

Male morphotypes in the Andean river shrimp *Cryphiops caementarius* (Decapoda: Caridea): morphology, coloration and injuries

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Abstract

In many species, different male morphotypes usually employ different tactics to access resources. Males with highly developed weapons are expected to fight and possibly incur higher levels of injury than males with less developed weapons, which usually avoid agonistic encounters. Discrete male morphotypes, where some males are very large and feature powerful chelae, have been reported for several large shrimp species, where males show a lobster-like monopolization of resources. During competitive interactions, these large males fight more vigorously than small males and, consequently, it is expected that they accumulate more injuries. Herein, we identified different morphotypes in the river shrimp *Cryphiops caementarius*, and we compared the percentage of body damage between large and small shrimps. We measured 26 morphometric data and 6 intensities of color on the chelipeds. Multivariate analysis based on a combination of morphometric and color data confirmed that there are two morphotypes. The 'dominant' morph is characterized by stout teeth on the cutting edges and by dark blue color on the external surface of the major cheliped. The two morphotypes can be distinguished based on the differences in the allometric relationships between several morphological traits and carapace length. Males from the large morphotype had a higher percentage of injuries on their chelipeds than other males. These results suggest that males from the large morphotype are dominant and compete aggressively for access to resources, a hypothesis to be tested in future, behavioral studies.

Introduction

In males of many species, different morphotypes can be distinguished, which often exhibit alternative reproductive tactics (Oliveira, Taborsky & Brockmann, 2008). Morphotypes with weapons or large body size usually fight for access to resources (e.g. reproductive females), while less developed morphotypes employ sneaking tactics. One of the main costs that fighting morphotypes incur during reproductive activities are injuries from agonistic encounters, as shown, for example, for insects (Siva-Jothy, 1987; Moore *et al.*, 2009), reptiles (Lailvaux *et al.*, 2004) and mammals (Mainguy *et al.*, 2008). More interestingly, in territorial fish, subordinate males often incur higher levels of injury than dominant territory holders (e.g. Ros *et al.*, 2006). In general, it is expected that in species where dominant morphotypes use powerful weapons to accede to resources, subordinates should avoid potentially costly fights and thus feature lower levels of injuries than dominants themselves.

A large number of crustacean species (many of them of commercial interest) show allometric growth of weapon struc-

tures, such as chelae in males (e.g. Jones, 1980; Nagamine & Knight, 1980; Mariappan & Balasundaram, 2004). Some of the more striking examples for allometric growth of chelae are found in the caridean shrimps, for example, in river shrimps from the genus *Macrobrachium* (Karplus, Malecha & Sagi, 2000; da Silva *et al.*, 2009) or in marine shrimps from the family Rhynchocinetidae (e.g. Thiel, Chak & Dumont, 2010).

Allometric growth of weapons (chelae) has been extensively studied in males of the giant Malaysian river shrimp *Macrobrachium rosenbergii*. Differential growth rates observed among adult individuals result in several types of morphologically distinguishable males (Ra'anan & Cohen, 1985; Kuris *et al.*, 1987; Ranjeet & Kurup, 2002). Cohen, Ra'anan & Brody (1981) distinguished three morphotypes of male *M. rosenbergii* based on the color of the chelipeds, relative size within the population and the length/height relationship of the cheliped. The 'small male' (SM) morphotypes of *M. rosenbergii* have smaller chelipeds compared with the other two morphotypes, which are termed 'orange claw' (OC) and 'blue claw' (BC) (Karplus *et al.*, 2000). Besides color and size, the BC morphotype is characterized by long and robust spines

whose orientation is different than in the other two morphotypes. However, the best morphometric discriminator between BC and OC was the relationship between carpus length and carapace length, followed by the relationship between propodus length and carapace length. By tracking individuals during ontogeny, Kuris *et al.* (1987) showed that the transition from SM to OC is gradual, while the change from OC to BC is dramatic and occurs in a single metamorphic molt. In other species, changes between morphotypes are less discrete and the identification of specific morphotypes requires multiple criteria (e.g. Correa *et al.*, 2000; Moraes-Riodades & Valenti, 2004; Thiel *et al.*, 2010).

The male morphotypes differ not only in morphology but also in behavior, especially during reproductive interactions. While the larger morphotypes with highly developed chelipeds usually fight for access to female-mating partners, which they defend before and during the copula, the smaller morphotypes are much less aggressive and attempt to gain access to females by sneaking tactics (Ra'anan & Sagi, 1985; Shuster, 1992; Correa *et al.*, 2003). Fights might cause chelae loss or damage, especially among the larger males. Indeed, high incidence of puncture wounds on chelae (Jones, 1980) or even lost chelae has been reported for large individuals in several littoral crab species (Smith & Hines, 1991; Abello *et al.*, 1994). Because dominant males of river shrimps frequently fight over females, it can be expected that they suffer higher degrees of injuries than SMs, which are rarely involved in fights (Ra'anan & Sagi, 1985).

One of the species for which large and possibly dominant males have been mentioned in the literature is the amphidromous river shrimp *Cryphiops caementarius* (Molina, 1782) (Viacava, Aitken & Llanos, 1978; Andrade, Valencia & 19 co-authors, 1998). Viacava *et al.* (1978) even distinguished a particular male morphotype of SMs with highly developed chelipeds for populations in Peru. However, no formal description of male morphotypes is available (Bahamonde & Vila, 1971). Apparently, ontogenetic growth and development of chelipeds and other structures are gradual, thereby complicating the identification of discrete morphotypes as known from *M. rosenbergii* and other species. There is also pronounced sexual dimorphism in *C. caementarius*, with males reaching a larger body size (up to 20 cm) and very much larger chelipeds than females (Hartmann, 1958; Viacava *et al.*, 1978; Andrade *et al.*, 1998). In natural populations of *C. caementarius*, there is a wide variation in the size composition of males (Bahamonde & Vila, 1971), and given the description of very large males (Hartmann, 1958; Bahamonde & Vila, 1971; Viacava *et al.*, 1978; Andrade *et al.*, 1998), it could be expected to find different male morphotypes in *C. caementarius*.

