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Distribution and abundance of sigmodontine rodents in relation to hantavirus in Neuquén, Argentina

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ABSTRACT**:** In order to estimate spatial distribution, temporal variation, and prevalence of Andes hantavirus antibody in the rodent community, and especially in *Oligoryzomys longicaudatus* populations, four different ecosystems were trapped seasonally between spring 2001 and winter 2002 in Neuquén, northwestern Argentinean Patagonia. Five peridomestic settings were sampled within the same period. The rodent *O. longicaudatus* had the widest distribution in Neuquén, as it was the only species captured at every sample site except for the High Andean steppe, and it was also the most common species captured. Rodents of 13 species were tested for hantavirus antibody prevalence, but *O. longicaudatus* and *Abrothrix longipilis* were the only seropositive species. Seropositive individuals were captured during spring and summer in the Subantarctic forest and in winter 2001 in a peridomestic setting in the Patagonian steppe. The dominant presence of *O. longicaudatus* throughout Neuquén must be incorporated into strategies to prevent human exposure to hantavirus. ***Journal of Vector Ecology* 30 (1): 119-125. 2005.**

Keyword Index: Andes hantavirus, Argentinean Patagonia, sigmodontine rodents, spatio- temporal variations, peridomestic settings.

INTRODUCTION

In recent decades, the appearance of many zoonotic diseases transmitted by rodents, ranging from minor episodes to huge epidemics, has indicated the great capacity of these agents to move out of natural habitats, and even, in some cases, to become an emergent disease under appropriate conditions (Enría 1998). In 1993, an outbreak of acute respiratory distress among people in the southwestern United States resulted in the identification of hantavirus in the New World (Nichol et al. 1993). From 1993 to 2004 more than 1,900 cases of hantavirus with pulmonary syndrome (HPS) have been reported in America with more than 1,400 cases in South America and about 590 cases in Argentina (Pan-American Health Organization 2004).

In Patagonia, HPS is caused by the Andes virus (ANDV) which is transmitted by the sigmodontine rodent *O. longicaudatus* (López et al. 1996, Levis et al. 1998). The rodent sheds virus into the environment through urine, feces, and saliva when the virus establishes a chronic infection (Lee et al. 1981, Hutchinson et al. 1998). Viral transmission to humans occurs through inhalation of infectious aerosols from secretions and excretions of infected mice (Tsai 1987, Glass 1997). In this region of Argentina (Enría et al. 1996, Wells et al. 1997, Padula et al. 1998) and Chile, person-to-person transmission of ANDV was proven by epidemic and genetic analysis (Toro et al. 1998). From February 1997 to April 2004, 31 cases of HPS were confirmed in Neuquén Province, northwestern Argentinean Patagonia, with a death rate of 45.2% (Elder, unpublished data). These cases occurred along a narrow western fringe of the province between 37.5º S and 41º S.

The first step in the study of the ecology of rodents acting as reservoirs and their relationship with the disease they transmit is to determine their geographic distribution and the range of the pathogen within the host range (Mills and Childs 1998). The general distribution of the main reservoir of ANDV in Patagonia, *O. longicaudatus,* is throughout the Andean region, from southern Tierra del Fuego up to the Argentinean-Bolivian border (Redford and Eisenberg 1992). In general *O. longicaudatus* seems to prefer moister areas with abundant cover (Pearson 1983, 1995), so it was always thought to inhabit mainly in forests and brushy areas. However, the specific distribution of *O. longicaudatus* within Neuquén province is poorly known. Preliminary studies of reservoir species conducted in the province after the first human cases appeared, between 1996 and early 1999, identified seropositive *O. longicaudatus* in the central-western (El Huecú and Trolópe) (Calderón et al. 1999) and southwestern (Villa La Angostura) regions of Neuquén (Public Health Subsecretary of Neuquén, unpublished data).

Occurrence of HPS cases in the region may be closely related to the periodic bloom of bamboo (*Chusquea culeou*) in the subantarctic forest. During 2001, this bloom, which reappears in the same patch once every 60-80 years, took place over 80,000 ha of a total of 378,000 ha in Lanín National Park which is located between 39º-40.5º S and the border with Chile-71º W. (Sanguinetti and Garcia, unpublished data).

