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Redescription of *Antarctophthirus lobodontis* (Anoplura: Echinophthiriidae) from the crabeater seal and identification key for Antarctic lice

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Abstract The Anoplura (Phthiraptera) is composed of lice parasitizing mainly terrestrial mammals, but a few members have been able to adapt to the marine environment. The latter are included in the family Echinophthiriidae, a particular group infecting pinnipeds worldwide. They also are of the few insects that managed to survive in the ocean. The study of Antarctic echinophthiriids flourished in the 1960s, but in the last 50 years, no advance has been made. Revision of Antarctic echinophthiriids is part of ongoing research on the systematics, phylogeny and ecology of these lice. During the summer season of 2014, we had the opportunity to collect lice from crabeater seals captured at Cierva Cove in the northern sector of the Danco Coast, Antarctic Peninsula. Since its original description was incomplete and the holotypes were lost, here we redescribe Antarctophthirus lobodontis based on these specimens. The present material can be distinguished from other Antarctic Antarctophthirus species by the presence of four marginal long hairs and in the basis of the head a line of eight spines and three hairs above the last row of four

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spines. Also in the present work, we provide a key to the identification of the Antarctic species of Echinophthiriidae based on morphological characteristics.

Keywords Antarctica · *Antarctophthirus* · Sucking lice · Taxonomic revision

Introduction

The Anoplurans (Insecta: Phthiraptera) or sucking lice are hematophagous ectoparasites present mainly in terrestrial mammals. However, a particular group of anoplurans managed to survive underwater, infecting amphibious hosts like pinnipeds and the river otter (Leonardi and Palma 2013). This group of lice are included in the family Echinophthiridae, which comprises 5 genera and 13 species: *Latagophthirus* (monotypic, from the river otter); *Proechinophthirus* (two species, infest fur seals and sea lions); *Lepidophthirus* (two species) and *Echinophthirus* (Leonardi and Palma 2013).

Seven species are grouped in the genus Antarctophthirus, thereby being the most diversified genus of the family. The genus is represented by A. callorhini from the northern fur seal, A. trichechi from walruses, A. microchir from sea lions, and the four species infecting Antarctic seals, i.e., A. ogmorhini from the leopard seal, A. carlinii from the Weddell seal, A. mawsoni from Ross seal and A. lobodontis in the crabeater seal.

Antarctophthirus lobodontis was described by Enderlein in 1909 based on one male and one female collected in Booth Island, Antarctica, by the Deutsche Südpolar Expedition. Ferris (1934) reproduced this information and pointed out that female length is 2.5 cm. However, as was

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highlighted by Leonardi and Palma (2013), the repository is unknown and the holotypes are missing. Moreover, by current standards, the description needs to be updated.

As part of an ongoing research on the ecology of packice seals conducted annually by the Argentinean Antarctic Institute, we had the opportunity to collect lice from crabeater seals. In view of the incomplete description of this louse species, the aims of the present study are to redescribe adults of *A. lobodontis* and to present an identification key for Antarctic echinophthirid species.

Materials and methods

Specimens examined

The samples were taken in the Antarctic Specially Protected Area (ASPA) No. 134 "Punta Cierva," (64°09′23″S, 60°57′17″W) in the northern sector of the Danco Coast during the austral summer of 2013/2014. Lice were collected from 4 crabeater seals, which were immobilized following the routine procedures (see details in Wheatley et al. 2006). Ten males and ten females of *Antarctophthirus lobodontis* were examined under light microscope. Voucher specimens are deposited at the Parasitology Collection, Centro Nacional Patagónico (Puerto Madryn, Argentina).

Light microscopy

Lice were prepared following the slightly modified protocol of Palma (1978). The specimens were treated with 20 % aqueous solution of potassium hydroxide (KOH) for 24 h. The KOH macerates the non-chitinous tissues and removes color from the sclerotin, distending the whole body. The KOH was removed and replaced by distilled water for 30 min and then by a 10 % aqueous solution of acetic acid. The acid neutralizes the remaining alkali, stopping maceration and avoiding damage by over treatment. The specimens were dehydrated in an ethanol series of 70, 80, 90 and 96 %, for 30 min, at each concentration. After dehydration, the alcohol was replaced by pure clove oil for 24 h. A coverslip was placed with some weight upon the lice to flatten them. Lice were finally mounted in Canada balsam.

