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A Revision of the Calicotylinae Monticelli, 1903 (Monogenea: Monocotylidae)

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Abstract

A total of 153 elasmobranchs (46 species), either freshly collected from the Gulf of Mexico, USA, and Tasmania, Australia, or museum specimens collected from various localities worldwide, were examined for calicotyline (Monocotylidae) monogeneans. Thirty-five elasmobranchs, representing 17 species, were infected with *Calicotyle* spp. which we identified as the following previously described species: *C. asterii* (Szidat, 1970) Timofeeva, 1985; *C. kroyeri* Diesing, 1850; *C. macrocotyle* Cordero, 1944; *C. similis* (Szidat, 1972) Timofeeva, 1985; *C. splendens* (Szidat, 1970) Timofeeva, 1985; *C. stossichi* Braun, 1899; and *C. urolophi* Chisholm, Beverley-Burton & Last, 1991. The Calicotylinae, which comprises the genera *Calicotyle* and *Dictyocotyle*, is revised based on supplementary material as well as deposited type-material. We consider 14 of the 17 nominal *Calicotyle* spp. to be valid. *C. rosinae* Kusnetzova, 1970 is synonymized with *C. macrocotyle*, *C. sjegi* Kusnetzova, 1970 is considered a *species inquirenda*, and *C. inermis* Woolcock, 1936 a *species incertae sedis*. Additional data and illustrations to show the morphological features of the hamuli and male copulatory organ, the form of the intestinal caeca, vaginae, and ovary, and the distribution of the vitellarium are provided for all valid species. The distribution of the 14 hooklets in the adult haptor of *Dictyocotyle coeliaca* Nybelin, 1941 is illustrated for the first time. We provide new host and locality records for *C. asterii*, *C. kroyeri*, *C. macrocotyle*, and *C. stossichi* and new locality records for *C. similis* and *C. splendens*. A key to species of the Calicotylinae is also included. Host-specificity, geographical distribution, and the need for information regarding the development of individuals from juvenile to adult are discussed.

Introduction

The monocotylid subfamily Calicotylinae currently comprises two genera, *Calicotyle* Diesing, 1850 and *Dictyocotyle* Nybelin, 1941 (see Chisholm et al., 1995). To date, 17 species of *Calicotyle* have been described from the cloaca, rectum, rectal gland, spiral valve, or oviducts of their chondrichthyan hosts. *Calicotyle* is distinguished from other genera in the family by the haptor, which is divided into one central and seven peripheral loculi; the presence of hamuli; the lack of other haptoral accessory structures such as sclerotized septal ridges, sclerites, or dorsal structures; and the absence of a marginal valve. *Dictyocotyle* is monotypic with *D. coeliaca* Nybelin, 1941 being found on the wall of the body cavity in a number of rajiform species. *D. coeliaca* is unique in that the haptor is divided into numerous random loculi, and hamuli are apparently absent.

Whereas much attention has focused on the description of some calicotylines, many of the species have not been examined closely since they were described originally. In some cases, the original descriptions are incomplete and the type-material is in poor condition or not available. Although the phylogenetic analysis of Chisholm et al. (1995) confirmed the validity of *Calicotyle* and *Dictyocotyle*, they stated that *Calicotyle* was in need of revision. In the present study, we obtained new material of seven previously described *Calicotyle* spp., most of which represent new host and locality records. Examination of this additional material and of type specimens has allowed us to revise the subfamily and genera, to provide a key to species, and to comment on host-specificity and the geographical distribution of the members of *Calicotyle*.

Materials and methods

Four specimens of *Raja olsenii* Bigelow & Schroeder (the spreadfin skate) were collected by otter trawl in the N.W. Gulf of Mexico (27°49'N, 94°57'W) in August 1985. The elasmobranchs were killed by pithing, preserved in 10% formalin, and then transferred to 70% ethanol for storage. Eighteen specimens of *Mustelus norrisi* Springer (Florida smoothhound) were obtained by trawl or by hook and line in the Gulf of Mexico between 28–29°N and 86–87°W. These hosts were usually dead when brought to the surface, were quickly preserved in 10% formalin and later transferred to 70% ethanol. *Calicotyle* spp. were subsequently recovered from the cloaca and transferred to fresh 70% ethanol. Eight specimens of *Mustelus antarcticus* Günther were caught on long-line at Upper Pittwater, Tasmania (42°48'S, 147°31'E), two specimens of a longnose skate *Raja* sp. A (see Last & Stevens, 1994) were caught by seine net in North West Bay, Tasmania (43°01'S, 147°16'E) and one specimen of *Urolophus paucimaculatus* Dixon was caught by seine net in Norfolk Bay, Tasmania (43°02'S, 147°59'E) in January 1997. The rays were killed by pithing and the skin, cloaca, and rectum were examined prior to dissection. The cloaca, rectum, rectal gland, and oviducts were removed and examined under a stereomicroscope. Additional calicotylines were recovered from preserved hosts from the Texas Cooperative Wildlife Collections (TCWC) of the Texas A&M University, College Station, Texas 17843-2258, USA and the Florida State Museum (UF) of the University of Florida, Gainesville, Florida 32611, USA.

(see Table I). Parasites were removed with a fine brush and transferred to fresh 70% ethanol.

The parasites recovered from hosts collected from the Gulf of Mexico and museum specimens were rehydrated, stained overnight in a solution of Van Cleave's hematoxylin with several drops of Ehrlich's hematoxylin, and dehydrated to 70% ethanol. A few drops of aqueous saturated lithium carbonate and then a few drops of 6% butyl-amine solution were added to the 70% ethanol solution for further differentiation. The specimens were then fully dehydrated in an ethanol series, cleared in clove oil, and mounted in Piccolyte. Live *Calicotyle* specimens retrieved from hosts from Tasmania were flattened under a coverslip and fixed in 10% formalin. They were either left unstained or stained in acetocarmine, dehydrated in an ethanol series, cleared in cedarwood oil, and mounted in Canada balsam. Additional material of *C. kroyeri* and *D. coeliaca* from the North Sea, given to the senior author by Dr. D. I. Gibson of The Natural History Museum, London (NHM), was cleared in Hoyer's medium to facilitate examination of the haptor for hooklets and hamuli. Additional host material, which was examined but contained no parasites, is listed in Table II.

Live *Calicotyle* from hosts collected in Tasmania were examined under a compound microscope for the presence of anterior glands. Whole-mounts were examined using a compound photomicroscope equipped with phase contrast optics, and drawings were made with the aid of a drawing tube. Measurements were made using a computerized digitizing system similar to that described by Roff & Hopcroft (1986). The measurements, in micrometers, are presented as the mean followed in parentheses, by the range and the number of structures measured. Measurements of the male copulatory organ are the total length including all curves. Haptoral terminology follows that of Chisholm et al. (1995) and the numbering of hooklets follows that of Llewellyn (1963). Terminology of the anterior glands follows that of Chisholm & Whittington (1996).

Type-material of *Calicotyle* and *Dictyocotyle* was obtained from the following institutions or personal collections; personal collection of Professeur L. Euzet (LE), Station Méditerranéenne de l'Environnement Littoral, 1, Quai de la Daurade, 34200 Sète, France; personal collection of Professor Klaus Rohde (KR), Department of Zoology, University of New England, Armidale, NSW 2351, Australia; Museo Argentino de Ciencias Naturales (MACN), Instituto Nacional de Investigación de Las Ciencias Naturales "Bernardina Rivadavia" Avda. Angel Gallardo 470, Casilla de Correo 10 – Sucursal 5, Buenos Aires, Argentina; The Natural History Museum (NHM), Cromwell Rd., London SW7 5BD, UK; Queensland Museum (QM), PO Box 3300 Brisbane, Queensland 4101, Australia; United States National Parasite Collection (USNPC), USDA, Beltsville, Maryland 20705, USA; Zoological Institute, Russian Academy of Sciences (ZISP), Laboratory of Parasitic Worms, St. Petersburg 199034, Russia. Voucher material is deposited in the NHM, QM, and USNPC.

Table I. Host-parasite list of *Calicotyle* spp. collected in the present study. The number in parentheses after the number examined represents the number of hosts infected and the number in parentheses after the parasite species represents the number of parasites collected. TCWC = Texas Cooperative Wildlife Collection, TJH = Collection of T. J. Hansknecht, UF = Florida State Museum.