Prodigious seed production after flowering produced an extraordinarily high nutritional resource that may have resulted in an increase in the rodent populations and therefore of the hantavirus reservoir species (Spotorno et al. 2000, Meserve et al. 1999, Jiménez et al. 1992). Approximately 47% of the total HPS cases in Neuquén were recorded during the bamboo blooming period (Elder, unpublished data).

In response to the appearance of HPS cases in Neuquén and insufficient data on the spatial ecology of the rodent-virus system, we began studies throughout the province to provide information on the temporal-spatial distribution of rodent species, as well as their densities and ANDV antibody seroprevalence. This is the first systematic study of the sigmodontine rodent assemblage throughout the different ecosystems of Neuquén. By identifying potential endemic areas, this study provides information that will aid in the development of mechanisms and strategies for preventing HPS.

MATERIALS AND METHODS

Sylvan study area and trapping

Sylvan small mammal trapping was conducted on six grids in four main ecosystems of Neuquén (Figure 1). The main characteristics of these environments and the geographic location of the sample sites are described in Table 1 (Cabrera and Willink 1980, León et al. 1998).

In each sample site, we set a ten by ten grid covering 0.81 ha with trap spacing of 10 m, consisting of 100 Sherman (8x9x23 cm) live traps for four consecutive nights, once per season, except in Villa La Angostura (VLA) and in Paraje Challacó. In VLA, because of the dense forest, we were able to set only eight lines of ten traps and sample in summer and winter. In Paraje Challacó, we set a twelve by twelve grid during spring and autumn. In all cases, traps were baited with rolled oats, bovine fat, and vanilla scent. To avoid mortality during cold seasons, we placed cotton bedding inside the traps. Traps were checked once a day during the morning. We replaced the bait or the cotton if necessary.

Peridomestic study area and trapping

Five additional sites were sampled in peridomestic settings or environments with some human use (Figure 1). In Paraje El Contra, traps were set, once per season, in a small farm on the south shore of Huechulafquen Lake. In Las Coloradas, ranch buildings were sampled during spring 2001, and an agricultural rural school was sampled during the summer. At Paraje Sauzal Bonito (38º 35´ 54´´ S and 69º 05´ 28.9´´ W), a lacustrine area inside the Monte shrublands, traps were set during autumn in farm dwellings. We sampled at Villa Pehuenia (subantarctic forest - 38º 51´ 3.3´´ S and 71º 13´ 31.5´´ W), a resort town, in an araucaria (*Araucaria araucana*) forest on Aluminé Lake. Ten traps were set for four nights in the vicinity of the Aluminé beach during summer, and in the autumn 40 more traps were set in an araucaria and radal (*Lomatia hirsuta*) forest. In Paraje San Cabao (Patagonian steppe - 39º 55´ 1.0´´ S and 71º 06´ 18.4 W), a periurban area on the banks of the Chimehuín River, at 6 km west from the town of Junín de los Andes, we set two parallel trap-lines of 41 traps (spaced 15 m) parallel to the river and separated by 30 m. Trap lines traversed llama (*Lama glama*) corrals and the bank of an artificial lagoon and were set for six consecutive nights during winter 2001 and winter 2002. Except for these last two sample sites, traps were set in typical rural buildings such as barns, storehouses, hen houses, greenhouses, and corrals for four nights. The number of traps set depended on the size and surface of the building.

Rodents were anesthetized with ethylic ether and marked with unique codes by ear cutting. Blood samples for seroprevalence determination were obtained from the retroorbital sinus and external morphologic characteristics (sex, weight, scars, breeding condition, etc.) were recorded. Sylvan animals were released at the exact site of capture. Animals captured inside buildings were subjected to the same procedure but were then sacrificed by cervical dislocation. Handling of rodents followed standardized safety guidelines described in Mills et al. 1995.

