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**Seleção sexual e evolução do dimorfismo sexual em duas
espécies de opiliões (Arachnida: Opiliones)**

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Orientador: Glauco Machado

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RESUMO

Ainda que existam diversos registros de formas extremas de dimorfismo sexual, a função do alongado quarto par de pernas nos machos de muitas espécies de opiliões permanece desconhecida. Neste estudo, testamos quatro previsões sobre a hipótese de que a seleção sexual está direcionando o alongamento do quarto par de pernas nos machos de *Longiperna zonata* e *Promitobates ornatus*: (1) o coeficiente alométrico do comprimento do fêmur IV em machos será maior do que o das fêmeas; (2) machos com pernas IV mais compridas vão residir próximos a grupos de fêmeas; (3) machos envolvidos em interações agressivas terão fêmures mais compridos do que o tamanho médio da população; (4) a diferença de tamanho de perna entre os oponentes será menor do que a diferença esperada entre pares de machos escolhidos ao acaso na população. Como o previsto, o coeficiente alométrico do fêmur IV foi significativamente maior em machos do que em fêmeas nas duas espécies. O comprimento do fêmur IV está positivamente relacionado à chance de um macho ser encontrado próximo a um grupo de fêmeas. Finalmente, a média do comprimento do fêmur IV dos machos envolvidos em brigas foi significativamente maior que a de machos selecionados ao acaso da população. Adicionalmente, a média da diferença de tamanho entre pares de machos que brigaram foi significativamente menor que a média da diferença de tamanho de perna entre pares de machos selecionados ao acaso na população. Assim, a seleção sexual na forma de competição entre machos parece estar direcionando o dimorfismo sexual extremo no comprimento do fêmur nessas duas espécies de opiliões.

Palavras-chave: Alometria, caracteres exagerados, briga, competição macho-macho, sistema de acasalamento, dimorfismo sexual.

ABSTRACT

Although there are many reports of extreme forms of sexual dimorphism in harvestmen, the function of the elongated fourth pair of legs in males of many species remains unknown. We tested four predictions to address the hypothesis that sexual selection is driving the enlargement of the fourth pair of legs in males of *Longiperna zonata* and *Promitobates ornatus*: (1) the allometric coefficient of femur IV length in males will be higher than in females; (2) males with longer legs IV will reside closer to groups of females; (3) males involved in aggressive interactions will have longer femur IV than the population average size; (4) the size difference between contenders will be smaller than the difference expected between randomly chosen pairs of males in the population. As predicted, the allometric coefficient of femur IV length was significantly higher in males than in females of both species. Femur IV length was positively related to the chance of a male being found close to a group of females. Finally, the mean femur IV length of the males involved in fights was significantly larger than the mean of random samples of males from the population, and the average difference between male contenders was smaller than the average random expected difference of the population. Therefore, sexual selection in the form of male-male competition seems to be driving the extreme sexual dimorphism in femur length in these species.

Key-words: Allometry, exaggerated trait, fight, male-male competition, mating system, sexual dimorphism.

INTRODUÇÃO GERAL

O cuidado parental constitui um importante aspecto na evolução das estratégias de acasalamento, uma vez que o comprometimento de cada sexo com o cuidado à prole pode influenciar fortemente os custos e benefícios da reprodução dos indivíduos (Trivers, 1972). Uma implicação do cuidado maternal é que as fêmeas, que maturam todos os ovos da ninhada simultaneamente, representam para os machos um recurso reprodutivo mais valioso do que fêmeas nas quais os ovos não se desenvolvem todos ao mesmo tempo (Austad, 1982, 1983). Portanto, em espécies com cuidado maternal pode-se esperar uma acentuada competição entre os machos por fêmeas ou por áreas propícias para oviposição ou alimentação das mesmas (Emlen & Oring, 1977). Nessas espécies espera-se também um acentuado dimorfismo sexual como consequência da seleção de estruturas envolvidas nas interações macho-fêmea ou em brigas entre machos (Andersson, 1994).

Machos de opiliões da subfamília Goniosomatinae (Gonyleptidae), por exemplo, apresentam um evidente dimorfismo sexual na armação da perna IV (Gnaspini, 1995, 2007) e no comprimento da perna II (Buzatto & Machado, 2008). Muito provavelmente todas as espécies da subfamília apresentam cuidado maternal (Machado, 2002) e alguns estudos incluem informações sobre suas estratégias de acasalamento (ver Buzatto & Machado, 2008). Um estudo detalhado com *Acutisoma longipes* (Gonyleptidae: Goniosomatinae) revelou que ao menos alguns machos defendem territórios na parede de cavernas que são visitados por fêmeas ovígeras a procura de sítios de oviposição. Existem haréns contendo até cinco fêmeas que são inspecionadas freqüentemente pelo macho dono do território (Machado & Oliveira, 1998). Brigas vigorosas entre machos foram recentemente observadas e, não surpreendentemente, estas brigas envolviam as pernas II e IV, que são dimórficas sexualmente (Buzatto & Machado, 2008).

Em espécies com cuidado paternal, a guarda dos ovos pode ser o critério selecionado pela fêmea e, nestes casos, o macho pode usar os ovos como uma exibição para as fêmeas (Tallamy, 2000). Não há uma razão óbvia para se esperar um desenvolvimento simultâneo dos ovos pelas fêmeas em espécies com cuidado paternal. De fato, em todas as espécies de opiliões que apresentam cuidado paternal as fêmeas são iteropáricas e não maturam todos os ovos simultaneamente (revisão em Machado *et al.*, 2004). Em espécies com cuidado paternal pode-se esperar também a evolução de estratégias que garantam a paternidade, como estruturas que removam esperma prévio da espermateca das fêmeas, cópulas múltiplas e coerção para as fêmeas oviporem logo após a cópula (Clutton-Brock, 1991). Observações de campo com *Ampheres leucopheus* (Gonyleptidae: Caelopyginae), uma espécie com cuidado paternal, corroboram todas essas previsões (Hara *et al.*, 2004). Nas espécies com reversão total de papéis sexuais, espera-se encontrar fêmeas competindo por machos e machos apresentando comportamentos de seleção de parceiras (Owens & Thompson, 1994; Parker & Simmons, 1996). Neste caso, como a competição entre machos deve ser menos intensa, nenhum dimorfismo sexual é esperado. De fato, machos do opilião *Zygopachylus albomarginis* (Manosbiidae), uma espécie com cuidado paternal, são cortejados pelas fêmeas e algumas vezes podem rejeitar cópulas (Rodríguez & Guerrero, 1976; Mora, 1990). Adicionalmente, a morfologia externa dos machos nessa espécie é praticamente idêntica à das fêmeas (Machado & Macías-Ordóñez, 2007).

Em espécies que não apresentam cuidado parental por nenhum dos sexos se espera que as fêmeas desenvolvam os ovos assincronicamente e que estes sejam postos individualmente ou em pequenos grupos (Tallamy & Schaffer, 1997). Em opiliões, esta é provavelmente a estratégia reprodutiva que maximiza o número de ovos produzidos pelas fêmeas na ausência de qualquer tipo de cuidado (revisão em Machado & Raimundo, 2001) e, diferentemente de espécies com cuidado maternal ou paternal, poucas previsões podem ser feitas quanto às estratégias de acasalamento e ao dimorfismo sexual. Entretanto, a existência

de dimorfismo sexual e a variação nos sistemas de acasalamento podem ocorrer devido a diferenças na dispersão de recursos, que irão exercer influência sobre a dispersão das fêmeas e, portanto, sobre a economia de defesa de territórios por parte dos machos (Emlen & Oring, 1977). Quanto mais agregados no espaço estiverem as fêmeas ou outros recursos reprodutivos, tais como sítios de oviposição, maiores serão as oportunidades de monopolizá-los e, portanto, espera-se a evolução de estratégias poligínicas (Emlen & Oring, 1977). Portanto, a dispersão desses recursos reprodutivos (incluindo as fêmeas) deve ser o principal aspecto na determinação das estratégias de acasalamento de espécies de opiliões que não cuidam da prole (Machado & Macías-Ordóñez, 2007).

