



Ecology

Anoplurans (Insecta: Psocodea: Anoplura) associated with rodents distributed in the neotropical region of Mexico

Anopluros (Insecta: Psocodea: Anoplura) asociados con roedores en la región neotropical de México

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Abstract

From April to December of 2010, we performed a cross sectional study in order to collect and identify the species of anoplurans associated with cricetid and heteromyd rodents from montane forests in 5 localities in Guerrero and Oaxaca, Mexico. We analyzed 147 rodents belonging to 10 cricetid species and 1 heteromyd species. A total of 378 sucking lice were collected (189 ♀, 106 ♂, 83 nymphs), distributed in 6 species (*Fahrenholzia microcephala*, *Hoplopleura emphereia*, *Hoplopleura ferrisi*, *Hoplopleura reithrodontomydis*, *Neohaematopinus neotomae*, *Polyplax auricularis*) and 2 families (Hoplopleuridae and Polyplacidae). Lice specimens were processed for morphological and molecular identification, using the mitochondrial gene cytochrome oxidase subunit I. Infestations were characterized based on the prevalence and mean abundance. Five of the 6 species were confirmed by molecular analysis. The highest levels of infestation were recorded for *H. emphereia* (66.7%; 4.4) on *Megadontomys thomasi*. All localities represent new records for the species studied.

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Keywords: Sucking lice; Cytochrome oxidase subunit I; Rodents; Mexico

Resumen

De abril a diciembre de 2010 desarrollamos un estudio con el objetivo de recolectar e identificar anopluros asociados con roedores cricétidos y heterómidos de bosques montañosos en 5 localidades en Guerrero y Oaxaca, México. Analizamos un total de 147 roedores pertenecientes a 10 especies de cricétidos y una especie de heterómido. Se recolectó un total de 378 piojos (189 ♀, 106 ♂, 83 ninfas), distribuidos en 6 especies (*Fahrenholzia microcephala*, *Hoplopleura emphereia*, *Hoplopleura ferrisi*, *Hoplopleura reithrodontomydis*, *Neohaematopinus neotomae*, *Polyplax auricularis*) y 2 familias (Hoplopleuridae y Polyplacidae). Los piojos fueron procesados para su identificación morfológica y molecular, usando el gen mitocondrial citocromo oxidasa subunidad I. Las infestaciones fueron caracterizadas con base en la prevalencia y la abundancia promedio. Cinco de las 6 especies fueron confirmadas molecularmente. Los más altos niveles de infestación fueron alcanzados por *H. emphereia* (66.7%; 4.4) sobre *Megadontomys thomasi*. Todas las localidades representan nuevos registros para las especies estudiadas.

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Palabras clave: Piojos; Citocromo oxidasa subunidad I; Roedores; México

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Introduction

Sucking lice are obligate hematophagous ectoparasites of eutherian mammals. Currently, 550 species of Anoplura distributed in 16 families and 49 genera have been recorded worldwide (Durden & Musser, 1994; Light, Smith, Allen, Durden, & Reed, 2010); two-thirds of these arthropods belong to the families Polyplacidae and Hoplopleuridae, both including species parasites of rodents (Durden, 2002). The inventory of Mexican sucking lice is conformed by 44 species distributed in 8 genera (*Antarctophthirus* Enderlein, 1906; *Enderleinellus* Fahrenholz, 1912; *Fahrenholzia* Kellogg and Ferris, 1919; *Hoplopleura* Enderlein, 1904; *Linognathoides* Cummings, 1914; *Linognathus* Enderlein, 1905; *Neohaematopinus* Mjöberg, 1910 and *Polyplax* Enderlein, 1904) and 5 families (Echinophthiriidae, Enderleinellidae, Hoplopleuridae, Linognathidae and Polyplacidae). Forty two of these species (95.5%) have been associated with 61 species of rodents belonging to 4 families (Cricetidae, Heteromyidae, Muridae and Sciuridae), and 21 genera distributed in 28 states of the Mexican Republic (Sánchez-Montes, Guzmán-Cornejo, León-Paniagua, & Rivas, 2013). As part of a project to describe the metazoan fauna associated with cricetid rodents from montane forests of Mexico, we determined the richness and abundance of sucking lice associated with cricetid rodents from forest in the mountains of Guerrero and Oaxaca, Mexico. For this purpose we identified the specimens morphologically and molecularly (using the cytochrome oxidase subunit I [COI] gene) and additionally, we calculated the prevalence and mean abundance for each lice species.

