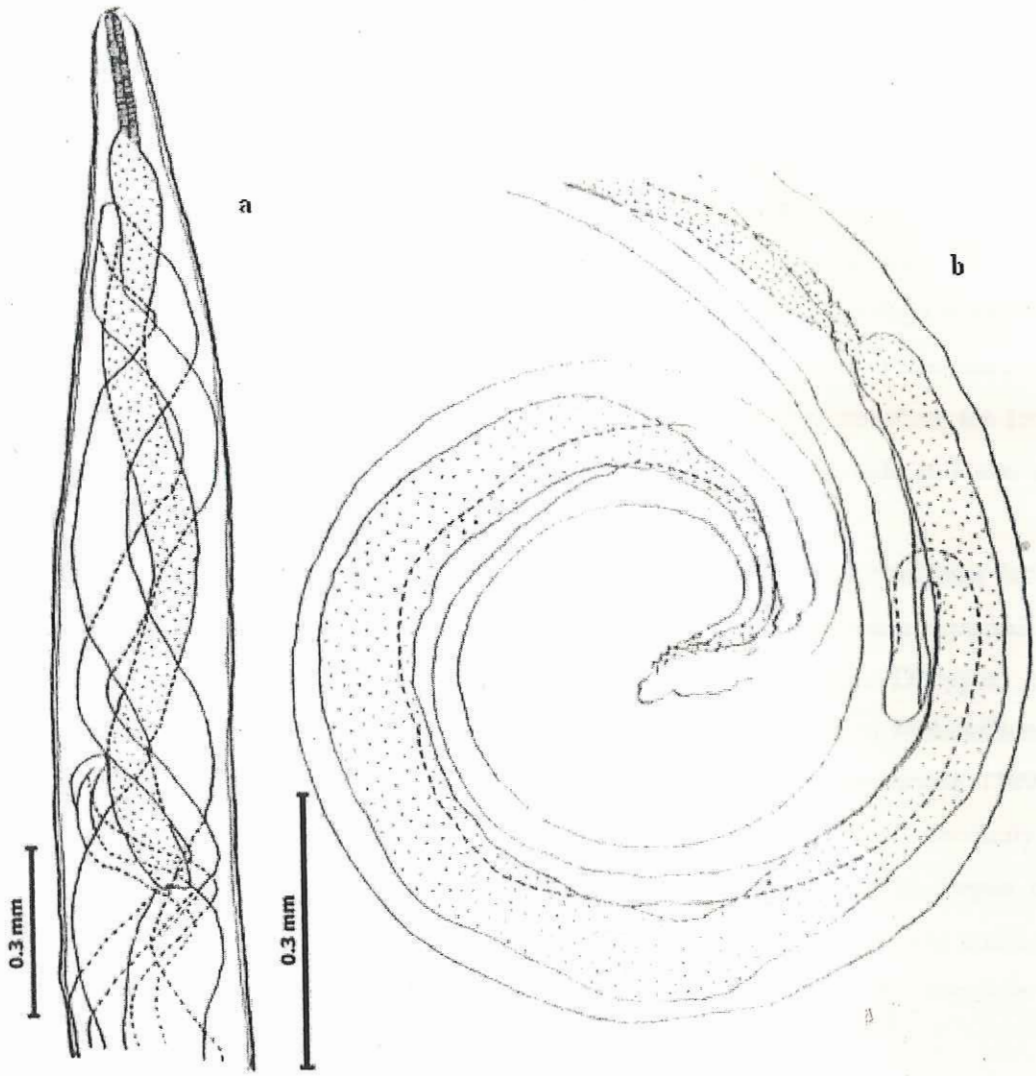


Fig. 255 ***Icosiella*** sp. (Male)

a. Anterior end of male

b. Posterior end of male, showing the coiled tail and the post-anal papillae

Remarks:

The present form is assigned to the genus ***Icosiella*** on account of the following features: body filiform, cuticle thin and transparent, lateral flanges absent, long oesosphagus comprising a short anterior muscular region and a long glandular posterior region, vulva opening at level of glandular oesophagus. It was also considered different from the earlier described species by the following features; the shape of the body is thread-like and filiform, the cuticle thin and transparent, the body length was more, the vagina was not muscular and the genital opening at the anterior region of posterior half of the glandular region of the oesophagus and the habitat was tissue-peritoneal cavity, therefore, the form was assigned to ***Icosiella*** sp.2. Other than addition of the same host, ***Icosiella*** sp.2 was not encountered from other new host.

In earlier survey and study of helminth parasites of anuran host in the Northeast India, Imkongwapang (1997) had encountered two filaroid nematodes that were reported as ***Icosiella*** sp.1 (from intestine of *E. cyanophlyctis* and *P. leucomystax*, Dimapur) and Icosiella sp.2 (from peritoneal wall tissue of *A. mamoratus*, that too from Mokokchung). The present form fully tallies in description with ***Icosiella*** sp.2 of Imkongwapang, (1997). The present observations also reconfirm the restricted distribution and host specificity of this filaroid nematode. However, since male specimens recovered were so fragile and disintegrated on handling and processing, good whole mounts of these specimens could not be prepared. Therefore, for want of more material of male specimens to be available for study, the species identification is being kept in abeyance.

ACANTHOCEPHALA

Family Echinorhynchidae (Cobbold, 1989)

Subfamily Echinorhychinae (Travassos, 1920, a)

Genus ***Acanthocephalus*** (Koelreuther, 1771)

Acanthocephalus bufonis (Shipley, 1903) Southwell et Maefie, 1925

(Syn. ***Echinorynchus bufonis*** Shipley, 1903; ***Acanthocephalus sinensis*** Van Cleave, 1937)

(Figs.256-262)

83 specimens (15 male and 68 female) of this form were collected during the study period, the range of infection per infected host being 1-8

Body size

Male: 8.82-14.53 x 0.98-1.56mm

Female: 11.52-52.52 x 1.74-1.94mm

Host: *E. cyanophlyctis*, *H. tigerinus*, *Rana* sp.1, *R. ievida*, *R. khare, A. marmoratus, M. glandulosa* , *P. leucomystax*, *P. taraensis*

Location: Intestine

Locality: Mokokchung, Tuensang, Kohima

Remarks:

Acanthocephalus bufonis was earlier reported from *H. tigerinus* of Meghalaya, by Diengdoh (1989) and from *E. cyanophlyctis* and *P. leucomystax* of Nagaland by Tandon *et al.* (2001). In the present study, *M. taraensis*, *R. khare*, *R. livida*, *A. marmoratu,* and *M. glandulosus* constitute new host records for this acanthocephalan species.

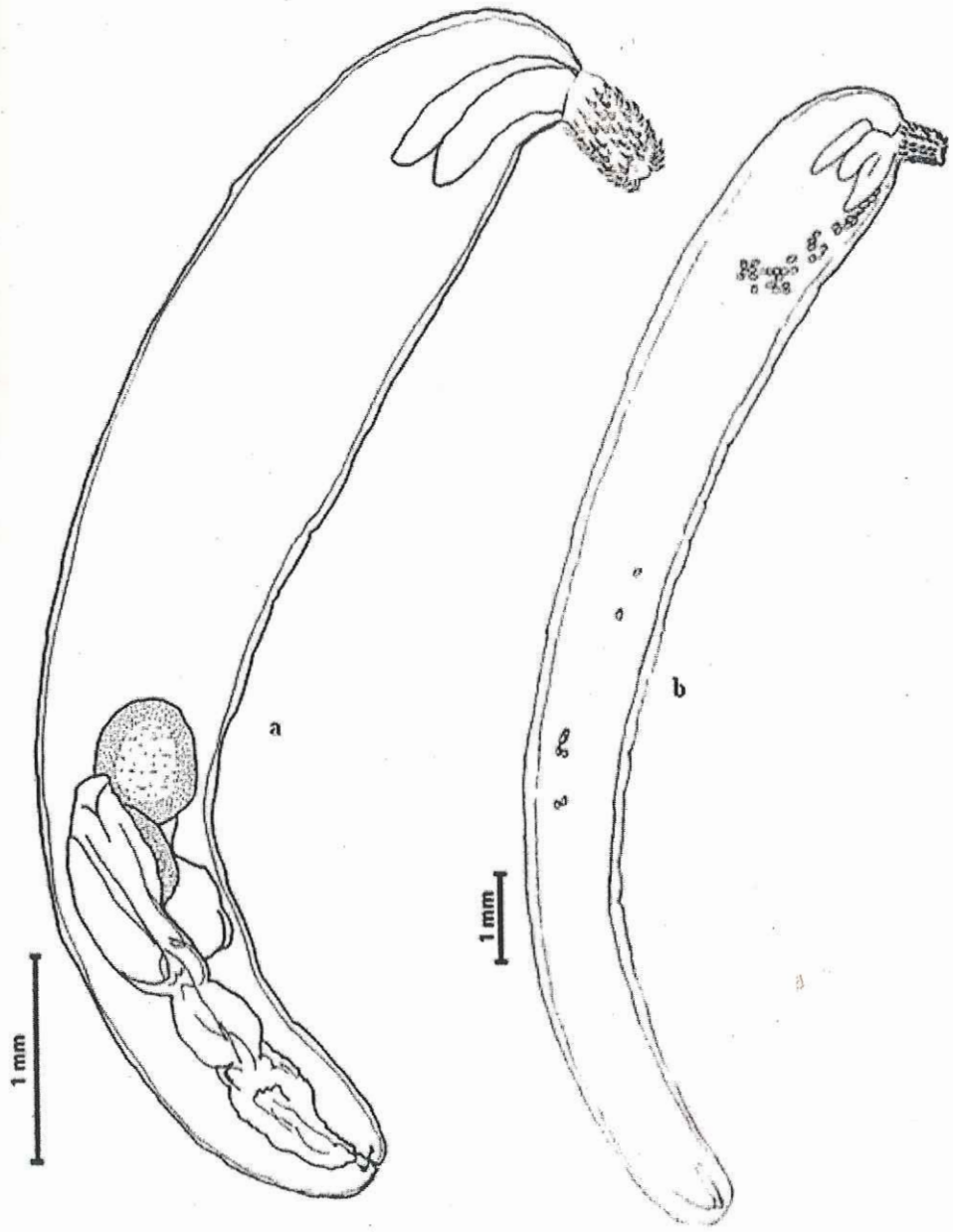
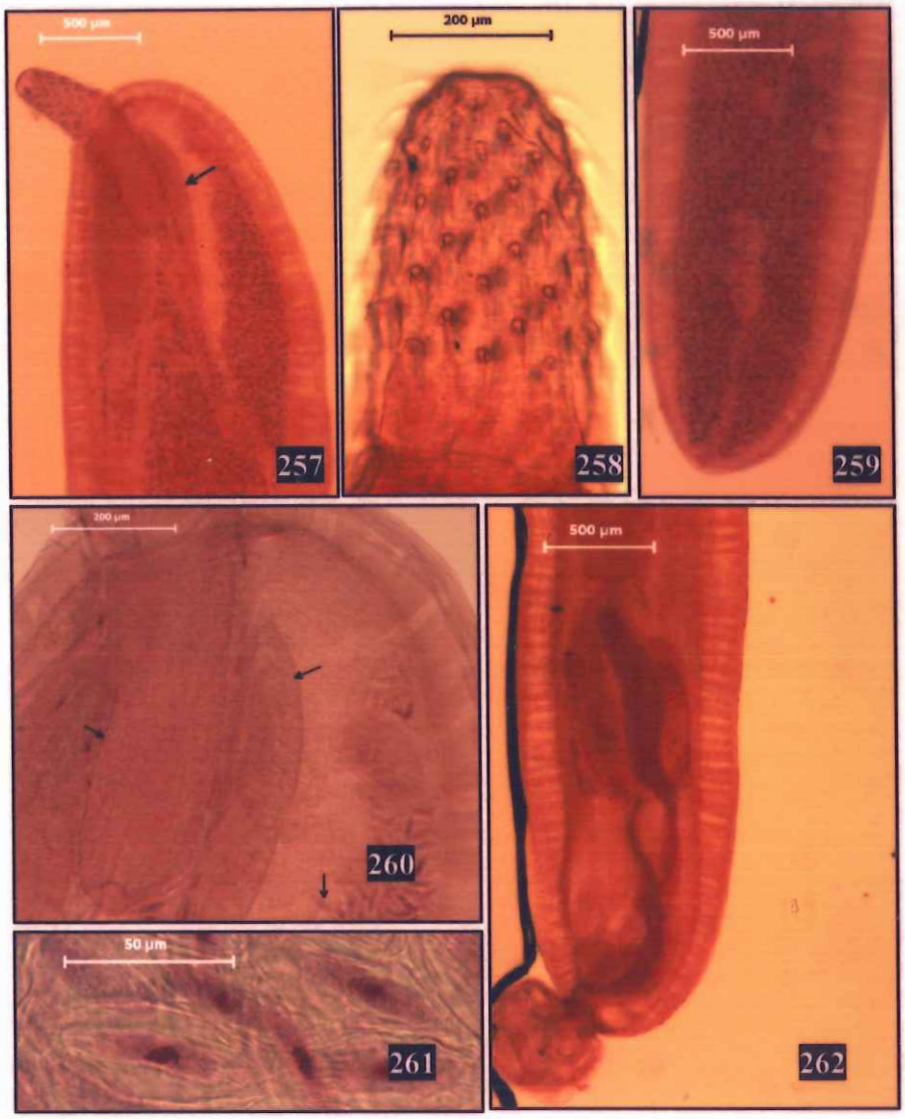


Fig. 256 *Acanthocephalus bufonis*

a. Whole male worm

b. Whole female worm



Figs. 257- 262 ***Acanthocephalus bufonis***- Photomicrographs

257. Anterior end of female worm, showing the (arrow)

258. A magnified view of the head, showing the spines

259. Posterior end of the female, showing the numerous eggs

260. Magnified anterior end of female, showing the eggs, empty sac of the retractable head and the limnisci (arrow)

261. Eggs

262. Posterior end of a male specimen

PREVALENCE AND STATUS OF HELMINTH PARASITES

Prevalence and status of helminth parasites

The present study is an extension of the earlier work carried out by R. Imkongwapang (1997); 16 more frog species and 2 newer localities (Tuensang and Zunheboto) were explored, with re-examination and resurveying of the earlier frog hosts and localities. Thus, the results presented herein are the compilation of overall data collected during the period 1997-2007.

Altogether, 1464 (one thousand four hundred sixty four) anuran hosts (frogs) comprising 29 species falling under 5 families and 15 genera from Nagaland were examined for helminth parasites infection in the study. The collection comprised a total of 839 anurans from Mokokchung (22 species), 291 from Kohima (10 species), 297 from Dimapur (10 species), 20 from Zunheboto (3 species) and 8 (4 species) from Tuensang. Only 1 species was found to occur throughout the year and was collected from 4 of the 5 localities surveyed.

Of the 29 host species, 13 (thirteen) were **arboreal** and 16 (sixteen) were **terrestrial** in habit.

The frog hosts from all the localities were found infected by at least 1 or more of the 39 helminth parasites reported herein viz., 5 monogeneans; 16 trematodes (13 adults and 2 metacercaria); 4 cestodes (3 adult and 1 larval); 14 nematodes and 1 acanthocephalan.

Data pertaining to host species examined and helminth parasites recovered during the study, are presented in Tables 1, 2, 3 , 18a and 18b. The results pertaining to the status of helminth infections presented herein are grouped into two categories: the terrestrial-habit hosts and arboreal-habit hosts; within each of the groups. Locality-wise prevalence, intensity, mean intensity and abundance of the parasites encountered are depicted in Table 17. Histograms, for locality-wise species richness of helminth infection in anuran hosts surveyed and overall prevalence of helminths group versus anuran families are in Figs. 263 and 264.

