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# Two new species of parasitic demodecid mites in the European polecat *Mustela putorius* and their co-infestation with *Miridex putorii* (Acariformes: Demodecidae)

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## Abstract

The biodiversity of parasitic mites of the Demodecidae, infesting mammalian carnivorans, is poorly understood. To date, 18 host-specific species have been described, including four each from domestic dogs and cats, and only 10 from wild carnivores, known from single or duplicate reports. No data is available on the level of infestation of wild populations, or the co-occurrence of different demodecids in the same host, as only single species have been identified in individual hosts. A convenient model for such studies turned out to be the European polecat *Mustela putorius*, in which a new genus and species, *Miridex putorii*, was recently described in the vibrissae region of the skin of the head. Our study reveals that *M. putorii* co-occur with other species: *Demodex putorii* sp. nov. (associated with the hairless skin of the head) and *Demodex foetorii* sp. nov. (associated with hairy skin). Thus, the present study provides descriptions of species new to science. It also provides the first analysis of the occurrence of Demodecidae in wild mammalian carnivorans at the species and individual animal level based on the co-occurrence of three demodecid mite species in the skin of the head in *M. putorius*. Demodecid mites were found in 75.7% of 37 polecats; *M. putorii* showed the highest prevalence (56.8%), and *D. putorii* showed the highest abundance in the skin (mean 10.8 mites in 9 cm<sup>2</sup>). The three species co-occurred in 5.4% of the polecats, and two species in 27.0%. Despite the very high abundances, infestation was not associated with gross skin pathology (a feature of stable parasite–host systems formed by long-term co-evolution). The co-occurrence of several species and the separation of microhabitats are also typical of Demodecidae of other mammalian groups and illustrate the optimal use of the host body as a habitat and food source with limited impact.

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**Keywords:** *host-parasite interactions, skin mites, taxonomy, wild mammals*

## Introduction

The Demodecidae (Acariformes: Prostigmata) are among the smallest parasitic arthropods; however, they are also among the least understood due to their lifestyle hidden in the skin or in other mammalian tissues and structures. They are likely to be common in host populations, exhibiting high host specificity (Izdebska & Rolbiecki 2020; Cierocka et al. 2022). In

individual host species, there may be several particular species of these mites, associated with different microhabitats: they have been found in various skin structures, tissues and organs, including normal and sensory hair follicles, various glands, ear canals, eyeball, tongue and gums (Izdebska & Rolbiecki 2020). At the same time, each species shows a clear preference regarding

location, inhabiting different regions of hairy skin (e.g. only the head, or only the limbs, abdomen, or back), sparsely hairy or hairless skin (e.g. the region of the lips or nose, hairless skin of the feet, the flight membranes of bats, genital or anal areas). Topical (related to the specific microhabitat) and topographic (distribution in various regions of the body) preference and the co-occurrence of synhospital Demodecidae species are mostly observed in rodents, especially cosmopolitan and synanthropic species like the house mouse *Mus musculus* Linnaeus, 1758 and the brown rat *Rattus norvegicus* (Berkenhout, 1769), or the widely distributed *Apodemus* spp. (Izdebska & Rolbiecki 2013a, 2013b, 2014b). There is little information on their occurrence in other mammals, and in the case of carnivorans, they refer only to the domestic dog *Canis lupus familiaris* Linnaeus, 1758 and the domestic cat *Felis catus* Linnaeus, 1758, in which four different species each have been described, showing different location preferences (Desch & Nutting 1979; Desch & Hillier 2003; Izdebska 2010; Izdebska & Rolbiecki 2018).

However, data from domestic animals do not necessarily provide an appropriate model for parasitological analyses for most members of the Demodecidae that are associated with wild mammals. In domestic mammals, infestation often has a different course, associated with the appearance of the disease symptoms of demodicosis (demodicosis), resulting from excessive multiplication of a particular demodecid species or, less frequently, from infestation by two or more species. Domestic animals live under different (artificial) environmental conditions, and such host–parasite systems probably have a much shorter evolutionary pedigree than in wild mammals; hence, this affects its instability – a greater chance of crossing the tolerance threshold of the host and the subsequent development of parasitosis (Izdebska et al. 2023).

However, in wild mammals, this type of analysis is hampered by a very poor understanding of the biodiversity of their Demodecidae. In individual hosts, only single representatives, usually of the genus *Demodex*, have so far been described based on isolated or sparse findings (Izdebska & Rolbiecki 2020). In the light of current research, a suitable example for such study has proved to be the European polecat *Mustela putorius* Linnaeus, 1758 (Carnivora: Mustelidae), in which a new species and genus *Miridex putorii* Izdebska, Rolbiecki et Rehbein, 2022 has recently been described residing in the vibrissae (Izdebska et al. 2022). The species is characterized by unusual structural features, making it one of the more mysterious and exceptional representatives of mites (Izdebska et al. 2022).

The aim of the current study was to describe two new species of Demodecidae which are typical and probably specific parasites of *M. putorius* and have been discovered in further research on the skin samples that resulted previously in the discovery of *Miridex putorii* (Izdebska et al. 2022). This study also provides the first analysis of the occurrence of Demodecidae in a mammalian carnivore based on the co-occurrence of three mite species from two genera in the skin of European polecat, both at the mite species level and at the individual host level. In addition, data on host associations and the global distribution of carnivore demodecid mites are updated.

## Material and methods

Skin from the head and adjacent neck area of 37 European polecats was collected during a survey of the animals' parasitic fauna in Germany from October 2013 to August 2015 (Table I) and preserved in 70% ethanol. The animals were trapped according to the hunting regulations in Germany, then placed in separate plastic bags and frozen (Kretschmar 2016).

Nine ~1 cm<sup>2</sup> fragments were taken from each skin sample representing eight head regions, viz. the area around the eyes, nose, area of vibrissae, lips, chin, cheeks, ear pinnae and vertex, plus the adjacent neck area (total ~9 cm<sup>2</sup> per animal). For the recovery of demodecid mites, skin fragments were individually digested in 10% potassium hydroxide solution as described previously (Izdebska 2004). The digest material was decanted and examined under phase-contrast microscopy (Nikon Eclipse 50i) with 1 cm<sup>2</sup> of skin sample yielding approximately 100 wet preparations. The mites were placed in polyvinyl-lactophenol solution. The following measurements were taken: total body length = length of gnathosoma, podosoma and opisthosoma; gnathosomal width (at base); podosomal and opisthosomal width (maximum width). All measurements are given in micrometers (µm).

The number of eggs and mite stages (larva, protonymph, deutonymph, adult male, adult female) per 1 cm<sup>2</sup> for each mite species were identified. The total number of specimens was counted to estimate the abundance of infestation as the number of mites (larvae + nymphs + adult mites) per examined host/skin samples.

To assess the relationship between presence of mite infestation (interpreted as prevalence – percentage of hosts infested with mites) and host demographic factors, all 37 polecats were considered, which

Table I. Sampling details for the European polecats examined for demodcid mites.

Host catalog no.	Locality (federal state, county)	Collection date	Sex (age)	Weight (g)	Demodcid mites	
75	Lower Saxony, Aurich, 53°28'15"N, 07°28'59"E	03 November 2014	M (1)	906.2	Mp	
76		18 December 2014	M (2)	1250.0	Mp	
80		03 January 2015	M (1)	989.6	Df, Dp, Mp	
81		13 November 2014	W (2)	779.4	–	
82		04 October 2014	M (2)	1278.1	Dp, Mp	
83		04 February 2015	M (1)	755.5	–	
85		20 January 2015	M (2)	1011.3	Mp	
89		01 October 2013	W (1)	573.6	Mp	
90		20 December 2014	W (2)	614.3	Dp, Mp	
95		14 December 2014	W (1)	628.4	–	
99		14 February 2014	W (1)	528.5	Df	
101		14 January 2014	M (2)	980.2	Mp	
110		Northrhine Westfalia, Bonn, 50°44'00"N, 07°06'00"E Northrhine Westfalia, Borken, 52°02'07"N, 06°49'28"E	18 August 2015	W (1)	622.4	–
79			28 December 2014	M (3)	1600.6	Mp
84	17 January 2015		M (3)	1221.2	Df, Dp	
86	29 December 2014		W (2)	740.3	–	
87	13 February 2015		M (2)	1647.5	Mp	
92	08 December 2014		M (2)	1185.4	Mp	
93	03 November 2014		M (2)	1477.5	Df, Dp	
96	07 November 2014		M (2)	1118.3	Mp	
97	05 February 2015		M (1)	1194.1	Dp, Mp	
98	01 November 2014		M (2)	1420.1	Dp, Mp	
102	Northrhine Westfalia, Heinsberg, 51°06'00"N, 06°09'00"E		09 December 2013	M (2)	1017.2	Df, Dp, Mp
111			18 August 2014	W (2)	672.3	Dp, Mp
112			06 August 2014	M (1)	688.6	–
113			14 February 2014	M (3)	1648.2	Dp
114		19 September 2014	W (2)	705.5	–	
115		18 December 2014	W (3)	901.9	Df	
116		26 November 2014	W (3)	697.7	Mp	
117		05 November 2014	M (2)	1297.7	Dp	
118		07 February 2015	M (2)	1220.3	Mp	
106		Hessia, Hersfeld-Rotenburg, 50°53'14"N, 10°00'20"E	15 October 2014	W (1)	730.0	–
105		Hessia, Wetteraukreis, 50°26'06"N, 08°40'08"E	12 February 2014	M (2)	1137.0	Dp, Mp
77		Thuringia, Nordhausen, 51°35'12"N, 10°39'46"E	?	M (1)	1081.0	Df, Mp
78		Thuringia, Wartburgkreis, 51°35'12"N, 10°39'46"E	?	M (1)	986.0	Df, Dp
94		Bavaria, Nürnberger Land, 50°45'31"N, 12°42'36"E	03 October 2013	M (1)	962.9	Mp
100	29 September 2014		M (1)	671.3	–	

M: male, F: female.

Age categories: (1) "juvenile": younger than 1 year; (2) "young adult": second and third year of life; (3) "old": older than 3 years.

