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Biodiversity and Conservation (2005) 14:3423–3436 DOI 10.1007/s10531-004-0548-8

Distribution, taxonomy, and conservation status of the perote mouse *Peromyscus bullatus* (Rodentia: Muridae) in México

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Received 5 December 2003; accepted in revised form 28 May 2004

Key words: Endemic, Mammals, México, Peromyscus bullatus, Rodents

Abstract. The Perote deer mouse *Peromyscus bullatus* is a rare species known only from a few specimens. It is endemic to the plains surrounding the Oriental Basin in the states of Veracruz and Puebla, Mexico, in the area where the Sierra Madre Oriental and the eastern end of the Transverse Volcanic Range merge. In past studies, a small area within the basin provided the only known habitat. During two recent surveys adjacent to this area, in habitats that matched the area in the state of Puebla where *P. bullatus* was previously found, we made new surveys. This mouse is differentiable from other species of the genus, mainly in the large of the auditory and the size of the ear. Intrinsic factors are highly discriminating in choice of habitat, low densities, and has restricted distribution, as well as the extrinsic factor of habitat deterioration, indicates a serious conservation problem.

Introduction

Mexican fauna include one of the world's most diverse groups of native rodents, with 148 species (Ramírez-Pulido and Müdespacher 1987; Ceballos and Rodríguez 1993). Unfortunately the endemic species, particularly those with restricted distributions, represent a disproportionately large number of endangered species (Ceballos and Rodríguez 1993).

One of the main reasons that some small mammal species are not provided special protection is the absence of biological information: systematics, distribution, relative abundance, and ecology. The real problems faced by those interested in biodiversity conservation are to collect biological data concerning these species before they reach at risk stage of extirpation and devising proper management schemes for conservation. The natural history of some species, especially small mammals, is largely unknown, mainly as a result of restricted distribution or scarcity of specimens available to biologists. The Perote mouse (*Peromyscus bullatus*) represents such a species.

The Perote mouse was first described by Osgood (1904), based on a specimen from Perote, Veracruz, Mexico. Hoffmeister (1951) discussed the taxonomy of the species in his analysis of *P. truei*, and added six specimens from two localities near El Limón, Veracruz, Mexico. Until the present surveys, *P. bullatus* was extremely rare, since it was known only from these seven specimens. Another particularity is the uncertain taxonomic position of this species. According to Hoffmeister (1951), it does not differ as much from *P. truei* as Osgood (1909) originally indicated. The support for this species is based on the size of the auditory bullae of the holotype. Carleton stated that: '... the status of this form has not been addressed. The proportion of the otic capsule of holotype is truly outsized for either *P. truei* or *P. difficilis...*' (Carleton 1989), although for that time, additional specimens of *P. bullatus* were not available.

No new information has been reported concerning the Perote mouse since Hall and Dalquest (1963) examined the same six specimens as Hoffmeister (1951). In September 2001, a population of *P. bullatus* was discovered near San José Alchichica in central Puebla state, less than 15 km from the type locality. Therefore examined all specimens in Mexican museums, and added 31 new specimens, all these were reviewed to clarify the taxonomic position of *P. bullatus* in reference to *P. gratus* and *P. truei*, species that can be mist identification. Additionally, we evaluated the present distribution, ecology, and natural history.

Materials and methods

Surveys were conducted in July and September 2001 in areas of the Oriental Basin where *P. bullatus* might be found (Figure 1). The two surveys represented a total effort of 2720 trap-nights using Sherman and Museum Special traps. Trap lines consisted of 80 traps at 5-m intervals. We used rolled oats as bait.

Rodent specimens (n = 466) were prepared and deposited in the mammalian collection of the Escuela Nacional de Ciencias Biológicas (ENCB). For analysis of the specimens, we reviewed the rodent collection of ENCB and of the Universidad Autónoma Metropolitana-Iztapalapa (UAM-I). A total of 31 specimens (11 collected and 20 from mammal collections; Appendix 1) of *Peromyscus bullatus* from seven different localities were identified and studied for stability of putatively diagnostic characteristics (Osgood 1904; Hoffmeister 1951).