Material and methods

From April to December 2010, hosts were collected under permission FAUT-0170 issued by Semarnat, Mexico from 5 localities, 2 in Guerrero and 3 from Oaxaca, Mexico (Table 1). Rodents were captured using 4 transects of 40 Sherman traps (Romero-Almaraz, Sánchez-Hernández, García-Estrada, & Owen, 2007), and sacrificed in compliance with the

guidelines of the American Society of Mammalogy for the Use of Wildlife Mammals in Research (Gannon & Sikes, 2007). Lice were recovered from the external surface of hosts, and were fixed and preserved in vials with 96% ethanol. Likewise, each host was brushed on a sheet of white paper to extract additional lice adhering to the fur, and was posteriorly processed in the laboratory. For morphological determination sucking lice were mounted on slides using the modified techniques of Kim, Pratt, and Stojanovich (1986) and Wirth and Marston (1968). Specimens were identified using the specialized keys of Cook and Beer (1959), Ewing (1935), Kim et al. (1986), Pratt and Lane (1951), Stojanovich and Pratt (1961a, 1961b) and Stojanovich and Pratt (1965). Prevalence and mean abundance were calculated according with (Bush, Lafferty, & Lotz, 1997). Additionally micrographs of specimens were taken using a Photomicroscope Olympus Provis AX70. Sucking lice were deposited in the collection of Laboratorio de Acarología, Facultad de Ciencias (LAFC), Universidad Nacional Autónoma de México.

DNA extraction was performed using the DNeasy Blood & Tissue Kit (QIAGEN Ltd., UK). Amplification of a partial segment of ≈ 620 of COI was done using primers Jerry 5'-CAACATTTATTTTGATTTTTTGG-3' and PatII 5'-TCCATTACATATAATCTGCCATATTAG-3' (Marsico et al., 2010).

The reaction mixture consisted of 2 μ l of primers (10 μ M, 1 μ l each), 0.4 μ l (1.25 units) of *Taq* DNA Axygen[®], 2.0 μ L of 10 \times Promega reaction buffer, 2 μ L of 25 mM MgCl₂, 0.8 μ L of 10 mM mix dNTPs, 12.3 μ L nuclease-free water and 5 ng DNA in a final volume of 19.5 μ L. PCR conditions were those used by Marsico et al. (2010). The PCR products were analyzed by electrophoresis on 1.5% agarose gels, using a 100 bp and 1 kb molecular weight marker (nucleic acid markers, Axygen) in 1 \times TBE buffer.

Purified amplification products were submitted for sequencing to Unidad de Síntesis y Secuenciación de DNA (USSDNA), Instituto de Biotecnología and Laboratorio de Biología Molecular y de la Salud, Instituto de Biología, Universidad Nacional Autónoma de México. Sequences were compared with other sequences of sucking lice available in GenBank using the basic

Table 1
Sampling sites of specimens collected in this study.

State	Locality	Geographic reference	Collection date
Guerrero	Parque Estatal Cerro del Huizteco, Municipality Taxco	18°36'08.17" N 99°36'30.63" W 2,499 m	30 July–4 August, 2010
	Puerto del Gallo, Municipality General Heliodoro Castillo	17°28'48.46" N 100°10'35.79" W 2,584 m	06–12 December, 2010
Oaxaca	La Yerba Buena, Municipality Santa Catarina Juquila	16°13'59.88" N 97°16'59.88" W 1,710 m	30 April–5 May, 2010
	3 km southern Punto Ixtepeji, Municipality Ixtlán de Juárez	17°12'06.37" N 96°35'28.21" W 2,537 m	22–25 November, 2010
	km 134.5 Highway 175 Oaxaca-Tuxtepec 21 km north of Guelatao	17°25'10.20" N 96°29'53.30" W 2,919 m	22–25 November, 2010

Table 2

Prevalence and mean abundance of sucking lice collected from rodents in 5 localities in the neotropical region of Mexico.

Locality/host species	<i>n</i>	HP	Sucking lice species	TSL	%	A
Guerrero						
<i>Parque Estatal Cerro del Huizteco</i>						
<i>Habromys schmidly</i>	6	3	<i>H. reithrodontomydis</i>	12	50.0	2.0
Romo-Vázquez et al., 2005						
<i>Liomys pictus</i>	5	2	<i>F. microcephala</i>	15	40.0	3.0
Thomas, 1893						
<i>Puerto del Gallo</i>						
<i>Megadontomys thomasi</i>	15	10	<i>H. emphereia</i>	66	66.7	4.4
Merriam, 1898						
<i>Neotoma mexicana</i>	2	2	<i>N. neotomae</i>	9	100.0	4.5
Baird 1855						
<i>Peromyscus beatae</i>	10	5	<i>P. auricularis</i>	7	50.0	0.7
Thomas, 1903						
<i>Peromyscus megalops</i>	15	9	<i>H. emphereia</i>	62	13.3	1.0
Merriam, 1898						
<i>Reithrodontomys bakeri</i>	1	1	<i>H. reithrodontomydis</i>	3	100.0	3.0
Bradley, Mendez-Harclerode, Hamilton and Ceballos, 2003						
<i>Reithrodontomys sumichrasti</i>	5	3	<i>H. reithrodontomydis</i>	5	60.0	1.0
Saussure, 1861						
Oaxaca						
<i>La Yerba Buena</i>						
<i>Peromyscus aztecus</i>	15	2	<i>H. ferrisi</i>	6	13.3	0.4
Saussure, 1860						
<i>Peromyscus melanurus</i>	35	4	<i>H. ferrisi</i>	19	11.4	0.5
Osgood, 1909						
<i>Reithrodontomys mexicanus</i>	1	1	<i>H. reithrodontomydis</i>	1	100.0	1.0
Saussure 1860						
<i>3 km al Sur del Punto Ixtepeji</i>						
<i>Peromyscus beatae</i>	4	1	<i>P. auricularis</i>	3	25.0	0.8
<i>Peromyscus megalops</i>	26	12	<i>H. emphereia</i>	133	46.2	5.1
Oaxaca-Tuxtpec						
<i>Km 134.5 de la Carretera 175</i>						
<i>Peromyscus aztecus</i>	2	1	<i>H. emphereia</i>	2	50.0	1.0
<i>Peromyscus beatae</i>	3	1	<i>P. auricularis</i>	2	33.3	0.7
<i>Reithrodontomys mexicanus</i>	2	1	<i>H. reithrodontomydis</i>	26	50.0	13.0