TABLE17. Locality surveyed Prevalence, Intensity and Abundance of various helminth parasites in anuran hosts

Locality: Dimapur

Name of host (no. examined/no. infected)	Parasite (no. infected)	Prevalence (M/F) %	Total no. of parasite recovered	Intensity	Mean Intensity	Abundance
E. cyanophlyctis(128/49)	*G. tigrinum*(20)	15.6(35/65)	82	2-8	4.1	0.64
	H. almorai (32)	25(34.3/65.6)	76	1-8	2.3	0.59
	P. gastroporus(2)	1.56(0/100)	6	1-5	3	0.04
	D. mehrai(8)	6.25(50/50)	32	1-8	4	0.25
	H. mehransis(3)	2.3(33.3/66.6)	12	2-7	4	0.04
	G. ellipticum (3)	2.34(39.3/66.6)	5	1-2	1.6	0.03
	Proalarioides sp.(5)	3.9(20/80)	20	1-7	4	0.15
	O. goezei(1)	0.78(100/0)	6	6	6	0.04
	A. gubernaculum(10)	7.8(50/50)	23	1-5	2.3	0.17
	Oxysomatium sp.(8)	14.0(83.3/16.3)	54	1-7	3	0.42
	Icosiella sp.I(3)	2.3(33.3/66.6)	8	2-4	2.3	0.06
	Plerocercoid larva(8)	6.25(37.5/62.5)	11	1-3	1.3	0.08
	A. bufonis(6)	4.6(50/50)	10	2-3	1.6	0.07
L. limnocharis (12/6)	*Oxysomatium* sp.(5)	41.6(80/20)	19	1-7	3.8	1.58
	Amplicaecum sp.(1)	8.3(0/100)	1	1	1	0.08
	G. tigrinum(1)	16.6(100/0)	5	2-3	2.5	0.41
M. ornata (6/0)	nil	-	-	-	-	-

Contd.

Name of host (no. examined/no. infected)	Parasite (no. infected)	Prevalence (M/F) %	Total no. of parasite recovered	Intensity	Mean Intensity	Abundance
Limnonectes sp. (3/3)	*G. tigrinum*(1)	33.3(0/100)	3	3	3	1
	Oxysomatium sp.(3)	100(66.6/33.3)	5	1-2	1.6	1.6
	Plerocercoid larva(2)	66.6(50/50)	7	2-5	3.5	2.3
Rana sp. 2(1/1)	*Oxysomatium* sp.(1)	100(100/0)	2	2	2	2
P. tarakensis (3/3)	*A. bufonis*(2)	66.6(50/50)	4	2-2	2	1.3
H. tigerinus (49/37)	*M. ranarum*(4)	8.16(25/75)	44	2-29	11	0.89
	G. tigrinum(16)	32.6(33.3/66.6)	63	1-19	3.93	1.28
	H. almorai(8)	16.3(37.5/62.5)	17	1-6	2.12	0.34
	D. mehrai(2)	4.09(0/100)	4	1-3	2	0.04
	P. gastroporus(7)	14.2(71.4/28.5)	41	2-23	5.85	0.83
	P. infrequentum(5)	10.20(60/40)	17	1-4	3.4	0.34
	Proalarioides sp.(7)	14.2(42.8/57.1)	53	3-12	7.57	0.1
	A. gubernaculum (11)	22.4(45.4/54.5)	40	1-9	3.63	0.81
	Oxysomatium sp.(12)	30.61(40/60)	53	1-15	4.4	1.34
	R. ranae(11)	22.4(27.2/72.7)	138	2-73	12.54	2.81
	Amplicaecum sp.(5)	10.2(20/80)	8	1-2	1.6	0.16
	P. tigrinus(1)	2.04(100/0)	2	2	2	0.04
	Nematotaenioides sp.(1)	2.04(100/0)	1	1	1	0.02
	Plerocercoid larva(21)	42.8(57.1/42.8)	71	1-6	3.38	1.4
	A. bufonis(3)	6.12(33.3/66.6)	8	2-3	2.66	0.16

Contd.

Name of host (no. examined/no. infected)	Parasite (no. infected)	Prevalence (M/F) %	Total no. of parasite recovered	Intensity	Mean Intensity	Abundance
H. crassus (27/21)	*G. tigrinum*(8)	29.6(22.2/77.7)	24	1-6	3	0.88
	D. mehrai(1)	3.7(0/100)	1	1	1	0.03
	P. gastroporus(4)	14.8(0/100)	15	1-5	3.75	0.55
	Oxysomatium sp.(3)	11.1(66.6/33.3)	24	1-17	8	0.88
	Amplicaecum sp.(2)	74(50/50)	2	1-1	1	0.07
	Icosiella sp.1(1)	3.7(100/0)	5	5	5	0.18
	Plerocercoid larva(2)	7.4(50/50)	4	1-3	2	0.o7
P. leucomystax (25/12)	*G. tigrinum*(2)	8(100/0)	6	2-4	3	0.24
	Oxysomatium sp.(7)	28(71.4/28.5	17	1-4	2.4	0.68
	R. ranae(3)	12(66.6/33.3)	14	2-6	4.6	0.56
	Icosiella sp.1(2)	8(100/0)	8	4	4	0.32
	Plerocercoid larva(12)	48(75/25)	19	1-7	1.9	0.17
	A. bufonis(6)	24(66.6/33.3)	19	2-6	3.16	0.76
Rana sp. I(43/30)	*G. tigrinum*(21)	48.8(52.3/47.6)	92	1-11	4.38	2.13
	H. almorai(10)	23.2(60/40)	25	1-6	2.48	0.15
	A. gubernaculum(5)	11.6(40/60)	19	2-8	3.5	0.44
	Oxysomatium sp.(80)	18.6(50/50)	21	1-6	2.6	0.48
	Icosiella sp.I(7)	16.2(57.1/42.5)	15	1-2	2.1	0.34
	Plerocercoid larva(7)	16.2(20/80)	20	1-7	4	0.18
	A. bufonis(15)	34.8(46.6/53.3)	25	2-8	1.66	0.58

Locality: Zunheboto

Name of host (no. examined/no. infected)	Parasite (no. infected)	Prevalence (M/F) %	Total no. of parasite recovered	Intensity	Mean Intensity	Abundance
E. cyanophlyctis (5/5)	*Oxysomatium* sp.(3)	60(33.3/66.6)	7	2-3	2.3	1.4
	O. macintoshii(2)	40(50/50)	5	2-3	2.3	1
	Cestode larva in sac(1)	20(100/0)	1	1	1	0.2
R. maximus (6/6)	*A. gubernaculum*(3)	50((33.3/66.6)	33	8-15	11	5.5
	R. ranae(4)	66.6(50/50)	10	2-3	2.5	1.6
	P. indicum(4)	66.6(50/50)	10	1-4	2.5	1.6
P. leucomystax (9/6)	*M. monas*(1)	11.1(100/0)	1	1	1	0.11
	D. amphichrus(2)	22.2(100/0)	2	2	1	0.22
	Plerocercoid larva(2)	22.2(50/50)	3	1-2	1.5	0.33
	R. ranae(1)	11.1(100/0)	1	1	1	0.11
	P. mucronata(1)	11.1(100/0)	8	8	8	0.88
	O. macintoshii(6)	66.6(66.6/33.3)	40	11-14	6.6	4.4
Locality:Tuensang						
P. leucomystax (2/1)	*M. monas*(1)	50(100/0)	3	3	3	1.5
R. bipunctatus (1/0)	*Nil*	-	-	-	-	-
A. marmoratus (3/3)	*Gorgoderina* sp.(1)	33.3(100/0)	3	3	3	1
	A. bufonis(1)	33.3(0/100)	2	2	2	0.66
	R. ranae(3)	100(33.3/66.6)	24	7-9	8	8
	Ophidascaris sp.(3)	100(33.3/66.6)	9	2-4	3	3
M. glandulosa (2/2)	*N. mokokchungensis* (2)	100(100/0)	2	1-1	1	1
	R. ranae(1)	50(100/0)	4	4	4	2

Locality: Mokokchung

Name of host (no. examined/no. infected)	Parasite (no. infected)	Prevalence (M/F) %	Total no. of parasite recovered	Intensity	Mean Intensity	Abundance
R. khare (13/9)	*P. gastroporus*(3)	23(66.6/33.3)	9	2-5	3	6.9
	B. nagalandensis(1)	7.6(0/100)	1	1	1	0.07
	R. ranae(5)	38.4(50/50)	22	3-7	4.4	1.6
	A. gubernaculum(4)	30.7(50/50)	15	1-6	3.75	1.6
	Oxysomatium sp.(3)	23.1(33.3/66.6)	30	2-11	6.6	2.3
	P. mucronata(1)	7.6(0/100)	1	1	1	3.8
	A. bufonis(1)	9.09(100/0)	3	3	3	0.39
R. livida(11/6)	*Oxysomatium* sp.(5)	45.4(40/60)	192	1-98	38.4	17.4
	A. bufonis(1)	9.09(100/0)	3	3	3	0.27
Rana sp.3(11/9)	*D. amphichrus*(1)	9.09(100/0)	3	3	3	0.27
	M. monas(1)	9.09(100/0)	4	4	4	0.36
	R. ranae(5)	45.4(0/100)	30	1-16	6	2.72
E. cyanophlyctis(118/45)	*G. tigrinum*(11)	9.32(81.8/18.1)	30	1-11	2.72	0.25
	H. almorai(44)	37.2(45.4/54.5)	150	1-22	3.4	1.27
	A. gubernaculum(7)	5.93(57.1/42.8)	38	2-6	5.4	0.32
	Oxysomatium sp.(24)	20.3(62.5/37.5)	61	1-7	2.5	0.51
	Rhabdias sp.(2)	1.69(50/50)	4	1-3	2	0.01
	B. baeri (5)	4.23(80/20)	9	1-3	2	0.01
	Plerocercoid larva(8)	6.7(62.5/37.5)	24	2-7	3	0.2
	A. bufonis(1)	0.8(100/0)	1	1	1	0.008
Lmnonectes sp.2(4/4)	*D. mehrai*(1)	25(0/100)	1	1	1	0.25

Contd.

Name of host (no. examined/no. infected)	Parasite (no. infected)	Prevalence (M/F) %	Total no. of parasite recovered	Intensity	Mean Intensity	Abundance
Rana danielii(12/11)	*A. gubernaculum*(2)	16.6(50/50)	6	1-5	3	0.5
	Oxysomatium sp.	66.6(25/75)	26	1-5	3.25	
	R. ranae(8)	66.6(12.5/87.5)	11	1-3	1.37	0.91
L.. limnocharis(84/52)	*G. tigrinum(3)*	3.5(33.3/66.6)	10	2-4	3.33	0.03
	Oxysomatium sp.(41)	54.7(73.9/26.0)	110	1-3	2.3	1.3
	Amplicaecum sp.(2)	2.3(50/50)	3	1-3	1.5	0.03
	Rhabdias sp.(14)	16.6(78.5/21.4)	20	1-2	1.4	0.23
	B.baeri(6)	7.1(100/0)	30	1-21	5	0.35
	Plerocercoid larva(4)	4.76(75/25)	6	1-2	1.5	0.07
	Cestode larva in sac(1)	1.19(100/0)	1	1	1	0.01
M. ornata(4/1)	*Oxysomatium* sp.1(3)	75(100/0)	12	2-8	4	3
A. marmoratus(151/107)	*O. indica*(12)	7.9(41.6/58.3)	38	1-18	3.16	0.25
	B. nagalandensis(11)	7.2(54.5/45.4)	16	1-4	1.45	0.1
	P. gastroporus(6)	3.9(50/50)	9	1-3	1.5	0.05
	M. monas(1)	0.66(0/100)	2	2	2	0.01
	Gorgoderina sp.(10)	6.62(30/10)	14	1-3	1.4	0.09
	R. ranae(84)	55.6(42.8/57.1)	387	7-9	4.6	2.56
	A. gubernaculum(60)	39.7(53.3/46.6)	422	1-40	2.79	2.79
	Ophidascaris sp.(58)	38.4(41.3/58.6)	194	1-8	3.3	1.28
	Icosiella sp.2(21)	13.9(47.6/52.3)	25	1-3	1.1	0.16
	A. bufonis(2)	1.32(0/100)	3	1-2	1.5	0.01

Contd.

Name of host (no. examined/no. infected)	Parasite (no. infected)	Prevalence (M/F) %	Total no. of parasite recovered	Intensity	Mean Intensity	Abundance
M. glandulosa(12/12)	*N. mokokchungensis* (9)	75(0/100)	11	1-3	1.2	0.91
	Oxysomatium sp.(8)	66.6(12.5/87.5)	146	2-32	18.2	12.16
	R. ranae(2)	16.6(0/100)	9	3-6	4.5	0.75
	A. bufonis(1)	8.3(100/0)	1	1	1	0.08
M. wuliangshanensis (26/23)	*Acanthocephalus* sp.(2)	7.6(100/0)	9	2-7	4.5	0.34
	Rhabdias sp.(1)	3,8(0/100)	1	1	1	0.038
	Oxysomatium sp.(6)	23.1(66.6/33.3)	42	2-8	7	1.61
	P. mucronata (4)	15.38(50/50)	20	1-5	5	0.76
	Icosiella sp.2(4)	15.3(25/75)	24	2-24	6	0.92
R. maximus(83/38)	*A. gubernaculum*(11)	27.5(63.6/36.3)	442	1-272		
	Aplectana sp.(7)	8.4(57.1/42.8)	20	1-5	2.8	0.24
	Oxysomatium sp.(6)	7.2(66.6/33.3)	15	1-6	2.5	0.18
	R. ranae(24)	28.9(45.8/54.1)	290	1-203	12.08	3.4
	Cosmocercella sp.(4)	4.8(50/50)	16	1-5	4	0.19
	D. amphichrus(7)	8.4(57.1/42.8)	22	2-7	3.14	0.26
	P. indicum(25)	30.1(36.5/64.4)	45	2-6	1.8	0.54
	Plerocercoid larva(4)	4.2(50/50)	16	1-6	4	0.8

Contd.

Name of host (no. examined/no. infected)	Parasite (no. infected)	Prevalence (M/F) %	Total no. of parasite recovered	Intensity	Mean Intensity	Abundance
R. bipunctatus(5/1)	*P. indicum*(1)	20(100/0)	4	4	4	0.8
T. asperum(3/1)	*P. indicum*(1)	33.3(100/0)	1	1	1	0.33
Theloderma sp.(4/4)	*Oxysomatium* sp.(3)	75(66.6/33.3)	15	1-7	5	3.75
	Pharyngodon sp.(1)	25(100/0)	3	3	3	0.72
	R. ranae (1)	25(0/100)	1	1	1	0.25
	B. baeri(2)	50(50/50)	11	1-10	5.5	2.75
C. vittatus(15/7)	*D. amphichrus*(1)	66.6(100/0)	1	1	1	0.06
P. annandalii (74/32)	*M. monas*(9)	12.1(33.3/66.6)	59	3-14	6.5	0.79
	Oxysomatium sp.(2)	9.4(71.4/28.5)	6	2-4	3	0.22
	O. macintoshii (2)	9.4(0/100)	10	2-8	5	0.13
	Pharyngodon sp.3(1)	1.3(0/100)	4	4	4	0.05
	R. ranae(13)	17.5(69.2/30.7)	20	1-6	1.5	0.27
	B. baeri(11)	14.8(27.2/72.7)	34	2-13	3.09	0.45
Philautus sp.3(13/8)	*R. ranae*(6)	46.1(100/0)	13	1-3	2.16	1
	B. baeri(2)	15.3(100/0)	2	1-1	1	0.15
Philautus sp. 1(23/17)	*A. gubernaculum*(3)	13.0(100/0)	21	4-10	7	0.94
	Oxysomatium sp.(4)	17.3(100/0)	8	1-5	2	0.34
	O. macintoshii(3)	13.0(100/0)	61	1-28	20.3	2.6
	R. ranae(3)	13.0(100/0)	17	1-12	5.6	0.73
	Polystoma sp.(2)	8.6(100/0)	10	4-6	5	0.43
	Pleurogenoides sp.(1)	4.3(100/0)	1	1	1	0.04
	B. baeri(6)	26.0(100/0)	24	1-9	4	1.04

Contd.