Host Species	Collection number	Number examined	Parasites recovered	Site	Locality
<i>Anacanthobatis folirostris</i> ^a	TJH 25	1	<i>C. kroyeri</i> (1)	cloaca	NW Gulf of Mexico, 27°21'N, 96°00'W
<i>A. folirostris</i> ^a	UF 40141	3(2)	<i>C. kroyeri</i> (3)	cloaca	NW Gulf of Mexico, 27°25'N, 95°56'W
<i>A. folirostris</i> ^a	UF 3984		1	<i>C. kroyeri</i> (1)	cloaca
<i>Dactylobatis armatus</i> ^a	UF 41274	1	<i>C. kroyeri</i> (2)	cloaca	NW Atlantic off Florida, 28°30'N, 79°46'W
<i>D. armatus</i>	UF 10157	2	<i>C. kroyeri</i> (1)	cloaca	No locality data
<i>Mustelus antarcticus</i>	n/a	8(2)	<i>C. stossichi</i> (2)	rectal gland	SE Tasmania, Upper Pittwater, 42°48'S, 147°31'E
<i>M. mustelus</i> ^b	UF 29945	1	<i>C. stossichi</i> (2)	rectal gland	SE Atlantic; Angola, Luanda, 08°47'S, 13°16'E
<i>M. norrisi</i> ^a	TJH	1	<i>C. asterii</i> (2)	cloaca	NW Gulf of Mexico, 28°16'N, 19°21'W
<i>M. norrisi</i> ^a	TJH	17(6)	<i>C. stossichi</i> (6)	rectum	Gulf of Mexico, 29°N, 86–87°W
<i>Raja agassizi</i> ^b	TCWC 3357.3	2 (1)	<i>C. splendens</i> (1)	cloaca	SW Atlantic
<i>R. australis</i>	TCWC 1.12050	1	<i>Calicotyle</i> sp. ^c (2)	spiral valve, rectum	SW Pacific, New South Wales, Australia
<i>R. batis</i>	TCWC 2819.5	1	<i>C. kroyeri</i> (1)	skin	NE Atlantic, off Britain
<i>R. brachyura</i> ^b	TCWC 2819.2	1	<i>C. kroyeri</i> (1)	cloaca	NE Atlantic, West English Channel near Plymouth
<i>R. cerva</i>	TCWC 38606	1	<i>Calicotyle</i> sp. ^c (3)	cloaca, rectum	SW Pacific, New South Wales, Australia
<i>R. cervigoni</i>	TCWC 11282	1	<i>Calicotyle</i> sp. ^c (1)	rectal gland	NW Atlantic, Caribbean Sea
<i>R. cyclophora</i> ^b	TCWC 396.1	2 (1)	<i>C. similis</i> (1)	cloaca	SW Atlantic off Brazil
<i>R. garmani</i> ^a	TCWC 188.1	15(1)	<i>C. kroyeri</i> (1)	cloaca	NW Atlantic, Virginia, 36°35'N, 74°42'W
<i>R. olseni</i> ^a	UF 31879	1	<i>C. kroyeri</i> (4)	cloaca	SW Gulf of Mexico, Campeche Banks, 19°41'N, 92°26'W
<i>R. olseni</i> ^a	UF 42596	2 (1)	<i>C. kroyeri</i> (4)	cloaca	Northern Gulf of Mexico, 29°12'N, 88°25'W
<i>R. olseni</i> ^a	TJH 50	4(1)	<i>C. kroyeri</i> (5)	cloaca	NW Gulf of Mexico, 27°49'N, 94°57'W
<i>Raja</i> sp. A	n/a	2(2)	<i>C. kroyeri</i> (29)	cloaca	SE Tasmania, North West Bay, 43°01'S, 147°16'E
<i>Sympterygia bonapartei</i> ^a	TCWC 3698.1	4(2)	<i>C. macrocotyle</i> (11)	cloaca, rectum	SW Atlantic, Rio Grande do Sul, Brazil
<i>S. bonapartei</i> ^a	TCWC 3358.1	4(2)	<i>C. macrocotyle</i> (1)	cloaca	SW Atlantic
<i>Urolophus paucimaculatus</i>	n/a	1	<i>C. urolophi</i> (1)	cloaca	SE Tasmania, Norfolk Bay, 43°02'S, 147°59'E

a. New host and locality record for parasite.

b. New locality record for parasite.

c. Species could not be identified due to poor condition of parasite.

Table II. Chondrichthyans (29 species) examined in which no calicotylinae parasites were found. Collection codes as in Table I.

Host species	Collection number	Number examined	Site examined	Locality
<i>Benthobatis marcida</i>	UF 41282	1	cloaca	NW Atlantic off Florida, 28°40'N, 79°44'W
<i>Breviraja spinose</i>	TCWC 42955	5	cloaca, rectal gland, oviduct	NW Atlantic
<i>Callorhynchus milii</i>	n/a	4	cloaca, rectum, rectal gland, body cavity	SE Tasmania, Upper Pittwater, 42°48'S, 147°31'E
<i>Cephaloscyllium laticeps</i>	n/a	1	cloaca, rectum, rectal gland, body cavity	SE Tasmania, Fortesque Bay, 43°05'S, 147°58'E
<i>C. laticeps</i>	n/a	1	cloaca, rectum, rectal gland, body cavity	SE Tasmania, Bruny Island, 43°06'S, 147°24'E
<i>Galeorhinus galeus</i>	n/a	1	cloaca, rectum, rectal gland, body cavity	SE Tasmania, Norfolk Bay, 43°02'S, 147°49'E
<i>Myliobatis australis</i>	n/a	1	cloaca, rectum, rectal gland, body cavity	SE Tasmania, Norfolk Bay, 43°02'S, 147°49'E
<i>Pristiophorus schroederi</i>	UF 24621	1	cloaca	NW Atlantic
<i>Psammobatis maculatus</i>	TCWC 030	1	cloaca	No information
<i>P. scobina</i>	USNPC 228152	1	cloaca	SE Pacific
<i>Raja alba</i>	TCWC 3093.4	2	cloaca, rectal gland	NE & SE Atlantic, England, South Africa
<i>R. clavata</i>	TCWC 309.1	1	cloaca, rectum, oviducts	NE Atlantic, British Waters
<i>R. eglanteria</i>	TCWC	13	cloaca	Gulf of Mexico, shallow water
<i>R. erinacea</i>	TCWC	2827.1	1	cloaca, rectum, rectal gland
<i>R. erinacea</i>	UF 4201, 29968, 32056	3	cloaca	No information
<i>R. fulginea</i>	TCWC 2812.1	1	cloaca	Atlantic, Caribbean Sea
<i>R. garmani lentiginosa</i>	TJH 51	1	cloaca, rectal gland, rectum	NW Gulf of Mexico, 27°49'N, 94°57'W
<i>R. laevis</i>	UF 13047, 13048	2	cloaca	No information
<i>R. lentiginosa</i>	UF 2221.9	1	cloaca	NE Gulf of Mexico, 29°19'N, 38°42'W
<i>R. microocellata</i>	TCWC 2819.3	1	cloaca	NE Atlantic, Plymouth Area, W. English Channel
<i>R. miraletus</i>	TCWC 3091.1	1	cloaca	SE Atlantic, off South Africa
<i>R. miraletus</i>	UF 29960(3), 29984 (4)	7	cloaca	No information
<i>R. montagui</i>	TCWC 2819.6	1	cloaca	NE Atlantic, Plymouth Area, W. English Channel
<i>R. senta</i>	TCWC	1	cloaca	NW Atlantic, 42°05'N, 70°11'W
<i>R. senta</i>	UF 4202	2	cloaca	NW Atlantic, St. Georges Bank, Massachusetts
<i>R. straeleni</i>	TCWC 3090.1	1	cloaca	SE Atlantic
<i>R. straeleni</i>	UF 29992	2	cloaca	No information
<i>R. teevani</i>	UF 32670	1	cloaca	No information
<i>R. texana</i>	TCWC 336.1	1	cloaca	NW Gulf of Mexico, off Freeport, Texas
<i>R. texana</i>	UF 29985	1	cloaca	NW Gulf of Mexico, off Texas
<i>Scyliorhinus rotifer</i>	UF 27984	1	cloaca, rectal gland	Gulf of Mexico
<i>Squalus acanthias</i>	n/a	4	cloaca, rectum, rectal gland, body cavity	SE Tasmania, North West Bay, 43°01'S, 147°16'E
<i>S. acanthias</i>	n/a	1	cloaca, rectum, rectal gland, body cavity	SE Tasmania, Bruny Island, 43°06'S, 147°24'E
<i>Sympterygia acuta</i>	TCWC 266.2	2	cloaca, rectal gland	SW Atlantic, Argentina
<i>S. brevicaudata</i>	TCWC 3118.1, 3119.1	6	cloaca, rectal gland	SE Pacific, Peru
<i>Urolophus cruciatus</i>	n/a	1	cloaca, rectum, rectal gland, body cavity	SE Tasmania, North West Bay, 43°01'S, 147°16'E

Monocotylidae Taschenberg, 1879
Calicotylinae Monticelli, 1903

Revised diagnosis

Monocotylidae. Haptor with one central and seven peripheral loculi or rarely with numerous unevenly distributed loculi. Marginal valve absent. Hamuli present or absent. Hooklets 14, distributed in muscular rim of haptor. Additional haptoral accessory structures lacking (septal sclerites, sinuous sclerotized septal ridge, marginal muscular papillae or dorsal accessory structures). Eye-spots in form of dispersed pigment granules present or absent. Pharyngeal glands prominent. Intestinal caeca smooth or with short lateral and medial pouches or longer diverticula, not confluent posteriorly. Testes follicular. Seminal vesicle present. Male copulatory organ a sclerotized narrow tube; accessory piece and accessory filaments absent. Ejaculatory bulb with two spherical internal chambers. Vagina double; vaginal pores unarmed. Vitellarium extending from level of pharynx to posterior portion of body, confluent medially in posterior part of body or not. Eggs tetrahedral or ovoid longitudinally with three flattened sides which are triangular in cross-section. Parasites of cloaca, rectum, spiral valve, rectal gland, oviducts, and inner wall of body cavity of chondrichthyan fishes.

***Calicotyle* Diesing, 1850**

Revised diagnosis

Monocotylidae: Calicotylinae. Haptor with one central and seven peripheral loculi. Hamuli present. Eyespots in form of dispersed pigment granules present or absent. Intestinal caeca smooth or with short lateral and medial diverticula. Eggs tetrahedral. Parasites of cloaca, rectum, spiral valve, rectal gland, and oviducts of chondrichthyan fishes.

Type-species: *C. kroyeri* Diesing, 1850.

Additional species: *C. affinis* Scott, 1911; *C. asterii* (Szidat, 1970) Suriano, 1977; *C. australiensis* Rohde, Heap, Hayward & Graham, 1992; *C. australis* Johnston, 1934; *C. macrocotyle* Cordero, 1944 (= *C. rosinae* Kusnetzova, 1970, new synonym); *C. mitsukurii* Goto, 1894; *C. palombi* Euzet & Williams, 1960; *C. quequeni* (Szidat, 1972) Timofeeva, 1985; *C. ramsayi* Robinson, 1961; *C. similis* (Szidat, 1972) Timofeeva, 1985; *C. splendens* (Szidat, 1970) Timofeeva, 1985; *C. stossichi* Braun, 1899; *C. urolphi* Chisholm, Beverley-Burton & Last, 1991.

Species inquirenda: *C. sjegei* Kusnetzova, 1970.

Species incertae sedis: *C. inermis* Woolcock, 1936.

***Calicotyle affinis* Scott, 1911**

(Figures 2A, 3A, 4A)

Synonym: *Calicotyle* (*Calicotylides*) *affinis* (Scott, 1911) Nybelin, 1941.

Type-host: *Chimaera monstrosa* Linnaeus (Chimaeridae).

