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ON THE REVIEW OF THE GENUS NEOPRONOCEPHALUS MEHRA, 1932 (TREMATODA)

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Abstract. The systematics of the species of the genus *Neopronocephalus* Mehra, 1932 is revised. A detailed redescription is given of the species *N. triangularis* and several signs have been found, by which the diagnosis of the genus *Neopronocephalus* and the subfamily Neopronocephalinae may be extended. In view of a variability inside the species *N. triangularis*, the species *N. gangeticus* Mehra, 1932, *N. mehrai* Chatterji, 1936 and *N. rotundus* Siddiqui, 1965 have been placed in synonymy with *N. triangularis* Mehra, 1932.

The genus *Neopronocephalus* was created by Mehra (1932) for *N. triangularis* and *N. gangeticus* obtained simultaneously from the gut of *Kachuga dhongoka* from Allahabad. Subsequently Chatterji in the year 1936 described the third species *N. mehri* from the gut of *Morenia ocellata* from Rangoon. Siddiqui (1965) described *N. rotundus* from *Cyclemys dentata* from Aligarh, India. Quite recently Gupta (1968) recorded *N. triangularis* Mehra, 1932 from a different host, a fresh water tortoise, *Geoclenys hamiltoni* again from India. Thapar in 1968 described the larval form of this genus, the cercaria of *N. indicus*.

A large number of trematodes of this genus *Neopronocephalus* were collected from *Morenia ocellata* from and around Fateh Sagar lake of Udaipur. These forms reveal new and interesting morphological data unnoticed and undescribed so far and also show many variable characters which perhaps led earlier workers to believe *N. triangularis* Mehra, 1932, *N. gangeticus* Mehra, 1932, *N. mehrai* Chatterji, 1936 and *N. rotundus* Siddiqui, 1965 to be distinct species. To the present authors these four species appear nothing but the form described as *N. triangularis* Mehra, 1932. The need to take up its study was appreciated when even the recent workers created the new species, *N. rotundus* solely on the basis of measurements which might vary and failed to notice the significant characters viz. the presence of the "canoe" and the filamented nature of the egg.

MATERIAL AND METHOD

The Baylis and Monro technique (1941) was followed for the collection and preservation of adult trematodes. The different regions of the alimentary canal of the host were examined piece by piece in physiological saline solution for the release of the worms. Flukes were thoroughly cleaned of debris in 1 % sodium chloride and then fixed in two lots one for making wholemounts and the other for

section cutting. More than two hundred specimens were studied both as wholemounts and serial sections. Fresh frozen sections were also obtained with the freezing microtome. For whole mounts the following fixatives were employed, Bouin's fluid, Corrosive acetic acid and F.A.A. Grower's acidified carmine was used as the stain and acid alcohol as the destaining reagent. The sections cut at 10 μ were stained either in Delafield's haematoxylin or iodine-ripened haematoxylin and were counterstained in eosin.