Name of host (no. examined/no. infected)	Parasite (no. infected)	Prevalence (M/F) %	Total no. of parasite recovered	Intensity	Mean Intensity	Abundance
Philautus sp. 2(6/1)	*R. ranae*(1)	16.6(100/0)	1	1	1	0.16
P. leucomystax(113/73)	*G. tigrinum*(7)	6.1(71.4/28.5)	22	2-6	3.1	0.19
	M. monas(9)	7.9(88.8/11.1	29	2-7	3.2	0.25
	D. amphichrus(9)	7.9(88.8/11.1)	24	1-5	2.6	0.21
	P. gastroporus (7)	0.8(100/0)	5	5	5	0.04
	P. indicum(18)	15.9(77.7/22.2)	33	1-6	1.8	0.29
	A. gubernaculum(27)	23.8(25.9/74.0)	229	1-118	8.4	2.02
	Aplectana sp.(19)	16.8(63,1/36.8)	131	1-37	6.8	1.15
	R. ranae(33)	29.0(66.6/33.3)	120	1-7	3.6	1.06
	Cosmocercella sp.(6)	5.30(50/50)	24	2-6	4	0.21
	O. macintoshii (3)	2.65(33.3/66.6)	43	9-17	14.33	0.38
	Oxysomatium sp.(25)	22.1(68/32)	363	1-120	14.5	3.21
	Plerocercoid larva(38)	33.6(68.4/31.5)	129	1-18	3.8	1.14
	A. bufonis(1)	0.88(100/0)	2	2	2	0.01
M. tarapensis(64/43)	*M. monas*(3)	4.6(66.6/33.3)	33	2-26	11	0.51
	P. indicum(2)	3.1(50/50)	3	1-2	1.5	0.04
	Oxysomatium sp.(16)	25(66.6/33.3)	201	1-30	10.5	3.14
	O. macintoshii(3)	34.(33.3/66.6)	22	5-10	7.6	0.34
	R. ranae(16)	25(50/50)	62	1-12	3.8	0.96
	B. baeri(1)	1.5(0/100)	10	10	10	0.15
	P. tigrinus(2)	3.1(100/0)	2	1-1	2	0.03
	A. bufonis(3)	4.6(33.3/66.0)	6	1-4	2	0.09

Locality: Kohima

Name of host (no. examined/no. infected)	Parasite (no. infected)	Prevalence (M/F) %	Total no. of parasite recovered	Intensity	Mean Intensity	Abundance
R. maximus(5/5)	*P. indicum*(2)	40(100/0)	5	1-4	2.5	1
	Oxysomatium sp.(2)	40(50/50)	8	2-6	4	1.6
	R. ranae(3)	60(66.6/33.3)	8	2-4	2.6	1.6
R. gongshanensis(13/7)	*P. kohimaensis*(7)	53.8(57.1/42.8)	12	1-3	1.7	0.92
	O. macintoshii (2)	15.3(50/50)	8	2-6	4	0.3
	R. ranae(1)	7.6(100/0)	2	2	2	0.51
R. bipunctatus(10/4)	*R. ranae*(2)	20(50/50)	3	1-2	1.5	0.3
	B. baeri	10(100/0)	1	1	1	0.1
	Plerocercoid larva	10(100/0)	1	1	1	0.1
H. annectans(106/32)	*M. monas*(2)	1.8(100/0)	56	7-49	28	0.52
	P. hylai(10)	9.4(70/30)	30	2-5	3	0.28
	B. baeri(21)	19.8(71.4/28.5)	37	1-9	2.8	0.44
	R. ranae(20)	18.8(60/40)	57	1-7	2.8	0.57
	Oxysomatium sp.(16)	15.0(68.7/31.2)	44	2-7	2.7	0.41
	O. macintoshii (2)	1.8(100/0)	12	3-9	6	0.11
Philautus sp.1(1/0)	*NIL*	-	-	-	-	-
P. leucomystax(36/16)	*M. monas*(1)	2.77(0/100)	2	2	2	0.05
	A. gubernaculum(4)	11.1(25/75)	16	2-6	4	0.44
	Aplectana sp. (7)	19.4(28.5/71.4)	17	2-3	2.4	0.47
	P. mucronata(2)	5.0(50/50)	7	2-5	3.5	0.19
	Oxysomatium sp.(9)	25(66.6/33.3))	27	1-10	1.5	0.75
	R. ranae(10)	27.7(40/60)	36	2-12	3.6	1

Contd.

Name of host (no. examined/no. infected)	Parasite (no. infected)	Prevalence (M/F) %	Total no. of parasite recovered	Intensity	Mean Intensity	Abundance
E. cyanophlyctis(88/30)	*G. tigrinum*(35)	39.7(37.1/62.8)	117	1-11	3.3	1.32
	H. almorai(23)	26.1(39.1/60.8)	62	1-6	2.6	0.7
	D. mehrai(16)	18.1(37.5/62.5)	33	2-6	2.06	0.37
	Cathaemasia sp.(2)	2.27(0/100)	6	2-4	3	0.06
	A. gubernaculum(4)	4.5(75/25)	10	1-4	2.5	0.11
	Oxysomatium sp.(24)	27.2(33.3/66.6)	50	1-7	2.08	0.56
	O. goezei(4)	4.5(50/50)	17	2-7	4.2	0.19
	B. baeri(2)	2.27(50/50)	11	2-9	5.5	0.12
	Plerocercoid larva(9)	10.2(33.3/66.6)	19	2-4	2.1	0.21
	A. bufonis(2)	2.27(100/0)	2	1-1	1	0.02
P. mokokchungensis(1/1)	*O. yunnanse*(1)	100(0/100)	6	6	6	6
L. limnocharis(30/)	*G. tigrinum*(3)	10(33.3/66.6)	11	2-4	3.6	0.36
	Oxysomatium sp.(15)	50(60/40)	46	2-10	3.06	1.5
M. wuliangshanensis(2/2)	*Oxysomatium* sp.(2)	100(50/50)	4	2-2	2	1
	Icosiella sp.2(1)	50(0/100)	1	1	1	0.5

Terrestrial habit species:

FAMILY: RANIDAE

Rana khare, available only in Mokokchung, harboured 8 helminth species that include 2 trematodes (*P. gastroporus* and *B. nagalandensis*) and 4 nematodes (*A. gubernaculum, Oxysomatium* sp., *P. mucronata* and *R. ranae*). The male hosts had higher prevalence for *P. gastroporus* than females. Only 1 *B. nagalandensis* was recovered from a female host. The nematodes, *P. mucronata* and *Oxysomatium* sp., showed more prevalence in female as compared to the male hosts, and the rest of the three nematode species were somewhat equally shared in both the sexes. Out of the 13 hosts examined, 5 were infected with 1 species of helminth parasite, 2 with 2, 1 with 3, and 1 with 4 species and the remaining 4 hosts were uninfected.

Rana livida (=***Limnonectes mawphlangensis***), which was collected only from Mokokchung, had 2 helminth species that includes 1 nematode, *Oxysomatium* sp. that showed more prevalence in female (60%) than male hosts (40%) and 1 acanthocephalan, *A. bufonis*. Of the 11 hosts examined, 5 were found infected with 1 species of parasite and 1with 2, respectively, and 5 were free of any infection.

Paa mokokchungensis (=***Rana leibigii***), a female host collected only from Kohima during the entire study period, harboured only 1 species of trematode *O. yunnanse*.

Again a species of the genus ***Rana*** from Dimapur is designated herein as ***Rana*** sp. 1 showed a wider spectrum represented by all the helminths groups except the monogenea and included 2 trematode and three nematode species. The trematode showed more prevalence for male hosts. Regarding the 3 nematode species, the female showed a higher prevalence for *A.*

gubernaculums (60%), while the infection of *Icosiella* sp. 1 was more prevalent among the male hosts. A plerocercoid cestode larva and *A. bufonis* also parasitized ***Rana*** sp. Male hosts had more infection by plerocercoid cestode larva (71.4%); the opposite was true for *A. bufonis*. Out of the 43 hosts examined, 24 were found infected, of which 2 hosts had infection with 1, 9 with 2, 8 with 3, 4 with 4 and 1 with 5 parasite species.

Rana sp.2, a new host, considered different from *Rana* sp.1 in having larger body size, skin texture and pattern with peculiar mating calls collected only from Dimapur, also harboured a single species of nematode, *Oxysomatium* sp.

Rana sp.3, yet another ranid collected from Mokokchung only, had the representation by 2 trematodes (*D. amphichrus* and *M. monas*) and 1 nematode (*R. ranae*). Both the trematodes were collected from one male host. The lung nematode was collected from female hosts only. 11 hosts were examined in the study, of which 8 host were infected with 1 parasite species, 1 with 3, and 2 were uninfected.

Haplobatrachus tigerinus, collected from Dimapur only, exhibited a rich diversity of parasitic fauna that includes 7 trematodes (6 adult and 1 metacercarial form). Except for *P. gastroporus* and *Prosostocus infrequentum*, all others had more prevalence in female than the male hosts; the 2 mentioned trematode species showed more prevalence in the male hosts (71.4% and 60%, respectively). Again, the nematodes also showed higher prevalence in female than in male hosts. The cestodes showed more prevalence in males than female hosts and the acanthocephalan had more female infection. Out of the 49 hosts examined, 9 hosts

were infected with 1 species of parasites, 4 with 2, 7 with 3, 8 with 4, 9 with 5, and the remaining 12 hosts did not show any infection.

H. crassus, collected only from Dimapur, was parasitized by 3 trematodes-*G. tigrinim, D. mehrai, and P. gastroporus* and 4 nematodes, namely, *Oxysomatium* sp., *Icosiella* sp. and *Amplicaecum* sp.; Plerocercoid larva. With plerocercoid larva somewhat equally represented in both the sexes, all the trematode species showed higher prevalence in female (77%, 100%, 100%, respectively) compared to male hosts. In contrast, the nematodes were more prevalent in males than females, except for *Amplicaecum* sp., which was recovered from one male and a female host. Of the 27 host examined, 10 were infected with 1 species of parasites, 5 with 2, 5 with 3, 1 with 4 and the remaining 6 were uninfected.

Euphlyctis cyanophlyctis, which was collected from 4 of the 5 localities surveyed, showed the richest and the highest helminth composition among the anuran host species examined in the study. This host, also available throughout the year was harboured by 17 species of helminth parasites in overall estimation; the parasite spectrum included four helminth groups, excluding only Monogenea. The forms encountered included 8 trematodes (*G .tigrinum, G. ellipticum, D. mehrai, H. mehransis, P. gastroporus, H. almorai* and 2 metacercariae-*Cathaemasia* sp. and *Proalarioides* sp.); 2 species of cestodes (*B. baeri* and 1 metacestode, plerocercoid larva); 7 nematodes (*A. gubernaculum, Oxysomatium* sp., *P. mucronata., O. goezei, Icosiella* sp. 1, *R. ranae*) and 1 acanthocephalan. Of the 4 localities, ***E. cyanophlyctis*** of Dimapur had the highest number of species represented by 7 trematode, 5 nematode, plerocercoid larva and one acanthocephalan; in comparison with the other 3 localities under

the study area (i.e., Kohima, Mokokchung and Zunheboto) and lacked only the metacercaria of *Cathaemasia* sp. & *B. baeri* from Kohima; *R .ranae & B.baeri* from Mokokchung and *P. mucronata* from *Zunheboto*. This host from Kohima, Mokokchung and Zunhebouto harboured 10, 8, and 2 species, respectively, and did not include *G. ellipticum, H. mehransis P. gastroporus*, metacercaria of *Proalarioides* sp.; *Icosiella* sp., from Kohima hosts; in addition to this species, *D. mehrai,* the metacercaria, *Cathaemasia* sp., *O. goezi* helminths were never record from Mokokchung. The hosts from Zunhebouto harboured only *Oxysomatium* sp and *O. macintoshii*. All the trematode species from Dimapur, excepting *D. mehrai* which were somewhat equally shared in both male and female hosts, had higher prevalence in female. The nematode species of *Oswaldocruzia* and *Oxysomatium* sp., showed higher prevalence in male (100% and 83.3%, respectively) than in female hosts, the opposite was true for *Icosiella* sp.1 and plerocercoid larva, which had higher prevalence in the female hosts (66.6%, 62.5%, respectively); *A. gubernaculum* and *A. bufonis* were found equally shared by both male and female hosts. The frogs of Kohima showed higher prevalence in female hosts for all the trematode species and did not presnt a definite pattern with regard to the infection of cestodes and nematodes; the *A. gubenaculum* showed higher prevalence in male hosts (75%) than in female (25%); the opposite was true for *Oxysomatium* sp. and plerocercoid larva (66.6% for both species), but in the case of *O. goezei* and *B. baeri* both male and female hosts had equal prevalence. Again, *A. bufonis* was recovered from two male hosts. In Mokokchung, this host was represented by 2 trematode species, *G. tigrinum* and *H. almorai*; the former had higher prevalence in male and the latter in female hosts. The cestodes, *B. baeri* and the plerocercoid larva, showed higher prevalence in male than in

female frogs. The nematodes showed somewhat different pattern; *A. gubenaculum* and *Oxysomatium* sp. showed higher prevalence in male hosts, with

R. ranae shared equally in both the sexes. Only 1 *A. bufonis* was recovered from a male host of Mokokchung. ***E. cyanophlyctis*** of Zunhebouto had 2 helminth species recovered from 5 hosts examined- *Oxysomatium* sp. from 1 male and 2 females and *O. macintoshii* from 2 hosts, represented in both the sexes.

Out of the 128 hosts examined from Dimapur, 15 were infected with 1 species of parasite, 19 with 2, 6 with 3, 2 with 4, 7 with 5, respectively, and the remaining 79 hosts were uninfected. Of the 88 hosts examined from Kohima, 4 were with 1, 4 with 2, 7 with 3, 8 with 4, 5 with 5, 2 with 6, respectively, and the rest of the hosts were free of infection. In Mokokchung, 2 were with 1, 19 with 2, 13 with 3, 5 with 4, 6 with 5 species, respectively, out of the 113 hosts examined and a total of 68 hosts were uninfected. In Zunheboto, out of the 5 hosts examined, 3 were infected with 1 parasite species and 1 with 2 and only one host was uninfected.

Limnonectes limnocharis was collected from 3 of the 5 localities surveyed and included 1 trematode species only, *G. tigrinum*, in all the localities and which was more prevalent in female than the male hosts. 2 nematode species *Oxysomatium* sp and *R. ranae* were recovered from the host of Mokokchung; and 2 cestodes, *B. baeri* and plerocercoid larva. All the nematodes showed more prevalence in male than in female hosts, whereas the cestodes (*B. baeri*, plerocercoid larva, metacestode) showed higher prevalence in males (100%, 75% and 100%, respectively). The frogs of Dimapur had 2 nematodes, *Oxysomatium* sp. showing higher prevalence in male hosts (80%) and an *Amplicaecum* sp. recovered from a female host. *Oxysomatium* sp. the lone nematode, from Kohima, had higher prevalence in males (60%).

Out of the 88 hosts examined from Mokokchung, 28 were infected with 1 species of parasite, 21 with 2, and 3 with 3 species. In Dimapur, 4 hosts were infected with 1 parasite speices and 2 with 2 of the total 12 hosts examined. Those from Kohima had 2 hosts with 1 and 7 with 2 of the 30 hosts examined, respectively.

Limnonectes sp., collected only from Mokokchung and Dimapur, was represented by 2 trematodes; the Mokokchung frogs had *D. mehrai*, the only helminth parasite recovered from a female of the 4 hosts examined. The hosts from Dimapur had *G. tigrinum*, recovered from a male of the 3 hosts examined. All the 4 hosts also harboured a nematode species, *Oxysomatium* sp. and two were also found infected with plerocercoid larva.

Rana danielii, collected only from Mokokchung, hosted only nematode parasites, namely, *A. gubernaculums*, *Oxysomatium* sp. and *R. ranae*. The former nematode was shared by both male and female hosts and the latter two species were more prevalent in female (75% and 87.5%, respectively) than in male hosts (25% and 12.5%). 4 hosts were found infected with 1 species and 7 with 2 and only 1 frog was uninfected.

Amolops marmoratus (***=A. afghanus***), collected from Mokokchung and Tuensang, harboured 10 helminth species. The hosts from Mokokchung included 5 trematodes (*P. gastroporus*, *O. indica*, *B. nagalandensis*, *M. monas* and *Gorgoderina* sp.), and 4 nematodes (*A. gubernaculum*, *R. ranae*, *Ophidascaris* sp., *Icosiella* sp.2). Three of the trematode species, *O. indica*, *M. monas* and *Gorgoderina* sp, were more prevalent in the female (58.3%, 100% and 100%, respectively) as compared to male hosts, the opposite was true for *B. nagalandensis*; and *P. gastroporus* was somewhat equally shared in both the sexes. Except *A. gubernaculum*,

the rest of the nematode species had higher prevalence in females than in male hosts and *A. bufonis* was recovered from 2 female hosts. This host from Tuensang harboured 1 trematode (*Gorgoderina* sp.) recovered from a male, and 2 nematodes (*R. ranae* and *Ophidascaris* sp.) and *A. bufonis* recovered from 2 female hosts. Of the 3 hosts examined from Tuensang, 1 was infected with 2 species of parasites and 2 with 3 species. Furthermore, 20 hosts were infected with 1 species of parasite, 50 with 2, 23 with 3, 10 with 4, and 4 with 5; and the remaining 44 of the 151 hosts examined from Mokokchung were uninfected.