Df: *Demodex foetorii* sp. nov., Dp: *Demodex putorii* sp. nov., Mp: *Miridex putorii*.

comprised 14 "juvenile" animals ( $\leq 1$  year; nine male, five female), 18 "young adult" animals ( $\sim 2$ –3 years old; 13 male, five female) and five "old" animals ( $> 3$  years old; three male, two female) (see Table I).

Associations between mite presence and variables representing host demographic factors (age

group, sex) were assessed using contingency tables and Fisher's exact test. Abundance of infestation (total demodcid mite count between sexes of polecats per age group) was analyzed using the Mann Whitney U-test. All testing was two-sided, and the level of significance for all analyses was set at  $p < 0.05$ .

The specimen depository is cited using the following abbreviation: UGDIZP, University of Gdańsk, Department of Invertebrate Zoology and Parasitology, Gdańsk, Poland (Zhang 2018).

The description of the species adopted the nomenclature commonly used for the family Demodecidae (Nutting 1976) and was completed with the nomenclature proposed by Bochkov (2008) for the superfamily Cheyletoidea (Acariformes: Prostigmata) and by Izdebska and Rolbiecki (2016). The scientific and common names of the hosts follow Wilson and Reeder (2005) and the Integrated Taxonomic Information System (2022).

Species descriptions were prepared including all specimens, and morphometric features were analyzed based on all (14 females and 10 males) *D. foetorii* specimens and 140 selected *D. putorii* specimens (100 females and 40 males) from different locations and different host individuals.

**Results**

*Infestation, and demodecid mites found in the European polecats*

A total of 629 specimens (including eggs) of Demodecidae were recovered from the examined skin samples, including 204 *Miridex putorii*, 401 *Demodex putorii* sp. nov. and 24 *D. foetorii* sp. nov. The overall prevalence of infestation (including animals infested with at least one of the three mite species) was 75.7% with the abundance (mean, median,

range) of mites 16.6, 4.0 (1–366) (Table II). The highest prevalence was exhibited by *M. putorii* (56.8%), with the highest abundance by *D. putorii* sp. nov. (10.8, 0.0, 1–360), where the most specimens (360) were found in one "old" male polecat from Northrhine Westfalia/county Borken; however, no skin lesions related to the infestation were observed.

Significant differences were found with regard to the prevalence of infestation with the three mite species (Fisher’s exact test;  $p = 0.007$ ) – *M. putorii* was recorded significantly more often than *D. foetorii* sp. nov. ( $p = 0.039$ ); however, there was no statistically significant ( $p \geq 0.1$ ) difference in the prevalence of *M. putorii* (56.8%) and *D. putorii* sp. nov. (35.1%) and of *D. foetorii* sp. nov. (21.6%) and *D. putorii* sp. nov. (35.1%), respectively.

Sixteen polecats (43.2%) exhibited single-species infestation. Ten (27.0%) animals demonstrated the co-occurrence of two demodecid mite species: *M. putorii* and *D. foetorii* sp. nov. in one animal, *M. putorii* and *D. putorii* sp. nov. in six animals, *D. foetorii* sp. nov. and *D. putorii* sp. nov. in three animals. Two animals (5.4%) were infested with mites of all three species (Table I).

While all ontogenetic stages were recovered for *M. putorii* and *D. putorii* sp. nov., only adult mites were isolated for *D. foetorii* sp. nov. Some variation in the adult male to female ratio was observed for the three mite species in individual hosts; in particular, a female predominated ratio was observed for *D. putorii* sp. nov. (1:5.5) (Table III, Figure 1).

Table II. Presence (prevalence, mean, median, and range abundance) of demodecid mites in nine ~1 cm<sup>2</sup> skin fragments from the head (eight fragments) and adjacent neck (one fragment) of *Mustela putorius*.

Mite species	European polecats			
	“Juvenile” (≤1 year, n = 14) N mite+	“Young adult” (~2 and 3 years, n = 18) N mite+	“Old” (>3 years, n = 5) N mite+	All animals (n = 37) N mite+ (infection)
<i>Miridex putorii</i>	6	13	2	21 (56.8%, 5.5, 1.0, 1–36)
<i>Demodex foetorii</i> sp. nov.	4	2	2	8 (21.6%, 0.6, 0.0, 1–10)
<i>Demodex putorii</i> sp. nov.	3	8	2	13 (35.1%, 10.8, 0.0, 1–360)
Total demodecid mites ( <i>Miridex putorii</i> , <i>Demodex foetorii</i> sp. nov., <i>Demodex putorii</i> sp. nov.)	8 (57.1%, 5.5, 2.0, 1–22)	15 (83.3%, 6.3, 4.5, 1–21)	5 (100%, 83.3, 19.0, 1–366)	28 (75.7%, 16.6, 4.0, 1–366)

N mite+: number of mite-positive animals.

“Total demodecid mites” refers to animals infested with at least one of the three mite species.

Table III. Recovery of demodecid mite eggs and ontogenetic stages (larvae, nymphs, adults) in the areas of head/adjacent skin (~1 cm<sup>2</sup> skin fragments each) of *Mustela putorius*.

Skin location	Number of mite positive animals – number of eggs and ontogenetic stages isolated			Total demodecid mites
	<i>Miridex putorii</i>	<i>Demodex foetorii</i> sp. nov.	<i>Demodex putorii</i> sp. nov.	
Area around the eyes	2 – 1L, 1F	1 – 1M	3 – 1E, 1L, 1DN, 2M, 17F	5 – 1E, 2L, 1DN, 3M, 18F
Nose	5 – 1L, 2PN, 10DN, 4M, 5F	0	1 – 3DN, 1F	5 – 1L, 2PN, 13DN, 4M, 6F
Area of vibrissae	18 – 11E, 4L, 3PN, 27DN, 56M, 53 F	0	3 – 2M, 3F	18 – 11E, 4L, 3PN, 27DN, 58M, 56 F
Lips	4 – 2PN, 2DN, 1M	0	5 – 4L, 3PN, 9DN, 24M, 116F	9 – 4L, 5PN, 11DN, 25M, 116F
Chin	3 – 1L, 3PN, 3DN, 7M, 7F	3 – 2M, 3F	4 – 2E, 3L, 2PN, 26DN, 25M, 137F	7 – 2E, 4L, 5PN, 29DN, 34M, 147F
Cheek	0	2 – 2M, 4F	1 – 1DN, 15F	2 – 1DN, 2M, 19F
Ear pinnae	0	3 – 1M, 4F	0	3 – 1M, 4F
Vertex	0	2 – 4M, 3F	0	2 – 4M, 3F
Neck	0	0	1 – 1DN, 2F	1 – 1DN, 2F
Total skin	21 – 11E, 7L, 10PN, 42DN, 68M, 66F	8 – 10M, 14F	13 – 3E, 8L, 5PN, 41DN, 53M, 291F	28 – 14E, 15L, 15PN, 83DN, 131M, 371F

E: egg(s), L = larva(e), PN: protonymph(s), DN: deutonymph(s), M: male(s), F: female(s).

“Total demodecid mites” refers to animals infested with at least one of the three mite species.

Regarding the location of occurrence, *M. putorii* and *D. putorii* sp. nov. were predominantly recovered from the skin surrounding the mouth (anterior part of the head – area of vibrissae, lips, chin, nose) while *D. foetorii* sp. nov. were mainly isolated from the skin of the posterior parts of the head (chin, cheek, ear pinnae, and vertex). In cases of infestation with single mite species (*M. putorii*, 21; *D. foetorii* sp. nov., 8; *D. putorii* sp. nov., 13), *M. putorii* and *D. putorii* sp. nov. were more often recorded from the skin of the anterior parts of the head than the posterior: 21/21 (anterior) vs. 2/21 (posterior),  $p = 0.0001$  and 12/13 (anterior) vs. 3/13 (posterior),  $p = 0.0002$ , respectively. No such differences were observed for *D. foetorii* sp. nov.: 5/8 (anterior) vs. 3/8 (posterior),  $p = 0.6193$  (Table III, Figure 2).

For total demodecid mite infestation (including animals infested with at least one of the three mite species) with respect to host age group, prevalence of infestation was higher among combined “young adult” plus “old” animals (i.e. sexually mature, capable of reproducing)

than for “juvenile” polecats (sexually immature) (20/23 vs. 8/14,  $p = 0.0569$ ). In addition, male polecats were significantly more likely to be infested than female polecats: all animals – 22/25 (male) vs. 6/12 (female),  $p = 0.0355$ ; combined “young adult” plus “old” animals – 16/16 (male) vs. 4/7 (female),  $p = 0.0198$ . Pairwise comparison of the total mite abundance by sex of the polecats demonstrated that “young adult” male animals harbored significantly more mites than “young adult” females ( $p = 0.02$ ), and that the total male polecats harbored significantly more mites than total females ( $p = 0.0063$ ). No significant difference in the abundance of demodecid mite infestation was noted between “juvenile” male and “juvenile” female animals ( $p > 0.1$ ); counts of “old” animals were not analyzed separately because of small sample size (Table IV).