The specimens of *Peromyscus bullatus* were identified initially by the characteristics given by Osgood (1904), and were compared morphologically and morphmetrically with two similar species, *P. difficilis* and *P. gratus*: (1) to clarify the uncertain taxonomic position of this species, (2) to explore the taxonomic relationships between these related species, (3) to give elements to the interested investigators for the correct identification of this species. Perote mouse is know to occur sympatrically with three species of *Peromyscus*

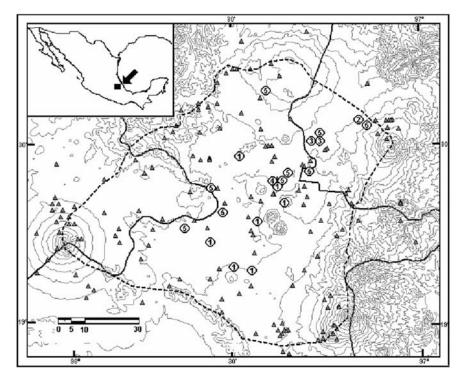


Figure 1. Geographic distribution of *Peromyscus bullatus*. The numbers indicate that: (1) collection localities where *P. bullatus* were found (Appendix 1); (2) type locality (Osgood 1904); (3) literature localities (Hoffmeister 1951; Hall and Dalquest 1963); (4) localities survey in this investigation and where *P. bullatus* were found (Table 3); (5) localities survey in this investigation and where *P. bullatus* were not found (Table 3) and triangles indicate the collection localities revised from the Oriental Basin were collected other species of rodents but not *P. bullatus* (total localities 177). The discontinuous line represents the limits of the Oriental Basin.

(*P. maniculatus*, *P. levipes*, and *P. difficilis*), and will probably be found with another species (*P. gratus*). No further comparison among *P. bullatus*, *P. levipes*, and *P. maniculatus* are presented nor are deemed necessary. In general size and external appearance, *P. bullatus*, *P. gratus*, and *P. difficilis* closely resemble one another, and their identification is difficult.

Interspecific comparisons were made using a sample of 31 *P. bullatus*, 45 *P. difficilis*, and 50 *P. gratus* all of them adults. Each species was compared with respect to the external and cranial measurements listed below, as well as several qualitative features of the pelage, and skull. We used *P. difficilis* specimens collected during the field trip from the same localities as those of *P. bullatus*. We used *P. gratus* specimens from Tlalpan and Pedregal de San Ángel, Distrito Federal, because there are no records from *P. bullatus* and *P. gratus* in the Oriental Basin area. Only adult specimens were used for comparison. The age were classified according with Hoffmeister (1951).

All measurements were taken with a dial caliper (0.01 mm accuracy). Cranial: occipito-nasal length (OL), brain case breadth (BC), postorbital constriction (PC), nasal length (NL), maxillary tooth row length (LX), bulla length (BL), bulla breadth (BW), and bulla depth (BD). Body measurements taken from the tag specimens: total length (TL), tail length (CL), hind foot length (HL), ear length (EL), and dry ear length (DL).

Measurements of eight bacula were taken with an ocular micrometer. Four specimens were stored and four were cleaned. Because the importance of the size of the ears and the bulla in *P. bullatus*, *P. difficilis*, and *P. gratus*. We used a ratio between hind foot length to ear length (HL–EL%) and occipito-nasal length with bulla length (OL–BL%) in the analysis between species.

Mean, range, and standard deviation were calculated for each measurement. The natural logarithm of each measurement was calculated for: analysis of differences among species, analysis of variance, and principal component analysis using a covariance matrix. Canonical analysis was performed using cranial measurements only. Uni- and multi-variate analyses of the data were performed with the Statgraphic v.5 computer software program (Statgraphics 1997).

Results and discussion

Morphometric comparisons

Univariate analysis. In 10 measurements, *P. bullatus* were smaller than *P. difficilis*, and nine were significantly smaller (p < 0.05). *P. bullatus* was larger in bulla length (BL), and significantly larger (p < 0.05) in the hind foot length to ear length ratio (HL–EL%), and occipito-nasal length with bulla length (OL–BL%). In relation to *P. gratus*, *P. bullatus* was slightly larger in 14 measurements and significantly greater (p < 0.05) in 11, *P. gratus* were larger only in the PC (Table 1).

Multi-variate analysis. Principal component analysis showed that the two first components account for 73.6% of the variations (Table 2). The first accounted for 62.7%, and can be attributed to size variation because the variables had positive correlation and similar values. The second component consisted of the three bulla measurements (BL, BW, and BD) and dry ear length (DL, Table 2), are positive and large coefficient, the other ones are negative, showing that the largest measurement of the bulla are distinctive for *P. bullatus*.