Host collected: HP, host parasitized; TSL, total of sucking lice; %, prevalence; A, mean abundance.

local alignment search tool [BLAST] (Altschul, Gish, Miller, Myers, & Lipman, 1990). The sequences obtained were submitted to GenBank.

Additionally to our sequences, we obtained another 25 from GenBank belonging to the families Hoplopleuridae and Polyplacidae, and 1 Ischnoceran (*Columbicola columbae*), which was used as an outgroup in accordance with the proposal of Light et al. (2010), with the following accession numbers: AF385003, AF545717, DQ324548, DQ324549, DQ324564, DQ324578, EU162163, EU375771, HM171425, HM171426, HM171427, HM171428, HM171429, HM171430, HM171431, HM171432, HM171433, HM171442, HM171443, HM171444, HM171445, HQ542195, HQ542196.

Sequences were edited and analyzed in Mega 5.1 software (Tamura, Peterson, Peterson, Stecher, Nei, & Kumar, 2011), all were aligned using Clustal W (Thompson, Higgins, & Gibson, 1994). Mega 5.1 was used to select the best nucleotide substitution model. A Neighbor-joining phylogenetic tree was generated using the Tamura 3 parameter distance model. Additionally, uncorrected pairwise 'p' divergences were calculated for comparative purposes.

Results

A total of 147 hosts pertaining to 10 species and 5 genera of Cricetidae (*Habromys schmidly*, *Megadontomys thomasi*, *Neotoma mexicana*, *Peromyscus aztecus*, *Peromyscus beatae*, *Peromyscus megalops*, *Peromyscus melanurus*, *Reithrodontomys bakeri*, *Reithrodontomys sumichrasti*, *Reithrodontomys mexicanus*) and 1 Heteromyidae (*Liomys pictus*) were reviewed. These hosts were infested by 378 sucking lice (189 ♀, 106 ♂, 83 nymphs), distributed in 6 species belonging to 2 families (Hoplopleuridae and Polyplacidae) and 4 genera (*Fahrenholzia*, *Hoplopleura*, *Neohaematopinus* and *Polyplax*). The heteromids were included since they were collected during the collection of rodents in Cerro del Huizteco, Guerrero, and also because they were parasitized by sucking lice.

Rodent species distributed in 2 or more localities were infected by the same lice species, excepting *P. aztecus* which was parasitized by a different species in the sites where it was collected (Table 2). Almost all rodent species harbored only 1 species of sucking lice, excepting *P. megalops*, which was co-infested by *H. emphereia* and *P. auricularis* in Puerto