### Systematics

*Demodex putorii* sp. nov. Izdebska, Rolbiecki et Rehbein (Tables V, VI, Figures 3–6)

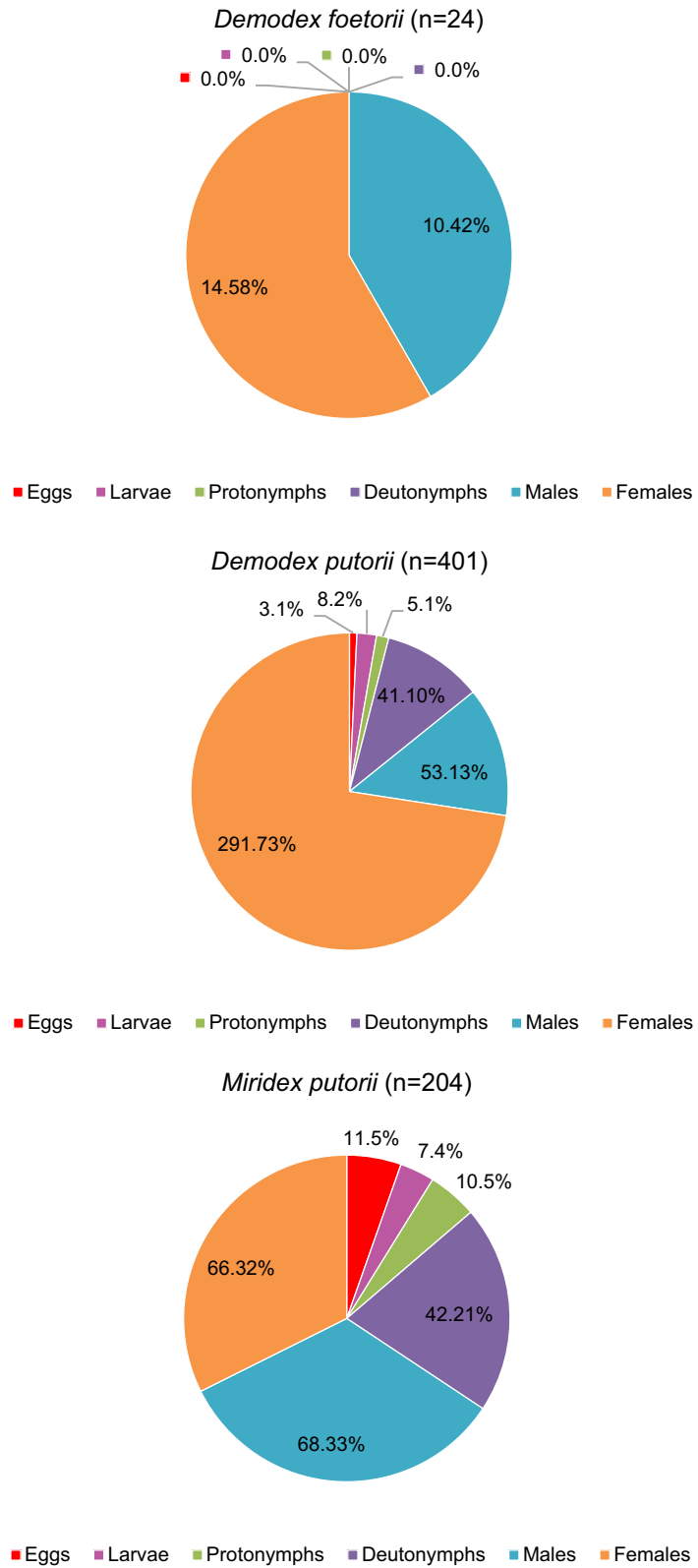


Figure 1. Population structure (number, %) of demodecid mites in European polecat.

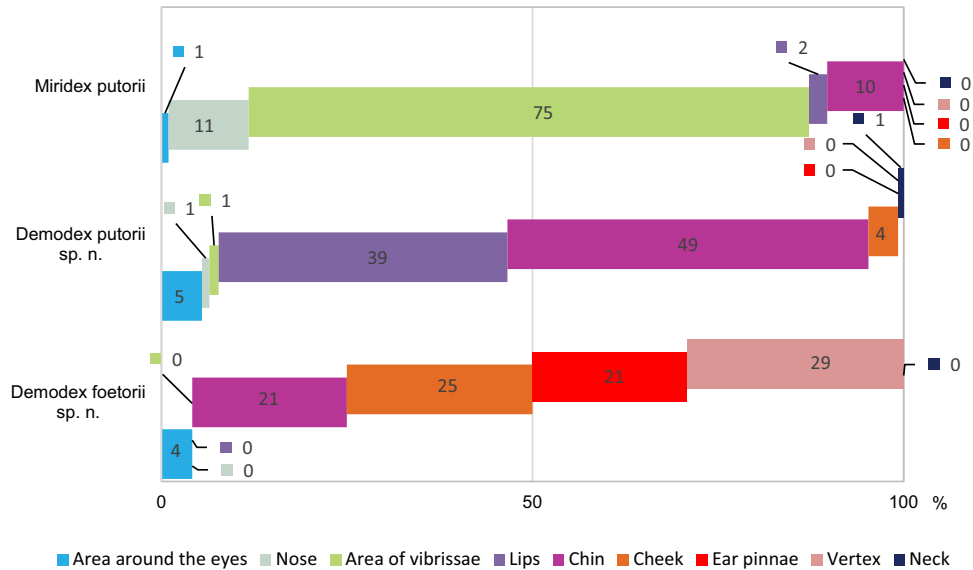


Figure 2. Topography (%) of demodecid mites in the European polecat.

Table IV. Number of demodecid mites (larvae + nymphs + adult mites; combined *Miridex putorii*, *Demodex foetorii* sp. nov. and *Demodex putorii* sp. nov.) per 9 cm<sup>2</sup> skin of head and adjacent neck of *Mustela putorius*.

Age group	Sex, N mite+/NE	Number of mites per 9 cm <sup>2</sup> skin, mean ± standard deviation (range), median
"Juvenile" (≤1 year)	Male, 6/9	7.89 ± 8.15 (3–22), 5
	Female, 2/5	1.20 ± 2.17 (1–5), 0
"Young adult" (~2 and 3 years)	Male + Female, 8/14	5.50 ± 7.3 (1–16), 2
	Male, 13/13	8.08 ± 6.56 (1–21), 7
	Female, 2/5	1.80 ± 2.68 (3–6), 0
"Old" (>3 years)	Male + Female, 15/18	6.33 ± 6.36 (1–21), 4.5
	Male, 3/3	132.67 ± 197.61 (2–366), 36
	Female, 2/2	10.0 ± 12.73 (1–19), 10
Total	Male + Female, 5/5	83.60 ± 155.18 (1–366), 19
	Male, 22/25	22.96 ± 70.73 (1–366), 7
	Female, 6/12	2.92 ± 5.48 (1–19), 0.5
Male + Female, 28/37		16.62 ± 58.63 (1–366), 4

N mite+/NE: number of mite positive animals/number of animals examined.

Table V. Body size (µm) for adults of *Demodex putorii* sp. nov.

Morphologic features	Males (n=40) mean (range) ± SD	Females (n=100) mean (range) ± SD
Length of gnathosoma	18 (16–20) ± 1	21 (16–24) ± 2
Width of gnathosoma (at base)	18 (16–20) ± 1	21 (17–24) ± 2
Length of podosoma	55 (48–63) ± 4	69 (60–93) ± 4
Width of podosoma	31 (27–35) ± 2	35 (30–41) ± 2
Length of opisthosoma	130 (110–150) ± 11	187 (150–215) ± 13
Width of opisthosoma	30 (24–33) ± 2	34 (27–41) ± 2
Aedeagus	23 (19–27) ± 2	–
Vulva	–	9 (5–11) ± 1
Total length of body	203 (177–229) ± 14	278 (231–309) ± 16

SD – standard deviation.



Table VI. Body size ( $\mu\text{m}$ ) for immature stages of *Demodex putorii* sp. nov.

Morphologic features	Larvae ( $n=8$ )	Protonymphs ( $n=12$ )	Deutonymphs ( $n=39$ )
	Mean (range) $\pm$ SD	Mean (range) $\pm$ SD	Mean (range) $\pm$ SD
Length of gnathosoma	12 (8–14) $\pm$ 2	14 (12–20) $\pm$ 2	18 (15–22) $\pm$ 1
Width of gnathosoma (at base)	14 (9–19) $\pm$ 3	18 (15–22) $\pm$ 2	22 (14–26) $\pm$ 3
Length of podosoma	25 (12–29) $\pm$ 5	39 (30–56) $\pm$ 7	62 (48–74) $\pm$ 6
Width of podosoma	25 (10–33) $\pm$ 7	28 (20–35) $\pm$ 6	41 (32–55) $\pm$ 5
Length of opisthosoma	52 (35–78) $\pm$ 14	107 (50–145) $\pm$ 26	173 (125–242) $\pm$ 28
Width of opisthosoma	22 (9–30) $\pm$ 6	24 (15–32) $\pm$ 6	36 (25–45) $\pm$ 5
Total length of body	89 (55–121) $\pm$ 19	160 (105–195) $\pm$ 28	253 (192–331) $\pm$ 30

SD – standard deviation.

Female ( $n = 288$  and 1 holotype). Body slender, elongated, spindle-shaped, distinctly separated gnathosoma, 278  $\mu\text{m}$  (231–309  $\mu\text{m}$ ) long and 35  $\mu\text{m}$  (30–41  $\mu\text{m}$ ) wide (holotype, 279  $\times$  33  $\mu\text{m}$ ). Gnathosoma trapezoidal with length close to width at base; on dorsal side of basal segments at the external edges, pair of small, conical supracoxal spines (setae *elc.p*) present, ca. 1.0–1.5  $\mu\text{m}$  long (holotype, 1.0  $\mu\text{m}$ ), directed outwardly. Palps 3-segmented, terminating in three spines (one small, conical, two larger, including one bifurcated) on tibio-tarsus; also setae *v*“*F*” present on middle segment (trochanter-femur-tarsus). On ventral surface of gnathosoma, horseshoe-shaped pharyngeal bulb with pair of very small (difficult to observe), conical subgnathosomal setae (setae *n*), situated anterior on both sides. Podosoma rectangular; four pairs of short legs, with coxa integrated into ventral idiosomal wall and five free, overlapping segments (trochanter-tarsus); two bifurcated claws, ca. 5.0  $\mu\text{m}$  long (holotype, 5.0  $\mu\text{m}$ ), with large, curved subterminal spur and large, rounded bulge on each tarsus; two small knobs at base of each claw; also one small solenidion (*o*) on each leg. Epimeral plates (coxal fields) distinctly sclerotized; all epimeral plates connect medially; pair I triangular, pair II trapezoidal, pairs III–IV rectangular; posterior edges of pair IV form slight arched incision. On dorsal side of podosoma, podosomal shield with distinctly vertical striation, reaches level of legs III. Opisthosoma constitutes 67 (61–70%) of body length (holotype, 67%); clearly elongated, conical, pointed at end. Whole opisthosoma distinctly annulated; annulations reach posterior edge of podosomal shield on dorsal side of podosoma; annuli relatively wide at ca. 1.0–1.5  $\mu\text{m}$ . Opisthosomal organ absent. Vulva 9  $\mu\text{m}$  (5–11  $\mu\text{m}$ ) long (holotype, 10  $\mu\text{m}$ ); located posterior to the incision of IV epimeral plates.

