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**Original Article**

# Original Article

# Sexual dimorphism in the Neotropical snakes genus *Chironius* (Serpentes: Colubridae)

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## ABSTRACT

Sexual dimorphism is a common phenomenon among snakes, with female snakes being larger than male snakes as a recurrent pattern . However, species that show male-male combat behaviour may impose a selective advantage by developing larger bodies in male specimens, like the diurnal *Chironius* snakes, which display courtship and male-male combat behaviours. In this study, we analysed sexual dimorphism in body, tail, and head size, and skull size and shape in twelve species of *Chironius*. We investigated whether sexual dimorphism patterns could be a result of allometric growth. The absence of sexual dimorphism regarding body and head length was the main pattern revealed by our analyses. Sexual dimorphism characterized by larger body and head sizes in males was observed in *Chironius bicarinatus*, *Chironius foveatus*, and *Chironius fuscus*. Only females of *Chironius exoletus* exhibited larger body and head sizes than males. Regarding the shape of the head, six species showed sexual divergences, with enlarged or robust heads. Sexual dimorphism in skull shape seems related to selection in *Chironius flavolineatus*, with no allometric influences. Larger tails in males of *C. fuscus*, *C. flavolineatus*, and *Chironius quadricarinatus* may represent an advantageous defensive strategy. Finally, the lack of divergence in tail length in the remaining species probably evolved due to arboreal habits in *Chironius*.

**Keywords:** static allometry; morphometric geometric; skull shape

## INTRODUCTION

Sexual dimorphism in snakes has been associated with fecundity, sexual selection, and intraspecific niche divergence (Darwin 1871, Camilleri and Shine 1990, Shine 1990b, Dubey *et al.* 2009). Regarding body size, the recurrent pattern corresponds to larger females. However, in terms of snake species that exhibit combat behaviour, males have larger bodies than females or, at least, there are no differences between sexes (Shine 1994). Therefore, such combative behaviour between males can impose a selective advantage by evolving larger body sizes, reflecting an advantage during interaction and, consequently, influencing reproductive success (Shine 1978, 1994). This behaviour is characterized by copulatory disputes between males to reproduce with females during mating and has been recorded in numerous snake species from different families. As for the ritual combat, it involves a behavioural repertoire mainly characterized by intertwining the body and different postures of head elevation (Shine 1978, 1994, Almeida-Santos *et al.* 2002, Pizzato *et al.* 2006, Senter *et al.* 2014, Missassi *et al.* 2022, Senter 2022).

Considering that most combatant species do not use their head nor jaws directly during a confrontation, it is unlikely that sexual differences observed in the head or skull size could be explained by sexual selection and ritual combat behaviour (Camilleri and Shine 1990, Shine 1991). Usually, dimorphic heads and skulls are associated with sexual divergences in the diet, depending on the different sizes and types of prey that males and females feed on (Shine 1990a, Houston and Shine 1993, Murta-Fonseca *et al.* 2019, Santos *et al.* 2022). However, in some combat species, males use their head and forebody to varying degrees during confrontations. Unfortunately, these observations are isolated, making more detailed interpretations of possible hypotheses that explain sexual divergences in the head in species with intraspecific interactions complex (Greene and Mason 2000, Almeida-Santos *et al.* 2002, Muniz-da-Silva and Almeida-Santos 2013).

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Regarding tail length, the dominant pattern in snakes is males with longer tails than females. This pattern may be related to the need for additional space to accommodate the hemipenis and associated muscles, body enlargement of females with secondary reduction of their tail, and tail utilization during courtship interactions, mating, or reproductive aggregations (Klauber 1943, Clark 1967, Kaufman and Gibbons 1975, Semlitsch and Gibbons 1982). In species that perform courtship, a greater caudal length can contribute to the success of male mating when used to displace other males and encourage females to copulate (Almeida-Santos *et al.* 1999). However, arboreal species generally do not show dimorphism in tail length, a fact related to their performance during locomotion that would favour both sexes (Lillywhite and Henderson 1993, Pizzatto and Marques 2007).

Sexual dimorphism in any of these traits (body, head, and tail) can reflect ecological differences, adaptations to reduce competition between sexes, or it may result from non-adaptive processes such as allometry, among other factors. Allometry describes variation in a structure (size or shape) associated with body size changes (Klingenberg and McIntyre 1998, Sanger *et al.* 2013). Allometric analysis, therefore, can suggest which processes are responsible for sexual dimorphism, whether those are linked to developmental processes or to a result of evolutionary adaptations (Strauss 1985).

The monophyletic Neotropical colubrine genus *Chironius* has 23 recognized diurnal species, which can be terrestrial or present different arboreality degrees that feed primarily on frogs (Hollis 2006, Klaczko *et al.* 2014, Hamdan *et al.* 2017, Torres-Carvajal *et al.* 2019, Roberto and Souza 2020, Uetz and Hallerman 2021, Banci *et al.* 2022). The genus, therefore, is an excellent model for a comparative analysis on sexual dimorphism because it includes species that exhibit combat behaviour between males [*Chironius bicarinatus* (Wied, 1820) and *Chironius carinatus* (Linnaeus, 1758)] and courtship behaviour [*Chironius flavolineatus* (Jan, 1863)] (Starace 1998, Feio *et al.* 1999, Almeida-Santos and Marques 2002).

In this study, we combined linear and geometric morphometrics to analyse patterns of sexual dimorphism in body, tail, and head size, and skull both size and shape, in twelve species of *Chironius*. We compared the allometric trajectories from the analysed structures between males and females to identify the allometric patterns in each sex and then verify if these could generate the observed patterns of sexual dimorphism. Considering intraspecific interactions, combat, and courtship in *Chironius* species, we hypothesize that sexual dimorphism mainly favours males with larger bodies and heads than females. Regarding the tail, we expect no sexual divergences due to arboreal habits (Lillywhite and Henderson 1993, Pizzatto and Marques 2007). Finally, for species with intraspecific interactions, we compared tail length between males of *C. flavolineatus* vs. *C. bicarinatus* and *C. carinatus* to test the hypothesis that courtship behaviour males would have a larger tail than males that combat (King 1989).

## MATERIAL AND METHODS

### Analysed material

We analysed 658 adult specimens of twelve species of *Chironius*, namely: *Chironius bicarinatus* Wied-Neuwied, 1820 (41 females and 39 males), *Chironius brazili* Hamdan & Fernandes, 2015 (22 females and eight males), *Chironius carinatus* Linnaeus, 1758 (17 females and 40 males), *Chironius exoletus* (Linnaeus, 1758) (39 females and 39 males), *Chironius flavolineatus* (30 females and 27 males), *Chironius foveatus* Bailey, 1955 (11 females and 25 males), *Chironius fuscus* (Linnaeus, 1758) (38 females and 33 males), *Chironius gouveai* Entiauspe-Neto, Lyra, Koch, Quintela, Abegg & Loebmann, 2020 (16 females and 27 males), *Chironius laevicollis* (Wied, 1824) (26 females and 26 males), *Chironius multiventris* Schmidt & Walker, 1943 (26 females and 25 males), *Chironius quadricarinatus* (Boie, 1827) (25 females and 22 males), and *Chironius scurrulus* (Wagler in Spix, 1824) (19 females and 37 males) (for the complete list Appendix, List of Materials analysed). We examined 69 skulls of six species, prepared under immersion in hot water, and the tissues removed with the aid of tweezers, being: 11 of *C. bicarinatus* (six females and five males), 15 of *C. carinatus* (seven females and eight males), 11 of *C. exoletus* (five females and six males), 10 of *C. flavolineatus* (four females and six males), 13 of *C. fuscus* (seven females and six males), and nine of *C. quadricarinatus* (five females and four males).

The analysed material belongs to the following Brazilian collections: Herpetological Collection of the Butantan Institute (IBSP), Herpetological Collection of the Federal University of Ceará (UFC), Herpetological Collection of the Pontifical Catholic University of Rio Grande do Sul (PUCRS), Herpetological Collection of the Museum of Natural History Capão da Imbuia (MHNCI), Collection of Reptiles at the Unicamp Museum of Zoology (ZUEC), Reptile Collection of the National Museum of Rio de Janeiro (MNRJ), Zoological Collection of the Federal University of Mato Grosso do Sul (ZUFMS), Museu Paraense Emílio Goeldi (MPEG), and Herpetological Collection of the Federal University of Mato Grosso (UFMT).

Considering the taxonomic uncertainties involving species that are recognized as part of cryptic species complexes, we restricted our analyses to the following populations: for *C. exoletus*, populations from the type locality in the state of Maranhão and in northeast and southwest of the state of Pará (Hamdan *et al.* 2017, Torres-Carvajal *et al.* 2019); for *C. flavolineatus*, we analysed specimens from the Cerrado-Amazon transition areas, from the Cerrado and also from the Amazon, in locations not corresponding to the lineages described by Hamdan *et al.* (2017); for *C. fuscus*, we analysed the population of the Brazilian Amazon, from the states of Rondônia, Amazonas, south of the Amazon River, Pará, and from Maranhão (Torres-Carvajal *et al.* 2019); as for *C. multiventris*, we have analysed specimens from eastern Amazonas, northern Rondônia, and Pará (Torres-Carvajal *et al.* 2019). Our objective was to characterize the sexual dimorphism for the species without considering population differences or seasonal differences, aiming to increase the analysed sample.

We verified the sex through an incision at the tail base to verify the presence or absence of a hemipenis. We only analysed adult individuals. We consider male and female adults, respectively, with a snout-vent length (SVL) greater than 46 cm and 55 cm for *C. bicarinatus* (Entiauspe-Neto *et al.* 2020); 53 cm and 57 cm for *C. fuscus* (Nascimento *et al.* 2013); 59 cm (Entiauspe-Neto *et al.* 2020) and 75.2 cm (this study) for *C. gouveai*; 47.4 cm and 60 cm for *C. exoletus* (this study); 58 cm and 56.2 cm for *C. flavolineatus* (this study); 51.1 cm and 55.9 cm (this study) for

*C. quadricarinatus*; 50.7 cm (Pinto *et al.* 2010) and 60 cm (this study) for *C. scurrulus*; and 50.7 cm and 60.6 cm for *C. brazili*, *C. carinatus*, *C. foveatus*, *C. laevicollis*, and *C. multiventris* (Pinto *et al.* 2010). We considered males mature when showing an opaque and coiled vas deferens and females showing vitellogenic follicles (Shine 1978, 1980, Slip and Shine 1988).

