

RESEARCH ARTICLE

Osteological postcranial traits in hylid anurans indicate a morphological continuum between swimming and jumping locomotor modes

Mónica Soliz¹ | María J. Tulli²  | Virginia Abdala³

¹CONICET-Universidad Nacional de Salta (UNSa), Facultad de Ciencias Naturales, Cátedra Vertebrados, Salta, Argentina

²Unidad Ejecutora Lillo (UEL)-CONICET-Instituto de Herpetología, Fundación Miguel Lillo, Tucumán, Argentina

³Instituto de Biodiversidad Neotropical (IBN)-CONICET, Universidad Nacional de Tucumán (UNT), Facultad de Ciencias Naturales, Cátedra Biología General, Tucumán, Argentina

Correspondence

Mónica Carina Soliz, Cátedra de Vertebrados, Fac. de Cs. Naturales, UNSa-CONICET, Av. Bolivia 5150, Salta, Argentina.

Email: monica.c.soliz@gmail.com

María José Tulli, UEL-CONICET- Instituto de Herpetología- Fundación Miguel Lillo, Tucumán, Argentina.

Email: majotulli@gmail.com

Abstract

Anurans exhibit a particularly wide range of locomotor modes that result in wide variations in their skeletal structure. This article investigates the possible correlation between morphological aspects of the hylid postcranial skeleton and their different locomotor modes and habitat use. To do so, we analyzed 18 morphometric postcranial variables in 19 different anuran species representative of a variety of locomotor modes (jumper, hopper, walker, and swimmer) and habitat uses (arboreal, bush, terrestrial, and aquatic). Our results show that the evolution of the postcranial hylid skeleton cannot be explained by one single model, as for example, the girdles suggest modular evolution while the vertebral column suggests other evolutionary modules. In conjunction with data from several other studies, we were able to show a relationship between hylid morphology and habitat use; offering further evidence that the jumper/swimmer and walker/hopper locomotor modes exhibit quite similar morphological architecture. This allowed us to infer that new locomotor modalities are, in fact, generated along a morphological continuum.

KEYWORDS

ecomorphology, frogs, habitat use, postcranial skeleton, skeleton

1 | INTRODUCTION

While considerable ecomorphological research has been conducted in the field of herpetology (Bauwens, Garland, Castilla, & Van Damme, 1995; Cruz et al., 2009; Herrel, Vasilopoulou-Kampitsi, & Bonneaud, 2014; Losos, 1990a, 1990b, 1990c; Losos, Andrews, Sexton, & Schuler, 1991; Tulli, Abdala, & Cruz, 2011), there is a gross imbalance between the extensive focus on squamates (Goodman, 2006; Goodman, Miles, & Schwarzkopf, 2008; Herrel, Vanhooydonck, Porck, & Irschick, 2008; Herrel et al., 2014; Kohlsdorf, Garland, & Navas, 2001; Kohlsdorf et al., 2004; Losos, 1990a, 1990b, 1990c; Losos, Jackman, Larson, de Queiroz, & Rodríguez-Schettino, 1998; Losos, Walton, & Bennett, 1993; Schulte, Losos, Cruz, & Nuñez, 2004; Tulli, Abdala, & Cruz, 2012; Tulli, Cruz, Herrel, Vanhooydonck, & Abdala, 2009; Tulli, Cruz, Kohlsdorf, & Abdala, 2016; Tulli et al., 2011; Vanhooydonck & Van Damme, 2011; Zani, 2000; among many others) and anurans (Aerts & Nauwelaerts, 2009; Emerson, 1979, 1988; Gillis & Biewener, 2000; Griep et al., 2013; Herrel et al., 2014; Jorgensen & Reilly, 2013; Nauwelaerts & Aerts, 2006; Nauwelaerts, Aerts, & D' Aout, 2001;

Nauwelaerts, Ramsay, & Aerts, 2007; Reilly & Jorgensen, 2011; Simons, 2008; Wells, 2007), with few of these papers addressing the biomechanics of locomotion in frogs (e.g., Bijma, Gorb, & Kleinteich, 2016; Cox & Gillis, 2015; Griep et al., 2013). This is especially surprising when considering the plethora of locomotor modes and habitat uses exhibited by anurans. Their ability to swim, hop, and walk allow them to inhabit arboreal, aquatic, and terrestrial environments and make them an ideal group to highlight relationships between morphology and ecology.

Simons (2008) suggested that relatively long hind limbs and short forelimbs coupled with a tail less, stout body define the conservative bauplan of anurans. However, the evolutionary study of the interaction between each particular organism and its biotic and abiotic environment illustrates how selection can mold specific phenotypic variations (Irschick, Meyers, Husak, & Le Galliard, 2008). For example, subtle structural differences in hind limb proportion could facilitate functional diversity, allowing for the exploitation of a broader range of environments (Bain, Lathrop, Murphy, Nikolai, & Cuc, 2003; Channing & Broadley, 2002; Dobrowolska, 1973; Duellman & Trueb, 1994;

Emerson, 1978, 1979, 1983, 1985; Emerson & De Jongh, 1980; Nauwelaerts et al., 2007; Zug, 1972, 1978). For example, habitat or microhabitat use, including interspecific or intraspecific interactions, competition, predators may also contribute to diversification (McPeck & Brown, 2000). Collar, Schulte, & Losos (2011) postulated that the locomotor functions demanded by a certain habitat may also vary depending on body size, adding another layer of complexity to the relationship between traits across habitats. For instance, the limb proportions of lizards such as the Caribbean *Anolis* are strongly associated with the particular habitat use on each of the islands they colonized, with arboreal anoles that live on narrow branches exhibiting shorter legs (Losos, 2009). Likewise in anuran species, variations in hind limb morphology appears to be strongly associated with habitat use, although the correlation has not been studied extensively (Enriquez-Urzelai, Montori, Llorente, & Kaliontzopoulou, 2015; Gomes, Rezende, Grizante, & Navas, 2009).

According to Emerson (1988), the same postcranial architecture has appeared repeatedly and independently across several families of anurans. The vertebral column and pelvic girdle play pivotal roles in locomotion and are both considered key factors for adaptation (Banbury & Maglia, 2006; Emerson, 1978, 1982, 1988; Jenkins & Shubin, 1998; Jorgensen & Reilly, 2013; Púgener & Maglia, 2009; Sheil & Alamillo, 2005; Simons, 2008; Trueb & Báez, 2006). Postcranial elements, such as the ilio-sacral articulation, permit distinct joint mobility patterns and, consequently, different locomotor behaviors (Emerson, 1979).

From a biomechanical point of view, the architecture of anurans suggests that they are specialized jumpers. Furthermore, an adaptation for one locomotor mode can result in a direct conflict with the performance of another (Cartmill, 1985; Kramer, 1951; Losos et al., 1993; Miles, 1994; Peterson, 1984). A better understanding of the functional significance of certain osteological traits could greatly enrich the interpretation of the interactions between these traits and habitat use or locomotor modes (Arnold, 1983).

All species selected for our focus group belong to the Hylidae, one of the most diverse anuran families with nearly 900 species described to date (Frost, 2016). Hylids are known for inhabiting a wide variety of environments, from ponds and streams to tropical rainforests and grasslands. Moreover, some species are strictly aquatic or specialized terrestrial or arboreal dwellers, while others are equally at home on land and in the water (Borges de Freitas et al., 2008; Cei, 1980; Duellman, 1970, 2001, 2003; Duré, 1999; Gallardo, 1993; Lucas, Fortes, & García, 2016; Pombal & Haddad, 1992; Vaira, 2001; Wells, 2007). Accordingly, hylids incorporate various locomotor modes (Manzano, Baldo, & Barg, 2004; Manzano & Barg, 2005; Vaira, 2001; Wells, 2007). The versatility and morphological diversity of this family make it ideal for ecomorphological research, which traditionally focuses on the locomotor system and its correlation with postcranial skeletal traits (e.g., jumping performance in relation to hind limb length; Emerson, 1979, 1988; Rand, 1952; Zug, 1972, 1978). In this context, we return to the question of whether relatively subtle changes in the morphology of frog's postcranial bones could have functional consequences, as occurs in other small bodied vertebrates such as rodents (Carrizo, Tulli, Dos

Santos, & Abdala, 2013). The main objectives of this study were to investigate whether (i) variations in osteological postcranial structures of select hylids indicate a specific evolutionary model, and whether (ii) the vertebral column, pectoral, and pelvic girdles of the selected species exhibit correlations between morphological patterns and ecological aspects such as locomotor mode and/or habitat use. Based on the outcomes of several ecomorphological papers studying the internal anatomy across different tetrapod taxa (Carrizo et al., 2013; Nauwelaerts et al., 2007; Tulli, Carrizo, & Samuels, 2015; Tulli et al., 2009, 2011, 2016), we hypothesize that hylid frogs will present a conservative postcranial morphology adequate for all those tasks requiring locomotion.

2 | MATERIALS AND METHODS

2.1 | Data collection

We examined 51 adult anuran specimens of both sexes, from 14 hylid species: *Hypsiboas riojanus* (Koslowky, 1895), *Hypsiboas raniceps* (Cope, 1862), *Hypsiboas pulchellus* (Dumeril & Bibron, 1841), *Scinax acuminatus* (Cope, 1862), and *Scinax fuscovarius* (Lutz, 1925), *Scinax nasicus* (Cope, 1862), *Dendropsophus nanus* (Boulenger, 1889), *Trachycephalus typhonius* (Linnaeus, 1758), *Lysapsus limellum* (Cope, 1862), *Pseudis platensis* (Gallardo, 1961), *Phyllomedusa azurea* (Cope, 1862), *Phyllomedusa boliviana* (Boulenger, 1902), *Phyllomedusa sauvagii* (Boulenger, 1882), and *Phyllomedusa tetraploidea* (Pombal & Haddad, 1992). In addition, five species of others families were included in the analysis: *Leptodactylus fuscus* (Schneider, 1799), *Pleurodema cinereum* (Cope, 1878), *Rhinella major* (Müller & Helmich, 1936), *Telmatobius ceiorum* (Laurent, 1970), *Telmatobius atacamensis* (Gallardo, 1962; for detail see Figure 1). Specimens were selected to represent a wide range of locomotor modes and habitat uses, including several specialized taxa belonging to independent evolutionary lineages. All the specimens examined are housed in systematic collections and listed in the Supporting Information (Appendix S1).