FAMILY: MICROHYLIDAE

Microhyla ornata was collected from Mokokchung and Dimapur; only the hosts from Mokokchung were found infected with a lone nematode species, *Oxysomatium* sp., recovered from one of the 4 male hosts examined.

FAMILY: PELOBATIDAE

Megophrys glandulosa, collected from Mokokchung and Tuensang, were represented by a monogenean species (*N. mokokchungensis* n. g, n. sp.), 2 nematodes (*Oxysomatium* sp. and *R. ranae*) and an acanthocephalan, *A. bufonis* from a male host. All the helminth forms showed higher prevalence in female than in male hosts. All the helminth forms collected from Mokokchung were also present in Tuensang hosts, except the acanthocephalan. Out of the 12 frogs examined, 2 were infected with 1 species of helminth, 3 with 2, 6 with 3 and 1 with 4 species, respectively.

M. wuliangshanensis, another host from the same family, was collected from Mokokchung and Kohima. The host frogs from Mokokchung were represented by 3 nematode species- *Oxysomatium* sp., *P. mucronata* and *R. ranae*) and an acanthocephalan. Except for *R. ranae*,

all the helminth forms were more prevalent in the male hosts and the *P. mucronata* was somewhat equally shared in both the sexes. Out of the 2 hosts examined from Kohima, *Oxysomatium* sp. was present in both sexes. Of the 26 hosts from Mokokchung, examined for helminth parasites, 8 were infected with 1 species, 7 with 2, and 8 with 3, respectively, and the remaining 3 hosts were uninfected.

Arboreal-habit species

FAMILY: HYLIDAE

Hyla annectans, collected only from Kohima, had 4 helminth groups representing 1 monogenean (*P. hylai* n sp.), 1 trematode (*M. monas*), 1 cestode (*B. baeri*) and 3 nematodes (*R. ranae*, *Oxysomatium* sp., and *O. macintoshii*). All these helminth parasites showed more prevalence in male hosts as compared to female. Out of the 106 frogs examined, 11 were infected with 1 species of parasite, 7 with 2, 8 with 3, 5 with 4, and 1 with 5; the remaining 74 hosts were free of any helminth parasite infection and no acanthocephalan species was ever recovered from this host species.

FAMILY: RHACOPHORIDAE

Rhacophorus maximus (=***R. nigropalmatus***), available only in Kohima, Mokokchung and Zunheboto sites, exhibited parasitization by 8 helminth species in all. The diversity of parasites in *R. maximus* in Kohima included the monogenean, *P. indicum*, and 2 nematodes , namely, *Oxysomatium* sp. and *R. ranae*, whereas in Mokokchung frogs it was represented by the trematode, *D. amphichrus*; the cestode plerocercoid larva; and the nematode *A.*

gubernaculums, Aplectana sp. and *Cosmocercella* sp. in addition to *P. indicum*, *Oxysomatium* sp. and *R. ranae*. The frogs of Zunheboto harboured only 3 helminth species that included *P. indicum*, *A. gubernaculum* and *R. ranae*. This host species had 2 males infected by *P. indicum* in Kohima. For *R. ranae* the male hosts showed higher prevalence than female hosts and *Oxysomatium* sp. was somewhat equally shared in both the male and female frogs. The opposite was true for *A. gubernaculums* from Zunheboto with higher prevalence in female hosts (66.6%) than male (33.3%) and the other two helminth species equally shared in both the sexes. In Mokokchung, the female frogs showed a higher prevalence than the male for *P. indicum* and for *D. amphichrus*, it was somewhat equally shared in both the sexes. Plerocercoid larva, *A. gubernaculum* and *Aplectana* sp., all the three were more prevalent in male when compared to female hosts. For *Oxysomatium* sp., the male hosts showed higher prevalence (66.6%), while *R. ranae* was more prevalent in female (54.1%); *Cosmocercella* sp. was equally prevalent in the two sexes. Only 5 hosts were examined from Kohima and out of these, 3 were infected with 1, 1with 2, and 1with 3 species, respectively. Of the 6 hosts examined from Zunhebouto, 2 were infected with 1 species of parasite, 3 with 2, and 1 with 3, respectively. Altogether, 83 host specimens were examined from Mokokchung, out of which 12 were infected with 1, 9 with 2, 4 with 4, 4 with 3, 4 with 4, 4 with 5 and 1 with 6 species, respectively. The remaining 45 hosts were uninfected.

R. bipunctatus (=***R. reinwardtii***), collected from Kohima, Mokokchung and Tuensang, was represented by 3 groups of helminths and Tuensang host had no parasite infection, Mokokchung hosts had only 1 parasite, represented by a monogenean (*P. indicum*) recovered from one of the 5 male hosts examined. The same host from Kohima had *R. ranae* and 2 cestode forms (*B. baeri* and plerocercoid larva) recovered from 2 female hosts; *R. ranae* were

equally prevalent in both the sexes. Out of the 10 hosts examined from Kohima, 2 were infected by 1 species of helminth parasite, 2 with 2 and the remaining 6 were free of any infection.

R. gongshanensis, collected only from Kohima, harboured a monogenean (*P. kohimaensis* n. sp.) and 2 nematodes (*O. macintoshii* and *R. ranae*). Except for *O. macintoshii* which showed equal prevalence in both the sexes, the other two helminths were more prevalent in female hosts. The monogenean had more prevalence in male than in female hosts. Out of the 13 host examined 6 were infected with 1 species of parasites and 1 with 2. The remaining 6 hosts were uninfected.

Theloderma asperum of Rhacophoridae family was collected only from Mokokchung and was represented by only *P. indicum*, recovered from a male host out of the 3 examined. However, another species of the same genus ***Theloderma*** sp. was collected from Mokokchung only and was represented by a cestode (*B. baeri*), 3 nematodes- namely, *Oxysomatium* sp., *Pharyngodon* sp. and *R. ranae,* recovered from a female. *Oxysomatium* sp. was more prevalent in male (66.6%) than in female hosts and *Pharyngodon* sp. was recovered from a male host. *B. baeri* was equally prevalent in both. Out of the 4 hosts examined, 1 was infected with 1 parasite species, 2 with 2 and 1 with 3, respectively.

Chirixalus vittatus, another representative of Rhacophoridae, was available only in Mokokchung and included only one trematode (*D. amphichrus*), which was more prevalent in male than in female host. Out of the 15 hosts examined, 7 were infected with this trematode.

Philautus annandalii was collected from Mokokchung and Kohima only. The host specimens from Kohima did not show any helminth infection, but those from Mokokchung supported only 1 adult trematode, *M. monas*; a cestode, *B. baeri* and 4 nematode species viz., *R. ranae*, *Oxysomatium* sp., *Pharyngodon* sp. and *O. macintoshii*. The nematodes, *Pharyngodon* sp. and *O. macintoshii*, and *B. baeri* were more prevalent in female (100%, 100% and 72.27%, respectively) than in male hosts, the opposite being true in the case of *Oxysomatium* sp. and *R. ranae*, which showed higher prevalence in male (71.4% and 69.2%, respectively). Out of the 74 examined for helminth parasite infections, 18 were infected with 1 species and 14 with 2, species of parasites respectively.

Besides, three more species of the genus ***Philautus***, designated herein as ***Philautus*** sp. 1, 2 and 3 (all different from one another and also from ***P. annandalii***) ***Philautus*** sp. 2, were collected from Mokokchung. ***Philautus*** sp.1 of Mokokchung showed a rich diverse helminth fauna and was represented by all the 5 groups, a monogenean (*Polystoma* sp.), 1 trematode (*P. gastroporus*), 1 cestode (*B. baeri*) 4 nematodes (*Oxysomatium* sp., *A. gubernaculum*, *O. macintoshii* and *R. ranae*) and the acanthocephalan represented by a cystacanth. Out of the 23 hosts collected, only 1 was female and did not show any infection. Of the 22 male hosts examined 10 were infected with 1 species of parasite, 5 with 2, and 1 with 3 species, respectively. The remaining 6 hosts were uninfected.

Philautus sp.2 was represented by a single nematode, *R. ranae* recovered from a male host of the 6 examined in the study.

Philautus sp.3, yet another host of the same genus collected only from Mokokchung, was represented by 2 helminth species, a cestode, *B. baeri* and a nematode, *R. ranae*. Both of these forms were recovered from male hosts only; of the 13 hosts examined, 1 had 2 helminth species and 7 only1 species.

Polypedates leucomystax was available in all the 5 localities surveyed and exhibited a rich parasitic fauna comprising 15 species that represented all the 5 groups of helminth parasites. The hosts from Kohima exhibited *M. monas*, *A gubernaculum*, *Aplectana* sp., *Oxysomatium* sp., *P. mucronata* and *R. ranae*. However, this host in Dimapur supported *G. tigrium*, plerocercoid larva, *Oxysomatium* sp., *R. ranae*, *Icosiella* sp. and *A. bufonis*. In Mokokchung, ***P. leucomystax*** harboured *P. indicum, D. amphichrus*, *M. monas*, *G. tigrinum*, *P. gastroporus*, plerocercoid larva; 5 nematodes namely, *A. gubernaculum*, *Aplectana* sp., *Cosmocercella* sp., *O. macintoshii*., *Oxysomatium* sp., and *R. ranae*; and an acanthocephalan, *A. bufonis*. In Zunheboto site the infection in this host was represented by 2 trematodes, *M. monas* and *D. amphichrus*. The cestodes were represented by Plerocercoid larva; the nematodes were *Cosmocercella* sp., *O. macintoshii*, *R. ranae*. The frog host ***P. leucomystax*** of Tuensang had *M. monas*, the lone representive of helminth recovered from a male. In Kohima, this host had only 1 female infected with *M. monas*; the nematode *Oxysomatium* sp. showed more prevalence in male (66.6%) but the other 3 nematode species showed higher prevalence in female when compared to male hosts, though for *P. mucronata* which was somewhat equally prevalent in both the sexes. The same host in Dimapur had only 2 male infected with *G. tigrinum*; the plerocercoid larva showed higher prevalence in male hosts (75%) and so did all the 4 nematodes and *A. bufonis*. The prevalence trend for helminths were

somewhat different in frogs of Mokokchung with monogenean and all the trematodes showing higher prevalence in males than in females, whereas the opposite was true for *A. gubernaculum* and *O. macintoshii* that showed less prevalence in males (25.9% and 33.3%, respectively) than in female (74.0% and 66.6%, respectively); *Cosmocercella* sp. and plerocercoid larva were equally prevalent in both the sexes. In Dimapur of the 25 hosts examined, 2 had infection with 1 species of parasites, 4 with 2, 3 with 3, 3 with 4 and 1 with 5, respectively. In Kohima, out of 25, 10 had infection with 1, 4 with 2, 1 with 4 and 1 with 5 species, respectively. Of the 113 host examined from Mokokchung, 35 had infection with 1, 20 with 2, 7 with 4, 8 with 5 and 3 with 6, respectively. The same host of Zunheboto had 3 with 1 species infection, 2 with 2 and 1 with 3, respectively and three were free. Of the 2 ***P. leucomystax*** examined from Tuensang, only 1 host was found infected by 1 species of parasite.

P. taraiensis was collected from Mokokchung and Dimapur localities. Only 2 hosts were examined from the latter site and both were found infected by *A. bufonis* only. The same host from Mokokchung showed rich species diversity with all the 5 groups of helminth parasites, a monogenean, *P. indicum*; a trematode *M. monas*; 5 nematodes, namely, *Oxysomatium* sp., *O. macintoshii*, and *R. ranae*; 2 cestodes, *B. baeri* and *P. tigrinus* and the acanthocephalan, *A. bufonis*. *P. indicum* was recovered from a male and a female host. *M. monas* and *P. tigrinus* had higher prevalence in male (66.6% and 100%, respectively) hosts. The same held true for *Oxysomatium* sp. and for *R. ranae*, it was equally prevalent in both sexes of host. In contrast, the cestode *B. baeri* and *A. bufonis* showed the opposite, with higher prevalence in female frogs (100% and 66.6%, respectively). Out of the 64 hosts

examined for helminth parasites, 18 were with 1 species, 16 with 2, 6 with 3, 3 with 4, respectively and the rest were uninfected.

In overall estimation of the host species, both trematode and nematode showed higher prevalence in female hosts in respect of ***H. tigerinus***, ***H. crassus***, ***E. cyanophlyctis*** (aquatic species) and ***A. marmoratus***, a wide ranging host from lotic systems. Only 4 species under the terrestrial habit species showed a different prevalence pattern in the two sexes, while in the rest a uniform sex-wise trend was not revealed. In the arboreal-habit species, the male hosts in majority showed higher prevalence for most of the trematode, nematode, cestode and acanthocephalan species. The anuran hosts with the habit of spending more time in land and trees showed higher nematode infection, whereas both the trematode and nematode were equally rich in the hosts that use water and land as habitat. Thus the habitat of different anuran species showed a prominent influence in the helminth parasites composition. Of the frog species examined in all the 5 localities, throughout the study period, ***P. leucomystax*** was the only host common to all and ***E. cyanophlyctis*** to 4 of the 5, explored.

The ranids surveyed from Dimapur study sites had the highest helminth species composition, followed by Mokokchung, Kohima, Tuensang and Zunheboto, in receding order. Pelobatids of the 3 localities also showed relatively richer helminths composition, with Mokokchung ranking the highest followed by Kohima and Tuensang. The microhylids of Mokokchung and Dimapur had the lowest helminth infection when compared to all the anurans examined during the study; all these 3 families fall under the terrestrial habit species.

Among the arboreal group, comprising 2 families of frogs, the rhacophorids of Mokokchung dominated in helminth species compositions over all the other localities. The hylids, restricted to Kohima only, also showed relatively rich helminth species compositon. In all the localities,

the terrestrial-habit species emerged as having richer parasitic fauna than the arboreal habit species. However, the latter group had three of the host species supporting all the 5 helminth groups; they are ***P. leucomystax***, ***Philautus*** sp.1 and ***P. taraensis***. Among the terrestrial-habit species, ***E. cyanophlyctis***, ***H. tigerinus*** and **A. *marmoratus*** had the richest overall helminths species composition, with the first mentioned host ranking the highest in overall estimation of helminth species, represented with 17 of the 39 helminth species prevalent, followed by 15 and 10 species, respectively, for the last 2 hosts. For the arboreal group, it was the rhacophorid, ***P. leucomystax*** that showed the highest species composition with 15 species, followed by *M. taraensis* and *R. maximus* with 8 species each.

In general, the present study revealed high intensity of infection in many of the anuran hosts examined. Nematodes showed the highest intensity per host with many species, followed by trematodes, cestodes, monogenea and acanthocephala, in receding order; the range being between 1 and less than 203 for nematode (*R. ranae*), 1-118 for (*A.gubernaculum*), 1-120 for (*Oxysomatiums* sp.); 1-29 for trematode (*M. ranarum*); 1-13 for cestode (*B. baeri*); 1-6 for monogenea (*P. indicum*) and acanthocephalans (*A. bufonis* and cystacanth), respectively. The mean intensity in respect of all the parasite species was low in general, excepting the nematode species, *Oxysomatium* sp. of *R. livida* (39.6), *A. gubernaculum* of *R. maximus* (33.9) and the trematode species, *M. monas* of *H. annectans* (29), all of which showed high mean intensity. The plerocercoid larval stage (metacestode) was found to occur in a wide range hosts (9 species).