Type-locality: North Sea.

Additional record: *Raja fullonica*, North Sea (see Brinkmann, 1940).

Sites: Spiral valve, rectal gland, cloaca, and gills (but see Remarks section for comments on gills).

Material examined: Seven voucher specimens (2 slides) NHM 1988.10.11-16.

Additional data

Hamuli as illustrated (Figure 2A); note not completely in profile. Hooklets 13 (12–14, $n = 10$) long, distributed as in *C. kroyeri* (Figure 1A). Eye-spots in the form of dispersed pigment granules anterodorsal to pharynx. Intestinal caeca with small medial and lateral pouches; caeca following path as illustrated (Figure 3A). Male copulatory organ 1,650 (1,379–1,900, $n = 4$) long, with at least 3 irregular loops (Figure 4A). Ovary loosely coiled, lobed at blind end (Figure 3A). Vaginae forming U-shape (Figure 3A); vaginal pores intercaecal at level of common genital pore (Figure 3A). Vitellarium not confluent posteriorly (Figure 3A).

Remarks

Calicotyle affinis was originally described by Scott (1911) from the gills of *Chimaera monstrosa* but with little detail of the internal anatomy. Brinkmann (1940) provided a comprehensive redescription of *C. affinis*, noting that Scott's record of *C. affinis* on the gills was probably "incidental" because he found specimens only in the rectal gland of the type-host. Brinkmann (1940) also noted that on one occasion he found *C. affinis* in the rectal gland of *R. fullonica*. Hooklets and eye-spots (as dispersed pigment granules), which were not noted by Scott (1911) or Brinkmann (1940), were observed in the present study. Brinkmann (1940) depicted the coils of the male copulatory organ as regular, but the coils are irregular in the voucher material we examined (see Figure 4A). For discussion on the close affinity of *C. affinis* with *C. australiensis*, see the "Remarks" section of *C. australiensis*.

Figure 1. Haptors. A. *Calicotyle kroyeri*; B. *Dictyocotyle coeliaca*. Note: hooklets not to scale.

Figure 2. Hamuli. A. *Calicotyle affinis*; B. *C. asterii*; C. *C. australiensis*; D. *C. australis*; E. *C. kroyeri*; F. *C. macrocotyle*; G. *C. mitsukurii* (redrawn from Goto, 1894); H. *C. palombi*; I. *C. quequeni* (redrawn from Suriano, 1977); J. *C. similis*; K. *C. splendens*; L. *C. stossichi*; M. *C. urolophi*. All drawings to same scale. Scale-bar: 100 μ m.

Figure 3. *Calicotyle* spp. Whole body illustrating path and form of intestine and ovary, path of vaginae, and distribution of vitellarium, ventral view. A. *C. affinis*; B. *C. asterii*; C. *C. australiensis*; D. *C. australis*; E. *C. kroyeri*; F. *C. macrocotyle*; G. *C. mitsukurii* (redrawn from Goto, 1894); H. *C. palombi*; I. *C. quequeni* (redrawn from Szidat, 1972); J. *C. ramsayi*; K. *C. similis*; L.C. *splendens*; M.C. *stossichi*; N.C. *urolphi*. Scale-bars: 1000 μ m.

Figure 3. *Continued.*

Figure 4. Male copulatory organs of *Calicotyle* spp., ventral view. A. *C. affinis*; B. *C. asterii*; C. *C. australiensis*; D. *C. australis*; E. *C. kroyeri*; F. *C. macrocotyle*; G. *C. mitsukurii* (redrawn from Goto, 1894); H. *C. palombi*; I. *C. quequeni* (redrawn from Suriano, 1977); J. *C. ramsayi*; K. *C. similis*; L. *C. splendens*; M. *C. stossichi*; N. *C. urolophi*. All drawings to same scale. Scale-bar: 100 μ m.

***Calicotyle asterii* (Szidat, 1970) Suriano, 1977**
(Figures 2B, 3B, 4B)

Synonyms: *Paracalicotyle asterii* Szidat, 1970; *P. asterri* Szidat, 1970.

Type-host: *Mustelus schmitti* Springer (but note comments in Remarks section) (Triakidae).

Type-locality: Puerto Quequén, Buenos Aires, Argentina.

Additional records: *M. schmitti*, Mar del Plata, Buenos Aires, Argentina (see Suriano, 1977); *M. norrisi* Springer, Northern Gulf of Mexico (present study) (Triakidae).

Site: Cloaca.

Material examined: Holotype MACN 27.951. 2 voucher specimens USNPC 87188 and NHM 1997. 6.12.1.

Additional data

Hamuli as illustrated (Figure 2B). Hooklets 9 (8–10, n = 4) long, distributed in haptor as for *C. kroyeri* (Figure 1A). Eye-spots (as dispersed pigment granules) anterodorsal to pharynx. Intestinal caeca without pouches or diverticula, following path as illustrated (Figure 3B). Male copulatory organ looping once (Figure 4B). Ovary tightly coiled, lobed at blind end (Figure 3B). Vaginae forming U-shape (Figure 3B); vaginal pores intercaecal at level of common genital pore (Figure 3B). Vitellarium not confluent posteriorly (Figure 3B).

Remarks

Szidat (1970a) erected the new genus *Paracalicotyle* to accommodate the newly described *P. asterii*. This genus was distinguished from *Calicotyle* by the parallel-sided body and the small haptor curled inward. Suriano (1977) did not accept *Paracalicotyle* and moved *P. asterii* to *Calicotyle*. The original description of *C. asterii* was incomplete, but the parasite was adequately redescribed and illustrated by Suriano (1977). The presence of eye-spots was not noted by Szidat (1970a) or Suriano (1977), but they were observed in the present study in both the type and additional material. Szidat (1970a) cited the type-host of the species as *M. asterias*, but Suriano (1977) noted that *M. asterias* is not found in the South Atlantic and stated that the host must have been *M. schmitti*. The finding of *C. asterii* in the cloaca of *M. norrisi* in the Gulf of Mexico in the present study represents both a new host and locality record.

***Calicotyle australiensis* Rohde, Heap, Hayward & Graham, 1992**
(Figures 2C, 3C, 4C)

Type-host: *Chimaera* sp. II (Chimaeridae) (species not identified).

Type-locality: Southeastern coast of Australia.

Sites: Rectum and rectal gland.

Material examined: Holotype NHM 1991.1.2.1.; KR (17 specimens on 17 slides).

Additional data

Hamuli as illustrated (Figure 2C). Hooklets 14 (14–15, $n = 10$) long, distributed in haptor as for *C. kroyeri* (Figure 1A). Eye-spots (as dispersed pigment granules) anterodorsal to pharynx. Intestinal caeca without pouches or diverticula, following path as illustrated (Figure 3C). Male copulatory organ long with at least 4 irregular coils (Figure 4C). Ovary loosely coiled, lobed at blind end. Vaginae forming U-shape, each with single loop along their length (Figure 3C); vaginal pores overlying intestinal caeca at level just posterior to intestinal bifurcation (Figure 3C). Vitellarium not confluent posteriorly (Figure 3C).

Remarks

Eye-spots which were not described by Rohde et al. (1992) were observed as dispersed pigment granules in the type-material examined here. The coils of the male copulatory organ were depicted as regular by Rohde et al. (1992), but they are irregular (see Figure 4C). It is unfortunate that the host species could not be positively identified. Rohde et al. (1992) noted that *C. australiensis* from *Chimaera* sp. II in Australia was very similar to *C. affinis* from *Chimaera monstrosa*, stating that the only difference was the greater total length of the male copulatory organ in *C. australiensis*. However, as they demonstrated, the length of the male copulatory organ can vary significantly depending on the age of the specimen (see Rohde et al., 1992). Measurements we made of the male copulatory organ from type material of *C. affinis* ranged from 1,379–1,985 μm and Rohde et al. (1992) quoted male copulatory lengths for *C. affinis* as 1,620 and 1,680 μm . Therefore, the upper limit of our measurements is almost 300 μm longer than those quoted by Rohde et al. (1992) and, more importantly, it is approaching the lengths noted for mature *C. australiensis* (2,140–2,413 μm). Furthermore, the hooklets in these two species are significantly longer (12–15 μm) and more robust than those observed in the haptors of other species of *Calicotyle* (8–10 μm long). However, the intestinal caeca of *C. affinis* have small lateral and medial pouches which we could not confirm in the type-material of *C. australiensis* and the loop observed in the vaginae of *C. australiensis* was not seen in *C. affinis*. While it is our opinion that *C. australiensis* is probably synonymous with *C. affinis*, as Rohde et al. (1992) noted, more material from chimaeroid fishes, preferably from regions between Australia and the North Sea, must be examined before they are synonymized.

Rohde et al. (1992) also found three specimens of a species of *Calicotyle* in the rectum of *Rhinochimaera pacifica* (Mitsukuri). Even though the male copulatory organ in the *Calicotyle* sp. from *R. pacifica* was much shorter than that of *C. australiensis*, the two species were similar in all other aspects. Therefore, Rohde et al. (1992) decided not to name this species until more material was collected.

***Calicotyle australis* Johnston, 1934**

(Figures 2D, 3D, 4D)

Type-host: *Trygonorhina fasciata* Müller & Henle (Rhinobatidae).

Type-locality: Glenelg, South Australia.

Additional records: *Aptychotrema rostrata* (Shaw & Nodder) (= *A. banksii*) Moreton Bay, Queensland, Australia (see Young, 1970); *Rhinobatos typus* Bennett (= *R. batillum*) (see Whittington et al., 1989) (Rhinobatidae).

Sites: Cloaca, rectum and skin (but see Remarks section for comments on the skin).

Material examined: Two voucher specimens (two slides) NHM 1988.10.17.1-2.

Additional data

Hamuli as illustrated (Figure 2D). Hooklets 9 (8–10, n = 5) long, distributed in haptor as in *C. kroyeri* (Figure 1A). Intestinal caeca without pouches or diverticula, following path as illustrated (Figure 3D). Male copulatory organ looping once (Figure 4D). Ovary not coiled or loosely coiled; blind end not lobed (Figure 3D). Vaginae running approximately parallel with transverse vitelline duct (Figure 3D); vaginal pores located lateral to intestinal caeca at level of common genital pore (Figure 3D). Vitellarium not confluent posteriorly (Figure 3D).

