

SMALL MAMMALS (LAGOMORPHA AND RODENTIA) AND FASCIOLIASIS TRANSMISSION IN THE NORTHERN BOLIVIAN ALTIPLANO ENDEMIC ZONE

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ABSTRACT: Helminthological studies of small mammal species from the Northern Bolivian Altiplano were carried out to ascertain whether lagomorphs and rodents inhabiting this *Fasciola hepatica* endemic zone may or may not participate in the transmission of the liver fluke and play a reservoir host role regarding human and domestic animal fascioliasis. A total of 6 lagomorphs and 152 rodents from 17 different localities were analyzed, including: the hare *Lepus capensis*, the rabbit *Oryctolagus cuniculus* and the rodents *Mus musculus* (Muridae), *Akodon boliviensis*, *Akodon* cf. *boliviensis*, *Akodon* sp., *Calomys lepidus*, *Phyllotis osilae*, *Auliscomys sublimix*, *Auliscomys pictus* (Cricetidae), *Cavia porcellus*, and *Galea musteloides* (Caviidae). The morphologic and ecologic characteristics of these small mammal species are described and discussed. Their helminth-fauna is analyzed. No small mammal studied harboured the liver fluke. It is concluded that both lagomorphs and rodents may be disregarded as playing the role of reservoir hosts of *F. hepatica* and, hence, that they need not be taken into account within the necessary control measures to be implemented against the disease in the Northern Bolivian Altiplano endemic zone.

KEY WORDS: *Fasciola hepatica*, transmission, lagomorphs, rodents, human fascioliasis, Northern Bolivian Altiplano.

INTRODUCTION

The Northern Bolivian Altiplano, between Lake Titicaca and the valley of the city of La Paz, at 3800-4100 m altitude, presents the highest prevalences and intensities of human infection by *Fasciola hepatica* (Linnaeus, 1758) (Trematoda: Fasciolidae) known worldwide: prevalences of up to 70% in coprological surveys (MAS-COMA *et al.*, 1995; ESTEBAN *et al.*, 1997a, b; ANGLES *et al.*, 1997) and even higher in immunological surveys (HILLYER *et al.*, 1992; MAS-COMA *et al.*, 1995; BJORLAND *et al.*, 1995; STRAUSS *et al.*, 1997); intensities, measured as egg output in stools, ranging from 24 to 5064 eggs per gram (epg), with arithmetic and geometric means of 474-1001 and 201-309 epg, respectively (ESTEBAN *et al.*, 1997b).

Among the several research activities carried out to understand such high human infection rates, large mammal species inhabiting the endemic area and representing potential reservoir hosts were surveyed. Goats, horses, llamas and alpacas proved to develop no role in the epidemiology of the disease (MAS-COMA *et al.*, 1997) and the prevalences detected in sheep and cattle (UENO & MORALES, 1973; UENO *et al.*, 1975; MAS-COMA *et al.*, 1995; BUCHON & MAS-COMA, 1995; HILLYER *et al.*, 1996; BUCHON *et al.*, 1997), as well as in pigs and donkeys (MAS-COMA *et al.*, 1997) were not high enough to

give rise to a very large environmental contamination with liver fluke eggs.

Small mammals belonging to the orders Lagomorpha and Rodentia were then considered as perhaps contributing to the liver fluke transmission in the Bolivian Altiplano. The capacity of lagomorphs (hares, rabbits) to play the role of reservoir hosts of fascioliasis is already known in other regions (BAILENGER *et al.*, 1965; ARRU, DEIANA & NUVOLE, 1967; ALCAINO *et al.*, 1990; APT *et al.*, 1993). Concerning rodents, numerous rodent species have been found infected in nature (MAS-COMA *et al.*, 1987, 1988) and several experimental studies proved that the liver fluke is able to reach a normal development in given species (DAWES, 1962a, b, c; DAWES & HUGHES, 1964; BORAY, 1982; VALERO, MARCOS & MAS-COMA, 1997). Recent studies have proved that murid species, mainly the black rat *Rattus rattus* (Linnaeus, 1758), may effectively contribute to *F. hepatica* transmission under natural conditions and, hence, act as reservoir hosts of the disease (MAS-COMA & BARGUES, 1997; VALERO *et al.*, 1997).