Não há nenhum estudo publicado abordando a descrição do comportamento de acasalamento em opiliões da subordem Laniatores que não apresentam cuidado parental, que devem corresponder à maioria das espécies no grupo (Machado & Raimundo, 2001). Entretanto, um estudo realizado com um representante da subordem Eupnoi, *Leiobunum vittatum* (Sclerosomatidae), uma espécie que não apresenta cuidado parental, mostrou que o acasalamento ocorre em rochas defendidas pelos machos e que o formato da rocha, bem como a presença ou ausência de musgos sobre ela, afetam a preferência dos indivíduos pelos territórios. O mecanismo sensorial dos opiliões permite que eles avaliem a topografia dos substratos por onde caminham, podendo assim reconhecer o formato e até mesmo saber se o ambiente possui condições adequadas de umidade. Rochas maiores normalmente são consideradas bons territórios, pois são mais facilmente encontradas pelas fêmeas, constituindo assim sítios preferenciais de acasalamento. Como seria esperado, os machos brigam entre si para defenderem rochas maiores e cobertas por musgos, que reúnem as características buscadas pelas fêmeas para depositarem seus ovos (Macías-Ordóñez, 1997, 2000; Machado & Macías-Ordóñez, 2007).

Modelo de estudo

Assim como as aranhas (o grupo mais bem estudado dentre os aracnídeos), os opiliões têm se mostrado organismos especialmente adequados como modelos para trabalhos comportamentais. A família Gonyleptidae, a segunda maior entre os opiliões, concentra a maioria dos estudos ecológicos e comportamentais realizados com espécies da ordem até o momento em regiões neotropicais (veja referências em Pinto-da-Rocha *et al.*, 2007). Muitas espécies dessa família foram usadas em manipulações experimentais diretamente no campo, permitindo testar hipóteses ecológicas de maneira refinada (e.g., Machado & Oliveira, 1998; 2002; Machado *et al.*, 2002, 2005; Buzatto *et al.*, 2007; Requena *et al.*, 2009). Outras espécies foram facilmente mantidas em cativeiro, onde executaram comportamentos similares aos observados no campo (e.g., Willemart & Chelini, 2007; Osses *et al.*, 2009; Willemart *et al.*, 2009; Nazareth & Machado, 2010). Dada a enorme variedade de formas de cuidado parental e estratégias de acasalamento nos Gonyleptidae, é provável que a seleção sexual tenha exercido um papel importante na evolução da enorme variedade de formas de dimorfismo sexual na família (Machado & Macías-Ordóñez, 2007).

O estudo de estratégias de acasalamento na ordem Opiliones só recebeu atenção recentemente (Machado & Macías-Ordóñez 2007). Todos os estudos, no entanto, focaram-se nas poucas, porém conspícuas, espécies que apresentam alguma forma de cuidado parental. Não existe nenhum estudo abordando as estratégias de acasalamento de espécies que não apresentam cuidado parental, como os representantes da subfamília Mitobatinae (Gonyleptidae). Entre as espécies dessa subfamília, o dimorfismo sexual ocorre no comprimento da perna IV, com machos apresentando segmentos, como o fêmur, cerca de três vezes mais longos do que as fêmeas (Kury, 1992). Observações de campo com *Longiperna zonata* e *Promitobates ornatus* indicam que essas pernas são usadas para desferir golpes em brigas entre machos. É possível, portanto, que o alongamento das pernas em espécies da subfamília tenha sido favorecido pela seleção sexual como armamento no combate entre

machos. Nesse sentido, as espécies da subfamília Mitobatinae são bons modelo de estudo para responder questões sobre a evolução do dimorfismo sexual e a função de características sexuais exageradas nos machos.

Objetivos

O objetivo geral desta dissertação é descrever aspectos básicos da biologia reprodutiva de duas espécies de opiliões da subfamília Mitobatinae, com enfoque nos seguintes tópicos: (a) função do dimorfismo sexual no comprimento das pernas, (b) determinação das suas estratégias de acasalamento e (c) significado das brigas entre os machos. A dissertação é composta por um capítulo apresentado na forma de um artigo formatado nos moldes da revista *Ethology*. O objetivo específico deste artigo é testar quatro previsões sobre a hipótese de que a seleção sexual está direcionando o alongamento do quarto par de pernas nos machos de *Longiperna zonata* e *Promitobates ornatus*: (1) o coeficiente alométrico do comprimento do fêmur IV em machos será maior do que o das fêmeas; (2) machos com pernas IV mais compridas vão residir próximos a grupos de fêmeas; (3) machos envolvidos em interações agressivas terão fêmures mais compridos do que o tamanho médio da população; (4) a diferença de tamanho de perna entre os oponentes será menor do que a diferença esperada entre pares de machos escolhidos ao acaso na população. Ao final da dissertação será apresentada uma conclusão geral na qual serão exploradas as implicações dos resultados obtidos aqui e serão apontados direcionamentos para estudos futuros.

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Sexual selection on male weaponry in two neotropical harvestmen (Arachnida: Opiliones)

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Introduction

In several species, males compete directly for the control of females, or indirectly by controlling the resources required by sexually receptive females (Emlen & Oring 1977). These situations lead to intense intrasexual selection among males, which can select for behavioral and morphological traits that enable males to be more competitive in contests for resources or females (Shuster & Wade 2003). Sexual selection on males that defend receptive females or resources that attract females is thought to have led to the evolution of elaborate male weaponry (Darwin 1871). In fact, males from several animal species exhibit sexually dimorphic structures that are primarily used as weapons in intrasexual contests (Andersson 1994). In arthropods, classical examples include the elongate mandibles, horns or antlers of certain beetles, the stalked eyes of diopsid flies, and the forceps of earwigs (references in Emlen & Nijhout 2000).

The allometric relationship between the sizes of a particular body part and the whole individual bearing that trait is an informative aspect of an organism's biology (Knell 2009). Positive allometry, meaning that larger individuals have disproportionately larger traits than smaller individuals, is usually defined as allometric values greater than 1.0 when scaled on body size (reviewed in Emlen & Nijhout 2000). Although sexual selection is generally thought to favor the evolution of strong positive trait allometries (Petrie 1988, 1992; Green 1992; Kodric-Brown et al. 2006), a recent study indicates that an optimal allometric pattern depends on the form of the function relating fitness to body size and trait size (Bonduriansky & Day 2003). In this way, positive allometry is expected to evolve only if the combined effect of sexual and viability selection on trait size and body size results in a greater relative advantage of increased trait size in larger individuals (Bonduriansky & Day 2003). However, regardless of the allometric value, sexually selected traits consistently exhibit significantly higher allometric slopes than nonsexually selected traits, supporting the notion that sexual

selection may elevate allometric slopes (Eberhard 2002; Kodric-Brown et al. 2006; Bonduriansky 2007).

Opiliones, commonly known as harvestmen, is the third largest order within the class Arachnida with more than 6,000 species (Machado et al. 2007). Sexual dimorphism in harvestmen is incredibly diverse (examples in Pinto-da-Rocha et al. 2007), suggesting that sexual selection may have played an important role in the evolution of morphological traits in the group. In some species from the Neotropical family Gonyleptidae, males have a strong fourth pair of legs, with several tubercles and spines on the coxa, trochanter, femur, and sometimes on the patella and tibia as well (Kury & Pinto-da-Rocha 2007). Males of the gladiator harvestman *Neosadocus maximus* (Gonyleptinae), for instance, use their spines and tubercles on femur and coxa IV in male-male fights (Willemart et al. 2009). Additionally, all structures directly involved in male-male fights in this species exhibit allometric coefficients significantly higher than nonsexually selected structures, reinforcing the notion that weaponry has evolved under intrasexual selection (Willemart et al. 2009). Apparently, *N. maximus* males fight for the ownership of territories with exclusive access to preferred oviposition sites, and thus to gravid females, much like other harvestmen (e.g. Macías-Ordóñez 1997, 2000; Buzatto & Machado 2008).

