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The family Opilioacaridae (Parasitiformes: Opilioacarida) in Mexico, description of two new species and notes on biology and geographical distribution

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Abstract

Two new species of Opilioacaridae from Mexico are described, *Neocarus chactemalensis* **sp. nov.** and *N. comalensis* **sp. nov.**, and new records for *N. texanus* Chamberlin & Mulaik and *N. veracruzensis* Vazquez & Klompen are presented. Relative positions of internal structures of the ovipositor, a highly variable character, are described based on comparisons of invaginated and evaginated ovipositors. A study of records of Opilioacaridae in Mexico shows that the group is distributed across a wide range of ecosystems and elevations, from dry, semi-desert to wet tropical forest, and coastal plains to the altiplano (>2,000m).

Key words: Neocarus, ovipositor, habitat distribution

Introduction

Mites in the family Opilioacaridae are relatively large, free-living scavengers and perhaps occasional predators (Walter & Proctor 1998). The family is widely distributed throughout the tropical and warm temperate regions of the world, although the number of described species is small (Walter & Proctor 2013). That being said, the group is interesting as one of the most basal, and unusual, lineages in the entire (super)order Parasitiformes (Grandjean 1969; Klompen 2010).

The genus *Neocarus* is both the most species-rich, with 14 described species, and most widely distributed genus, occurring from Florida and Texas (U.S.A.) to northern Argentina. The density of described species across that range varies dramatically, with the highest number in Mexico (Hoffmann & Vázquez 1986; Vázquez & Klompen 2002, 2009). Six species and subspecies have been described from Mexico, *N. bajacalifornicus* (Vázquez & Klompen), *N. bajacalifornicus chamelaensis* Vázquez & Klompen, *N. calakmulensis* Vázquez & Klompen, *N. nohbecanus* (Vázquez & Klompen), *N. siankaanensis* (Vázquez & Klompen), and *N. veracruzensis* Vázquez & Klompen, with an additional species to the north, *N. texanus* Chamberlin & Mulaik from the United States, and one to the south, *N. nicaraguensis* (Vázquez & Klompen) from Nicaragua. A final described species to be noted is *N. orghidani* (Juvara-Bals & Baltac) described from Cuba.

Additional collections from a variety of sites across Mexico allowed us to refine the distribution map for the family in Mexico, as well as describe two new species.

Materials and methods

Most material was studied as slide-mounted specimens. For this purpose, specimens were dissected (due to size), cleared in lactic acid and mounted on slides using Hoyer's medium (Walter & Krantz 2009). Frequently, parts of single specimens were mounted on multiple slides. Terminology for the palp tarsal sensilla follows Grandjean (1936) as modified by Vázquez & Klompen (2002).

Drawings were prepared using a Zeiss Axioskop 3 phase contrast microscope, connected to a drawing tube; measurements were taken using an ocular micrometer and are presented in micrometers (μ m). Measurements are summarized in Table 1. One caveat. Measurements of idiosomal length and width are distorted, as they are based on slide-mounted specimens.

	Sex	Ν	length body	width body	length shield	width shield	length palp	length subcap
N. chactemalensis	М	4	1280	794	444	477	529	325
	F	2	1512	1002	444	529	624	350
N. comalensis	*	8	2150	1214	553	657	657	366
N. texanus (Acapulco)	*	5	1710	1219	487	595	605	352
N. veracruzensis (Jalitla)	*	3	1915	1310	580	680	712	315

TABLE 1. Comparative measurements for species of Neocarus from Mexico in this study.

continued.

	Sex	N	width subcap	length ovip	width ovip	length male glands	width male glands
N. chactemalensis	М	4	198				
	F	2	223	378	331		
N. comalensis	*	8	234	536	372	711	333
N. texanus (Acapulco)	*	5	255	364	184	794	454
N. veracruzensis (Jalitla)	*	3	227	529	227	529	302

* differences between males and females very small; measurements combined for "adults". Subcap: subcapitulum; ovip: ovipositor

Georeference data provided are generally approximate. They include GPS data provided by collectors (assumed accurate), but also retroactive georeference data based on label information. In those cases, coordinates refer to the smallest unit (e.g. town) for which coordinates could be established. All are provided as decimal degrees.

Specimen depository abbreviations: CNAC: Acarology Collection of the Universidad Autónoma de Mexico, Mexico City, Mexico; OSAL: Ohio State University Acarology Collection, Columbus, OH, U.S.A.; UQRoo: Universidad de Quintana Roo, Chetumal, Quintana Roo, Mexico. Instar abbreviations: L= larva; PN= protonymph; DN= deutonymph; TN= tritonymph; F= female; M= male.

Taxonomic section

All specimens examined in this study are assigned to the genus *Neocarus* Chamberlin & Mulaik. Generic assignment is based on the following combination of character states. In the adults there are 3 setae on the penultimate body segment, 2 ventrolateral, 1 dorsal (none at all or >1 dorsal in all Old World taxa and *Amazonacarus* Vázquez, Araújo and Feres; shared with *Caribeacarus* Vázquez & Klompen and *Brasilacarus* Vázquez, Araújo & Feres), 4–6 foliate setae on the palp tarsus (>7 in *Caribeacarus* and *Brasilacarus*), and the sensillum with a "crown-like" tip in the main sensillar group of tarsus I (usually distal in *Caribeacarus* (Vázquez & Klompen 2009, Bernardi *et al.* 2013, Vázquez *et al.* 2015)).