Based on the above considerations, herein we examine the morphometrics of male *C. caementarius*. We expected to distinguish at least two discrete morphotypes, albeit discrimination of these probably requires the incorporation of more morphological traits than needed for the differentiation of male morphotypes in *M. rosenbergii*. Therefore, a wide range of body structures (including color patterns) was measured and quantified for male *C. caementarius*. Following the dis-

tingtion of morphotypes, we also examined the frequency of injuries to test the hypothesis that males from the large, more developed morphotype suffer a higher degree of damage than males from the other morphotype(s).

Materials and methods

Collection of specimens

Cryphiops caementarius inhabits the mountain rivers on the western slopes of the Andes from Peru to central Chile (from 6°S to 33°S). The species has considerable economic importance, and its conservation status has been identified as 'vulnerable to critically endangered' (Jara, Rudolph & González, 2006). Reduction of natural populations due to overexploitation and contamination has led to recent efforts in repopulation and small-scale aquaculture (Meruane *et al.*, 2006).

Shrimps were collected in the Choapa river (IV. Region of Coquimbo, Chile; 31°35'11''S, 71°28'38''W) from November 2010 to March 2011. These months are characterized by high reproductive activity (Bahamonde & Vila, 1971), as confirmed by a high proportion of ovigerous females during the samplings (pers. obs.). Water temperatures during night collecting were ~20°C. Water depth at the collection site ranged from about 0.5 to 1 m, with a high flow of water. The river bed at the collection site was characterized by gravel and medium-sized rocks.

Sampling was done at night by snorkeling or walking along the river edge, and scooping up shrimps with a dip net or catching them by hand. Shrimps were transported to the laboratory in large coolers with sponges that were soaked with river water and surrounded by ice packs. This transport method guarantees high survival and eliminates the risk of agonistic interactions. Shrimps were held in large freshwater tanks with shelters; no lost appendages were found in the tanks during the first days of laboratory maintenance. We collected a total of 119 males of *C. caementarius*, which were photographed within 1 or 2 days after arrival in the laboratory.

Measurements of shrimps

Photographs were taken of living individuals, with dorsal view of the whole body and interior and exterior views of the two main chelipeds (pereopod 2). All measurements were made from these photographs using image analysis software IMAGE-PRO PLUS version 6.0 (Media Cybernetics, Bethesda, MD, USA). For each of the 106 fully intact specimens, morphometric measurements were taken on 26 body and 6 color variables. For 13 additional males, only specific measurements were taken (length of carapace, length of carpus, width of propodus and length of segment 2 on the major cheliped) because these individuals had some appendages broken or missing. Following the photographic records, males were returned to large freshwater tanks where they were maintained for other studies.

Morphometric measurements

General body measurements were taken in dorsal view with the animal fully extended (Fig. 1a, Table 1). The measure-

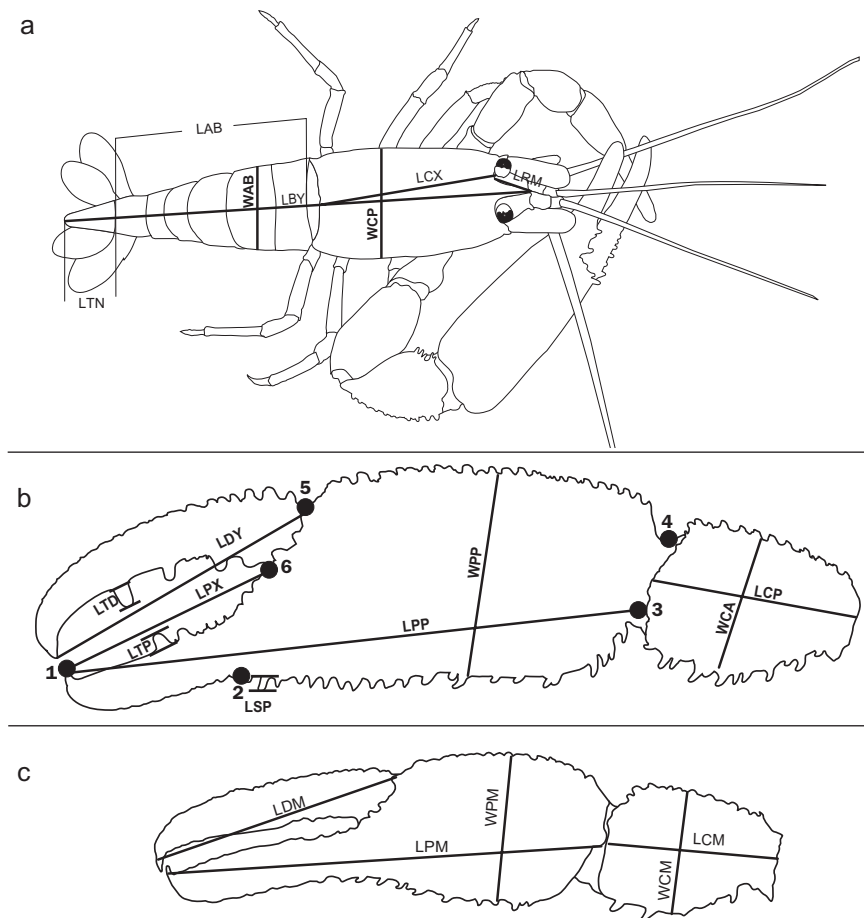


Figure 1 Morphometric measurements taken from each individual. (a) General body measurements. (b) Major cheliped measurements, black dots indicate the landmark location. (c) Minor cheliped measurements. For more details of each measurement, see Table 1.

ments of the major and minor cheliped were taken from the exterior surface. Six specific landmark points were used for the measurements of the major cheliped (Fig. 1b) in order to analyse the shape of the principal parts of the cheliped (e.g. length of pollex; Table 1). Basic length and width measurements were taken from the exterior surface of the minor cheliped (Fig. 1c, Table 1).