Male ( $n = 40$ ). Distinctly smaller than female, 203  $\mu\text{m}$  (177–229  $\mu\text{m}$ ) long and 31  $\mu\text{m}$  (27–35  $\mu\text{m}$ ) wide, but of similar shape. Gnathosoma shape

similar to female, but smaller. Pharyngeal bulb and morphological details of gnathosoma similar to those in female. Shape of podosoma and legs also similar to those in female, but epimeral plates I connect at one point, pairs II–IV separated. On dorsal side of podosoma, podosomal shield with distinctly vertical striation, reaches level of legs III. Opisthosoma relatively long, smaller than females, constitutes 64% (60–68%) of body length; distinctly annulated; annuli relatively wide at ca. 1.0–1.5  $\mu\text{m}$ . Opisthosomal organ absent. Aedeagus slender, elongated 23  $\mu\text{m}$  (19–27  $\mu\text{m}$ ) long, on dorsal surface, located between epimeral plates II and III. Genital opening located on dorsal surface, slightly above the border between epimeral plates I and II.

Egg ( $n = 3$ ). Non operculate, spindle-shaped, 79  $\mu\text{m}$  (74–84  $\mu\text{m}$ ) long and 20  $\mu\text{m}$  (19–20  $\mu\text{m}$ ) wide; shell surface smooth.

Larva ( $n = 8$ ). Club-shaped, 89  $\mu\text{m}$  (55–121  $\mu\text{m}$ ) long and 25  $\mu\text{m}$  (10–33  $\mu\text{m}$ ) wide. Gnathosoma distinctly separated, trapezoidal with length shorter than width at base. Supracoxal spines peg-like, small. Palp segments clearly separated, slender, terminating in three claw-like spines. Podosoma with three pairs of unsegmented, clearly separated legs; each leg equipped with two small 3-pointed claws; also three pairs of oval, weakly outlined, ventral scutes, located between I–III pairs of legs present. Opisthosoma conical, relatively short, constitutes 58% (48–64%) of body length.

Protonymph ( $n = 12$ ). Protonymph similar to larva but larger and more slender, 160  $\mu\text{m}$  (105–195  $\mu\text{m}$ ) long and 28  $\mu\text{m}$  (20–35  $\mu\text{m}$ ) wide. Gnathosoma distinctly separated, trapezoidal, similar to those in larvae but more massive. Morphological details of gnathosoma similar to those in larvae. Podosoma with three pairs of unsegmented, clearly separated

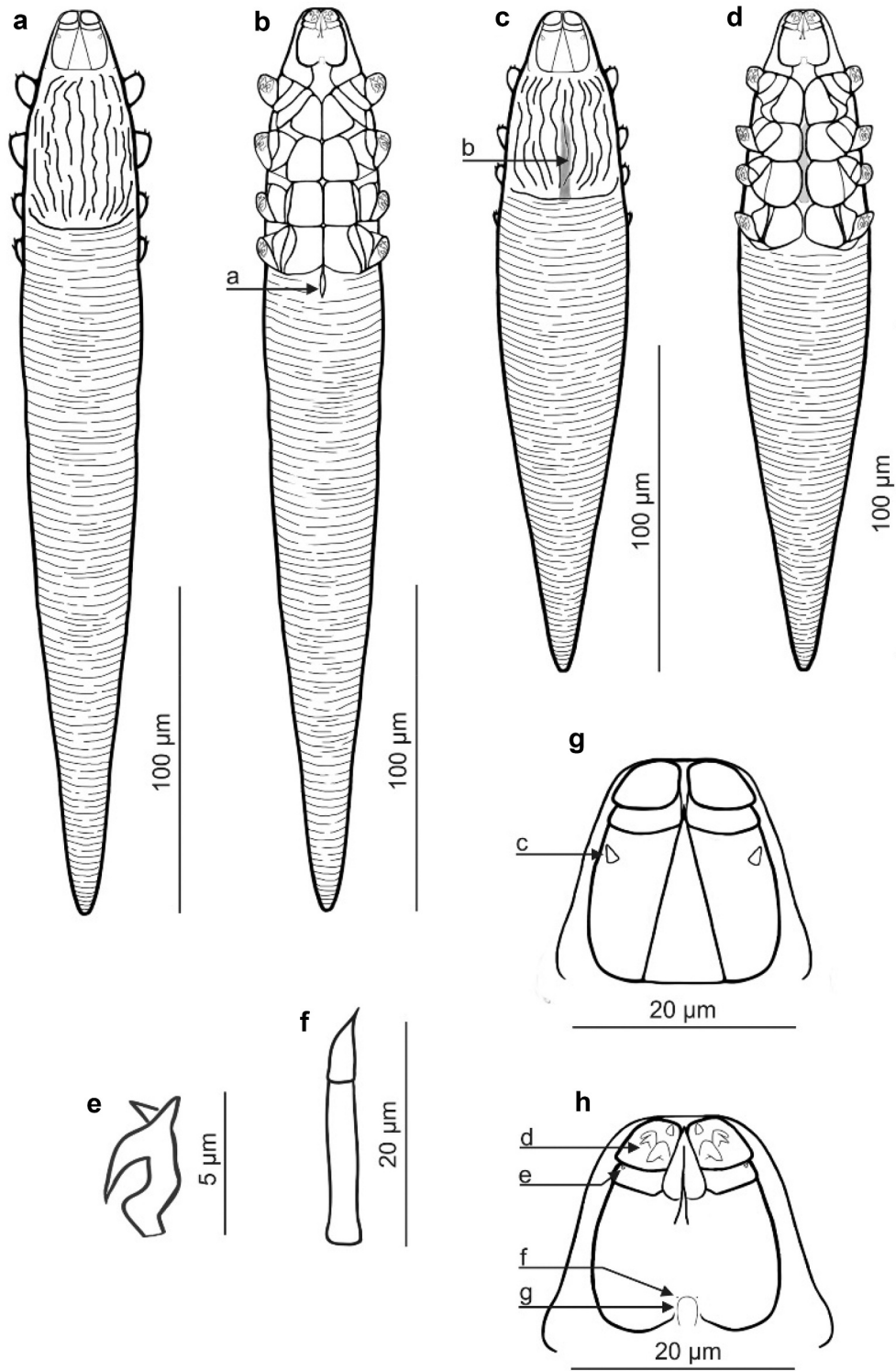


Figure 3. *Demodex putorii* sp. nov., female, dorsal view (a); female, ventral view (b); male, dorsal view (c); male, ventral view (d); claw on the leg (e); aedeagus (f); gnathosoma, female, dorsal view (g); gnathosoma, female, ventral view (h). Abbreviations: a – vulva, b – aedeagus, c – supracoxal spine (seta *elc.p*), d – spines on palps, e – seta *v*“F”, f – subgnathosomal seta (seta *n*), g – pharyngeal bulb.

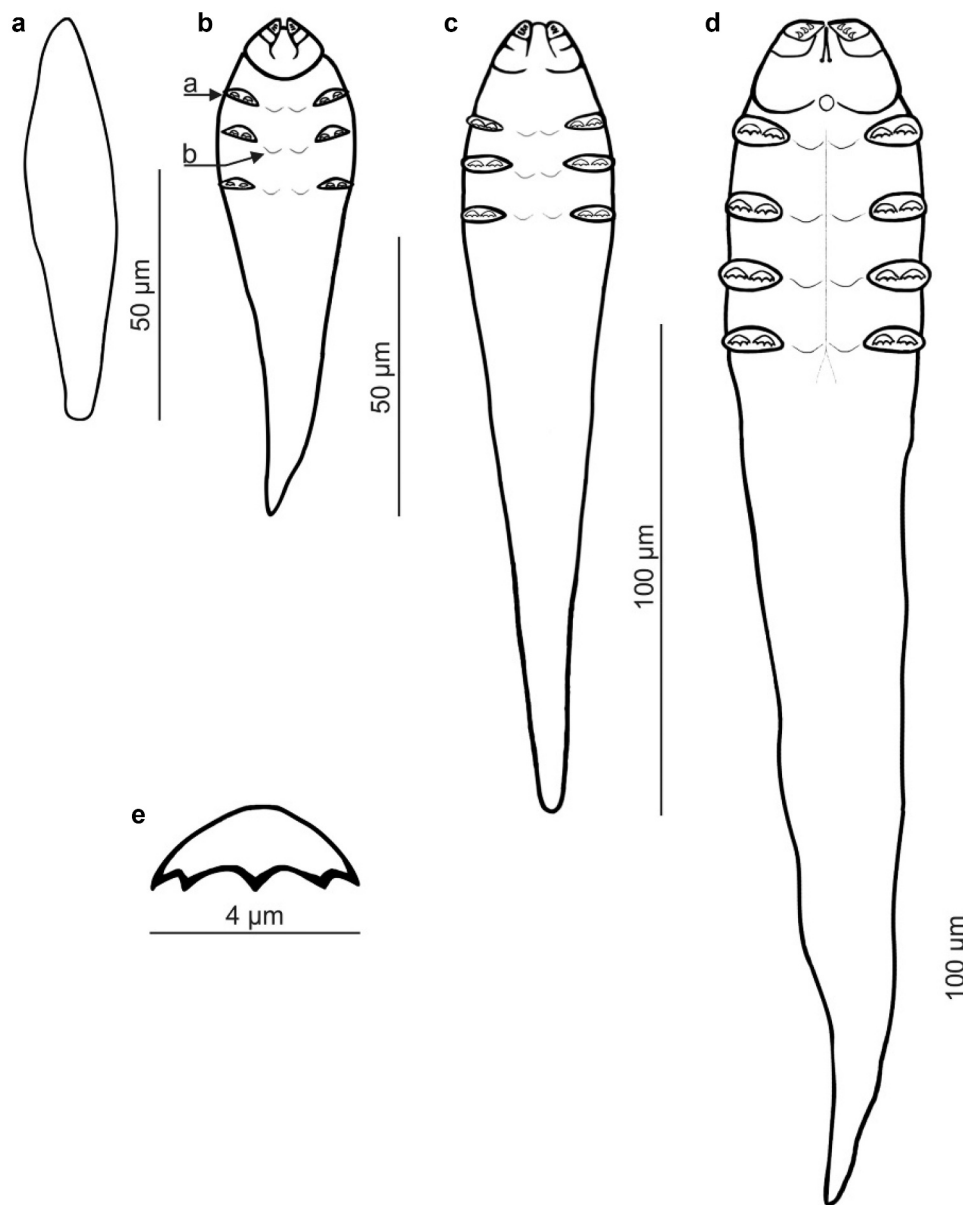


Figure 4. *Demodex putorii* sp. nov., egg (a); larva, ventral view (b); protonymph, ventral view (c); deutonymph, ventral view (d); claw (e). Abbreviations: a – leg with claws, b – ventral scutum.

legs; each leg equipped with two small denticled claws, larger (2–3 µm wide) than in larvae; also three pairs of oval ventral scutes, located between I–III pairs of legs present. Opisthosoma conical, elongated, constitutes 66% (48–76%) of body length.

