Original Article

Male dimorphism of a neotropical arachnid: harem size, sneaker opportunities, and gonadal investment

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Serracutisoma proximum is a harvestman with alternative male morphs. Large males use sexually dimorphic second legs in fights for the possession of territories on the vegetation, where females oviposit. Small males have short second legs and do not fight but rather sneak into the territories and copulate with egg-guarding females. We investigated the presence of male dimorphism across 10 populations of *S. proximum*, compared gonadal investment between male morphs, and assessed if the distribution of the sneakers is influenced by harem size. In all populations, there was male dimorphism, indicated by the bimodal distribution of the leg II length/body length. Gonadal investment did not differ between morphs and was not affected by male size, second leg length, and morph relative frequency in the populations. We found 361 territories, 90.0% containing 1 male, 9.7% containing 2 males (dyads), and 0.3% containing 3 males. The probability of encountering dyads increased with the number of females present in the territories. Moreover, the proportion of sneakers in territories containing dyads was higher than would be expected by chance. One possible reason for the ubiquity of alternative morphs in *S. proximum* could be the high mating opportunities experienced by sneakers in spatially structured populations with a resource defense polygyny system. Additionally, the high frequency of successful invasions by sneakers and hence the high sperm competition risk for both morphs may explain the similarity in gonadal investment between male morphs. *Key words:* alternative mating tactics, resource defense polygyny, sperm competition, testis mass. *[Behav Ecol]*

INTRODUCTION

Sexual selection on males has frequently originated a diverse array of phenotypic characters linked to intrasexual competition and mate choice (Andersson 1994; Berglund et al. 1996). As a result of these selective pressures, males of different species have recurrently developed weaponry such as antlers, horns, forceps, and claws that are employed both in agonistic interactions between rivals for female access and in sexual displays to females, which may use these morphological traits to screen males for potential partners (Andersson 1994; Berglund et al. 1996; Candolin 2003; Emlen 2008). Males with exaggerated traits used primarily as weapons increase their fitness via a higher resource-holding power associated to reproductive access to receptive females (e.g., Forsyth and Alcock 1990; Sneddon et al. 1997; Goddard and Mathis 2000; Bro-Jørgensen 2007). However, the expression of weapons in males is costly, and there is a trade-off between the investment in weapons and in other fitness components, such as somatic structures associated to viability (e.g., Zahavi 1975; Rowe and Houle 1996; Nijhout and Emlen 1998).

In systems in which there is male variation in weapon expression, females usually mate with a small subset of males that bear pronounced traits, thus providing the opportunity for alternative male tactics to evolve (Shuster and Wade 2003; Taborsky et al. 2008). Individuals of the alternative male morph (usually called "sneakers") typically do not invest in weapons and thus would be expected to have fewer mating opportunities when compared with individuals of the morph that express weapons. The probability of successful fertilization in each mating opportunity, however, is often higher for sneakers because they generally have adaptations to maximize fertilization of gametes (Taborsky 1998; Simmons et al. 1999). One of these adaptations includes investing proportionally more in structures associated to sperm competition, such as testis mass (e.g., Simmons et al. 1999, 2007). Gonadal investment in sneakers can be even higher than territorials because territory invaders are always in sperm competition with the owners of territories. Whenever males in a given population adopt sneaking tactics, it is expected that their copulations will nearly always occur with females that have previously mated with a territorial and that the ejaculates of both males will compete for fertilization of the eggs (Parker 1970, 1990; Simmons 2001).

Whereas theoretical models predict that sneakers will always be subject to sperm competition, the degree that territorials will also be subject to sperm competition, and hence their allocation in gonadal structures, can depend on the demographic parameters that modulate sperm competition, such as the relative frequency of sneakers in the population (Parker 1990; Simmons 2001). Indeed, the relative frequency of sneakers in the population modifies sperm competition risk, which in turn influences sperm expenditure of each male

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morph (Parker 1990; Simmons 2001). Asymmetries in gonadal investment between sneakers and territorials should be low when the relative frequency of sneakers is low. This pattern is generated due to minimal sperm competition risks for territorials and small selective pressure for sneakers to invest much energy in gonads at expense of somatic structures. When the relative frequency of sneakers is high, both territorials and sneakers should be under very high competition risk, and thus asymmetries in gonadal investment should be low again. However, when the relative frequency of sneakers is intermediate, sperm competition risk for territorials is lower than the risk faced by sneakers, thus the asymmetries in gonadal investment should be high, with sneakers investing proportionally more than territorials (Parker 1990; Simmons 2001).

In addition to the relative frequency of sneakers, ecological conditions, such as female distribution patterns, sex ratio, and population density can affect sperm competition risk (Shuster and Wade 2003). For instance, in species where females form harems of various sizes, sneakers may preferentially invade large harems where they are more likely to achieve copulations. This nonrandom distribution of sneakers' invasions will add variation to sperm competition risk for territorials (e.g., Shuster 1987; Wade and Shuster 2004). Moreover, population density can affect agonistic encounter rates between territorials and also the number of sneakers invading harems. Therefore, environmental and demographic parameters may influence the relative fitness of each male tactic and ultimately may determine whether the population will be composed of

dimorphic or monomorphic males (Tomkins and Brown 2004).

