

Geographical variation in sexual behavior and body traits in a sex role reversed wolf spider

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Abstract Mating partners need to recognize, assess each other, and exchange information through behavioral events that occur before, during, and after mating. Sexual signals, as well as life history traits, are influenced by selective pressures and environmental factors that can vary across distant geographical areas. *Allocosa senex* is a sand-dwelling wolf spider which constructs burrows along the sandy coasts of Argentina, Brazil, and Uruguay. Females are the mobile sex that searches for males and initiates courtship. They prefer males which construct longer burrows, and males prefer virgin females in good body condition. The objective of this study was to compare sexual behavior patterns, as well as body characteristics and burrow dimensions, between two geographically distant locations of *A. senex*, one in Uruguay (Uruguayan location) and the other from central Argentina (Argentinean location). We found differences in the number of male abdominal vibrations, male and female touches during mating, and number of erections of male leg spines, which all were higher in matings of Argentinean pairs. On the other hand, male body mass and female body condition were higher in Uruguayan individuals. The wide distribution of *A. senex* could be determining

variations in the biotic and abiotic features that affect the species, generating differences in the strength of selective forces acting on individuals from the two studied locations.

Keywords Intraspecific variation · Sexual selection · Sexual behaviors · Burrow size · Body traits · Lycosid

Introduction

Sexual behavioral patterns play roles in recognition between mates, especially during courtship, involving the exchange of signals in several sensory modalities such as vibratory, visual, acoustic, chemical, tactile, and the transmission of information about quality and species identity (Simmons 1988; Choe and Crespi 1997; Olivero et al. 2015). Courtship functions include the synchronization between the two sexes, orientation, persuasion, and assessment between the potential mating partners (Tinbergen 1953; Verrell and Mabry 2003). In general, mating behavioral events show similar “patterns” between individuals within the same species and, simultaneously, differ from the patterns shown by individuals from other species (Barlow 2002). In spite of this, we can expect certain levels of intraspecific variation in sexual behavior related to the occurrences, frequencies, and durations in which individuals perform the behavioral acts (Foster and Endler 1999). These variations may be associated—among other factors—with the wide geographic distributions that some species show that make them more likely to experience a diversity of environmental conditions and exhibit differences in their life history traits and behaviors in response to this variation (Foster and Endler 1999; Elias et al. 2011; Macedo and Machado 2013; Macías-Ordóñez et al. 2013). Even more, if some populations of the species are isolated either by geographical barriers or through colonization of new niches, this could lead them to

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allopatric or peripatric divergence, respectively. It would be expected that in those cases there could be heritable differences based on the variation in selection on each population, in agreement with the patterns shown by other species which live at the same habitat. Also, geographic wide distributions are good stages for the occurrence of behavioral plasticity, defined as ‘environmentally sensitive behaviors’ by Baldwin (1902) (cited in West-Eberhard 2003), that arise from adaptive behaviors to local environments. In this context, differences would not be heritable, but they could show similar patterns among related species under the same ecological conditions due to the fact that environment is determinant of the range of phenotypes exposed to selection (Alberch 1980; West-Eberhard 2003). Finally, differences observed in species with wide distribution could be explained by genetic drift and, in that case, variations would not be linked to environmental factors, and we would not expect consistent patterns within components of courtship of one species nor across species with similar ecologies (Lande 1976).

The Neotropics are a highly diverse and complex region in terms of climates, exhibiting a mosaic of environmental conditions which in turn result in complex selective pressures within taxa, producing diverse and unique reproductive behaviors and morphological traits (Brown 1991; Macedo and Machado 2013). Consequently, it is expected that a single species with wide distribution inhabiting this region can experience a wider set of environmental variables and a higher intraspecific variation in mating tactics than those species from the Paleotropics (Macías-Ordóñez et al. 2013). Geographical variations in sexual behavior have been reported in many taxa such as frogs (Ryan et al. 1990; Amezcuita et al. 2009), snakes (Gregory and Larsen 1993; Foster and Endler 1999), fish (Ishikawa et al. 2006), snails (Kuppfernagel and Baur 2011), insects (Briceño et al. 2007), scorpions (Olivero 2014), and spiders (Miller et al. 1998; Elias et al. 2006; González et al. 2013). Moreover, this possibility has also been discussed in humans (Laland and Brown 2002).