2.2 | Morphology

Initially, an osteological comparative analysis was performed to select specific characters conducive to the assessment of the degree of morphological specialization (Table 1). The vertebral column and pectoral and pelvic girdles were selected as they commonly reflect the mechanical and structural modifications necessary for exploitation of distinct environments (Emerson, 1982; Emerson & De Jongh, 1980; Enriquez-Urzelai et al., 2015; Gomes et al., 2009; Jorgensen & Reilly, 2013; Zug, 1978). The terminology used follows Trueb (1973), Emerson (1979), and Duellman & Trueb (1994). The number of adult specimens of each sex per species varied according to availability (range 1–5; Supporting Information Appendix S2). The specimens were cleared and whole-mounted for double staining of the cartilages and bones following the methodology of Wassersug (1976). They were then photographed with a high resolution digital camera (SonyDSC-H5, Sony Corp., Tokyo, Japan) using a binocular microscope. Images of the body size and postcranial skeletal bones were measured and analyzed using Image

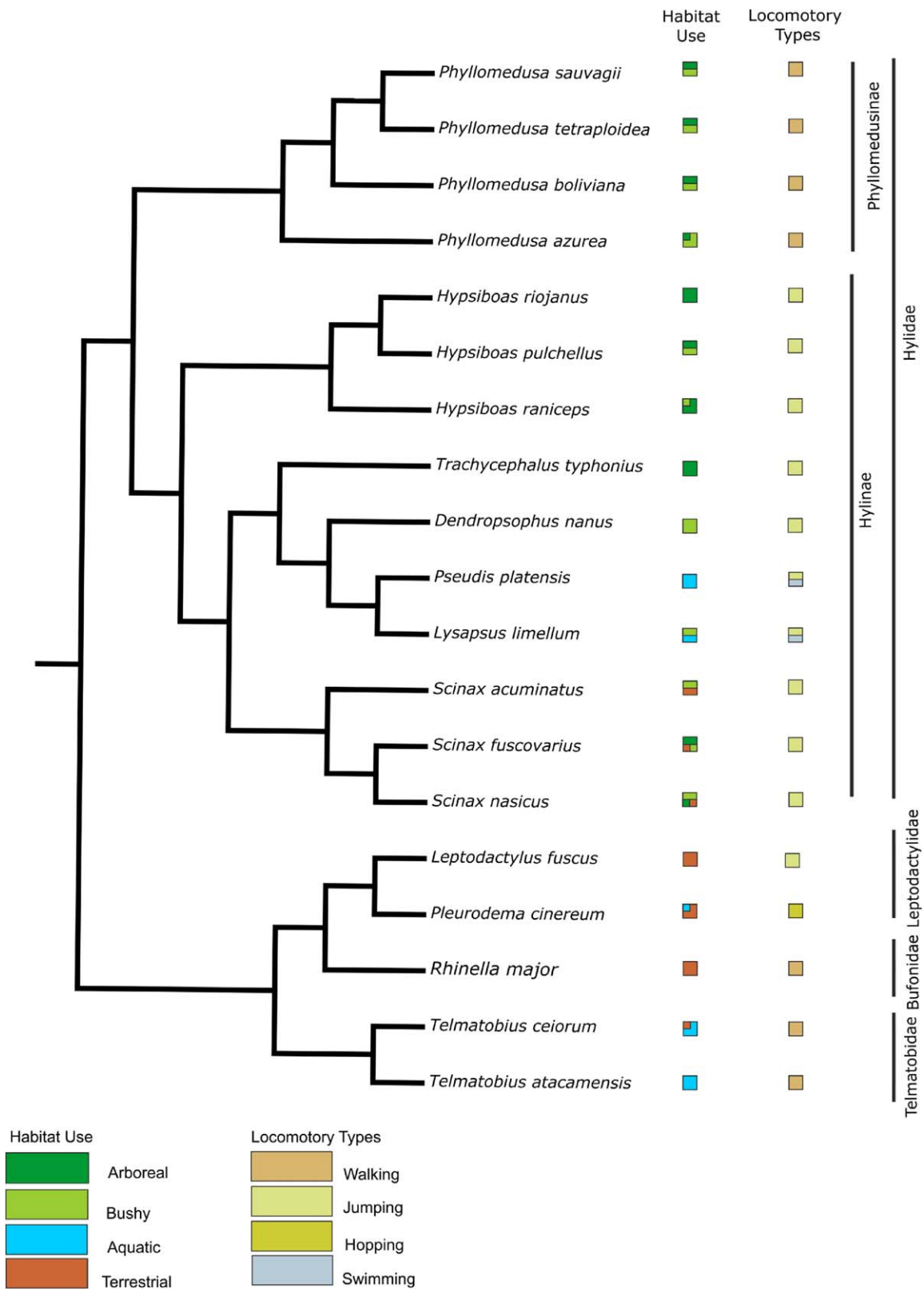


FIGURE 1 Composite tree based on Aguiar et al. (2007) and Wiens et al. (2010) [Color figure can be viewed at wileyonlinelibrary.com]

Tool 3.0 (University of Texas Health Science Center, San Antonio). Data related to body size were measured as the distance from cranial end of the vertebral column to the caudal end of urostyle in dorsal

view (hereafter referred to as TL). Measurements of the postcranial skeletal bones and body size are reported in the Supporting Information Appendix S2.

TABLE 1 List of postcranial measurements examined in this study, including vertebral column skeleton, and pectoral and pelvic girdles (following to Emerson, 1982; Simons, 2008; Trueb, 1977)

Measurements	Definition
Proximal sacral diapophysis width (PSDW)	The greatest length between the cranial and caudal edges of the sacral diapophysis adjacent to the vertebral centrum.
Distal sacral diapophysis width (DSDW)	The length between the distal cranial and caudal edges. Measured in dorsal view and including cartilage.
Distal expansion of the sacral diapophysis (DESD)	Angle between the sacral vertebral centrum and distal antero-postero edges of the sacral diapophysis. Measured in dorsal view and including cartilage.
Vertebral body width (VBW)	Length between the lateral edges of the vertebral body width. Measured in dorsal view.
Sacral diapophysis width, including cartilage (SDWic)	Length between the edges medial and lateral of the sacral diapophysis. Measured in dorsal view.
Orientation of the sacral diapophyses, including cartilage (OSDic)	Angle between the longitudinal axis of the vertebral column and antero-distal edges of the sacral diapophysis, measured from vertebral sacral centrum. Measured in dorsal view.
Urostyle length (UL)	Distance from cranial end to the caudal end of the urostyle. Measured in dorsal view.
Urostyle crest length (UCL)	Distance from cranial end of urostyle to the caudal end of the urostyle crest. Measured in dorsal view.
Vertebral column length (VCL)	Distance from cranial end of first vertebra to the caudal end of the eighth presacral vertebra. Measured in dorsal view.
Medial coracoid width (MCW)	Distance from cranial end to the caudal end of the coracoid. Measured along the longitudinal axis of body.
Coracoid length (CL)	The distance between the distal and proximal edge along the coracoids midline from the ventral surface.
Angle of the curvature of the clavicle (ACC)	Angle between the posterior end of the glenoid cavity to the medial and distal end of the clavicle. Measured in dorsal view.
Proximal scapula width (PSW)	Distance from anterior end to the posterior end of the scapula. Measured in the anterior end of the glenoid cavity.
Distal scapula width (DSW)	Distance from anterior end to the posterior end of the scapula. Measured in the articulation scapula-suprascapula.
Clavicle length (CIL)	The distance between the distal and proximal edge along the midline from the clavicle.
Scapula length (SL)	The distance between the distal and proximal edge along the midline from the scapula.
Ventral expansion of the ilium (VEI)	Angle between the posterior end of the ilial shaft and anterior edge of expansion of the acetabular plate.
Pelvic girdle length (PGL)	Distance from cranial end of the ilium to the caudal end of ischium. Measured in dorsal view.
Total length (TL)	Distance from cranial end of the vertebral column to the caudal end of urostyle. Measured in dorsal view.

2.3 | Ecological assignments

The assignment of locomotor modes and habitat uses were based on Cartmill (1985), Emerson (1985), Biewener (2003), Polly (2007), Wells (2007), Simons (2008), and on field observations (Tables 2 and 3).

Several lineages of anurans exhibit broad variations in habitat use and locomotor modes. To accommodate this we treated the ecological categories as a continuum (Grizante, Navas, Garland, & Kohlsdorf, 2010) and created indexes (ranging from 0 to 1) within each ecological category. For example, species that are strictly aquatic or specialized for terrestrial or arboreal habitats were assigned 1.0 for those categories and zero for the remainder, while others that are equally at home on land or in the water were assigned 0.5 for each (Tables 2 and 3).

Estimates of species substrate usage and locomotor mode were taken from relevant literature and personal observations (Table 2).