Deutonymph (n = 39). Deutonymph similar to protonymph but larger, 253 µm (192–331 µm) long and 41 µm (32–55 µm) wide. Gnathosoma distinctly separated, trapezoidal, similar to those in protonymph but more massive, with larger spines on palps; other morphological details of gnathosoma

similar to those in protonymphs. Podosoma with four pairs of unsegmented, clearly separated legs; each leg equipped with two small denticled claws, larger (3.5–4.0 µm wide) than in protonymphs; also four pairs of oval ventral scutes, located between I–IV pairs of legs present. Opisthosoma conical, elongated, constitutes 68% (61–74%) of body length.

#### Material deposition

Female holotype (reg. no. UGDIZPMMpDDp87f), 99 female paratypes reg. no. UGDIZPMMpDDp01f–86f, UGDIZPMMpDDp88f–100f), 40

male paratypes (reg. no. UGDIZPMMpDDp01m–40 m), eight larva paratypes (reg. no. UGDIZPMMpDDp01l–8 l), 12 protonymph paratypes (reg. no. UGDIZPMMpDDp01pn–12 pn), 39 deutonymph paratypes (reg. no. UGDIZPMMpDDp01dn–39 dn); skin around the eyes, nose, area of vibrissae, lips, chin, cheek, and neck; host *Mustela putorius* (reg. no. MCMMp78, MCMMp80, MCMMp82, MCMMp84, MCMMp90, MCMMp93, MCMMp97, MCMMp98, MCMMp102, MCMMp105, MCMMp111, MCMMp113, MCMMp117); counties Aurich, Borken, Heinsberg, Wartburgkreis, Wetteraukreis, Germany; December 2013, February 2014, August 2014, October 2014, November 2014, January 2015, February 2015, December 2014, and November 2014; parasites coll. J. N. Izdebska and L. Rolbiecki; host coll. S. Rehbein; the whole-type material (mounted microscope slides with the demodecid mites) deposited within the framework of the Collection of Extant Invertebrates in Department of Invertebrate Zoology and Parasitology, University of Gdańsk, Poland.

#### Etymology

The specific epithet *putorii* refers to the specific name of the host.

#### Location in the host

*Demodex putorii* sp. nov. was found in the skin around the eyes, nose, area of vibrissae, lips, chin, cheek, and neck (Figure 2). The observed mites did not cause any skin lesions in the examined European polecats.

#### Differential diagnosis of *Demodex putorii* sp. nov.

*Demodex putorii* sp. nov. is similar in shape and some features to other Demodecidae described from mustelid mammals, especially *D. lutrae* Izdebska et

Rolbiecki, 2014 (Table VII). However, both the female and male specimens of *Demodex putorii* sp. nov. are noticeably larger. The gnathosoma of both species is trapezoidal, but in *D. lutrae* it is elongated (length exceeds width at the base), while in *D. putorii* sp. nov., the length and width at the base are similar. The supracoxal spines on the gnathosoma are hammer-shaped and slightly larger in *D. lutrae* (1.5–2.0 µm in length), directed medially; in *D. putorii* sp. nov. they are smaller (1.0–1.5 µm in length), directed toward the outer edges of the gnathosoma. There are three spines each on the terminal segments of the palpi in both species, but they are conical and of different sizes in *D. lutrae*, and comprise one small conical, two much larger (one bifurcated) in *D. putorii* sp. nov. In addition, *v*“*F* setae are present on the palpi of *D. putorii* sp. nov., whereas in *D. lutrae* these setae are absent. The subgnathosomal setae in *D. lutrae* are located on both sides at the level of the posterior edge of the pharyngeal bulb; in contrast, they are located at the level of the anterior edge of the pharyngeal bulb in *D. putorii* sp. nov. The posterior edges of the epimeral plates IV of *D. lutrae* females form a triangular, distinct incision, and a slightly arched incision in *D. putorii* sp. nov.; in addition, the epimeral plates I–IV connect medially in *D. lutrae* males, while they are slightly separated in *D. putorii* sp. nov. males. The leg claws differ mainly in the shape of the spur (straight in *D. lutrae*, curved in *D. putorii* sp. nov.); in addition, no solenidia were observed in *D. lutrae*, but they are present in *D. putorii* sp. nov. Furthermore, the aedeagus of males of the two species are in similar locations but it is slightly longer in *D. lutrae*. The opisthosoma has a similar shape in the two species but is relatively shorter compared to body length in *D. lutrae* (i.e. 59% in the male and 61% in the female), and is more elongated (64%

Table VII. Morphometric comparison between *Demodex putorii* sp. nov. and *Demodex lutrae*.

Feature/species Source	<i>Demodex putorii</i> sp. nov.		<i>Demodex lutrae</i>	
	Present study		Izdebska & Rolbiecki (2014) and authors' unpublished data	
	Males ( <i>n</i> = 40) Mean (range) ± SD	Females ( <i>n</i> = 100) Mean (range) ± SD	Males ( <i>n</i> = 24) Mean (range) ± SD	Females ( <i>n</i> = 76) Mean (range) ± SD
Sex (simple size)				
Body total length	203 (177–229) ± 14	278 (231–309) ± 16	170 (158–186) ± 9	209 (183–240) ± 12
Body total width	31 (27–35) ± 2	35 (30–41) ± 2	34 (30–38) ± 2	35 (30–41) ± 3
Body length to width ratio	6.5:1 (5.4–8.1:1) ± 0.5:1	8.0:1 (6.8–9.6:1) ± 0.6:1	5.0:1 (4.4–6.0:1) ± 0.4:1	6.0:1 (4.9–7.1:1) ± 0.5:1
Opisthosoma length to body length ratio (%)	64 (60–68) ± 2	67 (61–70) ± 2	59 (57–63) ± 0.02	61 (56–66) ± 0.02
Aedeagus length	23 (19–27) ± 2	–	24 (20–30) ± 2	–
Vulva length	–	9 (5–11) ± 1	–	9 (8–14) ± 1

SD – standard deviation.

Table VIII. Body size ( $\mu\text{m}$ ) for adults of *Demodex foetorii* sp. nov.

Morphologic features	Males ( $n = 10$ ) Mean (range) $\pm$ SD	Females ( $n = 14$ ) Mean (range) $\pm$ SD
Length of gnathosoma	18 (16–20) $\pm$ 1	20 (17–22) $\pm$ 1
Width of gnathosoma (at base)	22 (19–25) $\pm$ 2	22 (20–25) $\pm$ 1
Length of podosoma	59 (50–70) $\pm$ 5	65 (57–68) $\pm$ 3
Width of podosoma	38 (34–41) $\pm$ 2	38 (38–40) $\pm$ 1
Length of opisthosoma	74 (70–78) $\pm$ 2	76 (65–80) $\pm$ 4
Width of opisthosoma	36 (33–39) $\pm$ 2	38 (35–40) $\pm$ 2
Aedeagus	24 (22–29) $\pm$ 2	–
Vulva	–	8 (7–10) $\pm$ 1
Total length of body	151 (142–165) $\pm$ 6	161 (139–166) $\pm$ 7

SD – standard deviation.

and 67%, respectively) in *D. putorii* sp. nov. The typical microhabitat is also different: *D. lutrae* was found mainly in the hairy skin of the head, and *D. putorii* sp. nov. in the sparsely hairy skin of the head (lips, chins).

*Demodex foetorii* sp. nov. Izdebska, Rolbiecki et Rehbein (Table VIII, Figures 5, 7)

Female ( $n = 13$  and 1 holotype). Body oval, stocky, with distinctly separated gnathosoma, 161  $\mu\text{m}$  (139–166  $\mu\text{m}$ ) long and 38  $\mu\text{m}$  (38–40  $\mu\text{m}$ ) wide (holotype, 162  $\times$  40  $\mu\text{m}$ ). Gnathosoma trapezoidal with length slightly smaller than width at base; on dorsal side in central part of basal segments, pair of large, wedge-shaped supracoxal spines (setae *elc.p*) present, ca. 5.0–6.0  $\mu\text{m}$  long (holotype, 5.0  $\mu\text{m}$ ), directed medially. Palps 3-segmented, terminating in three spines (one small, conical, two larger, curved) on tibio-tarsus. On ventral surface of gnathosoma, horseshoe-shaped pharyngeal bulb with pair of very small subgnathosomal setae (setae *n*), situated clearly below to anterior limit on both sides. Podosoma trapezoidal, slightly widens at end; four pairs of short legs, with coxa integrated into ventral idiosomal wall and five free, overlapping segments (trochanter–tarsus); two bifurcated claws, ca. 5.0  $\mu\text{m}$  long (holotype, 5.0  $\mu\text{m}$ ) with large, curved subterminal spur; two small knobs at base of each claws. Epimeral plates (coxal fields) pairs I–III distinctly sclerotized; all epimeral plates connect medially; pair I triangular, pairs II–IV trapezoidal; posterior edges of pair IV weakly sclerotized and form triangular incision. On dorsal side of podosoma, podosomal shield with distinctly vertical striations, reaches level of legs III. Opisthosoma oval, wide and relatively short, constitutes 47 (46–49%) of body length (holotype, 46%), rounded at end. Whole

opisthosoma distinctly annulated; annulation also reaches dorsal podosoma side (pair of legs III); annuli relatively wide at ca. 1.0  $\mu\text{m}$ . Opisthosomal organ absent. Vulva 8  $\mu\text{m}$  (7–10  $\mu\text{m}$ ) long (holotype, 10  $\mu\text{m}$ ); located behind incision of epimeral plates IV.

