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IDENTITY OF *DIPHYLLOBOTHRIUM* SPP. (CESTODA: DIPHYLLOBOTHRIIDAE) FROM SEA LIONS AND PEOPLE ALONG THE PACIFIC COAST OF SOUTH AMERICA

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ABSTRACT: Host specificity evidently is not expressed by various species of *Diphyllobothrium* that occur typically in marine mammals, and people become infected occasionally when dietary customs favor ingestion of plerocercoids. This report mainly concerns 2 species, *Diphyllobothrium pacificum* and *Diphyllobothrium arctocephalinum*, for which sea lions (Otariidae) are final hosts. The taxonomic status of those cestodes has not been clearly discernible because of misinterpretation of relationships; complex synonymies have resulted from misidentification(s). Stiles and Hassall in 1899 obtained, but did not describe, cestodes from the northern fur seal (Otariidae; Pribilof Islands). That taxon was subsequently studied by several investigators, with diverse conclusions. The valid designation is *D. pacificum* (Nybelin, 1931). In 1937, Johnston and Drummond described separately 2 conspecific cestodes from sea lions near Australia, designated *D. arctocephalinum* and *Diphyllobothrium arctocephali*. Both names have been listed incorrectly as synonyms of *D. pacificum*.

Species of *Diphyllobothrium* Cobbold, 1858 occur commonly in fur seals and sea lions (Otariidae). Those mammals inhabit the Pacific coasts of North America and South America from the sub-Arctic to the sub-Antarctic, and are also present on the coasts of Australia and New Zealand and the Atlantic coast of southern Africa. The geographic ranges of the 14 recognized species of otariids have been defined by Jefferson et al. (1993). Cestodes typically in those mammals have been recorded also from people along the Pacific coasts of South America and Japan.

For some years, we have been aware of uncertainty about the correct designation of a cestode, now usually called *Diphyllobothrium pacificum* (Nybelin, 1931), which occurs commonly in the northern fur seal, *Callorhinus ursinus* L., in the northern Pacific region. A cestode from that mammal was first recorded, but not described, by Stiles and Hassall (1899), by whom it was designated *Bothriocephalus* sp. It was subsequently described as *Adenocephalus septentrionalis* Nybelin, 1931. Baer (1967) reported *D. pacificum* from humans residing in coastal northern Peru, and the applicability of *Diphyllobothrium septentrionalis* (Nybelin, 1931) to the cestode from the northern fur seal remained uncertain. We attempt to resolve some taxonomic problems involving those cestodes in the present report.

We first review the status of the taxon *Clestobothrium glaciale* Cholodkovsky (Kholodkovskii), 1915, described on the basis of a 10-cm fragment of strobila obtained from the intestine of a northern fur seal killed near the coast of Kamchatka, in order to exclude that cestode from further consideration herein. Cholodkovsky (1915) assigned that specimen to *Clestobothrium* Lühe, 1899, which is characterized in part by having the opening of the genital ducts on the dorsal surface of the proglottid, and that of the uterus on the ventral surface (cf. Bray et al., 1994). Cholodkovsky concluded that the cestode represented an independent species differing from *Clestobothrium crassiceps* (Rudolphi, 1819), reported from fishes in the Atlantic.

Nybelin (1931) considered that Cholodkovsky's description was incomplete and that the location of the genital openings had become confused by reversal of the sections. He concluded that the cestode was a species of Diphyllobothriidae, but that Cholodkovsky's description could not permit the determination that *C. glaciale* is identical with the *Bothriocephalus* sp. of Stiles and Hassall (1899). On that basis, Nybelin provided a new name for the latter, which he described as *A. septentrionalis* Nybelin, 1931. Markowski (1952) transferred *C. glaciale* to *Diphyllobothrium*, concluding that it was identical with the *Bothriocephalus* sp. reported by Stiles and Hassall (1899). As its synonym, Markowski included *Adenocephalus pacificus* Nybelin, 1931 and *A. septentrionalis* Nybelin, 1931. The morphological characters considered by Markowski to justify transfer of *C. glaciale* to *Diphyllobothrium* are questionable. Moreover, Cholodkovsky's (1915) Figure 5 appears to show a large, sinus-like cavity on the ventral surface of the proglottid into which the uterus opens in species of *Clestobothrium*. The cestode reported by Stiles and Hassall (1899) represents another species, which is indicated also by its much larger egg (average = 61 × 42 μ for *A. septentrionalis*, as compared with a length of 40 μ for that of *C. glaciale* [Nybelin, 1931] [Cholodkovsky, 1915]). Because Cholodkovsky's collection in St. Petersburg was destroyed during World War II (Baer, 1967), re-examination of his material is not possible.

We conclude that the cestode described by Cholodkovsky was indeed a species of Bothriocephalidae, and consider that the fragment of strobila studied by him was probably the remains of a cestode from a fish that had been recently eaten by the seal. That probability is supported by the finding by Margolis and Dailey (1972) of *Abothrium gadi* van Beneden, 1871, a typical cestode of cod, in the intestine of a Steller sea lion, *Eumetopias jubatus* (Schreber), near the coast of British Columbia, Canada. In agreement with Deliamure et al. (1985), we consider *C. glaciale* Cholodkovsky, 1915 to be species inquirenda and clearly not a species of *Diphyllobothrium*.

The first recorded information regarding the helminth fauna of the northern fur seal was that of Stiles and Hassall (1899), based on material collected on the Pribilof Islands (Bering Sea) by F. Lucas in 1896. They reported cestodes of a single species that they assigned to *Bothriocephalus* Rudolphi, 1808. Unable at the time to undertake a study of the cestodes, Stiles and Hassall provided figures of entire specimens, much contracted, and deposited 4 lots of them in the U.S. National Parasite Collection.