### Linear morphometrics

We used a caliper and a millimetre ruler (1 mm precision) to obtain three linear measurements for the external morphology analysis: head length (HL—from the tip of the snout to the quadratemandibular articulation), snout-vent length (SVL—from the tip of the snout to the cloaca), and tail length (TL—from the cloaca to the end of the tail). All the variables were log-transformed for the following analysis. We tested the normality and homogeneity of the variables through Shapiro–Wilk and Levene's tests using the 'RVAideMemoire' and 'car' packages, respectively (Fox and Weisberg 2019, Hervé 2021). All analyses were performed in the R software (R Core Team 2021).

To verify the existence of sexual dimorphism in the head, body, and tail length, we used the *t*-test and Kruskal–Wallis test in the 'stats' package. To test the hypothesis that species with courtship behaviour show larger tails than species that combat, we compared the tail length of *C. flavolineatus* (courtship species) with *C. bicarinatus* and *C. carinatus* (species displaying combat behaviour). We have used a one-way ANOVA in the 'stats' package. We tested differences between species using *posthoc* analyses by Bonferroni and TukeyHSD in the 'PostHocTest' package (Signorell *et al.* 2022), with the results graphically demonstrated through boxplots produced in the 'ggplot2' package (Wickham 2016).

### Static allometry

We analysed the head and tail growth patterns in relation to body length (SVL), estimating the allometric slope (b) from the slope of a linear regression between each log-transformed morphometric variable and the log-transformed SVL. We obtained the allometric slope (b) using the equation $Y = aX^b$, where Y corresponds to head and tail length and X to body size. Then, we have tested to determine if the allometric slopes (b) were statistically different from isometry ($b = 1$), using the tilt test function in the 'Smatr' package ($P > 0.05$ indicates isometry) (Warton *et al.* 2012). This analysis suggests the different growth patterns of a structure in relation to the body length. Isometry occurs when the relative size of a structure grows in the same proportion as the body size ($b = 1$). Positive and negative allometric describe when a given structure grows at a higher ($b > 1$) or lower ($b < 1$) rate in relation to the body size, respectively (Bonduriansky 2007). We analysed the allometric relationship for each sex of each species separately to compare the allometric slopes (b) and the intercept (which reflect differences in the size of a structure) to test whether the sexes differ in the allometric trajectories of head and tail growth. This comparison was performed using an analysis of covariance (ANCOVA), with head and tail length as dependent variables, SVL as a covariate, and sex as an independent variable.

### Geometric morphometric analyses

For the geometric morphometric analyses, we obtained images of the dorsal view of the snakes' heads and skulls using a tripod coupled to a Nikon D90 camera and a Nikon macro 60 mm f 2.8 lens. We converted the images to dig format using the tpsUtil program v.1.81. We digitized 19 and 18 landmarks in the head and skull images, respectively, using the tpsDig2 v.2.32 software (tps series software: Rohlf 2008, 2015). All the landmarks were inserted at the contact points and ends of the cephalic shields, and at the meeting points and bone ends (Fig. 1; Supporting Information, Table S1). The anatomical landmarks were standardized by removing the effect of position, size, and orientation using the Procrustes Overlay method, now called Procrustes coordinates (Klingenberg 2011). The average shape for each species was obtained and used in the following analyses.

We investigated the presence of sexual dimorphism in the shape of both head and skull of the species using a Procrustes ANOVA. We visualized shape variation in head and skull between sexes using Canonical Variables Analysis (CVA) (Klingenberg and McIntyre 1998, Klingenberg *et al.* 2002, 2010, Klingenberg and Monteiro 2005). Sexual dimorphism in skull size was analysed based on centroid size, with the results graphically demonstrated through boxplots produced in the R program with the 'ggplot2' package (Wickham 2016).

We tested the allometry in the shape of both head and skull for each sex separately, utilizing multivariate regression of the shape variables (Procrustes coordinates) under the centroid size. The amount of size-dependent shape variation is expressed as the percentage of the total shape variation explained by size. We tested the statistical significance of the multivariate regression analysis using permutations with 10 000 iterations (Klingenberg 2011, 2022). We test differences in allometric trajectories between sexes by comparing the angles among the allometric slopes of the resulting vectors from the regressions. The comparison was performed using the Compare Vector Directions method, in which statistically significant results indicate similarities in the allometric trajectories. All geometric analyses were implemented in the MorphoJ program (Klingenberg 2011).

## RESULTS

### Linear morphometrics

Four of the twelve species analysed showed sexual dimorphism in head size. Males of *C. bicarinatus*, *C. foveatus*, and *C. fuscus* have larger heads than females, whereas females of *C. exoletus* are larger. Four of the analysed *Chironius* species exhibited sexual dimorphism in body size. *Chironius bicarinatus*, *C. foveatus*, and *C. fuscus* showed males larger than females, whereas in *C. exoletus*, females were larger than males. Regarding tail length, only *C. fuscus* showed sexual dimorphism, with males being larger than females (Fig. 2; Supporting Information, Table S2).

In the analysis comparing males of species with intraspecific interactions (courtship vs. combat), we observed significant differences in tail length ($P < 0.001$) between analysed species. *Chironius flavolineatus*, a species with courtship behaviour, has smaller tails, differing statistically from both combat species (*C. bicarinatus* and *C. carinatus*), while there are no significant differences between the latter (Supporting Information, Fig. S1; Table S3).

We observed negative allometry in the head length of females and males of *C. bicarinatus* and *C. exoletus*, and males of *C. fuscus*. This result was also observed for species that were

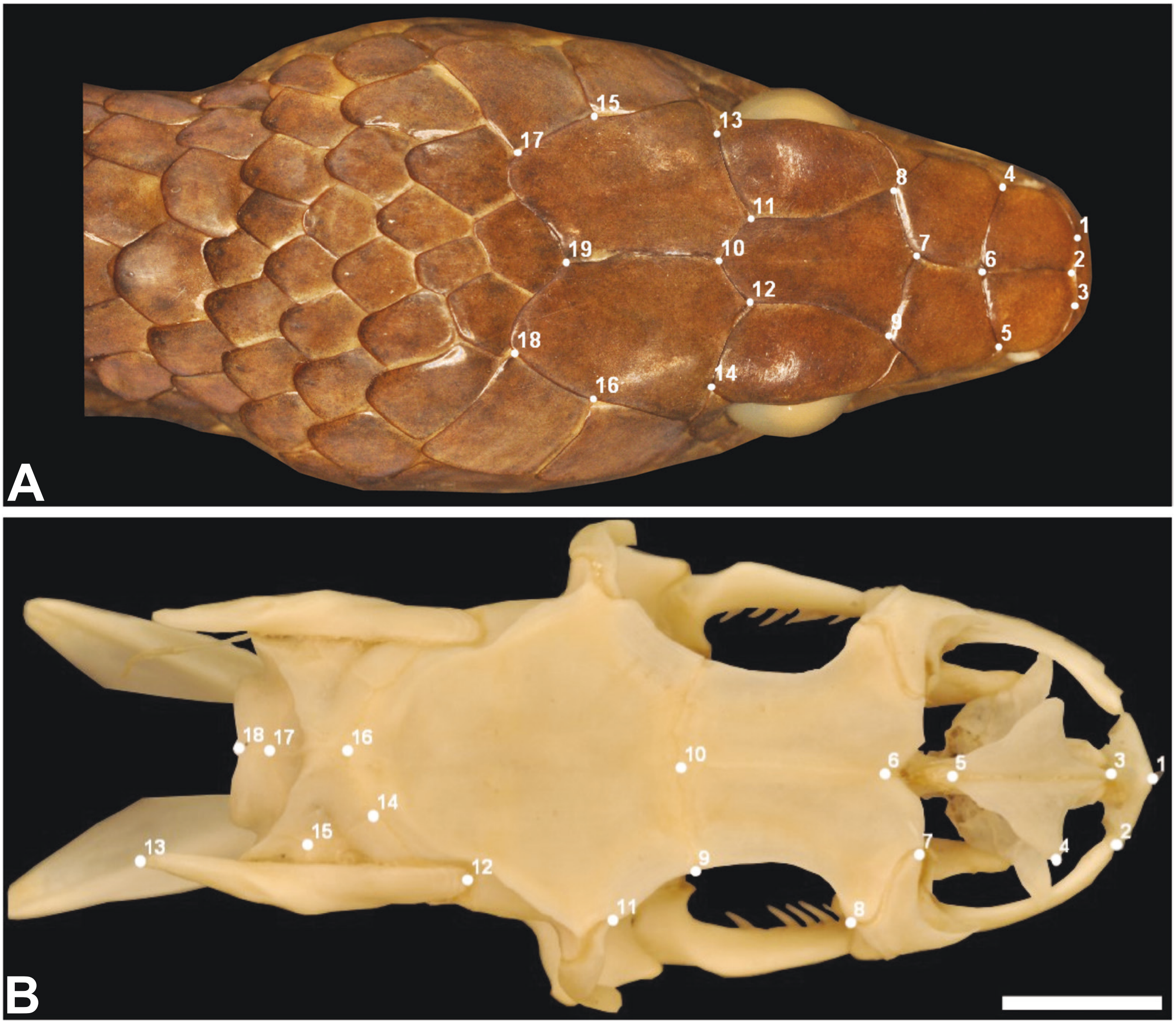

**Figure 1.** Landmark configurations used on the head (A) and skull (B) of species of *Chironius*. A, Head of *C. scurrulus*—MPEG 21 192. B, Skull of *C. carinatus*—MPEG 21 277. Scale: 5 mm.

not dimorphic: in females and males of *C. laevicollis* and *C. multiventris*; females of *C. brazili* and *C. scurrulus*; and males of *C. carinatus* (Supporting Information, Table S4). ANCOVA indicated significant differences in the allometric inclinations about the head growth of males and females of *C. exoletus*, but with borderline statistical significance ($P = 0.05$) (Supporting Information, Fig. S2; Table S5). For the other species, there were no significant differences between the sexes in relation to the allometric inclinations and between the intercept, indicating that the growth trajectories are shared among males and females.