2.3.1 | Locomotor modes

Four categories were considered: walking, hopping, jumping, and swimming. Walking is defined as a motion that involves the movement of both the fore- and hind-limbs alternatively from side to side, coupled with lateral undulations of the body. The main distinction between hopping and jumping is the distance traveled during locomotion. A frog that hops moves in a series of short leaps whereas a frog that jumps performs a leap greater than 10 times its snout vent-length (Emerson, 1979). Swimming species employ the same movement as jumpers; extending their hind limbs synchronously, while maintaining the fore

TABLE 2 Ecological index assigned to each species, according to the locomotor modes

Species	Locomotor modes				Literature source
	Jumper	Hopper	Walker	Swimmer	
<i>Scinax fuscovarius</i>	1.00	0.00	0.00	0.00	Pers. Obs.
<i>Scinax nasicus</i>	1.00	0.00	0.00	0.00	Pers. Obs.
<i>Scinax acuminatus</i>	1.00	0.00	0.00	0.00	Pers. Obs.
<i>Hypsiboas riojanus</i>	1.00	0.00	0.00	0.00	Pers. Obs.
<i>Hypsiboas pulchellus</i>	1.00	0.00	0.00	0.00	Pers. Obs.
<i>Hypsiboas raniceps</i>	1.00	0.00	0.00	0.00	Pers. Obs.
<i>Trachycephalus typhonius</i>	1.00	0.00	0.00	0.00	Pers. Obs.
<i>Dendropsophus nanus</i>	1.00	0.00	0.00	0.00	Pers. Obs.
<i>Phyllomedusa sauvagii</i>	0.00	0.00	1.00	0.00	Wells (2007)
<i>Phyllomedusa azurea</i>	0.00	0.00	1.00	0.00	Wells (2007)
<i>Phyllomedusa boliviana</i>	0.00	0.00	1.00	0.00	Vaira (2001), Wells (2007)
<i>Phyllomedusa tetraploidea</i>	0.00	0.00	1.00	0.00	Manzano et al. (2004), Wells (2007)
<i>Pseudis platensis</i>	0.50	0.00	0.00	0.50	Manzano & Barg (2005)
<i>Lysapsus limellum</i>	0.50	0.00	0.00	0.50	Manzano & Barg (2005)
<i>Leptodactylus fuscus</i>	1.00	0.00	0.00	0.00	Jorgensen & Reilly (2013)
<i>Telmatobius ceiorum</i>	0.00	0.00	1.00	0.00	Lavilla, personal communication
<i>Telmatobius atacamensis</i>	0.00	0.00	1.00	0.00	Lavilla, personal communication
<i>Rhinella major</i>	0.00	0.00	1.00	0.00	Emerson (1979)
<i>Pleurodema cinereum</i>	0.00	1.00	0.00	0.00	Fabrezi, Manzano, Lobo, & Abdala (2014)

limb adducted against the body (Table 2; Emerson & De Jongh, 1980; Peters, Kamel, & Bashor, 1996; Wells, 2007).

2.3.2 | Habitat use

Four categories of substrate usage were considered: arboreal, bush, terrestrial, and aquatic (Table 3). Arboreal species inhabit trees; bush species inhabit bushes or low growing vegetation such as bromeliads, grass, and so forth; terrestrial species live on land, inhabiting primarily the ground around waterholes and on hillsides; aquatic species inhabit water features almost exclusively (Table 3).

2.4 | Phylogeny

Phylogenetic comparative analyses were performed using a composite tree (Figure 1) based on Aguiar et al. (2007), Wiens, Kuczynski, Hua, & Moen (2010), and Pyron & Wiens (2011). Data from the last two studies were primarily used to fill the gaps of species not considered in Wiens et al. (2010). Branch lengths were not available, so we assumed all were equal to 1, and transformed branch lengths using Pagel's methods (see Tulli et al., 2012).

2.5 | Statistical analysis

Mean values of osteological variables were \log_{10} transformed prior to analyses to meet normality requirements (Zar, 1999). As habitat use is

expressed in proportions, ecological data were transformed to the arcsin of the square root of each value (Martin & Bateson, 2007). All statistical analyses were implemented in an R statistical environment (R Core Development Team, 2011). Morphological traits require body size corrections based on their phylogenetic context, so we performed the phylogenetic size correction analysis described by Revell (2009). Residuals were calculated from least square regression analyses of morphological traits on body size (SVL), while controlling for phylogenetic nonindependence by using *phylo.resid* (a module of *Phytools* for R developed by Revell, 2012). The resulting residuals were then used for the subsequent analyses. To reduce the number of variables a phylogenetically based principal component analysis (PCA) was conducted based on a variance-covariance matrix of the morphological residuals using a Varimax rotation, implemented with the module *Phyl.PCA* from the *Phytools* package for R (Revell, 2012). To detect the variables with the greatest contributions to each principal component (PC), we considered only the informative traits from the morphological loadings with higher absolute values (negative or positive; Table 4 in boldface).

Subsequently, we tested three different evolutionary models to explore the processes that best explain the evolution of these morphological traits. The first model, known as the Brownian Motion evolutionary model (BM), supposes that the evolution of a trait results from random fluctuations through time (Felsenstein, 1988; Harmon et al.,

TABLE 3 Ecological index assigned to each species, according to the habitat use

Species	Habitat use				Literature source
	Arboreal	Shrubby	Terrestrial	Aquatic	
<i>Scinax fuscovarius</i>	0.5	0.25	0.25	0.00	Cei (1980), Obs. Pers.
<i>Scinax nasicus</i>	0.25	0.5	0.25	0.00	Obs. Pers.
<i>Scinax acuminatus</i>	0.00	0.5	0.50	0.00	Cei (1980), Duré (1999)
<i>Hypsiboas riojanus</i>	1.00	0.00	0.00	0.00	Obs. Pers.
<i>Hypsiboas pulchellus</i>	0.50	0.50	0.00	0.00	Gallardo (1993)
<i>Hypsiboas raniceps</i>	0.75	0.25	0.00	0.00	Obs. Pers.
<i>Trachycephalus typhonius</i>	1.00	0.00	0.00	0.00	Duellman (1970), Emerson (1979)
<i>Dendropsophus nanus</i>	0.00	1.00	0.00	0.00	Cei (1980), Obs. Pers.
<i>Phyllomedusa sauvagii</i>	0.75	0.25	0.00	0.00	Cei (1980), Wells (2007), Obs. Pers.
<i>Phyllomedusa azurea</i>	0.25	0.75	0.00	0.00	Borges de Freitas et al. (2008), Lucas et al. (2016), Obs. Pers.
<i>Phyllomedusa boliviana</i>	0.50	0.50	0.00	0.00	Vaira (2001), Wells (2007)
<i>Phyllomedusa tetraploidea</i>	0.50	0.50	0.00	0.00	Manzano et al. (2004), Pombal & Haddad (1992), Wells (2007)
<i>Pseudis platensis</i>	0.00	0.00	0.00	1.00	Duellman (2001, 2003), Wells (2007)
<i>Lysapsus limellum</i>	0.00	0.50	0.00	0.5	Duellman (2001, 2003), Vera Candiotti, Wells (2007), Obs. Pers.
<i>Leptodactylus fuscus</i>	0.00	0.00	1.00	0.00	Ponssa (2008), Heyer (1969)
<i>Telmatobius ceiorum</i>	0.00	0.00	0.25	0.75	Cei (1980), Laurent (1970), Lavilla (1988), Lavilla & Barrionuevo (2005), Vellard (1954), Wells (2007), Barrionuevo Obs. Pers.
<i>Telmatobius atacamensis</i>	0.00	0.00	0.00	1.00	Lavilla (1988), Lavilla & Barrionuevo (2005), Vellard (1954), Wells (2007),
<i>Rhinella major</i>	0.00	0.00	1.00	0.00	Vellard (1954), Wells, 2007
<i>Pleurodema cinereum</i>	0.00	0.00	0.75	0.25	Vellard (1954), Lavilla, personal communication

2010). The second Ornstein–Uhlenbeck (OU) model focuses on sections of lineages where a trait varies in relation to an optimum or stabilizing selection (Butler & King, 2004; Harmon et al., 2010). Finally, the third hypothesis predicts intensified trait modifications early in the evolutionary tree followed by a gradual deceleration of the evolutionary rate, in a model called Early Burst (EB; Harmon et al., 2010). To test which evolutionary model best fits each variable, a fitContinuous analysis was run using GEIGER (Harmon et al., 2010) and analysis of phylogenetics and evolution (APE; Paradis, Claude, & Strimmer, 2004) packages for R. The command “fitContinuous” describes the rate of character modification within the three aforementioned evolutionary models, while also providing an Akaike value (AIC) for each procedure. The best fit among the candidate evolutionary models was then obtained from the AIC (Angilletta, 2006; Burnham & Anderson, 2002) using the weights (wAICc) as a measure of strength for each model, and indicating the probability that a given model is the best among a series of candidate models (Burnham & Anderson, 2002).

Due to their phylogenetic relationships species cannot be considered as independent data-points (Harvey & Pagel, 1991), so Pagel's

phylogenetic signal (λ) was estimated by using the residual errors simultaneously on the regression parameters of phylogenetic generalized least squares models (PGLS) analyses. These analyses were performed in “caper” (Orme et al., 2012) and “ape” (Paradis et al., 2004) packages for R. Models were built using the ecological data as the independent variable, as a proportion of habitat use, either individually or in combination (e.g., $PC \sim \text{arboreal} + \text{terrestrial} + \text{swimming} + \text{jumping}$), and using the morphological information as the dependent variable, represented by the species' scores of each one of the four retained PCs. Model choice was based on the model fit using the AIC aforementioned.

3 | RESULTS

The first two PC axes explained approximately 72% of the accumulated variance. PC1 loads show that the proximal sacral diapophysis width (PSDW), vertebral body width (VBW), sacral diapophysis width, including cartilage (SDWic), urostyle length (UL), vertebral column length

TABLE 4 Principal component scores of osteological characters (PC1 and PC2) that explained near 72% of variance

Variables	PC1	PC2
PSDW	-0,9494892	0,043205515
DSDW	-0,5754095	0,806866744
VBW	-0,9616448	-0,20964365
DESD	0,220454	0,95179795
SDWic	-0,9477486	0,113960093
OSDic	-0,1886238	-0,44646804
UL	-0,9690144	0,1225979
VCL	-0,9736428	0,112780242
MCW	-0,9671413	-0,019853012
CL	-0,9360136	-0,180119988
ACC	0,4492974	0,096426738
PSW	-0,9131004	-0,335359153
DSW	-0,9432721	-0,009342408
CIL	-0,9132546	-0,288266616
SL	-0,9429543	0,111759015
VEI	0,2603428	0,695962853
PGL	-0,9757635	0,078722801

(VCL), medial coracoid width (MCW), coracoid length (CL), proximal sacral diapophysis width (PSW), distal sacral diapophysis width (DSW), clavicle length (CIL), scapula length (SL), and pelvic girdle length (PGL) all contributed importantly, all displaying negative values (Figure 2, Table 4). In the case of PC2, distal sacral diapophysis width (DSDW), distal expansion of the sacral diapophysis (DESD), and ventral expansion of the ilium (VEI) had higher and positive loads (Figure 2, Table 4).

