Pearsonellum corventum, gen. et sp. nov. (Digenea: Sanguinicolidae), in Serranid Fishes from the Capricornia Section of the Great Barrier Reef

Vlaams Instituut voor de Zan

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#### Abstract

Pearsonellum corventum, gen. et sp. nov., is described from the heart of Plectropomus leopardus (type host), Epinephelus quoyanus, E. merra, and E. ongus from the vicinity of Heron I. in the Great Barrier Reef. It has body spines in ventrolateral transverse rows, an X-shaped intestine with long posterior caeca, a single testis, a unique auxillary external seminal vesicle, a well-developed cirrus sac, a post-testicular ovary, a partly preovarian uterus, and a well-developed metraterm. Those characteristics support a closer relationship among many piscine blood fluke genera than indicated by the seven subfamilies recognised in Yamaguti's 'Synopsis of Digenetic Trematodes of Vertebrates'.

## Introduction

During examination of fishes for parasites from and near Heron I. in the Capricornia Section of the Great Barrier Reef, we encountered a new species of blood fluke in serranid fishes. Examinations were made at the University of Queensland Heron Island Research Station for the Parasitology Workshop preceding the Sixth International Congress of Parasitology in August 1986, and for individual collections in January and April 1984. Of the 32 species of serranids listed from the Capricornia Section by Russell (1983), all but six are reported as rare or uncommon. We examined the hearts of five species, and all but Epinephelus fasciatus (Forsskål, 1775) had the parasite which we describe below. The response to the parasite by one host, Plectropomus leopardus (Lacepède, 1802), will be treated in a separate report by Overstreet and Thulin (1989).

### Materials and Methods

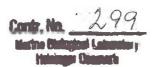
Individual blood flukes were removed from the heart; killed in hot saline; fixed in AFA (ethyl alcohol-formalin-acetic acid), Bouin's solution, or 5% formalin; and stained in Van Cleave's haematoxylin or carmalum. Other individual specimens and specimens within heart tissue fixed in buffered 5% formalin or in Bouin's solution were sectioned in paraffin and stained with Heidenain's azan or Harris' baematoxylin and eosin Y. Some fixed individuals were subsequently processed for scanning electron microscopy as described by Køie (1982).

Genus Pearsonellum, gen. nov.

Type species. Pearsonellum corventum, sp. nov., by monotypy.

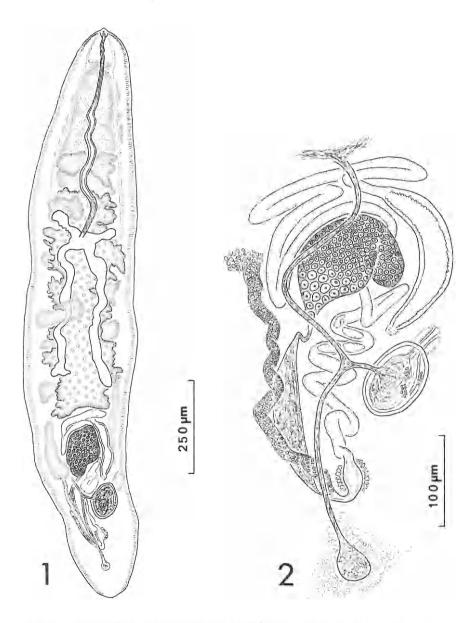
Etymology. The Latin diminutive in the neuter gender of Pearson, after John C. Pearson of the University of Queensland, who is being honoured for his influence on digenean helminthology.

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# Diagnosis

Sanguinicolidae von Graff, 1907. Body spatulate, spined in ventrolateral transverse rows, without relatively large rosethorn-shaped spines posteriorly. Oral sucker poorly developed; oesophagus relatively long; intestine X-shaped, with posterior caecal branches much longer than anterior ones. Testicular tissue in single mass, located mostly between caeca; auxiliary external seminal vesicle present; cirrus sac well developed. Ovary post-testicular, in posterior third of body; uterus extending anterior to ovary; metraterm conspicuously muscular; vitellarium follicular, extensive, extending along majority of body length. Genital pores separate, dorsal, postovarian. Excretory vesicle small, with 2 distensible anteriorly directed arms. Parasites of vascular system of marine teleosts.