On the other hand, the allometry analyses revealed isometric growth in the tails of all males and females analysed (Supporting Information, Table S4). *Chironius gouveai* males and females differed in allometric inclinations, whereas in *C. flavolineatus* and *C. quadricarinatus* males had larger tails than females (differ in intercept) (Supporting Information, Fig. S3; Table S5).

### Geometric morphometrics

The Procrustes ANOVA analysis revealed that *C. bicarinatus*, *C. carinatus*, *C. exoletus*, *C. foveatus*, *C. fuscus*, and *C. laevicollis* exhibited sexual divergences in the shape of the head (Supporting Information, Table S6). In general, CVA revealed two patterns of head shape: males with a broader head in *C. bicarinatus*, *C. carinatus*, and *C. foveatus*; and larger-headed females in *C. exoletus*, *C. fuscus*, and *C. laevicollis*. Enlargement in males' heads occurred in the following regions: between the prefrontals and the median portion of the parietals (*C. bicarinatus*) (Fig. 3A), between the supraoculars and the median portion of the parietals (*C. carinatus*) (Fig. 3B), and between the supraoculars and the anterior portion of the parietals (*C. foveatus*) (Fig. 3C). Enlargement in females occurred in the following regions of the head: in the posterior portion of the parietals (*C. exoletus*) (Fig. 3D), between the supraoculars and the median portion of the

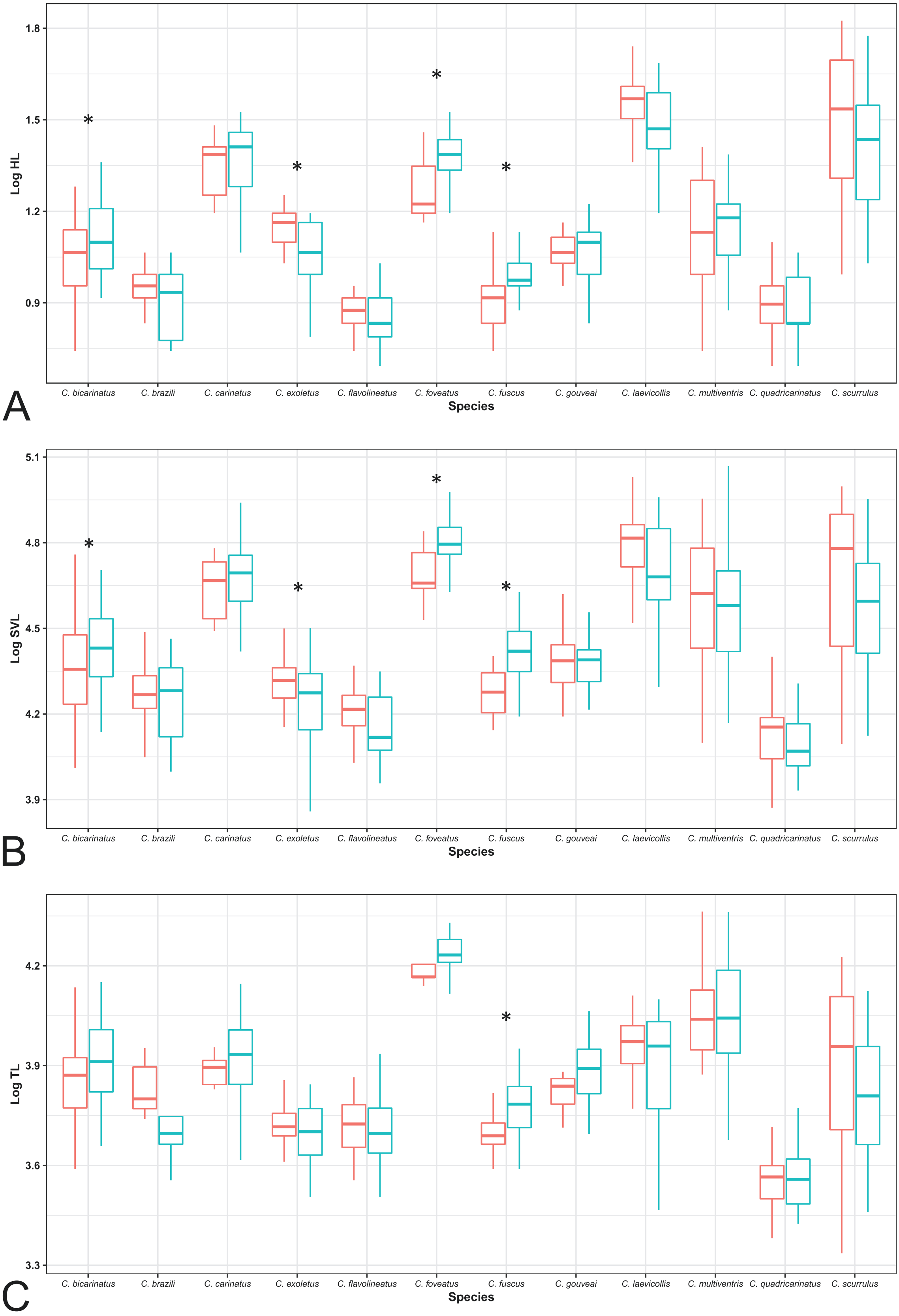

**Figure 2.** Boxplots representing variation in head (A), body (B), and tail (C) length in females (red) and males (blue) of species of *Chironius*. Asterisks represents statistically significant differences. The middle line represents mean values, boxes show standard deviation, and whiskers represent minimum and maximum values.

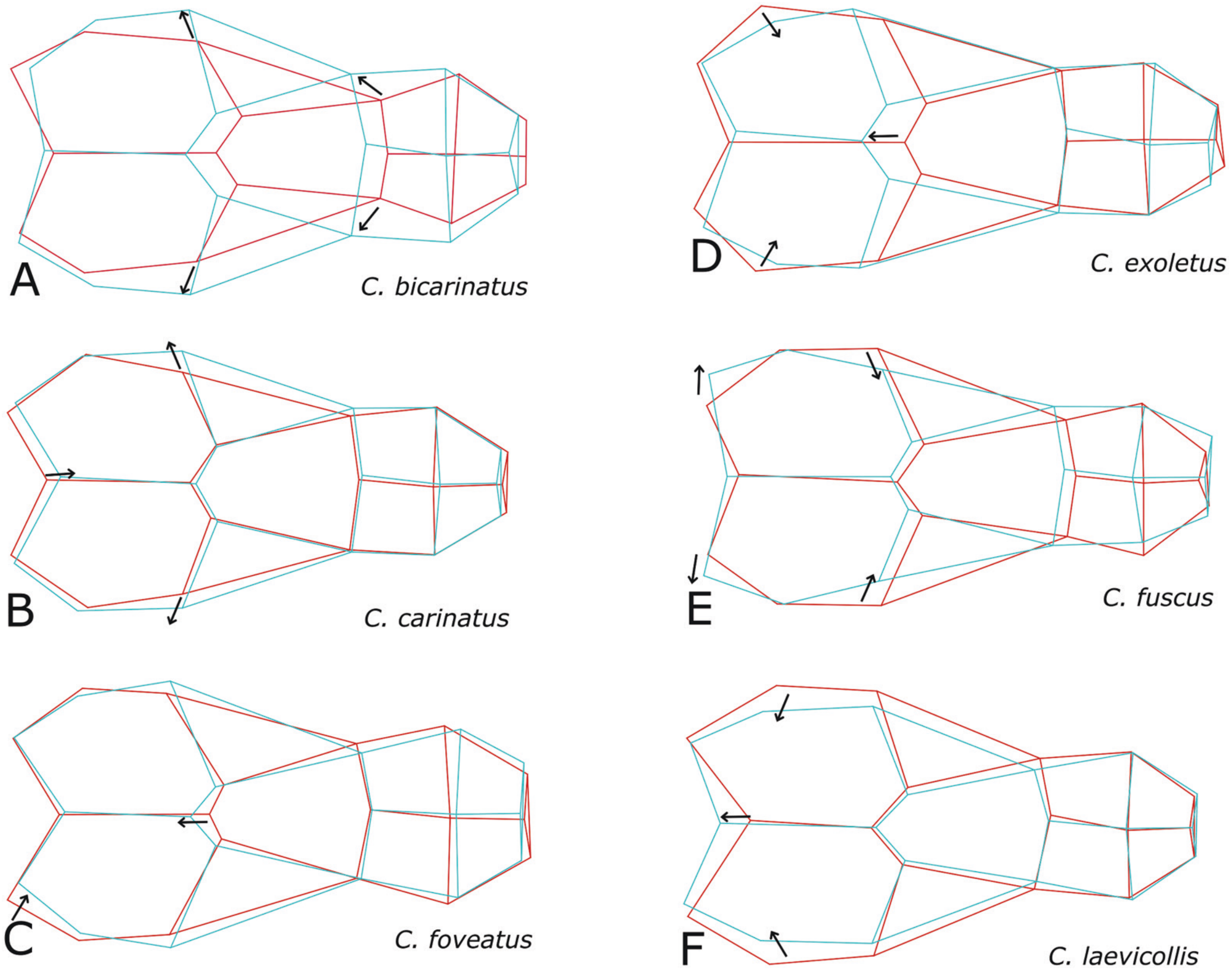

**Figure 3.** Transformation grids generated by CVA demonstrating head shape deformations for females (red) and males (blue) of *Chironius* species, at the positive and negative extremes of the first canonical variate axis. The black arrows in each grid indicate the major changes in shape of males relative to females.

parietals (*C. fuscus*) (Fig 3E), and between the prefrontals and the posterior portion of the parietals (*C. laevicollis*) (Fig. 3F).