The present paper concerns the results obtained in the helminthological studies of rodent species from the Northern Bolivian Altiplano. The aim was to ascertain whether rodents inhabiting this liver fluke endemic zone may or may not play a reservoir host role regarding human and domestic animal fascioliasis.

MATERIAL AND METHODS

Small mammal sampling: Concerning lagomorphs, hares and rabbits were collected. Wild hare specimens were sampled when found dead in roads. Domestic rabbits were bought from Aymara inhabitants of the endemic zone; only rabbits living free (in- and outdoors) and around fascioliasis transmission foci marked by fresh-water bodies presenting intermediate lymnaeid hosts were considered.

Rodent specimens were captured using small mammal traps placed at night. Rodent trapping was carried out throughout the Northern Bolivian Altiplano, mainly in localities where the presence of lymnaeid snails assuring *F. hepatica* transmission was previously verified. Localities in areas where human prevalences and intensities have proved to be high were preferentially surveyed. Domestic guinea pigs were bought from Aymaras living near fascioliasis transmission foci.

A total of 17 different localities of the Northern Bolivian Altiplano were prospected: Hichukkota, Huarina, Cuyahuani-Utavi, 3 km after Batallas on the road to Huarina, Matadero de Batallas, Karahuisa, 3 km before Batallas on the road from El Alto, Cohana, Chijipata Alto, Ancocahua, Ciudad Satélite El Alto, Laja, Tambillo, Corpa, Huankollu, Belén-Pituta, and Sullcata. The geographical distribution of localities furnishing lagomorphs and rodents for the present study is shown in Fig. 1.

Mammal specimen preparation and collection: Mammal (skull, skeleton and skin) specimens were prepared for collection according to the methodology described by GOSALBEZ (1987). On each lagomorph and rodent specimen, the following morphological data were measured: head and body length (HB); tail length (T); posterior leg length (PL); ear length (E); and weight (W). All specimens were deposited in the Colección Boliviana de Fauna of the Museo de Historia Natural of Cota Cota, La Paz, Bolivia.

Helminthological study of mammal hosts: Host specimens were dissected and the parasitological study performed according to well-known general methods (MAS-COMA, 1976; PRITCHARD & KRUSE, 1982). The search for helminth parasites was carried out by examining organs and viscera placed in a small water layer in a 7 cm-diameter Petri dish under 10x and 30x magnifications with a stereoscopic microscope. Initially, the bile duct was examined for parasites, followed by the thoracic and abdominal viscera and cavities, with thorough rinsing in water to secure recovery of all worms. All parasites recovered were noted in host individual record cards.

Helminthological techniques: The helminths collected were fixed and conserved in 70% alcohol until appropriate preparation for microscopic study, following techniques described by MAS-COMA (1976). Digenean specimens were fixed in Bouin's solution bet-