Serracutisoma proximum (Arachnida: Opiliones) is a large Neotropical harvestman and, at least in one population, we found 2 distinct male phenotypes that exhibit different behavioral reproductive tactics (Buzatto et al. 2011). Territorials use their long and sexually dimorphic second legs in ritualistic fights for the possession of territories on the vegetation, which may contain up to 6 females caring for their eggs (Buzatto et al. 2007, 2011; Buzatto and Machado 2008; Figure 1). Sneakers, on the other hand, possess shorter second legs and their reproductive behavior is based on invading territories and furtively mating with egg-guarding females (Buzatto et al. 2011; Figure 1). Intensive field observations indicated that sneakers can invade up to 5 harems and that nearly 70% of the invasions are successful and result in copulations. Finally, a long-term mark–recapture study showed that sneakers are more vagile than territorials, probably because their mating tactic relies on the invasion of different harems (see Figure 3 in Buzatto et al. 2011).

In this study, we investigate the presence of alternative male morphs across 10 naturally isolated populations of *S. proximum* and use data on male morphology, gonadal investment, and population ecology to test 2 hypotheses. 1) Gonadal investment of each male morph should vary according to the probability of a sneak mating. Asymmetries in gonadal investment between territorials and sneakers should be low when sneakers are rare or abundant in the population. On the other hand, when the relative frequency of sneakers is
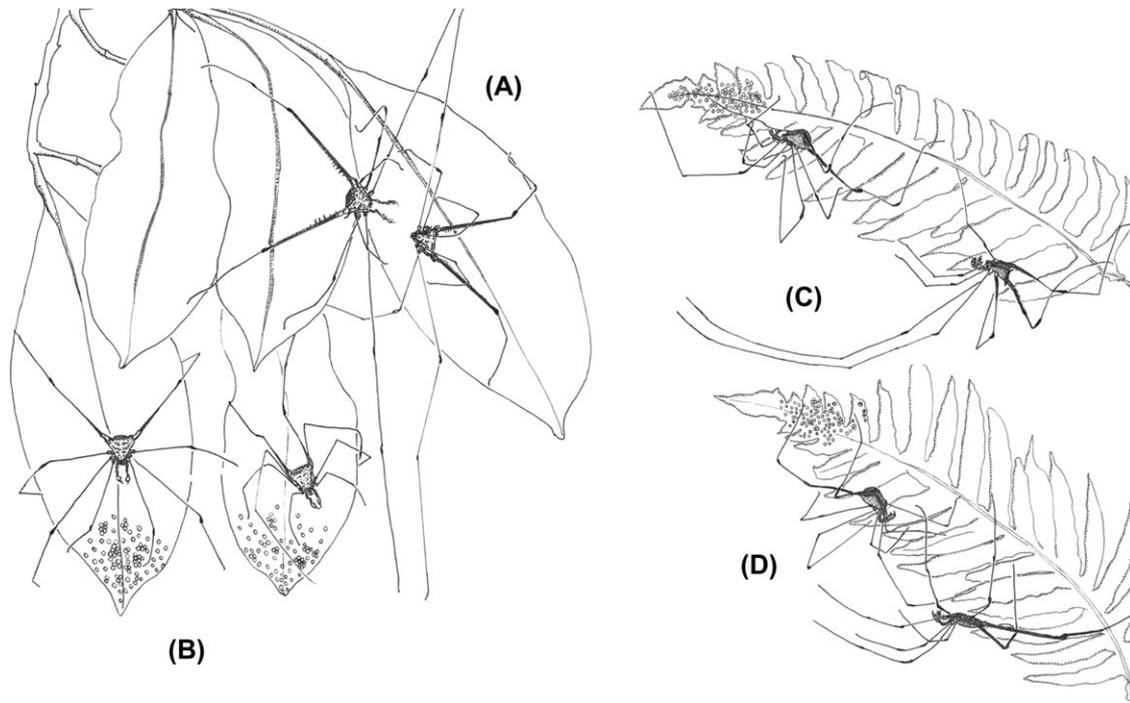
Figure 1
Scheme depicting the main features of the mating system of the harvestman *Serracutisoma proximum*. (A) At the beginning of the reproductive season, large males fight for territories on the vegetation, as in a typical resource defense mating system. During the fights, territorials hit each other with their elongated second pair of legs, which are much longer than those of females. (B) Females visit the territories, copulate with the territorials, and lay their eggs on the undersurface of the leaves. Although nearly 80–90% of the eggs are laid in the first 24 h after copulation with territorials, females may take up to 14 days to complete oviposition. (C) While the female oviposits, the territorial mate guards her with his second pair of legs extended toward her. After the arrival of several females in a harem, territorials concentrate their patrolling activity mostly on egg-guarding females. At this stage, the mating system seems to shift to a female defense polygyny. (D) Small males, with short second pair of legs, do not patrol or defend females and territories but instead adopt an alternative reproductive tactic based on invading harems and sneaking copulations with egg-guarding females. Sneak copulations generally occur when territorials are mate guarding another female or when they leave the territory to forage. The sneaking tactic is only possible because females have a long-lasting oviposition behavior, which allows sneakers to sire at least some of the last eggs females lay.

intermediate, the asymmetry in gonadal investment should be high (see Figure 7.5A in Simmons 2001). 2) Harems with a greater number of females will be more attractive to intruder males, that is, the sneakers (Shuster 1987). To our knowledge, this study is the first one to investigate sperm competition in many naturally isolated populations, which provides one of the most comprehensive empirical tests of Parker's (1990) model. Moreover, analyses of morph expression at the interpopulation level allow us to take into account environmental and demographic parameters that drive the local maintenance of the male dimorphism and modulate the intensity of sperm competition between male morphs.