Procrustes ANOVA for skull size and shape indicated sexual dimorphism only in *C. flavolineatus* (Fig. 4; Supporting Information, Table S7). The CVA analysis revealed that males of *C. flavolineatus* have a short skull and a more robust premaxilla than females (Fig. 5).

We observed allometry in head shape in four of the six dimorphic species: in females of *C. bicarinatus*, and in males of *C. carinatus*, *C. exoletus*, and *C. foveatus*. Allometry also affected species that did not show sexual divergence in head shape: males and females of *C. multiventris* and *C. scurrulus*; females of *C. flavolineatus* and *C. quadricarinatus*; and males of *C. gouveai* (Supporting Information, Fig. S4; Table S8).

In *C. bicarinatus*, *C. flavolineatus*, and *C. quadricarinatus* only females were allometric, with greater snout robustness as the head grew, and subtle narrowing of the median region of the head (*C. flavolineatus*); *C. carinatus* males show greater enlargement of the median and posterior region of the head (eyes and parietals), whereas the opposite pattern was observed in *C. exoletus* males; *C. foveatus* males also show modifications related to snout enlargement and eye enlargement; and in males of *C. gouveai* the changes involved a slight enlargement in the posterior region of the eyes, as well as elongation and widening of the snout. In females and males of *C. multiventris*, the increase in the head is related to the increase in the snout; and in females and males of *C. scurrulus*, head enlargement is related to greater robustness in the posterior region and more significant development of the snout. The angular comparison of the regression vectors indicated common allometric trajectories between the sexes in *C. bicarinatus*, *C. brazili*, *C. carinatus*, *C. exoletus*, *C. flavolineatus*, *C. multiventris*, and *C. scurrulus* (Supporting Information, Table S8).

We observed allometry in cranial shape in *C. bicarinatus* males and females, *C. fuscus* females, and *C. carinatus* males (Supporting Information, Table S9). These variations are related to the following changes: males and females of *C. bicarinatus* show an increase in the strength of the snout and a braincase shortening (Fig. 10A, B); females of *C. fuscus* show an increase in their nasal robustness and a shortening of the braincase (Supporting

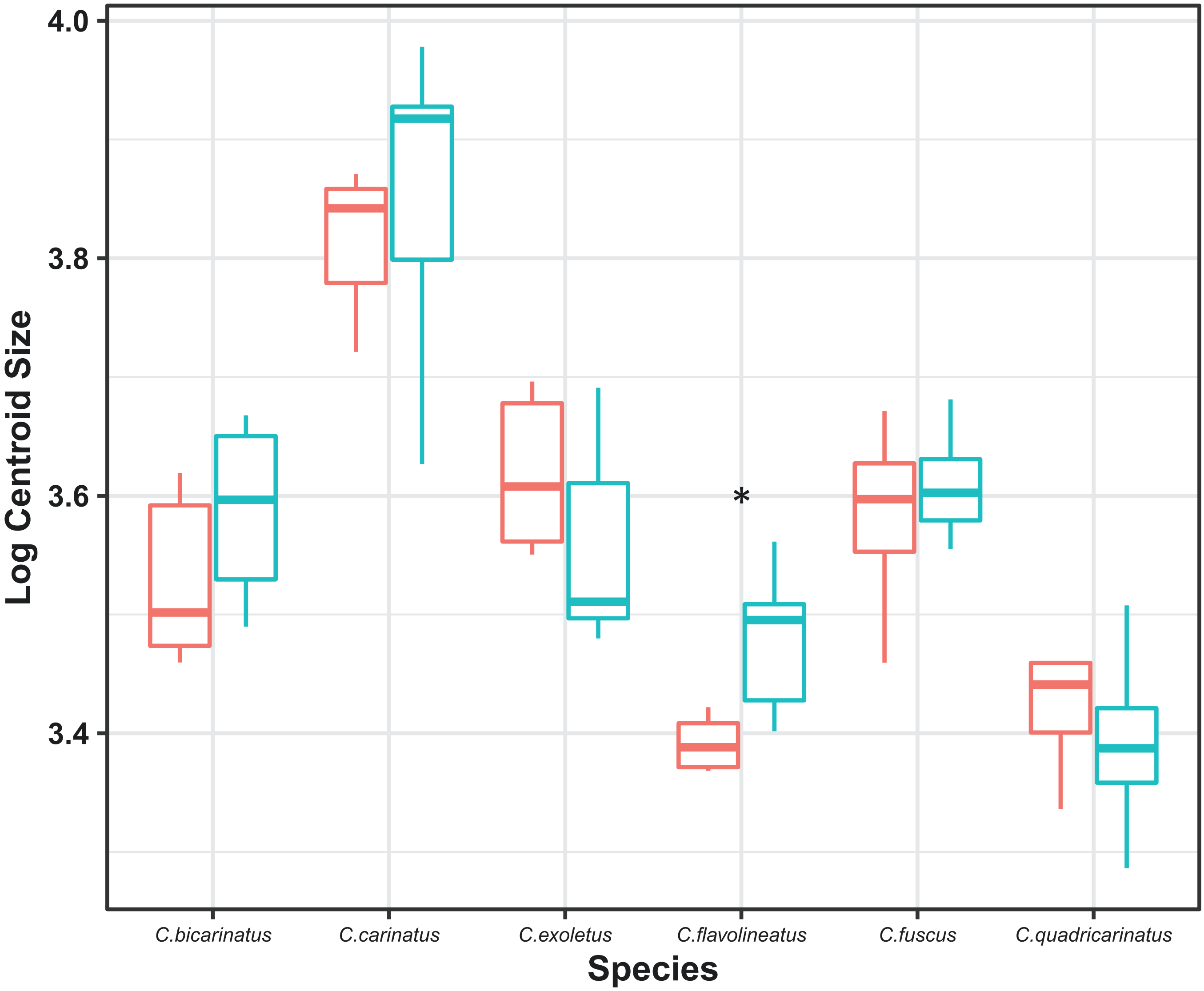

**Figure 4.** Boxplot representing variation in skull size (centroid size) in females (red) and males (blue) of *Chironius* species. The asterisk represents statistically significant differences. The middle line represents mean values, boxes show standard deviation, and whiskers represent minimum and maximum values.

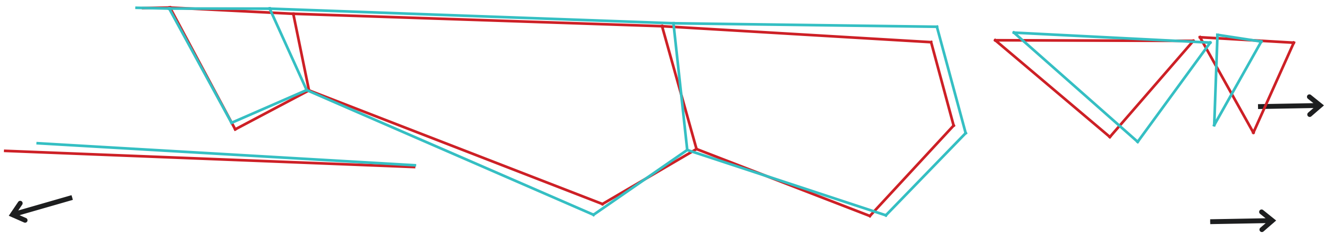

**Figure 5.** Transformation grids generated by CVA demonstrating skull shape deformations for females (red) and males (blue) of *C. flavolineatus* at the positive and negative extremes of the first canonical variate axis. The black arrows in each grid indicate the major changes in shape of females relative to males.

Information, Fig. S5C); and in males of *C. carinatus*, the snout is more robust, especially in the premaxilla (Supporting Information, Fig. S5F). In *C. bicarinatus*, *C. carinatus*, *C. exoletus*, and *C. quadricarinatus*, females and males share the same allometric trajectories (Supporting Information, Table S9).

## DISCUSSION

In our study, we identified distinct patterns of sexual dimorphism in head, body, tail, and head size, and skull both size and shape, in six of the twelve analysed species of *Chironius*.

### Body size

Sexual dimorphism in body size was observed in *C. bicarinatus*, *C. foveatus*, and *C. fuscus*, displaying males larger than females, and in *C. exoletus*, showing larger females. Larger body sizes in males can positively influence success during combat interactions, favouring their reproductive success (Shine 1994). This hypothesis was supported for colubrids from temperate areas and African vipers (Schuett 1997, Shine *et al.* 2000, Blouin-Demers *et al.* 2005, Glaudas *et al.* 2020). In Neotropical colubrines, studies addressing this issue are scarce. Sexual dimorphism favouring larger males in species that show combat behaviour has been reported for *Drymarchon couperi* (Holbrook, 1842) (Stevenson *et al.* 2009) and, in this study, for *C. bicarinatus.*

In the literature, there are descriptions of species that display males larger than females, despite having no reports of combat between males, as observed by Costa *et al.* (2013) for *Drymoluber dichrous* (Peters, 1863) and in this study for *C. foveatus* and *C. fuscus*. This pattern was previously reported for *C. fuscus* by Nascimento *et al.* (2013), suggesting the presence of combat behaviour in this species. However, these authors considered young and adult individuals in their analyses, making comparisons with our results unfeasible.

The larger body size of *C. exoletus* females may be directly related to the selective advantage of fecundity, considering the increase in space to accommodate a greater number of embryos (Darwin 1871, Trivers 1972, 1976). As for other Neotropical colubrines of which females are larger than males [such as *Mastigodryas boddaerti* (Sentzen, 1796), *Palusophis bifossatus* (Raddi, 1820), *Oxybelis fulgidus* (Daudin, 1803), and *Tantilla melanocephala* (Linnaeus, 1758)], the number of eggs produced by females was positively correlated with their body size, corroborating the hypothesis of advantages related to females fecundity (Darwin 1871, Santos-Costa *et al.* 2006, Leite *et al.* 2009, Scartozzoni *et al.* 2009, Pinto *et al.* 2010, Siqueira *et al.* 2012).

