Scientific Note

New records of *Aedes aegypti* at the southern limit of its distribution in Buenos Aires province, Argentina

Gabriela Zanotti[1], María Sol De Majo[1], Iris Alem[1], Nicolás Schweigmann[1], Raúl E. Campos[2], and Sylvia Fischer[1]✉

[1]*Departamento de Ecología, Genética y Evolución, and IEGEBA (UBA-CONICET), Facultad de Ciencias Exactas y Naturales, Universidad de Buenos Aires. Ciudad Universitaria, Buenos Aires, Argentina, sylvia@ege.fcen.uba.ar*
[2]*Instituto de Limnología "Dr. Raúl A. Ringuelet", Universidad Nacional de La Plata – CONICET, CCT La Plata, La Plata, Buenos Aires, Argentina*

Aedes (=Stegomyia) aegypti is the main vector of dengue in the Americas, and it is also a vector of urban yellow fever. This mosquito species has a global distribution, ranging from tropical and subtropical through temperate regions. Its continuously expanding distribution may be a critical factor in the constant increase in the incidence and geographic range of dengue (Gubler 2004). After the continental-wide control program during the twentieth century, *Ae. aegypti* was eradicated from Argentina and its neighboring countries in 1965 (Soper 1967). However, this species was detected again in the Misiones and Formosa provinces of Argentina in 1986 (OMS 1990). In 1991, *Ae. aegypti* reached the temperate region of this country, where it was detected in the locality of Quilmes, Buenos Aires province (Campos 1993), and in 1995 it reached various neighborhoods of Buenos Aires city for the first time (Junín et al. 1995). *Ae. aegypti* later showed a further expansion to localities within and near the metropolitan area of Buenos Aires (Curto et al. 2002). In recent years, this species has continued to increase its geographic limit at the cool margins of its range, where it has been detected in various cities of La Pampa province (Rossi et al. 2006, Diez et al. 2014), in the capital of Neuquén province (Grech et al. 2013), and in Dolores city in Buenos Aires province (Díaz-Nieto et al. 2013).

Temperature is a major environmental factor that influences the colonization and annual persistence of *Ae. aegypti* populations. The geographic distribution of *Ae. aegypti* is considered to fall roughly within the areas where the average winter isotherm is above 10° C (Christophers 1960). However, in Argentina, the mean annual temperature limit of 15° C has been suggested to be a better predictor of the distribution limits for this species (Otero et al. 2006). At the cooler margins of its geographic range, two different situations can be differentiated: localities where overwintering eggs can hatch and produce viable larvae in the spring, and others where overwintering eggs are unlikely to both hatch and produce viable larvae. In the latter case, the low temperatures (below 15.5° C mean annual temperature) could avoid the permanent colonization of this species (Eisen et al. 2014).

The aim of this study was to update the southern distribution of *Ae. aegypti* in Buenos Aires province in relation to a mean annual temperature gradient. Between 4 February and 14 April 2014, a total of 27 cities located in Buenos Aires province (Figure 1) with more than 10,000 inhabitants were selected for sampling. The selected sites have a mean annual temperature gradient from 16.1° C to 13.4° C. In each city, used tires stored outdoors within the urban limits were inspected. Up to a total of three or four tire deposits were examined in each city, providing that no larvae were found at the first deposit (in some of the smallest cities all available deposits were sampled). The maximum possible number of tires was inspected in each deposit, depending on availability and accessibility. Samples of 3rd and 4th instar larvae were collected from a representative number of tires (Table 1), and fixed in situ with 80% alcohol. Also, pupae were collected and individually placed in 3-ml glass tubes with 0.5 ml of water and reared until adults emerged. Samples were transported to the laboratory, larvae and adults were observed under a stereoscopic microscope, and specimens were identified to species level. The number of tires with mosquitoes was recorded and considered as evidence of favorable conditions for mosquito development (i.e., enough water permanence to ensure habitat colonization). All immature mosquitoes were identified as *Ae. aegypti* or *Culex* spp.

Immature or adult *Ae. aegypti* were collected in 13 of the 27 cities sampled (Table 1). The cities of General Alvear, Las Flores, Pehuajó, Roque Pérez, Saladillo, San Bernardo, San Carlos de Bolívar, San Clemente, Santa Teresita, and Villa Gesell constitute new records of *Ae. aegypti*. All cities with mean annual temperatures above 14.5° C (except for General Madariaga and Daireaux) were positive for this species, whereas in cities with lower temperatures the species was not detected (Table 1).