Note: previous studies all agree on the absence of lyrifissures on the prodorsal shield. In terms of shape, there are certainly no lyrifissures that resemble the common type observed on the posterior part of the body, but one pair of small, oval lyrifissures is present in all species (Bernardi *et al.* 2014). It is situated between the anterior tip of the shield and the eyes (Fig. 5B, arrow), a position similar to that observed for the anterior pair of lyrifissures in larval Ixodidae (associated with the lateral carina) and Mesostigmata (*id1* (Athias-Henriot 1975)).

Neocarus chactemalensis Vázquez & Klompen sp. nov.

(Fig. 1A, 3A, 4A, 5A, 6A, 7A, 8A)

Diagnosis. Palp tarsus with 4 foliate setae, each of which with 4 lobes; area between sternal and genital verrucae with 1 pair of long, tapering setae (*st3* stout and ribbed); female without setae in pregenital area; males with 4 pregenital and 6 genital setae, all of the short, stout, ribbed type; tip of ovipositor with 11 long, setiform sensilla with bifurcate tips. Glands in males not observed.

Description. Based on 2 females, 3 males, 3 tritonymphs, 5 deutonymphs, and 9 protonymphs.

<u>Gnathosoma</u>. Chelicera (Fig. 1A). Basal segment with 1 seta (*cht*; added in deutonymph), fixed digit with 3 in all instars examined. All setae lightly barbed, more prominently so in adults; fixed digit seta *ch2* ' (all instars) and basal segment seta *cht* bifurcate. Dorsal (*id*) and antiaxial (*ia*) lyrifissures on fixed digit well developed. Fixed digit with 1 tooth and a secondary, offset, protrusion, movable digit with 1 distinct large tooth with a distal hackle, a small proximal ridge-like structure, and a well developed terminal hook. Movable digit in adults and tritonymph with 1 large and 2 smaller ventral denticles (Fig. 1A, arrow); proto- and deutonymphs lacking the smaller denticles.

Palp adults. Trochanter with 3–4 spine-like, ribbed, tapering setae (= *r*-type); femur with 4–9 ribbed papilliform (= *p*-type) and 11–13 *r*-type setae; genu with 2–3 *p*-type and 25–31 *r*-type setae; tibia with 22–29 *r*-type setae and 27 lightly serrate and pointed setae (see Fig. 2). Tibia and tarsus partially fused. Tarsus (Fig. 3A) with 4 foliate (*d*), 7–8 *v*, 11–13 *ch*, and 10–11 *sm* setae, plus 3 solenidia (= *s* sensilla). Tarsal lyrifissures $i\pi$ and $i\alpha$ present (see Fig. 3B). Pretarsus in shape of a pair of well developed sessile claws, inserted more or less laterally. No distinct sexual dimorphism observed.

Subcapitulum (Fig. 4A). All 4 pairs of paralabial setae present: *pl1* (see Fig. 4D) small, conical; With's organ (*pl2*) membranous, discoid; rutellum (*pl3*) with 1 row of 5 teeth, inserted dorso-laterally; *pl4* tiny, inserted dorsally. With 4 circumbuccal (*cb*) and 11 median and subcapitular (*vm*, *lvm*, *ldm*, *vp*, *lvp*) setae in adults. Number of median and subcapitular setae in nymphs increases over development (PN 2; DN 4; TN 6). No sexual dimorphism observed. Lateral lips with distinct canals in all instars examined.



FIGURE 1. *Neocarus* species, chelicera. A, *N. chactemalensis* **sp. nov.**, female (OSAL0104233), arrow, ventral denticles; B, *N. comalensis* **sp. nov.**, female (OSAL 0105737); C, *N. texanus* Chamberlin & Mulaik, female (CNAC 007219c); D, *N. veracruzensis* Vázquez & Klompen, female (OSAL 0105757). Size bar: 50 µm.

Idiosoma. Color. With dark blue and violet stripes on both body and legs. A relatively small species (males average length 1280 (N=4), females 1512 (N=2)).

Dorsum. Prodorsal shield in adults with 2 pairs of eyes, 92 (M)–120 (F) stout, ribbed setae, and 1 pair of lyrifissures (Fig. 5A). Dorsal idiosoma between prodorsal shield and preanal segment without setae but with numerous lyrifissures arranged in transverse rows. Preanal segment with 1 dorsal and 2 ventro-lateral setae. Anal plates in females with 9–10, in males with 8, stout, ribbed setae.

Sternitogenital region adults (Fig. 6A, 7A). Sternal verrucae each with 2–3 medium-long, ribbed, and 1 distinctly longer, barbed and tapering seta (*st1*). Remaining sternal region with 6 pairs of stout, ribbed setae and, centrally, 1 pair of ribbed, tapering (*st2*) setae. Setae *st3* stout and ribbed, not distinct from remaining sternal setae. With 3 pairs of lyrifissures, 2 unusually large and distinct near sternal verrucae, and a smaller pair anterior to setae *st2*. A final pair sometimes present between setae *st3* and the pregenital area. Only the latter pair resembles the common opisthosomal lyrifissures, the anterior ones are larger and of a different shape. Pregenital capsules each with 1 long, tapering seta (*st5*?) and 3–4 (rarely 5) stout, ribbed setae. Pregenital and genital areas in male with, respectively, 4 and 6 short, stout, ribbed, and blunt-tipped setae. Pregenital and genital areas in female nude. Ovipositor (Fig. 8A) with 11 long, serrate, terminal setiform sensilla with bifurcate tips, and 2 pairs of glands; ovipositor terminating in 3 lobes. Glands in males not observed.