Color measurements

Color values (RGB) were obtained using the program CorelDRAW 10 (Corel Corporation, Ottawa, OT, Canada). We quantified the amount of red (R), green (G) and blue (B). Color measurements were obtained from the exterior surface of the propodus and dactylus, respectively, of the major cheliped (for details, see Table 1). We selected a homogeneously colored area (without damage or spines) in the central part of the propodus and dactylus, respectively, to determine the principal color combination for each structure. In these homogeneous areas, the three colors (red, green and blue) were quantified using the paint tool of CorelDRAW. To minimize the variation of light intensity between photographs, conditions of light and camera calibration were maintained identical throughout this study.

Injuries of chelipeds and missing appendages

We also recorded the injury marks on both sides of the major cheliped in all 119 specimens (including those not used in multivariate analyses due to missing or broken appendages). Missing or broken appendages were checked for melanized exoskeleton, which confirmed that all recorded damage had occurred in the field before collection. Injuries are classified into four categories: superficial marks on the chelipeds, without breaking the exoskeleton; punctures and cracks, which are local injuries breaking through the exoskeleton; dactylus or propodus broken, where parts of these structures are broken off; and missing appendage (chelipeds or pereopods).

Data analysis

To examine whether there are different morphotypes of males, we conducted a multivariate analysis (principal component analysis, PCA) for 106 males with all appendages complete. Analyses were carried out in the software PRIMER v. 6.0 (Clarke & Gorley, 2006). The first three axes of the PCA were used to visualize groups, and to corroborate differences between groups, we conducted a multivariate discriminant analysis using Hotelling’s T², which was analysed with the program PAST (Hammer, Harper & Ryan, 2001). In order to

Table 1 Abbreviation, name and explanation of measurements taken on *Cryphiops caementarius*

General measurements		
LBY	Length of body	From the end of rostrum to posterior margin of telson (with the animal fully extended)
LCX	Length of carapace	From the margin of the left eye socket to the posterior margin of the cephalothorax (middle)
LTN	Length of telson	From the posterior margin of last abdominal segment to the posterior margin of telson
LRM	Length of rostrum	From the outside end of the rostrum to the left eye socket
LAB	Length of abdomen	From the anterior margin of first abdominal segment to the posterior margin of telson
WCP	Width of carapace	Maximum width of carapace measured at level of fourth pereopods
WAB	Width of abdomen	Maximum width of the second abdominal segment
Major cheliped measurements		
LS1	Length of segment 1	From landmark 1 to landmark 2
LS2	Length of segment 2	From landmark 2 to landmark 3
LS3	Length of segment 3	From landmark 3 to landmark 4
LS4	Length of segment 4	From landmark 4 to landmark 5
LS5	Length of segment 5	From landmark 5 to landmark 6
LPX	Length of pollex	From landmark 6 to landmark 1
LPP	Length of propodus	From the base of the carpus and manus junction to tip of the pollex
WPP	Width of propodus	Maximum width of propodus in outside view
LSP	Length of the spine at landmark 2	Length of the largest spine (of three measured spines) at landmark 2
LTP	Length of tooth on the propodus	Length of the largest tooth measured from the base (junction with pollex) to the tip
LDY	Length of dactylus	From landmark 5 to tip of the dactylus
LTD	Length of tooth on the dactylus	Length of the largest tooth measured from the base (junction with dactylus) to the tip
LCP	Length of carpus	From anterior margin of carpus to posterior margin in outside view
WCA	Width of carpus	Maximum width of carpus in outside view
Minor cheliped measurements		
LPM	Length of propodus	From the base of the carpus and manus junction to tip of the pollex
WPM	Width of propodus	Maximum width of propodus in outside view
LDM	Length of dactylus	From the upper dactylus and propodus junction to tip of the dactylus
LCM	Length of carpus	From anterior margin of carpus to posterior margin in outside view
WCM	Width of carpus	Maximum width of carpus in outside view
Measurements of color in major cheliped		
RPP	Amount of red in propodus	Measured in the middle of propodus (outside view)
GPP	Amount of green in propodus	Measured in the middle of propodus (outside view)
BPP	Amount of blue in propodus	Measured in the middle of propodus (outside view)
RDY	Amount of red in dactylus	Measured in the middle of dactylus (outside view)
GDY	Amount of green in dactylus	Measured in the middle of dactylus (outside view)
BDY	Amount of blue in dactylus	Measured in the middle of dactylus (outside view)

Table 2 Percentages of variance explained by the three first axes of the principal component analysis (PCA) based on the 32 morphometric and color measurements

PC	Eigenvalues	% variance explained	% accumulated variance explained
1	0.421	63.2	63.2
2	0.127	19.1	82.4
3	0.037	5.6	88

confirm the reliability of the visual group assignment, we tested alternative groupings by a fuzzy c-mean clustering analysis (Bezdek, 1981). This method uses a fuzzy-algorithm to assign an individual to a given number of c-groups (here between 2 and 10). The precision of the classification provided by the visual analysis of the PCA and the different c-groups of the fuzzy-analysis was evaluated using the 'out-of-bag' error

estimate in a random forest analysis (Breiman, 2001). Analyses were repeated using only morphometric measurements (i.e. excluding color measurements). Fuzzy c-mean clustering and random forest analyses were carried out using the R libraries *cluster* (Maechler *et al.*, 2011) and *randomForest* (Liaw & Wiener, 2002), respectively.

