ON A NEW TREMATODE OPISTHORCHIS PEDICELLATA, SP. NOV. FROM THE INDIAN SILUROID FISHES RITA RITA AND BAGARIUS YARRELLII WITH A KEY TO THE SPECIES OF THE GENUS.

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(Plates XI, XII.)

INTRODUCTION AND HABITS.

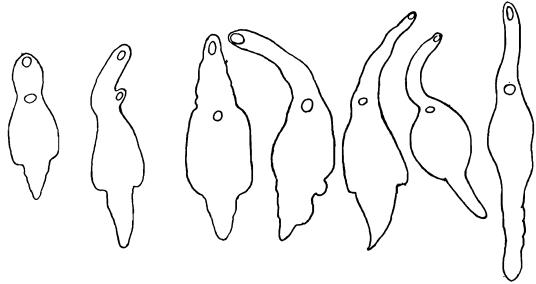
Opisthorchis pedicellata, sp. nov.

This species, apart from its remarkable anatomical features, is of great interest for it is only the second species of the genus known from fishes, and also the second to be described from India. The very brief description of the only adult piscine species, O. piscicola (Odhner 1902), that has hitherto been recorded does not appear to have attracted the attention of such well-known subsequent writers on Helminthology as Lühe (1909), Stephens (1916) and Brumpt (1922), for none of them has mentioned the occurrence of the genus Opisthorchis in fishes.

The present species was first encountered in the early winter of 1924 in the gall-bladder of three out of four Siluroid fishes of the species Rita rita (=Rita buchanani Day), obtained from the rivers Ganges and Jumna. Subsequently, about fifty more fishes were examined from time to time but only fifty per cent of them were found infected with this trematode. The worms are common in larger examples, of fifteen to twenty inches length, but in fishes of smaller size (less than 10-12 inches long) they are rare. The number of parasites in a host is very Some were found infected with a single adult fluke, while others had two, four or eight each; one example had as many as twelve (large mature forms 4, small mature 2, very small immature 6), and another as many as twenty-nine (large mature 2, small mature 7, very small immature 20). It is interesting to note that younger forms were obtained only from those fishes that were caught in the rivers in September, after the rains had stopped. Owing to their transparency, minute size and capacity of considerable elongation these immature specimens are apt to be missed in a cursory examination of the bladder

In the course of last year about half a dozen specimens of another fish, Bagarius yarrellii Sykes (=Pimelodus bagarius Ham. Buch.), of similar habits to Rita rita, were examined and every one of them carried a varying number of these parasites in the gall-bladder which, in these fishes, is closely attached to the duodenum. Apart from these two fishes I have had an opportunity of examining most of the local fishes, but so far I have not been able to obtain this fluke from any other species.

The worms are either attached to the thin membranous walls of the gall-bladder or keep moving about freely in the fluid contents by expanding and contracting their bodies in different planes. When placed in nutritive solutions or in water the movements and alterations of the body-form can be easily studied (vide text-fig. 1). At times the whole body is so much elongated that the worm looks like a nematode, but more often it is either the neck or the posterior region alone that is drawn out to a considerable extent. At one moment the body is completely straightened out but at another it is bent upon itself, being often produced in the region of the ventral sucker into a distinct protuberance or pedicel. In this respect this trematode very much resembles *Alla-canthocasmus varius* (Van Cleave 1922) which, "in spite of the fact that body musculature is not excessively developed," displays "both in the living and the preserved condition the results of unusual powers of generalised and localised contraction."



Text-fig. 1.—Outline sketches showing alterations in body-form of a living example of Opisthorchis pedicellata.

An attempt was made to keep examples alive, outside the host, at the temperature of the laboratory in various nutritive solutions which were changed every alternate day. The degree of success attained in these operations can be gathered from Table I, A and B.

TABLE I.

A. Mid-winter (December and January); temperature of Lab. 65°-70° F.

Nutritive solutions used.	Tap water.	Normal salt solution.	Dilute egg albumen.	Dilute yolk of egg.	Mix. of 20 per cent yolk and nor- mal salt sol. 1:1.	Mix. of albumen and nor- mal salt sol. 1:2.	Mix. of 0.5 per cent sugar sol. and normal salt sol. 1:1.
No. of trematodes kept.	2	4	2	2	4	3	3
No. of days trema- todes lived.	2 days	2,5 days 2,6 days	2 days	1,20 hrs. 1,2 days	2,4 days 1,5 days 1,8 days	1,1 day 2,2 days	1,1 day 2,2 days

B. Mid-summer (May and June); temperature of Lab. 110° F.

Nutritive solutions used.	Tap water.	Bile.	Normal salt sol.	Mix. of 20 per cent yolk and normal salt sol.
No. of trematodes kept	2	1	2	2
No. of days trematodes lived.	6 hrs.	8 hrs.	4 days	4 days

From the above table it is manifest that in summer none could survive beyond four days, whereas in winter some remained alive for six to eight days. This species is therefore more tenacious of life than Tremiorchis ranarum (Mehra and Negi 1926); but while the latter lived longest in a mixture of white of egg and salt solution, my species thrived best in a weak solution of yolk in normal salt solution. While agreeing with the above named authors that these parasites can live outside the body of the host on fluid food for a number of days, attempts are being made to discover some peptonised or other media in which they may be cultured under laboratory conditions. It may thus eventually prove possible to rear immature forms into adult flukes outside the body of the host if suitable conditions are provided.