Legs. Legs relatively long. Length legs I–IV in male, respectively, 3005, 1625, 1814, and 2741, in female, respectively, 4649, 1928, 1966, and 2911 μ m. Ratio of legs I to idiosoma in adults 2.3–3.1, legs IV to idiosoma 1.9–2.1. Tarsus I without distinct acrotarsus. Sensillum with "crown-like" tip in main sensillar field. Setae *lv* on acrotarsi II–IV with multiple distinct barbs. Coronidia limited to the basal 2/3 of basitarsi II–IV.

Deposition of types. Holotype female (CNAC 007215) and one paratype male (CNAC 007216) at CNAC; other paratypes at OSAL, UQROO.

Material examined. Due to their size, individual specimens may be represented by multiple slides (as indicated).

Holotype female CNAC (CNAC 007215; 3 slides): Mexico, Quintana Roo, Chetumal, campus Quintana Roo State University, 8m, 18.5230N 88.2703W, ex on campus, under stones and in litter, medium-high tropical forest, coll. Vázquez, M M, 7-Feb-2007. Same locality, collector and date: M, CNAC 007216; PN, OSAL 0104250; PN, OSAL 0104251; PN, OSAL 0104252; N, OSAL 0104249; same locality and collector: 15-Nov-2006: PN, OSAL 0104253; PN, OSAL 0104254; PN, OSAL 0104255; 20-Apr-2007: F, OSAL 0104233 (3 slides); M, OSAL 0104228 (2 slides); M, OSAL 0104230 (3 slides); M, OSAL 0104236 (2 slides); TN, OSAL 0104222 (2 slides); TN, OSAL 0104224 (2 slides); TN, OSAL 0104221 (3 slides); DN, OSAL 0104213; DN, OSAL 0104219 (2 slides); PN, OSAL 0104210; PN, OSAL 0104211; PN, OSAL 0104212; PN, OSAL 0104213; PN, OSAL 0104214; PN, OSAL 0104215; same locality and collector: 23-Jun-2007; DN, OSAL 0104217; U, OSAL 0104240; same locality: coll. Berenice, J, 24-Oct-2006, M, OSAL 0104238 (3 slides); PN, OSAL 0104216. Continued presence of this species at this locality is unlikely as the collecting site was recently developed as part of an expansion of university facilities.

Etymology. The specific name is derived from a Mayan word Chac-teh'-mal, "place where the rain god lives". The name of the city of Chetumal. the current capital of the state of Quintana Roo, has the same origin.

Taxonomic remarks. This species shares the presence of setiform sensilla on the ovipositor with *N*. *bajacalifornicus*, *N*. *bajacalifornicus chamelaensis*, and *N*. *nohbecanus*. It differs from the first two of those taxa by the presence of only 4, instead of 5, foliate setae on the palp tarsus, and the absence of setae in the pregenital region of the female. It differs from *N*. *nohbecanus* (and *N*. *orghidani*) by the absence, vs. presence, of modified *sm3*-type setae on the palp tarsus of the male, and by the structure of the setiform sensilla on the ovipositor: smooth in *N*. *nohbecanus*, with distinct barbs/hackles in *N*. *chactemalensis*.

Neocarus comalensis Vázquez & Klompen sp. nov.

(Figs. 1B, 2, 3B, 4B–C, 5B, 6B, 7B, 8B–D, 9B)

Diagnosis. Palp tarsus with 5 foliate setae; area between sternal and genital verrucae with two pairs of long, tapering setae; female with 2–3 well developed, stout and relatively blunt genital setae; males with 5–7 short, ribbed, blunt-tipped pregenital and 6–10 stout, round-tipped genital setae; ovipositor with numerous large and thick terminal setiform sensilla at the tip; glands in males unusually large and distinct.



FIGURE 2. Neocarus comalensis n. sp., palp femur - tibia, female (CNAC 007217b). Size bar: 100 µm.

Description. Based on 4 females, 5 males, and 1 tritonymph

<u>Gnathosoma</u>. Chelicera (Fig. 1B). Setation and dentition similar to *N. chactemalensis*, but seta *cht* not bifurcate. Dorsal and antiaxial lyrifissures on fixed digit well developed. Movable digit with 1 large ventral denticle.

Palp (Fig. 2, 3B). Trochanter with 4–5 (usually higher in females) ribbed, tapering setae (= *r*-type); femur with 7–9 ribbed papilliform (= *p*-type) and 12–16 *r*-type setae; genu with 4–8 *p*-type and 23–27 *r*-type; tibia with 21–33 *r*-type setae and 20–26 lightly serrated and pointed setae. Tibia and tarsus partially fused. Tarsus with 5 foliate (*d*), 7–8 *v*, 14–18 *ch*, and 9 *sm* setae, plus 3 solenidia. Tarsal lyrifissures $i\pi$ and $i\alpha$ present. Pretarsus in shape of a pair of well developed sessile claws.

Subcapitulum (Fig. 4B–C). All 4 pairs of paralabial setae present: *pl1* small, conical; With's organ (*pl2*) membranous, discoid; rutellum (*pl3*) with a single row of five teeth, inserted dorso-laterally; *pl4* tiny, inserted dorsally. With 4 circumbuccal (*cb*) and 8–12 median and subcapitular (*vm*, *lvm*, *ldm*, *vp*, *lvp*) setae. Females have more setae (9–12) than males (7–9) (compare Figs 4B and C). Lateral lips with distinct canals in all instars examined.

<u>Idiosoma</u>. Color. With dark blue and violet stripes on body and legs. Sexual differentiation distinct. Males can de distinguished (even in alcohol) by the presence of a pair of very large glands which are visible in dorsal view, while females carry an ovipositor. Relatively large, among Mexican species (adults average length 2150 (N=8)).