Given the behavioral and ecological importance of large chelipeds in this and other shrimps (Ra'anan & Sagi, 1985; Barki, Harpaz & Karplus, 1997; Correa *et al.*, 2003), we examined whether the two groups identified by the PCA showed allometric differences in the relationships between the size of specific cheliped measures and body size (carapace length). Differences in the intercept and slope of the allometric relationship were evaluated using an analysis of covariance (ANCOVA) with the 106 complete males (see above). Variables were natural-log transformed to ensure the homoscedasticity and normality of the data. Finally, to evaluate whether the proportion of body injuries differed

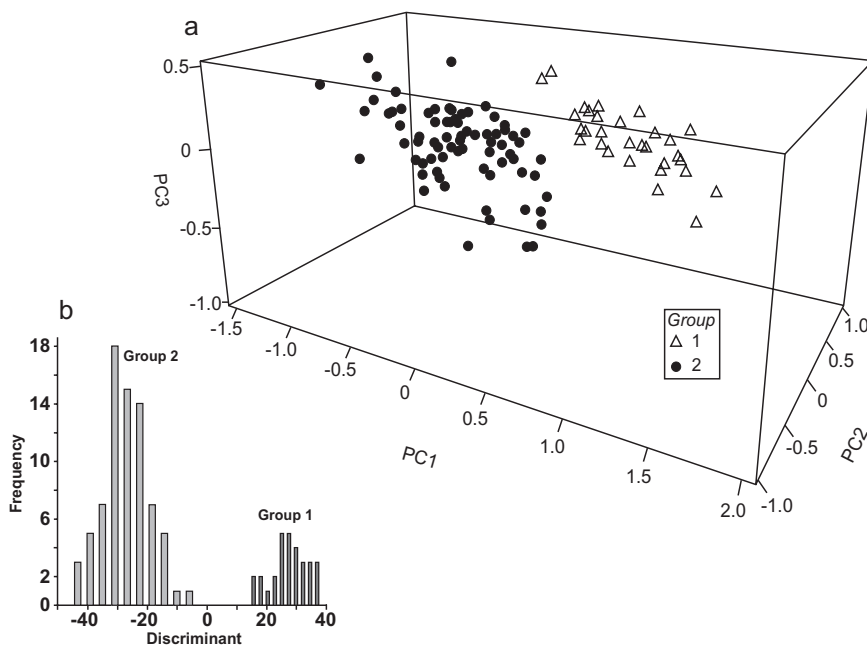


Figure 2 Principal component analysis (PCA) based on morphometric and color measurements of 106 males that had both chelipeds intact. (a) Three-dimensional graph of PCA formed by 32 variables (26 morphometric variables and 6 color variables). (b) Discriminant Hotelling analysis (T^2) for the two groups of PCA.

between the groups identified by PCA, we conducted a chi-square analysis for each type of injury separately. All analyses were carried out using the software R (R Development Core Team, 2011).

Results

Morphometric and colorimetric analyses

Based on the visual assessment of the first three axes of the PCA, two distinct male groups could be distinguished (Fig. 2a). The separation into two groups is supported by the result of the multivariate discriminant analysis of Hotelling's T^2 , which confirms significant differences ($P < 0.0001$) between the two groups (Fig. 2b). The variance explained by principal component 1 is 63.2%, and the first three principal components together explained 88% of the variance in the analysis (Table 2). Variables more related to the first principal component were the length of the teeth on the propodus and dactylus of the major cheliped, respectively (Table 3). Color variables contributed most to the second and third principal component. The error estimate of the random forest analysis was much lower for the two groups visually defined with the PCA plot compared with the groups assigned by the fuzzy c-mean clustering (using all variables). The error estimates varied between 1.9 and 6.5% (median = 2.8%) for the fuzzy groupings, regardless of the number of groups (2–10). This error increased greatly when we used only morphometric variables (3.7–16.8%, median = 11%). In contrast, the two groups defined with the PCA yielded 0 and 0.9% error estimates for all variables and morphometric variables only, respectively.

Table 3 Scores of the 32 morphometric and color measurements associated with each one of the first three axes of the principal component analysis. The two measurements with the largest scores associated with the PC1 are shown in bold. For abbreviations, see Table 1

Variable	PC1	PC2	PC3
LBY	0.056	-0.12	-0.096
LCX	0.087	-0.154	-0.128
WCP	0.092	-0.139	-0.114
LRM	0.03	-0.152	-0.11
LAB	0.049	-0.121	-0.103
WAB	0.066	-0.136	-0.109
LTN	0.069	-0.117	-0.101
LS1	0.179	-0.14	-0.051
LS2	0.222	-0.051	0.046
LS3	0.174	-0.122	0.043
LS4	0.24	-0.046	0.074
LS5	0.236	-0.03	0.061
LPX	0.177	-0.161	-0.081
LPP	0.208	-0.09	0.003
WPP	0.19	-0.048	0.042
LSP	0.234	-0.041	0.162
LTP	0.281	0.032	0.323
LDY	0.197	-0.135	-0.04
LTD	0.280	0.052	0.279
LCP	0.199	-0.078	0.014
WCA	0.185	-0.054	0.022
LPM	0.157	-0.139	-0.104
WPM	0.157	-0.111	-0.055
LDM	0.161	-0.151	-0.118
LCM	0.158	-0.119	-0.054
WCM	0.15	-0.112	-0.054
RPP	-0.198	-0.254	0.026
GPP	-0.213	-0.26	0.198
BPP	-0.122	-0.164	0.77
RDY	-0.209	-0.404	-0.098
GDY	-0.179	-0.433	0.007
BDY	-0.102	-0.415	0.077