Figures were made using a drawing tube mounted on a Leica light microscope at $\times 1000$ magnification. Measurements were taken from digital photographs using ImageJ software (Wayne Rasband, NIH, USA). Illustrations of the adults are a composite from a series of photographs. All measurements in the text are in millimeters unless otherwise stated and are given as the mean \pm standard deviation and followed by range, and the number of measurements.

Terminology

Species of Echinopthiriidae are characterized by their modified setae (Kim 1985). Names and abbreviations of setae used in this paper follow those of Kim and Ludwig (1978) and Leonardi et al. (2009): Spines are pointed setae, scales are flattened setae, and hairs are the long and thin setae.

Results

Antarctophthirus lobodontis Enderlein, 1909

"Antarctophthirus ogmorhini" Neumann 1907: 13. Not Antarctophthirus ogmorhini Enderlein, 1906.

Antarctophthirius [sic] lobodontis Enderlein, 1909; Ass 1934: 103. Misspelling.

Redescription

Type host: Crabeater seal, Lobodon carcinophaga

Type locality: Booth Island, Antarctica.

Type specimen/s data: Syntypes, \Im^{\bigcirc} ; repository unknown.

Other hosts: None.

Geographic distribution: Antarctica

Significant references: Freund (1928: 20, detailed figures); Ferris (1934: 488, synonymy, figures); Durden and Musser (1994: 7, synonymy, hosts, distribution); Leonardi and Palma (2013, synonymy, hosts, distribution, citations).

Site in host: Mostly in the hind flippers

Repository: Parasitology Collection, Centro Nacional Patagónico—CENPAT/CONICET. Puerto Madryn, Argentina. Collectors: MS Leonardi, S Poljak, P Carlini, J Galliari, M Bobinac, J Negrete.

Prevalence: 40 % (in four out of ten seals analyzed).

Mean intensity: 126 (intensity by individual: 35, 67, 99, 303)

Male: (Fig. 1) Total body length 2.60 ± 0.11 , 2.39-2.79, 10. *Head*: Slightly longer than wider (length: 0.55 ± 0.02 , 0.53-0.62, 10; width: 0.48 ± 0.04 , 0.39-0.52, 10); anterior margin heavily sclerotized; maxillary vestige very distinct; ventral labrum connected to long apodemes; postantennal angle developed, posterolateral angle not developed. The proboscis is prominent, formed by two pieces with two hooks on each one, around it two short and thin apical hairs in each side. Ventral head chaetotaxy less developed. Along the external border, four ventral principal head spines followed by a column of short spines. Short ventral posterior marginal spines of different sizes. Dorsally, the



Fig. 1 Light microscope micrograph of male Antarctophthirus lobodontis (scale bar 500 μ m)

pattern of chaetotaxy of *A. lobodontis* very characteristic (Fig. 2). Below the sclerotized anterior margin a conspicuous line of five dorsal anterior head spines. This is followed by three suprantennal central head spines and the dorsal and suprantennal head setae converging in six spines. Two rows of four sutural head spines below a row of spines, the most central and the most marginal are longer and stronger. The posterior margin of the head presents

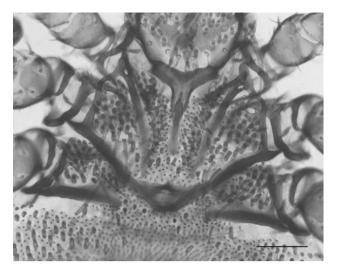


Fig. 3 Detailed of the thorax of Antarctophthirus lobodontis by light microscope micrograph (scale bar 200 μ m)