MATERIALS AND METHODS

Model system

The alternative male phenotypes of *S. proximum* seem to represent a polyphenism, that is, a dichotomous phenotypic plasticity (West-Eberhard 2003; Buzatto et al. 2011). Known in evolutionary game theory as the conditional evolutionarily stable strategy, polyphenic dimorphisms evolve and are maintained when individuals' phenotypes are decided through conditional decision rules (Hazel et al. 1990). If male body size (or any other proxy to status) determines the fitness of each phenotypic tactic, the alternative phenotypes are expected to evolve to become body size dependent, where individuals larger than an evolutionarily stable switchpoint benefit from adopting one phenotype, whereas individuals smaller than this switchpoint benefit from adopting the other (Hazel et al. 1990; Tomkins and Hazel 2007). The evolution and maintenance of conditional dimorphisms, as well as the potential role of density and frequency-dependent selection, have been reviewed and modeled under the light of the "environmentally cued threshold (ET) model" (Tomkins and Hazel 2007). We recently used this model to analyze male dimorphism in one population of *S. proximum* (Buzatto et al. 2011). This approach allowed us to infer that the alternative phenotypes in this species are body size dependent and that there is a large amount of genetic variation for the switchpoint between male morphs (Buzatto et al. 2011).

Study site

We did the study in 10 populations located at Intervales State Park, state of São Paulo, southeastern Brazil, during January–February 2009. The site is covered by a dense Atlantic Forest, has an annual rainfall from 2000 to 3000 mm, and the mean annual temperature ranges from 12 to 20 °C. Individuals of *S. proximum* are found on the vegetation along the margins of streams, and guarding females use mostly the ferns *Blechnum binervatum* (Blechnaceae) and *Olfersia cervina* (Dryopteridaceae), and the tree *Gomidesia schaueriana* (Myrtaceae) as oviposition sites (Buzatto et al. 2007; Buzatto and Machado 2008). All 10 populations are located at spatially distant streams, each of them belonging to a different hydrologic microbasin: 1) Rio da Caçadinha (CAC) (lat 24°16′43.8″S, long 48°24′47.8″W), 2) Rio do Carmo (CAR) (lat 24°18′23″S, long 48°24′52″W), 3) Rio Capoava (CAP) (lat 24°18′16″S, long 48°26′03″W), 4) Rio Três Córregos (RTC) (lat 24°19′19″S, long 48°23′32″W), 5) Rio das Mortes (RDM) (24°20′18″S, 48°26′07″W), 6) Rio Poços Altos (RPA) (24°18′19″S, 48°20′54″W), 7) Cachoeira do Arcão (ARC) (24°17′20.8″S, 48°26′01″W), 8) Rio Novo (RNO) (24°21′0.7″S, 48°27′18.3″W), 9) Rio dos Burros (RDB) (24°15′46″S, 48°23′07″W), and 10) Cachoeira das Pedrinhas (PED) (24°18′50″S, 48°22′33.8″W). Considering that individuals of *S. proximum* are always associated to the margins of streams (Ramires and Giaretta 1994; Machado 2002;

Buzatto et al. 2007; Buzatto and Machado 2008), it is likely that the genetic flow between the studied populations is minimal or nonexistent.

Sampling populations in the field

In each population, we searched for one day (from 10:00 to 17:00 h) adult individuals of *S. proximum* at both margins of the streams, looking for males and females on the vegetation, stones, and logs along transects ranging from 160 to 450 m (mean = 288 m). Transect length variation was due to differences in the density of individuals, so that populations with lower densities required longer transects to obtain a minimum number of males ($n = 35$) to perform the morphological analyses. In each transect, we captured and counted males and females, so that we could estimate sex ratio and density for each population. Because we inspected the marginal vegetation at a maximum distance of 2 m from the water, we calculated population density as the total number of individuals divided by transect length multiplied by 4 to account for both margins of the streams. After each sampling, we released all females and collected and transported all males to the laboratory for further morphological and gonadal investment analyses (see below). At the end of the fieldwork, we released all males that were not dissected to their original populations.

Although most gonyleptid harvestmen are strictly nocturnal, males of *S. proximum* are known to actively patrol and defend territories on the vegetation during daylight, especially during the peak of the reproductive season, between November and February (Buzatto and Machado 2008). Because we performed our observations in January, we considered that all active males we encountered were defending territories, defined as areas on the vegetation, rocks, or tree trunks patrolled and defended by males of the larger morph (Buzatto and Machado 2008). Based on extensive field observations that we conducted in a previous study with *S. proximum*, we defined a territory as a circular area with 1 m of radius (see Figure 4 in Buzatto and Machado 2008), the center of which was defined by the position of the territorial (see "Male morph discrimination" below). We considered all females encountered inside a territory as belonging to the male's harem.

Male morph discrimination

As an exploratory visual analysis, we performed scatterplots with the natural logarithms of leg II length and dorsal scute length across the 10 populations. Subsequently, to discriminate between male morphs in each population, we fitted a kernel density function to the frequency distribution of the ratios of the right leg II length to the dorsal scute length, using dorsal scute length as an estimate of body size (according to Willemart, Farine, et al. 2009; Buzatto et al. 2011). All populations showed a clear bimodal distribution of the ratios (see RESULTS), and thus we used the minimum value of leg II length/dorsal scute length ratio found between the 2 modes as the criterion to separate males in sneakers and territorials (Knell 2009). Given that male morphology and behavior are phenotypically integrated in *S. proximum*, the analytical discrimination of the morphs provides reliable information on the mating tactic (Buzatto et al. 2011).