Table 4 Results of the analysis of covariance testing for differences in the intercept and slope of the relationships length of segment 2 on propodus (LS2), propodus width (WPP) and carpus length (LCP) versus carapace length (LCX) between the two groups of male *Cryphiops caementarius*. All variables were log-natural transformed. Significant values in bold

Response variable	Source	d.f.	MSS	F	P	Overall R^2
LS2	log (LCX)	1	5.35	275.95	<0.0001	0.84
	Group (intercept)	1	5.08	261.97	<0.0001	
	Group (slope)	1	0.02	0.88	0.35	
WPP	log (LCX)	1	4.06	395.01	<0.0001	0.88
	Group (intercept)	1	3.77	366.24	<0.0001	
	Group (slope)	1	0	0.41	0.52	
LCP	log (LCX)	1	5.59	757.35	<0.0001	0.93
	Group (intercept)	1	3.71	503.51	<0.0001	
	Group (slope)	1	0	0.46	0.5	

MSS, mean sum of squares.

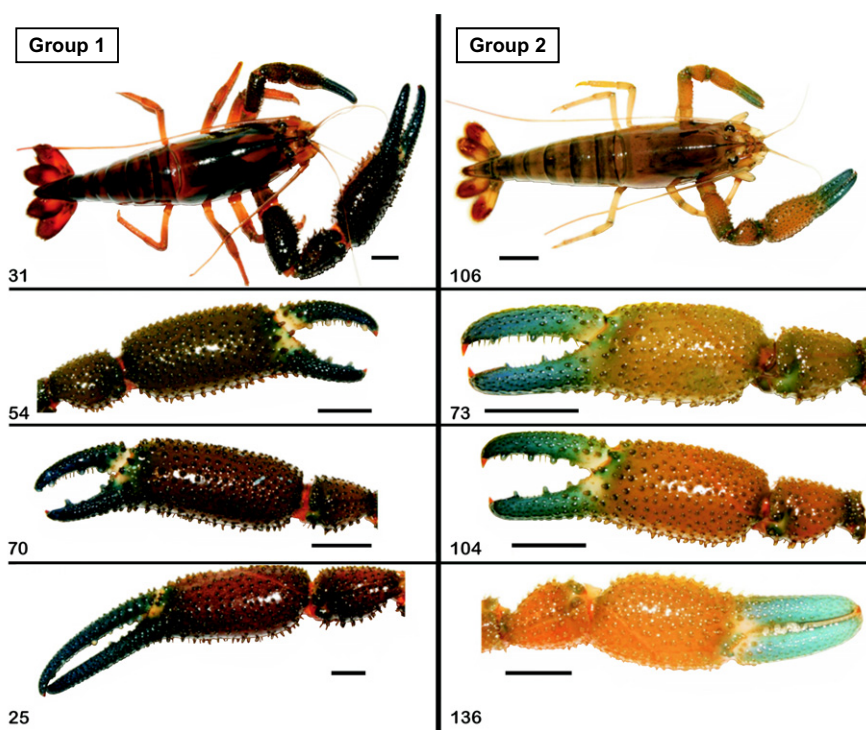


Figure 3 Individuals and major chelipeds of male *Cryphiops caementarius* belonging to the two groups distinguished by principal component analysis. Scale bars represent 1 cm in each picture; numbers in the lower left corner identify the different individuals. All photographs were taken with the same light conditions.

Color patterns and morphometry of males in the two groups

In general, males from the two groups differed in the coloration of the major cheliped. Males from group 1 were generally darker and had more intense colors than males from group 2 (Fig. 3). The dactylus of males from group 1 was often dark blue, almost black, while that of males from group 2 was bright blue. Similarly, the propodus of males from group 1 was of very dark color, while males from group 2 often had a brightly orange-colored propodus.

When comparing single morphometric relationships (three selected measurements on the major cheliped vs. carapace

length), the males from the two groups also exhibit different relationships (Fig. 4, Table 4). In all three cases, the ANCOVA revealed that the two groups differed only in terms of the intercept ($P < 0.0001$ in all cases, Table 4), not the slope ($P > 0.35$ in all cases, Table 4). Males of group 1 showed comparatively a much larger segment 2 [i.e. length of segment 2 (LS2)/length of carapace (LCX), Fig. 4a], a wider propodus [i.e. width of propodus (WPP)/LCX, Fig. 4b] and larger carpus [i.e. length of carpus (LCP)/LCX, Fig. 4c] than males of group 2 for a similar carapace length. The fit of the ANCOVA model was highest for the LCP/LCX relationship ($R^2 = 0.93$, Table 4); consequently, we used this model to classify the 13 males that had lost other appendages (e.g.