Remarks

C. australis was originally described by Johnston (1934) and Williams (1958) noted that the species needed reexamination. Therefore, Whittington et al. (1989) redescribed this species and confirmed its validity. They stated that hooklets were absent in the adult haptor, but we observed 14 hooklets distributed as in *C. kroyeri* (see Figure 1A). Comparison of the voucher specimens of *C. australis* with the description and drawings of *C. mitsukurii* shows that the form of the vaginae, ovary, and intestinal caeca are very similar between the two species. The only differences are the larger hamuli and the longer male copulatory organ in *C. mitsukurii*. While the male copulatory organ is longer in *C. mitsukurii*, its morphology is similar to that of *C. australis* because both loop once (compare Figures 4D and 4G). The description of *C. mitsukurii* (see Goto, 1894) seems to have been based on a single specimen because no range of measurements was given. Because *C. australis* and *C. mitsukurii* differ only in the length of the hamuli and male copulatory organ, characters shown to vary with age in *Calicotyle* spp. (see Kearn, 1987; Rohde et al., 1992), it is possible that these species are synonymous. However, we refrain from synonymizing *C. australis* with *C. mitsukurii* until additional material of *C. mitsukurii* can be obtained. There have been records of *C. australis* from the ventral surface of the host (see Whittington et al., 1989). This site is erroneous because, as noted by Kearn (1987) and Whittington et al. (1989), it appears that when the host dies the parasites migrate out of the cloaca onto the ventral body surface.

***Calicotyle inermis* Woolcock, 1936**

Synonyms: *Calicotyle* (*Gymnocalicotyle*) *inermis* (Woolcock, 1936) Nybelin, 1941; *Gymnocalicotyle inermis* (Woolcock, 1936) Yamaguti, 1963.

Type-host: *Pristiophorus cirratus* Latham (Pristiophoridae).

Type-locality: Port Philip Bay, Victoria, Australia.

Site: Oviducts.

Material examined: None.

Remarks

Type-material of this species could not be located to verify the original description. Woolcock (1936) differentiated *C. inermis* from other species in the genus by the longer coiled male copulatory organ and the absence of hamuli. Nybelin (1941) moved this species into the subgenus *Gymnocalicotyle*, which Yamaguti (1963) later raised to generic status without comment. Chisholm et al. (1995) did not consider *Gymnocalicotyle* a distinct taxon. Until additional material of *C. inermis* can be examined to confirm the absence of hamuli, we follow the decision of Chisholm et al. (1995) and consider *C. inermis* a *species incertae sedis*.

***Calicotyle kroyeri* Diesing, 1850**

(Figures 1A, 2E, 3E, 4E, 5A)

Synonym: *Calicotyle (Calicotylides) kroyeri* (Diesing, 1850) Nybelin, 1941.

Type-host: *Raja radiata* Donovan (Rajidae).

Type-locality: Kattegat, Denmark, North Sea.

Additional records: See Lawler (1981) and Llewellyn et al. (1984); *Anacanthobatis folirostris* Bigelow & Schroeder, northwest and southwest Gulf of Mexico (present study); *Dactylobatis armatus* Bean & Weed, northwest Atlantic off Florida, USA (present study); *Raja batis* Linnaeus, northeast Atlantic, off Britain (present study); *Raja garmani* Whitley, northwest Atlantic off Virginia, USA (present study); *Raja olseni* Bigelow & Schroeder, southwest Northern Gulf of Mexico (present study); *Raja* sp. A (see Last & Stevens, 1994), southeast coast of Tasmania (see Table I for detailed information on new records).

Sites: Cloaca, rectum and rectal gland.

Material examined: Voucher (1 slide) NHM 1932.11.22.14, 1 slide NHM 1959.10.14.28, 3 slides NHM 1976.4.8.54-56, 7 slides NHM 1976.4.8.57-63, 1 slide (6 specimens) 1976.1.5.19-34, 1 slide (6 specimens) NHM 1988.10.14.20.26. Voucher specimens NHM 1997.6.12.12-14, QM G213849-60, USNPC 87189, 87190.

Additional data

Hamuli as illustrated (Figure 2E). Hooklets 9 (8–10, n = 7) distributed in haptor as in Figure 1A. One pair of anterolateral glands containing granular secretion present on each side of body just posterior to level of pharynx (Figure 5A). Single duct from each gland runs anteriorly to open on margin of anterior end (Figure 5A). Intestinal caeca without pouches or diverticula, following path as illustrated (Figure 3E). Male copulatory organ curved (Figure 4E). Ovary not coiled or very loosely coiled, not lobed at blind end (Figure 3E). Vaginae running approximately parallel to transverse vitelline ducts (Figure 3E); vaginal pores located lateral to intestinal caeca at level of common genital pore (Figure 3E). Vitellarium not confluent posteriorly (Figure 3E).

Figure 5. Anterior region of *Calicotyle* showing glands. A. *C. kroyeri*; B. *C. urolophi*. Both drawings to same scale. Scale-bar: 500 μ m. Abbreviations: alg, anterolateral gland containing granular secretion; amg, anteromedian gland containing granular secretion; vi, vitellarium.

Live observations

Adult *C. kroyeri* from *Raja* sp. A from Tasmania were attached to the lining of the cloaca by their hamuli. One host was heavily infected with 23 parasites attached to the cloaca. When the haptor was dislodged, the parasite could attach to the host by pulling a plug of host tissue into its mouth.

Remarks

Much attention has been paid to the description of *C. kroyeri* due to the previous controversy surrounding the validity of *Dictyocotyle coeliaca* (see Dawes & Griffiths, 1958). However, studies on the oncomiracidia of both species clearly demonstrated that the two species were distinct (see Kearn, 1970). Lawler (1981) gave a comprehensive account of the numerous host and locality records for *C. kroyeri* and Llewellyn et al. (1984) provided additional information. Our finding of *C. kroyeri* in the Gulf of Mexico and the northwest Atlantic (Florida, USA) are the first records of this species in North America. Our record of *C. kroyeri* from southeastern Tasmania is the first record of this species occurring in the southern hemisphere. The lengths of the body, hamuli, and male copulatory organ of the specimens we obtained from *R. olseni* in the Gulf of Mexico and *Raja* sp. A from Tasmania

are less than those in the type-material we examined from the North Sea. However, the appearance of the internal anatomy is similar to that of *C. kroyeri* and we feel it is unwarranted to propose a new species based on small morphometric differences.

Anterior glands containing granular secretion were observed in live *C. kroyeri* from *Raja* sp. A from Tasmania and some strongly flattened voucher specimens using a phase contrast microscope. This confirms the presence of anterior glands in *Calicotyle* as was previously described in *C. mitsukurii* (see "Remarks" section for *C. mitsukurii*).

***Calicotyle macrocotyle* Cordero, 1944**

(Figures 2F, 3F, 4F)

Synonyms: *Calicotyle megacotyle* Cordero, 1944; *Austrocalicotyle macrocotyle* (Cordero, 1944) Szidat, 1972; *C. rosinae* Kusnetzova, 1970, new synonym.

Type-host: *Sympterygia bonapartei* Müller & Henle (Rajidae).

Type-locality: Montevideo, Uruguay.

Additional records: *Raja brachyurops* Fowler, Patagonian Shelf, Argentina (see Kusnetzova, 1970); *S. bonapartei*, Puerto Quequén, Buenos Aires, Argentina (see Szidat, 1972); Mar del Plata, Buenos Aires, Argentina (see Suriano, 1977); Rio Grande do Sul, Brazil (present study).

Sites: Spiral valve, rectal gland, and cloaca.

Material examined: Holotype, MACN 27.955b; paratype (*C. rosinae*) ZISP 4291; 5 vouchers NHM 1997. 6.12.15-16, USNPC 87191.

Additional data

Hamuli as illustrated (Figure 2F). Hooklets 8 (8–10, n = 6) long, distributed in adult haptor as in *C. kroyeri* (Figure 1A). Intestinal caeca without pouches or diverticula, following path as illustrated (Figure 3F). Male copulatory organ coiled (Figure 4F). Vaginae run posteriorly then turn sharply to run medially parallel with transverse vitelline ducts (Figure 3F); vaginal pores just lateral to intestinal caeca at level of ejaculatory bulb (Figure 3F). Ovary tightly coiled, blind end lobed (Figure 3F). Vitellarium confluent medially in posterior part of body (Figure 3F).

Remarks

Since *C. macrocotyle* was originally described by Cordero (1944), it has been redescribed by Szidat (1972) and Suriano (1977). Szidat (1972) cited the host as *Psammobatis microps*, which is a junior synonym of *S. bonapartei* (see McEachran, 1982). Szidat (1972) illustrated juvenile specimens of *C. macrocotyle*, but unfortunately he made no comment on the development of the male copulatory organ or hamuli, and these details cannot be seen adequately in his illustrations. The vitellarium does not appear to be confluent posteriorly in Szidat's illustrations of the most juvenile specimen. It appears to join later in the developmental sequence, but this needs to be verified.

Kusnetzova (1970), who described *C. rosinae* from the cloaca of *Raja brachyurops* off the coast of Argentina, was apparently unaware of the work by Cordero (1944) because no

reference was made to the South American material. The form of the ovary, vaginae, male copulatory organ, hamuli, and distribution of the vitelline follicles is identical to that of *C. macrocotyle*. Kusnetzova (1970) did not illustrate or describe the ovary looping the right intestinal caecum; however, we observed the looping similar to that in all monocotylids. Close examination of the type-material of *C. rosinae* has shown it to be synonymous with *C. macrocotyle*.

***Calicotyle mitsukurii* Goto, 1894**

(Figures 2G, 3G, 4G)

Type-host: *Rhina* sp. (Rhynchobatidae).

Type-locality: Mitsugahma, Japan.

Site: Cloaca and rectum.

Material examined: None.