Nybelin (1931) described a diphyllobothriid from a sea lion, *Arctocephalus australis* (Zimmermann) (=*Arctocephalus philippi* (Peters)), collected in the Juan Fernandez Islands during the Swedish Expedition of 1916–1917. For that cestode, Nybelin

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established the genus *Adenocephalus*, and designated the species *A. pacificus* Nybelin, 1931, although he recognized that it closely resembled the cestode reported by Stiles and Hassall (1899) from the northern fur seal, having studied some ''. . .junge Exemplare von 23–35 mm Länge. . .'' and more relaxed specimens, 50–66 mm in length. Nonetheless, Nybelin described the Stiles and Hassall cestode as *A. septentrionalis* Nybelin, 1931, distinguishing it from *A. pacificus* on the basis of its smaller strobila and some minor morphological differences, none of which appears to be of taxonomic significance; he also attached importance to the occurrence of the 2 taxa in hosts of different species that were widely separated geographically (Nybelin, 1931). Thereafter, the identity of the common cestode in the northern fur seal has been subject to diverse opinions.

Afanas'ev (1941) studied cestodes from 10 of 16 fur seals from the Komandorskie Islands (Bering Sea). He identified them as *A. septentrionalis*, but recognized that they were morphologically very similar to *A. pacificus*. Wardle et al. (1947) examined cestodes from St. Paul Island (Pribilof Islands) fur seals, retaining Nybelin's (1931) *Adenocephalus* for *A. pacificus*, but transferring *A. septentrionalis* to *Cordicephalus* Wardle et al., 1947, and placing it in synonymy with *Diphyllobothrium arctocephalinus* Johnston, 1937, from *Arctocephalus forsteri* (Lesson) in Australia. As synonyms of *Cordicephalus arctocephalinus*, they also listed *Cordicephalus arctocephali* (Drummond, 1937), *C. glaciale* (Cholodkovsky, 1915), and *Cordicephalus grandis* (R. Blanchard, 1894). Diphyllobothriids from the Pribilof Island seals were also examined by Stunkard (1948), who compared them with the cestodes reported by Stiles and Hassall (1899) and concluded that they were conspecific. Stunkard rejected *Cordicephalus* as a taxon and transferred the cestode from the northern fur seal to *Diphyllobothrium* Cobbold, 1858, not assigning a specific name, but giving designation only as ''Species No. 2,'' because he considered that discrimination of species in *Diphyllobothrium* ''presents a particularly difficult problem'' (Stunkard, 1948). By that time, however, differential characters for cestodes in the genus had been well defined by Russian helminthologists, but their literature was generally unknown in North America. (From 1558 to 1972, 2,173 publications on *Diphyllobothrium* and diphyllobothriasis had appeared in the Russian literature, as compared with 1,015 in other languages [Rozenberg, 1977].) Yamaguti (1951) obtained cestodes from the northern fur seal from Onahama, Hukusima Prefecture, in Japan, to which he assigned the name *A. pacificus*. For cestodes from seals from St. George Island (Pribilof Islands), Markowski (1952) applied the name *Diphyllobothrium glaciale* (Cholodkovsky, 1915), listing as synonyms *Bothriocephalus* of Stiles and Hassall (1899), *C. glaciale* Cholodkovsky, 1915, *A. pacificus* Nybelin, 1931, and *A. septentrionalis* Nybelin, 1931. In his list of helminths from marine mammals along the Pacific coast of Canada, Margolis (1954) accepted the specific name applied by Markowski.

Deliamure (1955) listed *C. glaciale* Cholodlovsky, 1915 as a species of uncertain status. He evidently had not seen Markowski's (1952) publication, as it was not included in the literature that he cited. Deliamure (1955) described *Diphyllobothrium krotovi* Deliamure, 1955 from fur seals from Tiulen Island (Sea of Okhotsk). He was aware of Stunkard's (1948) ''Species No. 2.'' In his second review of helminths of marine mammals along the Pacific coast of Canada, Margolis (1956) provided evidence for the morphological agreement of cestodes from a Steller sea lion with Nybelin's (1931) description *of A. pacificus*, and accepted the designation *D. pacificum* for the cestode occurring typically in the northern fur seal.

Subsequently, following Margolis (1956), the common cestode in the northern fur seal usually has been designated *D. pacificum* (Nybelin, 1931) (see Machida, 1969; Kamo et al., 1982; Deliamure et al., 1985; Kamo, 1999). One exception was Yamaguti (1959), who retained the name *D. glaciale* (Cholodkovsky, 1915). As synonyms, Yamaguti listed *A. pacificus* Nybelin, 1931 (incorrectly dated 1929); *A. septentrionalis* Nybelin, 1931 (incorrectly dated 1929); and *C. arctocephalinus* (Johnston, 1937) ''(=*arctocephali* Drummond, 1937).'' In the most comprehensive recent works (Deliamure et al., 1985; Kamo, 1999), descriptions of *D. pacificum* were provided, and *D. krotovi* Deliamure, 1955, also from the northern fur seal, was included as 1 of its synonyms. In addition to its 3 previously known hosts, it has been recorded from the California sea lion, *Zalophus californianus* (Lesson) by Dailey and Hill (1970). It was first reported from a person (not considering the incorrect report by Baer [1967]) in Japan by Kamo et al. (1982). Five additional infections in people were mentioned by Tsuboi et al. (1993). A detailed description of *D. pacificum* has been given by Maejima et al. (1981).

MATERIALS AND METHODS

The cestodes studied are listed below by the names designated in the collections from which they were provided to us. (1) An entire specimen and 17 sections of the cestode reported by Stiles and Hassall (1899) as *Bothriocephalus* sp., USNPC No. 2817, leg. F. A. Lucas, from the northern fur seal, Pribilof Islands, in 1896, designated *D. glaciale* in the USNPC; (2) 2 specimens of *A. pacificus* Nybelin, 1931 (paratypes) in alcohol, a slide of stained proglottids, and serial sections (transverse, sagittal, and frontal), leg. K. Bäckström, from a South American sea lion, Juan Fernandez Islands, Natural History Museum, Göteborg; (3) 7 immature specimens of *D.* cf. *pacificum*, from a South American sea lion, 20 XII 1992, Masatierra, Juan Fernandez Islands, leg. Maria Soledad Sepulveda; (4) 10 cestodes, *D. pacificum*, from 3 northern fur seals obtained off Sanriku, northern Honshu, Japan, leg. M. Machida; (5) 1 *D. pacificum* (identified by LM) from a northern fur seal, off the coast of Vancouver Island, Canada, leg. I. MacAskie; (6) 3 *D.* cf. *pacificum* from a Steller sea lion, Pleiades Island, Knight Island Passage, Alaska, 27 X 1977, leg. F. H. Fay and L. M. Shults; (7) slides of cestodes from people in Peru (leg. H. Miranda C.), kindly sent (to RLR in 1968) by the late Professor Jean-G. Baer, Université de Neuchâtel, Switzerland, on which his identification of *D. pacificum* was based. That material consisted of 7 slides with short series of stained proglottids, and 1 scolex, as well as a large series of serial sections, sagittal and transverse. Those included sections of the cestode identified as *D. pacificum* (Baer, 1967, Fig. 5A). The material provided by Professor Baer will be sent to the University of Geneva, where his collection formerly at Neuchâtel has been deposited. (8) A single cestode, lacking a scolex, and somewhat contracted, from a person in Chile (leg. I. Tagle) was generously provided by the late Dr. A. Neghme. That cestode had been identified as *Diphyllobothrium latum* (Linnaeus, 1758) by Neghme et al. (1950).

