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HOST-FEEDING PATTERNS OF ARGENTINE MOSQUITOES (DIPTERA: CULICIDAE) COLLECTED DURING AND AFTER AN EPIZOOTIC OF WESTERN EQUINE ENCEPHALITIS

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Abstract. Blood meals from 277 engorged mosquitoes collected in CDC light traps during December 1982 in Santa Fe Province, Argentina, during a western equine encephalitis (WEE) epizootic were identified by precipitin test. Blood meals from 348 engorged mosquitoes collected in the same general area during February 1984 were also identified, along with blood meals from 18 engorged mosquitoes from Rio Negro Province and 58 from Chubut Province. The total of 701 mosquitoes was composed of 297 *Aedes albifasciatus*, 16 *Ae. scapularis*, 1 *Anopheles albitarsis*, 4 *Culex* (*Melanoconion*) *delpontei*, 102 *Culex* (*Culex*) spp., 99 *Mansonia titillans*, 15 *Mansonia* spp., 3 *Psorophora albigena*, 6 *Ps. ciliata*, 4 *Ps. confinnis*, 50 *Ps. cyanescens*, 1 *Ps. dimidiata*, 3 *Ps. discrucians*, 16 *Ps. pallescens*, and 84 *Ps. varinervis*. Most of the mosquitoes (679 specimens) had fed on mammals, especially cattle; a few (19) had fed on birds. One *Cx.* (*Mel.*) *delpontei* had fed on an amphibian, and 2 mosquitoes contained a mixture of avian and mammalian bloods. In addition, the species composition of 20,697 mosquitoes collected in chicken-baited traps and 2,752 collected in a horse-baited trap is discussed, along with data on engorgement rates following entry of mosquitoes into the bait traps. A few mosquitoes also were collected in Nutria-baited traps (6 specimens) and in traps baited with domestic rabbits (143 specimens).

Outbreaks of equine encephalitis have occurred at irregular intervals in Argentina since the early part of the century (Sabattini et al. 1985). Since 1977 we have conducted field studies in Argentina with the objective of identifying the components of maintenance and epizootic cycles of encephalitis viruses. Initial studies were conducted during an interepizootic period (Calisher et al. 1985; Mitchell et al. 1985b; Monath et al. 1985; Sabattini et al. 1985). A widespread epizootic of western equine encephalitis (WEE) occurred in several provinces of Argentina during 1982–1983 (Mitchell et al. 1987). The outbreak began in Santa Fe Province in late 1982 and reached Rio Negro Province in the south by March 1983. Mosquito collections were made in Santa Fe and Rio Negro provinces during the epizootic and in these provinces and Chubut Province during the following virus transmission season (Fig. 1). Since the vector(s) of WEE virus in Argentina are unknown, it was important to study host-feeding patterns of mosquito species collected during and after the epizootic. Blood meals from engorged mosquitoes collected in CDC light traps were identified by precipitin test, and data on relative abundance and engorgement rates of mosquitoes from bait collections were analyzed. Prior to our investigations (Mitchell et al. 1985a,b, and present study), few studies had been conducted on the feeding habits of Argentine mosquitoes (Davis & Shannon 1928; Mayer et al. 1961), and these dealt only with *Anopheles* and 1 species of *Haemagogus*.

MATERIALS AND METHODS

Collecting and processing specimens. Engorged mosquitoes saved for blood-meal identification were collected in CDC light traps supplemented with ca. 1 kg of dry ice per trap (Sudia & Chamberlain 1962; Newhouse et al. 1966). In addition, mosquitoes were collected in lard-can traps (Bellamy & Reeves 1952) baited with chicken, rabbit, or Nutria, and in a net trap baited with a horse (Mitchell et al. 1985a). Following collection, arthropods were anesthetized with CO_2, placed in glass tubes with rubber stoppers, and transported on dry ice to the CDC laboratory in Fort Collins, Colorado.