Remarks

The description and illustrations by Goto (1894) of *C. mitsukurii* are excellent, but unfortunately type specimens are not available. There is no mention in the original description of how many specimens were collected, but, because no range of measurements was given, we assume that the description was based on a single specimen. The presence of eye-spots or dispersed pigment granules was not noted. The hamulus (Figure 2G) is reproduced from the original illustration, but Goto (1894) noted that this was not completely in profile. The intestinal caeca appear to be smooth following the path illustrated (Figure 3G). The male copulatory organ loops once (Figure 4G), the ovary is only loosely coiled and the blind end is not lobed (Figure 3G). The vaginae run approximately parallel to the transverse vitelline ducts and the vaginal pores are lateral to the intestinal caeca at a level posterior to the common genital pore (Figure 3G). The vitellarium is not confluent posteriorly (Figure 3G). We have discussed above the similarity between *C. mitsukurii* and *C. australis* (see "Remarks" section of *C. australis*) and the possibility that these two species may be synonymous.

Goto (1894) illustrated *C. mitsukurii* with four anterior glands (two on each side of the pharynx) opening at four places on the anterior margin of the head (see figures 1 and 4 of Goto, 1894). This arrangement is identical to that which we observed in live specimens of *C. kroyeri* collected from Tasmania in the present study (see above). Additional material of *C. mitsukurii* is required to examine for eye-spots or dispersed pigment granules and to confirm the validity of *C. australis*. The host record for *C. mitsukurii* must also be confirmed (see Whittington et al., 1989).

***Calicotyle palombi* Euzet & Williams, 1960**

(Figures 2H, 3H, 4H)

Synonym: *Paracalicotyle palombi* (Euzet & Williams, 1960) Szidat, 1970.*Host:* *Mustelus mustelus* (Linnaeus) (= *Mustelus canis*) (Triakidae) (see "Remarks" for discussion on type-host).*Type-locality:* Sète, France, Mediterranean Sea.*Sites:* Cloaca and rectum.*Material examined:* LE (3 specimens on 2 slides).*Additional data*

Hamuli as illustrated (Figure 2H). Hooklets 9 (8–9, n = 3) long distributed in haptor as for *C. kroyeri* (Figure 1A). Eye-spots in form of dispersed pigment granules anterodorsal to pharynx. Intestinal caeca without pouches or diverticula, following path as illustrated (Figure 3H). Male copulatory organ curved (Figure 4H). Ovary loosely coiled with 6–9 lobes at blind end (Figure 3H). Vaginae forming U-shape (Figure 3H); vaginal pores intercaecal at level of common genital pore (Figure 3H). Vitellarium not confluent posteriorly (Figure 3H).

Remarks

Eye-spots were not described by Euzet & Williams (1960), but we observed dispersed pigment granules anterodorsal to the pharynx in the material examined. As noted by Euzet & Williams (1960), *C. palombi* closely resembles *C. stossichi* by the form of the vaginae and the male copulatory organ is very similar (compare Figures 4H and 4M). The hamuli of *C. palombi* are much larger than those of *C. stossichi* (compare Figures 2H and 2L). Although the use of hamulus size can be a questionable character (see Kearn, 1987), the hamuli of what appear to be fully mature *C. palombi* (using the development of the vitellarium as an indicator) are always considerably larger than those of *C. stossichi*. The blind end of the ovary of *C. palombi* also appears more lobed than that of *C. stossichi*, but again this structure may vary depending on the stage of development of the specimen. We agree with Euzet & Williams (1960) that, until intermediate forms are found, *C. palombi* appears to be a valid species.

Euzet & Williams (1960) did not designate a type-host for *C. palombi*, stating only that it inhabits the cloaca and rectum of *M. mustelus* and *M. canis*. Euzet has confirmed that the guide used to identify *M. canis* was incorrect and that *M. canis* is in fact synonymous with *M. mustelus* (personal communication). Euzet & Williams (1960) also included a table listing *Centrophorus uyato* as having a single parasite in the cloaca. While there is no further discussion of this host record in their paper, Lawler (1981) included a footnote stating that, in personal correspondence with Professor Euzet, it was confirmed that the parasite collected from *C. uyato* was *Calicotyle palombi* but that it could have been an "accidental infestation," as they had not found it again in that host.

***Calicotyle quequeni* (Szidat, 1972) Timofeeva, 1985**
(Figures 2I, 3I, 4I)

Synonym: *Austrocalicotyle quequeni* Szidat, 1972.

Host: *Raja castelnaui* Ribeiro (Rajidae).

Type-locality: Puerto Quequén, Buenos Aires, Argentina.

Additional record: *R. castelnaui*, Mar del Plata (see Suriano, 1977).

Sites: Cloaca and rectal gland.

Material examined: None.

Remarks

Type-material of this species could not be obtained for examination, but this species was redescribed by Suriano (1977), and she considered it a valid species. The hamuli are as illustrated (Figure 2I). We could not confirm the presence, in the adult, of either hooklets in the haptor or eye-spots, but eye-spots were reported to be absent in the oncomiradicium (see Suriano, 1977). The intestinal caeca do not appear to have pouches or diverticula and they follow the path as illustrated (Figure 3I). The male copulatory organ is short and straight (Figure 4I). The ovary is tightly coiled and lobed at the blind end (Figure 3I). Szidat (1972) did not illustrate the ovary looping the right intestinal caecum. We have reproduced the full-body drawing of *C. quequeni* from Szidat (1972) but have depicted the ovary as looping the right intestinal caecum as is the case for all monocotylids. The path of the vaginae appear to form a shallow U-shape, but this is difficult to determine from the illustration of Szidat (1972). The vitellarium is confluent in the posterior region of the body. Vitelline cells are present in the median part of the body (see Figure 3I), but extensions leading from the lateral vitelline fields to the median vitelline cells as seen in *C. similis* (Figure 3K) and *C. splendens* (Figure 3L) were not illustrated or described by Szidat (1972) for *C. quequeni*.

***Calicotyle ramsayi* Robinson, 1961**
(Figures 3J, 4J)

Host: *Squalus acanthias* Linnaeus (= *Squalus lebruni*) (Squalidae).

Type-locality: Cook Strait, New Zealand.

Site: Cloaca.

Material examined: Holotype USNPC 39429.

Additional data

Eye-spots present in form of dispersed pigment granules anterodorsal to pharynx. Intestinal caeca with distinct medially directed diverticula following path as illustrated (Figure 3J). Proximal end of male copulatory organ sharply curved (Figure 4J). Ovary loosely coiled, lobed at blind end (Figure 3J). Vaginae forming V-shape (Figure 3J); vaginal pores lateral to intestinal caeca at level of anterior part of ejaculatory bulb (Figure 3J). Vitellarium apparently not confluent posteriorly (Figure 3J).

Remarks

The description of *C. ramsayi* was based on a single specimen. The holotype is in poor condition, and some of the structures are difficult to see. The hamuli were not illustrated by Robinson (1961) or by us because they are not oriented laterally in the holotype. We observed a few hooklets on the margin of the haptor, and we assume that 14 hooklets are present in this species, like in all other monacotylids. Eye-spots, which were not noted by Robinson (1961), appear as dispersed pigment granules. The medial intestinal diverticula appear distinct in *C. ramsayi*. Robinson (1961) illustrated the vitellarium as not confluent in the posterior region of the body, but we could not confirm this in the holotype. We consider *C. ramsayi* a valid species, but additional material is required to verify the features of the hamuli and the form of the vitellarium. This is the only record of a species of *Calicotyle* from a dogfish (Squalidae). We examined four specimens of *S. acanthias* (see Table II) from southeastern Tasmania but found no specimens of *C. ramsayi*.

***Calicotyle similis* (Szidat, 1972) Timofeeva, 1985**

(Figures 2J, 3K, 4K)

Synonym: *Austrocalicotyle similis* Szidat, 1972.

Type-host: *Raja cyclophora* Regan (Rajidae).

Type-locality: Puerto Quequén, Buenos Aires, Argentina.

Additional records: *R. cyclophora* Mar del Plata, Buenos Aires, Argentina (see Suriano, 1977); *R. cyclophora*, southwest Atlantic, off Brazil (present study).

Site: Cloaca.

Material examined: Holotype MACN 27.954a; 1 voucher specimen USNPC 87192.

Additional data

Hamuli as illustrated (Figure 2J). Hooklets 9 (8–10, $n = 7$) long, distributed in haptor as for *C. kroyeri* (Figure 1A). Eye-spots in form of dispersed pigment granules anterodorsal to pharynx. Intestinal caeca without diverticula, following path as illustrated (Figure 3K). Male copulatory organ curved (Figure 4K). Ovary tightly coiled, lobed at blind end (Figure 3K). Vaginae forming U-shape (Figure 3K); vaginal pores opening just lateral to intestinal caeca at level of common genital pore (Figure 3K). Vitellarium confluent posteriorly (Figure 3K); vitelline cells also present in mid-body region with small extensions entering from lateral vitelline fields (Figure 3K).

Remarks

The original description and illustrations of *C. similis* (see Szidat, 1972) were not complete, and the species was redescribed by Suriano (1977). We observed dispersed pigment granules anterodorsal to the pharynx in both the type-specimen and additional material examined here, which neither Szidat (1972) nor Suriano (1977) noted. Szidat (1972) did not illustrate vitelline cells in the mid-body region or the extensions between the lateral and median vitelline fields, but they are present in the voucher specimen. The discovery of *C. similis* off Brazil is a new locality record.

***Calicotyle sjegi* Kusnetzova, 1970**

Type-host: *Raja brachyurops* Fowler (Rajidae).

Type-locality: Patagonian Shelf, Argentina.

Site: Oviduct.

Material examined: None.