Figs 1, 2. Pearsonellum corventum, sp. nov.: I, holotype, ventral view; 2, terminal genitalia, modified slightly after those in holotype to clearly portray all ducts and a more representative ovary.

vedentás Pelipolitál szítelé Parapoli inperiori

# Differential Diagnosis

Body spines in ventrolateral transverse rows. Intestine X-shaped with long posterior caeca. Testis single; auxiliary external seminal vesicle present; cirrus sac well developed. Ovary post-testicular; uterus extending preovarially; metraterm well developed.

# Pearsonellum corventum, sp. nov.

(Figs 1-13)

### Material Deposited

Holotype, from ventriculus of *Plectropomus leopardus* (Lacepède, 1802), Heron Island, Great Barrier Reef, R. M. Overstreet, Queensland Museum (QM) GI.10091.

Paratypes: QM GL10092 and British Museum (Natural History) No. 1988.2.10.1.

#### Localities

Type locality: Reefs off Heron I. Other locality: off Wistari Reef. All locations in Capricornia Section of the Great Barrier Reef.

### Hosts

Type host, *Plectropomus leopardus* (Lacepède, 1802) (Serranidae). Other hosts, *Epinephelus quoyanus* (Valenciennes, 1830), *E. ongus* (Bloch, 1790), *E. merra* Bloch, 1793 (Serranidae).

Sites in Host. Ventriculus and bulbus arteriosus of heart.

### Etymology

The Latin 'corventum' is a perfect participle that means 'having come from the heart' (cor, heart; venire, to come).

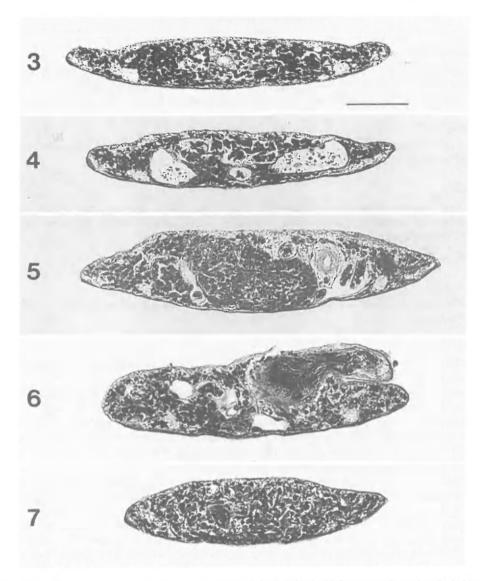
## Description

(Based on holotype, six paratypes, and portions of a few sectioned specimens. Measurements are of holotype and paratypes, in micrometres.)

Body flat, elongate fusiform, bluntly pointed at both ends in most specimens, spined, 832-1595 long, 127-348 wide, 15-22 × longer than wide, with nearly uniform width along middle half of most specimens. Spines in transverse rows at or near margin on ventral side, c. 4-5 long along most of body but c. 3 long anteriorly and posteriorly, susceptible to erosion; rows 8-22 wide along most of body length but typically c. 17 wide in most specimens, narrower and closer together near posterior end, continuing along posterior tip, containing 5-13 spines per row with 8-11 in most rows, not supporting rosethorn-shaped spines posteriorly. Nerve cords conspicuous laterally, joining posteriorly c. 10 from end of body; anterior commissure dorsal, 78-104 or 6-11% of body length from anterior end. Mouth subterminal, located ventrally, 4-10 from anterior end, surrounded by weakly muscular protrusile spherical to pyriform oral sucker (oral disc). Oseophagus slightly sinuous, 340-558 long, thick-walled except at short thin anterior portion, with distinct proximal portion; proximal portion 42-123 long, constituting 11-22% of oesophageal length; glandular cells surrounding oesophagus at level  $\frac{2}{3} - \frac{3}{4}$  distance from its anterior end, located in relatively narrow band several cells thick (Fig. 3). Intestine X-shaped, joining oesophagus 29-44% of body length from anterior end; anterior caecal arms sinuous or swollen depending on state of contraction when fixed, roughly equal in length, 32-78 long; posterior caeca sinuous, 200-450 long, unbranched, 23-30% of body length, 4-8× longer than anterior caeca, separated from anterior caeca in some specimens by relatively short common gut-space, with distal tip usually swollen.