The absence of sexual dimorphism in body size is common among snakes and was observed in *C. brazili*, *C. carinatus*, *C. flavolineatus*, *C. gouveai*, *C. laevicollis*, *C. multiventris*, *C. quadricarinatus*, and *C. scurrulus*. Snakes that display combat behaviour could have an absence of sexual dimorphism in body size (Shine 1994). Several authors corroborate this suggesting that combat may occur in Neotropical colubrine species in which body size dimorphism is absent, as reported for *Spilotes pullatus* (Linnaeus, 1758) (Muniz-da-Silva *et al.* 2013), *Mastigophis flagellum* (Shaw, 1802) (Schuett and West 2020), *Dendrophidion dendrophis* (Schlegel, 1837) (Prudente *et al.* 2007), *Drymarchon corais* (Boie, 1827) (Prudente *et al.* 2014), *Drymoluber brazili* (Gomes, 1918) (Costa *et al.* 2014), *Oxybelis brevirostris* (Cope, 1861) (Montgomery *et al.* 2011), *Opheodrys aestivus* Linnaeus, 1766 (Goldsmith 1984), *Rhinobothryum lentiginosum* (Scopoli, 1785) (Arruda *et al.* 2015), and corroborated in this study for *C. carinatus.* Snakes are naturally difficult to find in nature, so it is possible that combat behaviour between males is more frequent than observed and may occur in *Chironius* species that did not show body size dimorphism. Whether they show this behaviour, dimorphism favouring larger males or the absence of dimorphism in body size is a consistent pattern among Neotropical colubrines.

### Tail length

Regarding caudal length, our results suggested sexual dimorphism in *C. fuscus*, *C. flavolineatus*, and *C. quadricarinatus*. The absence of sexual differences could be explained by the advantage of long tails to arboreal life (Lillywhite and Henderson 1993, Pizzatto and Marques 2007). Except for *C. laevicollis*, considered terrestrial, the other *Chironius* species are semi-arboreal (Dixon *et al.* 1993). A recent study with species of *Chironius* from the Atlantic Forest (*C. bicarinatus*, *C. exoletus*, *C. foveatus*, *C. fuscus*, and *C. laevicollis*) revealed that morphological variation and diet reflect different degrees of arboreality. *Chironius laevicollis* was the most terrestrial species among those studied by Banci *et al.* (2022), and despite mainly using the soil, it can occupy the lower strata of the vegetation, which could explain the absence of differences in tail length even by more terrestrial species. Our results on the absence of sexual dimorphism in tail length may reflect the importance of substrate use under morphology for both sexes, corroborating to the literature.

On the other hand, like *C. fuscus*, *C. flavolineatus*, and *C. quadricarinatus*, in the colubrine species *Drymoluber dichrous* and *Palusophis bifossatus*, males have longer tails than females (Leite *et al.* 2009, Costa *et al.* 2013, 2014). These species have urotomy, a tail breakage and a defence mechanism against predation (Costa *et al.* 2013, Dourado *et al.* 2013). Although there are no studies on caudal urotomy in *Chironius*, the greater tail length in males of *C. fuscus*, *C. flavolineatus*, and *C. quadricarinatus* could represent an advantageous defensive strategy, considering that males are more exposed to predation due to their more significant movement during the breeding season (Aldridge and Brown 1995).

The allometric analysis of tail length revealed an isometric pattern in males and females of all species. As previously discussed, this may corroborate that for tail-dimorphic species (*C. fuscus*, *C. flavolineatus*, and *C. quadricarinatus*), other pressures drive sexual dimorphism in this character, acting more strongly under males. Unexpectedly, males and females of *C. gouveai*, which did not differ in tail length nor show allometry, diverged in the allometric trajectories, which may suggest the role of sex-specific selection pressures (Howard 2009).

King's (1989) hypothesis that males of species with courtship behaviour have longer tails than males that combat was not supported, considering that both *C. bicarinatus* and *C. carinatus* had tails longer than *C. flavolineatus*. This prediction probably applies to species that use the caudal actively during courtship interaction, *Natrix helvetica*, which have a mating system based on aggregations and formation of mating 'balls' (Luiselli 1996). According to descriptions of courtship behaviour in *C. flavolineatus*, there are no indications of using the tail to move other males away from the female or encourage her in any way (Feio *et al.* 1999).

### Sexual dimorphism in head size

Sexual dimorphism in lizards favouring males with larger heads is suggested to be the result of sexual selection by intra- and intersexual competition (Bull and Pamula 1996, Kratochvíl and Frynta 2002, Gvozdík and Van Damme 2003). In snakes, sexual dimorphism in the head size and shape could be associated with dietary divergences, such as in *M. boddaerti*, which shows sexual differences in their intake of prey of different sizes (Shine 1991, Siqueira *et al.* 2012, Costa *et al.* 2013). However, when the trophic structure is used during interactions, male-biased dimorphism can evolve in response to sexual selection (Shine 1991).

*Chironius* species are known for their relatively homogeneous diet, primarily composed of frogs from the families Hylidae and Leptodactylidae (Nascimento *et al.* 2013, Roberto and Souza 2020, Banci *et al.* 2022). As such, the greater head length in males of *C. bicarinatus*, *C. foveatus*, and *C. fuscus* could result from sexual selection. Among these species, *C. bicarinatus* show the greatest variation in ingested prey (anurans, birds, and lizards) (Roberto and Souza 2020). However, in males that show combat behaviour, the head plays a relevant role considering that it participates along with the 'neck' when subjugating the opponent at combat (Almeida-Santos and Marques 2002). Therefore, sexual dimorphism in head size may have evolved in response to non-dietary pressures. If combat behaviour occurs in *C. foveatus* and *C. fuscus*, and if larger heads in males play an important role during the interaction, it is reasonable to expect that sexual dimorphism favouring males with larger heads in these species has evolved due to sexual selection. However, to strengthen this prediction, studies would be needed to indicate no sexual differences related to diet. On the other hand, in *C. exoletus*, sexual divergences in growth trajectories and intercepts suggest sex-specific growth patterns. Therefore, sexual dimorphism in this species likely results from allometric growth.

### Sexual dimorphism in head shape

Among the species with head shape dimorphism biased towards males (*C. bicarinatus*, *C. carinatus*, and *C. foveatus*), *C. bicarinatus*, a taxon with combat behaviour, exhibited the most significant broadening. More robust heads may be directly related to visual communication during combat, representing an intimidation mechanism by the male with a larger head. Among species that perform mating aggregations, larger males have an advantage over smaller ones, as the larger body size would exert dominance over the opponent, even with few evident signs of aggression and, such as the body, more robust heads could also play this role during intraspecific interactions (Schuett and Gillingham 1989, Ross and Marzec 1990).

Vulnerability to predation during reproductive activities may also drive the evolution of sexual dimorphism in the head (Shine 1990a). During the mating season, males move more, becoming more exposed to predators than females; in these cases, selection may favour the enlargement of trophic structures as a visual anti-predator stimulus (Shine 1990a, Aldridge and Brown 1995, Keogh *et al.* 2007, Dubey *et al.* 2008). In *Chironius* species, a series of defensive mechanisms have already been reported, including immobilization, tail vibration on the ground, cloacal discharge, dilation of the gular region, head triangulation, and delivering bites, among others (Santos-Costa *et al.* 2015, Muscat and Entiauspe-Neto 2016). Under these circumstances, it is unlikely that the head enlargement has evolved due to defensive mechanisms, considering the behavioural repertoire diversity already known among the genus species. We hypothesize that expanded heads may be related to visual communication during combat; however, we lack evidence to corroborate this hypothesis. In addition to visual communication, head expansion may also serve secondarily as an anti-predator visual stimulus.

The head enlargement in males may reflect differences concerning muscle mass. Sexual divergences were observed mainly in the orbitotemporal region, where the mandible adductor muscles are inserted, except for *C. bicarinatus* males, where enlargement generally occurred throughout the head (Zaher 1994). The same pattern reported for males was observed for females of *C. exoletus*, *C. fuscus*, and *C. laevicollis*. If the increased robustness in the head resulted from increased muscle mass, a more substantial bite force performance would favour both sexes. The greater head enlargement is a pattern compatible with anurophagy and may be associated with ingesting large and robust prey (Hampton 2014, Andjelković *et al.* 2016, Tamagnini *et al.* 2018). With the exception of *C. laevicollis*, the other dimorphic species, in addition to amphibians, can occasionally prey on considerably large and robust items: *C. bicarinatus*, *C. carinatus*, and *C. fuscus* can ingest birds, mammals, and reptiles, *C. foveatus* can prey on birds, and *C. exoletus* on reptiles (Roberto and Souza 2020, Banci *et al.* 2022).

Notably, although males of *C. fuscus* have longer heads, females have more considerable head enlargement. This pattern could not be associated with sexual differences in diet, considering the absence of differences between sex (Nascimento *et al.* 2013). Although enlargement provides greater bite force, which allows the ingestion of larger and more robust prey, it may also represent a decisive factor in prey handling time, which could favour *C. fuscus* females during prey ingestion (Verwaijen *et al.* 2002, Herrel *et al.* 2017).

Allometry could not explain the sexual variation in head shape in four (*C. bicarinatus*, *C. carinatus*, *C. exoletus*, and *C. foveatus*) of the six dimorphic species, since only one of the sexes shows allometry and the percentage at which allometry did explain shape changes was relatively small (6.5% for *C. bicarinatus* females, and 7.6%, 8%, and 10.2% for *C. carinatus*, *C. exoletus*, and *C. foveatus* males, respectively). In *C. bicarinatus*, *C. carinatus*, and *C. exoletus*, males and females share similar allometric trajectories ($P < 0.01$), that is, the covariation between shape and size is similar between sexes, which may corroborate our hypothesis that sexual dimorphism in head shape in these species may have evolved by sexual selection. Common allometric trajectories between sexes and allometry affecting only males were reported for *Vipera berus* (Linnaeus, 1758). The increased venom production in females was suggested as an explanation for the absence of head shape allometry in females (Tamagnini *et al.* 2018). Similar results have already been reported for other snakes, lizards, and mammals (Ljubisavljević *et al.* 2010, Milenković *et al.* 2010, Abegg *et al.* 2020).