The Neotropical sand-dwelling wolf spider *Allocosa senex* (Mello-Leitão, 1945, synonym of *Allocosa brasiliensis* Petrunkevitch 1910 according to Simó et al. 2017) is a nocturnal species that constructs burrows along the coasts of rivers, lakes, and along the Atlantic Ocean at Argentina, Brazil, and Uruguay (Capocasale 1990). Individuals stay in their burrows during daylight and in winter, and they are most active during summer nights (Costa 1995). This spider shows a reversal in typical sex roles and sexual size dimorphism expected for spiders (Aisenberg et al. 2007; Aisenberg 2014). Contrary to the expectations in spiders, females of *A. senex* are smaller than males and they are the mobile sex that searches for males and initiates courtship. Mating takes place inside male burrows, and it includes approximately nine mounts and dismounts (Aisenberg et al. 2007). Both sexes are selective when they are taking mating decisions; females prefer males with longer burrows and males

prefer virgin females in good body condition (Aisenberg et al. 2011a). Furthermore, rejected females can be cannibalized by the males (Aisenberg et al. 2011a). When mating occurs and after the final mount, the male exits and blocks the burrow entrance before leaving, while the female releases silk and cooperates in closing the entrance from inside. The female will stay in the male burrow for oviposition and will finally emerge for spiderling dispersal (Postiglioni et al. 2008). Previous observations of the copulatory sequences in *A. senex* from a location in Argentina showed that females performed body shakings during mount and inter-mount periods, which could indicate the occurrence of copulatory communication between the sexes in this species (García Díaz et al. 2015). However, female body shakings during mating had not been described for *A. senex* Uruguayan location (Aisenberg et al. 2007). Also, casual observations of the authors indicated subtle sexual behavioral differences in matings of this species in Argentina and Uruguay.

Although *A. senex* is a well-known model system for studies on sexual selection in spiders (Aisenberg 2014), little is known about how the geographical distribution could vary their sexual repertoire. The objective of the present study was to compare courtship and copulatory behavioral patterns, as well as body characteristics and burrow dimensions, in two geographically distant locations of *A. senex*, one of the coast of the Río Anisacate and San Antonio (Mar Chiquita Basin, Córdoba Province, Argentina) and the other from the coast of the Río de la Plata (Canelones Province, Uruguay). Considering disparities in factors such as temperature between the two locations, we expected to find differences in the occurrences and/or durations of sexual behaviors in response to variations in availability of resources. Thus, we predicted that variations would be observed in both sexes due to the active participation of both females and males in sexual interactions compared to species which do not show reversed sex roles. Finally, we expected to observe larger and heavier individuals in the Uruguayan location due to the more stable temperatures to which they are exposed, similarly to what has been reported for a scorpion (Olivero 2012) and a spider species (González et al. 2013) with similar distributions. This study will provide information about intraspecific geographical variation of sexual and ecological traits on a sex-role reversed spider species, enlightening the discussion on the mechanisms shaping those atypical behaviors.

Materials and methods

Collecting and housing

We collected 66 sub-adults of *A. senex* at the coastal sand beaches of San José de Carrasco (−34° 50′ 34″ S, −55° 59′ 34″ W) and Marindia (−34° 46′ 52.3″ S, −55° 49′ 29.6″ W), Canelones, Uruguay (Uruguayan locations), and 91 sub-

adults at the coasts of river beaches of Copina ($-31^{\circ} 33' 0''$ S, $-64^{\circ} 42' 0''$ W), Cuesta Blanca ($-31^{\circ} 28' 56''$ S, $-64^{\circ} 34' 21''$ W), and Anisacate ($-31^{\circ} 43' 05''$ S, $-64^{\circ} 24' 01''$ W), Córdoba Province, Argentina (Argentinean locations) (Fig. 1), from November to March 2007–2008 and 2008–2009. Spiders were located during the night using headlamps to capture them while walking or leaning out from the burrow entrances.