Excepting specimens received on loan, cestodes were stained in acetic carmine or Ehrlich's hematoxylin, dehydrated in ethanol, cleared in terpineol, rinsed in xylene, and mounted permanently. Transverse and sagittal sections were cut by hand from series of gravid proglottids. Tegument and superficial musculature were removed from ventral surfaces of selected proglottids to facilitate study of the internal organs.

RESULTS

The taxonomic status of *D. pacificum* from the northern fur seal, and of taxa considered to be its synonyms, has been indefinite. Baer's (1967) study of cestodes from humans along the coast of northern Peru was made difficult by the quality of the

specimens (usually expelled spontaneously) and by longstanding concepts concerning synonymies. We conclude that the cestode identified by Baer as *D. pacificum* (synonym: *D. septentrionalis*) is instead *D. arctocephalinum*, which occurs in sea lions and sometimes in people in the southern hemisphere. Descriptions of the 2 species are included here. Because the anatomy of *D. pacificum* has been well portrayed elsewhere (Stunkard, 1948; Deliamure, 1955, as *D. krotovi*; Deliamure et al., 1985), we do not include a figure of a proglottid of that species. All dimensions are in microns unless otherwise stated.

REDESCRIPTION

Diphyllobothrium pacificum (Nybelin, 1931)

Diagnosis: Length of strobila 150–226 mm, with maximum of ca. 230 proglottids; maximal width 3–5 mm. Scolex rounded–ellipsoidal in lateral view; 3.7–4 mm long by 3.5–3.7 mm in maximal width. Neck 936–1.1 mm long. Longitudinal excretory canals, bilateral, 20–30 in diameter. Proglottids with egg-filled uteri about 4–5 times wider than long. Genital atrium, 230–350 in diameter, situated about 1/3 of length of proglottid from its anterior margin. At anterior end of atrium, transverse flap extending posteriad over anterior part of opening of cirrus sac. Opening of vagina, usually ca. 60 in diameter, 30–40 posterior to opening of cirrus sac. Immediately dorsal to its opening, vagina making abrupt turn dorsad, enlarging to diameter of 140–160, then extending to ventral surface of seminal vesicle, there turning ventrad; then decreasing to diameter of ca. 10, taking convoluted course posteriad ventral to gravid uterus. On ventral surface of proglottid, between its anterior margin and anterior limit of genital atrium, 1–4 transverse recesses or pits, 50–120 in length, present between lobes, with small canals possibly communicating externally. Testes in single, dorsal layer, forming 2 lateral fields not confluent at anterior and posterior margins of proglottid; slightly overlapping ends of loops of gravid uterus. Testes more or less subspherical, 90–140 in greater diameter. Cirrus sac clavate, 300–310 long by 80–90 in maximal diameter; opening in genital atrium just anterior to vaginal opening. Seminal vesicle 150–160 in maximal diameter by 100–150, arising posteriorly at dorsal end of cirrus sac, with its long axis directed posteroventrad. Walls of seminal vesicle ca. 20 in thickness. Uterine pore, 46–56 (average = 53) in diameter, opening 180–250 posterior to vaginal opening. Distal portion of uterus, containing fully developed eggs, forming 4 or 5 transverse loops bilaterally, anteriormost occasionally with ends extending anteriad to level of anterior margin of genital atrium. In more posterior gravid proglottids, uterus usually occupying posterior 2/3 of proglottidal length. Vitelline glands abundant and closely apposed; variable in diameter, 75–112 in ventral view. Reticulate ovary situated at posterior margin of proglottid, 2.1–2.6 mm in width, and ca. 750 in maximal length. Lateral margins of ovary extending slightly beyond lateral ends of uterine loops. Eggs ellipsoidal, 48.7–56.0 in length by 38.9–48.7 in maximal diameter (average = 53.1 × 41.3). Shells of eggs pitted.

Taxonomic summary

Type host: Juan Fernandez fur seal, *A. philippi* (Peters).

Type locality: Masatierra, Juan Fernandez Islands (ca. 800 km west of the coast of Chile at 34°S).

Taxonomic history: First recorded as *Bothriocephalus* sp. by Stiles and Hasssall (1899). Described as *A. septentrionalis* Nybelin, 1931; synonymous with *A. pacificus* Nybelin 1931 on the basis of page priority. *Adenocephalus pacificus* described from *A. australis* (Zimmermann) (= *A. philippi* (Peters)), Masatierra, Juan Fernandez Islands; leg. Kåre Bäckström, 1916–1917. *Adenocephalus pacificum*, as Species No. 2, was placed in *Diphyllobothrium* Cobbold, 1858, by Stunkard (1948).

Site of infection: Large intestine.

Voucher specimen: RLR No. 36554. Leg. M. Machida, off Sanriku Island, Japan. USNPC No. 102429.

Remarks

In the following description of *D. arctocephalinum*, the morphological details that could not be discerned in Baer's (1967) slides have been obtained from descriptions of that cestode by others (as indicated). The most detailed information is that of Deliamure and Parukhin (1968), who described that taxon as a new species, designated *Diphyllobothrium atlanticum* Deliamure and Parukhin, 1968. It was determined by Deliamure et al. (1985) to be synonymous with *Diphyllobothrium arctocephalinum*.

