

## *Demodex phocidi* (Acariformes: Demodecidae) from *Phoca vitulina* (Carnivora: Phocidae) – the second observation in the world and a supplement to the species description

by

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DOI: [10.1515/ohs-2020-0005](https://doi.org/10.1515/ohs-2020-0005)

Category: **Original research paper**

Received: **May 27, 2019**

Accepted: **July 26, 2019**

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### Abstract

The present study describes a finding of the demodecid mite *Demodex phocidi* in the seal *Phoca vitulina* in the Baltic Sea. This is the first identification in Europe and the second in the world. This is also the first observation of the Demodecidae family in the pinnipeds outside North America. A high density of demodecid mites was observed in the skin of the examined seal, but no symptoms of parasitosis were observed. Our findings also supplement the taxonomic description and morphometry of *D. phocidi*.

**Key words:** Demodecid mites, Pinnipedia, harbor seal, marine mammals, Baltic Sea

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## Introduction

Demodecidae (Acariformes: Prostigmata) are mammal-specific parasites found in representatives of almost all orders and ecological groups. Most of the 125 described species were found in terrestrial mammals, primarily rodents and bats (Izdebska et al. 2019). Only four species were found in aquatic mammals: two species in semiaquatic mammals, i.e. *Demodex lutrae* Izdebska et Rolbiecki, 2014 in the Eurasian otter *Lutra lutra* (Linnaeus, 1758) and *D. castoris* Izdebska, Fryderyk et Rolbiecki, 2016 in the Eurasian beaver *Castor fiber* Linnaeus, 1758, described from Europe (Poland), and two species associated with marine mammals were found in Pinnipedia seals, i.e. *D. phocidi* Desch, Dailey et Tuomi, 2003 in the harbor seal *Phoca vitulina* Linnaeus, 1758 and *D. zalophi* Dailey et Nutting, 1979 in the California Sea lion *Zalophus californianus* (Lesson, 1828) in the United States of North America (Dailey & Nutting 1980; Desch et al. 2003; Izdebska & Rolbiecki 2014a; Rolbiecki & Izdebska 2014; Izdebska et al. 2016). However, *D. phocidi* was described on the basis of material collected from one seal originating from the USA (Alaska, Seward, Sea Life Center; Desch et al. 2003). The present paper confirms the presence of *D. phocidi* in the harbor seal population from the Baltic Sea (Europe) in numbers large enough to enable an analysis of the variability of meristic features. It also presents morphological features that have hitherto been associated with mites from the Cheyletoidea superfamily and the Demodecidae family (Bochkov 2008; Izdebska & Rolbiecki 2018) and have not been included in previous descriptions of species representing this mite family.

## Materials and methods

The study material consisted of harbor seal skin samples from the collection of the Professor Krzysztof Skóra Hel Marine Station of the Institute of Oceanography of the University of Gdańsk in Hel (Poland). The dead seal, found on 29 April 2015 on a beach at the town of Jantar (Pomerania Province; 54°20'39"N; 19°02'06"E), was frozen and then dissected on 08 October 2018. It was a female weighing 49 kg with a total length of 148 cm, including fins.

To identify the presence of skin mites, skin fragments were taken from several areas of the seal body, including the head (eyelid, ear region, cheek, forehead, chin, lips, vibrissae area, vertex, back of the head), regions of the neck, the trunk (abdomen, back), limbs/flippers and the tail. The mites were prepared using the digestion and decantation method (Izdebska

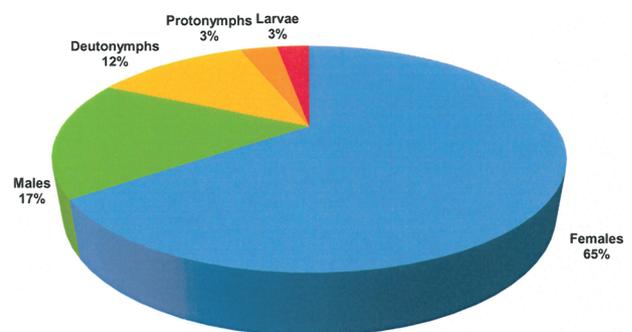
2004). Skin samples were preserved in 70% ethanol and digested in 10% KOH solution. The obtained samples were decanted (examination of 1 cm<sup>2</sup> of skin equal to the analysis of approximately 50–100 wet preparations, i.e. in the liquid state) and analyzed using phase-contrast microscopy (Nikon Eclipse 50i). Specimens were placed in polyvinyl-lactophenol solution and measured (measurements expressed in micrometers). All measurements were taken in the following way: total body length = length of gnathosoma, podosoma and opisthosoma; gnathosomal width = width at the base; podosomal and opisthosomal width = maximum width.