Males belonging to the subfamily Mitobatinae exhibit the most impressive form of sexual dimorphism in the family Gonyleptidae. They have an elongated fourth pair of legs that may reach 39 times the body length, as occurs in *Metamitobates squalidus* (Kaestner 1968). On the other hand, females have much shorter legs and are so different from the males that in *M. squalidus* each sex has been described as a different species (see Kury 1992). Although this extreme form of sexual dimorphism is known since the beginning of the last century, the function of the elongated fourth pair of legs in males, as well as all information on their mating system remains unknown. In this study, we tested four predictions to address the hypothesis that sexual selection is driving the enlargement of the fourth pair of

legs in males of two closely-related Mitobatinae harvestmen, *Longiperna zonata* and *Promitobates ornatus*.

The first prediction postulates that if the length of male fourth pair of legs is under sexual selection, the allometric value of this trait will be significantly higher than the third pair of walking legs and also higher than the allometric value of the females' fourth pair of legs because trait allometry is expected to increase with sexual selection intensity. The second prediction postulates that if larger males are more likely to monopolize access to females or be preferred by them, then adult males with longer legs IV will reside close to groups of adult females and this residency will be related to femur length. Our two last predictions specifically address the hypothesis that male-male competition is driving the enlargement of the fourth pair of legs. Given that body or weapon size predict fighting success (Arnott & Elwood 2009) and that larger males may exclude smaller males from the opportunity to fight for access to females (e.g. Williams 1978; Kelly 2006; Mendoza-Cuenca & Macías-Ordóñez 2010), the third prediction postulates that males observed in aggressive interactions will be larger than the population average size. Finally, the fourth prediction postulates that the size difference between males involved in contests will be smaller than the average difference expected between any two males in the population. Contests among similarly matched contenders are known to last much longer than contests among dissimilar contenders (reviewed in Arnott & Elwood 2009), which may be resolved even without fighting (e.g. Macías-Ordóñez 1997; Kelly 2006). We tested these four predictions by (1) comparing the allometric coefficients of legs III and IV between males and females, (2) relating male residency to the length of femur IV, (3) comparing the length of femur IV of males involved in contests with the rest of the male population and (4) comparing the size difference between contenders to the random difference expected among males in the population.

Methods

Study site

The study was conducted in an Atlantic Forest fragment at the Intervales State Park (24°14' S; 48°04' W; 800 m of altitude), located in the São Paulo state, southeastern Brazil. The region has high precipitation levels, with average annual rainfall varying between 2000 and 3000 mm/year, and mean annual temperature ranging from 12-20 °C. The weather is mainly subtropical, with a warm-wet season lasting from October to March and a cold-dry season from April to September. Naturalistic observations were done along a stream, which is approximately 5 m wide and is flanked by abundant vegetation that partially covers the stream bed in some places.

Studied species

Longiperna zonata and *Promitobates ornatus* (hereafter referred to only by the genus name) belong to the subfamily Mitobatinae, which comprises 11 genera and nearly 45 species (Kury 2003). Both species exhibit a marked sexual dimorphism, with males bearing a long and straight fourth pair of legs. The only information available about the reproductive biology of the Mitobatinae is that females of *P. ornatus* lay eggs individually on the substrate and cover them with debris - a possible strategy to camouflage the eggs and decrease dehydration (Willemart 2001). At the Intervales State Park, individuals of *Longiperna* and *Promitobates* are mainly found along the margins of streams, where they forage, reproduce and take shelter. Both species are very abundant and are found active during most part of the day during the wet-warm season.

Voucher specimens of both studied species were deposited in the arachnological collections of the Museu de Zoologia da Universidade de São Paulo (MZSP, São Paulo state).

Mark-recapture study

The reproductive behavior of *Longiperna* and *Promitobates* was studied by monthly inspecting a 170 m long transect along the river from October 2008 to April 2009. The vegetation flanking both sides of the river was inspected twice a day (from 14:00 to 18:00 h and from 20:30 to 01:30 h) during four consecutive days per month. At the time of the first capture, males and females were measured (see below) and individually marked with enamel paint applied to the dorsal scute (males and females), legs III (males and females), and/or legs IV (only females). This marking procedure has been widely used in studies with harvestmen because it does not affect the behavior of these organisms and last for more than two years (Buzatto et al. 2007). In each inspection, it was recorded sex and location along the transect (to the nearest 1 m) for each individual.

The mark-recapture data was used to estimate a residency index (RI) for all individuals in the transect, using the equation $\frac{N_m}{n} * 100$, where N_m is the number of times one individual was found in its modal recapture site (i.e., the point in the transect where this individual was recaptured more frequently), and n is the number of times each individual was recaptured during the study period. RI tends to zero when an individual was never found twice in the same place (i.e., $N_m = 1$), and may reach the value of 100%, when an individual was always found in the same place along the transect. Since many individuals of *Longiperna* ($n = 346$; 54.2% of the total) and *Promitobates* ($n = 228$; 83.8% of the total) were recaptured just a few times during the study period, RI was calculated only for individuals recaptured four times or more.

Data on residency was used in two different ways to test our second prediction, which states that larger males will monopolize reproductive territories and reside close to groups of adult females. First, male RI was correlated with femur IV length using a Pearson correlation test, and a positive correlation between these two variables was expected. Second, the

distribution of females was mapped in the transect using each female's modal recapture site. Females had high RI (see Results) and most of them were found more than 50% of times exactly in the same places, so that relatively stable female groups (or "harems", sensu Shuster & Wade 2003) were clearly identified in the transect. After detecting the harems, the distribution of the males was also mapped using their modal recapture sites. In some cases, however, more than one male was found to show a high residency value in the same harem at the same time. The criterion to assign harem ownership to a single male, the "resident" male, in these cases was to select the individual that was found more times in the harem (i.e., the one with a higher value of N_m). Finally, the influence of male size on his probability of holding a harem was tested using a logistic regression, in which the femur IV length was the independent variable and harem ownership (1) or not (0) was the dependent variable.

Morphometric measures

All captured individuals were measured to the nearest 0.01 mm using calipers for the length of the following structures: (a) dorsal scute, which is an indicator of body size, (b) right femur III, which is a structure not directly involved in male-male contests (see Results), and (c) right femur IV, the longer and stronger leg segment used as weapon in male-male contests (see Results). These measures were used in morphometric analyses to compare males and females of both species. First, t tests were done to compare dorsal scute length, femur III and femur IV length between males and females. To test our first prediction, which states that the allometric value of femur IV length in males will be significantly higher than walking legs III and also higher than the allometric value of the females' femur IV, the estimation of the slopes of bivariate lines between the \log_{10} of the dorsal scute length and the \log_{10} of the leg length (III and IV) was done using a standardized major axis estimation (Warton et al. 2006). Comparisons between the allometric coefficients of femur IV and femur III in males and between the allometric coefficients of femur IV in males and females were

done using the IC95% associated to the estimation of the slopes. Analyses were carried out using the “smatr” module version 2.1 (Warton 2005) of the R statistical package version 2.6.1.

Behavioral observations

Throughout the fieldwork, all relevant behavioral events, such as male fights, oviposition events, and copulations, were continuously recorded. At night, behavioral observations were done using a flashlight covered with a red filter to avoid disturbing the individuals. To increase sample size of male-male fights, two field trips exclusively devoted to behavioral observations were done in January and February 2009, lasting 10 and 15 days, respectively. These months were chosen because they correspond to the peak of the reproductive season for both harvestman species. In these two field trips, the study transect was searched from 09:00 to 02:00 h. For each fighting male, the following data were recorded (to the nearest 0.01 mm) in the field: (a) right and left femur IV length, (b) total right and left legs IV length, and (c) dorsal scute length. Fights were described by taking notes of the entire sequence of events ($n = 13$), based on the behavioral acts exhibited by the males (see Supplementary Material 1).