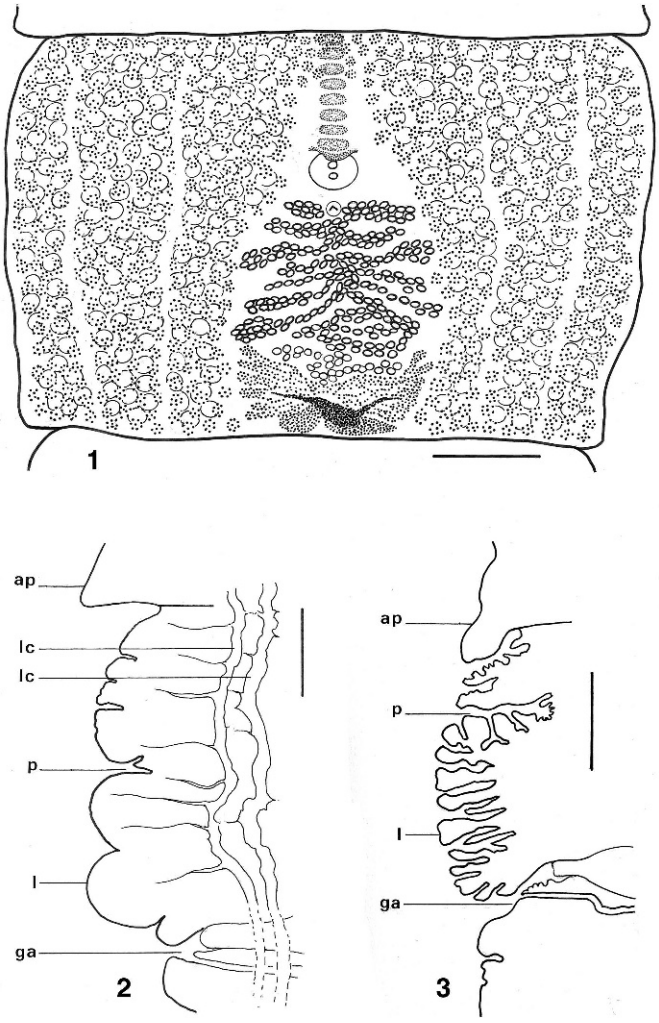

FIGURE 1. Gravid proglottid of *Diphyllobothrium arctocephalinum*, from a resident of northern Peru. (Drawn from J. G. Baer's slide No. 120.) Scale bar = 1 mm.

FIGURE 2. Thick sagittal section of *Diphyllobothrium pacificum*, with ventral median pits. ap, adjacent anterior proglottid; ga, genital atrium; l, lobe; lc, longitudinal canal; p, pit. Scale bar = 100 μ.

FIGURE 3. Sagittal section (Baer 119/84) of *Diphyllobothrium arctocephalinum* (=*D. pacificum* sensu Baer, 1967), showing ventral median pits. ap, adjacent anterior proglottid; ga, genital atrium; l, lobe; p, pit. Scale bar = 300 μ.

REDESCRIPTION

Diphyllobothrium arctocephalinum Johnston, 1937 (= *D. pacificum* sensu Baer, 1967)

(Fig. 1)

Diagnosis: Length of strobila not less than 54 cm (Johnston, 1937); 23 cm (Drummond, 1937); and 2 specimens 32 and 35 cm (the latter having 292 proglottids) (Deliamure and Parukhin, 1968). Maximal width of strobila 6 mm (Johnston, 1937); 6 mm (Drummond, 1937); 5–6 mm (Deliamure and Parukhin, 1968); and 6 mm (Baer's material). Gravid proglottids about 4 mm long. Scolex rather ellipsoidal in lateral view,

2.0 mm to 2.5 mm by l.5 mm to 2.1 mm in maximal width (Deliamure and Parukhin, 1968). Two longitudinal excretory canals, each situated laterally in medullary parenchyma; 43–65 in diameter (Deliamure and Parukhin, 1968). Genital atrium 280–350 in length by 350–400 in transverse diameter; situated 390–470 posterior to anterior margin of proglottid, or at about l/3 of proglottidal length from anterior margin. Genital atrium with anterior semilunar flap, covering atrium nearly to level of opening of male genital pore. On midline of proglottid ventrally, extending from anterior margin of genital atrium nearly to anterior margin of proglottid, series of 8–9 rounded lobes separated by transverse recesses or pits. Recesses or pits up to 300 long by 10–40 wide. Vaginal orifice in genital atrium about 150 posterior to orifice of cirrus sac. Uterine pore about 250 posterior to and somewhat dextral or sinistral to vaginal opening. Cirrus sac pyriform; as viewed in sagittal section, 434–455 long by 217–260 in maximal diameter (Deliamure and Parukhin, 1968). Testes in single layer, forming 2 lateral fields not confluent at anterior margin of proglottid and not overlapping margins of ovary nor ends of uterine loops. Number of testes stated to be 702–1,224 by Deliamure and Parukhin (1968). Reticulate ovary bilobed, situated at posterior margin of proglottid; isthmus of ovary about 22 in width, anterior to Mehlis' gland and seminal receptacle. Ovary 1.65–1.87 mm wide; ovarian lobes 1.16–1.27 mm in length (Deliamure and Parukhin, 1968). In Baer's material, 1.87–2.0 mm wide, and l.9–2.6 mm in length of lobes. Laterally, ovarian lobes extending anteriad adjacent to early uterine loops; not extending laterad to level of longest uterine loops. Posteriorly, inner ends of ovarian lobes nearly enclosing Mehlis' gland and seminal receptacle. Vitellaria abundant and closely apposed, forming 2 lateral fields confluent rather broadly at anterior end of proglottid, reaching but not extending over row of ventral lobes. Vitellaria not extending mediad over ends of uterine loops, leaving clear field around all uterine loops, genital atrium, and posteriormost of transverse grooves of lobes. Vitellaria more or less subspherical to spherical in ventral view, 30–61 in greater diameter (average = 42). Uterus containing fully developed eggs forming 7–8 transverse loops bilaterally, and occupying about posterior 1/3 of length of proglottid. Uterine loops usually not extending anteriad as far as posterior margin of genital atrium. Eggs ellipsoidal, 41.4–56.0 in length by 36.5–43.8 in diameter (average = 51.3 × 40.6). Dimensions of eggs from Drummond (1937) 60 × 35; from Johnston (1937) 52 to 57 × 35–38; from Deliamure and Parukhin (1968) 51–60 × 34–38.