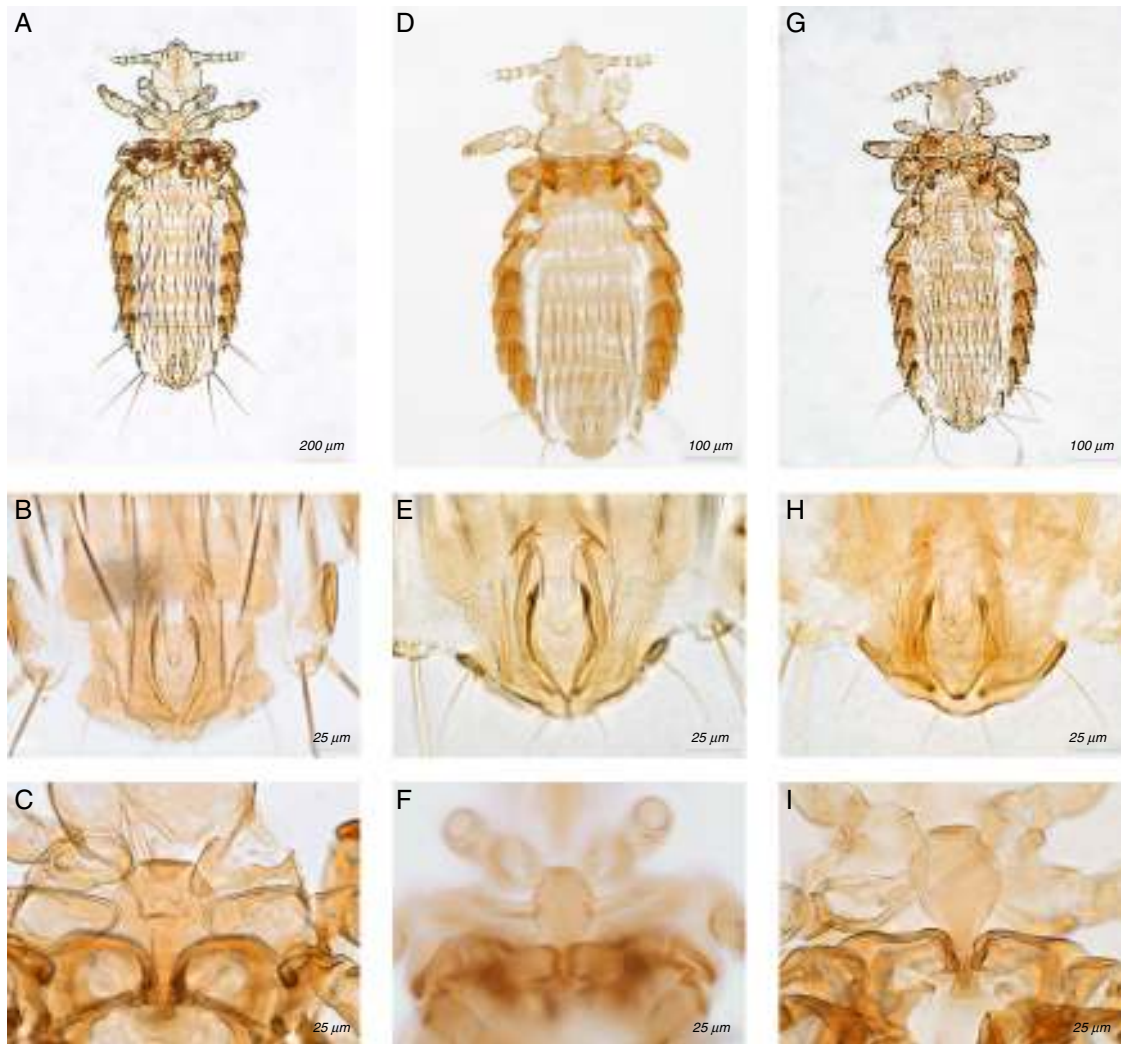


Figure 1. *Hoplopleura emphereia*: (A) male, (B) genitalia, (C) thoracic sternal plate; *Hoplopleura ferrisi*: (D) male, (E) genitalia, (F) thoracic sternal plate; *Hoplopleura reithrodontomydis*: (G) male, (H) genitalia, (I) thoracic sternal plate.

del Gallo, Guerrero. *Hoplopleura reithrodontomydis* showed the highest geographic distribution, being found in 4 of the 5 sampled localities. These sucking lice were the most generalist species and were found in association with 4 cricetids species (*H. schmidly*, *R. bakeri*, *R. mexicanus*, and *R. sumichrasti*). On the other hand, *N. neotoma* and *F. microcephala* were found in only 1 host species at a single locality. The number of host species collected among localities varied from 1 to 6; the highest specific richness was recorded in Puerto del Gallo, Guerrero with 4 species of lice associated with 6 species of cricetids; in contrast, Parque Estatal Cerro del Huizteco exhibited the lowest species richness, as only 2 species were collected infesting 2 host species. Considering only populations of rodents represented by 10 or more specimens, prevalence ranged from 50 to 66.7%, while mean abundance varies from 4.4 to 5.1; among these populations, the highest levels of prevalence and mean abundance were reached by *H. emphereia* in *M. thomasi* in Puerto del Gallo and *P. megalops* in 3 km southern Punto Ixtepeji, respectively (Table 2). Below, we present previous geographic distribution and the new

records obtained in this study for each species of sucking lice recovered.

Family Hoplopleuridae

Hoplopleura emphereia Kim, 1965 (Fig. 1A–C)

Material studied

4♂, 7♀, 3 km southern Punto Ixtepeji, Municipality de Ixtlán de Juárez, Oaxaca, ex *P. megalops*; 4♂, 7♀, 1N, Puerto del Gallo, Municipality General Heliodoro García, Guerrero, ex *M. thomasi*; 2♂, 6♀, 3N, Puerto del Gallo, Municipality General Heliodoro García, Guerrero, ex *P. megalops*.

Distribution

Guatemala, Mexico, Nicaragua and Panama (Castro & González, 1997).