3.1 | Evolutionary models

Hylid morphology appears to have evolved through various different processes, as no single evolutionary model could explain all the studied traits (Table 5). The majority of the traits were best explained by the Ornstein-Uhlenbeck model of evolutionary change (Butler & King, 2004; Harmon et al., 2010). The BM model, conversely, which predicts a random rate of change, better explained the evolution of only one trait of the sacral vertebra (Table 5). Using the AIC, none of the skeletal variables showed a best fit with the Early Burst evolutionary model (Table 5).

3.2 | Phylogenetic signal

λ -values were significant for most of the recovered associations (Table 6). Three of the five variables associated to PC1, bush and swimmer and hopper locomotor modes showed a λ -value = 1 or closer to 1. All other associations exhibited a λ -value lower than 1, suggesting that

variability was a consequence of adaptation to ecological requirements (Table 6).

3.3 | Ecomorphological relationships

The PGLS analysis produced a total of 256 possible models for each PC (Supporting Information Appendix S3). Only five of these (four for PC1 and one for PC2) were informative according to the Akaike criterion and provided evidence for an association between morphology and ecology (Table 6). These models described significant slopes for some of the habitat use and locomotor mode variables (Table 6). Frogs exhibiting longer vertebral column, broad vertebral body, broad proximal sacral diapophysis, longer urostyle, and longer pelvic girdle present a relationship between PC1 and bush habitat use, and jump and swim locomotor modes (Table 6). The pectoral girdle shows a trend toward broad proximal and distal scapula, longer clavicle, and longer scapula and coracoid (Tables 4 and 6). Species that inhabit arboreal environments and walk exhibit narrow proximal sacral diapophyses, narrow vertebral bodies, narrow proximal and distal scapula, accompanied by a shorter clavicle, pelvic girdle, scapula and coracoid, and a shorter urostyle and vertebral column (Tables 4 and 6). We have also identified an association between PC2 and frogs that inhabit bush, walk, and hop; they exhibit narrow distal sacral diapophyses and smaller sacral diapophyses expansion angles (Figure 3).

4 | DISCUSSION

In this study, we addressed the evolutionary models that can explain variations in the postcranial morphology of hylids, and examined whether the identified variations correlate with habitat use and/or locomotor modes.

4.1 | Morphology and evolutionary models

Our results show that the evolutionary model that best fits most of the morphological variables related to the pelvic girdle is the Ornstein-Uhlenbeck model. It emphasizes a stabilizing selection that tends to generate hierarchical patterns, which is congruent with previous studies of hylids (Jorgensen & Reilly, 2013; Nauwelaerts et al., 2007) that suggest they are morphologically and ecologically conservative frogs. The pelvic girdle length has been established as a conservative character between frogs with different locomotor modes, with greater variation in the shape of the sacrum (Jorgensen & Reilly, 2013; Nauwelaerts et al., 2007). Interestingly, one of the traits related to the sacrum are better explained by a BM model of evolution that indicates stochastic processes. The fact that different modes of evolution drive different structures of the same body could be an indication of the modular evolution of the vertebrae and pectoral and pelvic girdles. Modular theory predicts that developmental processes can generate phenotypic units that are capable of independent modification (Buchholz, 2007). Our results could indicate that the girdles and the vertebral column constitute independent modules.

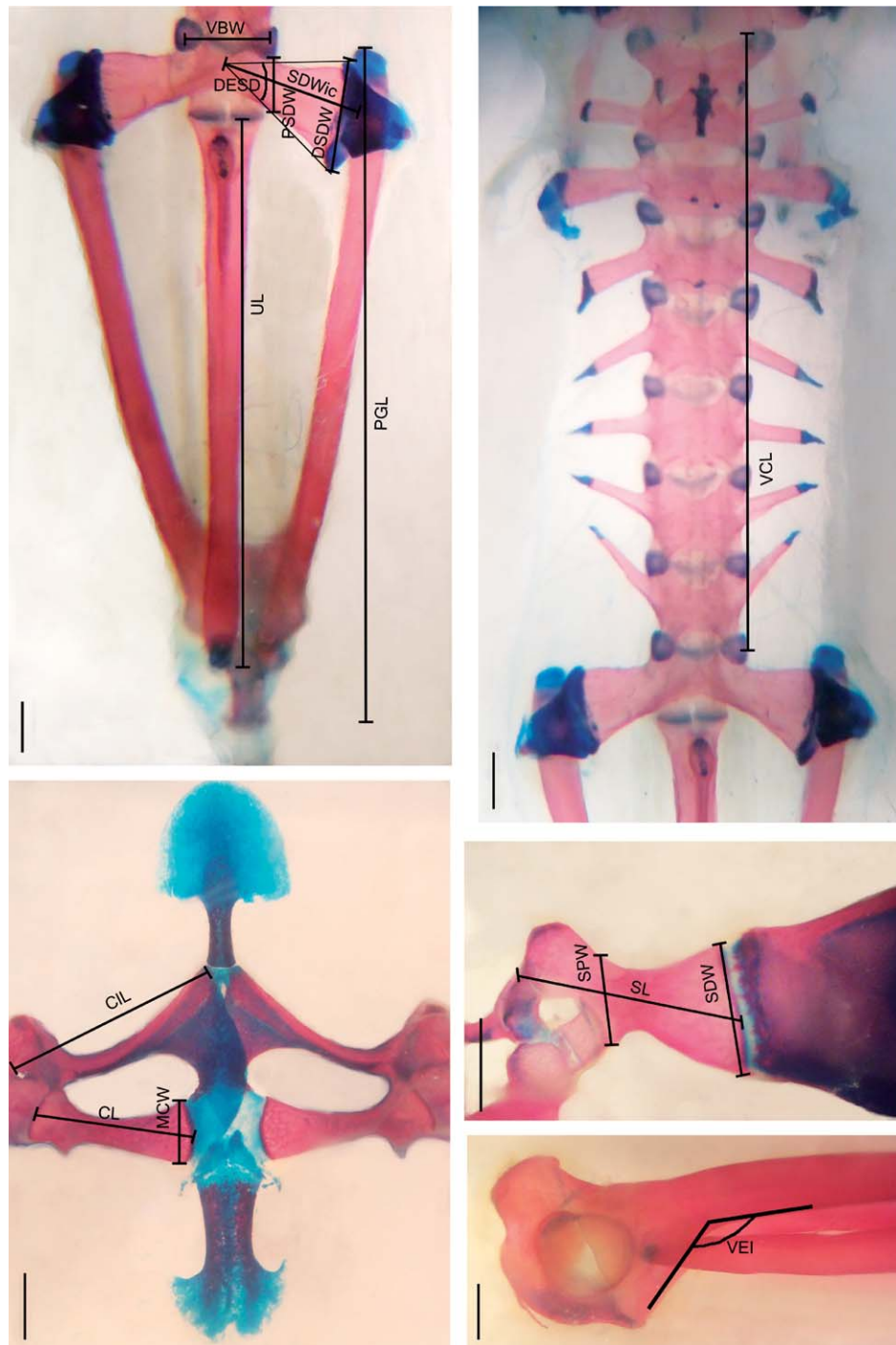


FIGURE 2 Osteological variables of *Scinax nasicus* included in our model analyses. VBW = Vertebral body width; SDWic = Sacral diapophysis width, including cartilage; DSDW = Distal sacral diapophysis width; PSDW = Proximal sacral diapophysis width; UL = Urostyle length; PGL = Pelvic girdle length; VCL = Vertebral column length; CIL = Clavicle length; CL = Coracoid length; MCW = Medial coracoid width; SPW = Proximal scapula width; SL = Scapula length; DSW = Distal scapula width; VEI = Ventral expansion of the ilium. Scale bars: 1 mm

4.2 | Ecomorphological relationships

Another outcome of our analysis is the identification of an association between features of the pelvic and pectoral girdles and the vertebral column, and particular ecological aspects. For example, we found that

frogs that live in the bush and swim and jump, exhibit broad proximal sacral diapophyses, broad vertebral bodies, and longer urostyles. Studies have previously shown an association between the morphological traits with swimming and jumping (Emerson & De Jongh, 1980; Nauwelaerts et al., 2007; Peters et al., 1996), and this is again

TABLE 5 Values of AICc and Log Likelihood (LogL) corresponding to the evolutionary models tested (Brownian motion [BM], Ornstein-Uhlenbeck [OU]) of the two principal components (PC1 and PC2)

Morphology	LogL	AICc	wAICc	LogL	AICc	wAICc	BeMo
VBW	0.591	3.566	0.098	4.20	-0.801	0.877	OU
DSDW	-4.878	14.506	0.620	-4.443	16.487	0.725	OU
DSW	-2.200	9.152	0.117	1.205	5.188	0.854	OU
SDWic	-1.184	7.118	0.209	1.500	4.599	0.739	OU
DESD	6.412	-8.075	0.667	6.477	-5.354	0.171	BM
MCW	-0.697	6.144	0.197	2.070	3.459	0.755	OU
PSDW	-3.740	12.23	0.037	-0.649	8.899	0.952	OU
PSW	2.201	9.152	0.117	1.205	5.188	0.854	OU
CL	6.323	-7.896	0.223	8.920	-10.24	0.722	OU
CIL	1.46	1.828	0.194	4.249	-0.899	0.759	OU
VCL	5.513	-6.27	0.306	7.644	-7.68	0.620	OU
PGL	4.436	-4.123	0.209	7.124	-6.648	0.740	OU
SL	-0.175	5.100	0.300	1.809	3.981	0.585	OU
UL	5.209	-5.669	0.625	7.371	-7.143	0.627	OU

wAICc is the weight of the different models. BeMo indicates the evolutionary model that best fitted the data based on the wAICc values. α and σ^2 , estimate evolutionary rate.