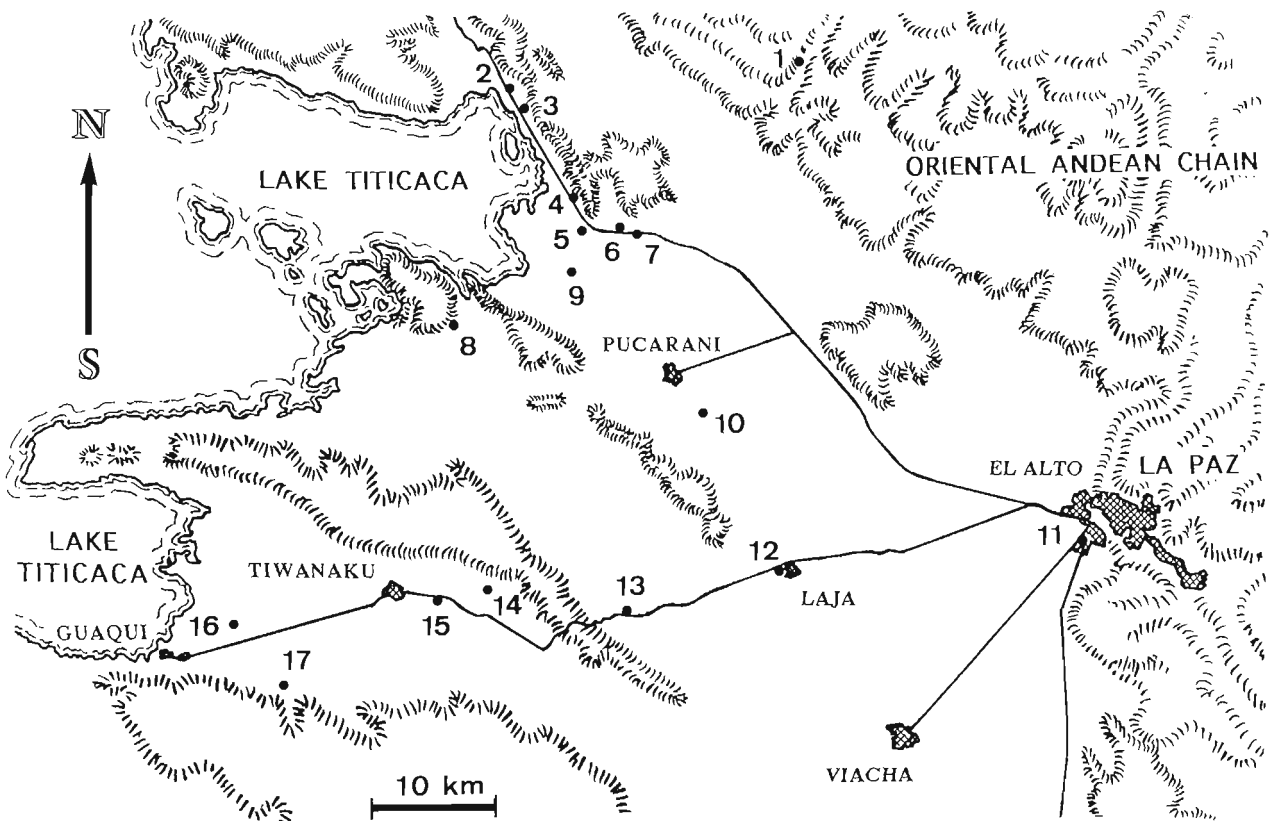


Fig. 1—Map of the Northern Bolivian Altiplano including the fascioliasis endemic zone and the localities in which lagomorphs and rodents were obtained: 1) Hichukkota; 2) Huarina; 3) Cuyahuani-Utavi; 4) 3 km after Batallas on the road to Huarina; 5) Matadero de Batallas; 6) Karahuisa; 7) 3 km before Batallas on the road from El Alto; 8) Cohana; 9) Chijipata Alto; 10) Ancocahua; 11) Ciudad Satélite El Alto; 12) Laja; 13) Tambillo; 14) Corpa; 15) Huankollu; 16) Belén-Pituta; 17) Sullcata.

ween slide and coverslip (with or without applying pressure, depending on specimens). Cestodes were fixed in hot 70% alcohol with hard manual shaking until appropriately relaxed specimens are obtained. Trematodes were stained with Grenacher's boracic carmine, and cestodes with alcoholic chlorhydric carmine. Plathelminths were afterwards differentiated with acidified alcohol, dehydrated in an alcohol series, cleared with xylene and mounted in Canada balsam between slide and coverslip. Nematodes were fixed in cold 70% alcohol and conserved in it until studied by direct examination in non-definitive extemporaneous preparations made directly in lactophenol clearing fluid, followed by recovery and preservation again in 70% alcohol.

RESULTS

Mammal hosts

Specimens of several different small mammal host species were obtained: 6 lagomorphs and 152 rodents. Within each mammal species the following data are noted: the number of specimens sampled (total, males and females) and localities in which specimens were collected. Their age according to approximate age groups (only noted if known), and their morphological measures (range and mean for each characteristic) are noted in Table 1. The classification of the mammal species was made following ANDERSON (1993). Systematics follows HONACKI, KINMAN & KOEPL (1982).