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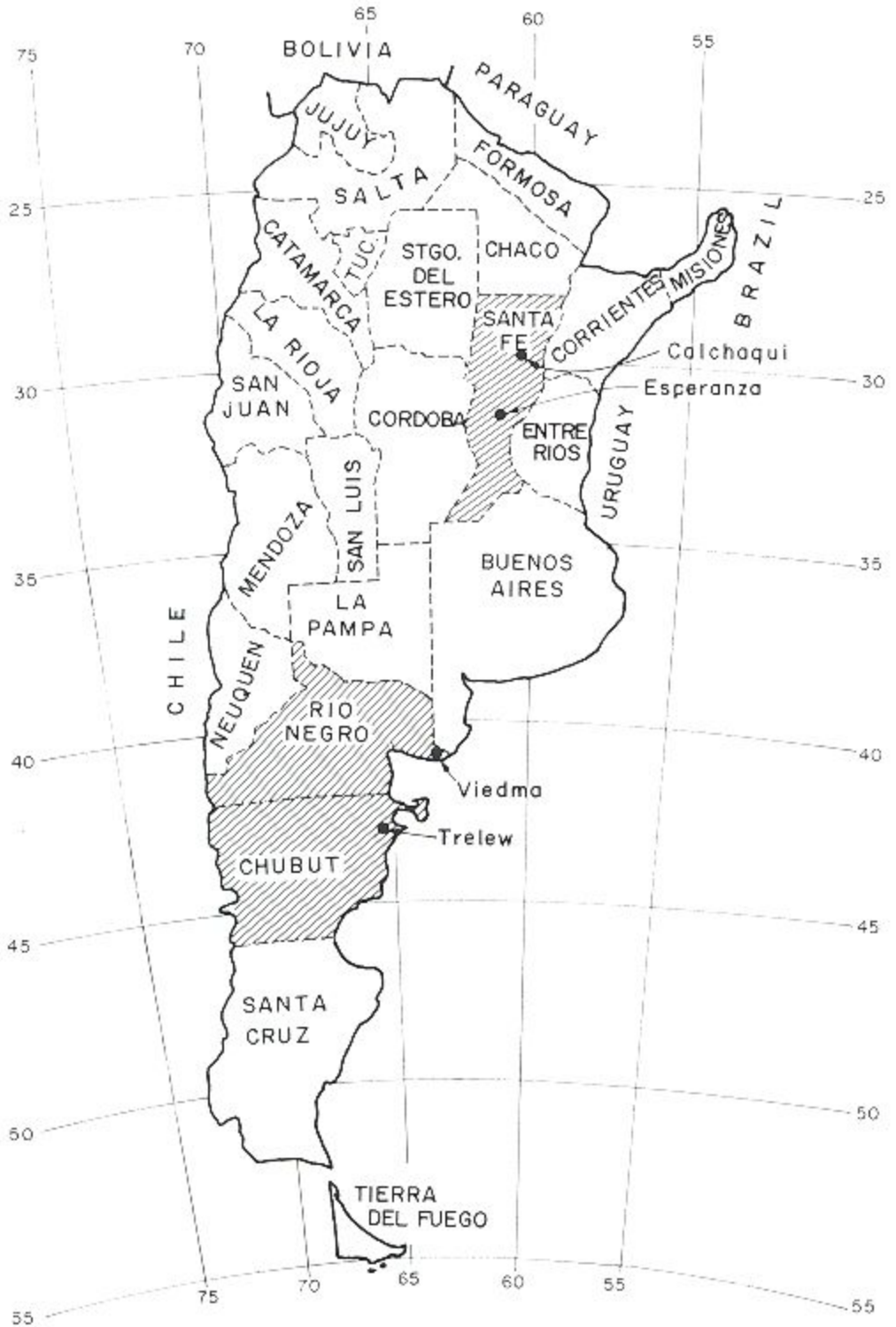

FIG. 1. Provinces of Argentina and principal areas where mosquito collections were made during 1982–1984.

TABLE 1. Identification of blood meals from engorged mosquitoes collected in Santa Fe Province, Argentina, during a western equine encephalitis outbreak, December 1982.