Graphic representation of the two first components (Figure 2) shows the differences in size and shape of the three species. In the first component, the relative sizes of *P. gratus*, *P. bullatus*, and *P. difficilis* ranged from small to large. In the second component, differences in size of the bulla and ear were measured. The discriminant analysis graph shows differences among skull characteristics of species (Figure 3), as shown in component analysis.

	Peromyscus	<i>us difficilis</i> $(n = 45)$	(Peromyscus	Peromyscus bullatus $(n = 30)$	(Peromyscu	Peromyscus gratus $(n = 50)$	
	Average	Min-Max	SD		Average	Min-Max	SD		Average	Min-Max	SD
OL	30.71	29.6–32.2	0.54	* *	27.99	27.4–28.6	0.38	*	27.09	25.8-28.3	0.60
BC	13.99	13.6 - 14.9	0.29	* *	13.34	12.9 - 15.0	0.39	*	12.86	12.3–13.4	0.29
PC	4.59	4.2-4.9	0.15	* *	4.43	3.7-4.8	0.19		4.45	3.6 - 4.9	0.22
NL	12.13	10.8 - 13.2	0.51	* *	10.41	9.5-11.1	0.36	*	10.00	9.0 - 10.9	0.41
LX	4.66	4.2 - 5.0	0.18	* *	4.14	3.8 - 4.4	0.15		4.06	3.7 - 5.9	0.30
BL	6.76	6.4-7.2	0.18		6.84	6.5-7.2	0.18	* *	6.05	5.7 - 6.4	0.18
BW	5.73	5.4 - 6.1	0.15		5.67	5.3 - 6.0	0.17	*	5.16	4.8 - 5.5	0.16
BD	5.16	4.9-5.5	0.16		5.16	4.8-5.7	0.27	*	4.67	4.2 - 5.3	0.25
TL	227.67	210.0 - 245.0	8.64	* *	197.04	178.0 - 224.0	10.52		194.78	158.0 - 214.0	11.14
CL	121.07	105.0 - 170.0	10.41	* *	103.64	87.0 - 120.0	7.08		101.12	61.0 - 116.0	9.94
HL	24.98	24.0 - 27.0	0.66	* *	22.93	19.0-25.0	1.46	*	22.08	19.5-27.0	1.21
EL	24.55	20.0 - 27.0	1.27	* *	25.67	23.0 - 28.0	1.35	*	20.95	17.0-24.0	1.34
DL	22.76	20.3 - 25.2	1.24		22.63	19.9 - 24.5	1.30	* *	19.35	10.9 - 21.8	1.71
% HL-EL	98.43	80.0 - 108.3	5.38	* *	112.55	92.0–142.1	11.53	*	94.96	81.5-105.0	6.25
% OL-BL	22.03	20.7–23.3	0.58	* *	24.46	23.2–25.7	0.68	* *	22.32	20.9–23.7	0.56
<i>n</i> , number of differences at	, number of specimens; SD, standard lifterences at $p < 0.05$, two at $p < 0.01$), standard deviatic at $p < 0.01$.	m; min, mir	nimum; n	aax, maximun	a, and <i>p</i> , probabili	ity of the AI	NOVA a:	nalysis. One a:	standard deviation; min, minimum; max, maximum, and p , probability of the ANOVA analysis. One asterisk indicates significant t $p < 0.01$.	gnificant

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Table 1.	

Variable	CP1	CP2	VC1	VC2
OL	0.343	-0.129	0.750	0.478
BC	0.317	-0.044	0.329	-0.111
PC	0.138	-0.154	-0.095	0.225
NL	0.329	-0.166	0.142	0.135
LX	0.284	-0.239	0.310	0.256
BL	0.297	0.408	0.067	-0.871
BW	0.316	0.296	-0.063	-0.264
BD	0.268	0.332	-0.006	-0.365
TL	0.313	-0.327		
CL	0.275	-0.369		
HL	0.283	-0.078		
DL	0.244	0.509		
Egenvalues	7.5	1.3	12.4	3.1
% Variance	62.7	10.9	80.1	19.9
% Accumulated	62.7	73.6	100.0	90

Table 2. Results of principal component analysis (CP1, CP2) and canonical variables of the discrimination analysis (VC1, VC2) for measurements of *Peromyscus bullatus*, *P. difficilis*, and *P. gratus*.