Dorsum. Prodorsal shield in adults with 2 pairs of eyes, 128 (M)–164 (F) setae, and 1 pair of lyrifissures (Fig. 5B). Dorsal idiosoma between prodorsal shield and preanal segment without setae but with numerous lyrifissures arranged in transverse rows. Preanal segment with 1 dorsal and 2 ventro-lateral setae. Anal plates in adults each with 9–12 (usually 10–11) stout, ribbed setae. Females tend to have slightly higher numbers (11–13) than males (9–11).

Sternitogenital region (Fig. 6B, 7B). Sternal verrucae in adults each with 2–3 large, ribbed, and 1 composite seta (*st1*). Remaining sternal region with 6–7 pairs of stout, ribbed and blunt-tipped setae and, centrally, 2 pairs of ribbed, tapering setae (*st2*, *st3*). Pregenital capsules each with 1 long, tapering seta and 4–5 stout, ribbed setae. Pregenital area in males with 5–7 stout, barbed, blunt-tipped setae, genital area with 6–10 stout, round-tipped setae. Pregenital area in female with 3 medium sized, stout, ribbed setae, genital area nude. Ovipositor with 40–46 large setiform sensilla, a pair of gland-like structures, and a pair of channels. When fully evaginated, the sensilla area

terminal, but when invaginated, they appear more median. Some females carried eggs near ovipositor. Males show two pairs of unusually large glands (largest set illustrated in Fig. 9B).

<u>Legs</u>. Legs relatively short. Length legs I–IV in adults (1 female, 1 male), respectively, 3440–3742, 1588–1928, 1644–2306, and 2476–2930 μ m. Ratio of legs I to idiosoma in adults 1.4–1.7, legs IV to idiosoma 1.1–1.2. Tarsus I without distinct acrotarsus. Sensillum with "crown-like" tip in main sensillar field. Setae *lv* on acrotarsi II–IV with multiple distinct barbs. Coronidia limited to the basal 2/3 of basitarsi II–IV.



FIGURE 3. *Neocarus* species, palp tarsus, female. A, *N. chactemalensis* **sp. nov.** female (OSAL0104233); B, *N. comalensis* **sp. nov.** (OSAL 0105737); C, *N. texanus* Chamberlin & Mulaik (OSAL 0105712); E, *N. veracruzensis* Vázquez & Klompen (CNAC 007221c). Size bar: 50 µm.

Deposition of types. Holotype female (CNAC 007217) and one paratype male (CNAC 007218) at CNAC; paratypes at OSAL, UQROO.

Material examined. Due to their size, individual specimens may be represented by multiple slides (as indicated). Holotype female CNAC (CNAC 007217; 4 slides): Mexico, Guerrero, El Comal, elevation 1749m (in Guerrero Mountains on Pacific side), 18.4600N 99.4100W; colls. Francke, O & Montaño, H; under stones in low tropical forest; coll. event no. CNAC 005975. Same collection event data: M, CNAC 007218 (3 slides); F, OSAL 0105726 (4 slides); F, OSAL 0105737 (4 slides); M, OSAL 0105734 (3 slides); M, OSAL 0105745 (3 slides); M, OSAL 0105748 (3 slides); M, OSAL 0105730 (4 slides); TN, OSAL 0105751 (3 slides).

Etymology. The specific name is derived from the primary collection locality, El Comal, Guerrero, Mexico. "Comal" is a Nahuatl word for an extended disk used by Mexicans to cook tortillas.



FIGURE 4. *Neocarus* species, subcapitulum. A, *N. chactemalensis* **sp. nov.**, female female (OSAL0104233); B, *N. comalensis* **sp. nov.**, female (CNAC 007217b). Size bar: 100 μm.



FIGURE 5. *Neocarus* species, prodorsal shield, female. A, *N. chactemalensis* sp. nov. (OSAL 0104233); B, *N. comalensis* sp. nov. (CNAC 007217a), arrow, prodorsal lyrifissure. Size bar: 100 µm, detail 50 µm.

Taxonomic remarks. This species differs from *N. calakmulensis*, *N. nicaraguensis*, *N. orghidani*, *N. siankaanensis*, *N. texanus*, and *N. veracruzensis* by the presence, vs. absence, of distinct setiform sensilla on the tip of the ovipositor, and from *N. chactemalensis* and *N. nohbecanus* by the presence of 5 (not 4) foliate setae on the palp tarsus, and the high number of setiform sensilla on the ovipositor (>40 vs. 7–12). Neocarus comalensis is quite similar to *N. bajacalifornicus* and *N. bajacalifornicus chamelaensis* but differs from those two taxa by the number of setiform sensilla on the ovipositor (>40 in *N. comalensis*, vs. 21–23 in *N. bajacalifornicus* and 18–20 in *N. bajacalifornicus chamelaensis*), and the fine-tipped genital setae in the male (blunt-tipped in *N. bajacalifornicus*).

In addition to the collections of these two new species, a number of individuals from two additional populations were examined. They show small differences with previously described species, but these differences are considered insufficient to recognize them as separate species, at least at this point. If the species are indeed conspecific, it does result in species ranges that are geographically and biologically very diverse. For this reason we are summarizing the differences with type specimens below. Some additional drawings are also included to provide comparative data for future studies.



FIGURE 6. *Neocarus* species, sternitogenital area, female. A, *N. chactemalensis* **sp. nov.**, female (CNAC 007215a); B, *N. comalensis* **sp. nov.** (OSAL 0105726); C, *N. texanus* Chamberlin & Mulaik (CNAC 007219a); D, *N. veracruzensis* Vázquez & Klompen (OSAL 0105757). Size bars: 100 μm; A–C same scale.