demonstrated in the current work. Remarkably, most anurans are capable of swimming, even those that do not live in water (Calow & Alexander, 1973; Emerson & De Jongh, 1980; Gal & Blake, 1988a, 1988b; Gillis & Biewener, 2000; Peters et al., 1996). Interestingly, we found that frogs that live in the bush and swim exhibit strong

phylogenetic signal, evidencing again some degree of morphological conservatism. Jenkins & Shubin (1998) argued that the caudopelvic mechanism needed for swimming compromises the movement of the urostyle, and requires elongated iliac shafts and mobile iliosacral and sacro-urostylic joints. A study by Herrel et al. (2014) however,

TABLE 6 Best fitting PGLS models for principal components of morphology (PC1, PC2), the proportion of the ecological categories

Model	λ	Adj r^2	Intercept	Variable	Slope	Pp	Pt	AICc	Wi
PC1~bush	1	0.59	1.48	bush	-1.39	0.000	0.000	33.005	0.39
PC1~jumper+bush+tree	0.001	0.52	0.60	jumper	-0.41	0.001	0.000	30.774	0.38
				bush	-0.79	0.000			
				tree	0.68	0.000			
PC1~walker+bush+tree	0.001	0.68	0.01	walker	0.45	0.002	0.000	26.189	0.38
				bush					
				tree					
PC1~swimmer+bush	0.96	0.66	0.04	swimmer	-1.29	0.03	0.000	30.187	0.39
				bush	-1.46	0.000			
PC2~bush+walker+hopper	1	0.67	3.36	bush	-0.44	0.009	0.000	7.46	0.96
				walker	-0.84	0.018			
				hopper	-0.73	0.003			

See Appendix S2 for all models. Pagel's λ (phylogenetic signal), adjusted r^2 (Adj r^2), intercept, slopes were considered for those informative variables based on the Akaike criterion (AICc and Wi). Pp: means the partial p value for each variable; Pt: is the p value for the complete model. Boldface denotes significant results.

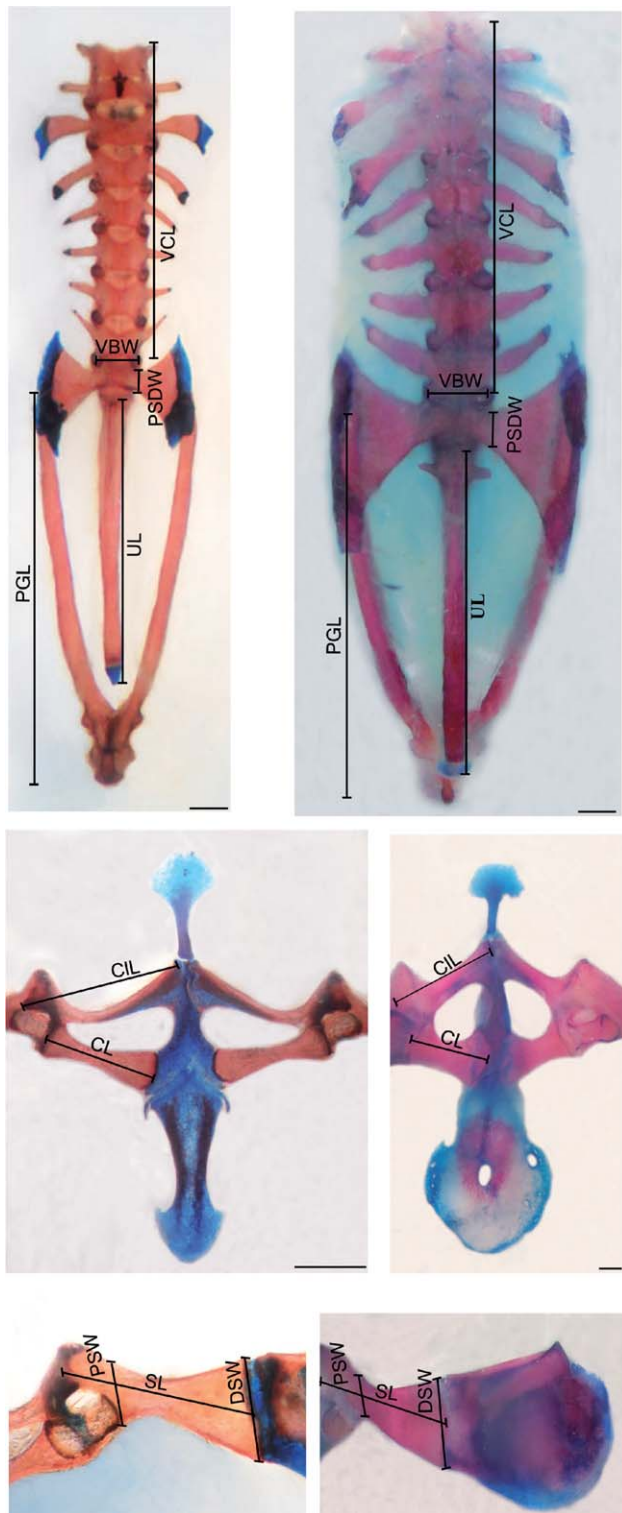


FIGURE 3 Left: Frogs that use bush and jump, *Dendropsophus nanus*. Right: Frogs that use arboreal habitat and walk present, *Phyllomedusa tetraploidea*. VBW = Vertebral body width; VCL = Vertebral column length; PSDW = Proximal sacral diapophysis; UL = Urostyle length; PGL = Pelvic girdle length; CIL = Clavicle length; CL = Coracoid length; PSW = Proximal scapula width; SL = Scapula length; DSW = Distal scapula width. Scale bars: 1 mm

postulates that the caudopelvic mechanism is actually the best predictor of the maximum resultant force in jumping and swimming, and suggests that it may in fact play an even more important role in jumping than in swimming. Likewise, Reilly et al. (2016) postulated that the pelvic girdle might play an important role in landing. In the present study we also identified several osteological postcranial characters that while adaptive for swimming, are also advantageous for jumping. These observations indicate the simultaneous optimization of two tasks that might be considered mutually incompatible. It is interesting to note, however, that the bauplan of frogs seems to be ancestrally developed for jumping. This interpretation is based on the study of fossil anurans, such as *Triadobatrachus* and *Prosalirus bitis* (Rage & Roček, 1986, 1989; Shubin & Jenkins, 1995; Wake, 1998) that indicate the strong influence that the plesiomorphic frog anatomy has on their current morphology (Essner, Suffian, Bishop, & Reilly, 2010). It is possible that this phylogenetic inertia, also shown in the significant phylogenetic signal, imposes limitations on the adaptation to a more aquatic life style (Nauwelaerts et al., 2005) and explains the morphological similarity despite differences in habitat use. It could also be inferred that the anuran bauplan, in fact, is an intermediate phenotype, designed to perform all tasks reasonably well (Nauwelaerts et al., 2007; Tulli et al., 2012, 2015, 2016). Many ecomorphological studies have found a tendency toward the conservation of gross structure traits across taxa (e.g., Abdala, Tulli, Russell, Powell, & Cruz, 2014; Gans, 1993; Nauwelaerts et al., 2007; Tulli et al., 2012, 2015, 2016). This may represent a response mechanism to the demands of a broad array of environmental and mechanical challenges by permitting adequacy in all circumstances (Abdala et al., 2014; Gans, 1993). From this perspective, organisms can be considered a “jack of all trades and master of none” (Tulli et al., 2012), as they retain an all-purpose morphology allowing them to access a variety of habitats and locomotor modes. This generalized morphology could represent a morphological optimum because natural selection is acting on several traits and several surfaces simultaneously without taking into account the available environments (Sathe & Husak, 2015).

The sacral vertebrae are among the structures modified for both jumping and swimming, as they provide attachments for key muscles involved in the general displacement of the urostyle. The m. longissimus dorsi, the m. coccygeiliacus, and the m. coccygeosacralis are specifically involved in the urostyle movement/stabilization. The m. coccygeiliacus during the muscular contraction also participates in the posterior gliding of the ilium along the sacral diapophysis, projecting the vertebral column forward (Příkryl, Aerts, Havelková, Herrel, & Roček, 2009). All three are strap and parallel fibered muscles, and their longer fibers enable greater muscle contraction that produces specific joint motion (Oatis, 2009). Thus, it could be inferred that a longer urostyle and larger diapophyses and sacral vertebrae would require longer muscles and thereby enhance jumping and swimming performance. It should be noted, however, that degree of sacral diapophyseal expansion has produced conflicting results in different studies (e.g., Jorgensen & Reilly, 2013; Soliz &

Ponssa, 2016), thereby identifying it as an important ecomorphological trait that requires further consideration.

The components of the pectoral girdle also tend to be longer and slender (compare the pectoral girdles in Figure 3) in frogs that inhabit the bush and jump and swim. The Slender-Body Theory has been developed in the hydrodynamic field, based in the advantages of having a slender body to face the problem of the movement in a viscous fluid (Cox, 1970). Interestingly, viscous drag is most critical at small size and slow speed (Biewener, 2003); likewise, slenderness has been associated to more effective long-distance running (Åstrand, Rodhal, Dahl, & Strømme, 2003). Taken together, these data suggest that a slender body would be better adjusted to face any challenging locomotor modes. The increased size of these elements has been reliably linked to the important role of the pectoral girdle during jumping, as it operates like a damper to absorb the impact of landing (Emerson, 1983; Griep et al., 2013; Nauwelaerts & Aerts, 2006). Acting as a buffer, the pectoral girdle elements can shift relative to one another during landing, as in the dorso-ventral flexion of the scapula-suprascapula joint (Emerson, 1983; Griep et al., 2013). It has also been suggested that the function of impact absorption upon landing could be carried out by the clavicle or the coracoid (Emerson, 1984). The entire pectoral girdle can rotate as it is connected to the spine by muscles, engaging the glenoid joint as a pivot (Emerson, 1983; Griep et al., 2013). Emerson (1983) suggested that during landing several muscles, such as the rhomboideus, serratus and levator scapulae, actively fasten the suprascapula dorsally to the vertebral column to prevent its movement. Additionally, it has been shown that several shoulder muscles are also important during the pre-landing activity, being critical for resisting the forces associated with impact (Akella & Gillis, 2010; Ekstrom & Gillis, 2015). These data are in concordance with our results, as we found jumper species like *Dendropsophus nanus* to have broad proximal and distal scapulae, longer clavicles, longer scapulae and longer coracoids. Shearman (2008) demonstrated that the jumper *Rana pipiens* exhibits longer scapulae than the swimmer *Xenopus laevis* and both have longer scapulae than the other species studied here.