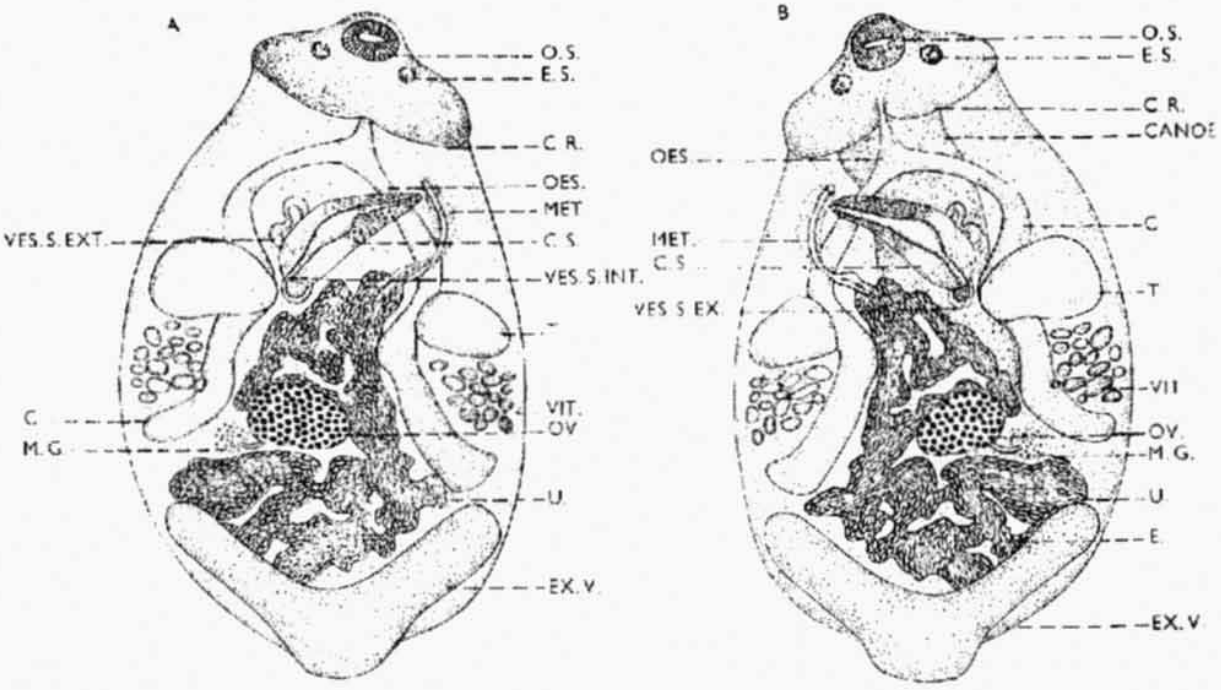

Fig. 1. *Neopronocephalus triangularis* Mehra, 1932. A — dorsal view, B — ventral view.
C. — caecum; CA. — canoe; C.S. — cirrus sac; C.R. — collar region; E. — egg; E.S. — eye spot; EX.P. — excretory pore; EX.V. — excretory vesicle; MET. — metraterm; M.G. — Mehlis' gland; OV. — ovary; O.S. — oral sucker; OES. — oesophagus; POST. REGION — posterior region; T. — testis; U. — uterus; VIT. — vitellaria; VES.S.EX. — vesicula seminalis externa; VES.S. INT. — versicula seminalis interna.

REDESCRIPTION OF *N. TRIANGULARIS*

Host: *Morenia ocellata;* locality: Udaipur, India; location: intestine.
Redescription based on 210 specimens examined. The specimens deposited in author's personal collection.

Body small rather plump with bluntly rounded to truncate posterior end. Cuticle* thin, aspinose. Musculature well developed especially in the cephalic region. A prominent constriction separates it from the rest of the body. While the constriction is fairly prominent in partially pressed and unpressed specimens, in pressed forms it becomes obscured by the muscle fibres surrounding this area. Head collar complete dorsally and laterally but incomplete ventrally. The groove, thus formed by the lobes of the collar, varies in its depth and may even extend to be very near the oral sucker thus giving the appearance that the two lobes of the collar are entirely separated (Fig. 2A),

The "canoe" extends from the base of the collar to the level of vitelline follicles, never crossing this region (Fig. 2A). Its breadth varies due to different orientation of the two lateral margins. Body is 0.600—3.070 mm long and 0.360—1.110 mm wide

*) Now called epidermis (after the study of fine structure).

at the vitelline region. The oral sucker is rather small and subterminal and measures 0.075—0.105×0.090—0.150 mm. Pharynx absent. Oesophagus comparatively long, measuring 0.105—0.405 mm. Intestinal bifurcation 0.165—0.525 mm from anterior extremity of the body. Caeca simple, arch over cirrus sac and undulate to terminate a little behind vitellaria about one third of the body length from the posterior extremity. Genital pores separate, situated on the left side of the median line, a little behind the intestinal bifurcation. Male opening ventral, occasionally extracaecal to left caecum. Female pore slightly away from the male but definitely extracaecal.