Hoplopleura ferrisi Cook & Beer, 1959 (Fig. 1D–F)

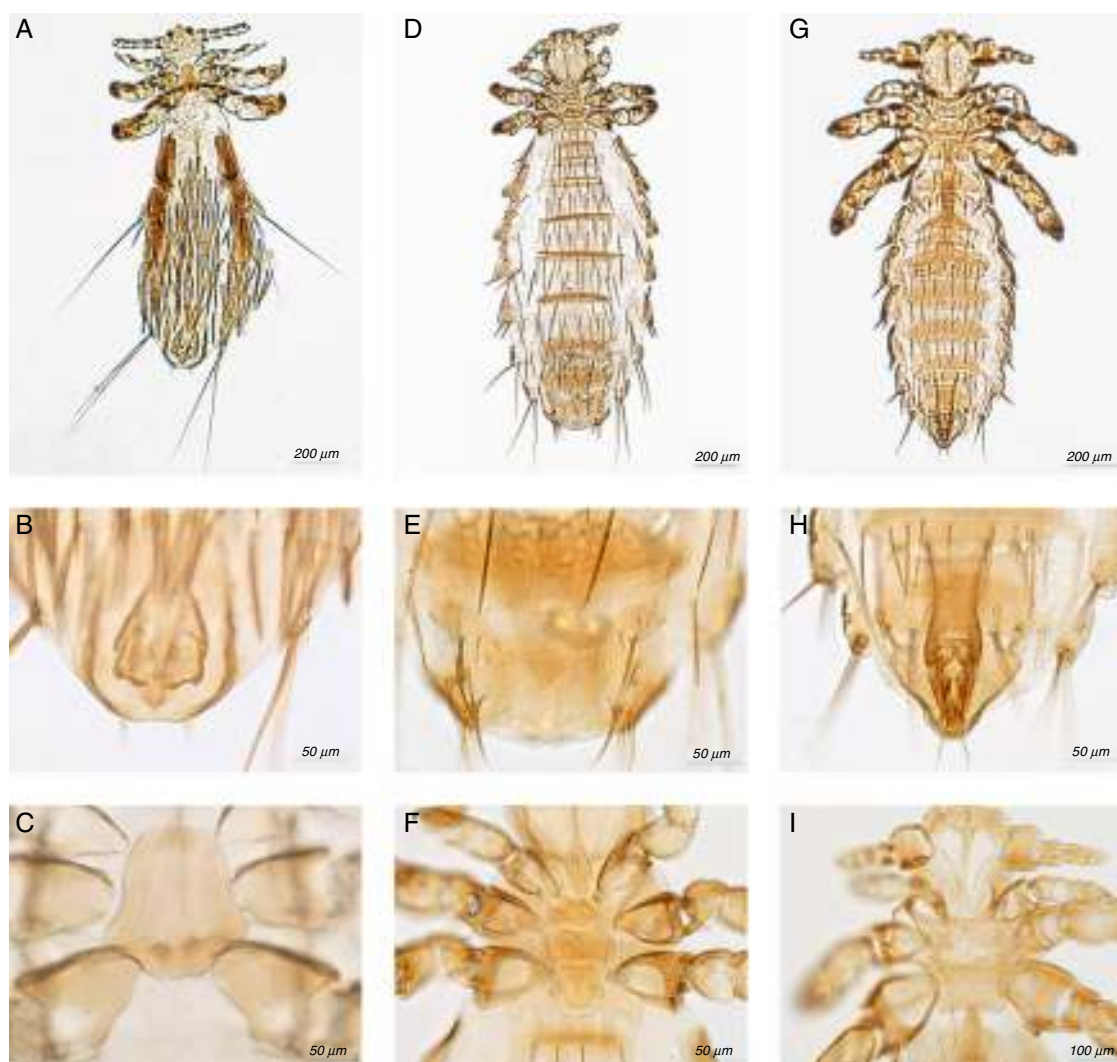


Figure 2. *Fahrenholzia microcephala*: (A) male, (B) genitalia, (C) thoracic sternal plate; *Neohaematopinus neotomae*: (D) female, (E) genitalia, (F) thoracic sternal plate; *Polyplax auricularis*: (G) male, (H) genitalia, (I) thoracic sternal plate.

Material studied

2♂, 4♀, La Yerba Buena, Municipality Santa Catarina Juquila, Oaxaca, *ex P. aztecus*; 5♂, 21♀, 1 N, La Yerba Buena, Municipality Santa Catarina Juquila, Oaxaca, *ex P. melanurus*. 2♀, Km 134.5 de la Carretera 175 Oaxaca-Tuxtepec a 21 Km al Norte de Guelatao, Oaxaca, *ex P. aztecus*.

Distribution

Southeastern of United States (Arizona, Nuevo Mexico) to Mexico (Kim et al., 1986).

Hoplopleura reithrodontomydis Ferris, 1951 (Fig. 1G–I)

Material studied

3♂, 3♀, Parque Estatal Cerro del Huizteco, Municipality Taxco, Guerrero, *ex H. schmidly*. 1♂, La Yerba Buena, Municipality Santa Catarina Juquila, Oaxaca, *ex R. mexicanus*. 1♂, 1♀, 7 N, 3 km al sur del Punto Ixtepeji, Municipality Ixtlán de Juárez, Oaxaca, *ex R. mexicanus*. 1♂, 1♀, 1 N, Puerto del

Gallo, Municipality General Heliodoro Castillo, Guerrero, *ex R. sumichrasti*.

Distribution

Southeastern of United States, Mexico to Central America (Kim et al., 1986).