Arboreal and walker species in this study presented smaller and shorter vertebral bodies, coracoids and clavicles than the jumper and swimmer species, along with narrow proximal and distal sacral diapophyses. Ours models showed that these traits associated to jump and walk might be more evolutionarily flexible, as their phylogenetic signal tend to be low. These results are in accordance with the traits described for hopping anurans: relatively short and stout bodies with pelvic girdles and hind limbs shorter than in jumping frogs (Emerson, 1978). Emerson (1978) also called attention to the similarity of the morphological requisites for walking and hopping, pointing out that these locomotor modes tend to occur together, as some frogs can switch from hopping to walking gaits. Interestingly, Jorgensen & Reilly (2013) also considered a similar transition from hopping to jumping in Neobatrachia. Soliz & Ponssa (2016) also found narrow diapophyses in walker-aquatic species such as *Telmatobius ceiorum* and *Telmatobius atacamensis*, and in the swimmer and jumper-aquatic species *Pseudis platensis*. They surmised that the presence of narrow sacral diapophyses appears to be more closely

related to aquatic habitat use than to any particular locomotor mode. Thus, it could be inferred that new locomotor abilities are actually acquired along a morphological continuum (Fontanarrosa & Abdala, 2016), a process that could be compared to evolutionary gradient in the acquisition of the forelimb control landing in frogs, which radiates into diverse locomotor modes and habitat uses (Reilly et al., 2016). Interestingly, Richards (2010) found a functional continuum between rotational and translational motion in four frog species, that could be easily associated to the idea of gradual morphological changes proposed here.

In conclusion, our work shows that the evolution of elements of the postcranial hylid skeleton, specifically the girdles and vertebral column, are explained by distinct evolutionary models, suggesting the influence of modular evolution. In support of findings from previous studies, we also detected a relationship between morphology and locomotor modes in hylid frogs. On one hand, jumping and swimming species, and on the other, walking and hopping species, exhibited evidence reaffirming similar morphological architectures for these locomotor modes. These findings lead to the inference that there is no perfect match between a particular locomotor ability and internal morphology, at least in hylids. The traits associated to ecological categories are in fact overlapped and generated on a morphological continuum and not as discreet adaptations.

ACKNOWLEDGMENTS

We thank to Sonia Kretzschmar and Marta Cánepa of the collection of Herpetology Institute from Fundación Miguel Lillo. This study was funded by a PIP CONICET 0284 to VA. MCS was supported by a Consejo Nacional de Investigaciones Científicas y Tecnológicas (CONICET) fellowship. For helpful for improving the English language, we thank Jennifer Richardson.

REFERENCES

- Abdala, V., Tulli, M. J., Russell, A. P., Powell, G. L., & Cruz, F. (2014). Anatomy of the crus and pes of Neotropical iguanian lizards in relation to habitat use and digitally based grasping capabilities. *The Anatomical Record*, 297, 397–409.
- Aerts, P., & Nauwelaerts, S. (2009). Environmentally induced mechanical feedback in locomotion: Frog performance as a model. *Journal of Theoretical Biology*, 261, 372–378.
- Aguiar, O., Jr., Bacci, M., Jr., Lima, A. P., Rossa-Feres, D. C., Haddad, C. F. B., & Recco-Pimentel, S. M. (2007). Phylogenetic relationships of *Pseudis* and *Lysapsus* (Anura, Hylidae, Hylinae) inferred from mitochondrial and nuclear gene sequences. *Cladistics*, 23, 455–463.
- Akella, T., & Gillis, G. B. (2010). Hopping isn't always about the legs: Forelimb muscle activity patterns during toad locomotion. *Journal of Experimental Zoology*, 315, 1–11.
- Angilletta, M. J. (2006). Estimating and comparing thermal performance curves. *Journal of Thermal Biology*, 31, 541–545.
- Arnold, S. J. (1983). Morphology, performance and fitness. *American Zoologist*, 23, 347–361.
- Åstrand, P.-O., Rodhal, K., Dahl, H. A., & Strømme, S. B. (2003). *Textbook of work physiology (4th ed.)*. In *Human Kinetics*. USA: McGraw Hill.
- Bain, R. H., Lathrop, A., Murphy, R. W., Nikolai, L. O., & Cuc, H. T. (2003). Cryptic species of a cascade frog from Southeast Asia: Taxonomic revisions and descriptions of six new species. *American Museum Novitates*, 3417, 1–60.

- Banbury, B., & Maglia, A. M. (2006). Skeletal development of the mexican spadefoot, *Spea multiplicata* (Anura: Pelobatidae). *Journal of Morphology*, 267, 803–821.
- Bauwens, D., Garland, T., Jr., Castilla, A. M., & Van Damme, R. (1995). Evolution of sprint speed in lacertid lizards: Morphological, physiological and behavioral covariation. *Evolution*, 49, 848–863.
- Biewener, A. A. (2003). *Animal locomotion* (p. 281). Oxford: Oxford University Press.
- Bijma, N. N., Gorb, S. N., & Kleinteich, T. (2016). Landing on branches in the frog *Trachycephalus resinifictrix* (Anura: Hylidae). *Journal of Comparative Physiology A*, 202, 267–276.
- Borges de Freitas, E., De Carvalho, C. B., Gomes Faria, R., de Carvalho Batista, R., de Carvalho Batista, C., Araújo Coelho, W., & Bocchiglieri, A. (2008). Nicho ecológico e aspectos da história natural de *Phyllomedusa azurea* (Anura: Hylidae, Phyllomedusinae) no Cerrado do Brasil Central. *Biota Neotropica*, 8, 1–9.
- Buchholtz, E. A. (2007). Modular evolution of the Cetacean vertebral column. *Evolution & Development*, 9, 278–289.
- Burnham, K. P., & Anderson, D. R. (2002). *Model selection and multimodel inference a practical information-theoretic approach*. New York: Springer-Verlag.
- Butler, M. A., & King, A.A. (2004). Phylogenetic comparative analysis: A modeling approach for adaptive evolution. *The American Naturalist*, 164, 6.
- Calow, L. J., & Alexander, R. M. (1973). A mechanical analysis of a hind leg of a frog (*Rana temporaria*). *Journal of Zoology*, 171, 293–321.
- Carrizo, L. V., Tulli, M. J., Dos Santos, D. A., & Abdala, V. (2013). Interplay between postcranial morphology and locomotor types in Neotropical sigmodontine rodents. *Journal of Anatomy*, 224, 469–481.
- Cartmill, M. (1985). Climbing. In M. Hildebrand, D. M. Bramble, K. F. Liem, & D. B. Wake (Eds.), *Functional vertebrate morphology* (pp. 73–88). Cambridge: Belknap Press of Harvard University.
- Cei, J.M. (1980). Amphibians of Argentina. *Monitore Zoologico Italiano Monographs*, 2, 1–609.
- Channing, A., & Broadley, D. G. (2002). A new snout-burrower from the *Barotse floodplain* (Anura: Hemisotidae: Hemisus). *Journal of Herpetology*, 36, 367–372.
- Collar, D. C., Schulte, J. A., II, Losos, J. B. (2011). Evolution for extreme body size disparity in monitor lizards (*Varanus*). *Evolution*, 65, 2664–2680.
- Cox, R. G. (1970). The motion of long slender bodies in a viscous fluid. Part 1. *General Theory*. *Journal of Fluid Mechanics*, 44, 791–810.
- Cox, S. M., & Gillis, G. B. (2015). Forelimb kinematics during hopping and landing in toads. *Journal of Experimental Biology*, 210, 3051–3058.
- Cruz, F. B., Belver, L., Acosta, J. C., Villavicencio, H. J., Blanco, G., & Canovas, M. G. (2009). Thermal biology of *Phymaturus* lizards: Evolutionary constraints or lack of environmental variation? *Zoology*, 112, 425–432.
- Dobrowolska, H. (1973). Body-part proportions in relation to the mode of locomotion in Anurans. *Zoologica Poloniae*, 23, 59–108.
- Duellman, W. E. (1970). The hylid frogs of Middle America. *Monograph of the Museum of Natural History, the University of Kansas*, 1, 1–753.
- Duellman, W. E. (2001). *The hylid frogs of Middle America*. *Society for the Study of Amphibians and Reptiles* (Vol. 2, p. 1180). New York: Ithaca.
- Duellman, W. E. (2003). An overview of anuran phylogeny, classification and reproductive modes. In B. G. M. Jamieson (Ed.), *Reproductive biology and phylogeny of Anura* (pp. 1–18). Nefield, NH: Science Publishers, Inc.
- Duellman, W. E., & Trueb, L. (1994). *Biology of amphibians*. Baltimore, London: The Johns Hopkins University Press.
- Duré, M. (1999). Interrelaciones en los nichos tróficos de dos especies sintópicas de la familia Hylidae (Anura) en un área subtropical de Argentina. *Cuadernos de Herpetología*, 13, 11–18.
- Ekstrom, L., & Gillis, G. B. (2015). Pre-landing wrist muscle activity in landing toads. *Journal of Experimental Biology*, 218, 2410–2415.
- Emerson, S. B. (1978). Allometry and umping in frogs: Helping the twain to meet. *Evolution*, 32, 551–564.
- Emerson, S. B. (1979). The ilio-sacral articulation in frogs: Form and function. *Biological Journal of the Linnean Society*, 11, 153–168.
- Emerson, S. B. (1982). Frog postcranial morphology: Identification of a functional complex. *Copeia*, 3, 603–613.
- Emerson, S. B. (1983). Functional analysis of frog pectoral girdles. The epicoracoid cartilages. *Journal of Zoology*, 201, 293–308.
- Emerson, S. B. (1984). Morphological variation in frog pectoral girdles: Testing alternatives to a traditional adaptive explanation. *Evolution*, 38, 376–388.
- Emerson, S. B. (1985). Jumping. In M. Hildebrand, D. M. Bramble, K. F. Liem, & D. B. Wake (Eds.), *Functional vertebrate morphology* (pp. 58–72). Cambridge: Belknap Press of Harvard University.
- Emerson, S. B. (1988). Convergence and morphological constraint in frogs: Variation in postcranial morphology. *Fieldiana Zoology*, 43, 1–19.
- Emerson, S. B., & De Jongh, H. D. (1980). Muscle activity at the iliosacral articulation of frogs. *Journal of Morphology*, 166, 129–144.
- Enriquez-Urzelai, U., Montori, A., Llorente, G. A., & Kaliontzopoulou, A. (2015). Locomotor mode and the evolution of the hindlimb in Western Mediterranean Anurans. *Evolutionary Biology*, 42, 199–209.
- Essner, L. R., Jr., Suffian, D. J., Bishop, P. J., & Reilly, S. M. (2010). Landing in basal frogs: Evidence of saltational patterns in the evolution of anuran locomotion. *Naturwissenschaften*, 97, 935–939.
- Fabrezi, M., Manzano, A., Lobo, F., & Abdala, V. (2014). Anuran locomotion: Ontogeny and morphological variation of a distinctive set of muscles. *Journal of Evolutionary Biology*, 41, 308–326.
- Felsenstein, J. (1988). Phylogenies and quantitative methods. *Annual Review of Ecology, Evolution, and Systematics*, 19, 445–471.
- Fontanarrosa, G., & Abdala, V. (2016). Bone indicators of grasping hands in lizards. *PeerJ*, 4, e1978. DOI 10.7717/peerj.1978.
- Frost, D. R. (2016). *Amphibian species of the world: An Online Reference*. Version 6.0. New York: American Museum of Natural History. Available at: <http://research.amnh.org/herpetology/amphibian/index.html>.
- Gal, J. M., & Blake, R. W. (1988a). Biomechanics of frog swimming I. Estimation of the propulsive force generate by *Hymenochirus boettgeri*. *Journal of Experimental Biology*, 138, 399–411.
- Gal, J. M., & Blake, R. W. (1988b). Biomechanics of frog swimming II. Mechanics of the limb-beat cycle in *Hymenochirus boettgeri*. *Journal of Experimental Biology*, 138, 413–429.
- Gallardo, J. M. (1993). Los anfibios de dos ecosistemas argentinos: Algunas estrategias en la reproducción y el desarrollo. *Boletín de la Sociedad Zoológica del Uruguay*, 8, 33–36.
- Gans, C. (1993). On the merits of adequacy. *American Journal of Science*, 293, 391–406.
- Gillis, G. B., & Biewener, A. A. (2000). Hind limb extensor muscle function during jumping and swimming in the toad (*Bufo marinus*). *Journal of Experimental Biology*, 203, 3547–3563.
- Gomes, F. R., Rezende, E. L., Grizante, M. B., & Navas, C. A. (2009). The evolution of jumping performance in anurans: Morphological