FIGURE 7. *Neocarus* species, sternitogenital area, male. A, *N. chactemalensis* **sp. nov.** (CNAC 007216); B, *N. comalensis* **sp. nov.** (CNAC 007218a); C, *N. texanus* Chamberlin & Mulaik (CNAC 007220a); D, *N. veracruzensis* Vázquez & Klompen. (CNAC 007222a). Size bar: 100 μm.



FIGURE 8. *Neocarus* species, ovipositor female. A, *N. chactemalensis* **sp. nov.** female (CNAC 007215b); B–D, *N. comalensis* **sp. nov.**: B, mostly inverted (OSAL 0105726); C, partially everted, frontal view (OSAL 0105737); D, fully everted, lateral view (CNAC 007217d). Size bars: 100 µm; B–D same scale; g= gland.

Neocarus texanus Chamberlin & Mulaik 1942

(Fig. 1C, 3C, 6C, 7C)

Material examined: 4 females and 1 male.

Description focusing on additions to, and differences with, the re-descriptions of *N. texanus* from Texas by Van der Hammen (1966) and Vázquez & Klompen (2002). States from those studies listed in square brackets.

<u>Gnathosoma</u>. Movable digit chelicera (Fig. 1C) with 1 large and 2 very small ventral denticles [just 1 large]. Palp trochanter with 3–6 ribbed, tapering setae (= *r*-type); femur with 6–9 papilliform (= *p*-type) and 12–15 *r*-type; genu with 6 *p*-type and 26 *r*-type setae; tibia with 26 *r*-type and 35 lightly serrate and pointed setae. Palp tarsus (Fig. 3C) with 5 foliate (*d*), 7–8 [8] *v*, 10–14 [14, 21] *ch*, and 9–10 [6, 8] *sm* setae, plus 3 solenidia. Tarsal lyrifissures $i\pi$ and $i\alpha$ present. Subcapitulum with 4 [5] circumbuccal (*cb*) and 8–10 [6–7] median and subcapitular (*vm*, *lvm*, *ldm*, *vp*, *lvp*) setae. Females have more setae (9–10) than males (8).

<u>Idiosoma</u>. Prodorsal shield in adults with 86 (M)–95 (F) [108–125] stout, ribbed setae, and 1 pair of lyrifissures. Anal plates each with 7–8 (9–10) stout, ribbed setae. Sternal verrucae in adults (Fig. 6C, 7C) each with 1–3 [2–4] ribbed, tapering, and 1 long, composite (*st1*) seta. Remaining sternal region with 5–7 [4–6] pairs of stout, ribbed, blunt-tipped setae and, centrally, 2 pairs of ribbed, tapering setae (*st2*, *st3*). Pregenital capsules each with 1 long, tapering seta and 3–4 [3–4M 5–6F] stout, ribbed setae. Pregenital and genital areas in male with, respectively, 6 [4–5] stout, ribbed, blunt-tipped and 8 [8–9] ribbed, pointy-tipped setae [fine]. Pregenital area in female with 2 [2] short, stout, ribbed setae, genital area nude. Ovipositor without terminal setiform sensilla or spine-like internal structures, but with 3 pairs of channels. Males with one pair of unusually large glands and one much smaller pair.

<u>Legs</u>. Legs moderately long. Length legs I–IV in adults (1 male, 1 female), respectively, 3194–3383, 1607–1644, 1663–1739, and 2495–2589 μ m. Ratio of legs I to idiosoma in adults 1.7–2.2 [2.1], legs IV to idiosoma 1.3–1.7 [1.8]. Tarsus I without distinct acrotarsus. Sensillum with "crown-like" tip in main sensillar field. Coronidia limited to the basal 2/3 of basitarsi II–IV. Distal solenidion ωd on basitarsi II–IV indistinct or absent [observed on legs III by Van der Hammen (1966)]; basal solenidion ωp distinct.

Deposition of specimens. One female (CNAC 007219) and one male (CNAC 007220) at CNAC; other specimens at OSAL, UQROO.

Material examined. Due to their size, individual specimens may be represented by multiple slides (as indicated). Mexico, Guerrero, Cumbre de Llano Largo, Mpio. Puerto Marquez, near coast close to Acapulco, 371m, 18.8251N 99.8333W; 19-Jun-2007; coll. Montaño, H.; ex litter under shrubs in tropical deciduous forest; coll. event no. CNAC 005974: F, CNAC007219 (4 slides); F, OSAL 0105705 (3 slides); F, OSAL 0105709 (3 slides); F, OSAL 0105712 (3 slides); M, CNAC 007220 (3 slides).

Taxonomic remarks. These specimens differ from *N. texanus* by the slightly smaller number of prodorsal shield (86–95 vs. 108–125 in *N. texanus*), and anal valve (7–8 vs. 9–10 in *N. texanus*) setae, and the common presence of a distinct distal solenidion ωd on basitarsi II–IV and absence of a distinct basal solenidion ωp (respectively indistinct and distinct in *N. texanus*). These differences are either small quantitative differences, or concern structures that are often very difficult to observe (leg solenidia). Given the relatively small sample size of available specimens from both Texas and Acapulco these differences are considered insufficient for specific status.

Neocarus veracruzensis Vázquez & Klompen 2009

(Fig. 1D, 3D, 6D, 7D, 9A)

Material examined: 2 females and 1 male.

Description focusing on additions to, and differences with, the description of *N. veracruzensis* by Vázquez & Klompen (2009). States from that study listed in square brackets.

Gnathosoma. Chelicera (Fig. 1D). Movable digit with a single large ventral denticle.