Each spider was individually raised in plastic transparent boxes (length 9 cm, width 5 cm, height 2 cm), with a layer of approximately 2 cm of sand as substrate and a piece of cotton soaked in water. We monitored individuals daily and recorded molting occurrence in sub-adults to determine the exact date of reaching adulthood. We obtained virgin females and males by maintaining them isolated since they were sub-adults under laboratory conditions. The spiders were fed twice a week with *Tenebrio* sp. larvae (Coleoptera, Tenebrionidae) and juveniles of *Blaptica dubia* cockroaches (Blattaria, Blaberidae). In both locations, rearing conditions were similar due to breeding training which was provided by the same person, supplying same food quantity and maintaining similar conditions of temperature and light/dark cycles.

Characteristics of the selected locations

The Argentinean location is characterized by small, freshwater beaches, a substrate with higher percentages of thick sand (1000 μ m vs. Uruguayan beaches with higher percentages of 125 μ m), greater range of temperatures, lower total annual precipitation, and higher density of *A. senex* individuals compared to the Uruguayan location. Moreover, the Argentinean location belongs to the Dry Chaco ecoregion, while the Uruguayan location corresponds to the Uruguayan Savanna ecoregion (taken from Terrestrial ecoregions (2016) <http://www.worldwildlife.org/biome-categories/terrestrial-ecoregions>).

Experimental trials and analyses of behavioral sequences

We performed the trials corresponding to the Argentinean location at the Laboratorio de Biología Reproductiva y Evolución, UNC, Córdoba, Argentina, and those corresponding to the Uruguayan location of *A. senex* at the Laboratorio de Etología, Ecología y Evolución, IIBCE, Montevideo, Uruguay. The average laboratory temperatures during the experiments did not differ significantly between locations (mean \pm SD Argentina 23.00 ± 3.00 $^{\circ}$ C, range 20–26 $^{\circ}$ C; Uruguay 25.58 ± 1.62 $^{\circ}$ C, range 21–26 $^{\circ}$ C; $t = 3.33$, $p = 0.99$). All the trials began after dusk coinciding with the period of activity reported for the species (Costa 1995). We used transparent glass cages for the trials (length 30 cm, width 16 cm, height 40 cm) with a layer of 15 cm of sand as substrate and a piece of cotton soaked in water. We moistened the first 5-cm layer of sand placed at the base of the glass cage to

provide humidity and resemble natural conditions, following Aisenberg et al. (2007) and Garcia Diaz et al. (2015). We randomly assigned females and males to the experimental pairs. We measured carapace width, a measurement considered representative of body size in spiders (Marshall and Gittleman 1994; Eberhard et al. 1998), abdominal width, and weighed each individual immediately before the trials. The index abdominal width/carapace width was considered as representative of body condition, as described by Moya-Laraño et al. (2003) for *Lycosa tarantula* (Linnaeus, 1758).

Virgin females and males were used 7–20 days post adult-molt, coinciding with the sexual receptivity period reported for this species (Aisenberg et al. 2011a). We did not reuse individuals. Each male was placed in the glass cage 48 h before the trial to allow burrow construction, which is usually dug against the glass walls allowing the observation of behaviors that occur inside the burrows (Aisenberg et al. 2007). The trial began when we introduced the female to the experimental cage and finished after the male exited the burrow and covered the burrow entrance in the case of mating; when mating did not occur, trials finished after 30 min without courtship or after 1 h with female and/or male courtship. We recorded 15 courtship and copulatory sequences for each location. As *A. senex* has been recently categorized as priority species for conservation in Uruguay (Ghione et al. 2017), we used moderate samples sizes of $n = 15$ to avoid extracting too many specimens from nature.

We performed the trials in total darkness and recorded the sexual interactions with Sony DCR-SR85 and Sony Handycam DCR-SR65E digital video cameras, both of them equipped with night-shot function. We analyzed the video recordings with JWatcher software (Blumstein et al. 2000). We named courtship and copulatory behaviors in agreement with Aisenberg et al. (2007) and Garcia Diaz et al. (2015). A list with recorded courtship and copulatory behaviors with their corresponding brief descriptions is included in Table 1. We recorded the number of occurrences and durations of courtship and copulatory behaviors. The number of erections of male leg spines was recorded as a possible indirect quantification of hematochocha inflations and ejaculations as had been previously applied for this species (Aisenberg et al. 2007) and for other wolf spider species (Costa 1979; González et al. 2013). We deposited voucher specimens at the Laboratorio de Biología Reproductiva y Evolución, LBRE, Facultad de Ciencias Exactas, Físicas y Naturales, Universidad Nacional de Córdoba, Argentina, and at the arachnological collection of Sección Entomología, Facultad de Ciencias, Montevideo, Uruguay.