Taxonomic summary

Type host: New Zealand fur seal, *Arctocephalus forsteri* (Lesson).

Taxonomic history: Described in May 1937 (Johnston, 1937). It was described later the same year (July 1937) by Drummond, as *Diphyllobothrium arctocephali*, from the Australian fur seal, *Arctocephalus tasmanicus* (Scott and Lord) (=*Arctocephalus pusillus* (Schreber)). It was later described as *Diphyllobothrium atlanticum* Deliamure and Parukhin, 1968 from *A. pusillus*, Atlantic Ocean, near the coast of southern South Africa.

Type locality: Pearson Island, 40 km off the west coast of Eyre's Peninsula, South Australia.

Site of infection: Intestine.

Voucher specimen: One slide with 5 proglottids (J. G. Baer No. 120), designated *D. pacificum* by Baer (1967, Fig. 5A); from a person residing near the coast of northern Peru (leg. H. Miranda C.). USNPC No. 102430.

REMARKS

In Baer's (1967) publication, his Figure 5A is clearly a photograph of proglottids of the cestode that we identify as *D. arctocephalinum*, whereas Figure 5B shows proglottids of another, unidentifiable species. The identity of proglottids in Figure 5A is clear also from comparison with other published figures of *D. arctocephalinum* (cf. Johnston, 1937, Figs. 5, 6; Drummond, 1937, Fig. 5; Deliamure and Parukhin, 1968, Fig. 5). Baer (1967) noted that the cestodes identified as *D. pacificum* from humans in Peru measured 30–196 cm in length (Miranda et al., 1967), whereas the type specimens of *Adenocephalus pacificus* (=*D. pacificum*), stated to have been complete and relaxed, were only 88 and 100 mm in length (Nybelin, 1931). Three specimens, all well extended, from Stiles and Hassall's material described as *A. septentrionalis* Nybelin, 1931, ranged in length from 50 to 66 mm. As indicated by their reported lengths, cestodes from sea lions along the Pacific coast of South America appear often to have been identified incorrectly as *D. pacificum* (Cattan et al., 1977). Baer (1967) recognized that the cestodes from people were considerably longer than those identified previously as *D. pacificum* from otariids. He stated (1967), ". . . this is very probably due to the fact that this species is usually found in large numbers in these hosts, whereas in the human cases there was only a single worm. There is no doubt whatever that in the genus *Diphyllobothrium*, the crowding effect is very pronounced."

Whether species of *Diphyllobothrium* are subject to "crowding effect" appears to be questionable, despite supporting statements in the literature. Baer (1967) stated that the strobilae obtained from humans varied from 300 to 1,960 mm in length, but further details were not provided. The cestodes studied by him were obtained by Dr. Miranda, and evidently consisted of portions of strobilae expelled spontaneously by infected persons (Miranda et al., 1967). Two, or more, species were present, of which the largest, represented by a 15-mm-long fragment, consisted of 8 proglottids having a maximum width of 13 mm. According to von Bonsdorff (1977), who reviewed literature concerning *D. latum* (Linnaeus, 1758), that cestode would appear to be subject to the crowding effect. In contrast, with respect to that species in northern Europe, Östling (1961) obtained 270 m of strobila and 18 scoleces from 1 patient, and 350 m of strobila and 16 scoleces from a second. Assuming that all scoleces had been obtained, the mean lengths of the cestodes from the 2 persons would have been 15.0 and 20.6 m, respectively. Of 17 complete specimens of *D. latum* from Yup'ik residents of southwestern Alaska, the longest measured 11.4 m (Rausch and Hilliard, 1970); the records indicated that those cestodes, all obtained by treatment with quinacrine, occurred singly. *Diphyllobothrium latum* appears to attain the greatest length of any cestodes of that genus that have an entirely freshwater life cycle.

Our observations seem generally to provide no support for the occurrence of the crowding effect. For example, 6 specimens of *Diphyllobothrium lanceolatum* (Krabbe, 1865) from a naturally infected sledge dog, *Canis lupus* forma familiaris were within the range of length of those cestodes in the bearded seal, the typical host, in which hundreds of the cestodes were often present in individual animals (Rausch and Hilliard, 1970). Of 523 cestodes from a brown bear, *Ursus arctos* L., on Kodiak Island, Alaska, the largest of several specimens of *Diphyllobothrium ursi* Rausch, 1954 was 11 m in length (Rausch, 1954). The many small cestodes also present in that animal, subsequently identified as *Diphyllobothrium dendriticum* (Nitzsch, 1824), were typical in length for the species obtained experimentally in gulls and dogs (Rausch and Hilliard, 1970).

Diphyllobothrium pacificum and *D. arctocephalinum* are readily distinguished morphologically (Deliamure et al., 1985), but they have in common a feature that appears not to occur in other species of the genus (Fig. 2). Each has a series of lobes, separated by recesses, on the ventral surface of the proglottid, on the midline between its anterior margin and the genital atrium. Those recesses or grooves, designated "pits" by Baer (1967), number from 1 to 4 in *D. pacificum* and 8 to 9 in *D. arctocephalinum*. The structures (elevated lobes and recesses) in *D. pacificum* are well shown in an electron micrograph by Maejima et al. (1981, Fig. 18). In their detailed description of that cestode, they suggested that the ventral

TABLE I. Comparison of morphological characteristics of *Diphyllobothrium pacificum* and *Diphyllobothrium arctocephalinum*.