In Villa Gesell city, *Ae. aegypti* was not found in the tire deposits inspected, but larvae were collected from small earthen pots in a residential garden. The presence of this species in Villa Gesell represents the most southern record for this species in Buenos Aires province and also the record at the lowest mean annual temperatures (14.5° C) within our study.

Our results show the presence of this species below the mean annual temperature limit of 15° C suggested by Otero et al. (2006), both in Dolores and in Villa Gesell. The mean annual temperatures in these cities are within the range where supposedly no successful overwintering for *Ae. aegypti* occurs (Eisen et al. 2014). Although it is not possible to infer from this study whether this species is able to persist through the unfavorable season and establish permanent populations in these cities, the fact that the presence of this species had already been reported in 2012, both in cemetery vases and

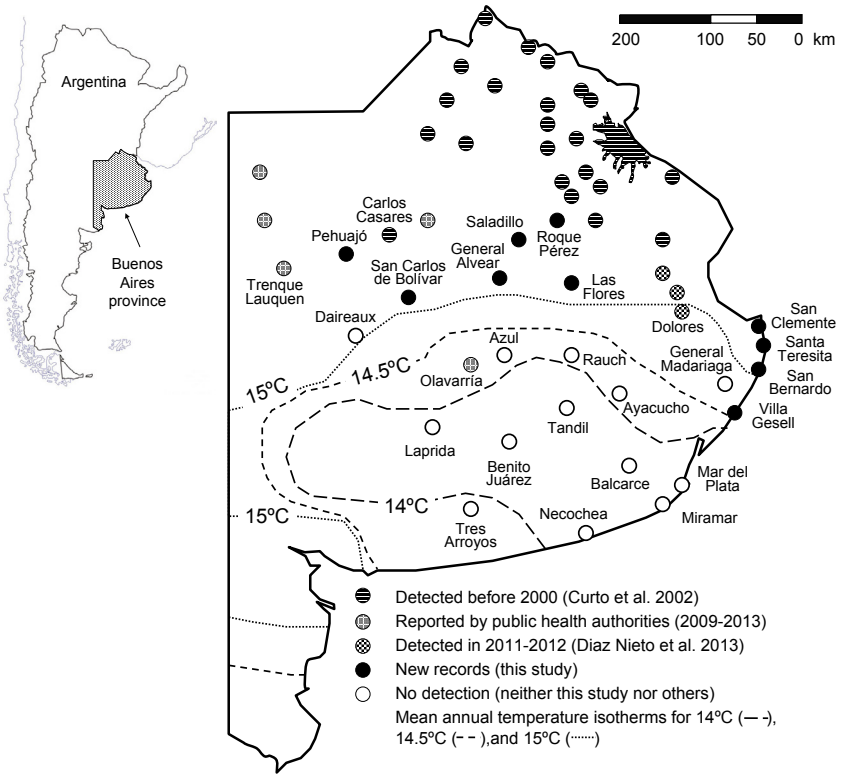

Figure 1. Location of Buenos Aires province in Argentina (small left). Previous and new records for *Ae. aegypti* in Buenos Aires province, and mean annual temperature isotherms for 14°, 14.5°, and 15° C according to data from climate-data.org. Cities sampled during the present study are identified with names (large center).

in tires in Dolores city (Díaz-Nieto et al. 2013), may indicate that overwintering occurs and a permanent population has successfully established. Another interesting fact is that, according to the owner of the tire deposit in Dolores city where *Ae. aegypti* adults were collected, used tires are sold to other cities located south towards the coast, thus facilitating the introduction of eggs within the tires to other nearby cities. Previous studies have reported the contribution of the trade of used tires to the dispersal of several mosquito species, including *Ae. aegypti* (Reiter and Sprenger 1987, Jupp and Kemp 1992, Becker et al. 2012), but the successful establishment of permanent populations may be restricted to those with temperature conditions above a critical threshold.

It is interesting that the identification of *Ae. aegypti* in our surveys roughly match the large-scale predictive maps of oviposition suitability based on temperature constraints developed by Brady et al. (2014). The only city where *Ae. aegypti* has been previously detected in Buenos Aires province that is not predicted by these maps is Olavarría, where the mean annual temperature is 14.15° C. Although we were not able to find this species in Olavarría during our survey, municipal public health authorities reported the presence of *Ae. aegypti* in 2009, 2013, and 2014, suggesting the possibility of a permanent population at still lower temperatures than those predicted by previous studies.