To test our third prediction, which states that individuals involved in aggressive interactions will have longer fourth pair of legs than the average population size, a null model approach was used (see the R codes in Supplementary Material 2). First, the mean femur IV length of the 26 males found fighting in the field was calculated. With the data on the femur IV length of all the males from the population measured during the study period, a randomization was done so that at each randomization 26 males were drawn from the total number of males ($n = 636$) without replacement. For each set of 26 males the mean length of their fourth pair of legs was calculated. This procedure was repeated 5.000 times and P value was estimated as the proportion of values generated by the null model that were equal or higher than the observed value. A similar null model approach was used to test our fourth

prediction, which states that the size difference between contenders will be smaller than the difference expected between randomly chosen pairs of males in the population. The mean difference in the femur IV length of the 13 pairs of males found fighting in the field was compared with the mean difference of a random sample of 13 pairs of males shuffled (without replacement) from the population. The randomization procedure was repeated 5.000 times and P value was estimated as the proportion of values generated by the null model that were equal or smaller than the observed value.

Results

Allometry

There is a clear sexual dimorphism in body size for both harvestman species regardless of leg length: female dorsal scute length is longer than male dorsal scute length in *Longiperna* ($t = -45.739$, $df = 1149$, $p < 0.001$, Table 1) and *Promitobates* ($t = 14.619$, $df = 631$, $p < 0.001$, Table 1). However, for both species male femur IV length is much longer than female femur IV length (*Longiperna*: $t = 59.785$, $df = 1169$, $p < 0.001$; *Promitobates*: $t = 65.586$, $df = 640$, $p < 0.001$, Table 1). Femur III is also longer in males than in females of *Longiperna* ($t = 63.125$, $df = 1171$, $p < 0.001$, Table 1) and *Promitobates* ($t = 72.349$, $df = 640$, $p < 0.001$, Table 1).

Allometric scaling also showed sex-specific patterns (Table 2; Fig. 1). The slope value of the bivariate line between dorsal scute length and femur IV length was significantly lower for females than for males of *Longiperna* (Figs 1b,d) and *Promitobates* (Figs 1f,h) (Table 2). Moreover, the slope value of the bivariate line between dorsal scute length and femur III was significantly lower than the slope of the bivariate line between dorsal scute length and femur IV in males of *Longiperna* (Figs 1a,b) and *Promitobates* (Figs 1e,f) (Table 2).

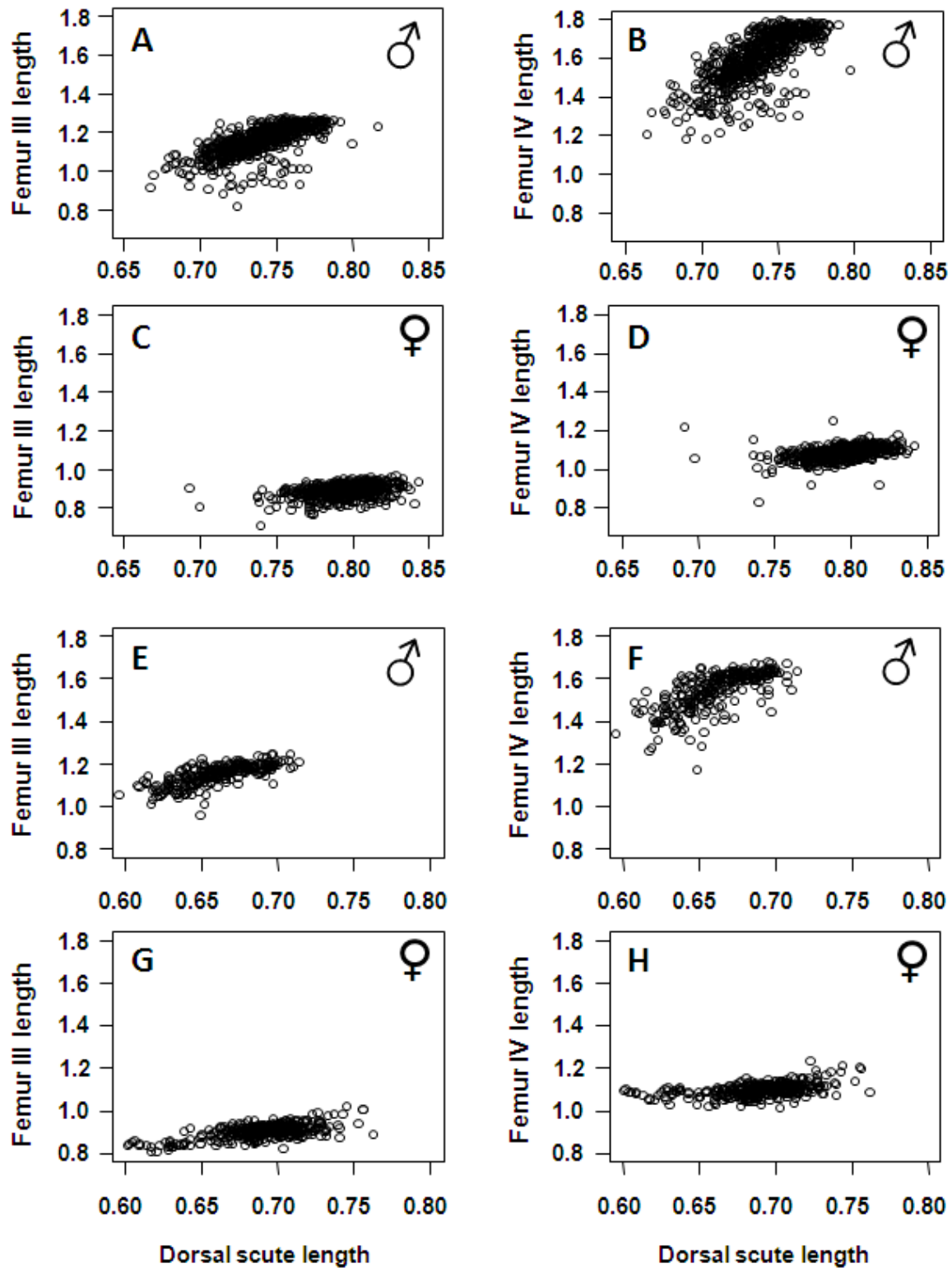


Fig. 1: Relationship between dorsal scute length (an indicator of body size) and the length of femurs III and IV for males and females of the harvestmen (a-d) *Longiperna zonata* and (e-h) *Promitobates ornatus*. All measures (in mm) were \log_{10} transformed.

Table 1: Mean and standard deviation of the size of three body parts (in mm) in males and females of the harvestmen *Longiperna zonata* and *Promitobates ornatus* — the coefficient of variation is presented in parentheses. Positive values for sexual dimorphism denote male-biased difference and negative values denote female-biased difference

Body parts	<i>Longiperna</i>			<i>Promitobates</i>		
	Male (n = 636)	Female (n = 535)	Sexual dimorphism	Male (n = 272)	Female (n = 370)	Sexual dimorphism
Dorsal scute length	5.49 ± 0.28 (0.05)	6.23 ± 0.27 (0.04)	- 11.87%	4.59 ± 0.24 (0.05)	4.91 ± 0.30 (0.06)	- 6.5%
Femur III length	14.39 ± 2.34 (0.16)	7.86 ± 0.51 (0.06)	+ 83.1%	14.15 ± 1.49 (0.10)	7.97 ± 0.59 (0.07)	+ 77.5%
Femur IV length	39.79 ± 10.68 (0.26)	12.08 ± 0.94 (0.07)	+ 229.4%	35.49 ± 6.67 (0.18)	12.48 ± 0.87 (0.07)	+ 184.4%

Table 2: Slope values of bivariate lines between the \log_{10} of the length of the dorsal scute and the \log_{10} of femurs III and IV assessed using standardized major axis estimation in males and females of the harvestmen *Longiperna zonata* and *Promitobates ornatus*. The numbers in parenthesis represent the inferior and the superior limits of the 95% confidence interval. The “R²” value is the coefficient of determination. The “p” represents significant differences (<0.05) from a slope value of 1