		Aves					Mammalia											
	No. tested	Amphibian	Avian	Anatidae	Phasianidae	Strigidae	Mammalian	Didelphidae	Leporidae	Felidae	Canidae	Perissodactyla	Artiodactyla	Bovidae	Suidae	Leporidae & Bovidae	Canidae & Perissodactyla	Bovidae & Perissodactyla
Ae. albifasciatus	107						2					15	1	88				1
Ae. scapularis	10				1							2		6	1			
An. albitarsis	1											1						
Cx. delpontei	4	1												3				
Cx. (Cux.) spp.	93		6	1	8	1			3	1	5	8		57	2	1		
Ma. spp.	15											2		12			1	
Ps. ciliata	2										1			1				
Ps. cyanescens	9						2					1	1	5				
Ps. dimidiata	1							1										
Ps. discrucians	3											1		2				
Ps. pallescens	5												1	4				
Ps. varinervis	27								2			3		21	1			
Total	277	1	6	1	9	1	4	1	5	1	6	33	3	199	4	1	1	1

Mosquitoes were sorted and pooled by species or genus in lots of 1 to 100. Abdomens were snipped from engorged mosquitoes as they were identified, placed in individual gelatin capsules, and shipped frozen to the Gorgas Memorial Laboratory, Panama, for blood-meal identification.

All antisera for blood-meal identification were produced in rabbits except anti-Leporidae, which was prepared in roosters. Class-, order-, and family-specific antisera were harvested after a series of injections into the axillary and inguinal lymph nodes of rabbits with equal quantities of animal plasma or sera and complete Freund's adjuvant. Titers ranged from 1:10,000 to 1:80,000; those that cross-reacted with sera diluted 1:1,000 from members of heterologous orders or families were absorbed overnight in a refrigerator with undiluted sera of these animals. For additional details, see Christensen & Vasquez (1981).

Engorged mosquito abdomens were placed in 12 × 75-mm test tubes containing 0.4 ml of phosphate-buffered saline 0.85% (w/v) and refrigerated overnight. The following day, the blood meals were expressed from the abdomens with applicator sticks and centrifuged at 500 × *g* for 30 min. Antigens in the supernatant were then screened by class-, order-, and family-specific antisera by a microcapillary precipitin method (Tempelis & Lofy 1963).

Study sites. The study sites in Santa Fe Province lie within the Espinal and Chaco zones of Cabrera & Willink (1973), and the sites in Rio Negro and Chubut provinces are in the Monte Zone. The Espinal and Chaco zones are characterized by xerophytic vegetation; these zones have been modified greatly by agriculture and cattle grazing. The Monte Zone in Rio Negro and Chubut provinces is characterized by an arid climate and desert-scrub vegetation. The sites where our collections were made in the Monte Zone have been greatly modified by the development of irrigated agriculture.

Mosquito collections made during a WEE epizootic focused on farms and ranches where clinical cases of equine encephalitis were occurring or had occurred recently. Several of the same sites in Santa Fe and Rio Negro provinces were revisited during the following spring and summer (1983–1984), and additional mosquito collections were made.

RESULTS

Blood meals were identified from 277 engorged mosquitoes collected in Santa Fe Province during December 1982 and from 348 spec-

TABLE 2. Identification of blood meals from engorged mosquitoes collected in Santa Fe Province, Argentina, February 1984.

		Aves	Mammalia									
	No. tested	Phasianidae	Mammalian	Leporidae	Rodentia	Cavidae	Canidae	Perissodactyla	Artiodactyla	Bovidae	Suidae	Hominidae
Ae. albifasciatus	123		2	2	1			20	3	95		
Ae. scapularis	6							2		4		
*Ma. titillans**	98	2	4	1		1		20	2	67		1
Ps. albigenu	3								1	2		
Ps. ciliata	4				1			1		2		
Ps. confunnis	4			1				1		2		
Ps. cyanescens	41		4	2			1	3	3	27		1
Ps. pallescens	11		2					1	1	7		
Ps. varinervis	57		10	1				7	1	37	1	
Total	347	2	22	7	2	1	1	55	11	243	1	2

* In addition, 1 *Ma. titillans* contained a mixed blood meal from a columbid and a bovid.