Family Polyplacidae

Fahrenholzia microcephala Ferris, 1922 (Fig. 2A–C)

Material studied

3♂, 2♀, 2 N, Parque Estatal Cerro del Huizteco, Municipality Taxco, Guerrero, *ex L. pictus*.

Distribution

South-eastern of United States to Mexico (Kim et al., 1986).

Neohaematopinus neotomae Ferris, 1942 (Fig. 2D–F)

Table 3
Matrix of uncorrected pairwise 'p' distances.

Sequence	<i>Fahrenholzia microcephala</i>	<i>Hoplopleura emphereia</i>	<i>Hoplopleura reithrodontomydis</i>	<i>Neohaematopinus neotomae</i>	<i>Polyplax auricularis</i>
<i>Neohaematopinus neotomae</i> (HM171451.1)	0.27	0.22	0.21	0.02	0.29
<i>Neohaematopinus sciuropteri</i> (HM171452.1)	0.27	0.26	0.25	0.20	0.23
<i>Hoplopleura arizonensis</i> (HM171425.1)	0.32	0.22	0.18	0.25	0.23
<i>Hoplopleura erratica</i> (HM171426.1)	0.31	0.27	0.21	0.23	0.23
<i>Hoplopleura ferrisi</i> (HM171427.1)	0.31	0.10	0.10	0.21	0.27
<i>Hoplopleura ferrisi</i> (HM171428.1)	0.27	0.18	0.14	0.20	0.25
<i>Hoplopleura hesperomydis</i> (AF545717.1)	0.29	0.19	0.18	0.19	0.24
<i>Hoplopleura hesperomydis</i> (HM171429.1)	0.30	0.20	0.21	0.23	0.26
<i>Hoplopleura hirsuta</i> (HM171430.1)	0.31	0.23	0.21	0.24	0.25
<i>Hoplopleura onychomydis</i> (HM171431.1)	0.30	0.24	0.19	0.24	0.27
<i>Hoplopleura quadridentata</i> (EU375771.1)	0.31	0.21	0.19	0.23	0.28
<i>Hoplopleura reithrodontomydis</i> (HM171432.1)	0.29	0.15	0.04	0.21	0.27
<i>Hoplopleura reithrodontomydis</i> (HM171433.1)	0.29	0.15	0.04	0.21	0.27
<i>Fahrenholzia ehrlichi</i> (HM171442.1)	0.22	0.31	0.30	0.27	0.27
<i>Fahrenholzia ehrlichi</i> (HM171443.1)	0.20	0.31	0.30	0.30	0.27
<i>Fahrenholzia microcephala</i> (DQ324564.1)	0.04	0.32	0.29	0.26	0.31
<i>Fahrenholzia texana</i> (DQ324578.1)	0.23	0.32	0.30	0.27	0.25
<i>Fahrenholzia reducta</i> (HM171444.1)	0.25	0.27	0.30	0.23	0.26
<i>Fahrenholzia zacatecae</i> (HM171445.1)	0.25	0.29	0.25	0.25	0.25
<i>Polyplax auricularis</i> (DQ324549.1)	0.29	0.27	0.28	0.29	0.03
<i>Polyplax borealis</i> (DQ324548.1)	0.25	0.27	0.26	0.22	0.23
<i>Polyplax spinulosa</i> (HQ542196.1)	0.27	0.28	0.25	0.24	0.23
<i>Polyplax spinulosa</i> (HQ542195.1)	0.27	0.28	0.25	0.24	0.23
<i>Polyplax serrata</i> (EU162163.1)	0.27	0.30	0.27	0.28	0.23
<i>Fahrenholzia microcephala</i>		0.33	0.29	0.27	0.29
<i>Hoplopleura emphereia</i>			0.12	0.22	0.27
<i>Hoplopleura reithrodontomydis</i>				0.21	0.27
<i>Neohaematopinus neotomae</i>					0.28

Material studied

1♂, 1♀, Puerto del Gallo, Municipality General Heliodoro Castillo, Guerrero, ex *N. mexicana*.

Distribution

South-eastern of United States to Mexico (Kim et al., 1986).

Polyplax auricularis Kellogg & Ferris, 1915 (Fig. 2G–I)

Material studied

3♂, 2♀, km 134.5 on Highway 175 Oaxaca-Tuxtepec, 21 km northern Guelatao, Oaxaca, ex *P. beatae*; 28♂, 46♀, 17N, km 134.5 on Highway 175 Oaxaca-Tuxtepec, 21 km northern Guelatao, Oaxaca, ex *P. beatae*; 1♂, 2♀ Puerto del Gallo, Municipality General Heliodoro Castillo, Guerrero, ex *P. beatae*; 2♂, 2♀, 2N Puerto del Gallo, Municipality General Heliodoro Castillo, Guerrero, ex *P. megalops*.

Distribution

From Alaska to Southern United States, Mexico, Costa Rica and Venezuela (Kim et al., 1986).