EXTERNAL FEATURES.

The trematodes are small worms measuring 5-9 mm. in a state of ordinary contraction. Their proper dimensions in life are difficult to obtain owing to the constant contraction and expansion of body both in length as well as breadth, hence the measurements here given are generally from preserved specimens. An adult individual of average size, which measured 7 mm. in length after fixation, when alive showed the following dimensions:—

At maximum elongation—length of body 12.7 mm., greatest breadth 0.83 mm.

At maximum contraction—length of body 6.3 mm., greatest breadth 1.66 mm.

This would show that when fully elongated it is twice as long and half as broad as when it is fully contracted.

TABLE II.

Dimensions in mm. of adult specimens of O. pedicellata fixed without pressure.

Length.	Breadth in region of ventral sucker.	Greatest breadth in front of ovary.	Distance of ventral sucker from anterior end.	Diameter of oral sucker.	Diameter of ventral sucker.	Diameter of pharynx.
4.5	0.32	1.10	1.04	0.13	0.17	0.06
5-6	0.59	0.93	1.84	0.17	0.25	0.09
6.0	0.64	1.20	1.80	0.24	0.33	0.15
6.0	0.62	1.12	1.90	0.23	0.32	0.14
6.5	0.71	1.25	2.14	0.25	0.33	0.16

The trematodes were best killed in the expanded state in hot corrosive sublimate or in warm methylated spirit actively shaken for a few minutes. As soon as they appeared to be motionless the worms were removed to the desired fixative. Measurements given in the above table are of worms preserved in this way, and are taken from vertical and horizontal sections, and whole mounts. Specimens, which after flattening between two glass slides were fixed in Bouin's fluid or hot alcohol, were

mounted entire. Various stains were used, but the best results were obtained by alum carmine and borax carmine. Table III shows the dimensions of full-grown worms fixed and mounted after flattening, and table IV of small immature flukes treated in the same way.

TABLE III.

Dimensions in mm. of adult specimens of O. pedicellata fixed after flattening.

Length.	Breadth in region of ventral sucker.	Greatest breadth in region of ovary.	Distance of ventral sucker from anterior end.	Diameter of oral sucker.	Diameter of ventral sucker.	Diameter of pharynx.
7.0	0.94	1.25	2.16	0.24	0.29	0.13
9.0	1.20	1.40	3⋅10	0.32	0.39	0.16
9.5	1.12	1.50	2.60	0.26	0.33	0.15
10.0	1.28	1.50	3⋅50	0.30	0.40	0.15
10.5	0.96	1.60	3.40	0.35	0.45	0.18
12.8	1.45	2.0	3.30	0.38	0.47	0.19

TABLE IV.

Dimensions in mm. of immature specimens of O. pedicellata from balsam mounts.

Length.	Breadth at anterior end.	Breadth in region of ventral sucker.	Greatest breadth in region of ovary.	Distance of ventral sucker from anterior end.	Diameter of oral sucker.	Diameter of ventral sucker.
0.640	0.112	0.140	0.180	0.38	0.096	0.064
0:960	0.114	0.176	0.208	0.61	0.120	0.096
1.008	0.114	0.224	0.358	0.65	0.128	0.112
1:010	0.160	0.190	0.240	0.59	0.128	0.112
2.000	0.176	0.240	0-360	1.09	0.128	0.176

Opisthorchis pedicellata is therefore a much longer form than O. piscicola, O. obsequeus and O. caninus; it is also somewhat larger than O. interruptus and O. lancea, but is decidedly shorter than O. viverrini, O. longissimus, O. noverca, O. simulans, O. entzi and O. felineus. In Table V, I have given the characteristics of the principal species of the genus.

In life the immature forms are white in colour and quite transparent; mature ones are, however, less transparent, the middle portions of their bodies appearing yellowish-brown owing to the innumerable eggs contained in the uterine coils. The body of the present species, unlike that of most of the other species of the genus, is more or less distinctly divisible both in the adult and young conditions into three regions, a narrow cylindrically-built neck extending up to the ventral sucker, a median broader and dorso-ventrally flattened body proper, which

ends with its widest part in the region of the ovary, and a small and abruptly tapering hinder part, in which are lodged the two testes. The surface of the body, in mature as well as in young forms, is more or less uniformly covered with minute closely-set spines arranged in regular rows encircling the body. The spines are more numerous round the anterior sucker and the neck, and in fully-grown individuals the posterior end bears comparatively few spines excepting in the immediate vicinity of the genital pore. The spines are minute structures not more than 0.025 mm. long but can easily be seen under the low power of a microscope.