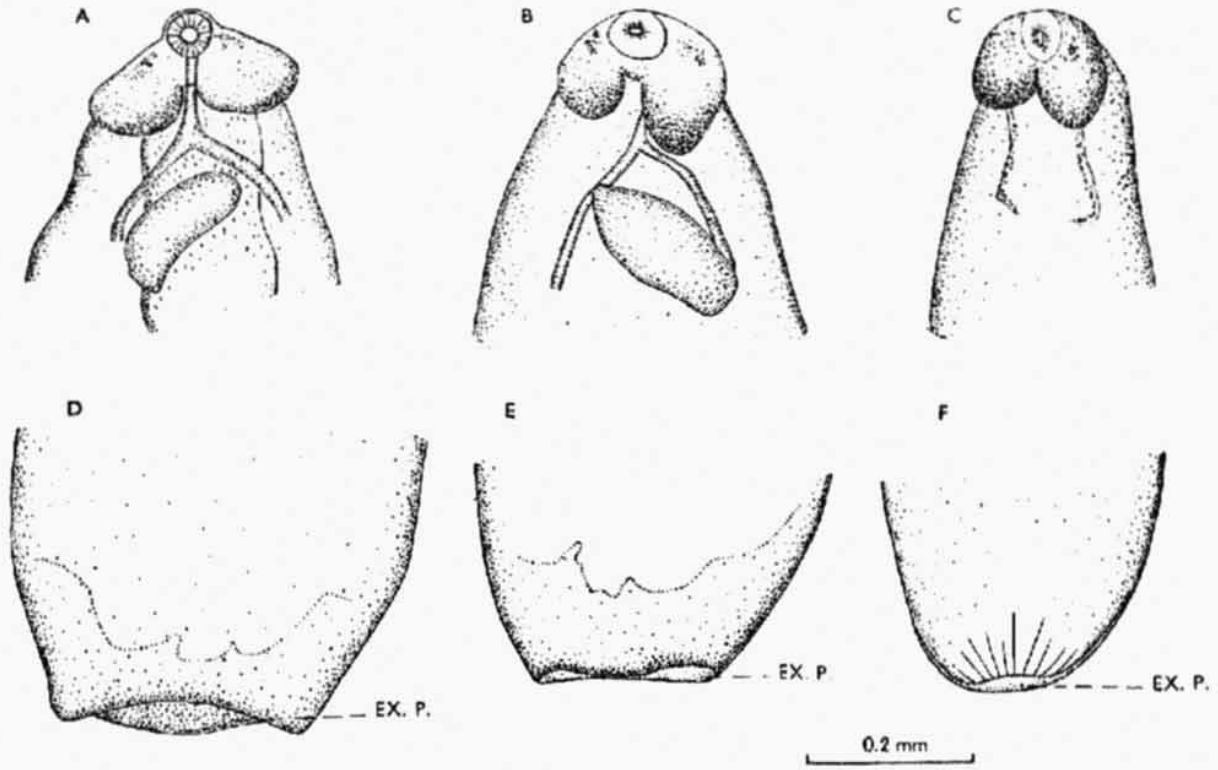

Fig. 2. *Neopronocephalus triangularis* Mehra, 1932. A — cephalic region with canoe and collar, B, C — cephalic region with collar, D, E, F — posterior region.

Testes two, symmetrical, slightly preequatorial, lateral to caeca except the inner lateral margin which is overlapped by the caeca in some specimens. Testes oval, almost equal, the left measuring 0.060—0.375×0.105—0.345 mm and right 0.060—0.360× ×0.120—0.345 mm. Cirrus pouch well developed obliquely placed in pretesticular, intercaecal field. It measures 0.105—0.495×0.060—0.225 mm. Vesicula seminalis externa coiled at the base of the cirrus sac. It extends into the latter for some distance as a straight tube and then swells up to form the pars prostatica, which narrows down anteriorly to form the ejaculatory duct. The whole of the interior of the cirrus sac is filled by glandular cells, the one surrounding the ejaculatory duct being most obvious.

The ovary is post-testicular and post-equatorial and is shifted from the median line to be opposite the genital openings. It is nearly oblong, sometimes with an irregular margin, it measures 0.060—0.210×0.075—0.195 mm in diameter. The common vitelline duct from the vitelline reservoir joins the oviduct. A Mehlis' gland surrounding the cotype is present. The vitelline follicles are massed together immediately behind each testis, on the right side 10 to 18 follicles while there are 8 to 16 on the left. Most of the follicles are lateral, few being ventral to caeca. The uterus is coiled, extending behind the ovary, the greater number of coils being between the left testis and the ovary and

then between the two testes. The metraterm is well differentiated, lying lateral to the caeca. Eggs numerous, bipolar, thin-shelled, non-operculate and thread like. These measure 0.090—0.105 × 0.015—0.021 mm. The excretory vesicle is V-shaped, the arms being fairly big extending to more than half the distance between the ovary and the posterior extremity of the body. Its lumen sometimes shows signs of branching. The excretory opening is subterminal and situated on the ventral side.