Molecular characterization

DNA sequences of the COI were obtained for 5 of the 6 species analyzed, *F. microcephala* (GenBank accession

number KT151124); *H. emphereia* (GenBank accession number KT151125), *H. reithrodontomydis* (GenBank accession number KT151126), *N. neotomae* (GenBank accession number KT151127) and *P. auricularis* (GenBank accession number KT151128). No DNA sequences were obtained for *H. ferrisi*.

The intraspecific uncorrected pairwise *p*-distances between COI sequences generated in this study and those obtained for GenBank ranged from 2% to 4%: *N. neotomae* (2%), *P. auricularis* (3%), and *F. microcephala* and *H. reithrodontomydis* (4%); no sequence for pair comparison was available for *H. emphereia* on databases. On the other hand, the interspecific variation ranged from 10% (*H. reithrodontomydis* vs *H. ferrisi*) to 27% (*H. emphereia* vs *H. erratica*) in *Hoplopleura*; from 20% (*F. ehrlichi* vs *F. microcephala*) to 25% (*F. microcephala* vs *F. zacatecae* and *F. reducta*) in *Fahrenholzia*; 23% in *Polyplax* (*P. auricularis* vs *P. borealis*, *P. spinulosa* and *P. serrata*); and 20% in *Neohaematopinus* (*N. neotomae* vs *N. sciuropteri* (Osborn, 1891)) (Table 3).

Four major groups can be recognized in the dendrogram constructed using the neighbor-joining method (Fig. 3). Each group contains species pertaining to the same genus (*Fahrenholzia*, *Polyplax*, *Neohaematopinus*, and *Hoplopleura*). The identity of 4 of the 5 lice species was confirmed by molecular analysis, since the sequences generated in this study joined with the respective sequence for each species obtained from GenBank, except for that of *H. emphereia* which was grouped with that of *H. ferrisi* (Fig. 3).

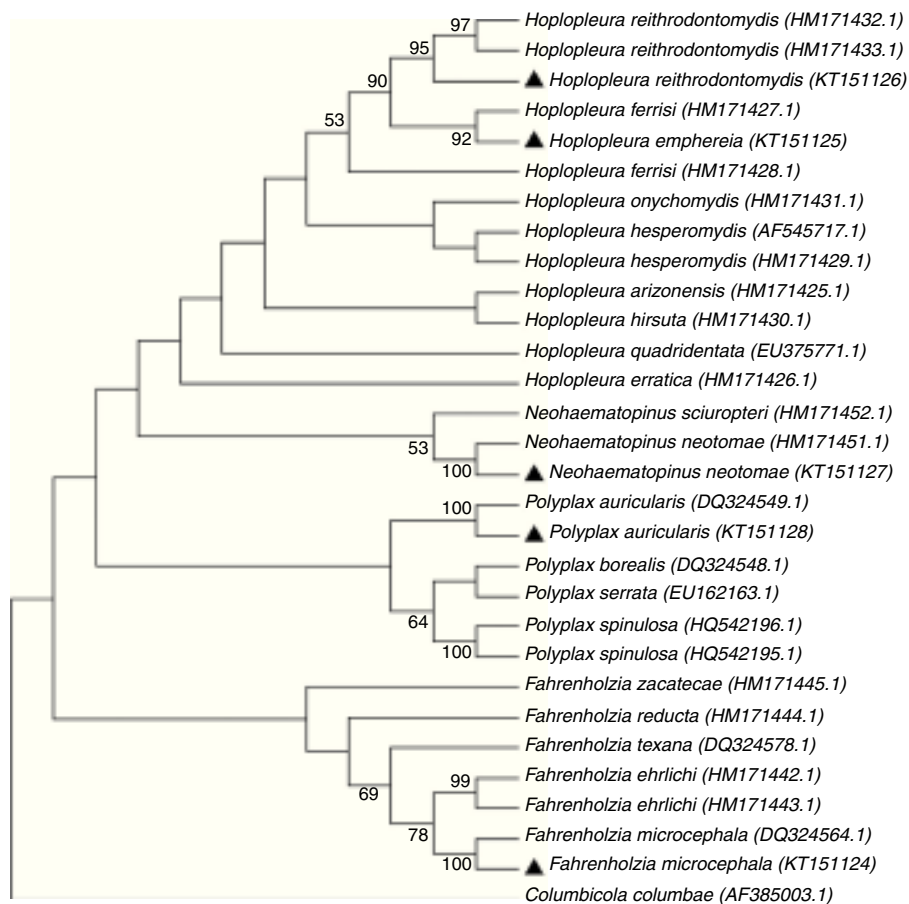


Figure 3. Neighbor-joining phylogenetic tree generated with partial sequences of the COI gene of sucking lice using Tamura 3 parameters distance model. Bootstrap values are indicated in the nodes.

Discussion

As a result of this study, the geographic distribution of the 6 species recorded is increased since we present 13 new locality records. Nine of the 10 cricetid rodents sampled (*H. schmidly*, *N. mexicana*, *P. aztecus*, *P. beatae*, *P. megalops* (for *P. auricularis*), *P. melanurus*, *R. bakeri*, *R. sumichrasti*, *R. mexicanus*) represent new host records for their associated sucking lice species (Sánchez-Montes et al., 2013).