Table 17a Helminth parasites and their anuran hosts in Nagaland: Terrestrial habit species

Helminth Parasites	HOST SPECIES															
	Megophrys glandulosus	*Megophrys wuliangshanensis*	*Microhyla ornata*	*Amolops marmoratus*	*Euphlyctis cyanophlyctis*	*Haplobatrachus tigerinus*	*Haplobatrachus crassus*	*Limnonectis limnocharis*	*Limnonectes* sp.	*Paa mokokchungensis*	*Rana khare*	*Rana danielii*	*Rana livida*	*Rana* sp.3	*Rana* sp.1	*Rana* sp.2
MONOGENEA: *Neoriojatrema mokokchungensis* n. sp.	+	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-
TREMATODE: Adult *Gorgoderina ellipticum*	-	-	-	-	+	-	-	-	-	-	-	-	-	-	-	-
Gorgoderina sp.	-	-	-	+	-	-	-	-	-	-	-	-	-	-	-	-
Diplodiscus amphichrus	-	-	-	-	-	-	-	-	-	-	-	-	+	-	-	-
Diplodiscus mehrai	-	-	-	-	+	+	+	-	+	-	-	-	-	-	-	-
Halipegus mehransis	-	-	-	-	+	-	-	-	-	-	-	-	-	-	-	
Mesocoelium monas	-	-	-	+	-	-	-	-	-	-		-		+		
Opisthioparorchis indica	-	-	-	+	-	-	-	-	-	-	-	-	-	-	-	-
Opisthioparochis yunnanse	-	-	-	-	-	-	-	-	-	+	-	-	-	-	-	-
Ganeo tigrinum	-	-	-	-	+	+	+	+	+	-	-	-	-	-	+	-
Haematoloechus almorai	-	-	-	-	+	+	-	-	-	-	-	-	-	-	+	-
Pleurogenoides gastroporus	-	-	-	+	+	+	+	-	-	-	+	-	-	-	-	-
Batrachotrema nagalandensis	-	-	-	+	-	-	-	-	-	-	+	-	-	-	-	-

Condt.

Helminth Parasites	*Megophrys glandulosus*	*Megophrys wuliangshanensis*	*Microhyla ornata*	*Amolops marmoratus*	*Euphlyctis cyanophlyctis*	*Haplobatrachus tigerinus*	*Haplobatrachus crassus*	*Limnonectis limnocharis*	*Limnonectes* sp.	*Paa mokokchungensis*	*Rana khare*	*Rana danielii*	*Rana lévida*	*Rana* sp.3	*Rana* sp.1	*Rana* sp.2
Prosotocus infrequentum	-	-	-	-	-	+	-	-	-	-	-	-	-	-	-	-
Mehraorchis ranarum	-	-	-	-	-	+	-	-	-	-	-	-	-	-	-	-
Metacercaria:												-				
Cathaemasia sp.	-	-	-	-	+	+	-	-	-	-	-	-	-	-	-	-
Proalarioides sp.	-	-	-	-	+	-	-	-	-	-	-	-	-	-	-	-
CESTODA: Adult												-				
Baerietta baeri	-	-	-	-	+	-	-	-	+	-	-	-	-	-	-	-
Proteocephalus tigrinus	-	-	-	-	-	+	-	-	-	-	-	-	-	-	-	-
Nemataenioides sp.	-	-	-	-	-	+	-	-	-	-	-	-	-	-	-	-
Plerocercoid larva	-	-	-	-	+	+	+	+	+	-	-	-	-	-	+	-
NEMATODA:Adult *Aplectana gubernaculum*	-	-	-	+	+	-	-	-	-	-	+	+	-	-	+	-
Aplectana. sp.	-	-	-	-	-	+	-	-	-	-	-	-	-	-	-	-
Oxysomatium sp.	+	-	+	+	+	+	+	+	?	-	+	+	+	-	+	-
Paracosmocerca mucronata	-	-	-	+	+	-	-	-	-	-	-	-	-	-	-	-
Oxysomatium macintoshii	-	-	-	-	+	-	-	-	-	-	-	-	-	-	-	-

Condt.

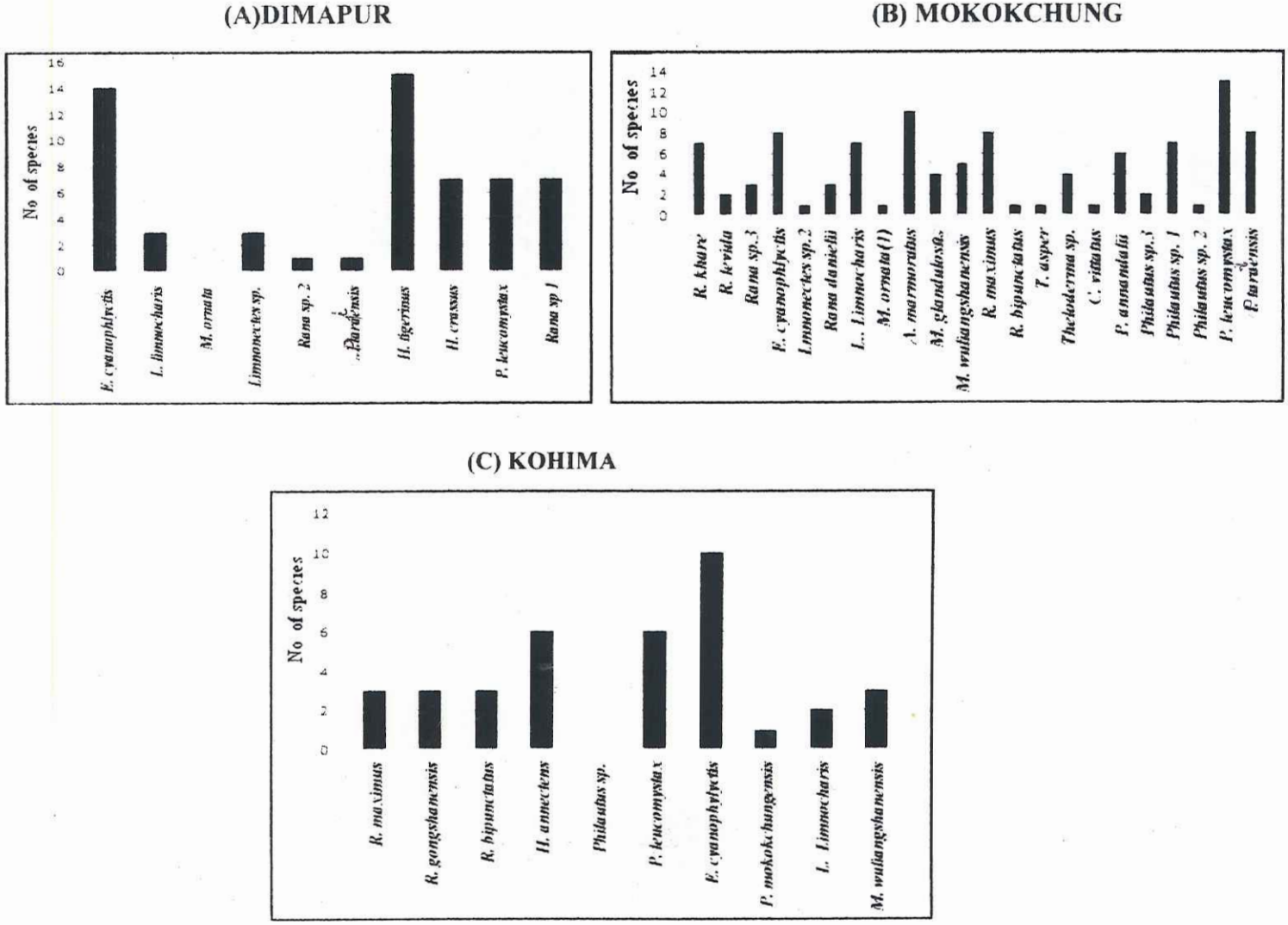
Helminth Parasites	*Megophrys glandulosa*	*Megophrys wuliangshanensis*	*Microhyla ornata*	*Amolops marmoratus*	*Euphlyctis cyanophlyctis*	*Haplobatrachus tigerinus*	*Haplobatrachus crassus*	*Limnonectis limnocharis*	*Limnonectes* sp.	*Paa mokokchungensis*	*Rana khare*	*Rana danielii*	*Rana livida*	*Rana* sp.3	*Rana* sp.1	*Rana* sp.2
Rhabdias ranae	+	+	-	+	+	+	-	+	-	-	+	-	-	+	+	+
Oswaldocruzia goezei	-	-	-	-	+	-	-	-	-	-	-	-	-	-	-	-
Ophidascaris sp.	-	-	-	+	-	-	-	-	-	-	-	-	-	-	-	-
Amplicaecum sp.	-	-	-	-	-	+	+	-	-	-	-	-	-	-	-	-
Icosiella sp. 1	-	-	-	-	+	-	-	-	-	-	-	-	-	-	-	-
Icosiella sp. 2	-	-	-	+	-	-	-	-	-	-	-	-	-	-	-	-
ACANTHOCEPHALAN:												-				
Acanthocephala bufonis	+	+	-	+	+	+	-	-	-	-	+	-	+	+	+	+

Table 18b Helminth parasite and their anuran host in Nagaland

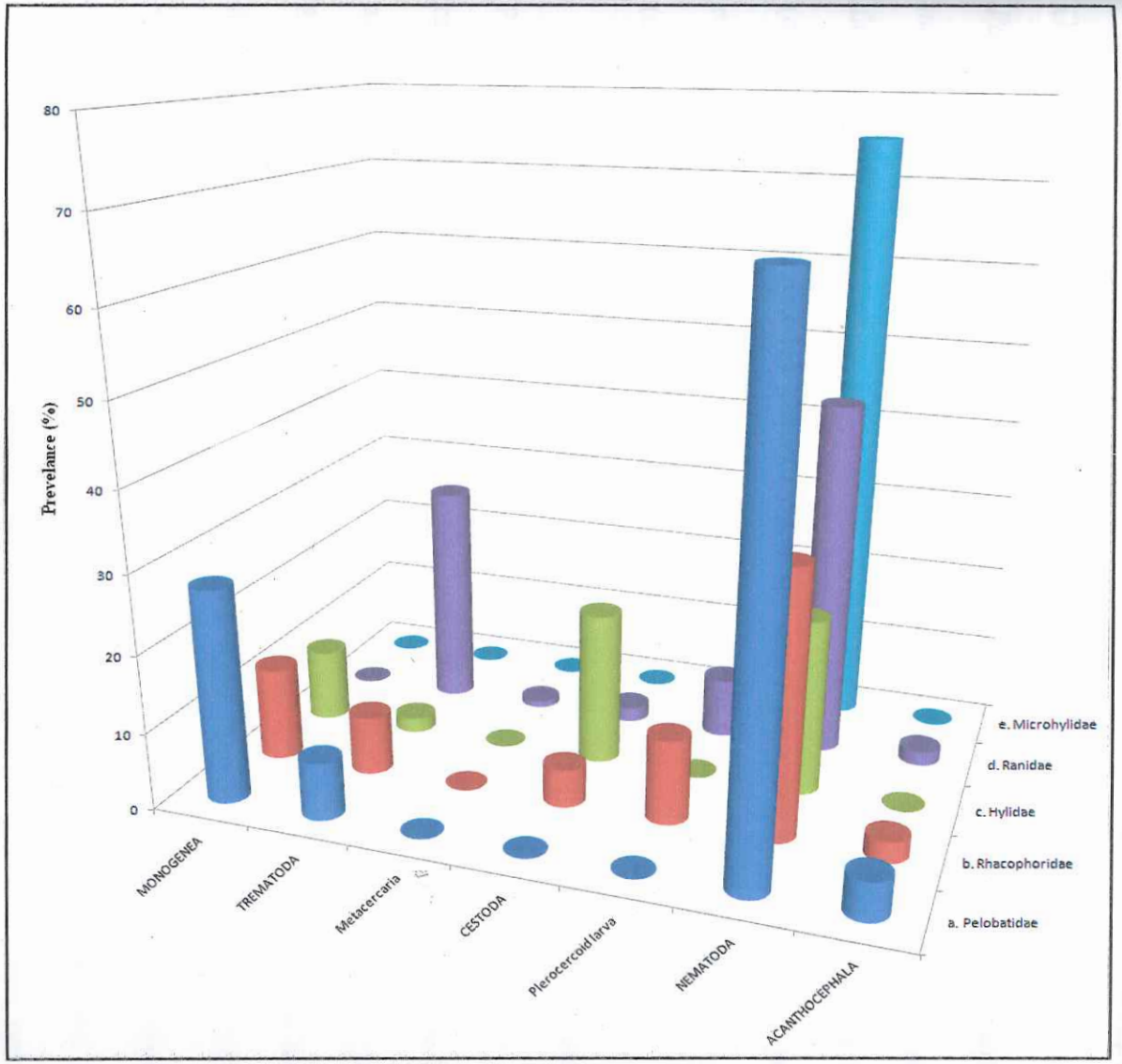
Arboreal habit species

HELMINTH PARASITE	HOST SPECIES												
	Hyla annectans	*Rhacophorus maximus*	*Rhacophorus bipunctatus*	*Rhacophorus gongshanensis*	*Chirixalus vittatus*	*Philautus annandalii*	*Philautus* sp.	*Philautus* sp.1	*Philautus* sp.2	*Theloderma asper*	*Theloderma* sp.	*Polypedates leucomystax*	*P. taraiensis*
MONOGENEA: *Polystoma indicum*	-	-	-	-	-	-	-	-	-	-	-	-	-
Polystoma sp.	-	-	-	-	-	-	-	+	-	-	-	-	-
Polystoma kohimaensis n. sp.	-	-	-	+	-	-	-	-	-	-	-	-	-
Polystoma hylai n. sp.	+	-	-	-	-	-	-	-	-	-	-	-	-
TREMATODA: Adult *Ganeo tigrinum*	-	-	-	-	-	+	-	-	-	-	-	+	-
Pleurogenoides gastroporus	-	-	-	-	-	-	-	-	-	-	-	+	-
Mesocoelium monas	+	-	-	-	-	+	-	-	-	-	-	+	+
Diplodiscus amphichrus	-	-	-	-	+	-	-	-	-	-	-	+	-
CESTODA: Adult *Baerietta baeri*	+	-	-	-	-	+	+	+	-	-	-	-	+
Proteocephalus tigrinus	-	-	-	-	-	-	-	-	-	-	-	-	+
Plerocercoid larva	-	+	+	-	-	-	-	-	-	-	-	+	-
NEMATODA: Adult *Aplectana gubernaculum*	-	+	-	-	-	-	-	-	-	-	-	+	-
Aplectana sp.	-	+	-	-	-	-	-	-	-	-	-	+	-
Oxysomatium sp.	+	+	-	-	-	-	-	+	-	-	-	+	+
Oxysomatium macintoshii	+	-	-	+	-	+	-	-	-	-	-	+	-
Cosmocercella sp.	-	+	-	-	-	-	-	-	-	-	-	+	-
Paracosmocerca mucronata.	-	-	-	-	-	-	-	-	-	-	-	+	-
Pharyngodon sp.					+						+		
Icosiella sp.1	-	-	-	-	-	-	-	-	-	-	-	+	-
Rhabdias ranae	+	+	+	+	-	+	+	+	+	+	+	+	+
ACANTHOCEPHALAN: *Acanthocephalus bufonis*	-	-	-	-	-	-	-	-	-	-	-	+	+

Figs: 278 A-E. Locality-wise species richness of helminth infection in the anuran hosts surveyed



Figs: 279. Overall prevalence of helminths group versus anuran families



Discussion

Of the 39 helminth species reported herein-4 monogenean, 2 trematode, 2 cestode and 3 nematode, all in adult stages are among the 29 species that have hitherto been reported from anuran Amphibia in Meghalaya by Diengdoh (1989) and in Nagaland by Imkongwapang (1997). The monogenean under the genus *Polystoma*, represented by 3 species, namely, *Polystoma* sp., *Polystoma kohimaensis* n. sp., *Polystoma hylai* n. sp. and *Neoriojatrema mokokchungensis* n. g., n. sp., all under the family Polystomatidae, are all presented here in as first records from the Indian subcontinent. *Gorgoderina* sp. and *Nematotaenioides* species (cestode) and the nematodes, *Pharyngodon* sp. and *Paracosmocerca mucronata* are new records from the country. *Prosotocus infrequentum* (trematodes), *Proteocephalus tigrinus* (cestode) and *Aplectana gubernaculums* as new locality record.