DISCUSSION

The genus *Neopronocephalus* was created by Mehra in 1932 for digenetic trematodes recovered from *Kachuga dhongoka* and assigned to a new subfamily Neopronocephalinae Mehra, 1932. The necessity, for the creation of a new genus was due to the topographical positions of the female reproductive system — (I) the ovary being post-testicular, (II) the vitelline follicles (though not mentioned in the subfamily character but mentioned in the generic diagnosis being post-testicular and (III) the presence of extensions of the uterus posterior to ovary.

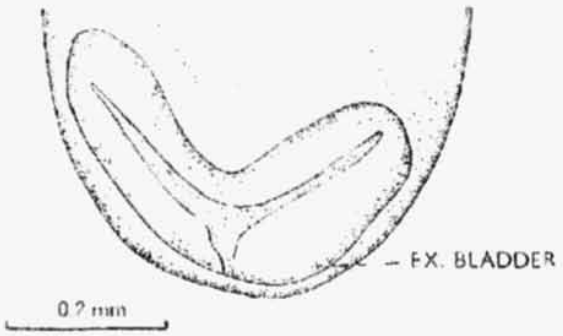

Fig. 3. *Neopronocephalus triangularis* Mehra, 1932 — posterior region.

In none of the genera and the subfamilies of Pronocephalidae, is there any account of the presence of a "canoe" which to the present authors appears to be a very significant feature. The "canoe" is morphologically quite distinct from the "scoop" or ventral anterior depression mentioned in some of the genera. The "canoe" highly characterizes Neopronocephalinae by its presence. It can clearly be seen in sections only. It is just like an excavation dug out from a solid mass whose lateral walls are contractile due to which the excavation may appear to be oriented differently. It extends from the anterior side just below the collar to the second third of the posterior side.

The oculate condition of the adult form was only reported in the genus *Macrovestibulum* Mackin, 1930 of Macrovestibulinae subfamily under the family Pronocephalidae. This character is also for the first time being reported in the subfamily Neopronocephalinae Mehra, 1932. It could remain unreported even in the diagnostic features of not only the subfamily but also the genus. Earlier species except *N. rotundus* under the genus *Neopronocephalus* show no record of the oculate nature of the adult form.

The filamented condition of the eggs is also a characteristic feature of the family Pronocephalidae. The nature of the egg sub-family-wise is as follows: One filament at each pole in Neopronocephalinae Looss, 1899, one filament at one pole in Cotiosaccinae Yamaguti, 1958, with a tuft of filament at each pole in Charaxicephalinae Price, 1931; one filament at each pole in Choanophorinae Caballero, 1942; with a tuft of long filament at each pole in Desmogoniinae Yamaguti, 1958; one filament at each pole in Diaschistorchiinae Yamaguti, 1958; with a heavy filament at each pole in Macrovestibulinae Yamaguti, 1958; while bioperculate in Metacetabulinae Yamaguti, 1958. It is only in cases of Neopronocephalinae Mehra, 1932 and Teloporinae Stunkard, 1934, that there is no mention of the nature of the egg, though in the case of Neopronocephalinae Mehra, 1932 in its diagnostic character there is a mark of interrogation against the egg which indicates that perhaps earlier workers failed to notice the exact nature and structure of the eggs while in the case of Teloporinae Stunkard, 1934 it is simply omitted. The present study clearly shows that the egg in the case of Neopronocephalinae possesses one filament at each pole (2b), though under the genus *Neopronocephalus*, the four species described do not throw light on this aspect of the egg. Even Siddiqui (1965) also failed to notice this character of the egg. In a sectional view of the worm the nature of the egg becomes quite

obvious. Lack of information about the canoe led to the confusion in differentiating the dorsal and ventral surfaces. The canoe is present on the ventral surface, so is the genital opening and the relationship between them determines the position of the genital opening which has been shown to be variable either on the left or the right. Interestingly enough the excretory arm shows a somewhat indented nature in its lumen, which has been shown in *Macrovestibulum obtusicaecalatum* Mackin, 1930. This species resembles specimens of genus *Neopronocephalus* in being oculate in the adult and having a filament at each pole of the egg. The excretory opening is fairly wide, prominent and highly contractile. Depending on its expansion, protrusion of the excretory vesicle is possible. The nature of the excretory opening, its width, protrusion of excretory vesicle all together determine the posterior shape of the parasite. This in turn modifies the shape of the fluke from triangular to oval.