Among the recorded sucking lice, *H. reithrodontomydis* was the most widely distributed species. This species has been recorded infesting only arboricolous and semi-arboricolous rodents of the genus *Reithrodontomys* (Ceballos & Oliva, 2005), which suggests a possible phenomenon of horizontal transmission between the arboreal host species. The finding of *H. reithrodontomydis* on *H. schmidly* is the first record for a host species other than from *Reithrodontomys* spp.; nonetheless, its presence on *H. schmidly* could be explained by its arboreal habits (Romo-Vázquez, León-Paniagua, & Sánchez, 2005) co-existing with 2 *Reithrodontomys microdon* Merriam, 1901; however, the latter host species was not infested by sucking lice.

In accordance with Durden (2002), sucking lice diversity is correlated with host diversity, due to the high specificity exhibited by this arthropod group toward their mammal hosts. In this context, our results fit with this statement, since Puerto del Gallo

the locality with the greatest rodent richness also exhibited the greatest sucking lice richness (6 species). In contrast, in Parque Estatal Cerro del Huizteco only 2 species of rodents and sucking lice were collected. Puerto del Gallo has been referred to as a well preserved area (Navarro, 1998); based on personal observations, we agree with Navarro (1998), while Cerro del Huizteco seems to be a more disturbed site. In this context, Saavedra-Millán (2009), mentioned that Cerro del Huizteco is an area with low floral richness, due to the removal of herbaceous strata as a result of the introduction of infrastructure for ecotourism. This probably could explain the differential rodent richness in both localities.

Although most rodent species were parasitized by only 1 lice species, we recorded the co-infestation of *H. emphereia* and *P. auricularis* on *P. megalops*; this particular association had been previously recorded on *Reithrodontomys creper* Bangs 1902 in Panama (Johnson, 1972). The heterogeneous sample sizes obtained in our study preclude any conclusion about the factors involved in the infection levels recorded; however, variation on ecological parameters has been attributed to different factors such as, host sex, age, immune response, gregarious habits (Durden, 2002). In most sucking lice species infestation levels are less than 15 lice per parasitized host, being an extreme case *H. emphereia* associated with *P. megalops* in 3 Km southern Punto Ixtepeji, whose range was 1–46 lice.

Morphological identification of sucking lice is complex as most of the descriptions are based on a single specimen, lacking information about intraspecific variation, an aspect that should be analyzed. Three of the 6 species studied are well characterized morphologically; however, there is controversy about the validity of 3 other species (*H. emphereia*, *H. ferrisi* and *H. reithrodontomydis*) included in the *Hoplopleura hesperomydis* complex by Kim (1965). This author mentioned that *H. emphereia* shares morphological characters with *H. ferrisi*; later, Johnson (1972) postulated that *H. emphereia* and *H. reithrodontomydis* could be the same species upon comparison of nymphal stages. In this work we identified the 3 species morphologically; *H. reithrodontomydis* differs of *H. emphereia* and *H. ferrisi* in the shape of abdominal paratergite 8, which presents a triangular shape, while in the other 2 species is rectangular. Likewise, *H. emphereia* males can be distinguished from *H. ferrisi* by the morphology of its thoracic sternal plate which have a posterior process abruptly pointed and paramers abruptly tapering posteriorly. In contrast, *H. ferrisi* males present thoracic sternal plate with posterior process gradually acute and paramers gradually tapering posteriorly (Johnson, 1972; Kim, 1965).

Intraspecific divergence values among DNA sequences of *F. microcephala*, *P. auricularis*, *N. neotomae* and *H. reithrodontomydis* (2–4%) obtained in this study and those from GeneBank, allowed us to consider them as valid taxa, since some authors have cited higher ranges for different species, e.g., 13% within *Hoplopleura tiptoni* Johnson, 1972 and *Hoplopleura rimae* Johnson, 1972, 14% for *Hoplopleura aitkeni* Johnson, 1972, and 18% for *Hoplopleura brasiliensis* Werneck, 1932 (Smith, Light, & Durden, 2008). For *H. ferrisi* no COI sequences were obtained and for *H. emphereia* no sequences for comparative purposes were available in GenBank.

On the other hand, the similarity between the COI sequence of *H. emphereia* from Mexico and 1 of the 2 sequences of *H. ferrisi* obtained from GenBank (associated to *Peromyscus difficilis* (Allen, 1891) from Puebla) showed a genetic divergence of 10%; this value suggests the misidentification of 1 of the 2 specimens involved; however, this situation can only be solved through more sampling of specimens of both taxa (Fig. 3).

In spite of the amount of information generated during the 20th century, the inventory of the sucking lice in Mexico remains scarce and fragmentary. To date, 88% of the mammals distributed in Mexico have been neglected as hosts of this arthropod group. Particularly for rodents, completing the inventory of sucking lice is a major challenge, as these mammals constituted the main group of hosts. Only by increasing the sampling of this group of vertebrates in Mexico, through systematic studies, and avoiding partial analysis of a particular group of ectoparasites, this host-parasite association will be understood.

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