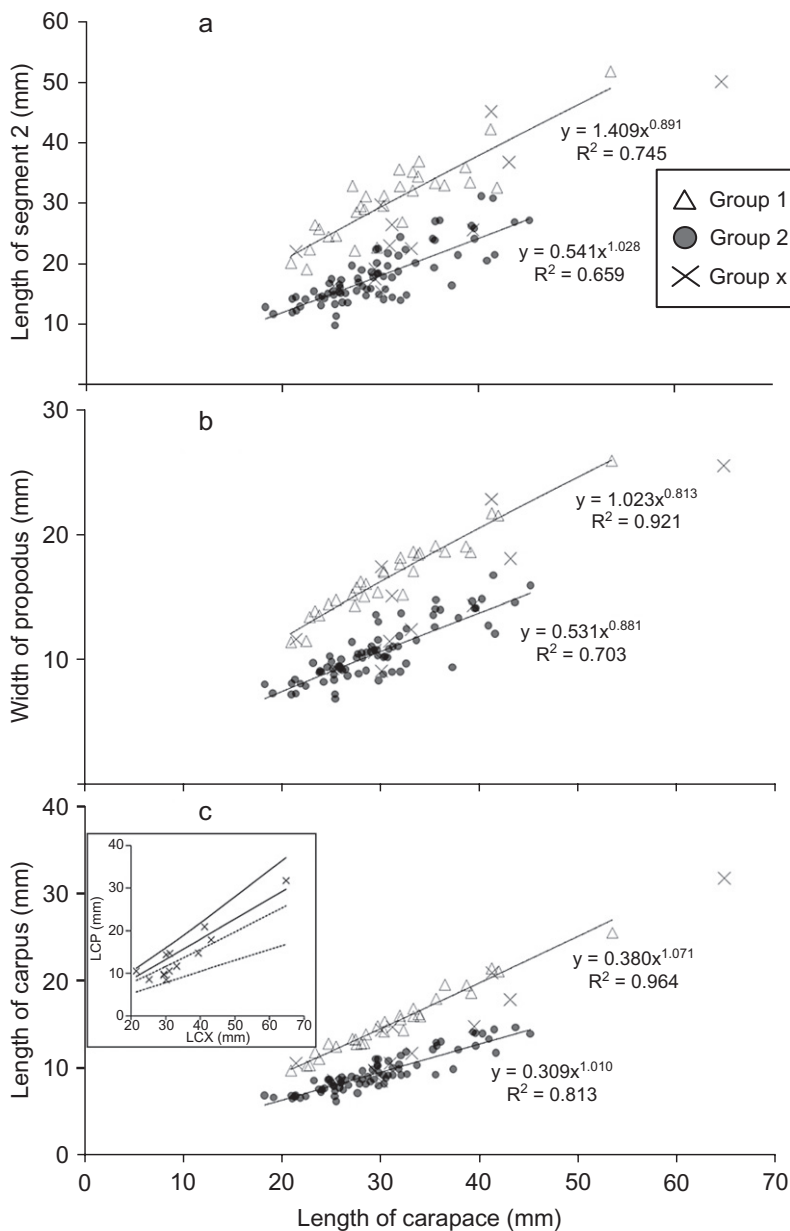


Figure 4 Relationship between selected morphometric measurements on the major chelipeds and carapace length of male *Cryphiops caementarius*. Equations were calculated for males from groups 1 and 2, respectively. The graph also includes individuals that were not used in the principal component analysis due to missing or broken appendages (labeled as a group X, $n = 13$). The inset shows the prediction intervals for the curves presented in (c) (dotted lines of different color) and the observed values for males of group X.

minor cheliped or pereopods) or broken some parts of the major cheliped (group X) into the two groups. Based on the prediction, confidence intervals were built for each group using the LCP/LCX relationship: 12 out of the 13 of males (92%) can thus still be classified into these groups (Fig. 4c, inset).

Injuries of chelipeds and missing appendages

Males of both groups suffered some degree of body injuries (Fig. 5), but males from group 1 generally had more injuries than males from group 2 (Table 5). Almost 75% of the

males from group 1 showed some form of damage, compared with the 29% of males in group 2, a threefold difference that was statistically significant (Table 5, $P < 0.0001$). However, when we analysed each type of injury separately, differences were significant only in the case of superficial marks (Table 5, $P < 0.0001$); more than 50% of males in group 1 showed superficial marks against less than 4% in group 2. Differences between groups were not significant for any other kind of body damage, but there was an overall tendency that the proportion of males with injuries was higher in group 1 than in group 2 (Table 5).

The injuries on the chelipeds were spread over the entire cheliped surface, but superficial marks were concentrated

Table 5 Number and percentage of individuals with different types of body injuries for male *Cryphiops caementarius* from the two groups distinguished by the principal component analysis (PCA). Superficial marks, puncture and cracks, and broken dactylus/propodus were counted on the major cheliped only. Individuals from group X were added to one of two groups (previously formed by PCA) based on the relationship length of carpus/length of carapace shown in Fig. 4. Differences between groups were evaluated using a chi-square analysis (significant values in bold)

Body damage	Group 1 (<i>n</i> = 35)	Group 2 (<i>n</i> = 84)	χ^2	<i>P</i> -value
Superficial marks	19 (54.3%)	3 (3.6%)	38.867	<0.0001
Puncture and crack	6 (17.1%)	6 (7.1%)	1.733	0.188
Dactylus/propodus broken	5 (14.3%)	5 (5.9%)	1.277	0.258
Appendages missing	6 (17.1%)	14 (16.7%)	0.004	0.949
At least one type of damage	26 (74.3%)	24 (28.6%)	19.358	<0.0001