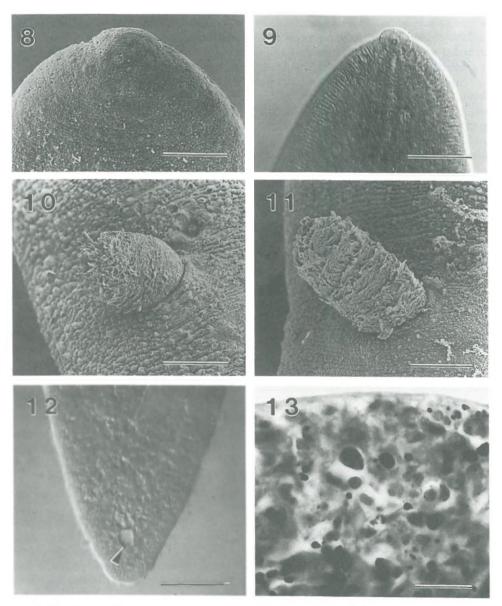
Testis elongate, irregular in shape but usually recognisable as single mass, with lateral and anterior lobes of various lengths, primarily confined between posterior caeca but with few or most lobes usually extending laterally beyond portions or all of sinuous caeca; testicular mass 253-656 long, 72-241 wide, 3-4× longer than wide, with length 30-44% of body length, containing sperm collection ducts often enlarged with abundance of spermatozoa,

accommodating narrow refractive primarily dorsoventral ducts. Post-testicular space 30-36% of body length. Vasa efferentia branched, with primary duct extending posteriorly to relatively short vas deferens and to 'auxiliary external seminal vesicle'; auxiliary vesicle spherical to teardrop-shaped, 27-30 long, 24-42 wide, 59-71 from posterior end, usually filled with degenerating sperm, surrounded by but not necessarily associated with eosinophilic



Figs 3-7. Pearsonellum corventum, sp. nov., cross-sections stained with haematoxylin and eosin (relate sections with levels in Fig. 1): 3, anterior section, approximately two-thirds distance down oesophagus, showing glandular cells with relatively clear and abundant cytoplasm surrounding oesophagus (relatively clear ventral structures near margins are nerve cords); 4, section through two anterior caecal branches and centrally located oesophagus at its posterior level (note partially digested blood in gut and clusters of spermatogonia in different stages of spermatogenesis in the testicular mass which occurs directly above oesophagus); 5, section through ovary showing progression in development of ova (round cross-section of eosinophilic muscular metraterm is located to right of ovary with two sections of egg-filled uterus to its right, one dorsal to it, and one to its left; vitelline duct is ventral, adjacent to uterus at left of ovary); 6, section showing dorsally everted cirrus packed with sperm (near ventral surface of worm, immediately to left of midline, is the sperm-filled oviduct with the vitelline duct to its left); 7, posteriormost section through auxiliary seminal vesicle located in central portion of worm to left of midline (note abundance of inclusions associated with some of cells). Scale line for all, 50 µm.

material and parenchymal cells (Figs 1, 7, 13); cells around auxiliary vesicle consisting of more than one type, occupying most of posterior end and apparently lateral areas of most of worm, with many containing strongly basophilic nuclei and inclusion bodies. Cirrus sac sinistral, 12–16% of body length from posterior end, 56–118 long, 34–103 wide, 1·1–1·8× longer than wide, with muscular wall 3–10 (usually 4) thick, containing relatively large