Order Lagomorpha: Specimens of the following two species belonging to the genera *Lepus* Linnaeus, 1758 and *Oryctolagus* Lilljeborg, 1871 (Leporidae) were obtained: the hare *Lepus capensis* Linnaeus, 1758 and the rabbit *Oryctolagus cuniculus* (Linnaeus, 1758).

–*Lepus capensis*: 2 specimens were studied, the sex of which could unfortunately not be determined. Localities: 3 km after Batallas on the road to Huarina: 1 indet.; 3 km before Batallas on the road from El Alto: 1 indet.

–*Oryctolagus cuniculus*: 4 specimens studied (2 ♂♂, 2 ♀♀). Localities: Tambillo: 2 ♂♂ + 2 ♀♀.

Order Rodentia: Specimens of the following eight species belonging to the genera *Mus* Linnaeus, 1766 (Muridae), *Akodon* Meyen, 1833, *Calomys* Waterhouse, 1837, *Phyllotis* Waterhouse, 1837, *Auliscomys* Osgood, 1915 (Cricetidae), *Cavia* Pallas, 1766 and *Galea* Meyen, 1832 (Caviidae), were obtained (in some *Akodon* specimens a definitive specific identification could not be made): *Mus musculus* Linnaeus, 1766; *Akodon boliviensis* Meyen, 1833; *Akodon* cf. *boliviensis* Meyen, 1833; *Akodon* sp.; *Calomys lepidus* (Thomas, 1884); *Phyllotis osilae* Allen, 1901; *Auliscomys sublimis* (Thomas, 1900); *Auliscomys pictus* (Thomas, 1884); *Cavia porcellus* (Linnaeus, 1758); and *Galea musteloides* Meyen, 1832.

–*Mus musculus*: 36 specimens studied (12 ♂♂, 24 ♀♀). Localities: Huarina: 1 ♀; Cuyahuani-Utavi: 1 ♀; Matadero de Batallas: 1 ♂; Karahuisa: 1 ♀; Cohana: 1 ♂;

Chijipata Alto: 4 ♀♀; Ancocahua: 1 ♂; Ciudad Satélite El Alto: 2 ♂♂ + 3 ♀♀; Laja: 1 ♂; Tambillo: 1 ♂ + 1 ♀; Corpa: 4 ♂♂ + 13 ♀♀; Sullcata: 1 ♂.

–*Akodon boliviensis*: 66 specimens studied (27 ♂♂, 39 ♀♀). Localities: Hichukkota: 4 ♂♂ + 4 ♀♀; Huarina: 1 ♂; Cuyahuani-Utavi: 1 ♂ + 1 ♀; Matadero de Batallas: 2 ♀♀; Karahuisa: 2 ♂♂; Cohana: 4 ♂♂ + 15 ♀♀; Chijipata Alto: 1 ♀; Laja: 9 ♂♂ + 8 ♀♀; Tambillo: 2 ♂♂ + 5 ♀♀; Corpa: 4 ♂♂ + 1 ♀; Huankollu: 1 ♀; Sullcata: 1 ♀.

–*Akodon* cf. *boliviensis*: 6 specimens studied (5 ♂♂, 1 ♀). Localities: Karahuisa: 1 ♂; Cohana: 2 ♂♂ + 1 ♀; Corpa: 2 ♂♂.

–*Akodon* sp.: 1 specimen studied (1 ♂). Locality: Laja: 1 ♂.

–*Calomys lepidus*: 8 specimens studied (4 ♂♂, 4 ♀♀). Localities: Chijipata Alto: 2 ♀♀; Laja: 2 ♂♂ + 1 ♀; Tambillo: 1 ♀; Huankollu: 1 ♂; Belén-Pituta: 1 ♂.

–*Phyllotis osilae*: 19 specimens studied (14 ♂♂, 5 ♀♀). Localities: Hichukkota: 1 ♂ + 1 ♀; Huarina: 1 ♀; Cuyahuani-Utavi: 2 ♂♂; Cohana: 5 ♂♂ + 1 ♀; Corpa: 1 ♂ + 2 ♀♀; Sullcata: 5 ♂♂.

–*Auliscomys sublimis*: 2 specimens studied (1 ♂, 1 ♀). Localities: Chijipata Alto: 1 ♀; Corpa: 1 ♂.