The oral and ventral suckers are nearly circular in outline, being slightly elongated in a transverse or an oblique direction. The ventral sucker in mature flukes is decidedly larger than the oral, having a mean diameter in uncompressed specimens of 0.28 mm. (Table II) and in flattened mounts of 0.39 mm. (Table III). The oral sucker is 0.204 mm. in diameter in uncompressed specimens and 0.308 mm. in flattened specimens of average size. The oral sucker, therefore, bears to the ventral a ratio of 2:3 in normal specimens, and of 3:4 in flattened mounts. But in immature flukes the two suckers are either of the same size (vide Table IV, last example), or the condition is reversed, the ventral sucker being decidedly smaller than the oral (vide Table IV. first four examples). In fact the younger the specimen the greater is the relative size of the oral sucker. In addition to this difference in the young and the adult stages there also exists a considerable difference in the relative position of the ventral sucker. In mature individuals it is situated at about the end of the anterior third of the body, but in immature ones it lies clearly behind the middle of the body as can be judged from the distance of the acetabulum from the anterior end given in tables II-IV and Pl. XI, figs. 1 and 2. This is due to the more rapid growth of the acetabulum and post-acetabular regions during the period of maturity, as has also been observed in Margeana californiensis (Cort 1919-20) and in Mesocoelium sociale (Lühe) [Sewell 1920]. The oral sucker lies at the anterior end of the body directed towards the ventral surface and has well-developed muscular walls.

The genital opening lies in the median line immediately in front of the ventral sucker. There is a short but rather wide genital atrium into which open both male and female ducts. The aperture of Laurer's canal cannot be made out in entire mounts owing to its minute size, but it is readily seen in sections, and particularly well in vertical ones, as a narrow pore lined by cuticle situated in the middle line on the dorsal surface of the body (vide Pl. XII, fig. 8).

The region of the ventral sucker together with the genital opening is capable of being protruded ventrally in a short process or pedicel, as is also the case in O. caninus Barker (=Paropisthorchis Stephens 1912) and Psilochasmus oxyurus (Lühe 1909) and certain other trematodes. A living distome of this species moving about in the nutritive solution often exhibits this pedicel projecting from the ventral region of the body, and if fixed in this condition it will show a curved pedicelled appearance. The creation, therefore, of a separate genus "Paropisthorchis" (Stephens 1912, as mentioned in a Animal Para-

sites of Man" 1916, p. 255) for the form described by Barker as O. caninus does not appear to be justifiable¹.

INTERNAL ANATOMY.

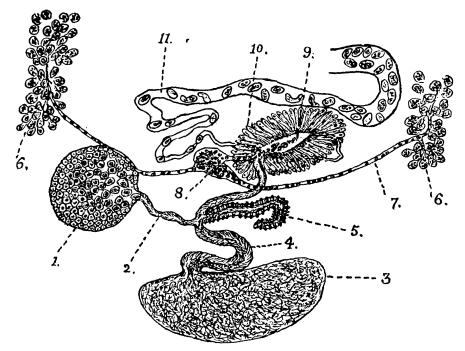
Alimentary System.—The mouth is subterminal and lies as usual at the bottom of the anterior sucker. It opens into a distinct thinwalled prepharynx which is followed by a thick-walled muscular, spherical pharynx having an average diameter of 0.13 to 0.19 mm. The pharynx leads into a narrow tubular oesophagus whose length varies from one to three times the diameter of the pharynx according to the degree of contraction of the part, but usually it is about one and a half times as long as the pharynx. The oesophagus is continued into the intestine which immediately bifurcates into two wider lateral caeca. The intestinal caeca are nearly straight tubes, with very faintly crenated walls, that run parallel to one another and terminate near the hinder end of the body in the vicinity of the terminal portion of the posterior testis; the left caecum is slightly shorter than the right one.

Reproductive System.—The principal genital organs all lie in the posterior fifth of the body, the two testes occupying the hindermost region. The posterior testis reaches to within 0.1 to 0.16 mm. of the hinder extremity: it is the larger of the two and is divided into 3 to 5 lobes occurring one behind the other. Its dimensions in mature specimens vary from 0.4 mm. to 0.56 mm. in length and from 0.192 mm. to 0.354 mm. in breadth. The anterior testis is roughly triangular in shape in surface view, and has its anterior arm slightly depressed backwards owing to the pressure of the large receptaculum seminis in front. It is also lobulated into three to five lobes and is usually displaced towards the left side, so that the right anterior margin of the posterior testis is in many cases flush with the posterior surface of the anterior testis and sometimes extends forwards even beyond it. In immature specimens both the testes are more or less circular in outline with clear lobes and are separated from one another by the thickness of the excretory bladder. This peculiar position of the testes, placed somewhat obliquely one behind the other in close proximity and partly alongside, is a characteristic feature of the species (vide Pl. XI, fig. I). The number of testicular lobes is not invariable as has also been observed in O. obsequeus (Nicoll 1914). The vasa efferentia arise as delicate tubes from the antero-lateral surface of the testes. They run forwards independently of one another to about the commencement of the posterior third of the body and then, approaching one another towards the middle line, come to lie side by side. After running together for some distance they unite to form a very small, inconspicuous (20 μ to 30 μ long) vas deferens, opening into the seminal vesicle. The vasa efferentia can be traced in transverse sections as minute ducts situated internally to the dorsal body-wall of the animal, but they are difficult to see in entire

¹ The author is collecting more specimens of this genus as he is of opinion that this pedicel may be present in some other forms also though, hitherto, not detected, as in many cases the specimens were not examined alive. Even if it were not so, he thinks that this characteristic is not of sufficient importance to warrant the creation of a separate genus; it is at best a sub-generic character. It is intended to discuss this further in another paper in which the author proposes to revise this and some other allied genera.

mounts unless very carefully differentiated. The seminal vesicle is a long coiled thin-walled tube full of sperms, occupying the space between the coils of the uterus. It commences about the middle of the body, runs straight for a little distance, then becomes twisted into several coils, and finally takes a more or less sinuous course along the right side of the ventral sucker, beyond which it turns inwards and slightly backwards to open into the genital atrium on a short muscular papilla, which probably functions as the copulatory organ (vide Pl. XII, fig. 4). A cirrus sac is absent—a feature common to all species of the genus—and no prostatic gland or cells appear to be present in this species.