Remarks

Kusnetzova (1970) proposed two species, *C. sjegi* for a calicotyline found in the oviduct of *R. brachyurops* and *C. rosinae* for a calicotyline found in the cloaca of the same host. We consider *C. rosinae* synonymous with *C. macrocotyle* (see "Remarks" section for *C. macrocotyle*). Kusnetzova (1970) noted that the form of the vagina and the "internal construction" in *C. sjegi* were identical to those of *C. rosinae*. Kusnetzova (1970) also stated that *C. sjegi* differed from *C. rosinae* only in the smaller body and hamulus size, the location of the central loculus on the haptor, and in the form of the male copulatory organ. Unfortunately, neither the number of specimens examined nor the state of maturity of *C. sjegi* was noted. The specimen identified as *C. sjegi* may be an immature specimen of *C. macrocotyle*, since there are few testicular follicles depicted in the illustration of *C. sjegi* (see Kusnetzova, 1970). If *C. sjegi* represents a juvenile specimen of *C. macrocotyle*, this would account for the smaller hamuli and undeveloped male copulatory organ. Although the vitellarium is not confluent posteriorly, in Kusnetzova's illustration of *C. sjegi*, this may also be because the specimen was immature. Szidat (1972) illustrated the development of *C. macrocotyle* and *C. splendens* from juvenile to adult, and the vitellarium in those species does not appear to join posteriorly until later in the development. Kearn (1987) noted that juvenile specimens of *C. kroyeri* are found in the rectal gland and then migrate to the cloaca where they reach sexual maturity. It is possible that juveniles of *C. macrocotyle* occur in the oviducts and then migrate to the cloaca. Since the identity of *C. sjegi* cannot be confirmed with type-material, we consider *C. sjegi* to be a *species inquirenda* until additional material can be examined.

***Calicotyle splendens* (Szidat, 1970) Timofeeva, 1985**

(Figures 2K, 3L, 4L)

Synonym: *Austrocalicotyle splendens* Szidat, 1970.

Type-host: *Raja agassizi* Müller & Henle.

Type-locality: Puerto Quequén, Buenos Aires Argentina.

Additional records: *R. agassizi*, Mar del Plata, Buenos Aires, Argentina (see Suriano, 1977); *R. agassizi*, southwest Atlantic (present study).

Site: Cloaca.

Material examined: Holotype MACN 27.952c; 1 voucher specimen NHM 1997.6.12.17.

Additional data

Hamuli as illustrated (Figure 2K). Hooklets 9 (8–10, $n = 9$) long, distributed in haptor as for *C. kroyeri* (Figure 1A). Eye-spots in form of dispersed pigment granules anterodorsal to pharynx. Intestinal caeca with short medial and lateral pouches, following path as illustrated (Figure 3L). Male copulatory organ short and straight (Figure 4L). Ovary tightly coiled, lobed at blind end (Figure 3L). Vaginae broadly U-shaped (Figure 3L); vaginal pores just lateral to intestinal caeca at level of common genital pore. Vitellarium confluent posteriorly (Figure 3L); vitelline cells also present in mid-body region with small extensions entering from lateral vitelline fields.

Remarks

Calicotyle splendens was described by Szidat (1970b) in his genus *Austrocalicotyle*, but the generic definition for that taxon was not given until later (see Szidat, 1972). Suriano (1977) redescribed *C. splendens* and confirmed the validity of the species. We observed eye-spots, in the form of dispersed pigment granules, which were not noted by either Szidat (1972) or Suriano (1977), in the type and additional material examined in the present study. Szidat (1970b) did not illustrate vitelline cells in the mid-body region or the extensions between the lateral and median vitelline fields, but they are present in the voucher specimen. *C. splendens* is similar to *C. quequeni*, both species having a short straight male copulatory organ (compare Figures 4L and 4I) and a vitellarium which is confluent medially in the posterior region of the body. *C. splendens* can be differentiated from *C. quequeni* by the path of the intestinal caeca and vaginae and by the presence of small lateral and medial gut diverticula (compare Figures 3L to 3I). The discovery of *C. splendens* on *R. agassizi* in the southwest Atlantic represents a new locality record.

***Calicotyle stossichi* Braun, 1899**

(Figures 2L, 3M, 4M)

Synonyms: *Calicotyle* (*Calicotylides*) *stossichi* (Braun, 1899) Nybelin, 1941; *Paracalicotyle stossichi* (Braun, 1899) Szidat, 1970.

Type-host: *Mustelus mustelus* (Linnaeus) (Triakidae).

Locality: Berlin Aquarium, Rovigno, Yugoslavia.

Additional records: *M. antarcticus* Günther, southeast coast of Tasmania (present study); *M. canis* (Mitchill), Sète, France, Mediterranean Sea (see Euzet & Williams, 1960); *M. mustelus*, Angola, Luanda, west African Coast (present study); *M. norrisi* Springer, northeast Gulf of Mexico (present study) (Triakidae).

Sites: Cloaca, rectum, and rectal gland.

Material examined: LE (10 specimens on 6 slides); voucher specimens NHM 1997.6.12.22-23, USNPC 87193, 87194.

Additional data

Hamuli as illustrated (Figure 2L). Hooklets 9 (8–10, $n = 6$) long, distributed in haptor as for *C. kroyeri* (Figure 1A). Eye-spots in form of dispersed pigment granules anterodorsal to

pharynx. Intestinal caeca without pouches or diverticula, following path as illustrated (Figure 3M). Male copulatory organ curved (Figure 4M). Ovary coiled, lobed at blind end (Figure 3M). Vaginae forming U-shape (Figure 3M); vaginal pores overlying intestinal caeca at level of common genital pore (Figure 3M). Vitellarium extending from anterior part of pharynx to posterior portion of body, not confluent posteriorly (Figure 3M).

Remarks

Calicotyle stossichi, which was originally described by Braun (1899), was redescribed by Euzet & Williams (1960). We observed dispersed pigment granules anterodorsal to the pharynx which were not previously described by these authors. The similarity between *C. stossichi* and *C. palombi* is discussed in the "Remarks" section for *C. palombi*. *Mustelus canis* is an incorrect host identification (see "Remarks" section of *C. palombi*). *C. stossichi* in *M. norrisi* from the Gulf of Mexico and in *M. antarcticus* from Tasmania represent both new host and locality records. *C. stossichi* in *M. mustelus* from Angola represents a new locality record.

***Calicotyle urolophi* Chisholm, Beverley-Burton & Last, 1991**

(Figures 2M, 3N, 4N, 5B)

Type-host: *Urolophus cruciatus* (Lacépède) (Urolophidae).

Type-locality: Coastal waters off southeastern Tasmania.

Additional records: *U. bucculentus* Macleay southeastern Tasmania (see Chisholm et al., 1991); *U. paucimaculatus* Dixon southeastern Tasmania (see Chisholm et al., 1991 and present study) (Urolophidae).

Site: Cloaca.

Material examined: Four paratypes NHM 1990. 8.23.1-7 (2 slides), 1990 8.23.8-9 (1 slide), 1990.8.23.10 (1 slide), 1 live specimen.

Additional data

Hamuli as illustrated (Figure 2M). Hooklets 9 (8–10, $n = 5$) long, distributed in haptor as for *C. kroyeri* (Figure 1A). Live specimen with single diffuse anteromedian gland containing granular secretion; numerous ducts from anteromedian gland run anteriorly and open on anterior margin of head (Figure 5B). One pair of anterolateral glands containing granular secretion located on each side of anterior end; single duct leads from each anterolateral gland to open on lateral margin as illustrated (Figure 5B). Eye-spots in form of dispersed pigment granules anterodorsal to pharynx. Intestinal caeca without pouches or diverticula, following path as illustrated (Figure 3N). Male copulatory organ sharply curved proximally (Figure 4N). Ovary loosely coiled, not lobed at blind end. Vagina running parallel with transverse vitelline duct (Figure 3N); vaginal pores lateral to intestinal caeca at level of common genital pore (Figure 3N). Vitellarium not confluent posteriorly (Figure 3N).

Remarks

Chisholm et al. (1991) stated that hooklets were absent in the haptor of *C. urolophi*, but we observed 14 hooklets distributed in the haptor as is characteristic for the genus (Figure 1A). Examination of a single live specimen revealed anterior glands not described previously.

Dictyocotyle Nybelin, 1941

Diagnosis

Monocotylidae: Calicotylinae. Haptor with numerous irregular loculi. Hamuli absent. Eye-spots absent. Intestinal caeca with short lateral and medial pouches. Vitellarium not confluent in posterior region of body. Egg ovoid longitudinally, pointed at each end but having three flattened sides, appearing triangular in cross-section (see figure 15a,b of Kearn, 1986). Parasitic on wall of body cavity of elasmobranchs.

Dictyocotyle coeliaca Nybelin, 1941

(Figures 1B, 6)

Type-host: *Raja radiata* Donovan (Rajidae).

Type-locality: Trondheim, Norway.

Additional records: See Lawler (1981).

Site: Wall of body cavity.

Material examined: One specimen NHM 1976.1.6.6.; 2 specimens on 1 slide NHM 1985.10.17.78-103; 4 specimens on 1 slide NHM 1989.9.4.6; 2 specimens on 1 slide NHM 1989.8.31.19. Two specimens cleared in Hoyer's mountant.

Additional data

Hooklets 9 (8–10, n = 5) long, distributed in muscular rim of haptor as illustrated (Figure 1B). Intestinal caeca with small lateral and medial pouches. Male copulatory organ long with irregular coils; slightly enlarged distally (Figure 6). Ovary loosely coiled, with multiclavate lobes at blind end. Vaginae forming V-shape; vaginal pores opening lateral to intestinal caeca at level of ejaculatory bulb. Vitellarium not confluent posteriorly.

Remarks

Hooklets which were previously believed to be absent or not present in the full complement of 14 were found to be present. No hamuli were found. The male copulatory organ is slightly enlarged distally (Figure 6), which has not been noted previously.

Figure 6. Male copulatory organ of *Dictyocotyle coeliaca* showing swelling at distal end (d).
Scale-bar: 200 μ m.