Palp (Fig. 3D). Trochanter with 5 [3–5] ribbed, tapering (= *r*-type) setae; femur with 9–12 [7–8] papilliform (= *p*-type), and 15 [12–17] *r*-type setae; genu with 11 [4–10] *p*-type and 26–29 [17–31] *r*-type setae; tibia with 34 *r*-type, 32 lightly serrate and pointed [65], plus two [4] thin smooth setae. Tibia and tarsus partially fused. Tarsus with 5–6 [5]foliate (*d*), 9 [7] *v*, 18 [13] *ch*, and 9–11 [5] *sm* setae, plus 3 solenidia (= *s* sensilla). Tarsal lyrifissures $i\pi$ and $i\alpha$ present. Pretarsus in shape of a pair of well developed sessile claws; claws with a line of short and acute denticles on internal side.

<u>Idiosoma</u>. Dorsum. Prodorsal shield in adults with two pairs of eyes, 132-135 [108–126] stout, ribbed setae, and 1 pair of lyrifissures. Anal plates each with 9-12 [7–14] stout, ribbed setae.

Sternitogenital region (Fig. 6D, 7D). Sternal verrucae in adults each with 2–3 [1–2] ribbed, tapering, and 1 long, composite seta (*st1*). Remaining sternal region with 6–8 [4–6] pairs of stout, ribbed, blunt-tipped setae and, centrally, 2 pairs of ribbed tapering setae (*st2*, *st3*). Pregenital capsules each with 1 long, tapering seta and 4 [3–4] stout, ribbed, blunt-tipped setae. Pregenital and genital areas in male with, respectively, 5 [6–8] medium-sized stout, ribbed, and blunt-tipped and 10 [6–8] medium-sized, stout, ribbed setae with rounded tips. Pregenital area in female with 2–3 [2] short, stout, ribbed setae, genital area nude. Ovipositor with two pairs of gland-like structures connected by small channels and one pair of larger, sac-like glands with large channels. Males with 2 pairs of large glands (Fig. 9A).



FIGURE 9. *Neocarus* species, glands male. A, *N. veracruzensis* Vázquez & Klompen overview idiosoma slide mounted specimen; inset, detail largest gland (CNAC 007222a); B, *N. comalensis* sp. nov. (CNAC 007218a). Size bars: 100 µm.

Legs. Legs moderately long. Length legs I–IV in adults (1 female, 1 male), respectively, 3515-3686, 1746-2060, 1928-2079, and 2986 (female only) μ m. Ratio of legs I to idiosoma in adults 1.8-2.0 [2.2], legs IV to idiosoma 1.5 [1.8]. Tarsus I without distinct acrotarsus. Sensillum with "crown-like" tip in main sensillar field. Coronidia limited to the basal 2/3 of basitarsi II–IV.

Deposition of specimens. One female (CNAC 007221) and one male (CNAC 007222) at CNAC; other female at OSAL.

Material examined. Due to their size, individual specimens may be represented by multiple slides (as indicated). Mexico, Guerrero., Mpio. Tepecoacuilco, Jalitla, elevation 580m, 18.1527N 99.5395W; 17-Jun-2007; coll. Francke, O & Montaño, H; ex under stones, dry low tropical forest; coll. event no. CNAC 005976. F, CNAC 007221 (3 slides); F, OSAL 0105757 (3 slides); M, CNAC 007222 (3 slides).

Taxonomic remarks. Our specimens are very close to *N. veracruzensis* but differ by the higher number of supernumeral sternitogenital setae, that is in addition to setae *st2* and *st3* (7–8 vs. 5–6 pairs in *N. veracruzensis*). Other quantitative characters (e.g. number of setae on the prodorsal shield) differ somewhat from the numbers listed for *N. veracruzensis*, but the ranges overlap. In the absence of conclusive evidence that the new population is different from *N. veracruzensis*, we assign it to that species.

Discussion

The ovipositor. The ovipositor has proven to be a useful structure in identification of opilioacarid species, with many species having their own characteristic ovipositor structure. Ovipositors are easily visible in adult females when evaginated, but even when invaginated they are distinct under the cuticle. That being said, dissections are generally necessary to study all details of their structure. Two problems remain with regard to use of the ovipositor in species identifications: 1) it contains relatively few heavily sclerotized parts, and the less sclerotized parts are more prone to distortion as a result of slide mounting; 2) it is not always clear how structures change position relative to their state of invagination or evagination. Here we consider each of these problems to aid future workers in interpreting this important structure.

Recognizable structures. Among the more recognizable structures are the terminal setiform sensilla. Their presence or absence, as well as shape and number are relatively easy characters (e.g. *N. chactemalensis, N. comalensis,* in this study). Internal sclerites can also be very informative. These sclerites can be very obvious, as in *Salfacarus* species (Vázquez & Klompen 2010) or some Brazilian taxa (Vázquez *et al.* 2014), but in Mexican species they are generally not well developed. Most other characters are less obvious, e.g. small glands inserted in

the middle of and/or at the base of the ovipositor, or channels connected with gland-like structures, or development of the terminal lobes. Most species have three terminal lobes, as in Acariformes, but the degree of development of the lobes varies.

The effect of evagination / invagination. This effect could be studied in some detail based on a series of females of *N. comalensis*, showing varying degrees of invagination / evagination of the ovipositor (Fig. 8B–D). When fully evaginated (Fig. 8D), the most prominent features of this ovipositor are the large number of setae at the tip. Notably, these setae are all inserted antero-dorsal of the ovipositor opening, not all around theopening. A number of gland-like structures open postero-ventral to the opening (Fig. 8C–D). Additional internal structures are absent or indistinct. An often underappreciated complication in studying "smaller" characters of the ovipositor structure is that slides based on fresh specimens allow recognition of structures that are often obscured in slides made from older, alcohol-preserved, specimens (insufficient clearing), or in slides that are getting older (gradual overclearing due to long-time exposure to Hoyer's medium). When evaginated, the ovipositor is quite long, often extending anteriorly to the level of the hypostome. When retracted, it is distinctly shorter. Even so, the same structures observed in an evaginated ovipositor can be observed on the invaginated one, but with some notable positional differences: the distal setae appear more or less median in the invaginated ovipositor (Fig. 8B), while the structures associated with the opening appear anterior. Gland openings are more difficult to observe, and even general positions tend to be far less clear.