- correlates and ecological implications. *Journal of Evolutionary Biology*, 22, 1088–1097.
- Goodman, B. (2006). *Ecomorphology, microhabitat use, performance and reproductive output in tropical iguonine lizards*. Thesis. James Cook University, Queensland.
- Goodman, B. A., Miles, D. B., & Schwarzkopf, L. (2008). Life on the rocks: Habitat use drives 495 morphological and performance evolution in lizards. *Ecology*, 89, 3462–3471.
- Griep, S., Schilling, N., Marshall, P., Amling, M., Hahne, L. M., & Haas, A. (2013). Pectoral girdle movements and the role of the glenohumeral joint during landing in the toad, *Rhinella marina* (Linnaeus, 1758). *Zoomorphology*, 132, 325–338.
- Grizante, M. B., Navas, C. A., Garland, T., Jr., & Kohlsdorf, T. (2010). Morphological evolution in Tropicurinae squamates: An integrated view along a continuum of ecological settings. *Journal of Evolutionary Biology*, 3, 98–111.
- Harmon, L. J., Losos, J. B., Davies, T. J., Gillespie, R. G., Gittleman, J. L., Jennings, W. B., ... Mooers AØ. (2010). Early burst of body and shape evolution are rare in comparative data. *Evolution*, 64, 2385–2396.
- Harvey, P. H., & Pagel, M. D. (1991). *The comparative method in evolutionary biology*. Oxford: Oxford University Press.
- Herrel, A., Vanhooydonck, B., Porck, J., & Irschick, D. J. (2008). Anatomical basis of differences in locomotor behavior in *Anolis* lizards: A comparison between two ecomorphs. *Bulletin of the Museum of Comparative Zoology*, 159, 213–238.
- Herrel, A., Vasilopoulou-Kampitsi, M., & Bonneaud, C. (2014). Jumping performance in the highly aquatic frog, *Xenopus tropicalis*: Sex-specific relationships between morphology and performance. *PeerJ* 2, e661. DOI 10.7717/peerj.661.
- Heyer, W. R. (1969). Studies on the genus *Leptodactylus* (Amphibia, Leptodactylidae) III. A redefinition of the genus *Leptodactylus* and a description of a new genus of Leptodactylid frogs. *Contributions in Science, Natural History Museum of Los Angeles County*, 155, 1–14.
- Irschick, D. J., Meyers, J. J., Husak, J. F., & Le Galliard, J.-F. (2008). How does selection operate on whole-organism functional performance capacities? A review and synthesis. *Evolutionary Ecology Research*, 10, 177–196.
- Jenkins, F. A., & Shubin, N. H. (1998). *Prosalirus bitis* and the anuran caudopelvic mechanism. *Journal of Vertebrate Paleontology*, 18, 495–510.
- Jorgensen, M. E., & Reilly, S. M. (2013). Phylogenetic patterns of skeletal morphometrics and pelvic traits in relation to locomotor mode in frogs. *Journal of Evolutionary Biology*, 26, 929–943.
- Kohlsdorf, T., Garland, T., Jr., & Navas, C. A. (2001). Limb and tail lengths in relation to substrate usage in *Tropidurus* lizards. *Journal of Morphology*, 248, 151–164.
- Kohlsdorf, T., James, R. S., Carvalho, J. E., Wilson, R. S., Silva, M. D. P., & Navas, C. A. (2004). Locomotor performance of closely related *Tropidurus* species: Relationships with physiological parameters and ecological divergence. *Journal of Experimental Biology*, 207, 1183–1192.
- Kramer, G. (1951). Body proportions of mainland and island lizards. *Evolution* 5, 193–206.
- Laurent, R. F. (1970). Dos nuevas especies argentinas del género *Telmatobius* (Amphibia: Leptodactylidae). *Acta Zoológica Lilloana*, 25, 207–226.
- Lavilla, E. O. (1988). The genus *Telmatobius* (Anura: Leptodactylidae) and its relations with montane wetlands. In G. H. Dalrympe, W. F. Loftus, & F. S. Bernardino (Eds.), *Wildlife in the Everglades and Latin America wetlands* (pp. 16–17 (english) and pp. 51–52 (spanish)). Miami: Florida International University.
- Lavilla, E. O., & Barrionuevo, J. S. (2005). El género *Telmatobius* (Anura: Leptodactylidae) en la república Argentina: Una síntesis. In E. O. Lavilla, & I. De la Riva (Eds.), *Estudios sobre las ranas andinas de los géneros Telmatobius y Batrachophrynus (Anura: Leptodactylidae)* (pp. 115–165). Valencia, España: Asociación Herpetológica Española. Monografías de Herpetología 7.
- Losos, J. B. (1990a). Ecomorphology, performance capability and scaling of West Indian *Anolis* lizards: An evolutionary analysis. *Ecological Monographs*, 60, 369–388.
- Losos, J. B. (1990b). Concordant evolution of locomotor behaviour, display rate and morphology in *Anolis* lizards. *Animal Behaviour*, 39, 879–890.
- Losos, J. B. (1990c). The evolution of form and function: Morphology and locomotor performance in indian *Anolis* lizards. *Evolution*, 44, 1189–1203.
- Losos, J. B. (2009). *Lizards in an evolutionary tree: Ecology and adaptive radiations of anoles*. Berkeley, CA: University of California Press.
- Losos, J. B., Andrews, R. M., Sexton, O. J., & Schuler, A. L. (1991). Behavior, ecology, and locomotor performance of the giant Anole, *Anolis frenatus*. *Caribbean Journal of Science*, 27, 173–179.
- Losos, J. B., Jackman, T. R., Larson, A., de Queiroz, K., & Rodríguez-Schettino, L. (1998). Historical contingency and determinism in replicated adaptive radiations of island lizards. *Science*, 279, 2115–2118.
- Losos, J. B., Walton, M. B., & Bennett, A.F. (1993). Trade-off between sprinting and clinging ability in Kenyan Camaleons. *Functional Ecology*, 7, 281–286.
- Lucas, E., Fortes, V., & Garcia, P. (2016). Amphian, Anura, Hylidae, Phyllomedusa azurea Cope, 1862: Distribution extension to southern Brazil. Available at: www.checklist.org.br.
- Manzano, A. S., Baldo, D., & Barg, M. (2004). Anfíbios del litoral fluvial argentino. *Miscelánea*, 12, 271–290.
- Manzano, A. S., & Barg, M. (2005). The iliosacral articulation in Pseudinae (Anura: Hylidae). *Herpetológica*, 61, 259–267.
- Martin, P., & Bateson, P. (2007). *Measuring behaviour: An introductory guide*. Cambridge: Cambridge University Press.
- McPeck, M. A., & Brown, J. M. (2000). Building a regional species pool: Diversification of the Enallagma damselflies in eastern North American waters. *Ecology*, 81, 904–920.
- Miles, D. B. (1994). Covariation between morphology and locomotor performance in sceloporine lizards. In L. J. Vitt & E. R. Pianka (Eds.), *Lizard ecology: Historical and experimental perspectives* (pp. 207–235). Princeton, NJ: Princeton University Press.
- Nauwelaerts, S., & Aerts, P. (2006). Take-off and landing forces in jumping frogs. *Journal of Experimental Biology*, 209, 66–77.
- Nauwelaerts, S., Stamhuis, E., & Aerts, P. (2005). Swimming and jumping in a semi-aquatic frog. *Animal Biol*, 55, 3–15.
- Nauwelaerts, S., Aerts, P., & D' Aout, K. (2001). Speed modulation in swimming frogs. *Journal of Motor Behavior*, 33, 265–272.
- Nauwelaerts, S., Ramsay, J., & Aerts, P. (2007). Morphological correlates of aquatic and terrestrial locomotion in a semi-aquatic frog, *Rana esculenta*: No evidence for a design conflict. *Journal of Anatomy*, 210, 304–317.
- Oatis, C. A. (2009). *Kinesiology: The mechanics and pathomechanics of human movement* (2nd ed.). Baltimore, MD: Lippincott Williams & Wilkins.
- Orme, C. D. L., Freckleton, R. P., Thomas, G. H., Petzoldt, T., Fritz, S. A., & Isaac, N. B. J. (2012). *Carper: comparative analyses of*