There were altogether 14 species of the ranid, 1 species of microhylid and 2 species of pelobatid examined in the study, all of which falls under the **terrestrial-habit species**. The ranids were collected from all the localities under survey, the microhylid from Mokokchung and Dimapur and the pelobatids from Mokokchung, Kohima and Tuensang only. Altogether 848 frogs were examined under the terrestrial-habit species, of which 798 were from the family RANIDAE, 10 from MICROHYLIDAE and 40 from PELOBATIDAE. The hylid comprising of only 1 species, was collected only from Kohima, whereas 12 species of rhacophorids, represented at least by 1 species from each locality were examined for the helminth parasites in the study; both the families fall under the **arboreal-habit species**. A total of 616 frogs were examined under these families, of which 106 were members of HYLIDAE and 510, of RHACOPHORIDAE. The present study, with a larger hosts sample size, revealed the trematodes and nematodes as the most prevalent among the helminth

groups, followed by cestodes, acanthcephalan and monogenean, in receding order, for both the terrestrial- and arboreal-habit species. The Tuensang and Zunheboto sites could not be explored thoroughly due to difficulties of various sorts prevailing in the area and therefore, the host sample size from the two localities was small; in the other three localities, however, a larger number of hosts could be collected and examined. The frogs of the 3 localities revealed a high prevalence of helminth infection, with Dimapur leading in helminth prevalence, followed by Mokokchung and Kohima, the percentage of infection being 61.27%, 59.8% and 36.42%, respectively. With regard to the size of the host, the largest and the heaviest was *H. tigerinus*-represented with the richest helminth infections; *M. microhyla* followed by *Philautus annandalii*, *Philautus* sp. 2, *Philautus* sp. 3, *M. wuliangshanensis*, and *Philautus* sp.1, are small-sized frogs and except for *P. annandalii* and the last 2 hosts, all had low helminth infections. In general, the terrestrial group in all the localities showed very rich helminth fauna in comparison to the arboreal group. But the most diversed helminth fauna, represented by all the 5 helminth groups was in two of the arboreal-host species.

The anurans, numbering 16 in all under the terrestrial-habit species showed adoption of varied habits in the habitat. All did not show strict adherence to a single habitat type only, in that 7 were strictly lentic or still-water-dwelling species and 7, strictly lotic or running-water species. There were also 2 more species with the ability to adapt to any water body type and thus were collected from both the lentic and lotic systems. They are: *E. cyanophlyctis*, occurring in 4 localities of the 5 surveyed and *R. khare* collected from Mokokchung only. Of these *R. khare* sustained 7 parasite species, two trematodes, four nematodes and one acanthocephalan. Earlier, the hosts were collected from a river that revealed a poor helminth infection (Tandon *et al.*, 2001); the hosts collected in the present study from a pond were

found infected with considerably rich nematode fauna. The helminths did not show a uniform trend of prevalence in the host sexes. *E. cyanophlyctis*, examined from all the localities, revealed the richest species composition- represented with 17 helminth species out of the total 39 prevalent and reported; and with trematode as the most dominant group. This host species from Dimapur showed the richest parasite fauna dominated by trematodes, followed by the nematode, cestode and acanthocephalan groups in receding order. This host from Kohima, mostly collected from terrace fields and represented with 10 species, was the second highest in helminth species composition from a single locality, both trematodes and nematodes being equally dominant over the cestodes and acathocephalan; majority of the parasite species were more prevalent in female than in male host. Again this frog host, collected from Zunheboto site, was found harbouring only two nematode species and a cestode larva enclosed in a sac. The hosts of Mokokchung locality numbering 118 were collected from a variety of habitats, such as fishery ponds, terrace fields, small streams, lakes and pot holes; they had comparatively a low species composition that included 8 helminth species only; moreover, in this locality the male hosts showed a higher prevalence for helminth parasites in contrast to the other localities. However, in overall estimation, the females showed a higher prevalence of parasites.

. This host caught from terrace fields in dry season (i.e., February-March) had poor helminth infections when compared with that of the rainy season. Parasite community structure can change seasonally with shifts in host habitat and in diet (King *et al.* 2008). The terrace fields that show a seasonal accumulation of water only during summer and also often used for other crops in winter season, may be affecting the survival of aquatic intermediate hosts of parasites (Tandon *et al.* 2001); anthropogenic activity may influence the natural

changes in transmission dynamics of different species (King *et al.* 2008). *E. cyanophlyctis* collected from river showed very poor helminth fauna. In contrast, many of the same hosts in Dimapur locality were also collected from fishery ponds and terrace fields in the present study, but were found instead harbouring very rich helminth infection. The tropical climatic conditions prevailing in Dimapur provide optimum conditions for the propagation of invertebrate host fauna and thus, indirectly for helminth parasites as well (Tandon *et al.*, 2001).

A total of 5 helminths representing the nematodes with *Oxysomatium* sp, *A. gubernaculum*, *Paracosmocerca mucronata* and *R. ranae* and the acanthocephalan had commonality in the two host species, i.e., *R. khare* and *E. cyanophlyctis*.

The hosts with strictly lentic or still-water dwelling habit; *H. tigerinus*, collected only from Dimapur, supported the second highest in helminth species composition, represented with 18 species and was also the highest in helminth species composition in a host species from a single locality, when compared with all the other anuran host species examined in the study. The trematodes were the dominant species, followed by nematode, cestode and acanthocephalan in receding order. Moreover, the female hosts showed higher prevalence for most of the parasite species except the cestodes, all of which had higher prevalence in male. Another aquatic species also collected only from Dimapur under the same genus but much smaller in size, *H. crassus*, was represented with only 7 species; and both the trematode and nematode groups emerged as equally dominant over the cestode and acathocephalan. The host *L. limnocharis,* collected only from three localities, revealed a very low infection of helminth parasites in two of the localities i.e., Kohima and Dimapur, and was represented with 2 and 3 species, respectively. The same host of Mokokchung had relatively richer species composition

represented with 7 helminth species; here the nematode was the dominant species, followed by cestode and trematode. Another host under the same genus, designated herein as *Limnonectes* sp., collected only from Mokokchung and Dimapur, also had a very low infection, represented with only 1 and 3 parasite forms respectively. Both the *Limnonectes* species were collected from lakes, terrace fields and pot-holes all of which show seasonal accumulation of water. *M. ornata,* the tiniest host specimen in the present study and collected from Mokokchung, was represented by one species of nematode, whereas the same host collected from Dimapur did not carry any infection.

H. tigerinus was the largest and heaviest of all the frogs examined in the present study, often found with crabs and even smaller frogs in the intestinal. With such appetite and feeding habits this frog may be exposing itself to a variety of transmission channels that would facilitate contracting many parasite species and thus contributing to its rich helminth fauna. Accommodation/sustenance of the parasite fauna seems to show an increase value with the length of intestine and also the body size, offering more feeding/dwelling ground for the parasites. Muzzal (1991a, b), in a study on newts, also observed that the helminth infracommunities become more diverge with increase in length or size of the host, suggestedly resulting from the larger number and wider range of intermediate hosts eaten by larger newts in comparison with smaller ones. Thus *H. tigerinus* was frequently found with multiple parasite species, the range being 7-9 species in a single host. Recruitment of most helminths occurs via food-web interactions. This is consistent with the large gape size and voraciously predatory habits of the bullfrog host which is known to ingest "nearly anything that that moves and that it can swallow" (Conant and Collins 1998). Moreover, it was the female host with higher prevalence of helminth than the male hosts. Full grown and larger size female *E.*

cyanophlyctis had more species richness (Tandon *et al.* 2001). *E. cyanophlyctis*, essentially an aquatic species, is an opportunistic and unselective feeder, preying on arthropods and also exhibits cannibalistic habit (Kumar, 1982). The island-size hypothesis of Holmes and Price (1996) also predicts that larger hosts support a richer helminth fauna than smaller ones.

The tropical climatic conditions of the host's collection site and especially the host is feeding habit and size as the contributing factors, might have led the anurans of Dimapur to host a rich parasite fauna. Hence, not only *H. tigerinus* but also *H. crassus* and *Rana* sp.1 showed relatively rich helminth species composition from this locality. All the ranids under this group -(strictly lentic or still-water species) were found co-existing in the habitat and was also observed in the earlier study, the host species collected from the same water body site did not share all the helminth species. The parasite species composition may also be determined by predilection for a particular host species, as exemplified by many parasitic species of Amphibia (Waitz, 1959; del Fosse and Whittaker, 1971; Frandsen, 1974).

Oxysomatium sp. was shared by 7 of the still water-dwelling host species. The trematode *G. tigrinum* was shared by 6 host species, *P. gastroporus*, *D. mehrai*, the lung worm *R. ranae* and *A. bufonis* were common to 4 host species, the nematode, *Aplectana* sp.1, *Icosiella* sp.1 and the lung fluke *H. almorai* in 3 host species; *Paracosmocerca* mucronata 1, *Aplicaecum* m sp. and the metacercaria *Proalarioides* sp. were hosted in two host species. The cestodes *B. baeri* was also shared by 2 host species and except for R. *khare* all hosts were found infected with plerocercoid larva.

There were still 7 more hosts under the group of terrestrial habit that were found using only rivers, small streams, waterfalls, ravines and cliffs in shady areas, forest floors and seldom lakes as their habitats; these hosts were *R. livida*, *Rana* sp.3, *A. marmoratus*, *R.*

danielii, *M. glandulosa* , *M. wuliangshanensis* and *Paa mokokchungensis*. All of them, but *Rana* sp.3 and *M. wuliangshanensis*, were collected mostly from strong current water bodies (co-existing with fish), cliffs, waterfalls and also lakes. In addition, *A. marmoratus* the widest habitat ranging host collected from many type of habitats (such as, large river to small streams, water falls to damped shady cliff or ravine, and even in fishery ponds), hosted the richest helminth species composition within this group represented by 13 helminth species. Both trematode and nematode groups were equally dominant over the other helminth groups; moreover, it was the female hosts that showed the higher prevalence of infection with most of the helminth species. Strong water currents may hinder helminth transmission, as a low diversity of helminth was seen in the case of the newt, *Leurognathus marmorata* that inhabits strong current (Aho, 1990). The host *A. marmoratus*, that covers wide range of area in the habitat in search of mate or while feeding, as was observed throughout the study period, may have the advantage in its dietary trend but exposing itself to a variety of infective stages of parasite species emerged as harbouring a richer helminth infracommunity among the ranid frogs of the lotic system (Muzzal, 1991a, b); The other hosts, *R. danielii* and *M. glandulosa* were collected from rivers, river banks, under the stones etc., and on one occasion even from the forest floor, were found hosting a rich nematode species composition, lacking trematode and cestode, *R. danielli* much smaller in size, also had only nematode infection and lacked all the other helminth groups *M. wuliangshanensis*, another pelobatid but smaller in size and collected from a variety of habitats, was found infected with relatively rich nematode fauna, represented with 4 species. Both the *Megophrys* species of Pelobatidae family from Mokokchung, *M. glandulosa* and *M. wuliangshanensis* were shared in 2 localities, the former and larger pelobatid in Tuensang and the latter in Kohima. The frogs from the lotic systems

showed more prevalence of helminth parasites in females when compared to the male hosts. Further, except for *Rana* sp.3 and *A. marmoratus*, which were equally dominanted by both trematode and nematode species over the other helminth species, the rest of the hosts under this habitat were predominated by nematodes. Relatively richer nematode fauna in the two pelobatids in the study may be attributed to the habit of spending more time in land. The larger of the two species, was often found with large earthworm, small crab and even small frogs in the intestinal contents, which may be a contributing factor for its rich helminth fauna; in this host, females had higher prevalence of helminths, predominated by nematode over the monogeanean species. Except for *R. danielii* and the smallest of this group *M. wuliangshanensis*, the rest of the hosts were large-sized hosts.

Altogether 13 species under the arboreal group were examined in the study and like the terrestrial hosts' varied habit, not all of this group were found to be strictly arboreal but were using both land and trees as their habitat in different seasons; however, another group emerged with strict arboreal habit, regardless of change in season. There were 6 species under the former group, and two of them were represented by all the 5 helminth groups; adopted to both terrestrial and arboreal habitats, they are *R. maximus* available in Mokokchung, Kohima, Zunheboto and Tuensang, shared in all of these localities by 1 monogenean and 1 nematode species; the nematode *Oxysomatium* sp. were shared in host from Mokokchung and Kohima and *Aplectana gubernaculum* in Mokokchung and Zunheboto localities. Trematode and cestode parasites were prevalent only in frogs of Mokokchung. The small sized rhacophorid, *Philautus* sp.1 from Mokokchung (Longkhum) was also collected from Kohima but did not carry any helminth infection; whereas the host from the former locality collected from a small lake surrounded by thick forest had relatively rich species composition represented by 4 of the

5 helminth groups. Another rhacophorid, *P. tardensis* available in Mokokchung and Dimapur harboured only 1 acanthocephalan species out of the 8 parasite forms represented from the former locality. *R. gongshanensis* and *H. annectans,* restricted only to Kohima locality, shared three helminth species viz., *R. ranae* and *O. macintoshii*. *P. leucomystax*, the only host collected from all the localities, was represented by 16 forms of the 39 helminth species reported herein, in overall estimation. This host from Mokokchung locality had the highest species composition, represented with 13 helminths, followed by Dimapur with 7, Kohima and Zunheboto with 6 each and only a trematode species from Tuensang (Noklak) locality. In all the 5 localities male hosts lead in prevalence of helminth infections for this host, with nematodes being the predominant helminth species. The trematode *M. monas* was lacking in the host of Dimapur wherin *G. tigrinum* was the lone trematode candidate, which also was shared by host in Mokokchung; the same frog from the latter site had *D. amphichrus* that was again shared only with frogs of Zunheboto. The nematode *R.ranae* was shared by hosts of all the localities except Tuensang; *Aplectana gubernaculum* and *Aplectana* sp. were prevalent in Mokokchung and Dimapur hosts only and *Oxysomatium* sp., in hosts of Mokokchung, Dimapur and Kohima localities. Kohima was also represented by *Paraosmocerca mucronata* shared with hosts of Zunheboto site. *O. macintoshii* was shared by hosts of Mokokchung and Zunheboto localities and along with these two sites the host of Dimapur was represented by plerocercoid larva. Throughout the study period, this particular group of arboreal hosts was collected from both lakes and trees; the presence of all the 5 helminth groups in some of these species may be attributed to their ability of adaptation to both habitats; especially in breeding season much time is spent in land and water, although they are arboreal in habit. Various

ecological factors, both biotic and abiotic, are well known to play an important role in influencing the host-parasite relationship (Kennedy, 1975).

There were still 7 more anuran species from the arboreal adaptation, which in the present study were categorized as strictly arboreal species and different from those described as arboreal species, in mode of adaptation to habitat, in that all of them were caught from varied types of habitat in the trees only and never on the ground; of these *P. annandalii, Philautus* sp.2, *Philautus* sp. 3, *C. vittatus* and *R. bipunctatus* were all restricted to Mokokchung locality only, except *R. bipunctatus,* which was also collected from Kohima and Tuensang. The 3 *Philautus* species were found completely adapted to small trees, thick bushes etc., such as coffee and tea plants that offer a perfect habitat within the tree itself; especially, in the rainy season where dampening and moistening of the fallen dead leaves within the tree itself occurs making it a perfect place, for dwelling and also mating, which was frequently encountered during the study period. Of the 3 *Philautus* species, *P. annandalii* had relatively rich helminth community, represented by 6 forms and lacking only the monogenea and acanthocephalan; all 3 species shared the lung worm *R. ranae* and the cestode *B. baeri* was shared in the former two hosts only. For *C. vitattus* and *R bipunctatus*, the former species collected from bamboo grooves and small vegetation in a marshy fishery pond, was represented by one helminth species, a trematode and a nematode species (for which the family identification was kept in abeyance for want of fresh specimens). Only 2 specimens of *R. bipunctatus* were caught near a water body from a tree in Mokokchung and were found harbouring a lone monogenean; comparatively hosts from Kohima had richer helminth community, with the representation of 3 helminth species and lacking any trematode and acanthocephalan. The other two species under this group of arboreal habit species were *T. asperum* and *Theloderma* sp., restricted to

Mokokchung locality only, of which the former species showed infection by a lone monogenean. *Theloderma* sp., collected from tree holes and bamboo stump, where rain-water had collected making a small pool of water, was represented by 3 nematodes, a cestode and a cystacanth. The nematode *Oxysomatium* sp., and *R. ranae* and the cestode *B. baeri* had commonality with the other arboreal hosts' parasites; *Pharyngodon mucronata* was shared with *P. annandalii*.