The ovary is rounded or pear-shaped in outline but in some specimens its posterior surface is somewhat depressed in the vicinity of the seminal receptacle. It is situated invariably to the right of the middle line and nearer the ventral than the dorsal surface at a distance of about one-fifth the body length from the hinder extremity. The oviduct arises from the mid-dorsal surface of the ovary and running a short distance inwards joins the duct of the seminal receptacle, into which also opens Laurer's canal from the opposite side. The receptaculum seminis is a prominent oval structure (with its anterior margin concave in some preparations), placed somewhat obliquely between the ovary and the anterior testis. The duct of the receptaculum is a thin-walled wide tube (0·18 to 0·25 mm. broad). It originates from its anterior margin and passes a little dorsalwards towards the ovary, then turns to the left and soon takes another turn to the right forming a depressed



TEXT-FIG. 2.—Diagrammatic view of female genitalia of Opisthorchis pedicellata.

1. Ovary; 2. Oviduot; 3. Seminal receptacle; 4. Duct of receptacle; 5. Laurer's canal; 6. Vitelline gland; 7. Vitelline duct; 8. Yolk-sac; 9. Ootype; 10. Shell-gland; 11. Uterus.

S-shaped loop before it meets the oviduct and Laurer's canal (vide text-fig. 2 and Pl. XII, fig. 7). The epithelial lining of the receptaculum does not appear to be ciliated, but that of the duct is strongly so, and the cilia are directed mostly towards the receptaculum. From its

junction with the duct of the receptaculum seminis Laurer's canal takes a short course to the left; it then bends towards the dorsal surface where it opens by a minute pore lined by cuticle which is continuous with that of the outer surface. The whole of Laurer's canal, with the exception of the cuticular external aperture, is lined with outwardly directed cilia which are only about half as large as those of the duct of the seminal receptacle. The canal is only 0.012 to 0.016 mm. thick, and the lumen is very narrow owing to the comparatively thick wall provided with powerful circular muscle-fibres.

The oviduct, beyond its junction with Laurer's canal and the duct of the seminal receptacle, continues its course towards the left; it then turns anteriorly and after crossing the left vitelline duct is joined by a short duct from the vitelline reservoir lying to its right. It soon enters the ootype on its dorsal aspect. The ootype is slightly dilated and is surrounded by prominent elongated shell-glands of the usual type, arranged radially round it. On the side opposite to that from which it receives the oviduct the ootype opens into a narrow duct which takes a backward turn before it abruptly broadens out as a transversly coiled uterus full of eggs, filling up the space between the ovary and the ventral In full-grown individuals the uterine coils form eighteen to twenty-four convolutions and are characteristically opisthorchid in character, touching but rarely overlapping the gut diverticula and never extending anteriorly beyond the ventral sucker. Its outgoing portion, the vagina, runs obliquely forwards along the left side of the ventral sucker as a narrower wavy tube with eggs arranged in single file, and passing forwards, beyond the genital aperture and the male duct, it turns backwards towards the middle line to open into the short but spacious genital atrium lying just in front of the sucker. The eggs are oval in outline and measure 30 μ by 16 μ in size. Their colour is at first white, but in the anterior half of the uterus they are distinctly reddish-brown and give the characteristic tinge to that part of the body in the living worm.

The yolk-glands occupy the lateral areas, beginning in the region of the ventral sucker and terminating at about the level of the ovary. They never extend behind the ovary and are confined to the dorsal and ventral surfaces of the body, lying partly to the outer side of the intestinal caeca and partly overlapping them. Both vitellaria consist of eight groups of follicles the hinder ones of which usually run together forming more or less continuous bands. The transverse vitelline duct of each side leaves the gland in front of the last group of acini, and runs inwards and backwards to open independently into a small but prominent vitelline reservoir or yolk-sac referred to above (vide Pl. XII, fig. 6). Text-figure 2 gives a dagrammatic sketch of the female genitalia.