Additionally, we estimated the relative frequencies of the 2 male morphs in each population. Because we know that sneakers are more vagile and have a lower probability of detection when compared with territorials (Buzatto et al. 2011), we took into account these systematic detection divergences between male morphs to avoid underestimating the relative frequency of sneakers in the populations. We did this by taking the recapture parameters corresponding to the peak of the

reproductive season (December–February) for sneakers and territorials from Buzatto et al. (2011) mark–recapture work in the CAC population and estimating their mean recapture probability ($\hat{p}$) for this period using the variance components approach. Next, we used the mean recapture probability of territorials ($\hat{p}_T = 0.687$) and sneakers ($\hat{p}_S = 0.503$) to assess their detectability using the ratio of these parameters ($\hat{p}_S/\hat{p}_T$). We found that territorials are 1.37 times more detectable than sneakers during the peak or reproductive season. Thus, to obtain a corrected relative frequency of the morphs, we multiplied the relative frequency of sneakers captured in the field by this detectability ratio for all populations we sampled.

Hypothesis 1: gonadal investment

Following other studies on sperm competition in arthropods (see examples in Simmons 2001), our proxy of gonadal investment was testis mass. To assess if gonadal investment of each male morph varies according to the relative frequency of sneakers in each population, we dissected 299 males and extracted the testis from a randomly defined subset of territorials ($n = 221$ individuals, population range = 18–26, population mean = 22.1). Additionally, we successfully extracted the testis from 95.1% of all sneakers we found ($n = 78$ individuals, population range = 2–16, population mean = 7.8). The number of sneakers (S) and territorials (T) dissected in each population was: CAC = 15 S/ 18 T, CAR = 9 S/21 T, CAP = 7 S/21 T, RTC = 8 S/22 T, RDM = 3 S/21 T, RPA = 16 S/20 T, ARC = 6 S/26 T, RNO = 6 S/25 T, RDB = 2 S/21 T, and PED = 6 S/26 T. We preserved the testis of all males in 100% ethanol and subsequently weighted them in an analytic balance with precision of 0.0001 g.

We constructed a hierarchical model with normal distribution, considering testis mass as a response variable, male morph, bogy size, leg II length, and their interaction as individual level predictor variables, and relative frequency of sneakers as a group level predictor variable. The inclusion of body size as a covariate was pertinent because sneakers are smaller than territorials, and we wanted to assess relative gonad investment between morphs. Population was considered a random variable. The analysis of gonadal investment we performed is an extension of the analysis of covariance suggested by Tomkins and Simmons (2002). However, the hierarchical models allowed us to take into account individuals from different populations and consider individual and population level predictors (Gelman and Hill 2007).

Hypothesis 2: harem invasion

In a subset of 7 populations, we assessed the mean number of females present in each territory defended by a male and its 95% confidence interval (Shuster and Wade 2003). We assessed if females within male territories were aggregated or randomly distributed. As the distribution of females become more aggregated, there will a clumping effect, that is, few territories will have a high number of females, whereas many territories will have a low number or no females at all (Shuster and Wade 2003). We tested if the distribution of females within male territories is aggregated or random fitting models with Poisson and negative binomial distribution for each population. We used the Akaike information criteria (AIC) to compare and select the models with Poisson and negative binomial distributions, choosing the model with the distribution that presented the lowest values of AIC and more than 2 units of difference from the model with the second lowest value of AIC (Johnson and Omland 2004).

Additionally, for each studied population, we estimated the harem size (H), that is, the mean number of females associated only with territories that had at least one female, excluding the territories of unsuccessful males (Shuster and Wade 2003). In the cases in which there were more than one male present in the harem (dyads), we considered that both had reproductive access to the females in the harem. Theoretically, these dyads could have 2 territorials, 2 sneakers, or 1 male of each morph. We estimated the mean number of females that were in successful males' territories using a modified version of Shuster and Wade (2003) equation:

$$H = \left(\sum K_i m_i w_i \right) \Big/ \left[\left(\sum m_i \right) - m_0 \right],$$

where K are classes defined by the number of females in each territory (ranging from 0 to n females), m is the number of territories in each class of K, w is the reciprocal number of males in the territory (i.e., if there is 1 male, $w = 1$, if there are 2 males, $w = 0.5$), and m_0 correspond to the number of territories without females. This formula avoids inflating the number of females when there is more than one male present in the harem.

We performed a generalized linear mixed model (GLMM) with binomial distribution with the presence of an additional male in the territory (dyad) as a binary response variable and the number of females in the territory as the predictor variable, considering the population origin as the random variable. Additionally, we assessed if the frequency of the combinations of morphs in the dyads "Sneaker–Sneaker" (S-S), "Sneaker–Territorial" (S-T), and "Territorial–Territorial" (T-T) occurring at random in a population differed from our observations. The null model estimated the frequencies of the 3 types of dyads taking into account the relative frequency of each morph in a population and considering the same number of dyads observed in the field. Thus, if a is the relative frequency of sneakers and b the relative frequency of territorials, we should expect a^2, $2ab$, and b^2 for the relative frequencies of S-S, S-T, and T-T dyads, respectively. This procedure is equivalent to what is done in population genetics analysis when genotypic frequencies in the next generation are calculated from gene frequencies to test Hardy–Weinberg equilibrium. As dyads originated in several populations and every population had a different relative frequency of morphs, we estimated the weighted mean of the relative frequency of sneakers across populations. To obtain the mean relative frequency of sneakers, the relative frequency of each population was summed after weighting the proportion of dyads that occurred at that specific population. We compared the predicted and observed frequencies of the 3 types of dyads using a chi-square test.