The nematodes dominated all the other helminth groups in the overall arboreal-habit host species, i.e., majority of the arboreal hosts had more nematodes or were represented by at least one species of nematodes per helminth infected host, excepting *Theloderma asperum* and *R. bipunctatus* of Mokokchung. The occurrence of more nematode species in all the arboreal host species can be attributed to thei habit, i.e., spending more time in dry land and trees than in water (Tandon *et al.*, 2001).

In the entire study period the 6 arboreal frogs were caught mostly from land and not trees, sitting behind a stone or in the bushes near by a water body making their mating calls. Except for *P. leucomystax* and *P. annandalii* for ehich the collection comprised both male and female frogs, nearly equal in number, the collection for the other arboreal species was mostly dominated by male hosts and a uniform trend of prevalence sex-wise could not be ascertained, because the male frogs spend more time in or near the water body in the process of making mating calls. For the hosts that were strict arboreal-habit species, especially *P. annandalii*, a relatively high intensity of *M. monas* was revealed; this may be due to the nature of its habitat where the hosts dwells with decaying matters of leaves that attract the intermediate hosts, i.e., snails or insects, for the parasite species.

With the addition of 3 more new species under the *Polystoma* from the region, this monogenean genus was revealed as restricted only in arboreal hosts i.e., rhacophorid and hylid frogs. *Neoriojatrema mokokchungensis* a new form of Monogenea described herein as a new genus and new species under the family Polystomatidae: Polystomatinae was encountered for the first time and the report of this monogenean fluke from the Indian subcontinent is an addition of new genus to the family and *M. glandulosus* of the Pelobatidae family a new host for the fluke.

The trematodes *Gorgoderina* sp. and *Prosostocus infrequentum* are new records from the region and the nematodes, *Aplectana gubernaculum, Paracosmocerca mucronata, O. macintoshii*, are new records from the region. *Proteocephalus tigrinus* and *Nematotaenaeioides* sp. are representatives of cestodes reported for first time from the region.

The present study revealed that the 2 dominant helminth groups were the trematode and nematode, followed by cestode, acanthocephala and monogenean. The overall estimation of the prevalence in host sexes did not show a definite pattern with all the hosts of terrestrial-habit species, except in *R. livida*, *R. danielii*, *H. tigerinus*, *H. crassus, E. cyanophlyctis*, *A. marmoratus* and *M. glandulosus,* which showed higher prevalence in female. For the arboreal-habit, species it was the male hosts that showed overall higher prevalence of helminth parasites, excepting *P. annandalii* and *C. vitattus,* which showed higher prevalence in female hosts. Overall estimation of cestode-infected hosts showed higher prevalence in male than in female hosts; of the 9 host species found infected by plerocercoid larvae, showed higher prevalence in male hosts, whereas in 2 host species, it was equally prevalent. The adult cestode species i.e., *B. baeri*, *Proteocephalus tigrinus* and *Nematotaenioides* sp. showed higher prevalence in male than in the female hosts, except *B. baeri* that was prevalent in 7

host species and which showed higher prevalence in female hosts of *P. annandalii* and *P. taraiensis*. The largest anuran host was from the terrestrial-habit species and showed the richest helminth infra community; the smallest/tiniest again from the same group had the lowest helminth infra community. In conformity with the earlier study the present study also revealed that the most widely distributed helminth parasites in the anuran host group, irrespective of the host habit and larger size samples are the nematodes (Tandon *et al.,* 2001. The trematodes were found parasitizing 17 host species followed by 15 and 12 host species, for cestode and acanthocephalan, respectively.

SUMMARY

1. The study reveals a total of 39 different helminth species occurring in anuran hosts of Nagaland. Twenty nine anuran host species representing 15 genera under 5 families were collected from five localities in the state and were examined for helminth parasites. Three families of Anuran in the study were terrestrial in habit and altogether, 13 species were under the family Ranidae, namely, *Rana khare*, *Rana livida*, *Rana danielii*, *Rana* sp.1, *Rana* sp.2, *Rana* sp.3, *Euphlyctis cyanophlyctis*, *Limnonectes limnocharis*, *Limnonectes* sp., *Haplobatrachus tigerinus*, *Haplobatrachus crassus*, *Amolops marmoratus and Paa mokochungensis*, one species of Microhylidae, *Microhyla ornata* and 2 species of Pelobatidae, *Megophrys glandulosa* and *Megophrys wuliangshanensis*. Ranids were collected from all the localities under survey, microhylid from Mokokchung and Dimapur and the pelobatids from Mokokchung, Kohima and Tuensang. 5 of the ranid species were from lentic system, 6 from lotic system and two of the species from both the systems. The arboreal-habit anurans were from two families Hylidae and Rhacophoridae. The former comprising of 1 species, *Hyla annectans* was collected only from Kohima and 12 species of rhacaphorids represented by at least 1 species from each locality, were examined for helminth parasites in the study; they were, *Rhacophorus maximus*, *Rhacophorus bipunctatus*, *Rhacophorus gongshanensis*, *Chirixalus vitattus*, *Philautus annandalii*, *Philautus* sp.1, *Philautus* sp.2, *Philautus* sp.3, *Theloderma asperum* *Theloderma* sp., *Polypedates leucomystax*, *Ptaraensis*. Of these arboreal anurans, 5 species were found inhibiting both terrestrial and arboreal habitats and 7 of the species only arboreal such as, trees, shrubs and bamboo grooves. The parasites recovered

include 5 monogeneans, 16 trematodes (14 adult and 2 metacercariae), 4 cestodes (3 adult and 1 larval forms), 15 nematodes (14 adults and 1 larval stage) and 1 acanthocephalan. Altogether there were 39 helminths, out of which 4 monogeneans, 2 trematodes, 2 nematodes and 2 cestodes all new reporting are along with the 29 already reported helminth species from the region.

2. The monogenean genus ***Polystoma*** were restricted to arboreal hosts only, and for ***Polystoma indicum*** new hosts are recorded; the ***Polytoma*** sp. ***Polystoma kohimaensis*** and ***Polystoma hylai*** are new species being reported from the Indian subcontinent, their description and validity are discussed. Again, under the family Polystomatidae, a new fluke was recovered from a pelobatid frog of Mokokchung and has been described as a new genus and new species, ***Neoriojatrema mokokchungensis***. The validity of this taxon is discussed and the name proposed after the type locality of the fluke. The trematodes were revealed to be slightly more diverse in the helminth spectrum, with 16 species representing 12 genera under 9 families. The trematode species namely, ***Gorgoderina ellipticum***, ***Diplodiscus amphichrus***, ***Diplodiscus mehrai***, ***Halipegus mehransis***, ***Mesocoelium monas***, ***Ganeo tigrinum***, ***Haematoloechus almorai***, ***Mehraorchis ranarum***, ***Opisthioparorchis indica***, ***Opisthioparorchis yunnanse*** and ***Batrachotrema nagalandensis*** are already known species from the region. For ***Prosotocus infrequentum*** and ***Gorgoderina*** sp. new locality (first record from Northeast India) and new host record have been made; For ***Gorgoderina*** sp. the species identification is kept in abeyance for pending comparison with the hitherto known species of the genus. For ***Diplodiscus amphichrus***, ***Diplodiscus mehrai***, ***Mesocoelium monas*** and the metacercaria ***Proalarioides*** sp. new

hosts are recorded. For the cestodes ***Barietta baeri*** and plerocercoid larvae, new host are recorded. ***Proteocephalus tigrinus*** and ***Nematotaenioides*** sp. are being reported for the first time from anuran hosts of Northeast India; for both the cestodes new hosts are recorded. Fifteen species of nematodes representing 10 genera under 6 families are reported. Of these ***Aplectana*** sp.1, ***Aplectana*** sp.2, ***Oxysomatium macintoshii***, ***Oxysomatium*** sp., ***Cosmocercella*** sp., ***Rhabdias ranae***, ***Oswaldocruzia goezei***, ***Ophidascaris*** sp., ***Amplicaecum*** sp., ***Icosiella*** sp.1, ***Icosiella*** sp.2 and ***Kalicephalus*** sp. are already known species from the region. For ***Oxysomatium macintoshii***, a new locality (Nagaland) and new host are recorded. ***Paracosmocerca mucronata*** is reported for the first time from Northeast India. New locality in the state and new host are recorded for ***Aplectana gubernaculum***, ***Oxysomatium*** sp., ***Rhabdias ranae***, ***Oswaldocruzia goezei***. For many of these nematode species identification could not be carried out due to pending comparison with the hitherto known species of the genus. For ***Acanthocephalus bufonis*** new host and new localities are recorded.

3. Surface fine topographic studies of 19 helminth species are revealed using scanning electron microscopy. The species include ***P. indicum***, ***P. hylai*** n. sp. and ***N. mokokchungensis*** n.g., n. sp. from urinary bladder; ***D. amphichrus***, ***D. mehrai***, ***O. indica***, ***B. nagalandensis***, ***M. monas***, ***M. ranarum***, ***Prosotocus infrequentum***, ***Aplectana gubernaculum***, ***Aplectana*** sp., ***O. goezei***, ***Ophidascaris*** sp., ***B. baeri*** and ***A. bufonis*** from intestine; ***H. almorai*** and ***R. ranae*** from lungs and plerocercoid larva from muscle tissue.
4. The prevalence, intensity and species richness of the helminths in all the anuran hosts species has been studied and the helminth spectrum analysed with regard to the host,

its sex and locality. The Tuensang and Zunheboto sites could not be explored thoroughly due to the unforeseen problems prevailing in the area and the host samples size from the two localities were small, whereas in the other three localities, maximum number of hosts could be collected and examined. The frogs of these 3 localities revealed a high prevalence of helminth infection; with Dimapur leading and followed by Mokokchung and Kohima, the percentage of infection was 61.27%, 59.8% and 36.42%, respectively. In general, the terrestrial group of hosts in all the localities showed very rich helminth fauna in comparison to the arboreal group and had representation by by all the 5 helminth groups. The same was also true, with the latter group revealing diverse helminth fauna, represented by all the 5 helminth groups in at least two of the host species. The largest host was *H. tigerinus* represented with the richest helminth infection from a single locality; the smallest was *M. microhyla* followed by *Philautus annandalii, P. garo, Philautus* sp. 2, *M. wuliangshanensis* and *Philautus* sp.1, with increasing order in size; except for *P. annandalii* and the last 2 hosts, all had low helminth infections. Of the 29 host species examined, in overall estimation ***E. cyanophlyctis*** showed the widest spectrum of helminth parasites, followed by ***P. leucomystax*** and ***H. tigerinus***, The occurrence of more helminth species in the anuran hosts of Dimapur and relatively less species in other localities has been discussed. Nematodes emerged as the more predominant group among the host species with the more terrestrial or semi-aquatic life, whereas the trematodes were found to predominate in those with more aquatic habitat.

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ON TWO NEW SPECIES OF THE TREMATODE GENERA, *OPISTHIOPARORCHIS* WANG, 1980 AND *BATRACHOTREMA* DOLLFUS AND WILLIAMS, 1966 (BATRACHOTREMATIDAE), WITH A REPORT OF A CHINESE SPECIES OF *OPISTHIOPARORCHIS* FROM ANURAN AMPHIBIAN HOSTS IN INDIA

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web supplement

ABSTRACT

*An examination of small samples of three species of anuran frogs (*Amolops afghanus, Rana liebigii *and* R. khare*) originating in Nagaland (northeastern India) revealed three species of trematodes, two of them being new to science. These include* Batrachotrema nagalandensis *sp. nov. (type host* A. afghanus*), Opisthioparorchis indica sp. nov. (type host* A. afghanus*) and* O. yunnanse *Li, 1996. This is the first record of their respective genera in India. The distinguishing features of* B. nagalandensis *sp. nov. are - testes located in the middle third of the body, and long cirrus sac extending posteriorly beyond the ventral sucker.* O. indica *sp. nov. is distinguished by having oral and ventral suckers of almost equal size, intestinal caeca extending beyond the ventral sucker up to the anterior border of testes, the genital pore at the level of the oral sucker and vitellaria confluencing medially in the pre and post testicular regions. Kohima (Nagaland) and* Rana liebigii *form a new locality and new host record, respectively, for* O. yunnanse.

KEYWORDS

Amolops afghanus, Batrachotrema nagalandensis *sp. nov., India, Nagaland,* Opisthioparorchis indica *sp. nov.,* Opisthioparorchis yunnanse, Rana khare, Rana liebigii, *Trematode*

During an exploratory survey of the helminth parasite fauna of anuran Amphibia in Nagaland (India) several digenetic trematode species were encountered. Two of these forms, on study, were found to be new to science and belong each to the genera *Batrachotrema* Dollfus and Williams, 1966 and *Opisthioparorchis* Wang, 1980. While the former genus has hitherto been reported from Africa (Dollfus & Williams, 1966), China (Wang, 1981; Zhang & Sha, 1985; Liang & Ke, 1988) and Vietnam (Moravec & Sey, 1989) the latter is represented so far in ranid frogs in China. The present communication deals with the description of a new species of each genus, and also forms the first record of the occurrence of these genera in the Indian subcontinent. The occurrence of *O. yunnanse* Li, 1996 is also reported from *Rana liebigii* as a new record.

MATERIALS AND METHODS

Eight specimens (range 1-4) of *Batrachotrema* were collected from the intestine of five *A. afghanus* and only one specimen, from *Rana khare* at Mokokchung in Nagaland. Twenty-one specimens of a form belonging to *Opisthioparorchis* were recovered from the intestine of three *Amolops afghanus* from Mokokchung, the maximum number of this parasite in a single host being 18. Two specimens, also belonging to the same genus but representing another species were collected from one *Rana liebigii* from Kohima.

Whole mount preparations of the flukes were made following standard procedure, using Mayer's carmalum or borax carmine as stains. For scanning electron microscopy (SEM) the specimens fixed in 4% cold neutral buffered formalin were processed and treated with tetramethylsilane as per the method described by Roy and Tandon (1991), metal coated and viewed under a JSM-35 CF (Jeol) scanning electron microscope at accelerating electron voltage ranging between 10-15kV.

Family Batrachotrematidae
Genus *Batrachotrema* Dollfus and Williams, 1966

***Batrachotrema nagalandensis* sp. nov.**

(Figs. 1-3; Images 1-2[w])

Material examined

Holotype: W 8338/1; from the intestine of *Amolops afghanus* (Günther), Mokokchung, Nagaland (25°-27°4'N & 93°2'-95°15'E).
Paratypes: W 8339/1, W 8340/1; from the intestine of *Amolops afghanus* (Günther) and *Rana khare* (Kiyasetuo & Khare), Mokokchung.
Deposited at the Zoological Survey of India, Kolkata.

Etymology

Named after Nagaland state, India.

Specific diagnosis

Body unspined, fusiform, testes located in middle third of body; cirrus sac long, extending beyond ventral sucker posteriorly, ovary rounded.