The density of parasites (number of parasites per unit area/skin) was calculated to determine the level of host infestation (Margolis et al. 1982).

## Results

Numerous mites taken from the analyzed harbor seal skin fragments were identified as *D. phocidi*: 214 males, 822 females, 157 deutonymphs, 39 protonymphs, 33 larvae and 30 eggs (Fig. 1). A total of 50 skin fragments (c. 3500 wet preparations) were examined; the presence of demodecid mites was found in 40 of them. The demodecid mites inhabited various parts of the seal body, particularly the hairy skin of the head region and the anterior part of the body (Table 1). Although considerable densities were observed, no symptoms of demodecosis could be found.

A representative sample (200 adult stages, 90 immatures, 30 eggs) was selected from the collected material, representing the best preserved and undamaged specimens that were subjected to



**Figure 1**

Quantitative contribution (%) of adult and immature stages of *Demodex phocidi* collected from *Phoca vitulina* in the Baltic Sea

morphometric analysis based on the body tagmata and various other elements (Tables 2, 3). Subsequently, morphological features considered to be significant

in Demodecidae taxonomy were analyzed and all features included in the species description by Desch et al. (2003) were found.

**Table 1**

Density of *Demodex phocidi* found in the skin of *Phoca vitulina*

Body region	Number of parasites in the examined skin samples	Mean density/1 cm <sup>2</sup> of skin
Hairy skin of the head	373	46.6
Regions of the neck	181	45.3
Back and abdomen	580	36.3
Fore-flippers	117	29.3
Hind-flippers	40	10.0
Tail	4	1.0
Total	1295	32.4

**Table 2**

Metrical features (micrometers) of *Demodex phocidi* adults and eggs collected from *Phoca vitulina* in the Baltic Sea in comparison with the original description

Feature	Present		Desch et al. (2003)	
	♂ (n = 100)	♀ (n = 100)	♂ (n = 12)	♀ (n = 20)
Gnathosoma length	13 (11–15), SD 1	15 (11–18), SD 1	13 (12–13)	15 (15–16)
Podosoma length	47 (40–58), SD 4	55 (43–63), SD 4	43 (40–46)	52 (47–54)
Opisthosoma length	114 (90–138), SD 11	137 (100–175), SD 17	115 (103–134)	117 (104–134)
Gnathosoma width	16 (13–20), SD 2	18 (13–22), SD 2	17 (15–19)	19 (18–22)
Podosoma width	38 (28–50), SD 4	45 (33–53), SD 4	36 (32–40)	45 (40–49)
Opisthosoma width	34 (25–43), SD 4	38 (28–48), SD 4	27 (24–29)	33 (29–38)
Vulva length	–	8 (7–13), SD 1	–	7 (6–7)
Aedeagus length	21 (18–28), SD 2	–	18 <sup>a</sup>	–
Opisthosoma length to body length ratio (%)	65 (59–70), SD 3	66 (60–71), SD 3	67 <sup>b</sup>	67 <sup>b</sup>
Body length to width ratio	4.6:1 (3.4–5.6:1), SD 0.5:1	4.6:1 (3.7–6.2:1), SD 0.5:1	4.8:1 <sup>c</sup>	3.9:1 <sup>c</sup>
Total body length	173 (148–202), SD 11	207 (159–249), SD 19	172 (162–193)	174 (169–203)
Egg	59 (50–65), SD 4 x 30 (23–36), SD 3 <sup>d</sup>		57 (53–60) x 31 (29–32) <sup>e</sup>	
Egg length to width ratio	2.0:1 (1.7–2.6:1), SD 0.2:1		1.8:1 <sup>f</sup>	

a – two aedeagi measured; b, c, f – based on means calculated from Desch et al. (2003); d – 30 eggs measured; e – two eggs measured

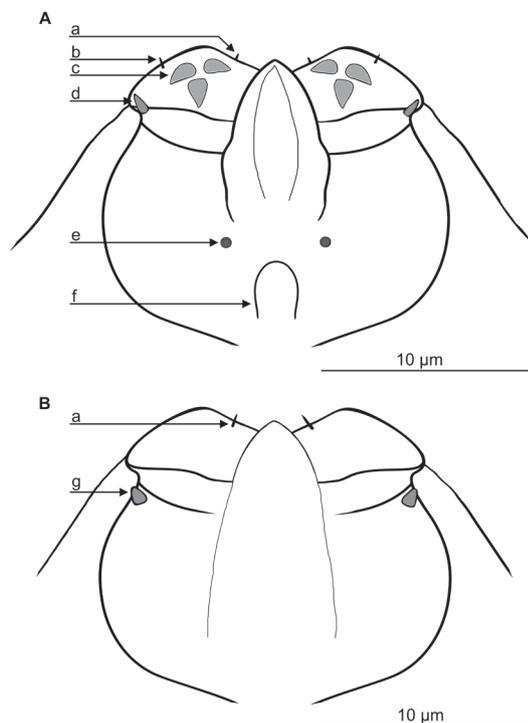
**Table 3**

Metrical features (micrometers) of *Demodex phocidi* immatures collected from *Phoca vitulina* in the Baltic Sea in comparison with the original description