imens collected during February 1984 (Tables 1 and 2). During 1982 in Santa Fe Province, 93.5% of the mosquitoes (259 specimens) contained mammalian blood; the remainder had fed on birds (17 specimens) and an amphibian [1 specimen of *Cx. (Mel.) delpontei* Duret] (Table 1). A high percentage (76.8%) of the mammalian blood meals were from cattle. The same pattern was observed during February 1984 (Table 2). However, the ratio of Perissodactyla : Artiodactyla (including bovids) was remarkably similar during 1982 (1:6) and 1984 (1:5) and probably similar to the ratio of horses to cows in the areas where collections were made. Only 3 mosquitoes contained blood meals from birds, and 1 of these was a mixed meal from a bird and a bovine. Again, cattle were the principal source (70.5%) of blood meals from mammalian hosts. *Aedes albifasciatus* (Macquart) made up the largest percentage of engorged mosquitoes collected and tested during 1982 (38.6%) and 1984 (35.3%). Only engorged abdomens from mosquitoes identified to species were submitted for blood-meal identification from the 1984 Santa Fe Province collection. This accounts for the absence of *Culex (Cux.)* spp. and *Mansonia* spp. in Table 2.

Only 18 engorged mosquitoes from Rio Negro Province and 58 from Chubut Province were obtained for blood-meal identification (Table 3), and the Chubut collection was biased by placement of 1 trap directly over a pig sty. Again, *Ae. albifasciatus* was the most common species in these collections and had fed exclusively on mammals, with the exception of a mixed blood meal from a bird and a pig. Although the few *Culex (Cux.)* in these collections were not identifiable to species because of the lack of suitable keys to the females, they were separable into 2 groups, designated as species "A" and "B."

A total of 20,697 mosquitoes was collected in chicken-baited traps in Santa Fe Province during 1982–1984 (Table 4). Most of these (91.5%) were *Culex (Cux.)*. Relatively few *Anopheles* were collected (101 specimens), and only 1 of these fed on the bait. Engorgement rates were greater

TABLE 3. Identification of mammalian blood meals from engorged mosquitoes collected in Rio Negro and Chubut provinces, Argentina, March 1984.

	Leporidae	Canidae	Perissodactyla	Bovidae	Suidae	Hominidae
Rio Negro Province						
*Ae. albifasciatus**	1	1	1	10	3	
Cx. (Cux.) sp. B					1	
Chubut Province**						
Ae. albifasciatus	2				48	
Cx. (Cux.) sp. A			2			1
Cx. (Cux.) sp. B					5	

* In addition, in Rio Negro Province, 1 *Ae. albifasciatus* contained a mixed blood meal from a bird and a pig.

** One CDC light trap was placed directly over a pig sty.

Table 4. Numbers of mosquitoes collected in chicken-baited traps in Santa Fe Province, 1982–1984, and percentages that fed on the bait.

	Dec. 1982		Feb. 1983		Feb. 1984		Total	
	n	% engorged	*n*	% engorged	*n*	% engorged	*n*	% engorged
Ad. squamipennis	47	98	252	83	21	95	320	86
Ae. albifasciatus	62	81	87	11	29	48	178	42
Ae. scapularis	118	17	6	0	12	58	136	20
Ae. serratus	3	100					3	100
Ae. stigmaticus	2	100					2	100
Aedes spp.	8	100	10	30	13	77	31	68
An. albitarsis			12	0			12	0
Anopheles spp.			89	1			89	1
Cx. bidens					168	89	168	89
Cx. maxi					18	100	18	100
Cx. pipiens complex	2	100	2	100	122	77	126	78
Culex (*Car.*) spp.	6	67					6	67
Culex (*Cux.*) spp.	6,291	66	3,058	95	9,273	97	18,622	86
Culex (*Mel.*) spp.	21	95	1	0	60	50	82	61
Ma. humeralis	1	0					1	0
Ma. titillans			101	11	256	97	357	73
Mansonia spp.	12	100	80	56			92	62
Ps. albigenu			1	0	1	0	2	0
Ps. ciliata	3	67			38	92	41	90
Ps. confinnis					17	65	17	65
Ps. cyanescens	10	40	5	40	92	35	107	36
Ps. dimidiata					1	100	1	100
Ps. discrucians	15	93					15	93
Ps. pallescens	1	100	1	0	30	73	32	72
Ps. paulli	3	100					3	100
Ps. varinervis	11	82	2	50	44	88	57	86
Psorophora (*Jan.*) spp.	14	79			110	76	124	77
Psorophora spp.			29	59	26	81	55	69
Total & % engorged	6,630	66	3,736	86	10,331	95	20,697	84