Statistics

We analyzed the results with NCSS software (2007) and RStudio (2016). We compared the number of occurrences of

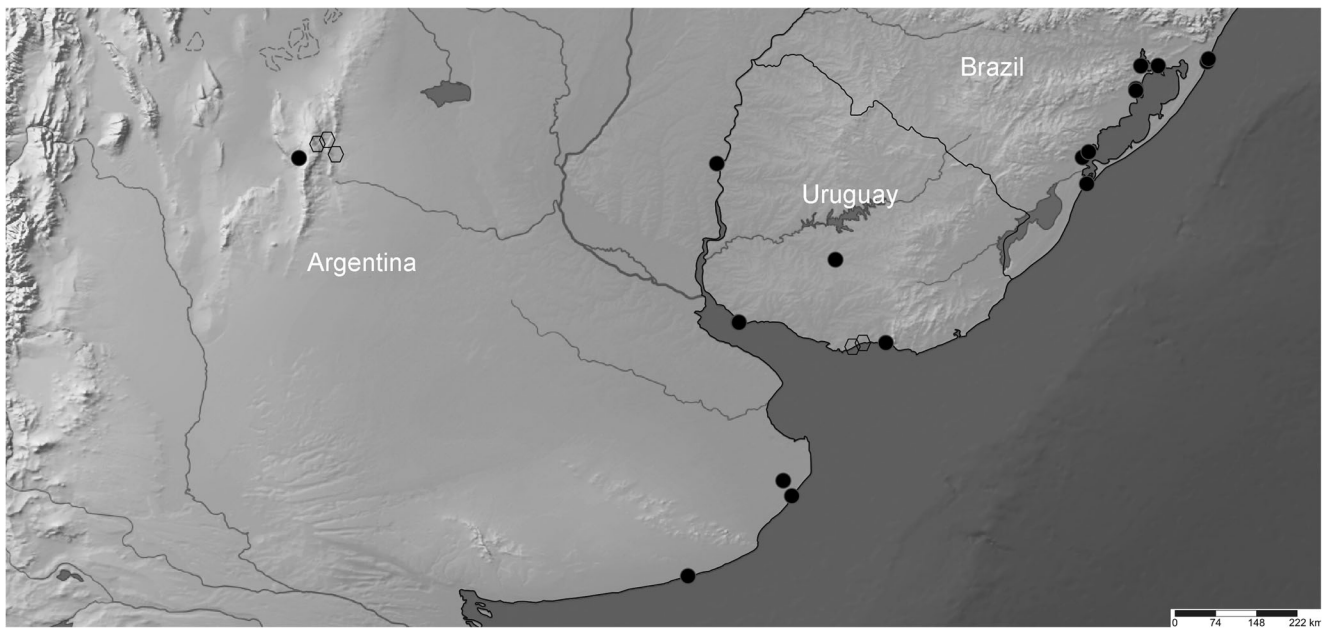


Fig. 1 *Allocosa senex* general distribution (solid black circles) and Uruguayan (Uruguay) and Argentinian (Argentina) locations used in the present study (open hexagons). The points corresponding to the

localities were downloaded from GBIF Secretariat: GBIF Backbone Taxonomy (<http://www.gbif.org/species/2145653>) and the map was drawn in <http://www.simplemappr.net> (Shorthouse 2010).

instantaneous sexual behaviors (male or female shakes, female waves, male taps, touches, male abdominal vibrations, palpal insertions, and erection of male leg spines) and durations of courtship, mating, palpal insertions, and mounts, between the two locations. If variables followed normal distribution and homogeneity of variances, we performed two-sample Student's *t* tests to compare the number of occurrences

and durations of behavioral units, body, and burrow dimensions, between the two groups. Those variables that did not adjust to conditions of normality or homogeneity were analyzed using the non-parametric Mann-Whitney *U* test. We performed an analysis of covariance (ANCOVA) with body mass as co-variable, sexual behaviors as dependent variables, and location as factor.