- phylogenetics and evolution in R. Methods. Ecology and Evolution* 3, 145–151.
- Paradis, E., Claude, J., & Strimmer, K. (2004). APE: Analyses of Phylogenetics and Evolution in R language. *Bioninformatics*, 20, 289–290.
- Peters, S. E., Kamel, L. T., & Bashor, D. P. (1996). Hopping and swimming in the leopard frog, *Rana pipiens*: I. Step cycles and kinematics. *Journal of Morphology*, 230, 1–16.
- Peterson, J. A. (1984). The locomotion of *Chamaeleo* (Reptilian: Sauria) with particular reference of the forelimb. *Journal of Zoology*, 202, 1–42.
- Polly, P. D. (2007). Limbs in mammalian evolution. In B. K. Hall (Ed.), *Fins in to limbs: Evolution, development, and transformation* (pp. 245–268). Chicago: University of Chicago Press.
- Pombal, J. P., Jr., & Haddad, C. F. B. (1992). Espécies de *Phyllomedusa* do grupo Burmeisteri do Brasil oriental, com descrição de uma espécie nova (Amphibia, Hylidae). *Revista Brasileira de Biologia*, 52, 217–229.
- Ponssa, M. L. (2008). Cladistic analysis and osteological descriptions of the frog species in the *Leptodactylus fuscus* species group (Anura, Leptodactylidae). *Journal of Zoological Systematics and Evolutionary Research*, 46, 249–266.
- Přikryl, T., Aerts, P., Havelková, P., Herrel, A., & Roček, Z. (2009). Pelvic and thigh musculature in frogs (Anura) and origin of anuran jumping locomotion. *Journal of Anatomy*, 214, 100–139.
- Púgener, L. A., & Maglia, A. M. (2009). Developmental evolution of the anuran sacro-urostylic complex. *South American Journal of Herpetology*, 4, 193–20.
- Pyron, R. A., & Wiens, J. J. (2011). A large-scale phylogeny of Amphibia including over 2800 species, and a revised classification of extant frogs, salamanders, and caecilians. *Molecular Phylogenetics and Evolution*, 61, 543–583.
- R Core Team. (2014). *R: A language and environment for statistical computing*. Vienna, Austria: R Foundation for Statistical Computing. Retrieved from <http://www.R-project.org/>
- Rage, J. C., & Roček, Z. (1986). *Triadobatrachus* revisited. In Z. Roček (Ed.), *Studies in herpetology* (pp. 255–258). Prague: Charles University Publishers House.
- Rage, J. C., & Roček, Z. (1989). Redescription of *Triadobatrachus massinoti* (Piveteau, 1936) an anuran amphibian from the early Triassic. *Palaeontogr A*, 206, 1–16.
- Rand, A. S. (1952). Jumping ability of certain anurans, with notes on endurance. *Copeia*, 1952, 15–20.
- Reilly, S. M., & Jorgensen, M. E. (2011). The evolution of jumping in frogs: Morphological evidence for the basal anuran locomotor condition and the radiation of locomotor systems in crown group anurans. *Journal of Morphology*, 272, 149–168.
- Reilly, S. M., Montuelle, S. J., Schmidt, A., Krause, C., Naylor, E., & Essner, R. L., Jr. (2016). Functional evolution of jumping in frogs: Interspecific differences in take-off and landing. *Journal of Morphology*, 277, 379–393.
- Revell, L. J. (2009). Size-correction and principal components for interspecific comparative studies. *Evolution*, 63, 3258–3268.
- Revell, J. L. (2012). Phytools: An R package for phylogenetic comparative biology (and other things). *Methods in Ecology and Evolution*, 3, 217–223.
- Richards, C. T. (2010). Kinematics and hydrodynamics analysis of swimming anurans reveals striking inter-specific differences in the mechanism for producing thrust. *Journal of Experimental Biology*, 213, 621–634.
- Sathe, E. A., & Husak, J. F. (2015). Sprint sensitivity and locomotor trade-offs in green anole (*Anolis carolinensis*) lizards. *Journal of Experimental Biology*, 218, 2174–2179.
- Schulte, J. A., II, Losos, J. B., Cruz, F. B., & Nuñez, H. (2004). The relationship between morphology, escape behavior, and microhabitat occupation in the iguanid lizard genus *Liolaemus*. *Journal of Evolutionary Biology*, 17, 48–420.
- Shearman, R. M. (2008). Chondrogenesis and ossification of the Lissamphibian pectoral girdle. *Journal of Morphology*, 269, 479–495.
- Sheil, C. A., & Alamillo, H. (2005). Osteology and skeletal development of *Phyllomedusa vaillanti* (Anura: Hylidae: Phyllomedusinae) and a comparison of this arboreal species with a terrestrial member of the genus. *Journal of Morphology*, 265, 343–368.
- Shubin, N. H., & Jenkins, F. A., Jr. (1995). An early Jurassic jumping frog. *Nature*, 377, 49–52.
- Simons, V. F. H. (2008). *Morphological correlates of locomotion in anurans: Limb length, pelvic anatomy and contact structures*. Thesis. College of Arts and Sciences of Ohio University, Athens.
- Soliz, M. C., & Ponssa, M. L. (2016). Development and morphological variation of the axial and appendicular skeleton in Hylidae (Lissamphibia, Anura). *Journal of Morphology*, 277, 786–813.
- Trueb, L. (1973). Bones, frogs, and evolution. In J. L. Vial (Ed.), *Evolutionary Biology of Anurans: Contemporary Research on Major Problems* (pp. 65–132). Columbia: University of Missouri Press.
- Trueb, L., & Báez, A. M. (2006). Revision of Early Cretaceous Cordicephalus from Israel and assessment of its relationships among pipoid frogs. *Journal of Vertebrate Paleontology*, 26, 44–59.
- Tulli, M. J., Abdala, V., & Cruz, F. B. (2011). Relationships among morphology, clinging performance and habitat use in Liolaemini lizards. *Journal of Evolutionary Biology*, 24, 843–855.
- Tulli, M. J., Abdala, V., & Cruz, F. B. (2012). Effects of different substrates on the sprint performance of lizards. *Journal of Experimental Biology*, 215, 774–784.
- Tulli, M. J., Carrizo, L. V., & Samuels, J. X. (2015). Morphological variation of the forelimb and claw in Neotropical Sigmodontine rodents (Rodentia: Cricetidae). *Journal of Mammalian Evolution*, 23, 81–91.
- Tulli, M. J., Cruz, F. B., Herrel, A., Vanhooydonck, B., & Abdala, V. (2009). The interplay between claw morphology and microhabitat use in neotropical iguanian lizards. *Zoology*, 112, 379–92.
- Tulli, M. J., Cruz, F. B., Kohlsdorf, T., & Abdala, V. (2016). When a general morphology allows many habitat uses. *Integrative Zoology*, 11, 473–489.
- Vaira, M. (2001). Breeding biology of the leaf frog, *Phyllomedusa boliviana* (Anura, Hylidae). *Amphibia-Reptilia*, 22, 421–429.
- Vanhooydonck, B., & Van Damme, R. (2011). Evolutionary trade-offs in locomotor capacities in lacertid lizards: Are splendid sprinters clumsy climbers? *Journal of Evolutionary Biology*, 14, 46–54.
- Vellard, J. (1954). *Études sur le lac Ticicaca v Les Telmatobius du haut-plateau interandin* (p. 157). Javier Prado: Mem Mus Hist Nat.
- Wake, M. H. (1998). Amphibian locomotion in evolutionary time. *Zoology*, 100, 141–151.
- Wassersug, R. J. (1976). A procedure for differential staining of cartilage and bone in whole formalin-fixed vertebrates. *Stain Technology*, 51, 131–134.
- Wells, K. D. (2007). *The ecology and behavior of Amphibians*. Chicago: The University of Chicago press.
- Wiens, J. J., Kuczynski, C. A., Hua, X., & Moen, D. S. (2010). An expanded phylogeny of treefrogs (Hylidae) based on nuclear and mitochondrial sequence data. *Molecular Phylogenetics and Evolution*, 55, 871–882.

- Zani, P. A. (2000). The comparative evolution of lizard claw and toe morphology and clinging performance. *Journal of Evolutionary Biology*, 13, 316–325.
- Zar, J. H. (1999). *Biostatistical analysis*. Upper Sadle River: Prentice Hall.
- Zug, G. R. (1972). Anuran locomotion: Structure and function. Preliminary observations on relation between jumping and osteometrics of appendicular and postaxial skeleton. *Copeia*, 1972, 613–624.
- Zug, G. R. (1978). Anuran locomotion: Structure and function 2. Jumping performance of semiaquatic, terrestrial, and arboreal frogs. *Smithsonian Contributions to Zoology*, 276, 1–31.

SUPPORTING INFORMATION

Additional Supporting Information may be found in the online version of this article.

How to cite this article: Soliz M, Tulli MJ, and Abdala V. Osteological postcranial traits in hylid anurans indicate a morphological continuum between swimming and jumping locomotor modes. *Journal of Morphology*. 2017;00:000–000. doi:10.1002/jmor.20654.