four marginal long hairs and the basis a line of eight spines and three hairs above the last row of four spines. Antennae: Typical of the genus. Five segments. The basal segment is the biggest and possesses three rough spines. The sclerotized margin of each segment has 3-4 thin and very short hairs. The terminal segment with sensoria at apex. Thorax: (Fig. 3) Trapezoidal, two times as long as broad and long as the head (length: 0.52 ± 0.02 , 0.50-0.55, 10; width: $1.00 \pm 0.04, 0.94$ –1.04, 10). Occipital apophyses typical of the genus, developed and parallel, above them a very sclerotized and notorious plate that cover the posterior margin of the head. Thoracic sternal plate covered by scales and spines. Dorsally, pleural apophysis very sclerotized, the space between them covered by scales and spines. Very conspicuous notal pit. Two long prothoracic long hairs and 3 meso- and metathoracic dorsal long hairs.

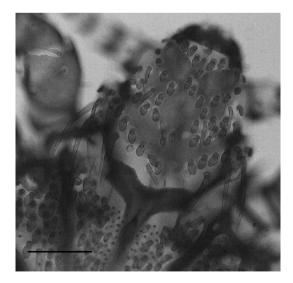


Fig. 2 Pattern of chaetotaxy on the dorsal side of the head of Antarctophthirus lobodontis by light microscope micrograph (scale bar 200 μ m)

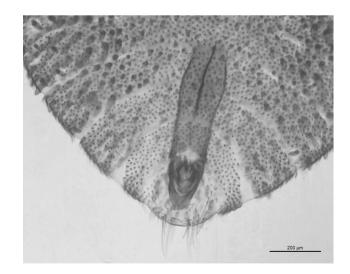


Fig. 4 Light microscope micrograph of male genitalia of Antarctophthirus lobodontis (scale bar 200 µm)

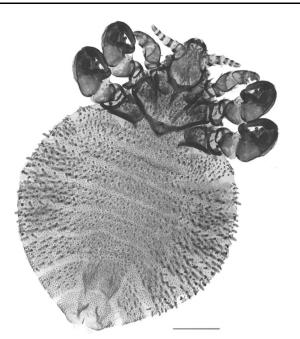


Fig. 5 Light microscope micrograph of female Antarctophthirus lobodontis (scale bar 500 μ m)

Legs: Fore legs characteristic of genus, small and weak; middle and hind legs very large and strong, very similar in shape and size. The femur of these legs possesses four spines. *Abdomen:* Very large, rounded, nearly as long as wide (length: 1.53 ± 0.10 , 1.34-1.67, 10; width: 1.58 ± 0.05 , 1.49-1.68, 10); without specific characteristics nor distinctive tergites or sternites; paratergal plates not developed; six spiracles present. The setae are mainly modified in scales and cover the abdomen entirely. Dorsal marginal abdominal setae and dorsal lateral abdominal setae modified in numerous spines. The size and density of spines decrease for the margin to the central of the abdomen. Eight apical long and thin hairs. Scales ovoid, uniform in shape and size. Spines pointed, spiral-shaped, vary in size but not in shape.

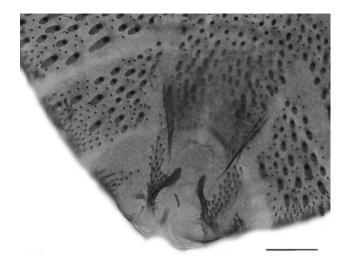


Fig. 6 Light microscope micrograph of female genitalia of Antarctophthirus lobodontis (scale bar 200 µm)

Genitalia (Fig. 4). Basal plate long, slightly wider in its insertion, long parameres; relatively short V-shaped pseudopenis, slightly less wide in its extreme; the arms of the pseudopenis articulate with bases of parameres.

Female (Fig. 5): Total body length: 3.22 ± 0.24 , 2.84–3.55, 10. Head (length: 0.59 ± 0.04 , 0.53–0.64, 10; width: 0.48 ± 0.03 , 0.41–0.53, 10), thorax (length: 0.53 ± 0.03 , 0.49–0.60, 10; width: 1.16 ± 0.08 , 1.03–1.27, 10), legs and abdomen as in male, except for genitalia and associated characters; abdomen bigger and more rounded (length: 2.1 ± 0.17 , 1.86–2.35, 10; width: 2.29 ± 0.22 , 1.96–2.55, 10). *Genitalia* (Fig. 6): Without distinct genital plate, gonopods and spermatheca; with a fringe of setae surrounding the genital opening, divided in a little group of few spines in the ninth abdominal segment and more abundant in the tenth abdominal segment to the apex.