Structure	Males			Females		
	Slope	R ²	p	Slope	R ²	p
<i>Longiperna</i>						
Femur III length	3.378 (3.189–3.578)	90.15	<0.001	1.499 (1.392-1.613)	43.28	<0.001
Femur IV length	5.751 (5.452–6.067)	97.11	<0.001	1.797 (1.663-1.940)	56.48	<0.001
<i>Promitobates</i>						
Femur III length	2.031 (1.873- 2.201)	75.23	<0.001	1.192 (1.103-1.288)	22.74	<0.001
Femur IV length	3.840 (3.524-4.185)	92.80	<0.001	1.089 (0.994-1.194)	9.58	0.06

Male residency, oviposition sites, and female distribution

Females from both species were found laying eggs and/or manipulating eggs with their first pair of legs (*Longiperna*, n = 21, *Promitobates*, n = 10), apparently covering them with debris, since the first sampling months (October and November) (Figs 2a,b). Oviposition sites were stones partially covered with moss (only for *Longiperna*, n = 12), fallen rotten trunks (*Longiperna*, n = 6; *Promitobates*, n = 7), leaves of ferns (*Longiperna*, n = 3; *Promitobates*, n = 1) and stems of giant tree ferns (only for *Promitobates*, n = 2). Other eggs covered with debris and early hatched nymphs were always observed close to the oviposition sites (within 20-30 cm). Marked *Longiperna* and *Promitobates* females rarely moved and, during most of the sampling time, they were re-captured exactly at the same stone or trunk (*Longiperna*, n = 76 females, 43.2% of the total; *Promitobates*, n = 40 females, 42.6% of the total).

Adult males of *Longiperna* were frequently found at the oviposition sites, sometimes copulating with females (n = 4). However, male residency in this species (median = 50.0%, range = 0-100%, n = 292) was lower than female residency (median = 90.0%, range = 33.3-100%, n = 176) (Mann-Whitney, U = 8,211.0, p < 0.001). Adult *Promitobates* males were also commonly found at the oviposition sites, but were observed copulating with females (n = 2) less frequently than *Longiperna*. Male residency in this species (median = 57.1%, range = 20-100%, n = 44) was also lower than female residency (median = 85.7%, range = 40-100%, n = 94) (Mann-Whitney, U = 862.5, p < 0.001). For both species, there was a positive correlation between femur IV length and male residency (Figs 3a,b). Moreover, the longer the femur IV length, the higher the chance of a male being the owner of a harem (Figs 3c,d).

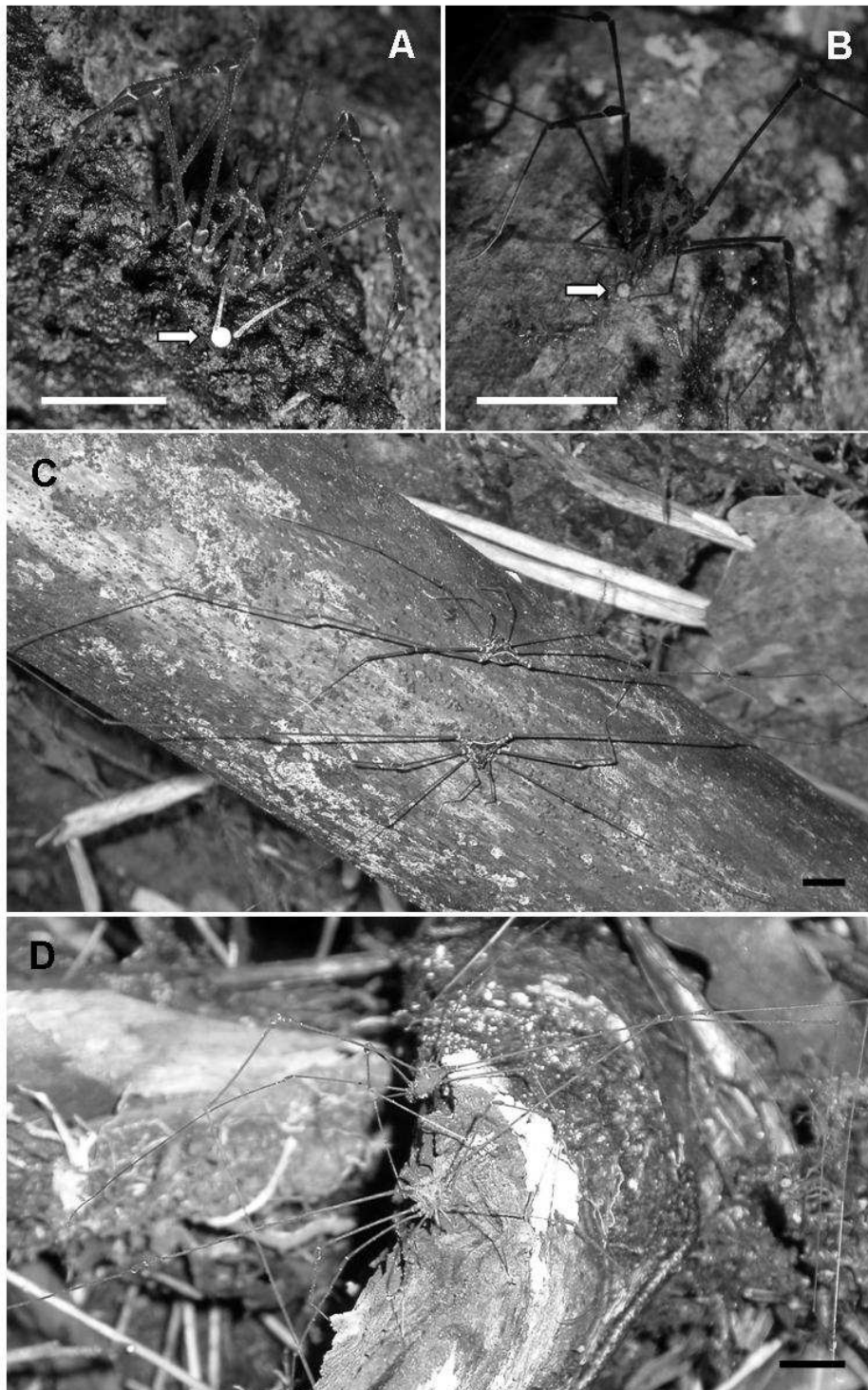


Fig. 2: Females of the harvestmen (a) *Longiperna zonata* and (b) *Promitobates ornatus* picking up debris and attaching particles to a recently laid egg (white arrow). In both photos the substrate is the bark of a rotten log. (c) Males of *L. zonata* fighting on a fallen trunk. Both males are exhibiting the “splits position”, in which they keep the fourth pair of legs widely opened, with right femur forming a 180° angle with the left femur. (d) Males of *P. ornatus* fighting in the same way on a fallen trunk. Scale bars in all photos = 1 cm.