Key to species of the Calicotylinae

1. Haptor with numerous irregular loculi; hamuli absent (*Dictyocotyle*) *D. coeliaca*
—Haptor with one central and seven peripheral loculi, hamuli present (*Calicotyle*) 2
2. Vitellarium confluent medially in posterior portion of body (Figures 3F, I, K, L) 3
—Vitellarium separate in posterior portion of body (e.g. Figure 3A) 6
3. Male copulatory organ short, straight tube (Figures 4I, L) 4
—Male copulatory organ longer, curved or coiled (Figures 4F, K) 5
4. Intestinal caeca with short lateral and medial pouches (Figure 3L); vaginae broadly U-shaped (Figure 3L) *C. splendens*
—Intestinal caeca lacking diverticula; vaginae U-shaped (Figure 3I) *C. quequeni*
5. Intestinal caeca without pouches or diverticula, with two prominent approximately 90° curves (Figure 3F); male copulatory organ looping twice (Figure 4F) *C. macrocotyle*
—Intestinal caeca with single approximately 45° medially directed curve distally (Figure 3K); male copulatory organ looping once (Figure 4K) *C. similis*
6. Vaginae U- or V-shaped; ovary coiled, lobed at blind end 7
—Vaginae approximately parallel to transverse vitelline duct, with pores opening lateral to intestinal caeca; ovary loosely coiled, not lobed at blind end 12

7. Male copulatory organ long, with at least three irregular coils (Figures 4A,C) 8
 —Male copulatory organ shorter, curved or looping only once 9
8. Intestinal caeca with small lateral and medial pouches; vaginae not looped (Figure 3A)
 C. affinis
 —Intestinal caeca without pouches or diverticula; vaginae with single loop (Figure 3C)
 C. australiensis
9. Male copulatory organ with one coil (Figure 4B) *C. asterii*
 —Male copulatory organ curved, not coiled 10
10. Intestinal caeca with five to six large medial diverticula (Figure 3J) *C. ramsayi*
 —Intestinal caeca without lateral and medial diverticula 11
11. Hamuli greatly reduced approximately 1/5 length of posterior radial septa (Figure 3M)
 C. stossichi
 —Hamuli approximately 1/2 length of posterior radial septa (Figure 3H) *C. palombi*
12. Proximal end of male copulatory organ arising from anterior part of ejaculatory bulb
 C. urolophi
 —Proximal end of male copulatory organ arising from posterior part of ejaculatory bulb . . 13
13. Male copulatory organ 9-shaped (Figure 4E) *C. kroyeri*
 —Male copulatory organ with single loop (Figure 4D, G) 14
14. Vaginal pores at level of common genital pore *C. australis*
 —Vaginal pores at level posterior to common genital pore *C. mitsukurii*

Discussion

The first reported monocotylid, *Calicotyle kroyeri*, was described from the rectum of *Raja radiata* by Diesing (1850). Sixteen additional species have since been described and the classification within Calicotylinae, which we outline below, has been modified numerous times. Nybelin (1941) separated *Calicotyle* into three subgenera: *Calicotyle (sensu stricto)* for those species with vaginae approximately parallel to the transverse vitelline duct and their body widest posteriorly (*C. australis*, *C. kroyeri*, and *C. mitsukurii*); *Calicotylides* for those species with U-shaped vaginae and their body widest near the mid-point (*C. affinis* and *C. stossichi*); and *Gymnocalicotyle* for the single species *C. inermis* which lacks hamuli. Nybelin's subgenera were generally ignored by subsequent workers, but Yamaguti (1963) raised *Gymnocalicotyle* to generic status without comment. Timofeeva (1985) and Chisholm et al. (1995) rejected Nybelin's subgenera *Calicotyle* and *Calicotylides*, and, because type-material of "*Gymnocalicotyle inermis*" was unavailable, Chisholm et al. (1995) did not consider *Gymnocalicotyle* a distinct taxon. We concur with these decisions.

Szidat (1970a) erected *Paracalicotyle* to include members of the Calicotylinae whose body was parallel-sided, had a relatively small haptor, and had been collected from the cloaca, rectum, or rectal gland of *Mustelus* spp. He designated *C. stossichi* as the type-species of

Paracalicotyle and included *C. palombi* and a new species, *P. asterii*, in the genus. Suriano (1977), who reexamined the South American calicotylines, rejected *Paracalicotyle* and moved *P. asterii* to *Calicotyle*. She failed to mention the fate of the other members of the genus.

Szidat (1970b) erected *Austrocalicotyle* to accommodate the newly described *A. splendens*, but the generic diagnosis of this new genus did not appear until two years later (see Szidat, 1972). Szidat (1972) designated *C. macrocotyle* as the type-species of the genus and included in it *A. splendens*, and two new species, *A. quequeni* and *A. similis*. He distinguished *Austrocalicotyle* from *Calicotyle* and *Paracalicotyle* by the presence of a transverse band of vitelline follicles in the posterior region of the body and an ovary which was lobed at the blind end. Suriano (1977) redescribed these species and accepted the validity of the genus. Suriano (1977) also described the oncomiracidium of *A. quequeni* and compared it to the only other *Calicotyle* species with a described oncomiracidium, *C. kroyeri* (see Kearn, 1970). Suriano (1977) noted that the larva of *A. quequeni* lacked pigment-shielded eyespots and possessed one fewer pair of flame-cells than the oncomiracidium of *C. kroyeri*, and interpreted those features as further support for the validity of *Austrocalicotyle*.

Because Szidat's diagnoses for *Paracalicotyle* and *Austrocalicotyle* were unclear, Timofeeva (1985) abandoned these genera in her revision of the Monocotylidae and transferred the species back to *Calicotyle*. This decision was followed by subsequent workers, and we concur. We feel it is unnecessary and inappropriate to erect new genera based on small differences in the shape of the body, the distribution of the vitellarium, and the lobation of the blind end of the ovary, especially since it is unclear how these characters vary with age and fixation.

Our revision shows clearly that the taxonomy of *Calicotyle* at the species level is in a state of great confusion. This confusion is exacerbated by the lack of knowledge regarding the development of individuals from juvenile to adult, the site of development within the host, and how other factors such as temperature, host, and site in the host affect the development and final size of the parasites. As for most monogeneans, calicotylines have been distinguished from each other primarily by the morphology of the hamuli and the male copulatory organ, both of which vary within a single species. For example, Kearn (1987) (for *C. kroyeri*) and Rohde et al. (1992) (for *C. australiensis*) have shown that the form of the hamuli differ considerably throughout the development of these species. Furthermore, Whittington et al. (1989) (for *C. australis*) and Chisholm et al. (1991) (for *C. urolophi*) both illustrated and commented on the fact that the morphology of the hamuli can vary between what they believed to be mature adult specimens of the same species. The male copulatory organ of *C. australiensis* was shown to differ in length and shape depending on the state of development of the individual (see Rohde et al., 1992). The male copulatory organ of smaller individuals of *C. australiensis* has a single loop and as the parasite matures, the length and number of loops increase. Changes in the morphology of the male copulatory organ with parasite age have also been shown for other monocotylids including *Dendromonocotyle ardea* Chisholm & Whittington, 1995 (see Chisholm & Whittington, 1995) and *Heterocotyle granulata* Young, 1967 (see Chisholm & Whittington, 1996). Kearn (1987) also has demonstrated that the site that *C. kroyeri* inhabits within the host differs as the parasite matures. Juvenile *C. kroyeri* which appear to live in the rectal gland of their host migrate to

the cloaca where they reach sexual maturity. It is unknown if this is also the case for other species of *Calicotyle*.

Choosing suitable characters to resolve relationships between the species in the Calicotylinae is difficult. For reasons outlined above, characters involving the shape of the hamuli and the male copulatory organ are questionable, since it is unclear how factors such as development and temperature influence them. Other characters involving the small differences in shape and path of the vaginae and the intestinal caeca can vary depending on the method of specimen preparation. Presence and absence of other features such as eyespots and anterior glands are also dubious, since these are often not visible in fixed material. More material of *C. inermis*, *C. mitsukurii*, and *C. ramsayi* is required to verify original descriptions. More information regarding interspecific variation in the morphology of the hamuli and the male copulatory organ as well as the effects of fixation on the form and path of the vaginae and intestinal caeca is needed before the phylogenetic relationships between members of the Calicotylinae can be examined.

We have noted in some of the "Remarks" sections above that a number of *Calicotyle* spp. currently regarded as valid may be synonymous. For example, *C. affinis* from *Chimaera monstrosa* in the North Sea is very similar to *Calicotyle australiensis* described from *Chimaera* sp. II off the eastern coast of Australia. The hooklets in the haptor are prominent in both species, the path of the intestinal caeca is similar, the vaginae are U-shaped in each species, and the male copulatory organ is long and coiled. When *C. australiensis* was proposed, it was differentiated from *C. affinis* by the slightly longer coiled male copulatory organ and the ratio of the guard to total hamulus length (see Rohde et al., 1992). However, these small differences could be due to other unknown factors such as temperature. Rohde et al. (1992) noted the close relationship between *C. affinis* and *C. australiensis* and stated that the discovery of intermediate forms could necessitate their synonymy. It is unclear whether the three specimens of a species of *Calicotyle* found by Rohde et al. (1992) in the rectum of *Rhinochimaera pacifica* (Mitsukuri) are a new species or an intermediate form of *C. australiensis*. We also believe that *C. australis* may be synonymous with *C. mitsukurii*, since these species differ only in the length of the male copulatory organ and the level at which the vaginal pores open. All other characters such as the path of the intestinal caeca, the path of the vaginae and the form of the vitellarium appear similar. Since type-material of *C. mitsukurii* is not available and the original description may have been based on a single specimen, additional material is required. The validity of *C. palombi* is also questionable. It is found in the cloaca and rectum of *Mustelus mustelus* and *M. canis* and is closely related to *C. stossichi* from the rectal gland of the same hosts. The form of the male copulatory organ, path of the intestinal caeca, and vaginae are very similar in these two species. The only significant difference between these taxa is the size of the hamuli, which are much smaller in adult specimens of *C. stossichi*. Euzet & Williams (1960) noted that *C. palombi* was never found in the rectal gland. It is possible that juvenile *C. stossichi* are located in the rectal gland and then migrate to the cloaca and rectum, where they continue to grow until they reach sexual maturity (see Kearn, 1987). However, using the development of the vitellarium as an indicator, all specimens we examined of *C. stossichi* and *C. palombi* appeared to be fully mature. It is unknown whether the hamuli continue to grow after the parasite

reaches sexual maturity. Euzet & Williams (1960) stated that *C. palombi* could be synonymized with *C. stossichi* if intermediate forms were found. Molecular studies of the *Calicotyle* spp. discussed above could help to resolve these taxonomic questions.