Character	*Diphyllobothrium pacificum*	*Diphyllobothrium arctocephalinum*
Hosts	Otariids; occasional in people	Otariids; occasional in people
Distribution	Pacific Ocean in both northern and southern hemispheres	Pacific Ocean in southern hemisphere; also southern Atlantic coast of South Africa
Length of strobila	88–226 mm	32 to ca. 54 cm
Maximal width of strobila	3.8–6 mm	5–6 mm
Number of proglottids	108–259	292 to unknown maximum
Form of gravid proglottids	4–5 times wider than long	About 1.5 times wider than long
Number of pits anterior to genital atrium	1–4	8–9; extending farther dorsad than in *D. pacificum*
Distribution of vitelline glands	Not confluent at anterior and posterior margins of proglottids; overlapping ends of uterine loops and lateral margins of ovary	Confluent at anterior margin of proglottids; not overlapping uterine loops or margins of ovary
Dimensions of egg*	Mean 53.1 × 41.3 μm	Mean 51.3 × 40.6 μm

* For both species, published dimensions of eggs differ.

lobes were unique to that species and could serve to distinguish it from all others of the genus, but remarked that those structures had been noted by Drummond (1937), Johnston (1937), and Baer (1967). The latter authors, as is now understood, were concerned with *D. arctocephalinum* (not *D. pacificum*). In their study of a specimen of *D. pacificum* expelled from a person, Tsuboi et al. (1993) observed the ventral structures, but did not provide details. The function of the anteroventral structures is not understood. They occupy a considerable proportion of the anterior surface of the proglottid and significantly increase its surface area.

In those 2 species of *Diphyllobothrium*, the recesses or pits were situated between the elevated areas; the lobes of *D. pacificum* were rounded, bordered by dense nuclei. In *D. arctocephalinum*, similar lobes were present, between transverse pits, 190–200 in length (Fig. 3). The pits in both were lined completely with an extension of tegument from the ventral surface of the proglottid. In our specimens of *D. pacificum*, the pits extended 22–32 dorsad, whereas those of *D. arctocephalinum* were deeper (see above). In both species, the diameter of pits decreased dorsad toward their ends in the cortical parenchyma.

As seen in sagittal sections of the 2 species, a canal in diameter to about 30 extended longitudinally in the cortical parenchyma just medial to the dorsal ends of the ventral pits. Also present was a network of smaller ducts, which connected with those of larger size. A similar pattern of canals occurred in the cortical parenchyma dorsally in the strobila. Study of sections indicated that small canals extended distad into the lobes from larger longitudinal canals in the ventral cortical parenchyma; further relationships could not be defined. The presence of canals in the cortical parenchyma was observed by Moniez (1881, Plate 8) in *D. latum*. Our comparisons, although not comprehensive, have shown that cortical canals appear to be relatively few in those species of *Diphyllobothrium* that have freshwater life cycles.

Size of strobila is well defined with respect to the discrimination of the 2 species. That of *D. pacificum* is smaller and less robust. Nybelin (1931) stated that well-relaxed specimens were 88 and 100 mm in length, with 195 and 270 proglottids, respectively. Stunkard (1948, p. 213) stated, ''. . . the longest worms measured about 120 mm in length, with a maximum of 210 proglottids.'' On the basis of 18 cestodes from the northern fur seal obtained off the coast of Japan, Maejima et al. (1981) stated that strobilae ranged from 75 to 160 mm in length, with 108 to 259 proglottids. Maximal widths of strobilae were 4.5 to 6 mm (Stunkard, 1948) and 1.95 to 3.50 mm (Maejima et al., 1981). Deliamure et al. (1985) recorded a maximal width of 3.77 mm. *Diphyllobothrium arctocephalinum* has a longer, more muscular strobila, with relatively long proglottids. The single ''mature'' specimen studied by Drummond (1937) was 23 cm long, with a maximal width of 6 mm. Johnston (1937) reported that in 1 otariid, ''a tangled mass of cestodes whose separation resulted in some fragmentation'' was found. He considered the length of an unbroken strobila to be not less than 54 cm, considerably greater than the maximum length known for *D. pacificum*. The 2 specimens studied by Deliamure and Parukhin (1968) were 32 and 35 cm in length.

The arrangements of internal organs in the 2 species are distinctive (Table I), in that the female genital organs in *D. arctocephalinum* are almost entirely surrounded by free space, whereas those of *D. pacificum* exhibit various areas of contact or overlap with other organs. The vitelline glands of *D. pacificum* typically are not confluent at the anterior margin of the proglottid, and never at the posterior margin; in *D. arctocephalinum*, the lateral fields of vitelline glands do not join posteriorly, but anteriorly the vitellaria often form a wide band of confluence extending about halfway to the genital atrium. In *D. pacificum*, vitellaria extend mediad over ends of major uterine loops bilaterally, and over lateral margins of ovary; neither is typical of *D. arctocephalinum*. In *D. pacificum*, testes also extend dorsal to ends of uterine loops; not in *D. arctocephalinum*. Numbers of uterine loops containing fully developed eggs usually number 4–5 bilaterally in *D. pacificum* and 7–8 bilaterally in *D. arctocephalinum*.

The cestode from a human in Chile, identified as *D. latum* by Neghme et al. (1950), lacked a scolex and was considerably contracted. With a beginning width of 2 mm, the strobila was 1,717 mm in length and consisted of 2,068 proglottids, 700–1 mm in length and 7 mm in maximal width. The genital atrium was situated at the anterior margin of the proglottid. The vitellaria formed lateral bands that did not overlap the ends of the uterine loops. Our study indicated that the cestode is not *D. latum* or *D. arctocephalinum*. We could not determine its identity.

DISCUSSION

Life cycles of *Diphyllobothrium* spp. fall into 1 of 3 patterns: (1) Freshwater, with piscivorous birds or mammals serving as final

host; (2) marine, with pinnipeds or whales as final hosts; and (3) in freshwater, development to early-stage plerocercoid in anadromous fishes and further development in marine habitat. Final hosts are usually terrestrial mammals, including people; infection of marine mammals cannot be excluded.

Most diphyllobothriids lack well-defined host specificity, although some species occurring in piscivorous birds have not been recorded from mammals. Those that characteristically are found in seals (Phocidae) in the Antarctic evidently have not been recorded from people, but that may be attributable to the lack of human population in that region. Also, indigenous fishes probably are rarely eaten by visitors to the southernmost regions. At least 10 species of *Diphyllobothrium* have been reported from people (Adams and Rausch, 1997). Yamane and Shiwaku (2003) listed 8 species of diphyllobothriids (6 of *Diphyllobothrium*) from people in Japan.