Excretory system.—A detailed study was made of the excretory system because no complete description of it is given in any species of this genus. It was studied mainly from young living specimens that had not yet developed the uterine coils and the eggs. The conclusions thus arrived at were verified independently by careful examination of permanent mounts of both vertical and transverse sections. The excretory bladder is clearly visible in younger specimens as an elongated

S-shaped structure running between the two testes and bifurcating behind the receptaculum seminis into a longer left and a shorter right horn or cornua. It continues backwards into a short narrow duct which opens to the outside by a terminal excretory pore surrounded by somewhat larger spines than those present on the adjacent parts of the body surface. Each horn of the bladder receives along its hinder outer margin a common collecting tube which runs obliquely outwards and after crossing the intestinal caecum continues forwards as a convoluted duct parallel to it. In the region of the prepharynx the collecting tube bends backwards and receives its first collecting tubule or branch (1) which appears to be connected by transverse connections with its fellow of the opposite side, and gives off one main branch towards the anterior side and another posteriorly. The main duct then continues backwards parallel to its ascending arm as a tube of gradually decreasing calibre to the posterior extremity of the worm, and receives in its course five more accessory collecting tubules (2-6) as indicated in Plate XI, figure 3. From each of these six accessory collecting tubules are given off two capillaries of the first order, which again divide into secondary and tertiary capillaries and end ultimately in flame cells. The latter were observed distinctly by means of an oil immersion microscope (Leitz. Eye-piece III, Objective 1/12 N.) in a permanent preparation made from a specimen that had been left in water after fixation for a very long time and from portions of which the cuticle could be easily removed by means of a fine brush. A diagram of the arrangement of the finer capillaries originating from flame cells is given in Plate XII, figure 5 which is drawn to scale as indicated.

The excretory system therefore consists of a Y-shaped bladder having unequal arms with curved stem and a pair of laterally placed collecting tubes, each consisting of an ascending portion receiving six accessory collecting tubules and a descending portion which ultimately opens into the bladder. The system therefore bears great resemblance to that of *Distomum tereticolle* (Rudolphi) described and figured by A. Looss (1894).

Systematic Position and Diagnostic Characters.

From the foregoing description it will be seen that in the topographic relationships of its organs this fluke conforms closely to the diagnosis of the genus Opisthorchis Blanchard 1895; but in possessing a retractile pedicel which carries on it the ventral sucker and probably also the genital pore it resembles the genus Paropisthorchis. As has already been pointed out, this character in itself does not appear to me to be sufficient ground for the creation of a separate genus, particularly as Stephens (1916, p. 255) admits that in all other structural features his genus agrees with Opisthorchis. Apart from the brief description given of this genus by Stephens in the book referred to above, I have not been able to find any reference to his paper published in 1912, in which he creates this genus; nor has the genus been taken cognisance of by Skrjabin (1913) in his important paper in which he gives diagnoses of the genera of the family Opisthorchidae. As my species is structurally much more opisthorchid than O. (=Paropisthorchis) caninus (Barker) I propose

for the present, to place it in the more stable genus Opisthorchis under the name O. pedicellata.

In Table V I have compared the diagnostic features of this trematode with other species of Opisthorchis including the closely allied form Opisthorchis (=Amphimerus) noverca and Opisthorchis (=Paropisthorchis) caninus. It can be seen from this table that the new species, in the possession of cuticular spines, resembles O. viverrini, O. piscicola, O. (=Amphimerus) noverca and O. (=Paropisthorchis) caninus and differs from the rest. It approaches the last named species in having in common with it the short pedicel—a structure that may be present in some other forms also. But apart from this and other features it can be readily differentiated from:—

- (i) O. viverrini by the ratio in the size of the two suckers, and by its simple undivided ovary.
- (ii) O. piscicola, the other piscine species, by the relatively larger size of its body, of the two suckers and of the vitellaria; also by its undivided ovary and the more posterior position of the ventral sucker in the body.
- (iii) O. noverca by its smaller size, by the relative size of the two suckers, the more posterior position of the ovary and the ventral sucker in the body, and above all in having no post-ovarial portion of the vitellaria.
- (iv) O. caninus by the reverse ratio in the size of the two suckers, by the presence of a prepharynx and a much longer oesophagus, and the more posterior position of the genital opening and the ventral sucker.

KEY TO THE SPECIES INCLUDED IN TABLE V.

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A. Body armed with spines
                                                                        В.
    Body unarmed with spines
                                                                        F.
B. Ovary simple, not divided into lobes
                                                                        O. pedicellata, n. sp. (14).
    Ovary not simple, divided into lobes
                                                                  .. <u>C</u>.
C. Oral sucker equal to or less than the ventral
                                                                       D.
                                                                   • •
    Oral sucker larger than the ventral
                                                                       E.
D. Oral sucker equal in size to ventral
                                                                   .. O. viverrini (4).
    Oral sucker smaller in size than ventral
                                                                   .. O. piscicola (9).
E. Vitellaria extend far behind ovary
                                                                       O. noverca (10).
                                                      ..
                                                                   • •
Vitellaria end in region of ovary .. F. Oral sucker smaller than ventral ..
                                                                        O. caninus (12).
    Oral sucker equal to (or nearly so) or larger in size than
      ventral ...
                                                                       Μ.
                            • •
G. Testes lobed
    Testes not at all lobed
                                                                        O. pseudofelineus (7).
H. Testes lobed deeply, almost dendritic
                                                                        O. obsequeus (13).
Testes lobed but not deeply .. .. .. .. K. Vitellaria disproportionate in size, one half much longer
      than the other
                                                                        O. entzi (11).
Vitellaria proportionate in size

L. Vitellaria long, extend far behind ovary
                                                                       O. lancea (1).
    Vitellaria short, do not extend behind ovary
                                                                       O. longissimus (2).
M. Oral sucker equal (or nearly so) to ventral ...
                                                                       N.
    Oral sucker larger than ventral
                                                                       0.
N. Ovary simple or but slightly lobed; length of eggs 30 \mu
                                                                        O. felineus (3).
Ovary usually three lobed; length of eggs 20 \mu . O. Ovary multilobed; vitellaria short, never extend behind
                                                                       O. geminus (6).
                                                                       O. simulans (5).
   Ovary not lobed, vitellaria long, extending far behind ovary O. interruptus (8).
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The type-slide and specimen are deposited in the collection of the Zoological Survey of India, at the Indian Museum, Calcutta. W 1376/1.