We made all the analyses and graphics using the R program (R Core Development Team 2009). More specifically, we used the package ggplot2 (Wickham 2009) for the scatterplots and the density kernel distributions and the packages lme4 (Bates and Maechler 2009) for the GLMM and the hierarchical model.

RESULTS

Male morph discrimination and population parameters

We found male dimorphism in the length of leg II in all 10 populations (Figure 2A). Moreover, there was a clear bimodal distribution of the leg II length/dorsal scute length, also indicative of intrasexual dimorphism in all 10 populations (Figure 2B). In the total, we found 625 males, 82 (13.1%) being sneakers and 543 (86.9%) being territorials. Relative frequency of sneakers (taking into account differences in detectability) ranged from 5.8% to 36.7% among populations, with a mean of 16.9%. Adult sex ratio ($\male/\female$) varied from 0.624 in PED to 1.291 in RDB (mean ± standard deviation [SD] = 0.931 ± 0.239), and population density varied from 0.042 ind/m^2 in RDB to 0.339 ind/m^2 in PED (mean ± SD = 0.139 ± 0.090 ind/m^2).
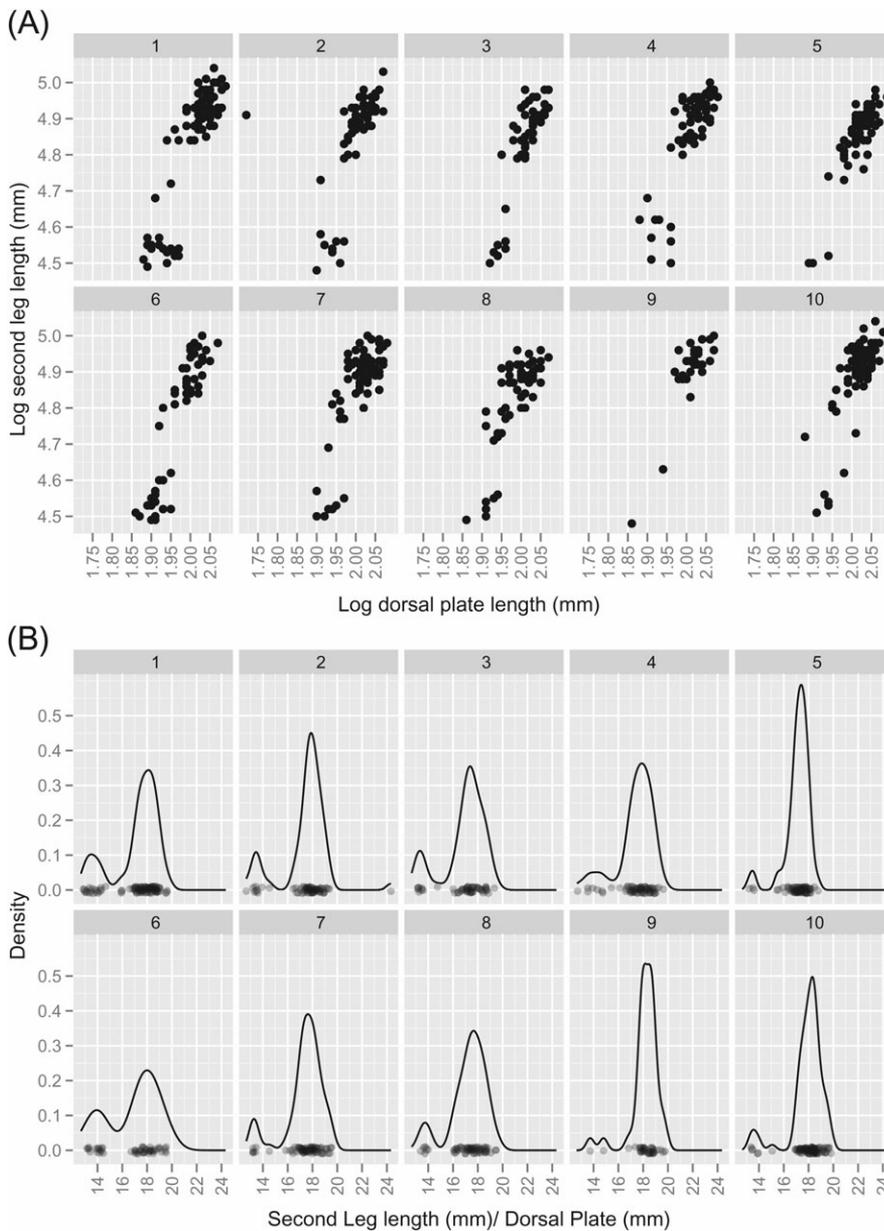
Figure 2
(A) Scatterplots showing leg II length as a sexual dimorphic character and dorsal scute as an estimate of body size in 10 populations of the harvestman *Serracutisoma proximum*. Both measures were transformed to natural logarithms. (B) Kernel density estimates of leg II length/dorsal scute ratio. Minimum kernel density values used for morph discrimination ranged from 14.51 to 15.96, with a mean of 15.35.