Male ( $n = 10$ ). Shorter than female, 151  $\mu\text{m}$  (142–165  $\mu\text{m}$ ) long, and 38  $\mu\text{m}$  (34–41  $\mu\text{m}$ ) wide, but of similar shape. Gnathosoma shape similar to female, but slightly smaller. Pharyngeal bulb and morphological details of gnathosoma similar to those in female. Also shape of podosoma and legs similar to those in female, but epimeral plates I and IV connect medially, while II and III are slightly separated; epimeral plates I–III distinctly sclerotized, pair IV weakly sclerotized. On dorsal side of podosoma, podosomal shield with distinctly vertical striations, reaches level of legs III. Opisthosoma similar to female, constitutes 49% (45–53%) of body length; distinctly annulated, annuli relatively wide at ca. 1.0  $\mu\text{m}$ . Opisthosomal organ absent. Aedeagus with shape resembling scorpion telson, 24  $\mu\text{m}$  (22–29  $\mu\text{m}$ ) long, located between epimeral plates I and III. Genital opening located on dorsal surface, above the border of epimeral plates I and II.

Immature stages and eggs – not found.

#### Material deposition

Female holotype (reg. no. UGDIZPMMpDDf03f), 9 female paratypes reg. no. UGDIZPMMpDDf01f–02f, UGDIZPMMpDDf04f–14f,) and 10 male paratypes (reg. no. UGDIZPMMpDDf01m–10); skin around the eyes, chin, cheek, ear pinnae, and vertex; host *Mustela putorius* (reg. no. MCMMp77, MCMMp78, MCMMp80, MCMMp84, MCMMp93, MCMMp99, MCMMp102, MCMMp115); counties Aurich, Borken, Heinsberg, Nordhausen, Wartburgkreis, Germany;

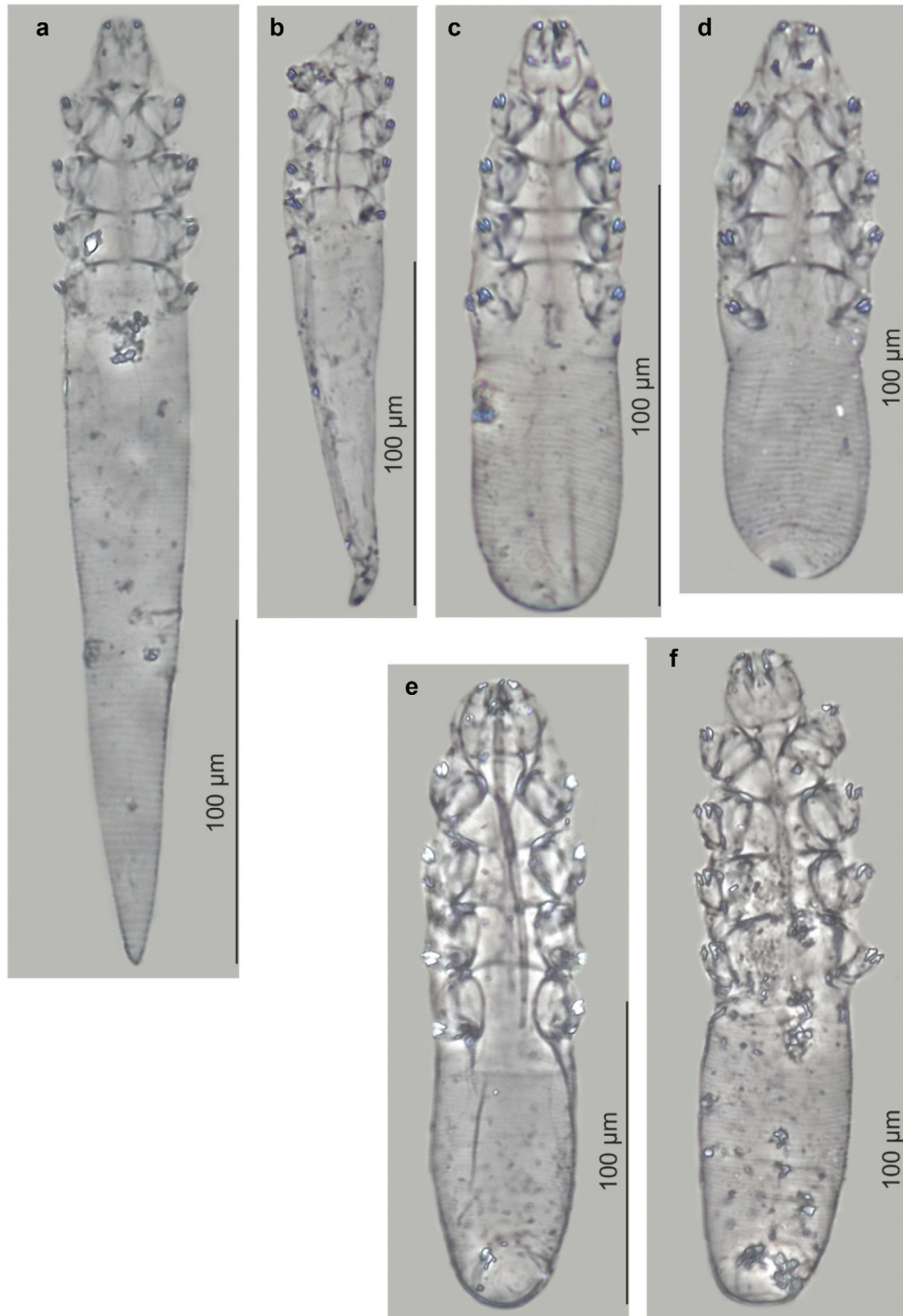


Figure 5. *Demodex putorii* sp. nov., female (a), male (b); *Demodex foetorii* sp. nov., female (c), male (d); *Miridex putorii*, male (e), female (f).

December 2013, February 2014, November 2014, January 2015, and December 2014; parasites coll. J. N. Izdebska and L. Rolbiecki; host coll. S. Rehbein; the whole-type material (mounted microscope slides with the demodecid mites) deposited within the framework of the Collection of Extant

Invertebrates in Department of Invertebrate Zoology and Parasitology, University of Gdańsk, Poland.

*Etymology*

The specific epithet *foetorii* is derived from one of the Latin names for the host, *Foetorius putorius*.



Figure 6. Group of specimens of *Demodex putorii* sp. nov.

#### Location in the host

*Demodex foetorii* sp. nov. was found in the skin around the eyes, chin, cheek, ear pinnae, and vertex (Figure 2). The observed mites did not cause any skin lesions in the examined European polecats.

#### Differential diagnosis of *Demodex foetorii* sp. nov.

*Demodex foetorii* sp. nov. differs significantly from *D. putorii* sp. nov. in its set of features and habit in the same host (Table IX). It is noticeably shorter, with different body proportions – *D. putorii* sp. nov. is strongly elongated, conical with a long, slender opisthosoma, while *D. foetorii* sp. nov. is short, broad, with an oval opisthosoma. The gnathosoma of both species is trapezoidal, but the length is close to the width at the base in *D. putorii* sp. nov., and the length is less than the width in *D. foetorii* sp. nov. The

supracoxal spines on the gnathosoma in *D. putorii* sp. nov. are small (1.0–1.5 µm in length), conical, and directed outwardly, while they are larger (5.0–6.0 µm in length), wedge-shaped, and directed to the center of the gnathosoma in *D. foetorii* sp. nov. There are three spines on the terminal segments of the palpi in both species, but one small, conical and two large, including one bifurcated in *D. putorii* sp. nov., compared to one small, conical and two large, curved, in *D. foetorii* sp. nov. In addition, a v“F seta is present on the palpi of *D. putorii* sp. nov., which was not found in *D. foetorii* sp. nov. The subgnathosomal setae are located at the level of the anterior edge of the pharyngeal bulb in *D. putorii* sp. nov., but they are located clearly below the anterior edge in *D. foetorii* sp. nov. Moreover, the posterior edge of epimeral plate IV has a slight arched shape in *D. putorii* sp. nov. females but a triangular shape in *D. foetorii* sp. nov. females; in addition, epimeral plates I–IV are slightly separated in *D. putorii* males, while only epimeral plates II and III are slightly separated, additionally epimeral plate IV is weakly sclerotized in *D. foetorii* sp. nov. males. The leg claws differ mainly in the shape of the spur, which is larger and more curved in *D. putorii* sp. nov. The solenidia are present in *D. putorii* sp. nov., but absent in *D. foetorii* sp. nov. Furthermore, the aedeagus in *D. foetorii* sp. nov. has the characteristic shape of a scorpion’s telson. The opisthosoma is conical and slender in *D. putorii* sp. nov., while it is short and oval in *D. foetorii* sp. nov. The microhabitat preferences are also a little different: *D. putorii* sp. nov. was mainly found in the sparsely hairy skin of the head (lips, chins), and mainly in the hairy skin of the head in *D. foetorii* sp. nov.