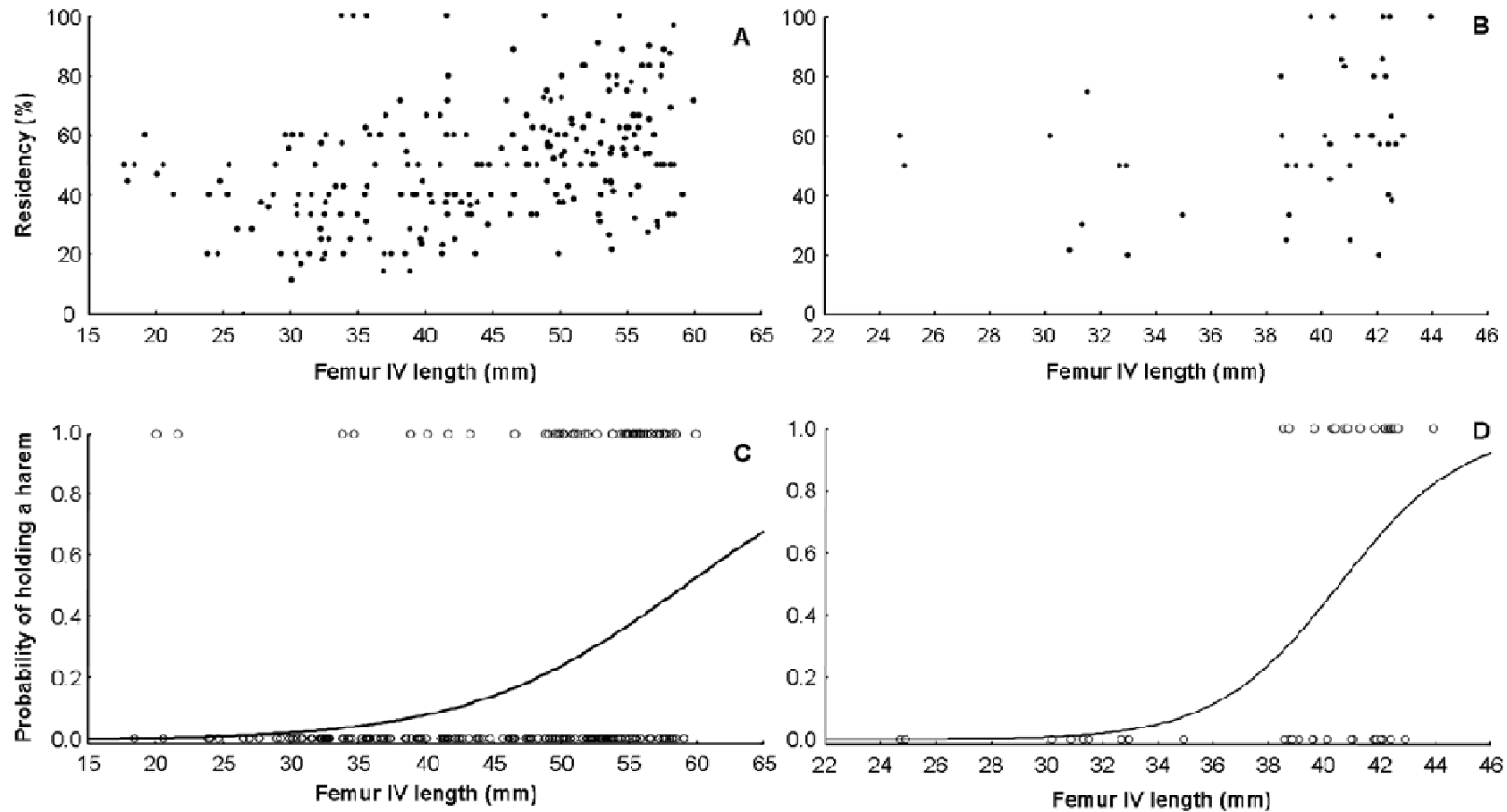


Fig. 3: Correlation between femur IV length and residency for the harvestmen (a) *Longiperna zonata* ($r = 0.34$, $n = 234$, $p < 0.001$) and (b) *Promitobates ornatus* ($r = 0.31$, $n = 44$, $p < 0.001$). Logistic regression showing how the probability of holding a harem increases with femur IV length for males of (a) *L. zonata* ($\chi^2 = 44.281$, $df = 1$, $p < 0.001$) and (b) *P. ornatus* ($\chi^2 = 14.864$, $df = 1$, $p < 0.001$).

Fighting behavior

Over the period devoted to behavioral observations, 13 fights were recorded for *Longiperna* and four for *Promitobates*. Despite the smaller number of observed fights of *Promitobates*, the general behavioral pattern is similar to that of *Longiperna* (see Supplementary Material 1). For both species, males were considered to be involved in a fight when the two individuals turned their backs to each other, within 20 cm, and with one of the males exhibiting a behavioral act called “splits”. A split occurs when males keep the fourth pair of legs widely opened, with the right femur forming a 100-180° angle with the left femur (Figs 2c,d). In the split position each male strikes the opponent in both metatarsi of legs IV using their own metatarsi IV. The loser is the male that clearly walks away from the other contender, whereas the winner is the male remains in the place where the fight occurred (see Supplementary Material 1).

The duration of *Longiperna* fights varied from 1 to 90 min (23.8 ± 29.1 min, $n = 13$). Conspecific females within 50 cm of the fight were recorded in only two fights. The mean femur IV length of the 26 *Longiperna* males involved in fights (49.10 ± 6.14 mm) is significantly larger than the mean of random samples of 26 males from the population (41.2 ± 10.7 mm, $p < 0.001$). Moreover, the mean difference in the femur IV length of males involved in fights (3.5 ± 2.5 mm) is significantly smaller than the mean difference of random samples of 26 males from the population (12.2 ± 2.4 mm, $p < 0.001$).

Discussion

Allometry

Femur IV in *Longiperna* and *Promitobates* is sexually dimorphic and the fourth pair of legs is likely to be related to the ability to hit the opponent in intrasexual combats (see Fighting behavior below). Moreover, in accordance with our first prediction, the slope of the bivariate line between dorsal scute length and femur IV length was significantly higher for males than for females in both species. Curiously, the allometric coefficient for the femur IV length was also significantly higher than 1 for *Longiperna* females and nearly significant in *Promitobates* females, none of which have been observed to fight. In some cases, when selection is practiced only in one sex, the focal character may respond in both sexes because selection in one sex does not limit the expression of the genes to that sex (Shuster & Wade 2003). Thus, femur IV of the unselected *Longiperna* females may increase in length as a result of the selection for increased leg length in males. In this case, the genetic correlation across the sexes is positive, and the direct selection on one sex results in a correlated response in the homologous trait in the other due to indirect selection (e.g., Petri 1988; Eberhard 2002; Kelly 2005). This scenario is expected to occur only when the costs of having a slightly longer fourth pair of legs is not high for females (Bonduriansky & Rowe 2005), and detailed analyses are currently in progress to test this prediction (see below).

Although male legs III are not directly used in combats, femur III length also showed a hyperallometric relationship with body size, and the slope of the bivariate line between dorsal scute length and femur III length was significantly higher for males than for females in both species. Nonsexual traits generally scale with negative allometry or isometry (review in Bonduriansky 2007), but positive allometry may be relatively common in locomotory structures (such as the legs III in harvestmen) as a result of biomechanical factors (Tseng &

Rowe 1999). Alternatively, direct selection favoring an increase in the length of legs IV may also result in indirect selection on the length of legs III because all appendices in arthropods are serial homologous (Akam 1998) and some degree of positive genetic correlation between the length of legs III and IV is expected, as occurs between the forewing and hindwing of butterflies (Frankino et al. 2007). This hypothesis can also explain why the length of female leg III also exhibited a hyperallometric relationship with body size, even though females do not fight.

According to Eberhard (2002), “there is nothing profoundly significant about the allometric value 1.0” because the balance between costs and benefits of especially large structures probably varies both among different structures and among different species (see also Bonduriansky 2007). Instead of emphasizing absolute allometric values, it seems more useful to use comparative values for different structures and sexes of the same species (Eberhard 2002). Thus, even though the allometric values for the length of males’ femur III length and also for females’ femurs III and IV in both species studied here are consistently higher than 1.0, the allometric value of the males’ femur IV is significantly higher than those of other traits. The ‘allocation trade-off model’ predicts that all else being equal, positive allometry is most likely to evolve when: (1) an increase in relative trait size yields greater mating success benefits for large individuals than for small ones, and (2) the viability costs of trait exaggeration are strongly dependent on body size (Bonduriansky & Day 2003; Bonduriansky 2007). In this study we demonstrate that males with long legs IV are more likely to hold a harem and probably experience higher reproductive success than males with short legs IV. Certainly, more behavioral information is needed to understand how the expression of relatively larger traits influences the mating success in *Longiperna* and *Promitobates*. Our next step, however, is to determine the precise form of selection acting on the length of legs IV estimating survival rates for individuals of different sizes. In this way, our mark-recapture data may add reliable information on the viability costs of the target trait

and provide an empirical test of the second prediction of the model. Harvestmen are an especially interesting model because, contrary to most arthropod species that exhibit strong positive allometry, their allometric sexual traits are not ‘dedicated’ secondary sexual structures (i.e., traits that serve sexual functions only) and certainly have viability-related functions (see discussion in Bonduriansky 2007).