Gonadal investment

The mean testis mass for territorials was 0.49 mg (range = 0.1–1.1 mg), whereas the mean testis mass for sneakers was 0.69 mg (range = 0.2–1.4 mg). Mean testis mass, however, did not differ between male morphs and was not affected by male size, sneaker relative frequency (range = 5–33%), or the interactions between morph and body size and morph and leg II length (Table 1).

Harem invasion

Female distribution in harems varied between populations, so that mean harem size values (H) ranged among populations from 1.351 in RDM to 3.375 in TCO (Table 2). A large proportion of territories did not contain females (n = 183 of 361, Figure 3A). When there were females in the territories, the majority of them had only 1–3 females (n = 167, Figure 3A). However, there was variation in the proportion of successful males (i.e., males with at least one female in their territory)

between populations. Successful territorial males ranged among populations from 26.5% to 71.1%, with a mean of 51.4% (Table 2).

We found a total of 361 territories, 325 of them (90.0%) containing only one territorial, 35 territories (9.7%) containing dyads, and 1 territory (0.3%) containing 3 males, being 1 territorial and 2 sneakers (Table 2). The GLMM model showed that the probability of having an additional male in the territory increases directly with the number of females present in it, and there is very low interpopulation variation, as reflected by the low value in the random effects (Table 3, Figure 3B).

We determined the composition of 35 of 36 dyads; in one dyad one male escaped at the moment of collecting, and thus we could not determine its morph. Of the 35 dyads, none was S-S, 18 were S-T, and 17 were T-T (Table 2, Figure 4). These combinations differed significantly from the expected values based only on the relative frequencies of the morphs in the populations (χ^2 = 10.862, degree of fredom = 2, P = 0.009).

Table 1

Hierarchical model analyzing gonadal investment of sneaker (*n* = 78) and territorial males (*n* = 221) of the harvestman *Serracutisoma proximum*

	Coefficient	Standard error	*t*	Probability
Fixed effects				
Intercept	−0.102	8.186	−0.124	0.902
Male morph	4.089	8.394	0.487	0.627
Dorsal scute	0.393	1.229	0.319	0.750
Leg II	0.050	0.059	0.839	0.403
Morph relative frequency	0.058	3.627	0.159	0.988
Dorsal scute: male morph	0.282	1.392	0.203	0.839
Leg II: male morph	−0.072	0.063	−1.148	0.252
Random effects				
Intercept (SD)	0.921			

Dorsal scute length, leg II length, and male morph are individual level predictors, whereas morph relative frequency is a group level predictor, and population origin is a random effect.

The number of the T-T dyads was less than the expected by chance (Figure 4). S-T dyads, on the other hand, were more frequent than the expected by chance (Figure 4).

DISCUSSION

In this study, we found that alternative male morphs are ubiquitous across all studied populations of the harvestman *S. proximum*, independently of variations in their demographic parameters, such as density or sex ratio. The only study published so far investigating the presence of alternative male morphs across many populations used the earwig *Forficula auricularia* as model organism and reported an evolutionary transition from monomorphic to male dimorphic populations over less than 40 km (Tomkins and Brown 2004). The ubiquity of alternative morphs in *S. proximum* could be related to high mating opportunities experienced by sneakers in spatially structured populations with a resource defense polygyny system (Buzatto and Machado 2008), leading to an equally high risk of sperm competition for both male morphs. Extensive behavioral observations in the field have shown that territorials are usually unsuccessful at repelling intruder males, so that in nearly 70% of the invasion attempts, sneakers achieve copulation with one female in the harems (Buzatto et al. 2011).

Considering that harvestmen are nonvisual organisms (Willemart, Osses, et al. 2009), *S. proximum* sneakers may take advantage of perceptual limitations of territorials in detecting medium-range intruders visiting females in the periphery of the harem. In fact, intruders cannot detect or be detected by a territorial until physical contact is established. Therefore, as harem size increases, it could become even more difficult for territorials to defend all females inside their harems. Moreover, territorials frequently perform postcopulatory mate guarding (Buzatto et al. 2011; Figure 1), so that sneakers

Table 2

Distribution of females and males among reproductive territories in 7 populations of the harvestman *Serracutisoma proximum*

Features	Populations						
	TCO	RDM	POA	ARC	RNO	RDB	PED
Number of territories	49	62	37	58	48	33	74
Number of dyads	8	5	3	6	4	1	9
Types of dyads (S-T, T-T)	3, 5	2, 3	2, 1	4, 2	1, 3	1, 0	5, 3
Number of females in the territories	24	50	31	75	40	18	90
Number of females outside the territories	24	31	28	24	37	12	43
Number of territorials	54	65	38	60	51	34	76
Number of successful territorials	16	42	17	42	27	9	54
Observed number of sneakers (inside and outside the territories)	8	3	16	7	6	2	7
Relative frequency of sneakers (corrected by detectability) (%)	16.87	5.95	3.66	13.78	13.88	7.46	11.20
Observed sex ratio	1.291	0.834	0.915	0.677	0.740	1.200	0.624
Mean number of females by territory (confidence interval 95%)	0.551 (0.285–1.132)	0.806 (0.603–1.051)	0.703 (0.428–1.129)	1.172 (0.873–1.560)	0.792 (0.566–1.071)	0.545 (0.217–1.661)	1.203 (0.932–1.539)
AIC Poisson model	118.2	145.4	90.0	177.9	118.4	93.6	227.4
AIC negative binomial model	96.4	147.4	89.3	174.9	119.8	60.1	224.1
k (overdispersion parameter)	0.242	—	1.284	2.420	—	0.2	2.7
Distribution	NB	Po	NC	NB	NC	NB	NB
H (mean harem size)	3.375	1.351	1.857	1.889	1.652	2.571	1.935