On the other hand, *C. foveatus* allometry could explain sexual dimorphism in head shape, considering that the allometric trajectories were divergent between males and females, which indicates differential growth between sexes (Mitteroecker *et al.* 2013). Divergences in allometric slopes can also occur due to strong directional selection. Such direction changes of allometric slopes may represent adaptations to different functional or ecological demands (Klingenberg *et al.* 2010, Wilson and Sánchez-Villagra 2010, Wilson 2013, Pélabon *et al.* 2014). In *C. foveatus*, for example, the patterns of sexual dimorphism in shape, partly, are consistent with the allometric analyses, indicating an increase in the eye size in males, a result different from that found in other colubrids, where females exhibit a slightly larger absolute eye size (Faiman *et al.* 2018). *Chironius foveatus* is a highly arboreal species among the species of the genus (Banci *et al.* 2022). Daytime arboreal snakes are more exposed to predators, which can approach from all directions. Therefore they depend much more on vision than nocturnal snakes, hence the eyes of arboreal species being larger compared to those of terrestrial and semi-aquatic species (Lillywhite and Henderson 1993, Senter 1999, Liu *et al.* 2012). The larger eye size in *C. foveatus* males may have evolved due to greater predation pressure on males, who are generally more exposed (Aldridge and Brown 1995).

### Sexual dimorphism in cranial shape

In snakes with feeding specialization, sexual dimorphism in cranial shape can be explained by intersexual differences in feeding habits (Borczyk *et al.* 2021, Borczyk 2023). The only species that showed sexual dimorphism in the skull was *C. flavolineatus*. The greater cranial length and robustness in the males' snout (premaxillary) may confer an advantage during intraspecific interactions. In this species, the head is relevant to tactile communication during courtship interactions (Carpenter 1977, Mascarenhas *et al.* 2020). In snakes, a greater snout robustness could be associated with the digging habit (Haines 1967). However, there is no information that *Chironius* species use their heads for digging during their foraging activities, and these modifications may be related to the behaviour of stimulation by rubbing the chin in *C. flavolineatus*. Therefore, sexual dimorphism in the skull in *C. flavolineatus* may have evolved due to sexual selection due to interactions, considering their head's participation in the behavioural repertoire of courtship. For *C. flavolineatus*, isometry in the shape of the skull can corroborate the importance of sexual selection in determining the observed sexual differences.

Historically, the main hypothesis used to explain sexual dimorphism in the head of snakes refers to dietary divergence (Shine 1986, Camilleri and Shine 1990). However, in a genus containing species with intraspecific interactions, sexual dimorphism in both head and skull size may have evolved in response primarily to sexual selection. Patterns of sexual dimorphism in the head and skull shape indicate that distinct forces may determine sexual divergence. According to Shine (1990a, 1991), sexual selection can cause initial divergences between the sexes, and niche divergences can amplify these differences. The lack of information on sexual divergences in the diet, mating system, and natural history for most species limits a better understanding of the main forces involved in divergences between the sexes.

## CONCLUSION

Among all the analysed species, *C. fuscus* presented sexual divergences in a more significant number of characters, being, therefore, the most dimorphic species, with males differing from females in relation to body size, and head and tail length . This may suggest intense pressure associated with sexual selection, especially when considering that there are no diet-related sexual differences in this species.

In this study, we identified distinct patterns of sexual dimorphism in body size, tail, and head size, and skull size and shape in *Chironius*. Determining which forces drive sexual divergences involves a complex number of factors, many of which are difficult to measure mainly because, for many species, information about various aspects of life is reduced. Our study revealed sexual dimorphism patterns in one of the most observed genera in nature and is especially interesting due to intraspecific interactions. The patterns documented in this study serve as a valuable reference for exploring sexual divergences linked to behavioural aspects in snakes. However, these patterns may be even better understood after studies of sexual divergences on a dietary level (mainly for species that present intraspecific interactions), as well as observations of aspects of natural history (intraspecific interactions and mating behaviour) for those species in which information is still scarce.

## SUPPORTING INFORMATION

Supplementary data is available at the *Biological Journal of the Linnean Society* online.

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## CONFLICT OF INTEREST

There is no conflict of interest.

## AUTHOR CONTRIBUTIONS

Marina Meireles dos Santos (collected and analysed the data, and wrote the manuscript), Ana Lúcia da Costa Prudente (revised the English, edited and approved the manuscript), and Julia Klaczko (edited and approved the manuscript)

## DATA AVAILABILITY

The data underlying this article will be shared on reasonable request to the corresponding author.

## REFERENCES

Abegg AD, Passos P, Mario-da-Rosa C *et al.* Sexual dimorphism, ontogeny and static allometry of a semi-fossorial snake (genus *Atractus*). *Zoologischer Anzeiger* 2020;**287**:95–104. https://doi.org/10.1016/j.jcz.2020.05.008

Aldridge RD, Brown WS. Male reproductive cycle, age at maturity, and cost of reproduction in the timber rattlesnake (*Crotalus horridus*). *Journal of Herpetology* 1995;**29**:399–407. https://doi.org/10.2307/1564990

Almeida-Santos SM, Marques OAV. Male-male ritual combat in the colubrid snake *Chironius bicarinatus* from the Atlantic Forest, southeastern Brazil. *Amphibia-Reptilia* 2002;**23**:528–33.

Almeida-Santos SM, Salomão MG, Peneti EA *et al.* Predatory combat and tail wrestling in hierarchical contests of the Neotropical rattlesnake *Crotalus durissus terrificus* (Serpentes: Viperidae). *Amphibia-Reptilia* 1999;**20**:88–96.

Andjelković M, Tomović L, Ivanović A. Variation in skull size and shape of two snake species (*Natrix natrix* and *Natrix tessellata*). *Zoomorphology* 2016;**135**:243–53. https://doi.org/10.1007/s00435-016-0301-3.

Arruda L, Carvalho M, Kawashita-Ribeiro R. New records of the Amazon banded snake *Rhinobothryum lentiginosum* (Serpentes: Colubridae) from Mato Grosso state, Brazil, with natural history notes. *Salamandra* 2015;**51**:199–205.

Banci KRS, Guimarães M, Siqueira LHC *et al.* Body shape and diet reflect arboreality degree of five congeneric snakes sympatric in the Atlantic forest. *Biotropica* 2022;**54**:839–51.

Blouin-Demers G, Gibbs HL, Weatherhead PJ. Genetic evidence for sexual selection in black ratsnakes (*Elaphe obsoleta*). *Animal Behaviour* 2005;**69**:225–34. https://doi.org/10.1016/j.anbehav.2004.03.012

Bonduriansky R. Sexual selection and allometry: a critical reappraisal of the evidence and ideas. *Evolution* 2007;**61**:838–49. https://doi.org/10.1111/j.1558-5646.2007.00081.x

Borczyk B. Sexual dimorphism in skull size and shape of *Laticauda colubrina* (Serpentes: Elapidae). *PeerJ* 2023;**11**:e16266. https://doi.org/10.7717/peerj.16266

Borczyk B, Paśko L, Kusznierz J *et al.* Sexual dimorphism and skull size and shape in the highly specialized snake species, *Aipysurus eydouxii* (Elapidae: Hydrophiinae). *PeerJ* 2021;**9**:e11311. https://doi.org/10.7717/peerj.11311

Bull CM, Pamula Y. Sexually dimorphic head sizes and reproductive success in the sleepy lizard *Tiliqua rugosa*. *Journal of Zoology* 1996;**240**:511–21. https://doi.org/10.1111/j.1469-7998.1996.tb05302.x

Camilleri C, Shine R. Sexual dimorphism and dietary divergence: differences in trophic morphology between male and female snakes. *Copeia* 1990;**1990**:649–65. https://doi.org/10.2307/1446430

Carpenter CC. Communication and displays of snakes. *American Zoologist* 1977;**17**:217–23. https://doi.org/10.1093/icb/17.1.217

Clark DR. Notes on sexual dimorphism in tail-length in American snakes. *Transactions of the Kansas Academy of Science* 1967;**69**:226–32. https://doi.org/10.2307/3627419

Costa HC, Moura MR, Feio RN. Taxonomic revision of *Drymoluber* Amaral, 1930 (Serpentes: Colubridae). *Zootaxa* 2013;**3716**:349–94. https://doi.org/10.11646/zootaxa.3716.3.3

Costa HC, Moura MR, Feio RN. A tale of lost tails: pseudoautotomy in the Neotropical snake genus *Drymoluber* (Serpentes: Colubridae). *Canadian Journal of Zoology* 2014;**92**:811–6. https://doi.org/10.1139/cjz-2014-0115

Darwin C. *The Descent of Man, and Selection in Relation to Sex*. London: John Murray, 1871.

Dixon JR, Wiest J, Cei JM. *Revision of the Neotropical Snake Genus Chironius Fitzinger (Serpentes, Colubridae). Monografie XIII*. Torino: Museo Regionale di Scienze Naturali, 1993.

Dourado ACM, Oliveira L, Prudente ALC. Pseudoautotomy in *Dendrophidion dendrophis* and *Mastigodryas bifossatus* (Serpentes: Colubridae): tail morphology and breakage frequency. *Copeia* 2013;**2013**:132–41. https://doi.org/10.1643/ch-12-008

Dubey S, Brown G, Madsen T *et al.* Male-biased dispersal in a tropical Australian snake (*Stegonotus cucullatus*, Colubridae). *Molecular Ecology* 2008;**17**:3506–14.