The cephalic collar is conspicuously developed and is interrupted ventrally. The head collar as also the ventral depression, is orientated in different directions (see Fig. 2A—C). Even the separated nature of the two lobes is also visible. The notch does not end exactly at the base of the oral sucker (see Fig. 2A—C). The nature and orientation of the head collar influences the shape of the worm from triangular to oval in appearance.

Length: The length of *N. triangularis*, *N. gangeticus*, *N. mehrai* and *N. rotundus* has been shown to vary from 2.9 to 4 mm. It is 2.4, 1 to 1.5 mm and 0.945 to 1.026 mm respectively. The authors in their studies found that the length varies from 1.035 to 3.070 mm. Point of bifurcation from anterior end (and size of cirrus sac): — On this character the four species have been differentiated and it is as 0.6—0.6; 0.46 –0.5; 0.30—0.38 and 0.272 mm.

Similarly the size of the cirrus sac 0.53—0.62 × 0.23 –0.30; 0.4 –0.52 × 0.25– 0.28; 0.25 —0.72 × 0.1—0.12 and 0.171—0.199 × 0.084—0.081 mm in *N. triangularis*, *N. gangeticus*, *N. mehri*, and *N. rotundus* respectively. The authors have noticed that the size of the worm causes variations in such measurements.

Number of vitelline glands: The number of vitelline glands as found in the four species is 11 to 17, 13 to 15, 11 to 13 and 13 to 15. Numbers may vary, overlapping or even rupture of the follicles may occur. The number 11 to 17 present in *N. triangularis* covers the figures mentioned in the others three species.

Eye spots: Eye spots were present in over 200 specimens studied.

Cephalic region: Its being triangular, entirely separated or triangular and entirely separated as mentioned in *N. triangularis*, *N. gangeticus* and *N. mehrai* are all due to changes in orientation due to contraction of the lateral body wall of the canoe (see Figs. 1, 2A—C). The condition of *N. rotundus* described as interrupted or uninterrupted is misleading.

Excretory pore: The controversial position of the excretory opening whether dorsal and subterminal or ventral and subterminal as figured in the species is mainly due to the confusion in the dorsal and ventral surfaces.

In view of all the facts *N. gangeticus* Mehra, 1932, *N. mehrai* Chatterji 1936 and *N. rotundus* Siddiqui, 1965 are suppressed and made synonyms to *N. triangularis* Mehra, 1932.

EMENDED DIAGNOSIS OF SUBFAMILY NEOPRONOCEPHALINAE

Neopronocephalinae: Body rather plump, truncated at posterior end. Canoe present ventrally. Head collar interrupted ventrally. Eye spots may be present. Oral sucker rather small, oesophagus comparatively long. Caeca simple, terminating at a considerable distance from posterior extremity. Testes two symmetrical, lateral or ventral to caeca in mid region of body. Cirrus pouch well developed, oblique, pre-testicular. Genital pore lateral or ventral to left caecum, a little post-bifurcal. Ovary massed together behind each testis, lateral or ventral to caeca. Uterus extending posterior to ovary and passing between two testes, eggs with polar filaments. Excretory vesicle V-shaped, arms short or long.

EMENDED DIAGNOSIS OF GENUS *NEOPRONOCEPHALUS* MEHRA, 1932

Pronocephalidae — Neopronocephalinae: Body rather plumb, truncated behind. Canoe present ventrally. Head collar interrupted ventrally. Eye spots present. Oral sucker rather small, oesophagus comparatively long; caeca simple terminating about one third of body length from posterior extremity. Testes two symmetrical lateral or ventral to caeca in mid region of body. Cirrus pouch thick, placed obliquely in pre-testicular

intercaecal field. Seminal vesicle convoluted around its base. Genital pore on the left, a little behind intestinal bifurcation. Ovary slightly to right of median line behind testes, postequatorial. Vitelline follicles massed together immediately behind testes, lateral or ventral to caeca. Uterus coiled behind ovary, passing between ovary and left testis and then between two testes; metraterm well differentiated; eggs bipolar filamented. Excretory vesicle V-shaped with short or long arm. Parasitic in intestine of turtles. Genotype: *N. triangularis* Mehra, 1932 (Synonym *N. gangeticus* Mehra, 1932, *N. mehri* Chatterji, 1936 and *N. rotundus* Siddiqui, 1965).

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