*Demodex foetorii* sp. nov. generally differs in its shape and features from known Demodecidae, including species described from mammal carnivores. It only resembles *D. erminae* Hirst 1919, described on the basis of two males specimens found in *Mustela erminea* Linnaeus, 1758, with regard to its habit and the presence of large supracoxal spines. This species, however, requires re-description, as its description lacks many of the features currently considered important in the taxonomy of Demodecidae, and no description exists of the females. Based on the laconic description, measurements and figures, it can be concluded that the males of *D. erminae* are smaller than those of *D. foetorii* sp. nov., but have slightly different body proportions and features of the gnathosoma (including supracoxal spines)

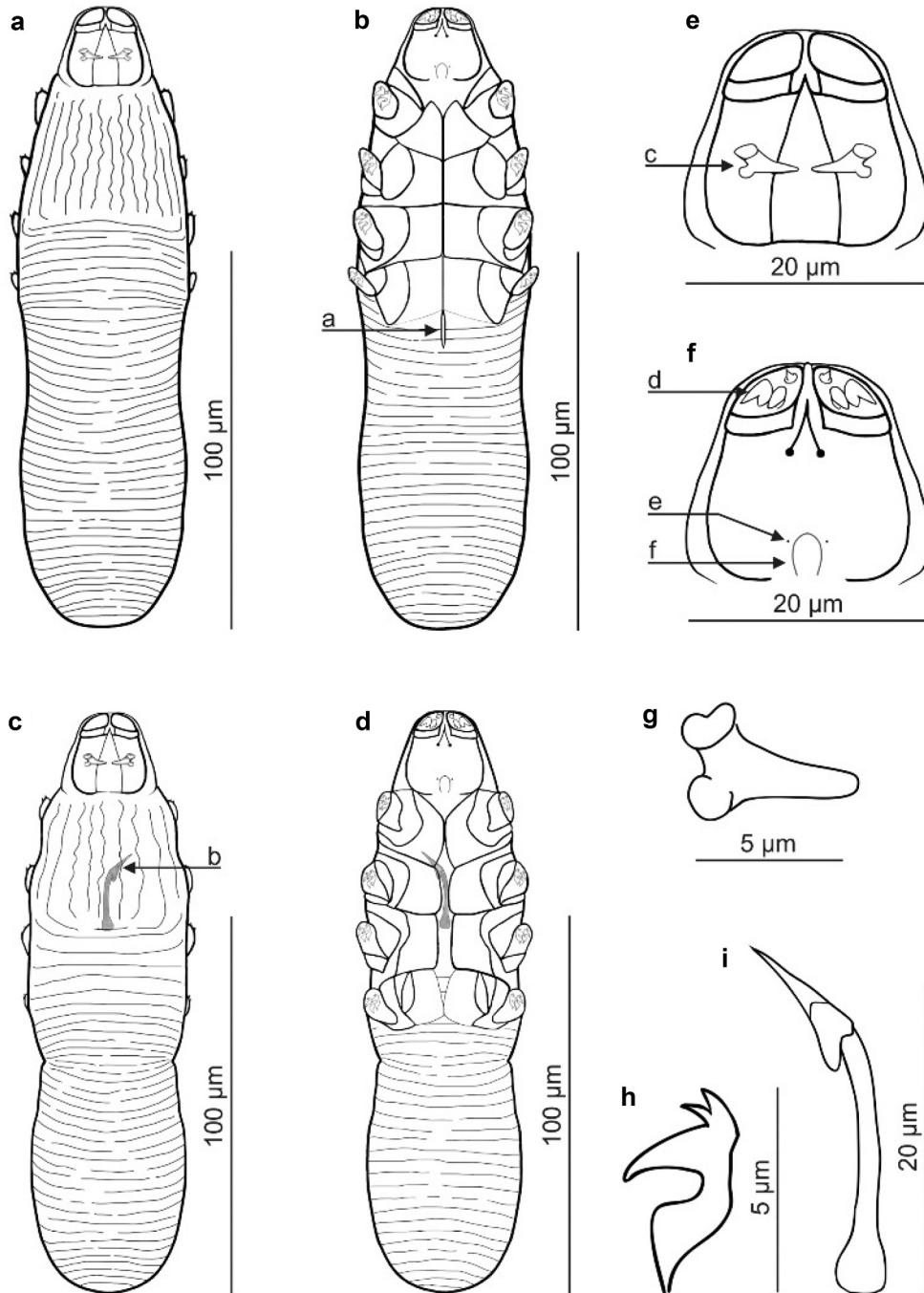


Figure 7. *Demodex foetorii* sp. nov., female, dorsal view (a); female, ventral view (b); male, dorsal view (c); male, ventral view (d); gnathosoma, female, dorsal view (e); gnathosoma, female, ventral view (f); supracoxal spine (g); claw on the leg (h); aedeagus (i). Abbreviations: a – vulva, b – aedeagus, c – supracoxal spine (seta *elc.p*), d – spines on palps, e – subgnathosomal seta (seta *n*), f – pharyngeal bulb.

and a shorter (higher located) and different-shaped aedeagus. It is likely that this is a morphologically similar species from a host belonging to the same genus, which is a phenomenon typical of Demodecidae.

*Distribution and associations of demodecid mites with families and species of carnivoran hosts*

Taking into account the currently described species of demodecid mites from the European polecat and the latest literature data on the occurrence of these



Table IX. Morphometric comparison between *Demodex foetorii* sp. nov. and *Demodex putorii* sp. nov.

Feature/species	<i>Demodex foetorii</i> sp. nov.		<i>Demodex putorii</i> sp. nov.	
	Present study		Present study	
Source				
	Males ( <i>n</i> = 10)	Females ( <i>n</i> = 14)	Males ( <i>n</i> = 40)	Females ( <i>n</i> = 100)
Sex (simple size)	Mean (range) ± SD	Mean (range) ± SD	Mean (range) ± SD	Mean (range) ± SD
Body total length	151 (142–165) ± 6	161 (139–166) ± 7	203 (177–229) ± 14	278 (231–309) ± 16
Body total width	38 (34–41) ± 2	38 (38–40) ± 1	31 (27–35) ± 2	35 (30–41) ± 2
Body length to width ratio	4.0:1 (3.7–4.5:1) ±0.3:1	4.2:1 (3.5–4.4:1) ±0.2:1	6.5:1 (5.4–8.1:1) ±0.5:1	8.0:1 (6.8–9.6:1) ±0.6:1
Opisthosoma length to body length ratio (%)	49 (45–53) ± 2	47 (46–49) ± 1	64 (60–68) ± 2	67 (61–70) ± 2
Aedeagus length	24 (22–29) ± 2	–	23 (19–27) ± 2	–
Vulva length	–	8 (7–10) ± 1	–	9 (5–11) ± 1

SD – standard deviation.

mites in carnivores, the list of Demodecidae species associated with Carnivora and their distribution in the world has been updated (Table X).

## Discussion

Although the Demodecidae have been studied for more than 180 years, knowledge of their biodiversity remains scant. Most of the data concern their occurrence in domestic mammals and humans, and knowledge of the Demodecidae fauna of wild animals is fragmentary. This is also true for the order Carnivora, a widely distributed and otherwise relatively well-studied group of mammals (Izdebska & Rolbiecki 2020; Izdebska et al. 2023). Of the 10 known species of Demodecidae of wild carnivorans, six have been described from single hosts kept in zoos (Table X), of which only *Demodex phocidi* has also been confirmed in a host from a natural population (Izdebska et al. 2020). Of the remainder, two (*D. erminae* Hirst, 1919, *D. lutrae*) are known from single findings (Hirst 1919; Izdebska & Rolbiecki 2014a), one (*D. melesinus* Hirst, 1921) has been found worldwide twice, in single hosts in the UK and Poland (Hirst 1921; Izdebska et al. 2018); only the recently described *Miridex putorii* was found in a sample of a dozen host individuals (Izdebska et al. 2022). Thus, data are lacking on not only the species diversity of Demodecidae of individual wild carnivores, but also the most basic information on their occurrence in host populations including the level of infestation. The current study, which resulted in the discovery of two more demodecid mite species in *Mustela putorius*, has made it possible to carry out such an analysis, taking into account the co-occurrence of these mites.

The European polecat is a common mammal carnivoran inhabiting Europe and North Africa. It also appears to be a good model for studying the occurrence of demodecid mites – it is less territorial than other mustelids, and is also polygamous, which undoubtedly promotes inter-individual contact and transmission of skin mites between individuals within a population. Despite having a relatively well-recognized parasitofauna (Kretschmar 2016), data on the occurrence of so-called skin and tissue mites of the order Prostigmata (Demodecidae, Psorergatidae, Epimyodecidae) in this mammal was lacking until recently. Unidentified *Demodex* sp. have only been recorded in the ferret *Mustela furo* (= *M. putorius furo* Linnaeus, 1758), the domesticated form of the European polecat, in the Netherlands and New Zealand (Nutting et al. 1975; Noli et al. 1996). However, no descriptions of specimens obtained from ferrets have been published, only metric data (Noli et al. 1996), which differ (length of adults in the range of about 60–70 µm) significantly from the dimensions of specimens found in the present study.

It was only recently that the first species of the family Demodecidae was discovered (in the European polecat), described and classified into a new genus: *Miridex putorii*. The analysis of taxonomic features considered important criteria in the systematics of Demodecidae mites (Fain 1959; Bukva 1994; Bochkov 2008; Izdebska & Rolbiecki 2016; Izdebska et al. 2022) confirms that the specimens observed herein in the European polecat represent two new species belonging to the genus *Demodex*; they are characterized not only by a different set of morphological structures, but also by the peculiar location and, like other species of this family, probably also by host specificity. This is

Table X. Carnivoran host associations and distribution of demodecidae of the world.