Figs 8-13. Pearsonellum corventum, sp nov.: 8, anterior end, dorsal view, SEM; 9, anterior end, ventral view showing oral disc and ventrolateral transverse rows of spines; Nomarski optics; 10, cirrus partly extruded adjacent to small female genital pore, dorsal view (note tegumental artifacts resulting from preparation; SEM); 11, cirrus almost completely everted (note similar rugged appearance of cirrus as in Fig. 10, but with less distinct female pore and better fixed tegument; SEM); 12, posterior end showing small excretory vesicle (arrowhead) with short portions of two arms distending anteriorly, dorsal view (note sperm-filled oviduct extending from near centre up toward right side; Nomarski optics); 13, cross-section of posterior portion of worm immediately below level of Fig. 7, showing wide distribution of densely stained bodies suspected of being haematin, haematoxylin and eosin. Scale lines: 8, 10, 25 μm; 9, 50 μm; 11, 20 μm; 12, 40 μm; 13, 15 μm.

seminal vesicle obscuring relatively short prostatic duct with associated small prostate cells and somewhat convoluted muscular male duct laterally; cirrus relatively large when everted through dorsal anterolateral male pore (especially large ones 86 long by 39 wide at base in  $108 \times 86$  sac and  $91 \times 44$  in  $105 \times 83$  sac), with rugged plicated projections 1-2 long; vas deferens entering sac mesially at about midlength.

Ovary spherical, irregular or arcuate, 47-175 long, 52-147 wide, containing narrow dorsoventral refractive ducts similar to those in testis, usually medial, at or slightly anterior to level of male pore, often separated from posterior margin of testis by caecal tips, located 22-25% of body length from posterior end. Oviduct usually crooked before conspicuously expanded with mature spermatozoa as a distinct descending organ 105-214 long, narrowing at posterior portion of descent before bending anteriorly and joining with common vitelline duct immediately before reaching ootype with its associated relatively inconspicuous Mehlis' gland. Uterus with few convolutions before extending anteriorly along margin of anterior half of ovary, retracing its path preovarially and separating ovary from testis in most specimens before joining metraterm; metraterm highly muscular, usually arcuate, separated from left side of ovary by uterus, 67-155 long, 17-34 wide, usually extending slightly above and below ovarian level. Female pore dorsal, slightly (10-30 µm in few measured specimens) anteromedial to male pore. Eggs up to 31 long by 17 wide, thinly shelled when in utero, 20-27 long by 10-15 wide when fixed in tissues; eggs embedded in host tissue conspicuously tanned, operculated. Vitelline follicles relatively small, mostly compacted in large masses, occupying most area between lateral nerve cords to or slightly anterior to dorsal nerve commissure, extending posteriorly to or occasionally past level of mid cirrus sac.

Excretory vesicle nearly spherical, 10-24 in diameter when filled, 22 from posterior end of body, with pair of excretory arms extending anteriorly; pore subterminal.

### Discussion

The few of specimens, usually one to four, collected from the lumen of the heart in each host suggests that *Pearsonellum corventum* does not occur in abundance. Sections from a portion of the ventricle from one specimen each of *Plectropomus leopardus* and *Epinephelus quoyanus*, however, suggest that the species occurs abundantly among trabeculae of the ventricular spongy layer (spongiosa). For example, a total of 11 individuals were found in three separated cuts of a small portion of the ventricle of the 569-mm-st specimen of *P. leopardus*. Worms, however, did not occur in the apex of the ventriculus. In most fish examined, the digeneans fitted in a restrained position between trabeculae and presumably did not readily dislodge. The few in the lumen of the bulbus arteriosus were seen readily and collected easily. Perhaps those specimens had dislodged from the ventricle, and such individuals ultimately could be pumped with the arterial blood flow from the bulbus arteriosus to recipient tissues where they settle. We did not see any such specimens.