Acknowledgments

We thank María Victoria Gonzalez Eusevi for her English review. This study was supported by grant PICT 1254-2012 (FONCyT). We are grateful to three anonymous reviewers for their comments on a previous version of the manuscript.

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Table 1. Mean annual temperature, location, and results for the cities sampled. Temperature data were obtained from climate-data.org.

City	Latitude (*)	Longitude (*)	Mean annual temperature	Number of sites inspected	Number of tires sampled	Number of tires with mosquitoes	Number of tires with *Ae. aegypti*	Number of samples collected	Total larvae collected (*Ae. aegypti* larvae)	Comments (citations of previous records are given)
Trenque Lauquen	35°57'50.29"S	62°44'7.98"W	16.11	2	14	6	2	6	35 (7)	provincial Public Health ministry 2014
Roque Perez	35°23'44.05"S	59°19'58.11"W	15.98	2	35	23	10	17	264 (55)	first record
Saladillo	35°38'38.90"S	59°47'12.90"W	15.71	2	24	21	13	12	305 (282)	first record
Pehuajó	35°49'18.78"S	61°53'44.09"W	15.59	1	6	3	2	2	28 (14)	first record
Carlos Casares	35°37'59.24"S	61°21'56.16"W	15.53	2	24	9	3	3	24 (12)	Curto et al. 2002
Las Flores	36° 1'33.20"S	59° 4'55.00"W	15.37	2	38	19	7	14	99 (33)	first record
San Clemente	36°21'30.80"S	56°43'46.20"W	15.36	1	4	2	2	1	9 (9)	first record
General Alvear	36° 1'15.30"S	60° 1'18.50"W	15.27	2	33	19	11	11	239 (50)	first record
Santa Teresita	36°32'9.20"S	56°42'11.20"W	15.23	2	10	5	2	4	43 (9)	first record
San Carlos de Bolivar	36°13'33.20"S	61° 6'18.50"W	15.19	2	40	25	12	15	234 (101)	first record
Daireaux	36°36'3.47"S	61°44'55.12"W	15.09	3	27	10	0	7	107	-
San Bernardo	36°41'3.20"S	56°41'6.90"W	15.08	1	15	12	11	11	71 (52)	first record
Dolores	36°19'14.60"S	57°41'2.50"W	14.96	1	1	1	1 **	1	1 (1)	Díaz Nieto et al. 2013
General Madariaga	37° 6'59.85"S	57°11'29.58"W	14.72	2	63	43	0	9	63	-
Villa Gesell	37°15'18.77"S	56°58'19.45"W	14.53	2	73	18	0	7	54 (1) ***	first record
Tres Arroyos	38°22'34.72"S	60°16'41.26"W	14.31	3	65	12	0	7	49	-
Rauch	36°46'29.70"S	59° 5'23.23"W	14.27	2	83	43	0	14	133	-
Ayacucho	37° 9'0.97"S	58°28'58.70"W	14.25	3	17	7	0	7	92	-
Olavarría	36°53'31.52"S	60°18'53.78"W	14.15	4	88	14	0	10	95	Municipal authorities
Azul	36°46'29.88"S	59°51'14.53"W	14.09	4	52	16	0	8	105	-
Balcarce	37°50'47.06"S	58°15'19.64"W	13.95	4	99	29	0	17	172	-
Laprida	37°32'43.52"S	60°47'55.34"W	13.92	3	45	25	0	7	97	-
Necochea	38°33'16.18"S	58°44'22,59"W	13.59	2	65	50	0	7	56	-
Tandil	37°19'4.11"S	59°9'1.40"W	13.48	3	58	47	0	14	127	-
Benito Juárez	37°40'40.12"S	59°48'27.93"W	13.48	3	47	29	0	7	62	-
Mar del Plata	37°58'47.49"S	57°35'23.29"W	13.45	1	12	2	0	1	8	-
Miramar	38°15'55.36"S	57°51'4.53"W	13.43	1	21	14	0	3	22	-

* Latitude and longitude data correspond to city center (negative cities) or collection site (positive cities).

** In Dolores, tires were stored in inaccessible piles, but *Aedes aegypti* adults flying around were also collected.

*** In Villa Gesell larvae of *Aedes aegypti* were collected in a residential garden, but not in the tire dumps inspected.

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