Specimens of *P. difficilis* were larger in greater occipito-nasal length (OL), brain case width (BC), and length of maxillary tooth row (LX, Table 2). In relation to *P. gratus, P. bullatus* OL and BC were smaller, but the species were distinguishable mainly by bulla measurements (BL, BW, BD) and occipito-nasal length (OL, Table 2).

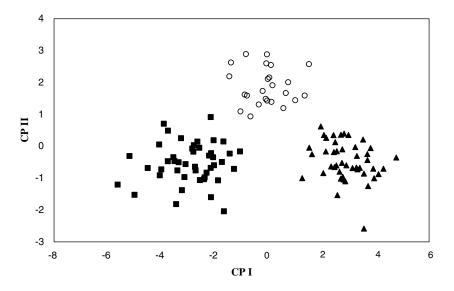


Figure 2. Projection of canonical variables I and II of *Peromyscus bullatus* (circles), *P. difficilis* (triangles), and *P. gratus* (squares).

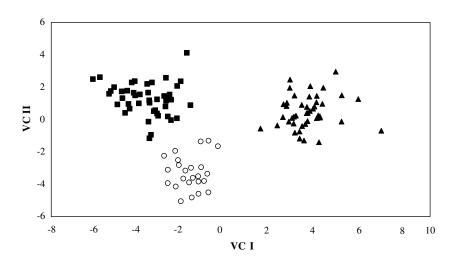


Figure 3. Projection of canonical variables I and II for discriminant analysis of Peromyscus bullatus (circles), P. difficilis (triangles), and P. gratus (squares).

Size ranking of *P. difficilis*, *P. bullatus*, and *P. gratus* was shown through uniand multi-variate analyses. Skull morphology of *P. bullatus* is more similar to *P. gratus* than to *P. difficilis*.

The *P. bullatus* can be distinguished easily from *P. difficilis* in most cranial and somatic measurements (Table 1). Some morphological overlap of *P. bullatus* and *P. gratus* is evident in all external and cranial measurements, making positive identification difficult. Nevertheless, *P. bullatus* and *P. gratus* fall into non-overlapping subsets in ANOVA analysis. But auditory bulla measurements provided evidence that *P. bullatus* is distinct from *P. gratus* at the specific level.

Hoffmeister (1951), Vial (1962), and Hall and Dalquest (1963) interpreted the large bulla and ear, relative to the auditory system of *P. bullatus*, as an evolutionary adaptation that provides an advantage for detecting predators in open habitat with sparse vegetation.

In the mammal collections, it is very common to confuse *P. bullatus* with *P. gratus* and *P. difficilis*. We found only six of the 20 specimens from these collections correctly assigned. Ramírez-Pulido et al. (2001) recorded *P. bullatus* specimens from El Limón, Veracruz (13929 UAM-I), and Minas de Arena La Caída, Puebla (13928 UAM-I), but we determined these specimens to be *P. levipes* and *P. difficilis*, respectively, according to the following characteristics.

Peromyscus bullatus was slightly smaller than *P. difficilis* (Table 1), and can be distinguished by a number of differences: occipito-nasal length was less than 28.6 mm, in contrast to 29.6 mm in *P. difficilis*. Ear length was greater, at 25.7 mm versus 24.6 mm (23.0–28.0 mm versus 20.0–27.0 mm, respectively),

and molars were heavier. The length of the bulla was similar in both species, with ratios of bulla to occipito-nasal length of 24.5% (23.2-25.7%) in *P. bullatus* and 22.0% (20.7-23.3%) in *P. difficilis*. In sympatric areas, juvenile *P. difficilis* have been mistaken for *P. bullatus*, but this study determined *P. bullatus* to be of lighter coloration, with heavier molars, and proportionately larger bulla.

Peromyscus bullatus was slightly larger than *P. gratus* (Table 1). The ear was longer than 23.0 mm (19.9 dry), whereas that of *P. gratus* was smaller than 24.0 mm (21.8 dry). The skull of *P. bullatus* was more elongated and the braincase was wider. The length of the bulla was over 6.5 mm, and the ratio of bulla to skull length was over 23.2%. In *P. gratus*, these measurements were less than 6.4 mm, and less than 23.2%, respectively. The species were more easily distinguished using the sum of bulla length, width, and depth as index. This index was greater than 16.1 in *P. bullatus*, and in *P. gratus* less than 15.4. The main characteristic that can be use for recognize *P. bullatus* from *P. gratus* and *P. difficilis* are the great value in the relation HL–EL and OL–BL. The color of *P. bullatus* was generally lighter, mainly on the sides and back and the ventral part of the bicolor tail was white, in contrast to the gray and white of *P. gratus*.