Table 1 Pre-copulatory and copulatory behaviors performed by *A. senex* with their corresponding descriptions

Behavioral event	Description
	Pre-copulatory phase
Female waves	The female performs bursts of tapping on the sand with forelegs and palps.
Male shakes	The male performs bursts of body shakes.
Male waves	The male performs bursts of tapping on the sand with forelegs and palps.
Female approaches	The female gets close to male.
Female touches	The female touches with legs 1 and 2, the corresponding forelegs or carapace of the male.
Male touches	The male touches with legs 1 and 2, the corresponding forelegs or carapace of the female.
	Copulatory phase
Mounts	The male mounts the female in the typical lycosid mating position, with the male on top of the female looking in opposite directions.
Male inserts	The male performs the palpal insertion with one palp at the time.
Male erections of leg spines	The male erects the spines of his legs. This behavior has been previously associated with hematodochal inflations and sperm transfer in this and other wolf spiders Aisenberg and Costa (2005), Aisenberg et al. (2007), and González et al. (2013).
Female shakes	The female performs bursts of body shaking.
Male vibrates	The male performs bursts of abdominal vibrations, moving his abdomen up and down.
Male taps	The male performs bursts of touches on the female carapace while inserting the palp.

Modified from Aisenberg et al. (2007) and Garcia Diaz et al. (2015)

Results

We did not find significant differences either in courtship or mating duration between mating pairs from the two studied locations (Fig. 2a). Data related to the number of occurrences and durations of courtship and copulatory behavioral units for Uruguayan and Argentinean sexual pairs, with the corresponding results of their statistical comparisons (shown whenever they were significant), are summarized in Fig. 2b. The general behavioral patterns were similar in the two groups. However, the number of erection of male leg spines, male abdominal vibrations, and the number of male and female

touches were all higher in the Argentinean pairs compared to the pairs from Uruguay (see Fig. 2b).

Data on body measures and burrow dimensions is shown in Table 2. We did not find significant differences in burrow length or diameter between the two locations. Body characteristics showed differences between the two study sites; Uruguayan females presented higher body condition indexes than Argentinean ones, and Uruguayan males showed higher body mass values compared to those from Argentina (Table 2).

Body mass did not show relation with the number of erection of male leg spines ($F = 1.18, p = 0.29$), male abdominal

Fig. 2 Mean values and standard deviations of the durations (a) and total number of cases (b) of courtship and copulatory behaviors, per trial in both Argentinean and Uruguayan couples. The corresponding results of statistical comparisons are shown whenever they were significant. * $0.05 > P > 0.01$, ** $0.01 > P > 0.001$, *** $0.001 > P > 0.0001$