–*Auliscomys pictus*: 1 specimen studied (1 ♂). Locality: Hichukkota: 1 ♂.

–*Cavia porcellus*: 3 specimens studied (1 ♂, 2 ♀♀). Locality: Chijipata Alto: 1 ♂ + 2 ♀♀.

–*Galea musteloides*: 10 specimens studied (7 ♂♂, 3 ♀♀). Locality: Chijipata Alto: 7 ♂♂ + 3 ♀♀.

Helminth species

No helminth was found in the hares and rabbits studied. The distribution of the trematodes, cestodes and nematodes found in the rodent specimens analyzed is noted in Table 2. The helminth species in question will be identified and described in a later paper. No acanthocephalans were found. No helminths were found in the specimens analyzed from the species *A. pictus*, *C. porcellus* and *G. musteloides*.

No liver fluke was found. The only digenean specimen detected was found in the intestine of a specimen of *P. osilae* from the locality of Sullcata. It was a very small, immature fluke, whose morphoanatomic characteristics enabled its classification within the superfamily Plagiorchioidea (see YAMAGUTI, 1971).

Only a very few cestodes were detected. Hepatic taeniid larvae were found in *M. musculus*, *A. boliviensis* and *P. osilae*, and hymenolepidid adult specimens were obtained from the intestine of *M. musculus* and *A. boliviensis*.

Nematodes belonging to the superfamilies Trichinoidea, Trichostrongyloidea, Oxyuroidea, Spiruroidea, Physalopteroidea, and Rictularioidea were found in all rodent species, but mainly in *M. musculus*, *A. boliviensis* and *P. osilae*.

Small mammal species	n	HB (mm)	T (mm)	PL (mm)	E (mm)	W (g)
LAGOMORPHA						
<i>Lepus capensis</i>	2					
age indet.	2	560 ^a	113-116 (114,5)	137-151 (144,0)	99-110 (104,5)	3350,0-4300,0 (3825,0)
<i>Oryctolagus cuniculus</i>	4					
age indet.	4	375-550 (425,0)	71-86 (77,3)	87-96 (91,0)	76-85 (80,0)	1200,0-1350,0 (1262,5)
RODENTIA						
<i>Mus musculus</i>	36					
young	1	65	65	18	12	9,6
subadult	4	66-83 (74,8)	63-83 (75,0)	14-18 (16,3)	11-15 (13,3)	7,5-17,0 (12,3)
adult	26	68-98 (86,3)	72-89 (81,4)	14-19 (17,7)	13-19 (14,4)	11,9-26,9 (19,4)
old	5	87-96 (92,0)	69-90 (80,6)	17-19 (17,8)	12-16 (14,6)	18,0-27,0 (21,3)
<i>Akodon boliviensis</i>	66					
young	3	67-72 (69,7)	46-52 (49,3)	18	12-14 (12,7)	9,9-10,8 (10,2)
subadult	3	75-83 (78,3)	58-66 (61,3)	18-20 (19,0)	12-14 (13,0)	12,8-19,0 (15,3)
adult	50	69-119 (90,1)	45-78 (63,8)	18-22 (20,4)	10-17 (13,8)	18,5-49,0 (24,8)
old	8	90-119 (105,6)	64-75 (70,3)	17-22 (20,4)	14-19 (15,0)	24,1-46,0 (38,0)
<i>Akodon cf. boliviensis</i>	6					
young	1	72	50	18	13	10,1
adult	4	86-105 (94,5)	61-78 (70,5)	20-21 (20,8)	14-15 (14,3)	22,7-26,7 (24,3)
<i>Akodon sp.</i>	1					
old	1	114	86	21	16	37,7
<i>Calomys lepidus</i>	8					
young	1	58	44	17	16	10,0
subadult	4	66-86 (74,3)	31-51 (37,0)	16-20 (17,3)	13-18 (15,0)	8,6-19,0 (12,2)
adult	3	83-91 (87,3)	43-47 (45,0)	16-17 (16,3)	14-17 (15,3)	14,9-30,0 (22,6)
<i>Phyllotis osilae</i>	19					
young	2	86-87 (86,5)	93-96 (94,5)	24-25 (24,5)	18-19 (18,5)	17,7-18,0 (17,8)
subadult	1	94	106	25	19	26,0
adult	14	101-135 (114,0)	110-140 (125,6)	25-29 (26,9)	19-25 (21,9)	30,0-54,9 (39,7)
old	2	118-134 (126,0)	92-122 (107,0)	27-28 (27,5)	22-23 (22,5)	35,2-50,0 (42,6)
<i>Auliscomys sublimis</i>	2					
subadult	1	88	49	21	19	21,0
adult	1	122	51	20	21	46,0
<i>Auliscomys pictus</i>	1					
adult	1	114	96	25	21	48,3
<i>Cavia porcellus</i>	3					
adult	3	280-510 (359,3)	— ^b	44-51 (48,3)	33-38 (35,3)	1000,0-1225,0 (1153,3)
<i>Galea musteloides</i>	10					
baby	2	100-105 (102,5)	— ^b	22-26 (24,0)	15	35,0-48,0 (41,5)
young	1	160	— ^b	31	21	104,0
adult	7	186-237 (210,7)	— ^b	33-38 (35,6)	22-26 (24,3)	272,0-660,0 (529,6)