Distribution

Peromyscus bullatus was previously known from three localities in western Veracruz, Mexico: Perote (Osgood 1904), 2 km W of El Limón, and 3 km W of El Limón (Hoffmeister 1951; Hall and Dalquest 1963). The surveys reported in this paper increased the known distribution of *P. bullatus* to the southwest, in the state of Puebla. The localities were: 9.5 km N, 6.2 km E Oriental; 6 km S, 8 km W San José Alchichica, 2250 m; 11 km S, 6 km W San José Alchichica, 2450 m; 4.5 km S, 9.5 km W San José Alchichica, 2350 m; 3 km NE Zacatepec; 3.5 km N, 9 km W San Salvador el Seco, 2500 m; 5 km NE San Salvador el Seco, 2300 m.

Peromyscus bullatus has been recorded from seven new localities in Puebla and three previously known (Hall and Dalquest 1963) from western Veracruz. These 10 localities encompass the north central part of the Oriental Basin in the states of Puebla and Veracruz (Figure 1). This study shows that the main occurrence (75% of the area) of *P. bullatus* is in Puebla, and may include eastern Tlaxcala, because it is part of the Oriental Basin and is relatively close to the locations in Puebla (Figure 1).

From the data here presented, we established the territory of this species from $19^{\circ}12'$ to $19^{\circ}34'$ N, and from $97^{\circ}14'$ to $97^{\circ}49'$ W, a relatively small area of 1212 km², less than that recorded for *Spermophillus perotensis* (2427 km²), another endemic species in the Oriental Basin (Valdéz and Ceballos 1997). Closer inspection shows that *P. bullatus* is more restricted because it is found only in flat, sandy, and arid areas with an elevation from 2250 to 2500 m.

Habitat

Mountains and volcanoes, including the notable Pico de Orizaba, La Malinche, Cofre de Perote, and Tlaxco and Soltepec ranges, surround the Oriental Basin (Reyes 1979). The vegetation is varied and there are great numbers of ecotones. The most common associations are grasslands and coniferous forests at higher elevation and arid scrub in the bottomlands (Gómez 1978; Gaona 1997).

Vegetation in the grasslands was variable, characterized in some areas by bunch grass or prostrate herbs and short grass. A dominant grass species in the alkaline grasslands is *Distichlis spicata*. In this setting, other species of herbs and grasses include *Actinella chysanthemoides*, *Atriplex pueblensis*, and *Bouteloua breviseta*. Most herbs in this grassland are annual and complete their life cycle in late autumn.

Structure and composition of the arid scrub varies in the region mainly in relation to soil, slope, and humidity (Alvarez and González-Medrano 1972; Gómez 1978). Dominant species include cacti, such as prickly pear and *Agave* obscura.

Peromyscus bullatus is restricted to gentle rises in the flat, low areas, and areas of fine, sandy soil with grasses and few trees. The most conspicuous plant species are *Juniperus dippeana* and *Yucca* spp. Hall and Dalquest (1963) describe the same vegetation for specimens collected near El Limón. The most abundant species in the same locations where Perote mice were collected was *P. maniculatus*, but on slopes over 30° . In these areas junipers (*Juniperus* sp.) are abundant, as are fields of wheat and pasture crops. *P. difficilis* were collected in rocky areas. *P. bullatus* were not collected in either of the habitats previously mentioned for *P. difficilis* and *P. maniculatus*. Intense collection effort was made in areas near crops, principally wheat, no specimens of the Perote mouse were found.

The habitat of *P. bullatus* seems to be very specific. We found specimens at six from 15 localities. All successful sites were nearly flat and sandy, with many annual plants and tufted grasses. The occurrence of P. bullatus was never over 20% of specimens trapped (Table 3). P. bullatus is sympatric with P. difficilis, but has a very specific differentiation at the microhabitat level (Hall and Dalquest 1963). We found that P. bullatus were more abundant in sandy flat areas in relation to P. difficilis, which occupies rocky areas, mainly volcanic areas with abundant xerophytes and steep slopes. In some localities, P. difficilis occurred in the forest (Pinus sp. and Juniperus sp.) with its nest between the leaves of the yuccas (Yucca sp.). In the study area, the most abundant rodent was P. difficilis, which were found in more diverse habitats than P. bullatus. In the microhabitat where Perote mice were found, no P. difficilis were collected. Other rodents collected on sites containing P. bullatus included: Dipodomys phillipsii, Perognathus flavus, Reithrodontomys megalotis, Peromyscus maniculatus, and a few number, Liomys irroratus and Mus musculus.