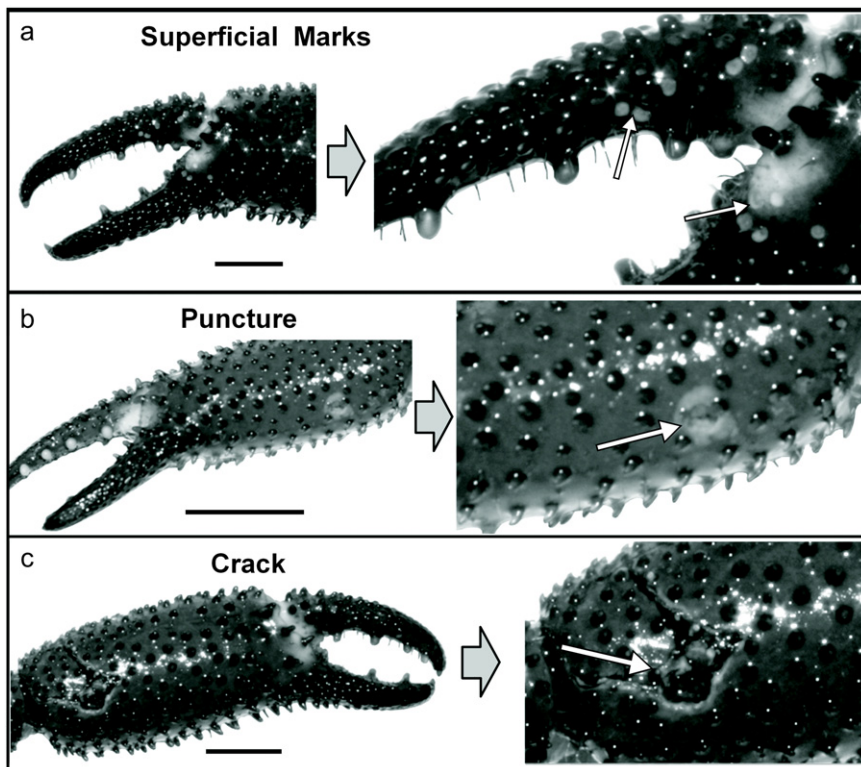


Figure 5 Different types of injuries on major chelipeds of male *Cryphiops caementarius*. Scale bars represent 1 cm.

near the joint between the propodus and dactylus (Fig. 6). Few injuries were found in the central (the widest) part of the propodus.

Discussion

Male morphotypes in *C. caementarius*

In the Andean river shrimp *C. caementarius* two male morphotypes can be distinguished, mainly based on morphological characteristics of the chelipeds. The singular morphometric trait that best separates males from group 1 or group 2 is the relationship between carpus length and carapace length, but also the relationship between the size (width) of the propodus and carapace length. This is comparable to the related species from the genus *Macrobrachium*, where male

chelipeds, and in particular, the carpus and propodus, exhibit marked allometric growth (e.g. Nagamine & Knight, 1980; Kuris *et al.*, 1987; Mariappan & Balasundaram, 2004). Also, in other caridean species with different male morphotypes, the length and width of the cheliped articles grow allometrically in large males, for example, in rhynchocinetid shrimps (Thiel *et al.*, 2010).

Other characteristics that contribute to the differentiation among male morphotypes of *C. caementarius* include primarily characters on the chelipeds such as the spines, teeth and the coloration. Spines on the chelipeds are proportionally longer in males from group 1 than in those from group 2. Similar spines, primarily on the carpus and propodus, have also been reported for male morphotypes in the genus *Macrobrachium* (Kuris *et al.*, 1987; Moraes-Riodades & Valenti, 2004; Short, 2004).

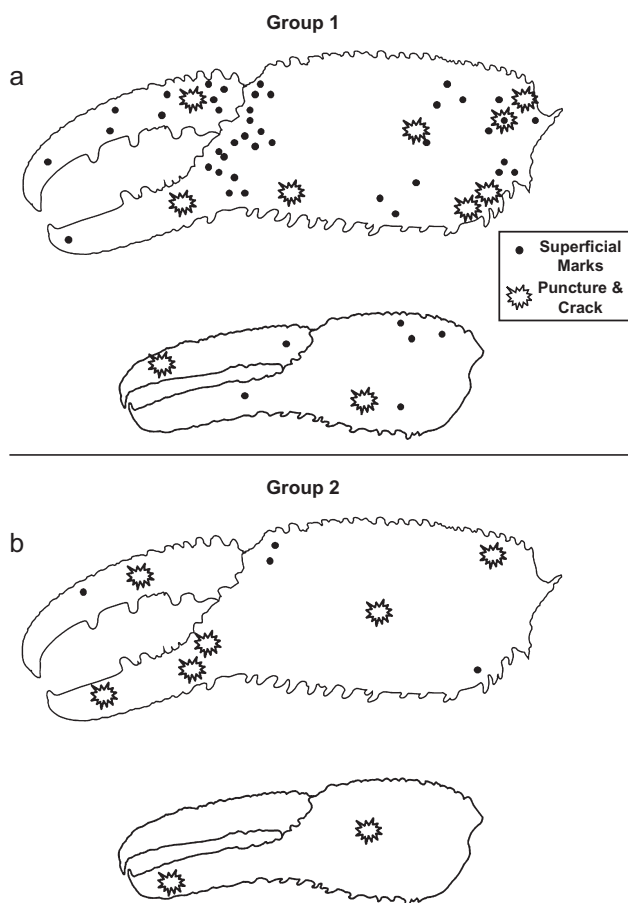


Figure 6 Superficial marks and puncture/crack injuries from the inside and outside surfaces of chelipeds of male *Cryphiops caementarius*. Each generalized cheliped drawing combines the injuries from all individuals belonging to the two groups formed by principal component analysis. (a) Major and minor chelipeds of individuals belonging to group 1 ($n = 35$). (b) Major and minor chelipeds of individuals belonging to group 2 ($n = 84$).

The teeth on the cutting edges of the chelae represent important weapons during agonistic interactions. These teeth are found in the fully developed males of a large number of river shrimps from the genus *Macrobrachium* (Short, 2004). Similar teeth are also found in the chelipeds of dominant males of other carideans, for example, in rhyncinetid shrimps (Thiel *et al.*, 2010), and other decapods, for example, in squat lobsters (Claverie & Smith, 2007). The teeth are much more developed in larger individuals and probably cause important damage during intrasexual combats.

Coloration also varies among the two morphotypes identified herein for males of *C. caementarius*. In *Macrobrachium* shrimps, male chelipeds usually show very distinct color patterns, and male morphotypes can be distinguished based on the coloration of their chelipeds (e.g. Kuris *et al.*, 1987; Moraes-Riodades & Valenti, 2004). The color might serve as

signal during intrasexual combats as suggested by Barki *et al.* (1997) for male morphotypes of *M. rosenbergii*, which feature pronounced and discrete color differences.