Decreasing numbers of marine mammals and commercially important fishes in the northern Pacific region may have the effect of reducing transmission of diphyllobothriids. In that area, the northern fur seal and, to a lesser degree, the Steller sea lion, are characteristic hosts of *D. pacificum*. Those mammals have decreased in numbers, probably as a result of commercial overfishing for pollack, *Theragra chalcagramma* (Pallas), and fishes of other species. According to reports in the public media in October 2008, numbers of orcas, *Orcinus orca* (L.), have undergone alarming decline in waters bordering the northwestern United States. Pollutants of anthropogenic origin might be the cause of their decline. That whale is the host of *Diphyllobothrium orcini* Hatsushika et Shirouzu, 1990.

The presence of eggs of *Diphyllobothrium* in coprolites indicates that residents of coastal Peru have been infected by diphyllobothriids since ancient times (Holiday et al., 2003). (Such eggs are not reliable for identification of species.) As is obvious from the slides studied by Baer (1967), species other than *D. pacificum* and *D. arctocephalinum* occur in people, as well as otariids along the Pacific coast of South America. Study of additional material in good condition is needed, with publication of adequate descriptions. In a survey involving the examination of 1,085 fishes from the seas bordering Peru, Tantalean (1975) found plerocercoids in sciaenids of 2 species. Of 188 *Sciaena deliciosa* Tschudi, 6.5% were infected, and 0.5% were infected in 180 *Polyclemus peruanus* (Steindachner). Fishes of both species are often consumed uncooked (Baer, 1967), and appear to be probable sources of plerocercoids infecting humans. Species of *Diphyllobothrium* are common symbionts in many species of birds and mammals. In people, infection often becomes evident only when portions of strobilae are expelled. A range of symptoms has been attributed to infection by such cestodes (e.g., Kamo, 1981; Yamane and Shiwaku, 2003). With the exception of the severe to fatal anemia caused by *D. latum* seen in patients in Finland (von Bonsdorff, 1977), descriptions of adverse effects have been only rarely reported. Curtis and Bylund (1991) found that the condition of severe anemia in people in Finland is no longer known to occur. Symptoms possibly related to infections by diphyllobothriids in Peru were listed by Miranda et al. (1967). Baer (1967, 1969) did not report adverse effects due to presence of *D. arctocephalinum*.

It is evident that the 2 species, *D. pacificum* and *D. arctocephalinum*, form a morphologically distinctive subgroup of *Diphyllobothrium*. They appear to occur typically in otariids of the genera *Callorhinus* and *Arctocephalinus*, both of which appeared during the early Pliocene, derived from the Otarioidea that arose during the early Miocene in the northern Pacific Oceanic region of North America (Thenius, 1980).

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LITERATURE CITED

Adams, A. M., and R. L. Rausch. 1997. Diphyllobothriasis. *In* Pathology of infectious diseases, Vol. II, D. H. Connor, F. W. Chandler, D. A. Schwartz, and H. J. Manz (eds.). Appleton and Lange, Stamford, Connecticut, p. 1377–1389.

Afanas'ev, V. P. 1941. Parazitofauna promyslovykh mlekopitaiuschchikh Komandorskikh ostrovov. Uchenie zapiski. Leningradskii Gosudarstvennyi Universitet, Seriia Biologii **74:** 93–117.

Baer, J. G. 1967. Identification of the species. Part I. *In* Human diphyllobothriasis in Peru, J. G. Baer, H. Miranda, W. Fernandez, and J. Medina (eds.). Zeitschrift für Parasitenkunde **28:** 277–289.

———. 1969. *Diphyllobothrium pacificum*, a tapeworm from sea lions endemic in man along the coastal area of Peru. Journal of the Fisheries Research Board of Canada **126:** 717–723.

Bray, R. A., A. Jones, and K. I. Andersen. 1994. Order Pseudophyllidea Carus, 1863. Part 10. *In* Keys to the cestode parasites of vertebrates, L. F. Khalil, A. Jones, and R. A. Bray (eds.). CABI International, University Press, Cambridge, U.K., p. 205–247.

Cattan, P. E., A. Atlas, B. B. Babero, and D. Torres. 1977. Helmintofauna de Chile: Primer hallazgo de *Diphyllobothrium pacificum* (Nybelin 1931) Margolis 1956, en lobos marinos de la costa Chilena. Revista Ibérica de Parasitologia **37:** 285–290.

Cholodkovsky, N. 1915. Cestodes nouveaux ou peu connus Annuaire du Musée zoologiques de l'Académie des Sciences de l'Union des Républiques Soviétiques Socialistes **19:** 516–523.

Curtis, M. A., and B. Bylund. 1991. Diphyllobothriasis: Fish tapeworm disease in the circumpolar north. Arctic Medical Research **50:** 18–25.

Dailey, M. D., and B. L. Hill. 1970. A survey of metazoan parasites infecting the California (*Zalophus californianus*) and Steller (*Eumetopias jubatus*) sea lion. Bulletin of the Southern California Academy of Sciences **69:** 126–132.

Deliamure, S. L. 1955. Gel'mintofauna morskikh mlekopitaiushchikh v svete ikh ekologii i filogenii. Gel'mintologicheskaia Laboratorii, Akademiia Nauk SSSR, Moskva, U.S.S.R., 517 p.

———, and A. M. Parukhin. 1968. Novaia difillobotriiida—parazit iuzhnoafrikanskogo morskogo kotika. *In* Biologiia Moria, No. 14, Parazity morskikh zhivotnykh, V. A. Vodianitskii (ed.). Naukova Dumka, Kiev, U.S.S.R., p. 25–34.

———, A. S. Skriabin, and A. M. Serdiukov. 1985. Difillobotriidy—lentochnye gel'minty cheloveka, mlekopitaiuschchikh i ptits. Vol. 11, Osnovy tsestodologii. Akademiia Nauk SSSR, Nauka, Moskva, U.S.S.R., 200 p.

Drummond, F. H. 1937. Cestoda. Proceedings of the Royal Society of Victoria **49:** 401–406.

Holiday, D. M., S. Guillin, and D. J. Richardson. 2003. Diphyllobothriasis of the Chiribaya culture (700–1476 AD) of southern Peru. Comparative Parasitology **70:** 167–171.