Localities	Rode	nt specie	Rodent species trapped	p									и	ΓV	Traps
	1	2	3	4	5	9	7	8	6	10	11	12			
Modified															
9.5 km N, 6.2 km E Oriental	1	11		1									12	в	
3.5 km N, 7 km W San Salvador El Seco	1	18	7				ю		-	1			30	в	
3 km NE Zacatepec	1	35		7									37	U	
6 km S, 8 km W San José Alchichica	ю	9	20	1		б	1	0					33	A	
11 km S, 6 km W San José Alchichica	4	10		4	-								15	в	
Surveyed in this study															
4.5 km S, 9.5 km W San José Alchichica	11	9	35	26			S						72	D	560
4.2 km S, 6.6 km W San José Alchichica		46	19	13			1	1					80	D	240
2 km S, 5 km W San José Alchichica		6	12	9									27	Щ	240
1.5 km S, 1.5 km E San José Alchichica	9									0		8	Ľ	240	
Cantona 16 km N Tepeyehualco		29		1				-			с		34	IJ	240
10 km NW Zacatepec		13	4	Ч									19	в	240
3 km SE Perote		1								9		1	8	Η	240
2 km W Oriental		39		٢				21					67	в	240
2.5 km N El Limón		14	ε										17	C	240
15 km N, 7.5 km W San Salvador El Seco		0		ς				ы					7	IJ	240
Total	21	245	100	99	1	ю	10	27	1	7	5	1	466		2720

Table 3. Localities with habitat characteristics suitable for *Peromyscus bullatus*.

Rodent species: (1) *Peromyscus bullatus*; (2) *P. alifficilis*; (3) *P. maniculatus*; (4) *Reithrodontomys megalotis*; (5) *R. sumicrasti*; (6) *Perognathus flavus*; (7) *Dipodomys phillipsii*; (8) *Liomys irroratus*; (9) *Mus musculus*; (10) *Neotomodon alstoni*; (11) *Neotoma mexicana*; (12) *Microtus mexicanus*. Vegetation types (VT): (A) Grassland with few trees; (B) Crop areas; (C) Secondary vegetation; (D) Stony areas with xerophytes; (E) Acahuales; (F) Stony areas; (G) Brush areas; (H) Pine forests.

Morphology and variation

Molar morphology of *P. bullatus* was very similar to that described by Hoffmeister (1951) for *P. gratus*. The pattern of crest variation was similar (Hooper 1957), but *P. gratus* had a high percentage of entolophids in m1 and m2, whereas none was found in the *P. bullatus* specimens.

The baculum of *P. bullatus* was thin along the length, and the base was wide and flat. The dorsal-ventral position was convex, but the curvature varied among specimens examined. Cartilaginous tips reported for others species in the *truei* species group (Tamsitt 1958) were not found. The mean baculum length (n = 6) was 15.3 mm (14.3–16.6), and width (n = 6) at the base was 1.5 mm (1.3–1.7). The ratio of the baculum length in relation to the body length was 16.1% (15.3–17.2). The baculum of *P. bullatus* was similar to those described by Burt (1960) and Tamsitt (1958) for the *truei* species group, but was larger than those of *P. gratus* and *P. nasutus* and smaller than those of *P. difficilis*.

Conservation status

The known distribution of *P. bullatus* may include areas to the south because the potential habitat and the Oriental Basin extend to the south (Figure 1). We regard its present distribution as relict. The southernmost-recorded *P. bullatus* specimen came from San Salvador El Seco, Puebla, but the Oriental Basin continues almost to Ciudad Serdan, Puebla, approximately 55 km to the southwest.

The relict distribution of *P. bullatus* can be explained by any of the following three factors: (1) The survey team did not make an adequate collection effort. However, the southwestern area is well represented in Mexican mammal collections, and no specimens have been previously found. (2) The historic distribution has always been restricted to the north-central portion of the Oriental Basin. (3) Agricultural activities, mainly in open areas, have had a deleterious effect on the distribution of *P. bullatus* because corn and beans radically alter the vegetation cover. Based on current data and analysis of the preferred habitat, we estimate that *P. bullatus* is limited to 1200 km², and that the species is threatened.