Feature	Present			Desch et al. (2003)		
	Larva (n = 30)	Protonymph (n = 30)	Deutonymph (n = 30)	Larva (n = 3)	Protonymph (n = 4)	Deutonymph (n = 9)
Gnathosoma length	12 (10–13), SD 1	12 (11–13), SD 1	14 (13–20), SD 2	14 (13–15)	15 (15–16)	14 (12–15)
Podosoma length	31 (18–44), SD 6	37 (28–48), SD 5	49 (38–58), SD 5	35 (32–37)	40 (37–43)	56 (47–60)
Opisthosoma length	44 (28–55), SD 6	71 (55–100), SD 13	95 (68–118), SD 13	46 (37–59)	89 (84–93)	96 (62–116)
Gnathosoma width	12 (10–16), SD 1	14 (11–20), SD 2	17 (13–23), SD 2	17 (16–18)	17 (15–19)	18 (16–21)
Podosoma width	26 (15–38), SD 5	31 (25–40), SD 4	37 (28–50), SD 5	26 (21–29)	29 (26–35)	35 (29–41)
Opisthosoma width	23 (15–35), SD 4	28 (23–35), SD 4	32 (23–48), SD 5	23 (21–25)	26 (23–31)	30 (24–37)
Opisthosoma length to body length ratio (%)	50 (42–56), SD 3	59 (51–68), SD 4	60 (52–65), SD 3	48 <sup>a</sup>	62 <sup>a</sup>	58 <sup>a</sup>
Body length to width ratio	3.4:1 (2.2–4.6:1), SD 0.6:1	4.0:1 (2.7–5.3:1), SD 0.7:1	4.4:1 (3.3–5.7:1), SD 0.6:1	3.7:1 <sup>b</sup>	5.0:1 <sup>b</sup>	2.9:1 <sup>b</sup>
Total body length	87 (63–110), SD 12	120 (97–148), SD 17	158 (120–191), SD 16	96 (82–109)	144 (135–148)	165 (128–191)

a, b – based on means calculated from Desch et al. (2003)

Adults of both sexes were found to possess previously undescribed gnathosoma elements, such as supracoxal spines (setae *elc.p*), as well as setae *dG* and *v''F* on trochanter-femur-genu segments and setae *l''G* on tibia-tarsus segments of the palpi. The supracoxal spines were located on the dorsal side of the gnathosoma, in the anterior margin of the coxal segment, at its lateral margin, and were found to take the form of very small rods less than 1  $\mu\text{m}$  in length (Fig. 2). Photographic documentation is provided for adult stages (Fig. 3).



**Figure 2**

*Demodex phocidi*: A – gnathosoma, male, ventral view; B – gnathosoma, male, dorsal view; a – seta *l''G*; b – seta *dG*; c – spines on palps; d – seta *v''F*; e – subgnathosomal seta (seta *n*); f – pharyngeal bulb; g – supracoxal spine (seta *elc.p*)

## Discussion

The present identification of *D. phocidi* represents the first record of its presence in Europe and only the second occurrence in the world. In addition, it is also the first reported finding of Demodecidae in European seals and the first report on their occurrence in pinnipeds outside North America. Our findings confirm the existence of *D. phocidi* as a valid species. Until



**Figure 3**

*Demodex phocidi*: A – male, various morphotypes; B – female, various morphotypes; C – group of specimens

now, its presence was limited to a single observation in a single host individual kept in captivity, in which conditions the transmission of parasites to atypical hosts was sometimes observed. The fact that the host examined in the present study originated from a wild population is therefore an important point to consider.

As Demodecidae are considered to be monoxenic parasites, i.e. those whose evolution has been convergent with the evolution of the host species, the same species can be found in host species in different, sometimes distant, areas of its distribution. For instance, *D. nanus* Hirst, 1918 described from Europe was also found in the brown rat *Rattus norvegicus* (Berkenhout, 1769) in North America (Bukva 1987; 1995; Desch 1987; Desch et al. 2010; Izdebska et al. 2013; Izdebska & Rolbiecki 2014b). In addition, in the case of the harbor seal, the same parasites were found in populations in very distant localities, e.g. the endoparasitic mite *Halarachne halichoeri* (Allman, 1847) and the seal louse *Echinophthirus horridus* (von Olfers, 1816) (Geraci et al. 1981; Thompson et al. 1998; Kadulski 2001; Rolbiecki et al. 2018). Although they are often oligoxenic parasites, i.e. their transmission does not have to be limited to exchanges between

*P. vitulina* populations, local populations may also acquire those parasites from neighboring populations of other seal species. Although *D. phocidi* is thought to be monoxenic and to be transmitted only within *P. vitulina*, the two observations of the species to date are insufficient to confirm this thesis and further studies on the presence of Demodecidae in other members of Phocidae are required.