Dubey S, Brown GP, Madsen T *et al.* Sexual selection favours large body size in males of a tropical snake (*Stegonotus cucullatus*, Colubridae). *Animal Behaviour* 2009;**77**:177–82. https://doi.org/10.1016/j.anbehav.2008.09.037

Entiauspe-Neto OM, Lyra ML, Koch C *et al.* Taxonomic revision of *Chironius bicarinatus* (Wied 1820) (Serpentes: Colubridae), with description of a new species. *Herpetological Monographs* 2020;**34**:98–115.

Faiman R, Abergil D, Babocsay G *et al.* A review of sexual dimorphism of eye size in Colubroidea snakes. *Vertebrate Zoology* 2018;**68**:91–108. https://doi.org/10.3897/vz.68.e32229

Feio R, Santos PS, Fernandes R *et al. Chironius flavolineatus*. Courtship. *Herpetological Review* 1999;**30**:99.

Fox J, Weisberg S. *An R Companion to Applied Regression*, 3rd edn. Thousand Oaks, CA: Sage, 2019. https://socialsciences.mcmaster.ca/jfox/Books/Companion/

Glaudas X, Rice SE, Clark RW *et al.* The intensity of sexual selection, body size and reproductive success in a mating system with male–male combat: is bigger better? *Oikos* 2020;**129**:998–1011. https://doi.org/10.1111/oik.07223

Goldsmith SK. Aspects of the natural history of the rough green snake, *Opheodrys aestivus* (Colubridae). *The Southwestern Naturalist* 1984;**29**:445–52. https://doi.org/10.2307/3670997

Greene MJ, Mason RT. Courtship, mating, and male combat of the brown tree snake, *Boiga irregularis*. *The Herpetological Journal* 2000;**56**:166–75.

Gvozdík L, Van Damme R. Evolutionary maintenance of sexual dimorphism in head size in the lizard *Zootoca vivipara*: a test of two hypotheses. *Journal of Zoology* 2003;**259**:7–13. https://doi.org/10.1017/s0952836902003308

Haines TP. Variations of colubrid skulls, their correlations and their value in taxonomy. *Herpetologica* 1967;**23**:142–5.

Hamdan B, Pereira AG, Loss-Oliveira L *et al.* Evolutionary analysis of *Chironius* snakes unveils cryptic diversity and provides clues to diversification in the Neotropics. *Molecular Phylogenetics and Evolution* 2017;**116**:108–19. https://doi.org/10.1016/j.ympev.2017.08.004

Hampton P. Allometry of skull morphology, gape size and ingestion performance in the banded watersnake (*Nerodia fasciata*) feeding on two types of prey. *Journal of Experimental Biology* 2014;**217**:472–8.

Herrel A, Petrochic S, Draud M. Sexual dimorphism, bite force and diet in the diamondback terrapin. *Journal of Zoology* 2017;**304**:217–24. https://doi.org/10.1111/jzo.12520

Hervé M. *RVAideMemoire: Testing and Plotting Procedures for Biostatistics*. R package version 0.9-79, 2021.

Hollis JL. Phylogenetics of the genus *Chironius* Fitzinger, 1826 (Serpentes, Colubridae) based on morphology. *Herpetologica* 2006;**62**:435–53. https://doi.org/10.1655/0018-0831(2006)62[435:potgcf]2.0.co;2

Houston DL, Shine R. Sexual dimorphism and niche divergence: feeding habits of the arafura filesnake. *Journal of Animal Ecology* 1993;**62**:737–48. https://doi.org/10.2307/5393

Howard RD. Ontogeny of a sexual dimorphism in tiger salamanders. *Canadian Journal of Zoology* 2009;**87**:573–80. https://doi.org/10.1139/z09-043

Kaufman GA, Gibbons JW. Weight length relationships in thirteen species of snakes in the southeastern United States. *Herpetologica* 1975;**31**:31–7. https://www.jstor.org/stable/3891983

Keogh JS, Webb JK, Shine R. Spatial genetic analysis and long-term mark–recapture data demonstrate male-biased dispersal in a snake. *Biology Letters* 2007;**3**:33–5. https://doi.org/10.1098/rsbl.2006.0570

King RB. Sexual dimorphism in snake tail length: sexual selection, natural selection, or morphological constraint. *Biological Journal of the Linnean Society* 1989;**38**:133–54. https://doi.org/10.1111/j.1095-8312.1989.tb01570.x

Klaczko J, Montingelli GG, Zaher H. A combined morphological and molecular phylogeny of the genus *Chironius* Fitzinger, 1826 (Serpentes: Colubridae). *Zoological Journal of the Linnean Society* 2014;**171**:656–67. https://doi.org/10.1111/zoj.12147

Klauber LM. Tail-length in snakes, with notes on sexual dimorphism and the coefficient of divergence. In: Klauber LM (ed.), *Tail-length in Snakes, with Notes on Sexual Dimorphism and the Coefficient of Divergence and a Graphic Method of Showing Relationships*. San Diego, CA: Zoological Society of San Diego, 1943, 1–60.

Klingenberg CP. MorphoJ: an integrated software package for geometric morphometrics. *Molecular Ecology Resources* 2011;**11**:353–7. https://doi.org/10.1111/j.1755-0998.2010.02924.x

Klingenberg CP. Methods for studying allometry in geometric morphometrics: a comparison of performance. *Evolutionary Ecology* 2022;**36**:439–70. https://doi.org/10.1007/s10682-022-10170-z

Klingenberg CP, McIntyre GS. Geometric morphometrics of developmental instability: analyzing patterns of fluctuating asymmetry with Procrustes methods. *Evolution* 1998;**52**:1363–75. https://doi.org/10.1111/j.1558-5646.1998.tb02018.x

Klingenberg CP, Monteiro LR. Distances and directions in multidimensional shape spaces: implications for morphometric applications. *Systematic Biology* 2005;**54**:678–88. https://doi.org/10.1080/10635150590947258

Klingenberg CP, Barluenga M, Meyer A. Shape analysis of symmetric structures: quantifying variation among individuals and asymmetry. *Evolution* 2002;**56**:1909–20.

Klingenberg CP, Debat V, Roff DA. Quantitative genetics of shape in cricket wings: developmental integration in a functional structure. *Evolution* 2010;**64**:2935–51. https://doi.org/10.1111/j.1558-5646.2010.01030.x

Kratochvíl L, Frynta D. Body size, male combat and the evolution of sexual dimorphism in eublepharid geckos (Squamata: Eublepharidae). *Biological Journal of the Linnean Society* 2002;**76**:303–14. https://doi.org/10.1111/j.1095-8312.2002.tb02089.x

Leite PT, Nunes SF, Kaefer IG *et al.* Reproductive biology of the swamp racer *Mastigodryas bifossatus* (Serpentes: Colubridae) in subtropical Brazil. *Zoologia* 2009;**26**:12–8.

Lillywhite HB, Henderson RW. Behavioral and functional ecology of arboreal snakes. In: Seigel RA, Collins JT (eds), *Snakes: Ecology and Behavior*. New York, NY: McGraw-Hill, 1993, 1–48.

Liu Y, Ding L, Lei J *et al.* Eye size variation reflects habitat and daily activity patterns in colubrid snakes. *Journal of Morphology* 2012;**273**:883–93. https://doi.org/10.1002/jmor.20028

Ljubisavljević K, Urošević A, Aleksić I *et al.* Sexual dimorphism of skull shape in a lacertid lizard species (*Podarcis* spp., *Dalmatolacerta* sp., *Dinarolacerta* sp.) revealed by geometric morphometrics. *Zoology* 2010;**113**:168–74. https://doi.org/10.1016/j.zool.2009.09.003

Luiselli L. Individual reproductive success in mating balls of the grass snake, *Natrix natrix*: size is important. *Journal of Zoology* 1996;**239**:731–740.

Mascarenhas W, Oliveira CR, Ávila RW *et al.* Defensive behaviour of *Chironius flavolineatus* Jan, 1863 (Serpentes: Colubridae) in northeastern Brazil. *Herpetology Notes* 2020;**13**:607–8.

Milenković M, Jojić-Šipetić V, Blagojević J *et al.* Skull variation in Dinaric-Balkan and Carpathian gray wolf populations revealed by geometric morphometric approaches. *Journal of Mammalogy* 2010;**91**:376–86. https://doi.org/10.1644/09-MAMM-A-265.1

Missassi AFR, Coeti RZ, Almeida-Santos SM *et al.* Intense male-male ritual combat in the *Micrurus ibiboboca* complex (Elapidae) from northeastern South America. *Herpetological Conservation and Biology* 2022;**17**:204–16.

Mitteroecker P, Gunz P, Windhager S *et al.* A brief review of shape, form, and allometry in geometric morphometrics, with applications to human facial morphology. *Hystrix* 2013;**24**:59–66.

Montgomery CE, Lips KR, Ray JM. Ontogenetic shift in height of sleeping perches of Cope's vine snake, *Oxybelis brevirostris*. *The Southwestern Naturalist* 2011;**56**:358–62. https://doi.org/10.1894/f08-rw-02.1

Muniz-da-Silva DF, Almeida-Santos SM. Male-male ritual combat in *Spilotes pullatus* (Serpentes: Colubrinae). *The Herpetological Bulletin* 2013;**126**:25–9.

Murta-Fonseca RA, Machado A, Lopes RT *et al.* Sexual dimorphism in *Xenodon neuwiedii* skull revealed by geometric morphometrics (Serpentes: Dipsadidae). *Amphibia-Reptilia* 2019;**40**:461–74. https://doi.org/10.1163/15685381-20191147

Muscat E, Entiauspe-Neto OM. *Chironius foveatus* (Atlantic Forest sipo snake) defensive behavior. *Herpetologica* 2016;**47**:475.

Nascimento LP, Siqueira DM, Santos-Costa MC. Diet, reproduction, and sexual dimorphism in the vine snake, *Chironius fuscus* (Serpentes: Colubridae), from Brazilian Amazonia. *South American Journal of Herpetology* 2013;**8**:168–74.