The auxiliary external seminal vesicle is unique, but not clearly apparent in poorly prepared specimens. Its function was not determined. It could serve as a storage receptacle for spermatozoa. In the testis of some specimens, numerous ducts were swollen with mature spermatoxoa. Since many sperm developed simultaneously, an excess was probably produced, and that excess could become established in the vesicle. Perhaps that auxiliary male vesicle acts as a structure analogous to Juel's organ in the female system of hemiuroids (Gibson and Bray 1979). Because spermatozoa in the vesicle often appeared in a state of degeneration, the structure may serve to concentrate and conserve raw materials of sperm for future sperm production rather than allowing the sperm to discharge from a non-mating individual or to maintain it as a reservoir. On the other hand, the structure may be homologous to the posterior testis found in a few species discussed below. The rear portion of the worm supporting the vesicle contained a few different kinds of cells (Figs 7, 13); their associations with the vesicle and their functions were not investigated, but should be. Many of the cells appeared superficially like pycnotic cells, but close examination of the dense large and small inclusion bodies revealed that some occurred in cells with healthy nuclei. All or some of this dark-appearing material may be haematin or some other product of the blood meals of the worm; it occurred within and between some parenchymal cells throughout the specimens. The caeca were not lined with abundant gastrodermal cells, but most of the few cells associated with the caeca contained the material.

Relatively narrow refractive, but not birefringent, ducts extend primarily in a dorsoventral course through the testis and ovary. These ducts or channels, also called perforations or pores in some descriptions, have been reported from the testis of several other blood flukes but apparently do not occur in all. For example, Manter (1947) reported them in the testis of *Deontacylix ovalis* Linton, 1910 and *Cardicola cardiocola* (Manter, 1947) [not *C. cardicola* as proposed by Short (1953) and Yamaguti (1971)] but later (1954) specifically reported them as absent in *C. coridodacis* Manter, 1954; Yamaguti (1970) reported them as present in the testis of *C. chaetodontis*, but apparently not in *C. mugilis* Yamaguti, 1970. They occur in the gonads of several blood flukes. For example, we saw them in both testis and ovary of *Neoparacardicola nasonis* Yamaguti, 1970, as well as in the ovary of *D. ovalis*. At least some of these channels in *P. corventum*, usually 1–3 µm wide, passed from the caeca through the testis to the tegument. Autoradiography and electron microscopy should help explain the function and phylogenetic significance of the structures.

Probably not unique among sanguinicolids is the presence in *P. corventum* of operculated eggs. Yamaguti (1971), however, considered the absence of an operculum in eggs to be diagnostic for the Sanguinicolidae.

The poorly developed oral sucker in *P. corventum* (Fig. 9) also occurs in several blood flukes, some of which are discussed below. Perhaps the sucker is more likely to develop in species normally occurring in the lumina of the heart, among ventricular trabeculae, or in some other relatively spacious areas than in species that lodge passively in vessels.

The systematic position of P. corventum is not as clear as its specific distinctness. The questionable phylogenetic status of some of its characters reaffirms the confused state of blood fluke systematics in general. We agree with Van der Land (1967) and Holmes (1971) that the four subfamilies established by Yamaguti (1958) obscure the true relationships among the genera of blood flukes. The problem is more strongly exemplified by the two additional subfamilies that Yamaguti established later (Yamaguti 1971). We also agree with Van der Land (1967) and Smith (1972) in considering Appropriatelylidae Odhner, 1912 a junior synonym of Sanguinicolidae von Graff, 1907. Since the recognition of 10 genera reported by Yamaguti (1971), seven additional genera have been described; Orchispirium Madhavi & Rao, 1970; Psettarioides Lebedev & Paruchin, 1972; Metaplehniella Lebedev & Paruchin, 1972; Paracardicoloides Martin, 1974; Plethorchis Martin, 1975; Hyperandrotrema Maillard & Ktari, 1978; and Pseudocardicola Paruchin, 1985. Until representative specimens of species of all 18 currently accepted genera can be re-examined critically for comparison of reproductive structures, especially terminal genitalia but also glandular and other structures. a cladistical phylogenetic reassessment of the group may be premature. Nevertheless, we can suggest relationships among P. corventum and members of genera in at least three 'subfamilies'. Pearsonellum corventum appears associated with Cardicola Short, 1953 because of the intestinal shape, single testis, and body spination. Its thick-walled cirrus sac, preovarian uterine loops, and auxiliary external seminal vesicle, however, separate it from members of that relatively large genus, even though the uterus in C. congruenta Lebedev & Mamaev, 1968 and C. whitteni Manter, 1954 extends slightly above the ovary. The cirrus sac appears to be thick-walled (Lebedev and Mamaev 1968) in both C. grandis Lebedev & Mamaev, 1968 and C. congruenta, and these and some other species should be re-examined.