than 50% in all species, or groups, represented by reasonable sample sizes, with the exceptions of *Ae. albifasciatus* (42% engorged), *Ae. scapularis* (Rondoni) (20%), and *Ps. cyanescens* (Coquillett) (36%).

We compared data from the 5 horse-bait collections with those from the 39 chicken-bait collections made in the same general area during 21–29 February 1984 (Table 5). This information gives a rough comparison of the attractiveness of these 2 animals to different mosquitoes. The data from the horse-bait collections were published previously in a different format as part of an evaluation of the trap (Mitchell et al. 1985a). As indicated above, the chicken-bait collections were principally *Culex* (*Cux.*), in this case 92.7% of the total collection. In contrast, the horse-bait collections contained significant percentages of *Ae. albifasciatus* (21.7%) and *Psorophora* (27.5%), as well as *Culex* (*Cux.*) (48.8%). In both types of bait collections, engorgement rates were generally high, except for *Ae. albifasciatus* (48% engorged) and *Ps. cyanescens* (35%) in the chicken-bait collections and *Culex* (*Mel.*) (none of 29) in the horse-bait collections. No *Anopheles* was collected in the chicken-bait traps during February 1984; 34 specimens were collected in the horse-bait trap, and all fed on the bait animals.

Lard-can traps baited with Nutrias yielded 6 unengorged *Cx. pipiens* complex mosquitoes in 2 trap-nights in Santa Fe Province during February 1984. At another site a few miles away and on a different night, 3 lard-can traps baited with domestic rabbits yielded 143 mosquitoes, of which 67% engorged on the bait. All of the *Psorophora* collected [3 *Ps. cyanescens* and 1 each of *Ps. pallescens* Edwards, *Ps. ciliata* (Fabricius), *Ps. confinnis* (Lynch Arribalzaga), and *Ps.* (*Jan.*) sp.] fed on the rabbits, as did 65% of the re-

TABLE 5. Relative abundance and comparative engorgement rates of mosquitoes collected in chicken- and horse-baited* traps in Santa Fe Province, Argentina, 21–29 February 1984.

	Relative abundance				Engorgement rates			
	Chicken bait		Horse bait		Chicken bait		Horse bait	
	No. coll.	% of total	No. coll.	% of total	No. engorged	% engorged	No. engorged	% engorged
Ad. squamipennis	21	0.2	1	—**	20	95	1	100
Ae. albifasciatus	29	0.3	596	21.7	14	48	563	94
Ae. scapularis	12	0.1	21	0.8	7	58	21	100
Ae. stigmaticus			2	0.1			2	100
Aedes spp.	13	0.1	22	0.8	10	77	21	95
An. albitarsis			18	0.7			18	100
Anopheles spp.			16	0.6			16	100
Cx. apicinus			3	0.1			3	100
Cx. bidens	168	1.6	205	7.4	150	89	190	93
Cx. maxi	18	0.2			18	100		
Cx. pipiens complex	122	1.2	19	0.7	94	77	14	74
Culex (*Cux.*) spp.	9,273	89.8	1,033	37.5	8,965	97	966	94
Culex (*Mel.*) spp.	60	0.6	29	1.1	30	50	0	0
Ma. titillans	256	2.5	29	1.1	249	97	29	100
Mansonia spp.			2	0.1			2	100
Ps. albigenu	1	—			0	0		
Ps. ciliata	38	0.4	284	10.3	35	92	284	100
Ps. confinnis	17	0.2	23	0.8	11	65	23	100
Ps. cyanescens	92	0.9	94	3.4	32	35	93	99
Ps. dimidiata	1	—	1	—	1	100	1	100
Ps. ferox			1	—			1	100
Ps. pallescens	30	0.3	198	7.2	22	73	198	100
Ps. paulli			1	—			1	100
Ps. varinervis	44	0.3	90	3.3	39	88	90	100
Psorophora (*Jan.*) spp.	110	1.1			84	76		
Psorophora spp.	26	0.3	64	2.3	21	81	62	97
Total	10,331	100.2	2,752	100.0	9,802	95	2,599	94