Male residency, oviposition sites, and female distribution

Although our study was not aimed to have a formal description of the mating system, our field data strongly suggest that the mating strategy adopted by at least some males from the studied population is a resource defense polygyny. In resource defense polygynies, males should be associated with a limiting resource used by females (such as oviposition sites), and should also have the same association with this limiting resource both in the presence and absence of breeding females. Consequently, if males fight for resources or territories, these fights should occur regardless of female presence (Ostfeld 1987). In fact, several males of both harvestman species were regularly recaptured in sites without females, and most *Longiperna* fights occurred without any female around the fighting males. Females of both species were highly phylopatric, being found in the same places used as oviposition sites, such as rocks and fallen trunks. The small groups of females associated to an oviposition site were monopolized by the largest males in the population, i.e., those bearing the longer fourth pair of legs. Thus, in accordance to our second prediction, length of males’ femur IV seems to be related to their resource holding power, such as the length of males’ legs II in the harvestman *Acutisoma proximum*, which use this long pair of legs to strike the opponent in territorial fights (Buzatto & Machado 2008).

One result of intense competition among males for mates is selection for alternative reproductive tactics in which two male phenotypes exhibit different ways of obtaining mates (Shuster & Wade 2003). The production of two or more distinct phenotypes can result in a discontinuous or multimodal distribution of characters such as weapon size (reviewed by Oliveira et al. 2008). Even though there was a great variation in the length of males' femur IV (both for *Longiperna* and *Promitobates*), apparently there is no bimodality in the distribution of this trait, and in the scatter plot showing the relationship between dorsal scute length and femur IV length it is not possible to distinguish two clear clusters of points, as should be expected in the presence of two male morphs (Figs 1b,f). Even if there were two mating strategies in these populations, they would not seem to be directly related to the existence of two male morphs in terms of the size of their weapons, as have been reported for many other arthropod species, including insects (Brockmann 2008), crustaceans (Shuster 2008), and arachnids (Vanacker et al. 2004; Radwan et al. 2003). Although many cases of alternative reproductive strategies are correlated with distinct morphological differences between male morphs, there are other cases in which no obvious differences exist (Brockmann 2008). For both species studied here, males with short femur IV had a lower residency and exhibited longer-range movements along the river margins (unpublished data). It is possible, therefore, that small males rely on a mating strategy based on furtively invading large males' harems where they sneak copulations with females, but the presence of this strategy in *Longiperna* and *Promitobates* needs to be tested in the field.

Fighting behavior

Here we demonstrated that the sexually dimorphic fourth pair of legs of two Mitobatinae species, *Longiperna* and *Promitobates*, is used as a weapon in male-male fights. Much as in the case of the gladiator harvestman *Neosadocus maximus* (Willemart et al. 2009),

fighting males turn their backs to each other, but they do not intertwine their fourth pair of legs, probably because the femur IV is long and lacks any spine or tubercle to hook on the legs of the contenders. Once males are back to back, they widely open their forth pair of legs and strike each other using these legs. Males of the harvestman *A. proximum* also use sexually dimorphic legs (in this case, the long second pair) to strike the opponent, but the fighting males remain in a front to front position (see Fig. 3a in Buzatto & Machado 2008). Because the sexual dimorphism in the length of the fourth pair of legs is a synapomorphy of the subfamily Mitobatinae (Kury 1991), it is likely that many other species from the group also exhibit similar fighting behavior.

In accordance to our third and fourth predictions, those males of *Longiperna* observed fighting had longer femur IV, and contenders were more closely matched in femur IV length than would be expected by chance if all possible pairs of males in the population had the same chance of being observed fighting. There are two possible explanations for the observed patterns, and they do not exclude each other. Smaller males may be avoiding fights altogether and following an alternative strategy to achieve matings, thus only larger males would be fighting. In such case, contenders would be closely matched since no small males would be involved. Alternatively, fights involving smaller males may last much less than fights involving larger males, in which case the probability of observing those fights would be much lower. If so, our results would be a consequence of a sample bias against short fights involving small males.

The “war of attrition” model predicts that contenders may be willing to fight for a period directly related to their own size (or any trait related to RHP), in which case the size of the smallest contender would be the best predictor of fight duration. This model also predicts long contests of endurance, with relatively low levels of escalation (Taylor & Elwood 2003; Arnott & Elwood 2009). A competing model is the “sequential assessment” (Enquist & Leimar 1983), which assumes that contenders sample each other’s RHP, and

predicts that fight duration will be inversely related to size differences between contenders since a smaller difference would require a longer contest in order for one of them to assess its disadvantage and give up. This model also predicts a sequence of stereotyped behavioral patterns and increasing escalation. If all *Longiperna* males in the population were fighting regardless of size, we may expect smaller size differences than expected by chance among fighting males only if fights between similar sized contenders were longer and our chances of finding them were higher. However, we would find males of all sizes fighting each other, although closely matched between them. However, all males observed fighting were clearly larger than the average, thus if our results are due to a sample bias in terms of fight duration, this would support a “war of attrition” mechanism of fight resolution. Furthermore, the stereotyped sequence of increased escalation predicted by the sequential assessment model was not observed, but rather a relatively unescalated interaction among contenders.

Another factor to consider is a resident-intruder role. It is well established that prior residence or ownership results in a higher chance of winning a contest regardless of size or any other difference between contenders (Kokko et al. 2006). Among other mechanisms suggested, owners may be willing to fight for longer, and residents may respect ownership and give up sooner when facing an owner; such a mixed strategy has long been shown to be evolutionary stable under some circumstances (Maynard-Smith & Price 1973). If large *Longiperna* males are better able to monopolize females or the resources needed by these females, territory owners will be larger on average than intruders. In such case, the usually smaller intruders may give up the fight even sooner if they follow a role-respecting fighting strategy, thus such fights would be even shorter than predicted by the “war of attrition” model. Only large intruders would engage in fights against the usually large residents. These fights would be much longer, either because they need longer time to assess their differences, or because the smallest contender is still relatively large.

Concluding remarks

Our results suggest that male *Longiperna*, and probably *Promitobates*, are subject to intrasexual selection on traits such as the length of femur IV, which enable them to better defend spots where females are more abundant, probably areas with adequate oviposition sites. The study of mating strategies in the order Opiliones has only recently been the subject of some attention (Machado & Macías-Ordóñez 2007). Most of these studies, however, have focused on the few but more conspicuous species showing some form of parental care, which plays an important role in the evolution of mating strategies. With very few exceptions, the mating strategies of harvestmen in the absence of parental care have been barely studied. This may be due to the fact that, in these species, the actual resource limiting female reproduction, and thus driving male mating strategies, is not as evident. Identifying such resources in species such as *Longiperna* and *Promitobates* may be the key to understand selective forces acting on these males, ultimately driving extreme sexual dimorphism.

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Supplementary Material 1

Definition of the behavioral acts accomplished by males of the harvestman *Longiperna zonata* during intrasexual fights. Behavioral acts marked with an asterisk were also recorded in fights of *Promitobates ornatus*.