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TABLE V.

Showing Characteristics of the Species of the Genus Opisthorchis and Allied Forms.

Species.	Host.	Size and shape of body.
1. O. lancea Diesing 1856 (Weski 1900).	"Delphinus tacuschi" (sta- notucuxi, Gray) (gall- bladder).	5.5—12.5 by 1.0—2.8: neck and body lancet-shaped, margin behind ventral sucker undulating.
2. O. longissimus Linstow 1883.	Botaurus (Ardea) stellaris (liver).	20.0 by 1.0: very long, cylindrical, posteriorly rounded.
3. O. felineus Rivolta 1885	man, cat, dog, fox, glutton and seal (liver, gall- bladder, bile and pancrea- tic duct).	8-11 by 1.5-2.0: flat, with a conical neck (varies according to contraction).
4. O. viverrini Poirier 1886	man and civet cats (gall- bladder, bile duct and intestine).	closely resembles O. felineus.
5. O. simulans Looss 1896 (1910).	Pernis apivorous and Anas penelope (gall-bladder); A. boschas and Fulix cris- tata, young ones (liver).	7.0 by 1.15 (15—16 by 1—1.5); very long, pointed posteriorly.
6. O. geminus Looss 1896 (1910).	Milvus parasiticus (liver and bile duct).	7-8 by 1.3; thin, long, rounded posteriorly.
7. O. pseudofelineus Ward 1900.	cat (gall-bladder and liver)	10—13 by 1.5: like O. felineus but somewhat longer and narrower.
8. O. interruptus Braun 1902.	Alcedo viridirufa (intestine)	7—8 by 0.5—0.7; elongated, band-shaped, posterior end broad; margin behind ventral sucker undulating.
9. O. piscicola Odhner 1902.	Gymnarchus niloticus (gall- bladder).	3—4 by 0.5: strongly tapering in front.
10. O. (=Amphimerus) noverca, Braun 1903 (Barker 1911).	man, dog and American fox (liver and bile duct).	9—12 by 2.5: lanceolate, more elongated in front.
11. O. entzi Ratz 1903	Ardea purpurea (gall-blad- der).	like that of O. longissimus but more pointed, an- teriorly lancet-shaped.
12. O. (=Paropisthorchis) caninus Barker 1911. Stephens 1912.	dog (liver)	2·75—5·75 by 1·5: oval ventro-dorsally concavo-convex.
13. O. obsequeus Nicoll 1914.	Hieracidea berigora and H. orientalis (liver).	2.6—5.1 by 0.8—1.0: flattened, with crenated edges; a knob-like tip frequently projecting from posterior
14. O. pedicellata, sp. nov. 1927.	Rita rita and Bagarius yar- rellii—India (gall-blad- der).	end. 5—9 by 1·0—1·5 (average 7 by 1·25) (varies according to contraction); divisible into three regions.

Showing Characteristics of the Species of the Genus Opisthorchis and Allied Forms—contd.

Cuticle.	Size and ratio of suckers (oral: ventral).	Nature and position of ven- tral sucker.
	(orar: ventual).	orar sucker.
l. unarmed	0.47 by 0.51: 0.5 by 1.2 1: 1.5	from less than one and a half times to twice as big as oral; at \frac{1}{3}rd of body length from anterior end.
2. unarmed	0·28: 0·34 1: 1·2	slightly larger than oral; in anterior the of body length.
3. unarmed	0·25: 0·25 1: 1	of same size as oral; at th to the of body length from anterior end.
4. armed; spines small, pointed	1:1	of same size as oral; as in O. felineus.
5. unarmed	0·5: 0·2 1: 0·4	less than half the size of oral; at \frac{1}{3}rd to \frac{1}{5}th of body length from anterior end.
6. unarmed	0·17: 0·17 1: 1	nearly as big as oral; just behind 4th of body length from anterior end.
7. unarmed	0·2: 0·33 (approximate, from fig.) 1: 1·65	little more than one and a half times as big as oral; in anterior 1th of body length.
8. unarmed	0·25 by 0·26; 0·135 1: 0·52	nearly half the size of oral, oblique; at ith to ith of body length from anterior end.
9. armed, spines minute	0·19 by 0·22: 0·026 by 0·3 1: 1·4	nearly one and a half times as big as oral; transversely oval; at end of the fith of body length from anterior end.
10. armed, spines, small	0.45 by 0.75: 0.35 (approximate, from fig.) 1: 0.4—0.7	nearly half the size of oral; at th of body length from anterior end.
11. **	**	**
12. armed, uniformly spiny except on pedicel	0·28: 0·176 1: 0 ·63	more than half as big as oral; at \$\frac{1}{8}\$th of body length from anterior end; on a short pedicel.
13. unarmed	0·16 by 0·20: 0·26 1: 1·3—1·6	about one and a half times as big as oral; at \frac{1}{2}rd of body length from anterior end.
11. armed, more or less uniformly spiny, spines minute (0.025)	0·204 by 0·308 : 0·28 by 0·39 1 : 1·3—1·5	in adult—about one and a half times as big as oral; at end of frd of body length from anterior end. in young—as big as or smaller than oral; behind middle of body on a short pedicel.