Key to species of Antarctic Echinophthiriidae

1	a. Antennae with 4 segments	2
	b. 5 segments	Antarctophthirus 3
2	a. Elongated abdomen; setae modified in spines and hairs, without scales	Proechinophthirus zumpti
		Host: Arctocephalus gazella
		Antarctic fur seal
	b. Rounded abdomen; scales covering the whole dorsal side	Lepidophthirus macrorhini
		Host: Mirounga leonina
		Southern elephant seal
3	a. Ventral side of the head with short hairs	A. mawsoni
		Host: Ommatophoca rossi
		Ross seal

	b. Ventral side of the head with spines	4
4	a. Central anterior head setae modified in spines	5
	b. Central anterior head setae modified in three short hairs	A. ogmorhini
		Host: Hydrurga leptonyx
		Leopard seal
5	a. Six (four marginal and two principal) dorsal posterior long hairs around the posterior border of the head	A. carlinii
		Host: Leptonychotes weddelli
		Weddell seal
	b. Four marginal long hairs and in the basis of the head a line of eight	A. lobodontis
	spines and three hairs above the last row of four spines	Host: Lobodon carcinophaga
		Crabeater seal

Concluding remarks

Sucking lice of the family Echinophthiriidae represent an integral element of the parasite fauna of pinnipeds, with worldwide records in species from all pinniped genera (Leonardi and Palma 2013). Echinophthiriids are peculiar among the Anoplura by their adaptations to the amphibious life of their hosts (Kim 1971; Murray 1976; Mehlhorn et al. 2002), being among the few insects that have been able to adapt to the marine environment (Leonardi and Lazzari 2014). The morphological, physiological and behavioral characteristics of echinophthiriids are the evidence of a long co-evolutionary process (Kim 1985). This association began with the terrestrial ancestors of the actual pinnipeds and the primitive forms of echinophthiriids that they harbored before they ventured into the marine realm. During the evolutionary time, echinophthiriids managed to adapt to the new conditions imposed by their hosts (Kim 1985; Leidenberger et al. 2007; Leonardi et al. 2012).

Revision of Antarctic echinophthirids is part of ongoing research on the systematics, phylogeny and ecology of these lice.

The final objective of our work is to determine the potential use of lice as indicators of both environmental changes and hosts' behavior and biology identifying those associations more sensitive to changes. We are studying the host–parasite association of three species of pack-ice seals and their lice species: the crabeater, the Weddell and the leopard seals around the Antarctic Peninsula. Currently, there is a warming trend in the Antarctic Peninsula region (Smith et al. 1999, 2003; Domack et al. 2003; Vaughan et al. 2003), and in this climate change context, the study of the association of parasites and pinnipeds will be useful to make inferences about potential effect and magnitude of that change in seals' behavior, ecology and population dynamic.

As a practical outcome of this work, we provided a key to the species of Echinophthiriidae from Antarctic hosts' to facilitate the identification on the basis of their main morphological characteristics. **Acknowledgments** The study was financially and logistically supported by the Dirección Nacional del Antártico, Instituto Antártico Argentino. This work's permit was granted by the Dirección Nacional del Antártico (Environmental Office). We are very grateful to Sebastián Poljak, Juan Galliari and Pedro Carlini for their assistance during the fieldwork. Special thanks goes to A. Moyano, L. Cataldo, G. Arce, C. Alvárez, J. Villalba, R. Moyano, M. Echazarreta, E. de los Santos and M.S. Cosenza. We specially thank to Paula Rivera, who helped us to improve the figures. Thanks are given to the Ministerio de Ciencia, Tecnología e Innovación Productiva and Ministerio de Educación de la Nación for the promotion of the Scientific Argentinean Program and the support of public education.

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