Although the identified mites are consistent in their features with the description of *D. phocidi* published by Desch et al. (2003), they exhibit a wider range of meristic features, which is probably related to the examination of a larger group. The present study has included measurements of 290 specimens and 30 eggs, while Desch et al. (2003) was able to measure 48 specimens (32 adults and 16 immatures) and two eggs. In addition, our mite specimens were obtained from a different harbor seal population that lives in a remote locality compared to the previous report. Even though Desch et al. (2003) did not specify a subspecies of the examined seal, other subspecies may be found in the area of North America (Berta & Churchill 2012), therefore the present seal may represent a different subspecies compared to the previous host. So far, five subspecies have been distinguished, of which the nominative subspecies, *P. v. vitulina* Linnaeus, 1758, occurs in the Baltic Sea, in Europe and in West Asia. On the other hand, North America was associated with *P. v. concolor* De Kay, 1842 (eastern region), *P. v. richardii* Gray, 1864 (western region) and *P. v. mellonae* Doult, 1942 (North-East Canada; Wilson & Reeder 2005). However, recent genetic analyses indicate that *P. v. concolor* is paraphyletic in relation to *P. v. vitulina* and this form should not be considered as a valid subspecies (Berta & Churchill 2012).

In the light of our present findings, it is recommended that the description of *D. phocidi* should be supplemented with information on the presence of gnathosoma structures. Similar recommendations for their inclusion in the description have been brought by a recent study on the occurrence of structures and other morphological elements in the phylogeny of families representing the Cheyletoidea superfamily (Bochkov 2008; 2009). The present study also demonstrated the presence of supracoxal spines, a structure not reported by Desch et al. (2003).

In the *D. phocidi* specimens obtained in the present study, these spines were found to be extremely small (less than 1  $\mu\text{m}$ ). In addition, their location at the lateral edge and anterior margin of the coxal segment of the gnathosoma makes observation difficult because they are located within the corner formed on the border of the edge and grooves between the segments (Fig. 2). They might have been absent from a small

number of previously examined specimens, and their completeness and preservation status is difficult to assess. Although certain morphological differences may occur between specimens from geographically-distant demodecid mite populations and from hosts classified as distinct subspecies, supracoxal spines constitute a significant feature. They have been indicated by Bochkov (2008) as a diagnostic feature for the family Demodecidae and are commonly observed in other known species.

It should be added that while the previous report on *D. phocidi* infestation related to a seal displaying showing skin symptoms caused by mite infection, the infestation currently described was asymptomatic. This is a typical phenomenon for Demodecidae, which rarely manifest their presence in the form of parasitosis/demodectosis, making it undoubtedly difficult to detect in hosts. Skin symptoms are typically only observed in cases of high parasite density, however, hosts can display high tolerance to these parasites and despite the relatively high local density observed in the present seal (c. 47 per 1  $\text{cm}^2$ ), no symptoms were observed. Similar observations have been reported for many other mammal mites, such as *D. lutrae* in *L. lutra* or *D. melesinus* Hirst, 1921 in the European badger *Meles meles* (Linnaeus, 1758), where the host may also show high densities and remain asymptomatic (Izdebska & Rolbiecki 2014a; Izdebska et al. 2018).

The fact that Demodecidae infestations are usually asymptomatic undoubtedly complicates their detection in host populations and is one of the reasons for the paucity of available data on their occurrence, particularly in wild mammal populations. In the case of demodecid mites in pinnipeds, an additional constraint on researchers is the difficulty of gaining access to the material, as increasing numbers of these species are endangered and protected. This is certainly the case for *P. vitulina*, which is a relatively rare species for the Baltic Sea, with a population estimated at only around 800 individuals (<http://www.hel.ug.edu.pl/animals/fp.html>). Of all the observations of the Baltic seals, 69% are the grey seal *Halichoerus grypus* (Fabricius, 1791), while only 6% are the harbor seal and 4% are the ringed seal *Pusa hispida* (Schreber, 1775), and the remaining 21% are unidentified seal species (Pawliczka 2016).

An additional constraint in research on the seal mite fauna, compared with research on other mammals based on dead specimens found in the field, is their aquatic habitat: dead seals found on beaches typically exhibit different levels of decomposition, which constitutes a considerable impediment to the study of dermal parasites. However, as shown by

studies of Demodecidae in other mammals, further research into the occurrence of skin mites in seals will provide more data to confirm their common occurrence in host populations with different distributions.

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