Showing Characteristics of the Species of the Genus Opisthorchis and Allied Forms—contd.

Prepharynx.	Pharynx.	Oesophagus.	Intestinal caeca.
1. absent	muscular, about half the size of oral sucker	short, slightly small- er than pharynx	rather wide, reach almost to posterior end.
2. not noticed, (probably absent)	slightly smaller than oral sucker	almost as big as oral sucker	do not quite reach posterior end.
3. absent	close behind oral sucker and about frd its size.	hardly any longer than pharynx	reach almost to pos- terior end.
4	••••	as in O. felineus	extend to posterior end.
5. absent	close behind oral sucker and about th its size	as long as pharynx	reach almost to pos- terior end.
6. absent	feebly developed, minute (dia. 0·1)	more than twice as long as pharynx, length 0.25	reach almost to pos- terior end, ends turned inwards.
7. absent	not prominent and smaller than oral sucker	small, nearly as long as pharynx	extend to posterior end.
8. probably absent or indistinct	close behind oral sucker and about half its s ize	as long as pharynx	wider than oesopha- gus; one slightly longer than the other, extending to posterior end.
9. not observed	less than half the size of oral sucker (dia. 0.08)	not described	*
10. probably absent	close behind oral sucker and about half its size	very short, half as long as pharynx	do not reach posterior end.
11. **	**	**	**
12. absent	close behind oral sucker and smaller than it (size 0.224 by 0.184)	minute, 0.04 in length	do not reach posterior end.
13. absent	close behind oral sucker and smaller than it (dia. 0·12)	almost one and a half times as long as pharynx	almost reach posterior end; inner walls crenated, ends turn- ed inwards.
14. distinct, thin- walled	nearly half the size of oral sucker (dia. 0·13—0·19)	prominent, one to three, usually one and a half times as long as phar- ynx	almost reach posterior end, walls very feebly crenated.

-	Excretory system.	Testes.
1.	bladder very long; main trunk S-shaped between the two testes but behind them straight, its narrow horns run forwards between uterine loops and gut forks	roundish, anterior 4 posterior 5 lobed, in hinder third of body, oblique; post testicular space large.
2.	not observed	roundish, anterior 5 posterior 6 lobed, in hinder fourth of body in median line.
3.	bladder elongated S-shaped, forked in front of anterior testis	roundish, anterior 4 posterior 5 lobed, in hinder fourth of body, oblique.
4.	••••	in hinder fourth of body.
5.	bladder forked just behind ovary, elon- gated	elongated, spheroidal (dia. 0.6), or partly rounded, partly irregularly shaped; in hinder fourth of body in median line.
6.	bladder elongated S-shaped; forks short, at level of seminal receptacle	roundish, anterior 4 posterior 5 lobed, in hinder fourth of body, oblique.
7.	bladder very long and curved.	ovoid, obliquely or transversely elon- gated; in hinder fourth of body in median line.
8.	bladder very long, S-shaped between the testes but hinder portion straight; horns not indicated	anterior 4 posterior little lobed, roundish; post testicular space 0.7—0.8.
9.	*	slightly lobed, lie in median line close behind ovary.
10.	bladder narrow, elongated, Y-shaped, bifurcates a short distance behind middle of ovary	anterior roundish, posterior distinctly lobed; at commencement of last third of body; post testicular space about sthe body longth.
11.	**	**
12.	bladder narrow, S-shaped, confined to last third of body; horns very short	usually ovoid, though may be regularly lobed, oblique; in last third of body.
13.	bladder sinuous; horns short and lie just behind ovary	deeply lobed, almost dendritic; anterior 5 posterior 4 lobed.
14.	bladder with two unequal horns and S-shaped stem whose hinder part is nearly straight; principal collecting tube on each side consists of an ascending and a descending portion	shape variable; in young, circular in outline; in adult, ovoid or elongated; lobes 3-5 but never deep; post testicular space 0·1—0·16.