Table 1—Lagomorph and rodent species studied from the Northern Bolivian Altiplano: species distributed according to approximate age groups (only noted if known); morphological measures with range and mean in parentheses for each characteristic; HB = head and body length; T = tail length; PL = posterior leg length; E = ear length; W = weight. n = number of specimens studied; ^a = only in 1 specimen could be measured; ^b = these species have no tail.

Host species	<i>Mus musculus</i> n = 36	<i>Akodon boliviensis</i> n = 66	<i>Akodon cf. boliviensis</i> n = 6	<i>Akodon</i> sp. n = 1	<i>Calomys lepidus</i> n = 8	<i>Phyllotis ositae</i> n = 19	<i>Auliscomys sublimis</i> n = 2
Hosts analyzed	No. (%)	No. (%)	No. (%)	No. (%)	No. (%)	No. (%)	No. (%)
TREMATODA	– (–)	– (–)	– (–)	– (–)	– (–)	1 (5.26)	– (–)
Plagiorchioidea	– (–)	– (–)	– (–)	– (–)	– (–)	1 (5.26)	– (–)
CESTODA	3 (8.33)	13 (19.70)	– (–)	– (–)	– (–)	1 (5.26)	– (–)
Taeniidae larvae	2 (5.56)	7 (10.61)	– (–)	– (–)	– (–)	1 (5.26)	– (–)
Hymenolepididae	1 (2.78)	7 (10.61)	– (–)	– (–)	– (–)	– (–)	– (–)
NEMATODA	14 (38.89)	46 (69.70)	5 (83.33)	1 (100)	1 (12.50)	15 (78.95)	1 (50.00)
Trichinelloidea	– (–)	– (–)	– (–)	– (–)	– (–)	1 (5.26)	– (–)
Trichostrongyloidea	2 (5.56)	13 (19.70)	2 (33.33)	– (–)	– (–)	13 (68.42)	1 (50.00)
Oxyuroidea	13 (36.11)	33 (50.00)	3 (50.00)	1 (100)	1 (12.50)	2 (10.53)	– (–)
Spiruroidea	– (–)	3 (4.54)	– (–)	– (–)	– (–)	– (–)	– (–)
Physalopteroidea	– (–)	7 (10.61)	– (–)	– (–)	– (–)	– (–)	– (–)
Rictularoidea	– (–)	3 (4.54)	– (–)	– (–)	– (–)	1 (5.26)	– (–)
TOTAL	16 (44.44)	49 (74.24)	5 (83.33)	1 (100)	1 (12.50)	15 (78.95)	1 (50.00)

Table 2– Helminth species (noted at superfamily level) found in the rodents harbouring parasites in the Northern Bolivian Altiplano. Lagomorph and rodent species studied but presenting no helminth are not included. No. = number of hosts infected; % = percentage of hosts infected.