Pélabon C, Firmat C, Bolstad GH *et al.* Evolution of morphological allometry. *Annals of the New York Academy of Sciences* 2014;**1320**:58–75. https://doi.org/10.1111/nyas.12470

Pinto RR, Marques OAV, Fernandes R. Reproductive biology of two sympatric colubrid snakes, *Chironius flavolineatus* and *Chironius quadricarinatus*, from the Brazilian Cerrado domain. *Amphibia-Reptilia* 2010;**31**:463–73. https://doi.org/10.1163/017353710x518423

Pizzatto L, Marques OAV. Reproductive ecology of boine snakes with emphasis on Brazilian species and comparison to pythons. *South American Journal of Herpetology* 2007;**2**:107–22. https://doi.org/10.2994/1808-9798(2007)2[107:reobsw]2.0.co;2

Pizzatto L, Almeida-Santos SM, Marques OAV. Biologia reprodutiva de serpentes brasileiras. In: Nascimento LB, Oliveira ME (eds), *Herpetologia no Brasil II*. Belo Horizonte: Sociedade Brasileira de Herpetologia, 2006, 201–221.

Prudente ALC, Maschio GF, Yamashina CE *et al.* Morphology, reproductive biology and diet of *Dendrophidion dendrophis* (Schlegel, 1837) (Serpentes, Solubridae) in Brazilian Amazon. *South American Journal of Herpetology* 2007;**2**:53–8.

Prudente ALC, Menks AC, Silva FM *et al.* Diet and reproduction of the western indigo snake *Drymarchon corais* (Serpentes: Colubridae) from the Brazilian Amazon. *Herpetology Notes* 2014;**7**:99–108.

R Core Team. *R: a Language and Environment for Statistical Computing*. Vienna: R Foundation for Statistical Computing, 2021. https://www.R-project.org/

Roberto IJ, Souza AR. Review of prey items recorded for snakes of the genus *Chironius* (Squamata, Colubridae), including the first record of *Osteocephalus* as prey. *Herpetology Notes* 2020;**13**:1–5.

Rohlf FJ. *tpsUtil, version 1.81*. Stony Brook, NY: Department of Ecology and Evolution, State University of New York, 2008.

Rohlf FJ. *TpsDig2, version 2.32*. Stony Brook, NY: Department of Ecology and Evolution, State University of New York, 2015.

Ross AR, Marzec G. *Reproductive Husbandry of Pythons and Boas*. 1st edn. Santa Barbara, CA: Institute of Herpetological Research, 1990.

Sanger TJ, Sherratt E, McGlothlin JW *et al.* Convergent evolution of sexual dimorphism in skull shape using distinct developmental strategies. *Evolution* 2013;**67**:2180–93. https://doi.org/10.1111/evo.12100

Santos MM, Klaczko J, Prudente ALC. Sexual dimorphism and allometry in malacophagus snakes (Dipsadidae: Dipsadinae). *Zoology* 2022;**153**:126026.

Santos-Costa MC, Maschio GF, Prudente ALC. Natural history of snakes from Floresta Nacional de Caxiuanã, eastern Amazonia, Brazil. *Herpetology Notes* 2015;**8**:69–98.

Santos-Costa MC, Prudente ALC, Di-Bernardo M. Reproductive biology of *Tantilla melanocephala* (Linnaeus, 1758) (Serpentes, Colubridae) from eastern Amazonia, Brazil. *Journal of Herpetology* 2006;**40**:553–6.

Scartozzoni RR, Salomão MG, Almeida-Santos SM. Natural history of the vine snake *Oxybelis fulgidus* (Serpentes, Colubridae) from Brazil. *South American Journal of Herpetology* 2009;**4**:81–9.

Schuett GW. Body size and agonistic experience affect dominance and mating success in male copperheads. *Animal Behaviour* 1997;**54**:213–24. https://doi.org/10.1006/anbe.1996.0417

Schuett GW, Gillingham JC. Male-male agonistic behaviour of the copperhead, *Agkistrodon contortrix*. *Amphibia-Reptilia* 1989;**10**:243–66. https://doi.org/10.1163/156853889x00412

Schuett GW, West Z. Natural history notes: *Masticophis flagellum* (Coachwhip). Male combat. *Herpetological Review* 2020;**51**:353–5.

Semlitsch RD, Gibbons JW. Body size dimorphism and sexual selection in two species of water snakes. *Copeia* 1982;**4**:974–6. https://doi.org/10.2307/1444113

Senter P. The visibility hypothesis: an explanation for the correlation between habitat and defensive display in snakes. *Herpetological Review* 1999;**30**:213.

Senter P. Phylogeny of courtship and male-male combat behavior in snakes: an updated analysis. *Current Herpetology* 2022;**41**:35–81.

Senter P, Harris SM, Kent DL. Phylogeny of courtship and male-male combat behavior in snakes. *PLoS One* 2014;**9**:e107528. https://doi.org/10.1371/journal.pone.0107528

Shine R. Sexual size dimorphism and male combat in snakes. *Oecologia* 1978;**33**:269–77. https://doi.org/10.1007/BF00348113

Shine R. "Costs" of reproduction in reptiles. *Oecologia* 1980;**46**:92–100. https://doi.org/10.1007/BF00346972

Shine R. Sexual differences in morphology and niche utilization in an aquatic snake, *Acrochordus arafurae*. *Oecologia* 1986;**69**:260–7. https://doi.org/10.1007/BF00377632

Shine R. Ecological causes for the evolution of sexual dimorphism: a review of the evidence. *The Quarterly Review of Biology* 1990a;**64**:419–61. https://doi.org/10.1086/416458

Shine R. Proximate determinants of sexual differences in adult body size. *The American Naturalist* 1990b;**135**:278–83. https://doi.org/10.1086/285043

Shine R. Intersexual dietary divergence and the evolution of sexual dimorphism in snakes. *The American Naturalist* 1991;**138**:103–22. https://doi.org/10.1086/285207. https://www.jstor.org/stable/2462535

Shine R. Sexual size dimorphism in snakes revisited. *Copeia* 1994;**1994**:326–46. https://doi.org/10.2307/1446982

Shine R, Olsson MM, Lemaster MP *et al*. Effects of sex, body size, temperature, and location on the antipredator tactics of free-ranging garter snakes (*Thamnophis sirtalis*, Colubridae). *Behavioral Ecology* 2000;**11**:239–45. https://doi.org/10.1093/beheco/11.3.239

Signorell A, Aho K, Alfons A *et al*. *DescTools: Tools for descriptive statistics*. R package version 0.99.45, 2022.

Siqueira DM, Nascimento LP, Santos-Costa MC. Feeding biology of Boddaert's tropical racer, *Mastigodryas boddaerti* (Serpentes, Colubridae) from the Brazilian Amazon. *South American Journal of Herpetology* 2012;**7**:226–32.

Slip DJ, Shine R. Habitat use, movements, and activity patterns of free-ranging diamond pythons *Morelia spilota spilota* (Serpentes: Boidae): a radiotelemetric study. *Australian Wildlife Research* 1988;**15**:515–31.

Starace F. *Guide des serpentes et amphibenes de Guyane Française*. Guadeloupe: Ibis Rouge Editions, 1998.

Stevenson DJ, Enge KM, Carlile LD *et al*. An eastern indigo snake (*Drymarchon couperi*) mark-recapture study in southeastern Georgia. *Herpetological Conservation and Biology* 2009;**4**:30–42.

Strauss RE. Evolutionary allometry and variation in body form in the South American catfish genus *Corydoras* (Callichthyidae). *Systematic Biology* 1985;**34**:381–96. https://doi.org/10.1093/sysbio/34.4.381

Tamagnini D, Stephenson J, Brown RP *et al*. Geometric morphometric analyses of sexual dimorphism and allometry in two sympatric snakes: *Natrix helvetica* (Natricidae) and *Vipera berus* (Viperidae). *Zoology* 2018;**129**:25–34. https://doi.org/10.1016/j.zool.2018.05.008

Torres-Carvajal O, Echevarría LY, Lobos SE *et al*. Phylogeny, diversity and biogeography of Neotropical sipo snakes (Serpentes: Colubrinae: *Chironius*). *Molecular Phylogenetics and Evolution* 2019;**130**:315–29. https://doi.org/10.1016/j.ympev.2018.10.022

Trivers RL. Parental investment and sexual selection. In: Campbell B (ed.), *Sexual Selection and the Descent of Man*. Chicago, IL: Aldine, 1972.

Trivers RL. Sexual selection and resource-accruing abilities in *Anolis garmani*. *Evolution* 1976;**30**:253–69. https://doi.org/10.1111/j.1558-5646.1976.tb00908.x

Uetz P, Hallerman J. *The Reptile Database*. 2021. http://reptile-database.reptarium.cz

Verwaijen D, Van Damme R, Herrel A. Relationships between head size, bite force, prey handling efficiency and diet in two sympatric lacertid lizards. *Functional Ecology* 2002;**16**:842–50. https://doi.org/10.1046/j.1365-2435.2002.00696.x

Warton DI, Duursma RA, Falster DS *et al*. Smatr 3- an R package for estimation and inference about allometric lines. *Methods in Ecology and Evolution* 2012;**3**:257–9. https://doi.org/10.1111/j.2041-210X.2011.00153.x

Wickham H. *ggplot2: Elegant Graphics for Data Analysis*. New York, NY: Springer-Verlag, 2016.

Wilson LAB. Allometric disparity in rodent evolution. *Ecology and Evolution* 2013;**3**:971–84. https://doi.org/10.1002/ece3.521

Wilson LAB, Sánchez-Villagra MR. Diversity trends and their ontogenetic basis: an exploration of allometric disparity in rodents. *Proceedings Biological Sciences* 2010;**277**:1227–34. https://doi.org/10.1098/rspb.2009.1958

Zaher H. Comments on the evolution of the jaw adductor musculature of snakes. *Zoological Journal of the Linnean Society* 1994;**111**:339–84. https://doi.org/10.1111/j.1096-3642.1994.tb01488.x