In a subset of territories, we found additional males (36 dyads and 1 triad). Distribution of females within the territories can be aggregated (NB = negative binomial) or random (Po = Poisson), so we compared these models using the AIC. When the AIC difference between the models is less than 2, there is not a clear support for a particular distribution (NC = not conclusive). Mean harem size (*H*) value indicates the average number of females attached to territories of successful males, that is, those territories containing at least one guarding female.
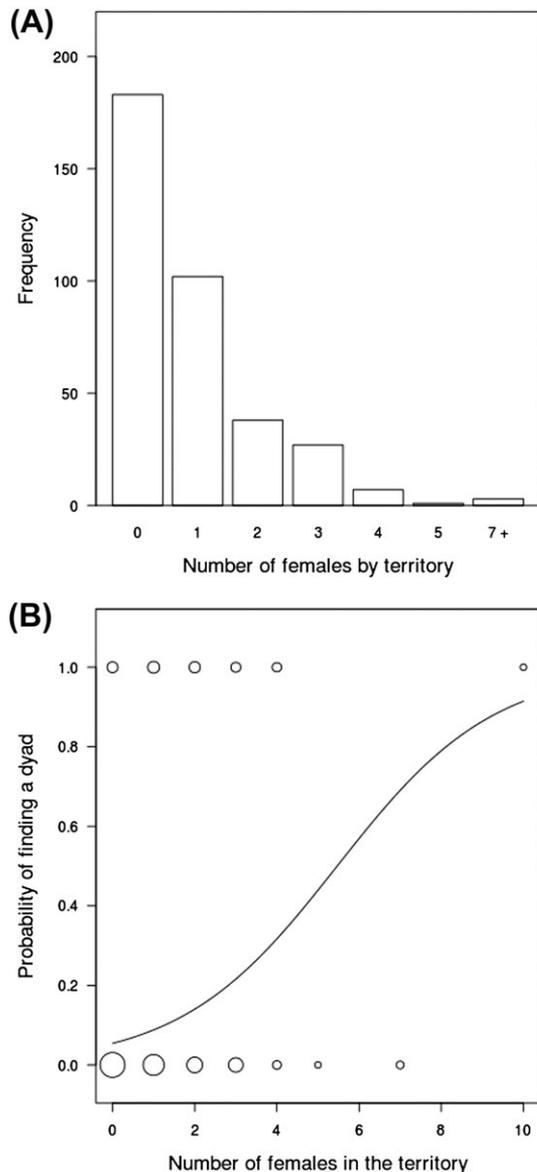
Figure 3
(A) Barplot showing the number of females in male territories of the harvestman *Serracutisoma proximum*. (B) Predicted probability values of encountering an additional male (dyad) as the number of females present in male territories increases. The line depicting the predicted values was obtained from a generalized linear mixed model with binomial distribution. The diameters of the circles are proportional to the number of observations falling at a particular point. We performed the analysis removing the point at the superior right corner (i.e., the territory with the highest number of females), and the general pattern was the same, with minimal changes in the coefficients.

harem (T-T dyads). After territorial fights, loser males may remain in the periphery of the territory without being noticed by the territory owner and strike again after some hours (Buzatto BA, Machado G, personal observation). However, given that we have never observed invasive territorial males copulating inside the harems, we suppose these males do not represent any risk of sperm competition for territory owners.

In a comparative study, Simmons et al. (2007) found that sneakers of the dung beetle *Onthophagus taurus* invest relatively more than territorials in gonads and that testis mass increases proportionally to the relative frequency of sneakers. Although the relative frequency of sneakers greatly varied among *S. proximum* populations, we did not find systematic differences in relative testis mass between male morphs. Compared with *O. taurus*, however, the costs of territorial invasion by *S. proximum* sneakers may be regarded as cheaper. In *O. taurus*, territorials have enlarged horns and compete for the possession of nests in the ground. Contrary to territorials, hornless sneakers do not assist in brood rearing but rather construct alternative tunnels to invade nests when searching for guarded females (Emlen 1997; Moczek and Emlen 2000). Therefore, *O. taurus* sneakers spend much more time and energy to assess potential sexual partners than *S. proximum* sneakers, which need only to walk few meters to access 3–5 harems where they can find up to 14 receptive females (see Figure 3 in Buzatto et al. 2011). The cost–benefit relationship experienced by sneakers in these 2 species with alternative male tactics could be the reason for the marked difference in gonadal investment between male morphs. In *S. proximum*, of 361 territories found in the field, almost 10% of them had an additional sneaker male in a time window of a single day. In the context of the entire reproductive season, which lasts nearly 7 months (Buzatto et al. 2007), sneaker invasions are probably very frequent (see below), which means that territorials constantly face intense sperm competition. Thus, high frequency of invasions into harems coupled with great chance of achieving copulations lead to high probability of sneak mating, which may explain the lack of difference in testis mass between male morphs (Simmons 2001).