For detection of ANDV specific IgG antibodies, enzyme linked immunosorbent assay (ELISA) tests were performed on rodent blood serum samples as described by Padula et al. 2000. Rodent samples and control sera were diluted 1:200 and twofold up to 1:1600. A recombinant ANDV

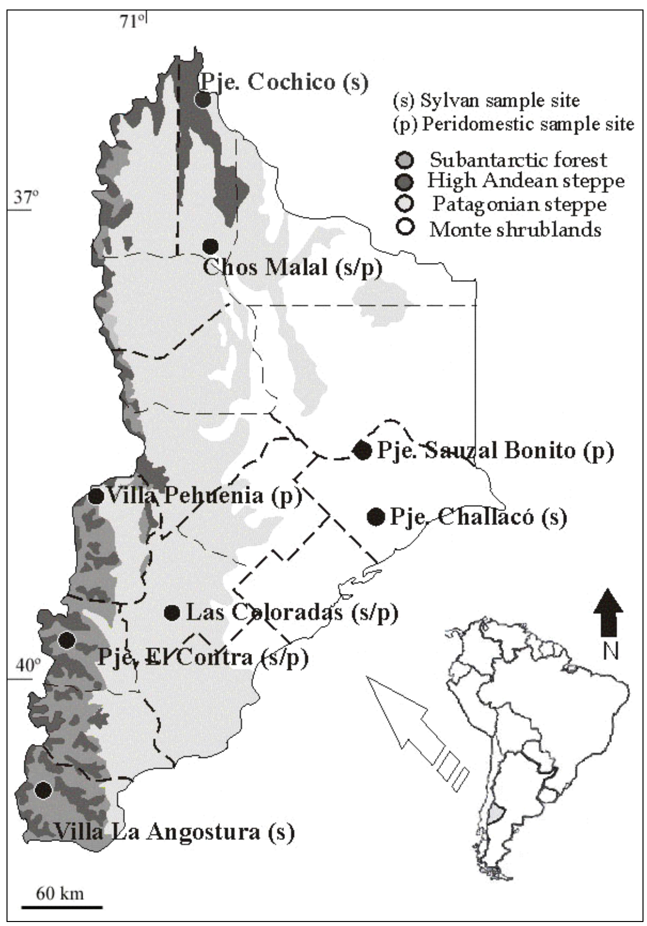

Figure 1. Geographical location of the sample sites in Neuquén province showing the four main ecosystems (or phytogeographic provinces) and the peridomestic or/and sylvan sample sites.

nucleoprotein was used as specific antigen. IgG in rodents was detected using peroxidase-labelled affinity-purified antibody to *Peromyscus leucopus* IgG (H+L) as the conjugate. ABTS (2.2'-azino-di [3-ethyl-benzthiazoline sulphonate]) was used as substrate for peroxidase and absorbance was measured at 405 nm. OD of each test sample was substracted by OD of the non-specific antigen corresponding dilution run on the same microtiter plate. Samples with OD less than 0.3 were considered negative. Signal reaction was detected using ABTS as substrate for peroxidase. All commercial reagents were from Kirkegaard and Perry Inc.

Data analysis

Abundance was estimated using an enumeration technique (Chitty and Phipps 1996) to determine the minimum number alive (MNA) and using the software 2CAPTURE (White et al. 1982) for closed population capture-recapture studies. 2CAPTURE evaluates the fit of the data to different models of patterns of capture probability for the population and indicates the most appropriate abundance estimator. Population densities (ind/ha) were estimated as a ratio between abundance and the area occupied by the grid. A strip of width equal to the average distance moved by individuals between captures was added to obtain the effective area of the grid and avoid overestimation of this parameter and "edge effect" (Seber 1982, Corley et al. 1995). In peridomestic settings, relative abundance of rodents was calculated as the number of individuals captured per 100 trap nights (ind/100 tn). Trap success was also determined using the total captures (captures and recaptures) per 100 trap night (capts/100 tn).

RESULTS

Sylvan areas

In 8,016 tn during the four seasons between September 2001 and August 2002, 297 sigmodontine rodents of seven species were captured a total of 508 times (Table 2). The overall trap success was 6.34 captures/100 tn ranging from no captures in the Monte shrublands to a peak of 32 captures/100 tn in Chos Malal during the autumn trapping event.