Host suborder	Host family	Host species	Demodecid mite species	Occurrence
Caniformia	Canidae	<i>Canis lupus familiaris</i> Linnaeus, 1758	<i>Demodex canis</i> (Leydig, 1859) re-description, Nutting et Desch, 1978	Probably cosmopolitan, e.g. Bangladesh (Ali et al. 2011), Colombia (Arroyo-Munive & Hincapié-Gutiérrez 2018), Cuba (Guerra et al. 2010), Germany (Leydig 1859), Great Britain (Hirst 1919), India (Sivajothi et al. 2013, 2015; Veena et al. 2017), Nepal (Sharma & Pokharel 2019), New Zealand (Thomson 1922), Pakistan (Ashfaq et al. 2019), Poland (Izdebska 2005, 2010; Izdebska & Fryderyk 2011; Pawelczyk et al. 2016; Izdebska & Rolbiecki 2018), Russia (Moskvina 2017), Thailand (Sakulploy & Sangvaranond 2010), Turkey (Pekmezci et al. 2018), USA (Nutting & Desch 1978) Probably cosmopolitan, Poland (Izdebska & Rolbiecki 2018)
			<i>Demodex cornei</i> Izdebska et Rolbiecki, 2018	Japan (Morita et al. 2018)
			<i>Demodex cyonis</i> Morita, Ohmi, Kiwaki, Ike et Nagata, 2018	
			<i>Demodex injai</i> Desch et Hillier, 2003	Probably cosmopolitan, e.g. Brazil (Sgarbossa et al. 2017), Poland (Izdebska 2010; Izdebska & Fryderyk 2011), Spain (Ordeix et al. 2009), USA (Desch & Hillier 2003)
Ursidae		<i>Ailuropoda melanoleuca</i> (David, 1869)	<i>Demodex ailuropodae</i> Xu, Xie, Liu, Zhou et Shi, 1986	China, zoological garden (Xu et al. 1986)
		<i>Ursus americanus</i> Pallas, 1780	<i>Demodex ursi</i> Desch, 1995	USA (Desch 1995; Foster et al. 1998)
Otariidae		<i>Zalophus californianus</i> (Lesson, 1828)	<i>Demodex zalophi</i> Dailey et Nutting, 1979	USA, Australia (Dailey & Nutting 1979)
Phocidae		<i>Phoca vitulina</i> Linnaeus, 1758	<i>Demodex phocitidi</i> Desch, Dailey et Tuomi, 2003	USA, sealife center (Desch et al. 2003), Poland (Izdebska et al. 2020)
Mustelidae		<i>Lutra lutra</i> (Linnaeus, 1758)	<i>Demodex lutrae</i> Izdebska et Rolbiecki, 2014	Poland (Izdebska & Rolbiecki 2014a)
		<i>Meles meles</i> (Linnaeus, 1758)	<i>Demodex melesinus</i> Hirst, 1921	Great Britain (Hirst 1921), Poland (Izdebska et al. 2018)
		<i>Mustela erminea</i> Linnaeus, 1758	<i>Demodex erminae</i> Hirst, 1919	Great Britain (Hirst 1919), New Zealand (Nutting et al. 1975)
		<i>Mustela putorius</i> Linnaeus, 1758	<i>Demodex foetorii</i> Izdebska, Rolbiecki et Rehbein	Germany (present study)
			<i>Demodex putorii</i> Izdebska, Rolbiecki et Rehbein	Germany (present study)
			<i>Miridex putorii</i> Izdebska, Rolbiecki et Rehbein, 2022	Germany (Izdebska et al. 2022)

(Continued)

Table X. (Continued).

Host suborder	Host family	Host species	Demodecid mite species	Occurrence
Feliformia	Felidae	<i>Felis catus</i> Linnaeus, 1758	<i>Demodex cati</i> Megnin, 1877 (re-description, Dailey et Nutting, 1979)	Probably cosmopolitan, e.g. Brasil (Valandro et al. 2016), Germany (Löwenstein et al. 2005), Great Britain (Hirst 1919), Italy (Marricotti & Maina 2017), New Zealand (Tengquist & Charleston 2001), Poland (Izdebska 2005; Izdebska et al. 2023), Republic of Bulgaria (Iliev et al. 2019), Spain (Ortúñez et al. 2009), USA (Desch & Nutting 1979; Bizikova 2014)
			<i>Demodex gatoi</i> Desch et Stewart, 1999	Probably cosmopolitan, e.g. Austria (Silbermayr et al. 2013), Finland (Saari et al. 2009), Poland (Izdebska 2005; Jańczak et al. 2017; Izdebska et al. 2023), Spain (Ortúñez et al. 2009), USA (Desch & Stewart 1999; Short & Gram 2016), Probably cosmopolitan, Poland (Izdebska et al. 2023)
			<i>Demodex murilegi</i> Izdebska, Rolbiecki et Fryderyk, 2022	Probably cosmopolitan, Poland (Izdebska et al. 2023)
			<i>Demodex obliquus</i> Izdebska, Rolbiecki et Fryderyk, 2022	
		<i>Panthera tigris amoyensis</i> (Hilzheimer, 1905)	<i>Demodex tigris</i> Shi, Xie et Hsu, 1985	China, zoological garden (Shi et al. 1985)
		<i>Uncia uncia</i> (Schreber, 1775) (= <i>Panthera uncia</i> (Schreber, 1775))	<i>Demodex uncii</i> Desch, 1993	USA, zoological garden (Desch 1993)

because the Demodecidae acquired many adaptations to inhabit different hosts over the course of evolution, including separate microhabitats (Izdebska & Rolbiecki 2020); however, the acquisition of high host specificity was conditioned by the low possibility of transfer of these microscopic, not very mobile parasites, with a hidden mode of life in the skin or other tissues.

In the light of the discovery of the new species, the question of co-occurrence of several species of Demodecidae in the same host seems interesting. Such synhospital species are known in mammals from other groups, especially rodents; for example, in the house mouse, seven species from two Demodecidae genera have been described so far. They inhabit different regions of the skin, in different areas of the body, thus making optimal use of the host's body (Izdebska & Rolbiecki 2015a, 2015b, 2016; Izdebska et al. 2016), while not exceeding its tolerance for the presence of skin parasites, resulting in infestations which are not associated with skin lesions. However, in the case of the European polecat, the described species inhabit the same region, i.e. the skin of the head, without gross pathology. In this case, what may be the correlation of the occurrence of synhospital species, allowing for optimal use of environmental resources? According to the analyses, all species can occur simultaneously in the same host, although relatively rarely, at different abundances and within different microhabitats. Thus, *M. putorii* prefers the vibrissae region, *D. putorii* the hairless or sparsely hairy parts of the head, and *D. foetorii* the adjacent but more intensely hairy regions. In general, a higher abundance was noted on the anterior (facial) part of the head, which is characterized by a greater diversity of potential microhabitats. An analogous diversity of species associated with adjacent head microhabitats was described in rodents, such as the field mouse *Apodemus agrarius* (Pallas, 1771), in which demodecid mites specific to the vibrissae region (*D. gracilentus* Izdebska et Rolbiecki, 2013), eye area (*D. huttereri* Mertens, Lukoschus et Nutting, 1983), ear canals (*D. agrarii* Bukva, 1994), or the rest of the hairy skin on the head (*D. apodemi* Hirst, 1918) were found; however, the possibility of co-occurrence at the individual level was not analyzed (Izdebska & Rolbiecki 2013b).

While *Miridex putorii* and *D. putorii* sp. nov. were more numerous and observed in all life stages, only adults were recorded for *D. foetorii* sp. nov. It is possible that these species engage in reproduction and population development during different periods or seasons, which prevents the simultaneous overgrowth of Demodecidae. Such seasonality has been observed in other Demodecidae, such as *D. plecoti* Izdebska, Rolbiecki, Mierzyński et

Bidziński, 2019 of the brown bat *Plecotus auratus* (Linnaeus, 1758) (Izdebska et al. 2019). *Miridex putorii* and *D. putorii*, although more numerous and represented by all life stages, also demonstrated differing dynamics of infestation: the former showed high prevalence, with a lower level of abundance in the skin, while the latter was much less frequent, but with a higher abundance. It appears that the host habitat can be optimized, not only by the separation of microhabitats (e.g., different species may use sensory hair follicles, normal hair follicles, glands, or other structures) but also by seasonal fluctuations in abundance. This may occur especially for long-established host–parasite relationships. In general, the stability of host–parasite relationships is related to the adaptation of parasites to function in the host, optimizing its use as a habitat and food source, and limiting nuisance to the host and thus pathogenicity; this is a feature of the old host–parasite relationships, developed as a result of long-term evolution.

Demodecidae were noted in nearly 76% of the studied European polecats. However, a higher level of infestation was observed in males; this may be a result of polygamy and, at the same time, greater activity of males in inter-individual contacts. This has been confirmed by studies of, for example, Demodecidae in rodents (Izdebska 2012; Izdebska et al. 2017). In addition, higher levels of infestation were observed in older individuals, which is also a typical phenomenon in Demodecidae infestations and is due to a greater number of contacts over a longer life span, i.e. greater opportunities to acquire parasites with age. The phenomenon also applies to demodecid mites of other mammals, including, for example, humans (Izdebska & Jankowski 2006).

Previous studies of infestation levels of Demodecidae in mammalian carnivores have focused only on domestic dogs and cats, although even here, few analyses of the typical non-clinical infestation associated with these mites have been carried out. Studies on domestic dogs and cats from Poland confirm frequent and even widespread occurrence of these mites in their hosts (Izdebska 2010; Izdebska & Rolbiecki 2018; Izdebska et al. 2023). However, in the case of pets from animal shelters or areas where they live in high densities (high possibility of contact between individuals), the transfer of parasites is easier, which determines a high prevalence of infestation. In the case of wild animals, which exhibit territorialism and where contacts between individuals mainly occur during the breeding season, the level of infestation observed in the present population can be considered very high. Interestingly, despite the high level of abundance of mites in the skin of some hosts, no signs of

demodecosis were observed in them. Similar observations apply to *D. melesinus* from badgers, *D. lutrae* from European otters and *D. phocidi* from common seals, where even very high abundances did not generate the occurrence of gross pathology (Izdebska & Rolbiecki 2014a; Izdebska et al. 2018, 2020). It is likely that the level of host tolerance, formed as a result of long-term co-evolution of the parasite and host, is much higher here than in the case of analogous relationships in domestic mammals or humans, where the abundance of even a few individuals per 1 cm<sup>2</sup> of skin determines clinical demodecosis (Forton & De Maertelaer 2017).

In summary, our findings extend the current knowledge of the biodiversity of the Demodecidae in wild mammalian carnivores from 10 to 12 species. They also provide new data on the co-occurrence of different species of this family in the same host, as well as on level of infestation among a larger number of individuals than in previous studies. They also confirm data obtained from studies of other host groups indicating high topical and topographic specificity associated with optimal use of available microhabitats within the host.

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### Disclosure statement

No potential conflict of interest was reported by the author(s).

### Compliance with ethical standards

All applicable international, national and/or institutional guidelines for the care and use of animals were followed.

### Data availability statement

The data analyses in this study are available from the corresponding author upon reasonable request.

### Authors' contributions

Design and methodology: JNI, LR, SR. Data collection: JNI, LR, SR. Morphological analysis: JNI, LR. Statistical/parasitological analysis: SR, LR, JNI. Original draft: JNI, LR, SR. Review and editing: JNI, LR, SR.

### Ethics approval consent to participate

All the fieldwork was done according to the ethical standards recommended by the relevant institutions.

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