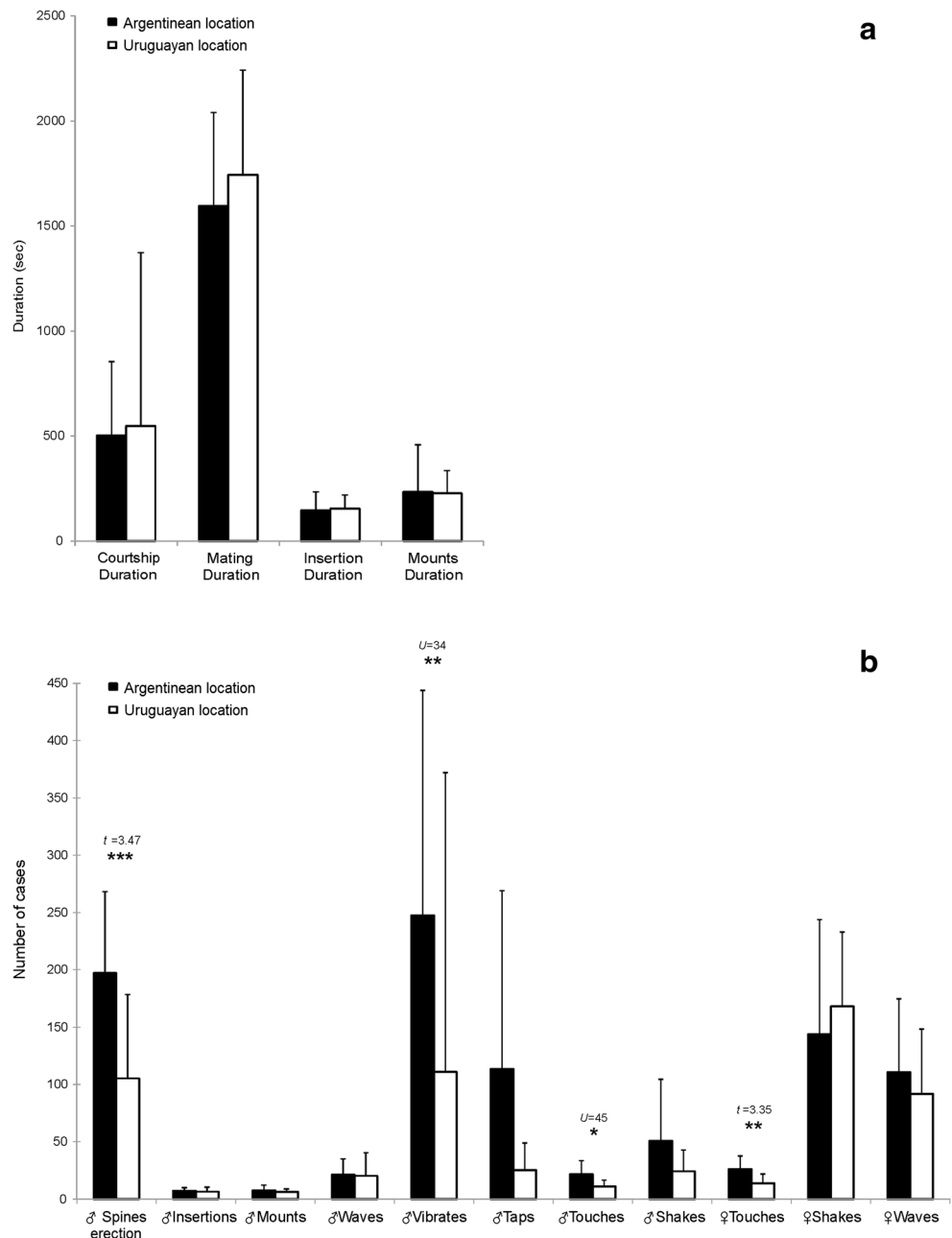


Table 2 Data (mean \pm SD) of body characteristics of females and males and male burrow dimensions for Uruguayan and Argentinean locations, with the corresponding results of the statistical comparisons between the two groups

	Argentinean individuals	Uruguayan individuals	Statistics
Male mass (g)	0.3323 \pm 0.1405 ($n = 19$)	0.5336 \pm 0.2784 ($n = 15$)	$U = 84, P = 0.04$
Male body condition index	9.1953 \pm 1.1873 ($n = 19$)	8.6425 \pm 2.5560 ($n = 16$)	$U = 147, P = 0.87$
Burrow length (mm)	136.316 \pm 38.4892 ($n = 19$)	124.125 \pm 62.3879 ($n = 16$)	$U = 109, P = 0.15$
Burrow diameter (mm)	11.8942 \pm 1.0530 ($n = 19$)	12.0000 \pm 2.7386 ($n = 13$)	$U = 116, P = 0.77$
Female mass (g)	0.2596 \pm 0.1093 ($n = 8$)	0.341 \pm 0.04040 ($n = 10$)	$U = 19, P = 0.06$
Female body condition index	1.0577 \pm 0.1579 ($n = 8$)	1.2582 \pm 0.1860 ($n = 10$)	$t = -2.43, P = 0.01$

vibrations ($F = 0.96, p = 0.34$), or male touches ($F = 1.10, p = 0.31$). We found a significant relation between the location and the number of erection of male leg spines ($F = 15.37, p = 0.0008$), number of abdominal vibrations ($F = 16.67, p = 0.0006$), and number of touches ($F = 10.89, p = 0.003$), which all were higher in the Argentinean location.