Comparing gonadal investment in the Wellington tree weta *Hemideina crassidens*, another arthropod species with dimorphic males, Kelly (2008) also found no difference between territorials and sneakers. This system has several similarities when compared with *S. proximum*: 1) territorials exhibit a defense polygynous mating system, guarding females inside tree cavities, whereas sneakers acquire matings by infiltrating harems (Kelly 2008); 2) because tree cavities are scarce and spatially scattered, female distribution is highly clumped (Kelly 2006); 3) females mate with different males between bouts of oviposition and store sperm from previous copulations, suggesting males face strong sperm competition against rivals, regardless of their morph (Kelly 2008). An important implication derived from the studies on *S. proximum* and *H. crassidens* is that the relative frequency of sneakers may not be a good proxy for sneaker mating opportunities. Moreover, differential reproductive access for sneakers conditioned to the existing variation of harem sizes can produce heterogeneity of sperm competition risk among territorials. In larger harems, there would be a selective pressure for territorials to invest more in gonads to offset both the higher invasion risk from sneakers that try to copulate with females inside the harems and the sperm competition inside the female spermathecae.

Populations of *S. proximum* where females were highly clumped in a few harems had the highest *H* index (Table 2), possibly making harems with a higher number of females more attractive to intruder males. This was confirmed by our data because the probability of encountering intruder males
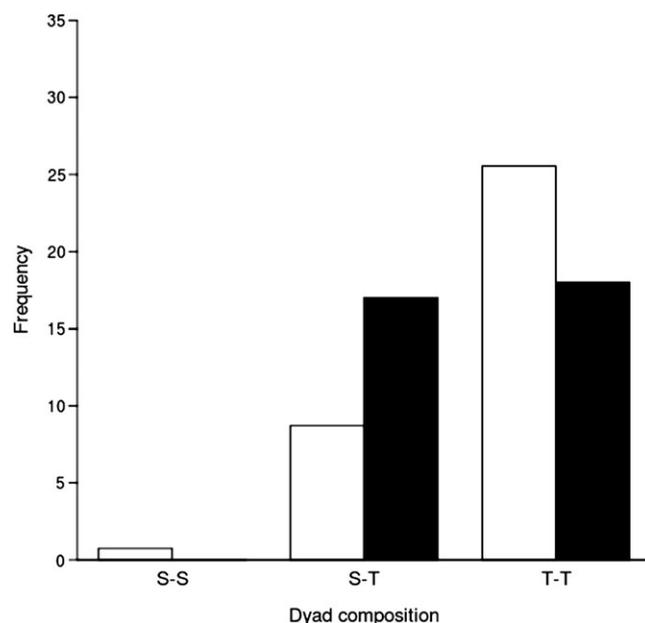
could seize the opportunity to copulate with one female when the territorial is copulating or mate guarding another female of the harem. The evolution of alternative male morphs that benefit from perceptual constraints of territorials is common in arthropods with a resource defense polygyny mating system, including examples of female mimicry in which sneakers are confounded with females by territorials (e.g., Shuster 1987; Forsyth and Alcock 1990). The fact that harvestmen are unable to detect long-range cues, such as visual and chemical stimuli, may also explain the presence of 2 territorials inside the same

Table 3

GLMM model used to analyze the formation of dyads according to the number of females present in territories in the harvestman *Serracutisoma proximum*

	Coefficient	Standard error	Z value	Probability
Fixed effects				
Intercept	−2.860	0.258	−11.073	<0.001
Number of females by territory	0.523	0.123	4.24	<0.001
Random effects				
Intercept (SD)	0.001			

The model considers the presence of an additional male in the territories as the binary response variable, the number of females by territory as the predictor, and the population origin as the random effect.

(mainly sneakers) in the territories increased with the number of females inside these territories and had very little variation among populations. This pattern has also been reported for the isopod *Paracerceis sculpta*, in which females are distributed in harems within spongocoels and one of the alternative morphs, the gamma males, are found more frequently in larger harems with receptive females (Shuster 1987; Wade and Shuster 2004). Sneakers are more vagile and have lower recapture rates than territorials in *S. proximum* (Buzatto et al. 2011). Instead of defending territories, sneakers are constantly walking on the vegetation, probably screening territories in order to maximize their chances of invasion. This could be accomplished by moving from one territory to another, depending on the number and reproductive status of the females inside these territories. Moreover, harem properties are probably dynamic because the receptivity of females is a transitory state (Shuster and Wade 2003). Therefore, sneakers may change the focus of invasion from one harem to another along the time. In fact, behavioral observations in the field indicate that a single sneaker can visit and successfully invade up to 5 harems during the reproductive season, acquiring copulation with a large number of females (Buzatto et al. 2011).

In conclusion, we showed that sneakers of the harvestman *S. proximum* do not invest more than territorials in gonadal structures, independently of the relative frequency of the 2 male morphs in natural populations. Moreover, we showed that territories with a greater number of females are more attractive to sneakers. We argue that spatial distribution of receptive females in the population and female capacity to store sperm from previous copulations play an important role in modulating asymmetries in gonadal investment between sneakers and territorials. Given that females mate multiple times and their clutches are generally composed of eggs from different oviposition events (Buzatto et al. 2011), *S. proximum* offers an ideal opportunity to investigate details of sperm storage and its subsequent use and to obtain field estimates of the reproductive success of territorials and sneakers based on paternity data.

Figure 4
Expected (white bars) and observed (black bars) values for dyads according to their morph composition in the harvestman *Serracutisoma proximum* (n = 35 territories with dyads).

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