Previously, there has been no consensus regarding the presence or absence of hooklets in the haptors of adult members of the Calicotylinae. Chisholm et al. (1991) stated that hooklets were absent in their revised diagnosis of the Calicotylinae, but we confirmed the presence of 14 hooklets in most specimens of *Calicotyle* and *Dictyocotyle* that we examined. The distribution of hooklets in all *Calicotyle* spp. we examined (except *C. ramsayi* in which we could not see the full complement of hooklets) corresponds to that shown by Bychowsky (1957; figure 274) for *C. kroyeri*. In *Calicotyle* spp. hooklet pairs I and II lie between the hamuli and the remaining five pairs are distributed as illustrated (Figure 1A). In *C. affinis* and *C. australiensis* the hooklets are comparatively large (12–15 μm) and relatively easy to find in the muscular haptor, but in the rest of the species the hooklets are smaller (8–10 μm) and easy to overlook. Furthermore, they are embedded in the thick muscular edge of the haptor which is often curled inwards (Figure 1A). Therefore what appears to be the edge of the haptor in whole-mounts is, in fact, not (see Figure 1A), and we suspect this is why they have been overlooked in the past.

In *Dictyocotyle*, the haptor is often indented at the points where the hooklets are embedded making them easier to locate (see Figure 1B). They are distributed approximately evenly around the edge of the haptor. We also made a concerted effort to locate the small sclerites which Kearn (1970) identified as hamuli in the haptor of the oncomiracidium of *D. coeliaca*. We examined adult haptors which had been cleared in Hoyer's and, although the 14 hooklets were clearly visible, there was no sign of any hamuli. We searched the entire haptor for hamuli but concentrated our efforts in the regions between hooklet pairs II and III. It appears that the vestigial sclerites which Kearn (1970) observed in larvae either develop no further or are lost in the adult.

The presence and absence of eye-spots is also difficult to determine in many *Calicotyle* spp. Eye-spots have only been recorded previously in *C. australis* (see Whittington et al., 1989), *C. kroyeri* (see Bychowsky, 1957), and *C. urolophi* (see Chisholm et al., 1991). We observed what we believed to be dispersed pigment granules anterodorsal to the pharynx in most of the species we examined, but they are very difficult to see in most preserved material. Because we could not obtain the type-material of *C. quequeni*, we could not confirm the presence or absence of eye-spots in this species, but Suriano (1977), who described the oncomiracidium of *C. quequeni*, stated that eye-spots are absent in the larva. It is unlikely that she would have overlooked these prominent structures, but this should be verified. Goto (1894) made no mention of the presence of eye-spots in *C. mitsukurii*. Fresh material of many of these species of *Calicotyle* is required to resolve these questions.

Goto (1894) described *C. mitsukurii* with one pair of "sticky glands" on each side of the pharynx and a single duct leading from each gland and opening on the anterior margin of the head (see figures 1 and 4 in Goto, 1894). Until the present study, this was the only record of anterior glands in an adult specimen of the genus. However, we observed anterior glands in live and in heavily flattened, fixed specimens of *C. kroyeri* and *C. urolophi*. The arrangement of the anterior glands in *C. kroyeri* is similar to that noted by Goto (1894) for *C. mitsukurii*. In *C. kroyeri*, a pair of anterolateral glands are present on each side level

with the pharynx (Figure 5A). These glands contain granular secretion and a single duct leads anteriorly from each gland and opens on the anterior margin (Figure 5A). It is difficult to see these glands in fixed material because they are in close proximity to the vitellarium (Figure 5A) but in live worms these glands are distinct. In *C. urolophi*, the arrangement of anterior glands is different. We observed a diffuse anteromedian gland containing granular secretion (Figure 5B). Numerous small ducts leading from this gland open on the anterior margin. This gland and its associated ducts are difficult to see in live material and they are not visible in fixed material. We did not observe an anteromedian gland in live, adult *C. kroyeri* and it was not described in *C. mitsukurii*, but Kearn (1970) illustrated anteromedian gland-cells in the larva of *C. kroyeri*. Furthermore, the anterolateral glands appear to be located more anteriorly in *C. urolophi* than in either *C. kroyeri* or *C. mitsukurii* (compare Figures 5A and 5B). However, since only a single live specimen of *C. urolophi* was observed, more material should be examined to confirm this arrangement.

In addition to anterior glands with granular secretion, anterior glands containing needle-like secretion have been described in many other monocotyliids (see Chisholm & Whittington, 1996, 1997; Cribb et al., in press). No glands with needle-like secretion were seen in any of the live *Calicotyle* spp. we examined. However, glands-cells with needle-like secretion were illustrated on either side of the pharynx in the oncomiracidia of *C. kroyeri* and *D. coeliaca* (see Kearn, 1970). Chisholm & Whittington (1996) observed lateral gland-cells containing needle-like secretion in the larva of *Heterocotyle capricornensis* Chisholm & Whittington, 1996. They demonstrated that glands with needle-like secretion are also found in the adult and they discussed the possible homology of these structures. A similar arrangement has been shown for some members of *Neoheterocotyle* Hargis, 1955 (see Chisholm & Whittington, 1997). Therefore, because glands with needle-like secretion are present in the oncomiracidium of *C. kroyeri* and *D. coeliaca*, it is possible that they are also present in adult calicotylines. Examination of more live material or a transmission electron microscopy study such as that done by Cribb et al. (in press) for another monocotyliid, *Monocotyle spiremae* Measures, Beverley-Burton & Williams, 1990, may confirm their presence or absence in *Calicotyle*. Kearn (1970) also noted an additional pair of glands in the posterior body region near the haptor in the larva of *C. kroyeri*, but we did not observe these in the adults. The posterior glands, if present, are probably obscured by the extensive, darkly colored vitellarium.

Chisholm & Whittington (in press) discuss the morphology of the monocotyliid haptor and relate the presence of accessory structures on the haptor to the microhabitat of the parasite on/in the host. They postulate that because the marginal valve is absent and the edge of the haptor is thick in species of *Calicotyle*, the haptor cannot maintain a good seal for suction. However, because species of *Calicotyle* live in the cloaca, rectum, or rectal gland of their hosts, they are not exposed to the same turbulence as monocotyliids, which live on the skin or gills. Therefore, Chisholm & Whittington (in press) considered that attachment by suction of the haptor may not be as important for internal monocotyliids and that, instead, *Calicotyle* spp., which live in the cloaca, rely more on their large hamuli for attachment. This is supported by our observations in the present study. *C. kroyeri* from *Raja* sp. A was found firmly attached to the cloacal lining by their hamuli. A fine needle could be slid easily under the anterior part of the haptor, but the posterior portion remained firmly

attached by the blades of the hamuli, piercing the cloacal lining. However, the host we examined for parasite attachment was infected with 23 worms and it is not known how this high intensity of infection might alter their mode of attachment. It is also not known whether the mode of attachment changes when the host dies, since species of *Calicotyle* are known to migrate out of the cloaca and onto the ventral body surface with the death of their host (see Kearn, 1987; Whittington et al., 1989). In contrast, we observed that specimens of *C. stossichi*, which inhabit the rectal gland and have very small hamuli, were attached weakly to the rectal gland tissue, but we could not determine if the blades of the hamuli pierced the tissue. It has been suggested that those parasites which inhabit the rectal gland, where turbulence is minimal, have smaller hamuli perhaps to prevent damage to the delicate tissue (see Timofeeva, 1985; Chisholm & Whittington, in press). Our limited observations in the present study suggest that the hamuli may be vestigial in those calicotylines that live in the rectal gland, since they do not appear to play a significant role in attachment. However, this requires further investigation. We also observed that when the haptor of *C. kroyeri* was detached from host tissue, the parasite could attach firmly to the host by the anterior end by pulling a plug of tissue into its mouth.

Six previously described species of *Calicotyle* were collected in this study. *C. asterii* previously known only from *Mustelus schmitti* off Argentina is recorded in *M. norrisi* from the Gulf of Mexico. We record five new hosts for *C. kroyeri*: *Anacanthobatis folirostris*, *Dactylobatis armatus*, *Raja olseni* from the Gulf of Mexico, *R. garmani* from the northwest Atlantic off Virginia, and *Raja* sp. A from the southeast coast of Tasmania near Hobart. These are the first records of *C. kroyeri* outside the northeast Atlantic and the first record of *C. kroyeri* in the southern hemisphere. It would be interesting in the future to compare specimens of *C. kroyeri* from the northern hemisphere with those in the southern hemisphere using molecular techniques. *C. macrocotyle* which was previously only known from Argentina is found on a new host, *S. bonapartei*, from Brazil. We record new localities for *C. similis* (Brazil) and *C. splendens* (southwest Atlantic) from their respective type-hosts. *C. stossichi*, found previously in the Mediterranean Sea, is recorded off Angola, Africa, from the type-host, from the Gulf of Mexico on *Mustelus norrisi*, and in *M. antarcticus* from southeastern Tasmania.

Lawler (1981), Llewellyn et al. (1984), and Chisholm et al. (1991) noted the lack of host-specificity in *C. kroyeri* and *C. urolophi*, respectively. Results from the present study show that the lack of host-specificity appears to span to other members of the subfamily. As noted by Chisholm & Whittington (1996), other monocotylids may also lack host-specificity and have a wide geographical range. *Heterocotyle chinensis* Timofeeva, 1983, which was originally described from the gills of *Dasyatis akajei* in the Yellow Sea, was recorded by Chisholm & Whittington (1996) from two new species of hosts in Moreton Bay, Queensland, Australia. Similarly, *H. pastinacae* Scott, 1904 has been recorded from the North and the Adriatic Seas. *Merizocotyle pseudodasybatis* (Hargis, 1955) Chisholm, Wheeler & Beverley-Burton 1995, which was originally described from *Aetobatus narinari* from the Gulf of Mexico off Florida (see Hargis, 1955), has been found on the same species of host in Moreton Bay, Australia (see Whittington, 1990).

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