Jefferson, T. A., S. Leatherwood, and M. A. Webber. 1993. FAO species identification guide. Marine mammals of the world. FAO, Rome, Italy, 320 p.

Johnston, T. H. 1937. Entozoa from the Australian hair seal. Proceedings of the Linnean Society of New South Wales **62:** 9–16.

Kamo, H. 1981. Present situation of human diphyllobothriasis in Japan. Yonago Acta Medica **25:** 144–155.

———. 1999. Guide to identification of Diphyllobothriid cestodes. Gendai Kikaku, Tokyo, Japan, 146 p.

———, J. Maejima, S. Yazaki, M. Otsuru, H. Hasegawa, S. Kuniyoshi, and R. Asato. 1982. Occurrence of human infection with *Diphyllobothrium pacificum* (Nybelin, 1931) Margolis, 1956 in Japan. Japanese Journal of Parasitology **31:** 165–170.

MACHIDA, M. 1969. Parasites of the northern fur seal and their relationship to the breeding islands. Proceedings of the Japanese Society of Systematic Zoology, No. 5, p. 16–17.

MAEJIMA, J., S. YAZAKI, S. FUKUMOTO, M. HIRAGA, AND H. KAMO. 1981. Morphological observation of *Diphyllobothrium pacificum* (Nybelin, 1931) Margolis, 1956 from fur seals, *Callorhinus ursinus*, in Japan. Yonago Acta Medica **25:** 69–79.

MARGOLIS, L. 1954. List of the parasites recorded from sea mammals caught off the west coast of North America. Journal of the Fisheries Research Board of Canada **11:** 267–283.

———. 1956. Parasitic helminths and arthropods from pinnipedia of the Canadian Pacific coast. Journal of the Fisheries Research Board of Canada **13:** 489–505.

———, AND M. D. DAILEY. 1972. Revised annotated list of parasites from sea mammals caught off the west coast of North America. Technical Report NMFS SSRF-647, NOPAA, Seattle, Washington, 18 p.

MARKOWSKI, S. 1952. The cestodes of pinnipeds in the Arctic and other regions. Journal of Helminthology **26:** 171–214.

MIRANDA, C. H., W. FERNANDEZ, AND J. MEDINA. 1967. Description of seven new cases of human diphyllobothriasis. Part II. *In* Human diphyllobothriasis in Peru, J. G. Baer, H. Miranda, W. Fernandez, and J. Medina. Zeitschrift für Parasitenkunde **28:** 283–289.

MONIEZ, R. 1881. Mémoires sur les cestodes. Première partie. Travaux de l'Institut Zoologique de Lille et de la Station Maritime de Wimereux. Tome III, Fascicule II. Octave Doin, Paris, France, 238 p.

NEGHME, A., R. DONCKASTER, AND R. SILVA. 1950. *Diphyllobothrium latum* en Chile. Primer caso autóctono en el hombre? Revista Médica de Chile **78:** 410–411.

NYBELIN, O. 1931. Säugetier- und Vogelcestoden von Juan Fernandez. *In* The natural history of Juan Fernandez and Easter Island, Vol. III, Zoology, C. Skottsberg (ed.). Uppsala, Sweden, p. 493–523.

ÖSTLING, G. 1961. Treatment of tapeworm infection with desapidin, a new phloroglucinol derivative isolated from Finnish fern. American Journal of Tropical Medicine and Hygiene **10:** 855–858.

RAUSCH, R. L. 1954. Studies on the helminth fauna of Alaska. XXI. Taxonomy, morphological variation, and ecology of *Diphyllobothrium ursi* n. sp. prov. on Kodiak Island. Journal of Parasitology **40:** 540–563.

———, AND D. K. HILLIARD. 1970. Studies on the helminth fauna of Alaska. XLIX. The occurrence of *Diphyllobothrium latum* (Linnaeus, 1758) (Cestoda: Diphyllobothriidae) in Alaska, with notes on other species. Canadian Journal of Zoology **48:** 1201–1219.

ROZENBERG, A. I. 1977. Difillobotriidy I difillobotriozy, imeniushchie medico-veterinarnoe znachenie. Ukazatel' otechestvennoi i inostrannoi literatury 1558–1972. E. M. Zhadena, and A. K. Koski (eds.). Kareliia, Petrozavodsk, U.S.S.R., 294 p.

STILES, C. W., AND A. HASSALL. 1899. Internal parasites of the fur seal, Chapter VII. In The fur seals and fur-seal islands of the North Pacific Ocean, Part 3. U. S. Government Printing Office, Washington, D.C., 629 p.

STUNKARD, H. W. 1948. Pseudophyllidean cestodes from Alaskan pinnipeds. Journal of Parasitology **34:** 211–228.

TANTALEAN, M. 1975. Hallazgo de larvas plerocercoides de Diphyllobothriidae Lühe, 1910 (Cestoda) en peces del mar peruano. Boletin Chileno de Parasitologia **30:** 18–20.

THENIUS, E. 1980. Grundzüge der Faunen- und Verbreitungsgeschichte der Säugetiere. Eine historische Tiergeographie. Gustav Fischer Verlag, Stuttgart, Germany, 375 p.

TSUBOI, T., M. TORII, AND K. HIRAI. 1993. Light and scanning electron microscopy of *Diphyllobothrium pacificum* expelled from a man. Japanese Journal of Parasitology **42:** 422–428.

VON BONSDORFF, B. 1977. Diphyllobothriasis in man. Academic Press, New York, New York, 189 p.

WARDLE, R. A., J. A. MCLEOD, AND I. E. STEWART. 1947. Lühe's "Diphyllobothrium" (cestoda). Journal of Parasitology **33:** 319–330.

YAMAGUTI, S. 1951. Studies on the helminth fauna of Japan. Part 47. Cestodes of marine mammals and birds. Arbeiten aus der Medizinischen Fakultät Okayama **7:** 307–316.

———. 1959. Systema Helminthum. Vol. II. The cestodes of vertebrates. Interscience, New York, New York, 860 p.

YAMANE, Y., AND K. SHIWAKU. 2003. *Diphyllobothrium nihonkaiense* and other marine-origin cestodes. Progress of Medical Parasitology in Japan, Meguro Parasitological Museum (Tokyo) **8:** 245–259.