* Data for horse-bait collections from Mitchell et al. 1985a.

** Less than 0.1% of total.

mainder of the collection composed entirely of *Culex* (*Cux.*).

DISCUSSION

The preponderance of mosquito blood meals from large mammals, especially bovines, in Santa Fe Province is probably a reflection, in part, of host abundance at the sites sampled. Nonetheless, domestic and wild birds were present, and often abundant, at most of the sites. It is somewhat anomalous that engorged *Culex* (*Cux.*) collected in CDC light traps in Santa Fe Province during December 1982 had fed predominantly on large mammals (Table 1), whereas *Culex* (*Cux.*) spp. were the most abundant mosquitoes in chicken-bait collections made simultaneously at the same sites (Table 4). It is apparent from the data that a wide variety of mosquito species are attracted to chickens and will feed on them, at least within the confines of the traps (Table 4).

Confining mosquitoes with the bait animal after they have entered the trap would be expected to increase the chances of engorgement. Therefore, low engorgement rates among *Anopheles* mosquitoes and *Ps. cyanescens* that entered chicken-baited traps (Table 4) probably indicate a reluctance of these mosquitoes to feed on chickens. The same rationale can be applied to the failure of 29 *Culex* (*Mel.*) to feed on equine bait following entry into the horse-baited trap (Table 5).

Perhaps the most revealing comparisons can be made by examining the data in Table 5. Although not abundant in the collections, *Aedeomyia squamipennis* (Lynch Arribalzaga) was attracted to the chicken bait and readily fed following entry into the traps. The latter is also

true for the genera *Culex* and *Mansonia*. *Culex* mosquitoes also made up a significant component of the horse-bait collections, and engorgement rates were high. In contrast, the genera *Aedes* and *Psorophora* were abundant in the horse-bait collections but not in the chicken-bait collections. Finally, species of *Anopheles* were absent from the February 1984 chicken-bait collections and, although not abundant in the horse-bait collections (34 specimens), all specimens fed following entry into the trap. In general, these results confirm what is known about the feeding habits of these groups of mosquitoes in Argentina (Mitchell et al. 1985b) and elsewhere in the world (Tempelis 1975; Washino & Tempelis 1983).

The demonstration that *Ae. albifasciatus* from Santa Fe, Rio Negro, and Chubut provinces had fed on Leporidae may be significant. *Aedes albifasciatus* would be an excellent candidate for the vector component of an *Aedes*–Leporidae–WEE virus cycle similar to that described for *Ae. melanimon* Dyar in California (Hardy & Bruen 1974). The European Hare, *Lepus europaeus*, has been introduced into Argentina and is abundant in the area of Rio Negro Province, where WEE virus activity has been documented and where our collections were made. WEE virus was isolated from *Ae. albifasciatus*, *Ps. pallescens*, *An. albitarsis*, and *Mansonia* spp. collected in Santa Fe Province during December 1982 (Mitchell et al. 1987).

Three blood meals were determined to be of human origin (Tables 2, 3). In the case of *Ps. cyanescens* and *Ma. titillans* (Walker), it is quite possible that these mosquitoes fed on the collectors during the early evening hours as the light traps were being placed for the nightly collections. These species readily bite humans, and they were quite abundant at some of the collection sites in Santa Fe Province. The distinctive *Ae. albifasciatus* was an aggressive human biter during the daytime in almost all sites sampled in Santa Fe, Rio Negro, and Chubut provinces.

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