Vesicula seminalis.	Ov ary.
1. very short, formed a little distance be- hind ventral sucker	simple, small, mostly biscuit-shaped; at commencement of last third of body.
2. long; wavy in front, sinuous behind	many lobed, median; at end of third fourth of body.
3. not described, in figure narrow and indistinct	simple or slightly lobed, median, transverse; at end of second third of body.
4	many lobed; at commencement of hinder fourth of body.
5. rather short, rarely sinuous; maximum dia. 0.1	irregular in outline, faintly lobed; behind commencement of posterior third of body.
6. voluminous, forming many transverse coils; maximum dia. 0.1	usually three lobed, almost median; near end of third fourth of body.
6. long, convoluted; indistinct in figure	simple, pear-shaped, median; within posterior third of body.
8. not observed	simple, elongated oval, transverse dia- meter equal to that of testes; at commencement of last third of body.
9.	more or less single-notched.
10. not observed	slightly lobed, median; just behind middle of body between horns of excretory bladder.
11. **	**
12. coils close and displace uterus to left	irregular in outline, six to eight lobed; in front of last third of body.
13. short, highly convoluted; extends beyond ventral sucker for about 0.3	usually irregular with three deep lobes; at \frac{1}{3}rd of body length from hinder end.
14. long thin-walled, coiled; anteriorly extends a little beyond genital pore	rounded or pear-shaped, simple, to right side of middle line; at 1th to 1th of body length from hinder end.

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_	Vitellaria.	Shell-gland.
1.	outside gut forks; from near the termination of gut forks to a little distance behind ventral sucker; ant-ovarial portion as long as post-ovarial	not visible in permanent preparations.
2.	overlapping gut forks; in third fourth of body, from in front of shell gland to half way between ovary and ven- tral sucker	prominent, median, extends in front of ovary.
3.	outside gut forks; in central third of body, from level of ovary to some distance behind ventral sucker	diffuse ; at same level as ovary.
4.	as in O. felineus	as in O. felineus.
5.	usually outside gut forks, close together; in third fourth of body, from level of ovary to midway between it and ventral sucker	not prominent, near ovary.
6.	outside gut forks; from hinder margin of ovary to some distance behind ventral sucker	not prominent, near ovary.
7.	usually outside gut forks; from front margin of hinder testis to end of first third of body; ant-ovarial portion much longer than post-ovarial	distinct, extends in front of ovary.
8.	overlapping gut forks; from posterior testis to the fith of body length behind ventral sucker: ant-ovarial portion a little longer than post-ovarial	not observed.
9.	extend from hinder border of ovary to commencement of middle third of body	*
10.	well outside gut forks; from posterior testis to behind ventral sucker: ant- ovarial portion equal to post-ovarial	
11,	disproportionately developed, one half always longer than the other	**
12.	entirely outside gut forks; acini distinct from one another; from ovary or front testis to region of ventral sucker	extensive, diffuse, in region of ovary.
13.	entirely outside gut forks; from middle or front border of anterior testis to region of ventral sucker	not observed.
14.	overlapping gut forks; from ovary to region of ventral sucker; groups of acini often merge into one another	large, rather compact, to left of ovary.

	Receptaculum seminis.	Laurer's canal.	Uterus.	Genital pore.	Eggs.
1. 1	petween horns of excretory blad- der; small spin- dle shaped, obli-	not observed	loops narrow, not filling the middle field	as usual in gonus	0·029—0·033 by 0·012— 0·014.
2. ł	que oetween ovary and anterior testis; large, ovoid, median and elongated	not observed	extensively convo- luted; rarely over- lapping gut forks	as usual in genus	0.026 by 0.015.
3. c	in the long axis close behind ovary; large, pear shaped or retort shaped, well in advance of front testis	present	convolutions occupy central field, rare- ly touching gut forks	as usual in genus	0.030 by 0.011 operculated.
4. a	is in O. felineus	••••	as in O. felineus	••	••
5. k	oehind ovary; small, oblique	rather short and broad, external opening in front of anterior testis, in mid-dorsal line	anteriorly loops narrow and wavy, posteriorly they fill up space bet- ween gut forks	on anterior border of ventral suc- ker	0.028 by 0.018 operculated.
6. 0	elose behind ovary and equal to it in size; curved and saccular	rather long and narrow; external opening in region of seminal recep- tacle	extensively convo- luted, filling up space between gut forks	as usual in genus	0.02 by 0.01 operculated.
7. (close to one side of ovary; large, oblique, fills up space between ovary and an- terior testis	long, narrow, exter- nal opening in ad- vance of anterior testis	extensively convo- luted throughout its length	male and female apertures side (fig. Stiles)?	
8. (close behind ovary and smaller than it; roundish or	not observed	convolutions hardly cross central field, at times oblique.	as usual in genus	0.023 by 0.01 darkish.
9.	pear shaped	*	*	*	0·021 by 0·011.
10.	••		convolutions barely spread beyond central field	as usual in genus	0.034 by 0.019 0.021.
11.	**	**	**	**	**
12.	dorso-lateral to posterior ovar- ian lobe; glo- bular	from end of recep- tacle takes a single curve medi- ally backwards	convolutions often overlap gut forks	on apex of a pedicel; surrounded with scales	• c
13.	alongside and slightly behind ovary; pear shaped, of mo- derate size	not observed	convolutions 14 to 16, occasionally touch gut forks	as usual in genus	0.028 by 0.0155.
14.	between ovary and anterior testis; large, oval and oblique	somewhat S-shaped, narrow, external opening minute and cuticular	convolutions 18-24, only at times over- lap gut forks	on a protru- sible pedi- cel; along with ventral sucker	0.030 by 0.016 brown.

[•] In these "respects it agrees with the diagnosis given by Looss of the genus and sub-family."
•• Complete description of this species published in Allatt. Kozl, II, Budapest, is not available to the Author.