Habitat description and species distribution. The first collections of Opilioacaridae were made in dry sites in the Mediterranean region (Algeria, Sicily, mainland Italy) and Yemen (With 1902, 1904), usually under stones in areas with some shrubby vegetation. Subsequent reports (Grandjean 1936; Chamberlin & Mulaik 1942) reinforced the idea that this was the typical type of habitat for the group. Since these early collections, the geographic and habitat range of the group has expanded considerably. Opilioacaridae have been recorded from Central Asia, Africa, Madagascar, India, S.E. Asia, Australia, and in the New World from the southern tier of states of the United States in the north to Uruguay in the south (Silvestri 1905; Redikorzev 1937; Chamberlin & Mulaik 1942; André 1947; Naudo 1963; Hammen 1977; Juvara-Bals & Baltac 1977; Lehtinen 1980; Leclerc 1989; Walter & Proctor 1998; Vázquez & Klompen 2002; Das & Bastawade 2006(2007); Vázquez & Klompen 2009, 2010; Bernardi *et al.* 2012; Vázquez *et al.* 2014). Superficially the notion that the group is limited to warm temperate and tropical regions was challenged by the discovery of two fossil species in Baltic amber (Dunlop *et al.* 2004; Dunlop *et al.* 2008), but it is worth noting that the climate in Europe in the Eocene was considerably warmer than today. There have been various theories on the origin of Opilioacaridae (Gondwanan or not) (Leclerc 1989; Dunlop & Bernardi 2014), a subject beyond the aims of this study, but the current geographical range of the family suggests a cosmopolitan distribution in tropical and warm temperate regions.

Perhaps more unexpected, the range of macro- as well as microhabitats has also expanded drastically from the early records. Additional documented macrohabitats include caves (Juvara-Bals & Baltac 1977; Leclerc 1989; Bernardi *et al.* 2012; Bernardi *et al.* 2013), humid tropical forests (André 1947; Naudo 1963; Lehtinen 1980; Leclerc 1989; Vázquez & Klompen 2002, 2009), coastal dunes (Vázquez & Klompen 2009) and even hot springs (Vázquez & Klompen 2002), in addition to dry, shrub sites (Hammen 1977; Walter & Proctor 1998; Thaler & Knoflach 2002) and high elevation pine-oak forests (unpublished records at CNAC, Vázquez & Klompen 2002). In terms of microhabitats, most collections are still from under stones and in litter, but a number of new collections have been made from under bark, sometimes of fallen trees, but also 1–2m high on trunks of living trees (in Belize & São Paulo state, Brazil, author observations). Taken together these observations shift the overall view of the group from a relict group specialized for particular areas and habitats, to a taxon that is cosmopolitan and, apparently, highly flexible in its habitat requirements.

Many of the above trends can also be observed in Mexico. Mexico is a good target to study opilioacarid distribution because at this point it is probably the best sampled area for Opilioacaridae in the world. So far, specimens of the group have been collected from 14 states (Table 2; Fig. 11). The initial pattern suggested an association with coastal regions, but new records from the land-locked states of Nuevo León, Querétaro, Hidalgo, and México, and inland localities in Guerrero and Oaxaca, do not support that hypothesis. Neither is there an association with lowlands, as species have been recorded from mid- to high-level mountainous regions in Hidalgo and Querétaro (>2,300m), as well as sea level sites in e.g. Veracruz and Quintana Roo. Associated with this elevational range are strong variations in temperature range. The mountain sites have average summer temperatures of 25°C, down to 0°C in winter, while the lowland forest sites (e.g. Calakmul, Chetumal) experience

average temperatures of 30°C throughout the year with near 100% humidity. Naturally, microclimates in sites where the mites actually live may be quite different from these numbers, but it seems reasonable to assume that Opilioacaridae have a broad tolerance for climate. Finally, while most collections of Opilioacaridae have been made in pristine, relatively undisturbed habitats, at least one collection was made from cultivated grasslands. The one notable aspect of current distribution records for Mexico is the absence of records from most of the northern tier of states (Baja California Norte, Sonora, Sinaloa, Chihuahua, Durango, Coahuila). It is unclear whether this reflects real distributions or lack of collecting effort. We expect the latter given records of Opilioacaridae in the U.S.A. from Texas to Arizona.



FIGURE 10. Distribution map of Opilioacaridae in Mexico. Full data for Mexican collections listed in Table 2. Localities for extra-limital records (U.S.A., Belize, Guatemala) listed in full.

As a final note, the geographical range of most species of Opilioacaridae, as currently recognized, is relatively small, with the possible exception of *N. texanus*, which may extend from Texas to Arizona (Hammen 1966). This model is severely perturbed by the current study suggesting that *N. texanus* may extend to SW Mexico (Acapulco), and that *N. veracruzensis* may occur on both coasts of Mexico. In both cases the same species would occur on both sides of the Sierra Madre, the mountain range running along the spine of Mexico. Additional studies, using additional characters, e.g. molecular data, will be needed to test if the older "highly endemic" or the alternative "some wide-spread species" models, are most appropriate for mapping true opilioacarid species distributions.