Discussion

According to the results of the present research, the two studied locations of *A. senex* show differences in their courtship (male touches) and mating behaviors (number of erection of male leg spines and number of male abdominal vibrations), and in all the cases, values are higher in mating pairs from Argentina. It is notable that three out of four of those behaviors correspond to males and only one to females. However, we only recorded female courtship behavior and copulatory body shakings so more attention to female sexual behaviors should be contemplated in future research on this species. Moreover, with the exception of the number of erections of male leg spines, behaviors are not directly related to sperm transfer per se and likely represent copulatory courtship. The fact that all those behaviors were more intense in Argentinean mating pairs could be explained by the fact that individuals from that location inhabit smaller beaches, with higher aggregations of individuals compared to the Uruguayan location (Bollatti personal observation). These circumstances could generate higher chances of finding mates and higher male-male competition too, similarly to what González (2015) reported for *Aglaoctenus lagotis* wolf spider. Thus, individuals should have higher pressures to be selected by the other sex, even more in this species in which both sexes are very choosy (Aisenberg et al. 2011a). Selective pressures can vary with the environment so we can expect that differences found in this study could be explained by geographical variations and environmental heterogeneity. Historical and geographical variations could be determining biotic and abiotic factors that affect each *A. senex* location. In this study, we did not find significant differences between the temperatures in which individuals of both locations were exposed to trials under laboratory conditions. However, in the wild *A. senex*, individuals of selected locations

are exposed not only to different temperature conditions but also to different conditions of availability of resources such as retreats or preys (Alonso-Paz and Bassagoda 2006; De Simone 2013; Postiglioni 2015; De Simone et al. 2015; Albín et al. 2016; Cingolani personal communication).

Many reports of intraspecific variations due to geographical divergence have been described for several arachnid species. Similarly to the present study, González et al. (2013) and Olivero (2014) compared sexual behaviors of Uruguayan and Argentinean populations of the wolf spider *A. lagotis* and the scorpion *Bothriurus bonariensis*, respectively (Table 3). *A. lagotis* showed sexual behaviors exclusive for each population and differences in patterns and frequencies of occurrence of courtship and copulatory behaviors common to both populations (González et al. 2013). These authors discussed the fact that the southern form of *A. lagotis* is exposed to more extensive winters, less pronounced rainfall regimes, and less extreme temperatures than the northern form, and differences could be acting as primary differentiation. Those characteristics could favor divergence processes or an exceptionally high level of flexibility in the sexual behavior of the species involved (González et al. 2013). The scorpion *B. bonariensis* also exhibited differences in durations and frequencies of occurrence of mating behaviors between Uruguayan and Argentinean pairs (Olivero et al. 2012). The previous authors suggest that those differences in sexual behavior could be explained by the ecoregions in which each population is found since the core population in Uruguay belongs to Pampean Province while the peripheral population in Argentina belongs to Espinal Province (Cabrera and Willink 1980; Ojanguren Affilastro 2005), showing different vegetation, soil, and climate (Cabrera and Willink 1973) that could affect the populations on different ways. The results of this study provide evidence against genetic drift due to similar pattern across species with similar ecologies; however, further studies are needed to confirm these patterns.

We found that Argentinean pairs of *A. senex* showed a higher number of erection of male leg spines than Uruguayan ones, but we did not find differences in mating and palpal insertion durations between the two locations. This result could suggest that Argentinean males could transfer more sperm than Uruguayan ones; however, this is not

Table 3 Comparison among sexual and morphological traits evaluated in two arachnids with similar distribution (the spider *Aglaoctenus lagotis* and the scorpion *Bothriurus bonariensis*) and *A. senex*

	<i>A. lagotis</i>	<i>B. bonariensis</i>	<i>A. senex</i>
Courtship duration	N > S	=	=
Mating duration	S > N	=	=
Insertion duration	N > S	–	=
Number of erections of male leg spines	N > S	Does not correspond	N > S
Sexual behavior frequencies	Δ	Δ	N > S
Body size	S > N	S > N	♀S > N
Body mass	–	–	♂S > N
Sexual periods	≠	=	=
Pigmentation	≠	≠	–
Mating system	≠	=	=