The most abundant rodents in the forest were *A. longipilis* and *O. longicaudatus,* comprising 92.6% of the individuals captured (Table 2). *A. longipilis* was also the rodent most recaptured (about 73% of recaptures in forests). Rodent density was significantly higher at Paraje El Contra in spring than during the rest of the sampling periods (χ^2 =28.8, p<0.01) (Table 2). Following spring, a major reduction in densities (between 47 and 10 times lower than the peak) occurred during subsequent trapping events. The VLA sample site had a high density in summer and a much lower density in winter (Table 2).

The highest rodent densities in the Patagonian steppe occurred during autumn (Las Coloradas and Chos Malal). These densities were significantly higher than during other seasons (Las Coloradas: χ^2=9.4, p=0.002 and Chos Malal: χ^2=57.55, p<0.05). *Eligmodontia* sp. and *A. iniscatus* were the most common rodents captured in the Patagonian steppe, comprising 89% of the captures, alternating their dominance seasonally. *O. longicaudatus*, in the Patagonian steppe, was captured only during autumn, the period of highest total rodent density. In Paraje Cochico, the highest densities were in winter (ä=12.4 ±0.37 ind/ha; Table 2) and the High Andean steppe was the only sample site where *Abrothrix xanthorhinus* was captured.

Peridomestic areas

During the same period in peridomestic settings, a total of 73 sigmodontine rodents were captured in 1,616 tn: 31 *Mus musculus*, 28 *O. longicaudatus*, 4 *A. olivaceus*, 4 *A. longipilis*, 3 *Eligmodontia* sp., 1 *Rattus* sp., 1 *Phyllotis darwini,* and 1 *Calomys musculinus*. The overall trap success was 4.5 captures/100 tn. The highest trap success was registered during autumn in Paraje Sauzal Bonito and during summer in Las Coloradas (34.6 and 32.5/100 tn, respectively) and the lowest in winter 2002 in Paraje Contra (0 captures/100 tn). *M. musculus* and *O. longicaudatus* were the rodents most frequently captured. *O. longicaudatus* was the only species captured in every trapping event, in all ecosystems,

Table 1. Main characteristics and geographic location of the sylvan sample sites.

Ecosystem	Sylvan sample site	Geographic location	Vegetation types	Climate	Key plant species
Subantarctic forest	Pje. El Contra	39º 47´ 5.7´´ S 71º 22´ 21´´ W	Dense forests and water meadows.	Temperate and humid. Rain from 750 to 4,000 mm, snows from autumn to spring.	Southern beech (*Nothofagus* spp.), bamboo (*Chusquea* spp.), and *Berberis* spp.
	Villa La Angostura	40º 44´ 58.3´´ S 71º 38´ 16.5´´ W			
Patagonian steppe	Chos Malal	37º 19´ 5.2´´ S 70º 09´ 40.7´´ W	Shrub grasslands.	Dry and temperate-cold with very strong westerly winds. Precipitation between 130 - 1,500 mm, snow in winter.	Neneo (*Mulinum spinosum*), bunchgrasses (*Festuca* spp., *Stipa* spp., and *Poa* spp.)
	Las Coloradas	39º 33´ 30.5´´ S 70º 35´ 20.3´´ W			
High Andean steppe	Pje. Cochico	36º 30´ 12.6´´ S 70º 11´ 16.9´´ W	High grasslands and semideserts over 2,200 m.a.s.l.	Snow precipitation or hail. Low temperature and intense solar radiation. Variable and intense winds.	Coliguay (*Colliguaya integerrima*), molle (*Schinus* polygamus), senecio (*Senecio* spp.), and uña de gato (*Nassauvia* sp.)
Monte shrublands	Pje. Challacó	38º 58´ 22.17´´ S 68º 57´ 45.6´´ W	Shrub grasslands with a vegetation cover of 40 to 60%.	Dry and warm. Precipitation from 80 to 250 mm distributed over the whole year.	Jarillas (*Larrea* sp.), algarrobos (*Prosopis* sp.), and matasebo (*Montea aphylla*)