In some species of river shrimps, for example, in *M. rosenbergii* and *Macrobrachium amazonicum*, the male morphotypes can be identified relatively easily based on the morphological and color characteristics (Kuris *et al.*, 1987; Moraes-Riodades & Valenti, 2004). The change from one morphotype to the next can be accompanied by discrete changes in morphology and color, especially during the change towards the most developed male stages. These are usually dominant over the less developed male morphotypes (Lee & Fielder, 1982; Ra'anán & Sagi, 1985; Barki, Karplus & Goren, 1991). In *C. caementarius*, the change between the two male morphotypes appears to be more gradual, and only the relationship carpus length versus carapace length represents a discrete difference between the two groups. In particular, the color changes are gradual and color alone does not allow identifying the two morphotypes in *C. caementarius*. While the behavioral significance of morphotype coloration in river shrimps (*Macrobrachium* and *Cryphiops*) is not known at present, the relatively subtle differences in coloration between the morphotypes of *C. caementarius* suggest that in this species color displays might only play a minor role during intra-male interactions and that conflicts over resources (food, shelter and mates) might be resolved by direct agonistic interactions. The high frequency of injuries supports this suggestion because effective color communication likely would serve to mitigate conflicts and reduce the probability of escalating fights.

Body injuries

The comparatively high proportion of males with injuries on their chelipeds indicates a high frequency of agonistic interactions among male *C. caementarius*. The present study was conducted during the peak of the annual reproductive period, suggesting that aggressive encounters during mate competition cause the observed injuries, similar as in many other taxa (Siva-Jothy, 1987; Lailvaux *et al.*, 2004; Mainguy *et al.*, 2008; Moore *et al.*, 2009). Most injuries on the chelipeds likely result from the grip with the teeth or tips of the cutting claws, similar as suggested for porcelain crabs or squat lobsters (Claverie & Smith, 2007; Rypien & Palmer, 2007). In estuarine crabs, the injuries are mostly distributed along the border of the chelae (Jones, 1980; Rypien & Palmer, 2007), suggesting that combatants grab their opponents in a different way than in *C. caementarius*, where injuries could be found in most parts of the chela.

Herein, we found that the individuals from group 1, that is, the morphotype with the more developed chelipeds, had a higher proportion of individuals with injured chelipeds than the individuals from group 2. This is in contrast to the observations from *P. cinctipes*, in which small individuals had a higher incidence of cheliped injuries than large individuals (Rypien & Palmer, 2007). In their study, aggressive interactions most likely were caused by competition for food and

space, and injuries were especially frequent at high population densities. Intertidal porcellanid species gather in common refuges during low tide, and when forced into these shelters, small individuals might suffer more from aggressive interactions than large individuals. In river shrimps, small individuals might have more opportunities to avoid encounters with dominant individuals or to escape by tail-flips into the open water.

The higher proportion of injuries in the more developed morphotype of *C. caementarius* might also be due to the fact that these compete more aggressively for access to reproductive females than less developed, smaller males. In crayfish *Orconectes rusticus*, large males fight more intensively than SMs, and during prolonged and intense fights, the large males are thought to incur a high risk of injuries (Schroeder & Huber, 2001). This interpretation is supported by observations on dominant males of *Rhynchocinetes brucei*, which had suffered more body damage than other individuals (Thiel *et al.*, 2010).

Surprisingly, among the less developed males of *C. caementarius*, there is a comparatively high proportion of males with lost appendages. Decapods may autotomize appendages during intraspecific fights (e.g. Parzefall & Wilkens, 1975; Sneddon *et al.*, 2003). In estuarine crabs, large males had a higher incidence of lost chelae than SMs (Smith & Hines, 1991; Abello *et al.*, 1994). This is in contrast to *C. caementarius*; possibly in river shrimp, SMs autotomize appendages more readily than large individuals. When gripped by other males, SMs might lose chelipeds, while large males (which molt less frequently than SMs) might hold onto their chelipeds and, consequently, be more susceptible to injuries. The high proportion of accumulated cheliped injuries in more developed, large *C. caementarius* could thus be due to a higher threshold for autotomy.

Conclusions and outlook

Two male morphotypes can be distinguished in *C. caementarius*. Differentiation of the two morphotypes is primarily based on the characteristics of the chelipeds, which include weapons (teeth) and coloration. Presently available evidence suggests that the two observed morphotypes reflect dominant and subordinate individuals. Differences in cheliped injuries between the two morphotypes also suggest differences in agonistic behaviors and autotomy thresholds during fights. This interpretation is supported by previous observations on other natural populations of *C. caementarius*. Hartmann (1958) and Viacava *et al.* (1978) reported several old and large males (total length: 25 cm) in the river populations of Peru. Similarly, Bahamonde & Vila (1971) observed a small proportion of large males (carapace length: 48–62 mm) in populations from rivers in northern Chile. Most likely these large males (with the major cheliped of large size and intense color, e.g. Viacava *et al.*, 1978) represent 'dominant' males, which appear to be less common (29.4% in the present study) than 'subordinate' males (70.4%). According to anecdotal reports from shrimp collectors, these large males (called 'garrudo') are highly territorial and occupy the best refuges in the river,

further suggesting that they are dominant over other males. Future behavioral studies are needed to test these hypotheses.

The present study was conducted at a particular site and during a particular season. While the presence of different male morphotypes in *C. caementarius* likely is independent of site or season (earlier, anecdotal reports already suggested their existence), their proportions might vary depending on the particular context. Also, the observed differences between the morphotypes in injury levels may vary depending on intrinsic (e.g. sex ratio, population density) or extrinsic factors (e.g. habitat structure and refuge availability). Examining these factors will help one to understand why some studies observed higher injury levels in subordinate individuals (e.g. Ros *et al.*, 2006) and others in dominant individuals (e.g. Moore *et al.*, 2009; this study).

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