In all the cases, the Uruguayan location (S) is located in Uruguay and Argentinean location (N) is located in central Argentina. One triangle (Δ) indicates the bidirectional variation in such trait. Sexual periods means the period in the year in which individuals are sexually active. The greater-than (>) symbol after the location denotes the higher values for that locality, not equal to (≠) symbol denotes the differences in when the sexual periods occur between the two localities, equals sign (=) denotes the similarity in when the sexual period occurs, and minus sign (–) stands for the absence of enough data. Information about *A. lagotis* and *B. bonariensis* is based on González et al. (2013) and Olivero et al. (2012), respectively. Data about *A. senex* correspond to the present study

necessarily true (see Herberstein et al. 2011). Theoretical models have postulated that males can increase sperm investment in agreement with the level of sperm competition (Parker et al. 1997; Engqvist and Reinhold 2006, 2007). On the other hand, Immler et al. (2011) found that males perform a trade-off between pre-copulatory competition for mating and sperm competition. Moreover, the level of sperm competition in a population can vary from low risk (when females generally have one mate per clutch but eventually can mate with two males) to high intensity (when females mate with numerous and different males) (Parker and Pizzari 2010). In this frame, considering the higher aggregation of individuals at the Argentinean location sperm competition could be higher at that location. New studies are required to test this prediction.

In the present study, Uruguayan males were heavier than Argentinean males, while Uruguayan females showed higher indexes of body condition. Nevertheless, none of these body characteristics affected the sexual behavior of the individuals. One explanation that could agree with our results is what Entling et al. (2010) found about body size in spider fauna of Europe that would decrease from warm/dry to cool/moist environments. However, regarding males, other abiotic characteristics such as the type of substrate—considering that males are good diggers—or biotic variables as prey abundance, vegetation coverage, and/or density, could be determining the differences in body mass between individuals from each locality. Furthermore, the intensity of sexual selection forces acting on a certain population can show marked variances according to environmental characteristics (Claridge et al. 1985; Miller et al. 1998; Elias et al. 2006; Ishikawa et al. 2006; Macías-Ordoñez et al. 2013; González et al. 2013). Differences in size or weight can be related with the degree of direct male-male competition, sperm competition, or

female choice, as has been reported for other animal groups (Andersson 1994; Blankenhorn 2000).

Though there was a strong trend, we did not find differences in burrow length or width between the two locations. Male burrows are the mating refuges that need to be stable enough to resist the changing of positions between females and males prior to mating and the frequent mounts and dismounts during the sexual encounter (Aisenberg et al. 2007). Furthermore, male burrows also function as breeding nests for the future progeny, so they need to provide adequate temperature and humidity conditions for development of the eggs (Aisenberg et al. 2011a; Aisenberg 2014). Those conditions depend on burrow length (Aisenberg et al. 2011b), so male burrow dimensions could be relatively conservative to fulfill the requirements for all the previous functions.

All the traits studied here (reproductive behaviors, body size and mass, and burrow dimensions) are labile traits, which means that they can change repeatedly along the life of the individuals (Gomulkiewicz and Kirkpatrick 1992; Nussey et al. 2007). This capacity to alter the expression of a changeable trait in response to environmental conditions is phenotypic plasticity (Réale et al. 2003; Nussey et al. 2007). In this sense, phenotypic plasticity could explain the differences found in this research. The best stage to validate this hypothesis should be to test the variation of those characteristics in wild populations and/or cross-fostering experiments (Nussey et al. 2007; Forsman 2015). Whether differences in sexual behavior between pairs from each location are fixed or plastic behaviors that can be adjusted remain to be tested in the near future. Considering that Peretti and Aisenberg (personal observation) have observed matings involving males and females from different locations, another interesting point could be to perform mixed matings with individuals

from the two locations and evaluate courtship and copulatory patterns to determine if individuals are able of modulating their behavioral patterns when they are exposed to ‘foreign’ mating partners. Finally, this sex-role reversed wolf spider is a promising model for testing if the differences in sexual behavior represent genetic divergence or if they can be explained by phenotypic plasticity, enlightening the discussion about the causes driving the evolution of the atypical sexual behavior found in this species.

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