Table 2. Densities (ind/ha), minimum number alive (MNA), and prevalence of ANDV infection among sigmodontine rodents captured in sylvan settings of four different ecosystem sampled during four trapping events.
SF: Subantarctic forest, **PS**: Patagonian steppe, **HA**: High Andean steppe, **MS**: Monte shrublands, **Ab prev**: Antibody prevalence, **VLA**: Villa La Angostura, ***Ol***: *Oligoryzomys longicaudatus*, ***Al***: *Abrothrix longipilis*, ***Ao:*** *Abrothrix olivaceus*, ***Ai:*** *Akodon iniscatus*, ***E***: *Eligmodontia* sp., ***Pd:*** *Phyllotis darwini*, ***Ax:*** *Abrothrix xanthorhinus.*

Trapping event	Ecosystem	Location	Trap nights	Total rodent density± SE	*Ol* MNA (# tested)	*Ol* Ab prev (%) (n)	*Al* MNA (# tested)	*Al* Ab prev (%) (n)	Other species MNA (# tested)				
									Ao	*Ai*	*E*	*Pd*	*Ax*
Spring 2001	SF	Pje. El Contra	400	93.9 ± 23.60	**31** (29)	**3.4** (1)	**37** (34)	**8.8** (3)	4 (4)	0	0	0	0
	PS	Las Coloradas	400	6 ± 2.60	0	0	0	0	0	2 (2)	3 (3)	2 (2)	0
	PS	Chos Malal	400	3 ± 0.09	0	0	0	0	0	2 (2)	3 (3)	0	0
	HA	Pje. Cochico	400	0.8 ± 0.03	0	0	0	0	0	0	0	1 (1)	0
	MS	Pje. Challacó	576	0	0	0	0	0	0	0	0	0	0
Summer 2001-2002	SF	Pje. El Contra	400	8.8 ± 2.70	**3** (2)	0	**4** (2)	0	2 (2)	0	0	0	0
	SF	VLA	320	39.3 ± 12.10	**17** (17)	**29.4** (5)	**19** (19)	0	0	0	0	0	0
	PS	Las Coloradas	400	1 ± 0.05	0	0	0	0	0	2 (1)	0	0	0
	PS	Chos Malal	400	21 ± 5.00	**3** (3)	0	0	0	0	14 (13)	9 (8)	1 (1)	0
	HA	Pje. Cochico	400	1 ± 0.05	0	0	0	0	0	0	0	1 (1)	0
Autumn 2002	SF	Pje. El Contra	400	8.5 ± 2.50	**2** (2)	0	**4** (4)	0	3 (3)	0	0	0	0
	PS	Las Coloradas	400	18 ± 4.90	**4** (4)	0	0	0	0	3 (2)	4 (4)	7 (6)	0
	PS	Chos Malal	400	43.5 ± 6.30	**3** (2)	0	0	0	0	8 (6)	43 (42)	1 (0)	0
	HA	Pje. Cochico	400	9.6 ± 0.09	0	0	0	0	0	0	2 (2)	5 (5)	5 (5)
	MS	Pje. Challacó	576	0	0	0	0	0	0	0	0	0	0
Winter 2002	SF	Pje. El Contra	400	2 ± 0.02	**1** (1)	0	**1** (1)	0	1 (1)	0	0	0	0
	SF	VLA	320	13 ± 2.26	**4** (4)	0	**3** (3)	0	0	0	0	0	0
	PS	Las Coloradas	400	1.4 ± 0.07	0	0	0	0	0	3 (3)	0	0	0
	PS	Chos Malal	400	6.52 ± 1.72	0	0	0	0	0	3 (3)	8 (8)	2 (2)	0
	HA	Pje. Cochico	400	12.4± 0.37	0	0	0	0	0	0	6 (6)	3 (3)	8 (8)
TOTAL			**8016**		**68** (64)	**9.3**	**68** (63)	**4.8**	**10** (10)	**37** (32)	**78** (76)	**23** (21)	**13** (13)

Table 3. Prevalence of ANDV infection among rodents captured in peridomestic settings of three different ecosystems during winter 2001 to winter 2002.
Ol: *Oligoryzomys longicaudatus*, ***Al***: *Abrothrix longipilis*, **other spp**.: Total of other species captured. **SF**: Subantarctic forest, **PS**: Patagonian steppe, **MS**: Monte shrublands, **Ab prev**: Antibody prevalence.