Act	Definition
1. Standing still*	To remain in the same place supported by legs III and IV (sometimes also supported by legs I).
2. Standing still with forward legs in movement (FLM)*	As described in 1, but displaying alternating or simultaneous movements of legs I and/or II, waving in the air or tapping the substrate, an opponent or a female.
3. Walking*	To change location supported by legs III and IV (sometimes also supported by legs I). The movement may be forward, lateral or backwards.
4. Walking with FLM	As described in 3, but displaying FLM.
5. Splits*	To keep the fourth pair of legs widely opened, with right femur forming a 180° angle with the left femur. At this position, males are supported by legs I and III, with the posterior part of the body raised, kept at 30°-70° in relation to the substrate. Males exhibiting “splits” may be either standing still or walking backwards. Moreover, fighting males exhibiting “splits” may be in contact or not.
6. Half-splits*	As described in 5, but with legs IV opened in a 90°-170° angle.
7. Splits with FLM	As described in 5, but displaying FLM.
8. Aggressive splits*	As described in 5, distance between opponents ranges to 1-4 cm and both have their backs turned to each other, and to strike the opponent in metatarsus of leg IV. Usually males display an up and down movement, much like the <i>bobbing</i> of <i>Eupnoi</i> .
9. Touched half-splits	As described in 6, but the legs IV of both opponents are in contact. The right leg IV of one male touches the left leg IV of the other male and vice versa.

Continued.

Act	Definition
10. Touched half-splits with FLM	As described in 9, but displaying FLM.
11. Bite*	To use the pedipalps and/or the chelicerae to grasp some part of the opponent's body. In our observations only the IV legs of a male were bitten.
12. Mutual bite*	To use the pedipalps and/or the chelicerae to grasp the pedipalps and/or the chelicerae of the opponent.
13. Fleeing*	When one male clearly walks away from the contender. This behavior may define the end of the fight, the winner remains and the loser flees.
14. Mutual fleeing	When both males walk to opposite sides and leave the place where the fight occurred. This behavior may also define the end of the fight, but no winner emerges.

Supplementary Material 2

R code for null models constructed to evaluate the probability of observing the recorded mean average leg IV length of males involved in fights and the mean leg IV length difference between fighting male pairs. Each command includes an explanation following the “#” sign.

```
#####These lines read the spreadsheet file "population.csv", which is made of a single  
column with the leg length of all males in the population, named "leglength" on the  
first row
```

```
DATA <- read.table("population.csv", header = TRUE, sep = ",")  
attach(DATA)
```

```
#####These lines create empty numeric objects
```

```
avgleg <-numeric() #a vector that will store 5000 averages of 26 leg length values, one for  
each time 26 values are randomly sampled from the variable "leglength"
```

```
avglegdif <- numeric() #a vector that will store 5000 averages of leg length difference values  
between members of 13 randomly formed pairs
```

```
pair<- numeric() #a vector that will store the 13 leg differences in each replicate
```

```
countleg <- 0 #a counter of the number of times that the average leg length of 26 randomly  
sampled males is larger than the observed average of fighting males
```

```
countlegdif <- 0 #a counter of the number of times that the average leg length difference of  
13 randomly sampled pairs is smaller than the observed average of fighting male pairs
```

```

####These lines generate 5000 random samples and compare them with observed values

for(i in 1:5000) { # this first loop generates 5000 replicates, "i" stores the replicate number
  sample26<-sample(leglength, 26, replace = FALSE, prob = NULL) #this line samples 26
    values from the variable "leglength" and stores it in the object "sample26"
  avgleg[i] <- mean(sample26) #this lines stores the average leg length of the replicate i and
    stores it in the previously created vector "avgleg" in the ith position
  countleg<-countleg + ifelse(avgleg[i]>49.08,1,0) #"countleg" increases its value by one unit if
    the average leg length of the ith replicate is larger than the average leg length of those
    males observed fighting (49.08), otherwise its value remains the same
  for(j in 1:13){ #this second loop (nested in the first loop) generates the leg length differences
    between 13 value pairs from the 26 values sampled in the ith replicate
    pair[j]<-abs(sample26[j]-sample26[j+13]) #the 13 differences are stored in the previously
      created vector "pair"
    } # end of the second loop
  avglegdif[i] <- mean(pair) #this lines stores the average leg length difference of the replicate i
    and stores it in the previously created vector "avglegdif" in the ith position
  countlegdif<-countlegdif + ifelse(avglegdif[i]<3.53,1,0) #"countlegdif" increases its value by
    one unit if the average leg length difference of the ith replicate is smaller than the
    average leg length difference of those male pairs observed fighting (3.53), otherwise its
    value remains the same
  } #end of the first loop

```

```
#### These lines provide the mean and standard deviation of random samples
```

```
mean(avgleg) #mean random leg length
```

```
sd(avgleg) #random leg length standard deviation
```

```
mean(avglegdif) #mean random leg length difference
```

```
sd(avglegdif) #random leg length difference standard deviation
```

```
####These lines estimate the probability that the observed values may be randomly  
observed
```

```
pleg<-countleg/5000 #the number of times (out of 5000) that the random sample of leg  
lengths was larger than the average leg length of males observed fighting, divided by  
5000, is stored in "pleg"
```

```
pleg #p value of observed leg length of fighting males to be randomly obtained from the  
population
```

```
plegdif<-countlegdif/5000 #the number of times (out of 5000) that the random sample of leg  
length differences was smaller than the average leg length difference of male pairs  
observed fighting, divided by 5000, is stored in "plegdif"
```

```
plegdif #p value of observed leg length differences of fighting male pairs to be randomly  
obtained from the population
```


CONCLUSÃO GERAL

Os resultados obtidos nesta dissertação indicam que o comprimento do fêmur IV em machos de duas espécies da subfamília Mitobatinae está sujeito à seleção intrasexual, que favorece o alongamento do quarto par de pernas. Os dados morfológicos indicam claramente que, assim como seria esperado pela teoria, o coeficiente alométrico do fêmur IV é significativamente maior em machos do que em fêmeas, tanto em *Longiperna* quanto em *Promitobates*. Mais informações comportamentais são necessárias para entender como a expressão de pernas desproporcionalmente mais longas em relação ao corpo influencia o sucesso de acasalamento em ambas as espécies. Nosso próximo passo, entretanto, é determinar se a seleção natural age negativamente sobre o comprimento da perna IV por meio de estimativas de sobrevivência de indivíduos de tamanhos diferentes. Nesse sentido, nossos dados de marcação-recaptura podem prover informações valiosas sobre os custos de sobrevivência impostos pelo comprimento da perna IV, tanto nos machos quanto nas fêmeas.

O comprimento do fêmur IV está positivamente relacionado à chance de um macho ser encontrado próximo a um grupo de fêmeas em *Longiperna* e *Promitobates*. Machos com pernas IV mais longas devem possuir maior capacidade de defender pontos onde as fêmeas ovígeras são mais abundantes — provavelmente áreas com sítios de oviposição adequados. O sistema de acasalamento parece, portanto, ser baseado em uma poliginia por defesa de recursos. Em outras espécies de opiliões territoriais, o tamanho do armamento dos machos está positivamente relacionado com a qualidade do sítio reprodutivo e com o número de fêmeas dentro do harém. Estudos futuros devem, portanto, identificar variações na qualidade dos recursos reprodutivos usados por *Longiperna* e *Promitobates* a fim de entender como essas variações influenciam o sucesso reprodutivo dos machos.

A média do comprimento do fêmur IV dos machos de *Longiperna* envolvidos em brigas é significativamente maior que a de machos selecionados ao acaso da população. Adicionalmente, a média da diferença de tamanho entre pares de machos que brigaram também é significativamente menor que a média da diferença de tamanho de perna entre pares de machos selecionados ao acaso na população. Existem duas possíveis explicações não mutuamente excludentes para esses padrões. Machos pequenos poderiam evitar brigas adotando uma tática reprodutiva alternativa para obter cópulas, e somente machos grandes seriam vistos brigando. Alternativamente, brigas envolvendo machos pequenos poderiam durar muito pouco tempo e a probabilidade de observar essas brigas no campo seria muito mais baixa. Nossos dados, portanto, abrem portas para estudos comportamentais focados exclusivamente nas brigas a fim de entender os fatores que influenciam o resultado das brigas em machos da subfamília Mitobatinae. Além disso, a possibilidade de existência de táticas reprodutivas alternativas em *Longiperna* faz da espécie um modelo adequado para investigar questões sobre intensidade de seleção intrasexual e oportunidade de seleção em opiliões.