Diagnosis

(Based on measurements of 5 mature specimens and SEM observations on 1 specimen.). Body elongated or some what spindle shaped, broadest at shoulder, tapering and terminating bluntly towards anterior and posterior regions; surface devoid of spines. Oral sucker subterminal. Ventral sucker pre-

[w] Also see SEM Images in the web supplement at www.zoosprint.org

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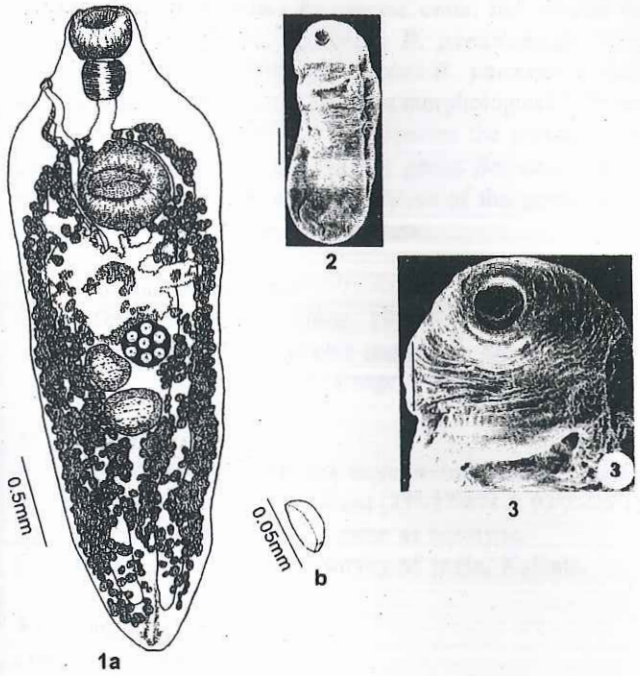


Figures 1-3. **Batrachotrema nagalandensis *sp. nov.***
1a - Whole mount, dorsal view; 1b - Egg; 2-3 - Scanning electron micrographs: 2 - Full worm, ventral view (scale 100mm); 3 - A magnified view of anterior end, showing the oral end and ventral sucker (scale 100mm).
[See Figures 2 & 3 on the web as Images 1 & 2]

Table 1. Measurements (in mm) of **Batrachotrema nagalandensis *sp. nov.***

Characters	Range (Mean)	± S.D.
Length of Body	2.38-3.89 (2.93)	0.49
Maximum width of body (at level of ventral sucker)	0.80-1.27 (1.01)	0.202
Oral sucker:		
Length	0.17-0.27 (0.21)	0.043
Breadth	0.26-0.37 (0.33)	0.034
Ventral Sucker:		
Length	0.30-0.49 (0.42)	0.07
Breadth	0.39-0.56 (0.47)	0.07
Pharynx:		
Length	0.17-0.25 (0.22)	0.032
Breadth	0.17-0.31 (0.20)	0.036
Length of Oesophagus	0.24-0.46 (0.41)	0.023
Distance of intestinal caeca from hind end	0.07-0.25 (0.22)	0.037
Intestinal bifurcation from anterior end	0.46-0.75 (0.64)	0.01
Testis I:		
Length	0.20-0.22 (0.22)	0.017
Breadth	0.22-0.34 (0.27)	0.035
Testis II:		
Length	0.24-0.46 (0.31)	0.077
Breadth	0.19-0.36 (0.31)	0.058
Ovary:		
Length	0.22-0.29 (0.26)	0.012
Breadth	0.19-0.28 (0.24)	0.022
Extent of Vitellaria	1.89-3.05 (2.37)	0.419
Eggs	0.03-0.04 x 0.02-0.023 (0.036 x 0.022)	0.002

equatorial, located in broader part of body, three times larger than oral sucker, covering half of body in width, strongly muscular. Prepharynx indistinct; pharynx muscular; oesophagus of moderate length or short; intestinal caeca reaching near posterior end. Testes rounded or oval, lying in tandem in middle third of body; anterior testis at level of ovary, slightly smaller than posterior; cirrus sac long, extending posteriorly beyond ventral sucker, lying on its left side, extending anteriorly up to and ending in level with pharynx. Ovary rounded, pretesticular, postacetabular; oviduct, Mehlis' gland complex in region anterior to ovary; uterine coils mainly limited in ovarian and postacetabular area, overlapping intestinal caeca. Genital pore at left side of body, marginal or sub marginal, male and female pores opening into genital atrium. Vitellaria follicular, extending from level of intestinal bifurcation up to posterior most part of body, overlapping intestinal caeca, also scattered in intercaecal fields. Eggs numerous, large, oval. The measurements of this form are given in Table 1.

Discussion

In having a pretesticular ovary, which is nearer to the anterior testis than the ventral sucker and the genital pore lying laterally to the median line in level with the pharynx, the present form bears a close morphological resemblance to the genus *Batrachotrema*. Dollfus and Williams (1966) proposed a new family Batrachotrematidae for the genus. Yamaguti (1971) retained the family as proposed by Dollfus and Williams and Wang (1980) also accepted its validity. However, Prudhoe and Bray (1982), on the basis of its close morphological relationship with the members of Opecoeliidae, placed the genus under the latter family.

The genus *Batrachotrema* so far includes five species: *B. petropedetis* Dollfus and Williams, 1966 from *Petropedetes natator* in Sierra Leone, Africa; *B. pseudobagri* Wang, 1981 (from *Pseudobagrus fulvidraconis* and *Pseudogastromyzon zebroidus* in Fujian Province, China); *B. yaanensis* Zhang and Sha, 1985 (from *Rana phrynoides* in Yaan, Sichuan, China); *B. opistosacca* Liang and Ke, 1988 (from *Rana spinosa* in Meixian, Guangdong, China); *B. vietnamensis* Moravec and Sey, 1989 (from *Rana kuhlii* in Hanoi, Vietnam).

A comparison of the present form with the type species reveals several differences between the two. In the present form the testes lie in the middle third of the body and the cirrus sac is long extending posteriorly beyond the ventral sucker. In the type species the testes are located much posteriorly and the claviform cirrus sac is quite small extending posteriorly only up to the anterior margin of the ventral sucker. Both *B. petropedetis* and *B. opistosacca* have an elongated oval body shape and have intestinal caeca extending posteriad a little beyond the testes. In having a fusiform body, the present species resembles *P. pseudobagri*, a species described from piscine hosts, but distinctly differs from it in having a larger body size, a rounded ovary (lobate in *P. pseudobagri*), and a long cirrus sac that extends posteriad much beyond the ventral sucker. In possessing an unspined body, testes situated in the middle third of the body and immediately pretesticular ovary that is

not separated from testes by uterine coils, the present form comes close to *B. petropedetis*, *B. pseudobagri* and *B. opistosacca* and stands apart from *B. yaanensis* and *B. vietnamensis*. In view of the apparent morphological differences and also different amphibian host species the present form is considered a new species under the genus *Batrachotrema*. It is for the first time that a representative of the genus is being reported from the Indian subcontinent.

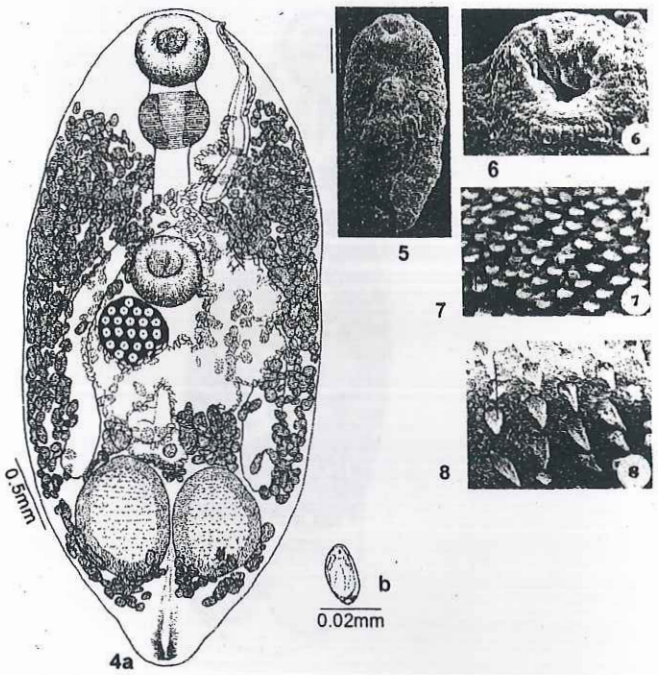
Family Batrachotrematidae
Genus *Opisthioparorchis* Wang, 1980

***Opisthioparorchis indica* sp. nov.**
(Figs. 4-8; Images 3-6[w])

Material examined
Holotype: W 8341/1; from the intestine of *Amolops afghanus* (Günther), Mokokchung, Nagaland (25°-27°4'N & 93°2'-95°15'E)
Paratypes: W 8342/1, 8343/1; same as holotype.
Deposited at the Zoological Survey of India, Kolkata.

Etymology
Named after the country.

Specific Diagnosis
Intestinal caeca extending much beyond ventral sucker up to anterior level of testes, ventral sucker almost equal to oral sucker in size, genital pore in level with anterior margin of oral sucker, vitellaria extending beyond testes posteriorly, confluent medially in pre-and posttesticular regions.



Figures 4-8.* Opisthioparorchis indica *sp. nov.
4a. Whole mount, ventral view, b. Egg; 5-8. Scanning electron micrographs: 5 - Whole worm, ventral view (scale 100mm); 6 - Oral end, magnified view (scale 10mm); 7 - A magnified view of the tegumental spination in the mid ventral region (scale 10mm); 8 - Tegument of the posterior part of the body depicting conical spines (scale 10mm) [See Figures 5-8 on the web as Images 3-6]

Diagnosis
(Based on measurements of 10 mature flukes and SEM observations on 2 specimens.). Body elongate or oval with rounded anterior end, rounded or slightly conical posterior end, spinose. Oral sucker subterminal, spherical. Ventral sucker pre-equatorial or equatorial almost equal to oral sucker in size. Prepharynx indistinct; pharynx muscular; oesophagus of moderate length; intestinal caeca extending in postacetabular region up to level of anterior margin of testes. Testes round or oval, lying symmetrically juxtaposed in posterior region of body; cirrus sac well developed, elongated, somewhat S-shaped, recurved, lying on left side of body, extending from near junction of oesophagus and intestinal caeca up to anterior margin of oral sucker. Ovary rounded, pretesticular, postacetabular; receptaculum seminis conspicuous, lying posterior to ovary; uterus pretesticular, occupying major area between ovary and testes, extending anteriad partially overlapping intestinal caeca. Genital pore marginal on left at level of anterior margin of oral sucker. Vitellaria in varying follicular size, extending in lateral fields from level of pharynx posteriorly up to post-testicular region, becoming confluent medially in intercaecal, pretesticular and post testicular fields. Eggs numerous, ovoid, operculate.

SEM revealed the surface fine topography of the fluke. The body surface is studded throughout with dense spination except for the anterior circum-oral region, which appears devoid of spines. Whereas the spines covering the tegument in most parts of the body are scale like with rounded tips, those abounding in the posterior region are conical, with broad base and pointedly tapering distal end. The non-spinous tegument of the anterior region presents a spongy texture. The measurements of this form are given in Table II.

Discussion
In having a well-developed cirrus sac and marginal or submarginal genital pore located in the region of the oral sucker or pharynx, the present form belongs to the family Batrachotrematidae.

The genus *Opisthioparorchis* was created by Wang (1980) for an intestinal fluke of *Rana spinosa*, which was characterized by having juxtaposed testes located at the posterior end of the body and intestinal caeca extending up to just near the anterior border of testes. At present the genus includes six species, all described from China: *O. ranae* Wang, 1980 (type species) from *Rana spinosa* in Fujian Province South; *O. pleurogenitus* Wang, 1980 from the same host and locality as those of the type species; *O. boheansis* Wang, 1980 from *Staurois wuyiensis* also from Fujian; *O. megaloonis*. Liang and Ke, 1988 and *O. meixianensis* Liang and Ke, 1988, both from *R. spinosa* in Changsha (Meixian, Guangdong Province) and *O. yunnanse* Li, 1996 also from *R. spinosa* from Yunnan Province.

In having the vitellaria extending in the posttesticular region,

Table 2. Measurements (in mm) of* Opisthioparorchis indica *sp. nov.

Characters	Range (Mean)	± S.D.
Length of Body	1.80-2.33 (2.05)	0.18
Maximum width of body (at level of ventral sucker)	0.89-1.08 (0.97)	0.08
Oral sucker:		
Length	0.21-0.26 (0.24)	0.02
Breadth	0.23-0.30 (0.26)	0.026
Ventral Sucker:		
Length	0.21-0.25 (0.23)	0.014
Breadth	0.21-0.27 (0.24)	0.018
Pharynx:		
Length	0.19-0.23 (0.21)	0.018
Breadth	0.2-0.24 (0.22)	0.016
Length of Oesophagus	0.14-0.23 (0.18)	0.03
Distance of intestinal caeca from hind end	0.58-0.77 (0.68)	0.06
Intestinal bifurcation from anterior end	0.53-0.65 (0.61)	0.04
Testis (right):		
Length	0.33-0.46 (0.39)	0.04
Breadth	0.26-0.33 (0.29)	0.023
Testis (left):		
Length	0.33-0.41 (0.38)	0.03
Breadth	0.25-0.32 (0.28)	0.029
Cirrus sac Length	0.52-0.66 (0.57)	0.05
Ovary:		
Length	0.17-0.25 (0.22)	0.027
Breadth	0.17-0.23 (0.20)	0.022
Receptaculum seminis:		
Length	0.36-0.53 (0.44)	0.07
Breadth	0.03-0.065 (0.047)	0.013
Eggs	0.027-0.037 x 0.011-0.018 (0.031 x 0.015)	0.002

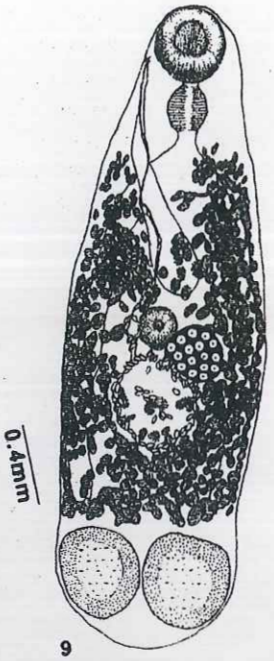


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Figure 9.* Opisthioparorchis yunnanse *Li, 1996* *Whole mount, ventral view

***Table 3. Measurements (in mm) of* Opisthioparorchis yunnanse**

Characters	Measurement
Length of Body	2.5
Maximum width of body (at level of ventral sucker)	1.94
Oral sucker:	
Length	0.28
Breadth	0.29
Ventral Sucker:	
Length	0.08
Breadth	0.08
Pharynx:	
Length	0.18
Breadth	0.20
Length of Oesophagus	0.09
Distance of intestinal caeca from hind end	0.58
Intestinal bifurcation from anterior end	0.6
Testis (right):	
Length	0.37
Breadth	0.35
Testis (left):	
Length	0.38
Breadth	0.31
Cirrus sac Length	0.97
Ovary:	
Length	0.23
Breadth	0.23
Receptaculum seminis:LengthBreadth	0.420.047
Eggs	0.041 x 0.021

the present form resembles the type and other species but stands apart from *O. pleurogenitus* and *O. yunnanse,* in both of which the vitelline follicles are distributed only in the pretesticular region and the testes are the posterior most structures in location in the fluke body. It also differs from all those species having posttesticular vitellaria in several characters; the latter species, have a much smaller body size, the ventral sucker is smaller than the oral and unlike the present form, the vitellaria do not become confluent medially in the pretesticular, inter-intestinal and posttesticular regions.

In view of the conspicuous differences of the present form with the hitherto known species of *Opisthioparorchis*, it is proposed to consider this as a new species named *O. indica.*

***Opisthioparorchis yunnanse* Li, 1996**
(Fig. 9)

Material examined
From the intestine of *Rana liebigii* (Günther), Kohima (Nagaland, 25°-27°4'N, India).

Diagnosis
(Whole mount). Body elongate, somewhat narrow anteriorly, broader posteriorly, with bluntly rounded ends, spinose. Oral sucker subterminal, almost twice as large as ventral sucker, latter equatorial in position. Prepharynx short, pharynx muscular; oesophagus short; intestinal caeca extending posterial up to a little in front of testes. Testes round or oval, juxtaposed near posterior extremity of body; cirrus sac club-shaped, elongated, extending from a little in front of ventral

sucker anteriad up to level of oral sucker. Ovary rounded, just by side or slightly posterior to ventral sucker; uterus mainly in near post ovarian region, extending as ascending narrow tube anteriad. Genital pore marginal, on left at level of oral sucker. Vitellaria extending extensively from level of oesophagus posteriad up to just in front of testes. Eggs elliptical, operculate. The measurements of this form are given in Table III.

Remarks

The whole mount preparation of one specimen collected from *Rana liebigii*, though apparently resembled *Opisthioparorchis* species, turned out to be different from the new species of the genus described above. In its general morphology (elongated body, oral sucker larger than the ventral sucker- almost double the size, vitellaria limited to only pretesticular zone and long club-shaped cirrus sac), the present specimens stands close to *O. yunnanse* Li, 1996, which was originally described from *Rana spinosa* from Yunnan Province, China. However, the only conspicuous deviation it shows from this species is the length of the oesophagus; in *O. yunnanse* the oesophagus is quite long, though short in the present form. However, considering that just one character i.e. length of the oesophagus, is not enough to erect a new species, the present form is considered representing *O. yunnanse*.

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