The Perote mouse is very specific in habitat selection, and the species in threatened by habitat conversion and degradation. This habitat is not common, and is isolated from similar habitat types outside the Oriental Basin. Distribution probably has been undergoing natural reduction since the Pleistocene from climatic desiccation (Reyes 1979). Current agricultural activity appears to have increased isolation and decline in population. Evidence for this is that eight of ten locations that provided museum specimens in the 1980s yielded no Perote deer mice in 2001. The type localities of Perote, Veracruz (Osgood 1904) and near El Limon, Veracruz (Hoffmeister 1951), which have

been transformed to urban areas and factories; the area 9.5 km N, 6.2 km E Oriental is today covered by irrigated alfalfa fields; 3 km NE Zacatepec is near a cement mine, and the two localities near San Salvador el Seco have cattle grazing activity and different crops. In two of the three localities near Alchichica, we found *P. bullatus*, the third area is a cultivated peach orchard, and only *P. difficilis*, *P. maniculatus*, and *R. megalotis* were found (Table 3).

The combined loss of a small area that the species can inhabit, the natural isolation of the remaining suitable habitat, and the low densities found in the sampled areas, suggest that gene flow among the remaining populations will be reduced in a short time. There is evidence that isolated and/or small populations often have higher risk of extinction from loss of genetic diversity, environmental processes, or disease (Wilcox and Murphy 1985; Ceballos and Rodríguez 1993). *P. bullatus* should be registered as threatened, and placed under special protection (SEMARNAT 2002).

Peromyscus bullatus occupies in a region that does not have protected areas, probably because it is believed to be an area of low biodiversity with few endemic species. However, the region has three endemic species: *Peromyscus bullatus, Spermophillus perotensis*, and *Neotoma nelsoni* that probably had wider distribution in the Pleistocene. Clearly, we have conservation problems (Valdéz and Ceballos 1997; SEMARNAT 2002). In this area, we found *Dipodomys phillipsii, Peromyscus mekisturus*, and *Reithrodontomys chrysopsis*, species not restricted to this region, but which are endemic to Mexico. Conservation efforts should not focus on high biodiversity regions only (Arita 1993; Ceballos and Rodríguez 1993), but also on areas valuable for their information on historical biogeographic changes of the last few centuries as well.

Acknowledgments

We gratefully acknowledge two anonymous reviewers for their valuable comments and criticisms, and Brett Riddle for reviewing early draft and providing helpful comments, to the CIBNOR editing staff for providing helpful comments and editing the text. To José Ramírez Pulido of the Universidad Autónoma Metropolitana for allowing use of the mammals collection, to Jorge Villalpando, Nansy Sánchez, Francisco J. Navarro, and Aurelio Ocaña for valuable field and laboratory assistance, and to the Consejo Nacional de Ciencia y Tecnología (CONACyT) for Grant 39467Q that funded this research.

Appendix 1

Peromyscus bullatus: PUEBLA. 9.5 km N, 6.2 km E Oriental (ENCB 15061-15064); 6 km S, 8 km W San José Alchichica, 2250 m (ENCB 15617, 15619, and 15622); 11 km S, 6 km W San José Alchichica, 2450 m (ENCB 11809,

11811, 11812, and 11814); 4.5 km S, 9.5 km W San José Alchichica, 2350 m (ENCB 42324-42334); 3 km NE Zacatepec (ENCB 7934); 3.5 km N, 9 km W San Salvador el Seco, 2500 m (ENCB 15767); 5 km NE San Salvador el Seco, 2300 m (UAM-I 9466-9472).

Peromyscus gratus: DISTRITO FEDERAL. 2 km N, 1.5 km W Tlalpan, 2240 m (16); Pedregal de San Angel (13). ESTADO DE MÉXICO. Santa María Cuevas, 2480 m (21).

Peromyscus difficilis: PUEBLA. 6 km S, 8 km W San José Alchichica, 2250 m (2); 11 km S 6 km W San Salvador el Seco, 2450 m (8); 9.5 km N, 6.2 km E Oriental (2); 3 km NE Zacatepec (27); 3.5 km N, 7 km W San Salvador el Seco (6).

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