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No ¹	Species	Depository ²	Locality	General habitat	Specific habitat	climate
1	Neocarus	UQRoo	Baja California Sur, Presa de la Buena Mujer,	semi-desert	soil under shrubs	
	(unidentified)		Mpio. La Paz 24.0838N 110.1905W			
7	N. bajacalifornicus		Baja California Sur, Sierra de La Laguna 23.593611N 109.9686W	pine-oak forest	under bark	temperate
б	<i>Neocarus</i> (unidentified)	CNAC	Nuevo Leon, Sierra de Apanco, 2027m 23.9171N 99.9798W	high elevation		temperate
4	Neocarus (unidentified)	CNAC	Querétaro, San Joaquin, km 14.5, 2381m 20.9892N 99.6581W	high elevation pine- oak forest	under bark	temperate
2	Neocarus (unidentified)	CNAC	Hidalgo , El Cardenal, 2301m 20.6713N 99.1042W	high elevation pine forest		temperate
9	N. bajacalifornicus chamelaensis		Jalisco, Estacion de Biologia de Chamela 19.5333N 105.0833W	medium-high temperate forest	soil and litter	
L	Neocarus (unidentified)	CNAC	Colima, Cueva (Fuera), 8km S Coguimatlán 19.2983N 104.1018W	entrance cave	under stones	warm- tropical
8	Neocarus (unidentified)	CNAC	México, Mpio Tonatico, 1614m 18.7721N 99.5660W	grassland with acacia		temperate
6	N. comalensis	current	Guerrero, El Comal, Mpio. Buenavista de Cuellar, 1749m 18.4600N 99.4100W	medium-high temperate forest	under stones	temperate
10	N. veracruzensis	current	Guerrero, Jalitla, Mpio Tepecoacuilco, 580m 18.1527N 99.5395W	low-high tropical; forest	under stones	warm- tropical
11	N. texamus	current	Guerrero, Cumbre de Llano Largo, Mpio Puerto Marquez, nr coast nr Acapulco, 371m 16.8287N 99.8489W	low dry tropical forest	under stones and rocks	warm- tropical
12	N. veracruzensis		Veracruz, El Morro de la Mancha 19.8432N 96.4533W	coastal dunes	on soil under shrubs	warm- tropical
13	Neocarus (unidentified)	OSAL	Veracruz, Veracruz, ~15km SSW of; 19.0372N 96.1763W			•
4	<i>Neocarus</i> (unidentified)	OSAL	V eracruz, Catemaco, Playa Azul 18.4215N 95.1111W			
15	<i>Neocarus</i> (unidentified)	CNAC	Oaxaca, San Melchor Betaza, Districto Villa Alta, 1432m 17.2514N 96.1538W	mid-elevation low dry tropical forest	under stones	warm-dry
					continued o	n the next page

TAB	LE 2. (Continued)					
No	Species	Depository ²	Locality	General habitat	Specific habitat	climate
16	Neocarus (unidentified)	CNAC	Oaxaca, San Juan Tabaá, 9km camino del entronque de la carretera Diaz Ordaz, Villa Alto, 1354m 17.3048N 96.2064W	low dry tropical forest	under stones	warm-dry
17	Neocarus (unidentified)	CNAC	Chiapas, E of Ocosingo, 888m 16.7146N 90.9055W	high tropical forest		warm tropical
18	N. calakmulénsis		Campeche, Calakmul Biosphere Reserve 18.1083N 89.8000W	high tropical forest	litter, under bark & stones	warm- tropical
19	Neocarus (unidentified)	UQRoo	Campeche, Calakmul Biosphere Reserve, 5km from Arroyo Negro, ~160m 17.8241N 89.2815W			warm- tropical
20	N. calakmulensis		Campeche, Calakmul Biosphere Reserve, Bel-Ha, ~150m 18.9473N 89.3155W	high tropical forest	litter	warm- tropical
21	N. calakmulensis		Quintana Roo, Cenote "Cocodrilo Dorado", La Union 17.9000N 88.8833W	medium-high tropical forest	litter	warm- tropical
22	N. chactemalensis	current	Quintana Roo, Campus UQRoo, Chetumal, 8m; 18.5229 -88.2703	medium-high tropical forest	under stones, in litter	warm- tropical
23	Neocarus (unidentified)	UQRoo	Quintana Roo, Peninsula, ~8m 18.8314N 88.2267W			warm- tropical
24	Neocarus (unidentified)	UQRoo	Quintana Roo, Chacchoben, ~22m 19.0261N 88.2540W			warm- tropical
25	N. nohbecanus		Quintana Roo, Noh Bec 19.1234N 88.3390W	high hardwood tropical forest	litter	warm- tropical
26	N. siankaanensis		Quintana Roo, Sian Ka'an Biosphere Reserve 19.6833N 87.8833W	low tropical forest	under stones	warm- tropical
27	N. siankaanensis		Quintana Roo, Lol Beh restaurant, 2 km from road Tulum to Cobá 20.4239N 87.6499W	medium-high tropical forest	under stones	warm- tropical
28	<i>Neocarus</i> (unidentified)	UQRoo	Quintana Roo, Laguna Colombia, Cozumel Island 20.4638N 86.8788W	mangrove forest	in litter and sandy soil	warm- tropical
29	Neocarus (unidentified)	UQRoo	Quintana Roo, Puerto Morelos, Botanical Garden 20.8442N 86.8807W	botanical garden	litter	warm- tropical
30	Neocarus (unidentified)	UQRoo	Yucatan, Muná 20.9000N 89.2500W		litter	warm- tropical

¹ Numbers corresponds to numbers in Fig. 10. ² Listed only for unpublished records.

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