DISCUSSION

Lagomorphs

The total lack of helminth parasites in the hares and rabbits studied is surprising. At any rate, it may be explained by taking into account that both lagomorph species were introduced into South America (CORBET, 1978; HONACKI, KINMAN & KOEPL, 1982). The hare *L. capensis* is originally from the Palearctic Region and, according to Aymara inhabitants, it has only very recently been introduced into the Bolivian Altiplano from Peru. The original range of the rabbit *O. cuniculus* was probably limited to Iberia and N.W. Africa. Most likely, after being domesticated there, it was introduced worldwide. Moreover, the hare and the rabbit are typical inhabitants of lowlands and their presence at very high altitude in the Altiplano evidently represents an extreme in their range. All this suggests that both lagomorphs have lost their helminth parasites when colonizing the Altiplano.

The possibility of the hare and the rabbit becoming infected by the liver fluke in the Bolivian Altiplano cannot be disregarded. At any rate, in the Altiplano the low populations of both lagomorphs, the ecology of the hare far away from zones presenting fresh-water collections with lymnaeid snails, and the marked peridomestic habits of the rabbit, allow us to conclude that they are not able to significantly participate in the transmission of the disease. Consequently, it is not necessary to consider them when implementing control measures against fascioliasis.

Rodents

All rodent species captured are typical South American species inhabiting Andean zones, except the house mouse *M. musculus*. The latter has its original range in the Palearctic Region and was able to expand worldwide through commensal forms (REICHSTEIN, 1978). Interestingly, it is apparently the only murid species able to reach the extreme altitude of the Altiplano; at a lower altitude, besides the house mouse, only one other murid species has been cited in the city of La Paz: the black rat *Rattus rattus alexandrinus* (Geoffroy, 1803) (MERCADO TUDOR & MIRALLES GOYTIA, 1991).

Although specimens from 8 rodent species were studied, the rodent species spectrum detected is far from the 18 species known to be present in this Andean area (MERCADO TUDOR & MIRALLES GOYTIA, 1991). However, the rodents here analyzed may be considered as the species living in or near *F. hepatica* rural endemic zones in the Bolivian Altiplano.

Among these species, *M. musculus* and *A. boliviensis* seem to be the most abundant around transmission foci. At any rate, our observations suggest that, in the Altiplano, no rodent species has a behaviour including frequent visits to water-collections or an evident preference for humid habitats. The almost total absence of digenans can be understood in this way, since no xerophilous terrestrial snail species exist in the Altiplano. The only trematode specimen found, a plagiorchoid, is most probably a bird parasite.

Moreover, the 6 species of the family Cricetidae are all small sized species, too small to be able to develop the

role of a definitive host able to harbour enough liver fluke adults to be considered a reservoir host in the disease transmission. VALERO *et al.* (1997) demonstrated that, in black rats, only animals presenting a head-body length of more than 140 mm and a weight of more than 51.5 g were infected by liver flukes under natural conditions.

The 2 Altiplanic species of the family Caviidae, *C. porcellus* and *G. musteloides*, are larger in body size and weight, but their ecological characteristics are not appropriate to participate in the transmission of *F. hepatica*. *C. porcellus* appears in the Bolivian Altiplano only as a domesticated animal which rarely goes out of the house and the garden of its human owners. Moreover, only a relatively few Aymara families have guinea pigs in the fascioliasis endemic zone. *G. musteloides* is a subterranean species which inhabits zones, usually plains with agricultural fields, far away from water collection transmission foci.

The parasitological findings suggest that the helminth-fauna of rodents inhabiting the Northern Bolivian Altiplano is very poor both qualitatively (only a few helminth species present) and quantitatively (only a few helminth specimens were found in each infected rodent host individual, if any). Taking into account that the cricetid and caviid species found are rodents adapted to live in high altitude conditions (MERCADO TUDOR & MIRALLES GOYTIA, 1991), it can be deduced that only a few parasite species of these Andean rodents are able to adapt to the high altitude environmental characteristics.

Finally, the results of this study allow us to conclude that the rodents may be disregarded as playing the role of reservoir hosts of *F. hepatica* and, hence, that they need not be taken into account within the necessary control measures to be implemented against fascioliasis in the Northern Bolivian Altiplano endemic zone.

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