Members of *Deontacylix* Linton, 1910 have an anteriorly looping uterus and probably have a close phylogenetic relationship with *P. corventum*. The structure in *D. ovalis* referred to as a seminal receptacle by Linton (1910) and others is not an auxiliary seminal vesicle but the sperm-packed duct leading directly from the ovary which we and many other contemporary authors call the oviduct. To compare *P. corventum* with *D. ovalis*, we examined 23 specimens of the latter reported by Overstreet (1969) and here provide supplemental data on the species. The cirrus sac has a relatively thick wall and contains a moderate number of prostatic cells, and the testis varies among individuals. The testis is generally a single irregularly shaped mass with refractive ducts mentioned earlier, and often with other ducts swollen with sperm. It did not necessarily consist of 'two symmetrical lateral wings which may or may not be continuous in median line', a feature used in the subfamily

diagnosis by Yamaguti (1971). Neither did it appear as multiple, irregularly arranged testes as diagrammed by Van der Land (1967). The illustration of *D. ovalis* by Manter (1947, fig. 150) showed a dextral lobed ovary as reported by Linton (1910), but Manter (1947) stated it occurred to the left of the midline. In our examination of the ovary in the 23 specimens, we found it was always dextral.

In life, *D. ovalis* is chalky white like *P. corventum*, but *D. ovalis* is wider and more elliptical, with a yellowish tint because the intestine contains different food. Those features reflect the different habitat of *D. ovalis*; it lives in the body cavity rather than in the blood vascular system like *P. corventum* and typical blood flukes. Its spines occur over most of the ventral, thick, muscular surface and dorsal margins rather than on the delicate ventrolateral margins as in *P. corventum*. Those features allow the species to move rapidly on the viscera and peritoneum in an inchworm manner, rather than to passively attach to heart trabeculae. Also, unlike *P. corventum*, it and the other known member, *D. kyphosi* Yamaguti, 1970, produce a large number of eggs and lack the auxiliary seminal vesicle and the conspicuously muscular metraterm.

Most species in monotypic blood fluke genera also suggest a close relationship among many genera. Three that exemplify relationships with P. corventum are discussed below. Neongracardicola nasonis has a muscular cirrus sac, even though the sac is anterior rather than posterior to the ovary and the cirrus everts posterolaterally rather than anterolaterally. The anterior of two testes in that species is not as dendritic as reported by Yamaguti (1970). An examination of the holotype (USNM Hel. Coll. No. 63554) revealed the illustrated (Yamaguti 1970, fig. 24) 'four simple or ramified main branches' to be ducts swollen with sperm rather than the entire testis. It also has a weakly developed oral sucker as in P. corventum and in other species such as Paracardicoloides vamagutii Martin, 1974 (from blood vessels of an eel in Queensland), which has two testes, a completely preovarian uterus, and differently shaped caeca. Metaplehniella lethrini Lebedev & Paruchin, 1972 from the stomach (probably in the wall) of a lethrinid off India is related to P. corventum; but it might be much more closely related that its description reveals if the illustrated (Lebedev and Paruchin 1972) postcaecal testis was an atypical condition, if the cirrus sac has a highly muscular wall, and if an auxiliary seminal vesicle exists. The auxiliary vesicle and the highly muscular metraterm in combination with various other features (many indicated above) in P. corventum clearly separate Pearsonellum from all other genera.

Probably, too many monotypic blood fluke genera have been erected, but most fishes have not been examined for blood flukes, life cycles for members of most blood fluke genera have not been elucidated, several species require a critical re-examination to clarify the presence and status of some characters, and the taxonomic significance of all the morphological features has not been fully appreciated.

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