Trapping event	Ecosystem	Location	# Trap nights	# *Ol* (# tested)	*Ol* Ab prev (%) (n)	# *Al* (# tested)	*Al* Ab prev (%)	# Other spp. (# tested)
Winter 2001	PS	Pje. San Cabao	**492**	**4** (4)	**25.0** (1)	0	0	**5** (5)
Spring 2001	SF	Pje. El Contra	**112**	**15** (15)	**13.3** (2)	**3** (3)	0	**2** (2)
	PS	Las Coloradas	**28**	0	0	0	0	1(0)
Summer 2001-2002	SF	Pje. El Contra	**40**	0	0	0	0	**1** (1)
	SF	Va. Pehuenia	**40**	**2** (2)	0	**1** (1)	0	0
	PS	Las Coloradas	**40**	0	0	0	0	**14** (7)
Autumn 2002	SF	Pje. El Contra	**80**	0	0	0	0	**1** (1)
	SF	Va. Pehuenia	**200**	**2** (2)	0	0	0	0
	MS	Pje. Sauzal Bonito	**52**	**2** (2)	0	0	0	**16** (15)
Winter 2002	SF	Pje. El Contra	**40**	0	0	0	0	0
	PS	Pje. San Cabao	**498**	**3** (3)	0	0	0	**1** (1)
TOTAL			**1616**	**28** (28)	**10.7**	**4** (4)	**0**	**41** (32)

and comprised 79.3% of the individuals captured (Table 3).

Hantavirus antibody prevalence

Subantarctic forest was the only ecosystem that harbored seropositive rodents in sylvan areas, and seropositive animals were captured only during spring in Paraje El Contra and summer in VLA (Table 2). In Paraje El Contra, hantavirus antibodies appeared in *O. longicaudatus* (n=1) and *A. longipilis* (n=3), with a higher seroprevalence in the second species (Table 2), whereas in VLA all the ANDV positives rodents trapped were *O. longicaudatus*. None of the seropositive rodents were recaptured in subsequent trapping events. At Paraje El Contra, a seropositive *O. longicaudatus* was captured inside a rural house during spring. In this area, two HPS cases had occurred during winter 2001. Although no seropositive rodents were captured in sylvan settings in the Patagonian steppe, one seropositive *O. longicaudatus* was trapped in the Paraje San Cabao peridomestic setting.

The seropositive *O. longicaudatus* in VLA were all adult males (scrotals) and three of five had obvious wounds (lack of an eye or part of the tail). At Paraje El Contra, two seropositive *A. longipilis* (n=3) were infected females. None of the seropositive *O. longicaudatus* had any distinct morphological characteristics or wounds.

DISCUSSION

This study provides the first data on spatial distribution and temporal variation in abundance of the sigmodontine assemblage and its relation to ANDV antibody prevalence. No previous data on the overall provincial distribution of the most important ANDV host, *O. longicaudatus*, exists for Neuquén province. Our study suggests that this species is the sigmodontine rodent with the widest distribution in Neuquén, as it was captured in every ecosystem we studied except for the High Andean steppe. Our data do not imply that this rodent is absent in this ecosystem. It was also the most abundant species captured, representing 26% of all individuals captured (in sylvan and peridomestic settings). *O. longicaudatus* and *A. longipilis* were the predominant species in forests trapped during every sampling period (Tables 2 and 3). The presence of *O. longicaudatus* throughout most of Neuquén province may represent evidence of its ability to survive in diverse types of habitats. In the Monte shrublands ecosystem, the presence of *O. longicaudatus* was verified at Paraje Sauzal Bonito, although most of the rodents captured there were domestic species (*M. musculus*). Paraje Sauzal Bonito is a more mesic area within the arid shrublands ecosystem, and as *O. longicaudatus* was not captured in the sylvan grid of the shrublands, we cannot confirm its presence throughout this habitat type. Our lack of captures in this habitat may have been due to an insufficient trapping effort required to capture rodents where densities were very low.

Some authors have estimated rodent densities in subantarctic forest of between 10-114 ind/ha under normal environmental conditions (Jaksic 1997), but there is no quantitative definition of what constitutes an outbreak. However, more than 100 ind/ha for total small mammals or more than 50 ind/ha for a single small mammal species has been suggested as an outbreak threshold (Jaksic and Lima 2003). In Las Chinchillas National Reserve, Chile, 404 ind/ha were recorded in the summer of 1988 (Jiménez et al. 1992), and 422 ± 33 ind/ha were recorded in April 1994 at San Martín Experimental Preserve, Chile (González et al. 2000). These are two examples of the many outbreaks or “ratadas” in South America but there are very few of them described for Argentina (Jaksic and Lima 2003). Although the maximum density at Paraje El Contra during spring 2001 is below that threshold, several factors indicate that our study began at the end of an outbreak. These factors include the subsequent “crash” in rodent densities in this region, the large number of rodents sighted daily by the sides of the roads, paths, and houses during the trapping period, and dead rodents on the shores of lakes in Argentina (Huechulafquen Lake:11,033 individuals counted in 72 days [O. Robotti, pers. comm.] and Lácar Lake: 6,776 individual in 32 days [San Martín de los Andes Municipality, pers. comm.]), as reported during Chilean outbreaks (Jaksic and Lima 2003, Gallardo and Mercado 1999, Murúa et al. 1996). This population probably peaked in the middle of the winter of 2001 before our trapping began. The proposed outbreak is thought to be due to the effect of the enormous bamboo seed set that was available during spring 2000 after the flowering/mast seeding of the bamboo.

The presence of ANDV antibodies was verified only in the forest and coincided with the period of highest reservoir density in each site. Therefore, the probability of humans contracting HPS may be greater in forest habitats, which is actually where most cases have occurred. In spite of *O. longicaudatus* being the main reservoir for ANDV, in the Paraje El Contra we observed that *A. longipilis* showed the highest antibody prevalence (8.8% vs. 3.4%) during spring 2001 (outbreak ending). After September 2001, no seropositive *A. longipilis* were captured. The disappearance of antibodies in *A. longipilis* suggests that the high densities of rodents during the outbreak produced a “spillover” effect of the virus from its main reservoir, *O. longicaudatus*, to other species. Mills et al. (1997) suggested that high rodent densities and/or increased interspecific interactions may provide opportunities for spillover to infect other species of rodents. We also found hantavirus antibody in blood samples from a male *A. olivaceus* in Paraje Piedra Mala (Subantarctic forest, north shore of Paimún Lake, at aprox. 20 km northwest from Paraje El Contra, 39º 43´ 17.6´´ S and 71º 32´ 7.3´´ W) during an associated study carried out during the spring of 2001 (Piudo et al. unpublished data). This is the first seropositive *A. olivaceus* confirmed in Neuquén province. This may be another similar spillover event (Paraje Piedra Mala was also suffering the outbreak ending) as described in Toro et al. (1998). Alternatively, *A. olivaceus* and *A. longipilis* may act as carriers and/or potential reservoirs of ANDV in Patagonia. However, more viral genetic studies are needed to elucidate these alternatives.

No seropositive animals were captured at the steppe sites, as occurred in studies carried out in the same habitat in Rio Negro province (Cantoni et al. 2001). However, the infected *O. longicaudatus* captured in the vicinity of Junín de los Andes

(Paraje San Cabao) represents the most eastern seropositive rodent in Neuquén to date. It is notable that this area is where cases of HPS occurred between May 2001 and March 2002. No HPS cases occurred in other areas sampled within the same ecosystem.

The presence of *O. longicaudatus* in most of our sample sites, including areas where it was not previously found or even considered in Neuquén province should be taken into consideration when designing preventative strategies against HPS. The number, geographical distribution, and abundance of known and potential reservoir species presented here was previously almost unknown in the province. The distribution of reservoir species may indicate the maximum potential extent of HPS (Mills and Childs 1998), indicating the potential for cases to occur in areas other than the forest if rodent populations increase. As a result of these data new prospected studies were started by the Public Health Service to monitor possible ANDV advancing fronts within the Monte shrublands ecoregion near Neuquén city (38º 57´ S and 68º 03´ W), the largest one in Patagonia. However, more in-depth peridomestic and sylvan studies of population dynamics of ANDV reservoir species in Neuquén are required to understand the ecological behavior of the rodent-hantavirus system. A thorough understanding of the ANDV system is necessary to develop effective measures to reduce human exposure.

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