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Colour change in Darwin's frogs (*Rhinoderma darwinii*, Duméril and Bibron, 1841) (Anura: Rhinodermatidae)

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In the laboratory, wild-caught male *Rhinoderma darwinii* frogs, but not females, changed body colour from brown to green starting with the appearance of green dorsal spots or a greenish dorsal tone on a brown body colour. After a year in captivity, most males exhibited a greenish or complete green dorsal colour, probably induced by the terrarium's swamp (green) vegetation and season.

Keywords: anuran; Darwin's frogs; *Rhinoderma*; colour change; Chile

Introduction

Colour change has been suggested as an alternative solution to the problem of background colour matching in an environment consisting of spectrally heterogeneous microhabitats. Alternatively, it may disrupt the outline of animals, allowing them to use more than one microhabitat while reducing predation risk (Tordoff 1980; Endler 1988; Stevens and Cuthill 2006).

Colour change occurs in a number of green amphibians and reptiles. The green colour is produced by a combination of structural blue colour and yellow/orange/red pterines or carotenoids, which can change to other colours like brown in anolis lizards or tree frogs (Bagnara and Matsumoto 2006; Bagnara et al. 2007). A colour change for example from brown to green, or vice versa, can be accomplished by frogs over periods of weeks to months as in *Hyla regilla* (Mackey 1958; Haugen 1992). A change from a green stripe to a grey stripe, or vice versa, took 6 months in *Acris crepitans* (Pyburn 1961).

Colour changes in amphibians can be non-reversible, as ontogenetic colour changes that are limited to a particular period of development, or reversible as in physiological colour changes, where frogs can lighten or darken existing tones (all frogs; Hoffman and Blouin 2000), day and night colouration (e.g. *Hyla arborea*), colour changes after sloughing (e.g. *Neobabrachus pictus*; Henle 2010), pathological colour changes (e.g. by chytrids; Fellers et al. 2001), and breeding colouration, such as blue in male moor frogs (*Rana arvalis*) or lemon yellow in male frogs (*Rana lessonae*; Günther

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1996; Ries et al. 2008, and *Scinax* spp.; Haddad et al. 2008). Colour traits can also function in mate recognition and mate choice, thereby selecting traits such as mimetic colouration (Jiggins et al. 2001), brightness (Maan and Cummings 2009) or specific colouration (Summers et al. 1999) causing reproductive isolation (Jiggins et al. 2001) shaped by sexual selection.

Darwin's frog, *Rhinoderma darwinii*, is characterized by a nasal prolongation, a variable body colouration (between green and brown) and dorsal patterning (two to five patterns in a population), and most of all a unique brood system, in which males brood their offspring after hatching in their vocal pouch until metamorphosis is complete. This frog inhabits the south of Chile and occurs in the leaf litter of the cool temperate rainforest (Valdivian forest). Unfortunately, knowledge of *R. darwinii*'s ecology is limited to the study of Crump (2002). Darwin's frogs are currently endangered and declining (Young et al. 2001), probably because of sickness caused by chytrid fungus (Bourke et al. 2010) and of habitat destruction (Neira et al. 2002)

In populations of Darwin's frog the background colour (Wilhelm 1927; Kilian 1965) as well as temperature (Kilian 1965; Crump 2002) correlate with body colour hue, but individual colour changes have only been mentioned by Janvier (1935) and Busse (2003), but they supplied no data. The aim of the present study was to confirm whether *R. darwinii* individuals are capable of colour change.

Materials and methods

Twenty individual Darwin's frogs were collected in Coñaripe (39°23' S, 71°23' W) (Figure 1). In 2009 they were collected in a secondary forest creek at 400 m above sea level and in 2010 in a primary forest at 800 m above sea level. A total of 11 individuals were collected in 2009 and nine in 2010. All individuals were brought into indoor terraria with swamp vegetation (moss, ferns and leaf litter) to initiate the captive rearing facility located in Concepción, Chile. All individuals from 2009 and 2010 were held under the same conditions of light regimen, water source and diet. The only condition that differed between the 2009 and 2010 samples was the housing. Individuals from 2009 were held in groups of two males and one female, and individuals from 2010 in groups of two females and one male. For the 2009 collection, each individual was photographed in autumn at the time of collection (March 2009), one year later (February 2010) and then again seven months later in spring (October 2010). Individuals from 2010 were photographed at the time of collection in autumn (February 2010) and then again seven months later in spring (October 2010). Individuals were photographed dorsally and ventrally to document their colouration and ventral pattern, respectively. The latter is unique for each individual and remains constant during adult life allowing reliable individual recognition.

Results

Six of the eleven individuals collected in 2009 (54.5%) exhibited colour change, which included all brown males, but not females or completely green males (Table 1, Figure 2). The colour change started with green irregular spots or a greenish tone (hue) located in the middle dorsal area between the flanks (Figure 2) and, in general, took a couple of weeks or months (C B., personal observation). None of the nine individuals

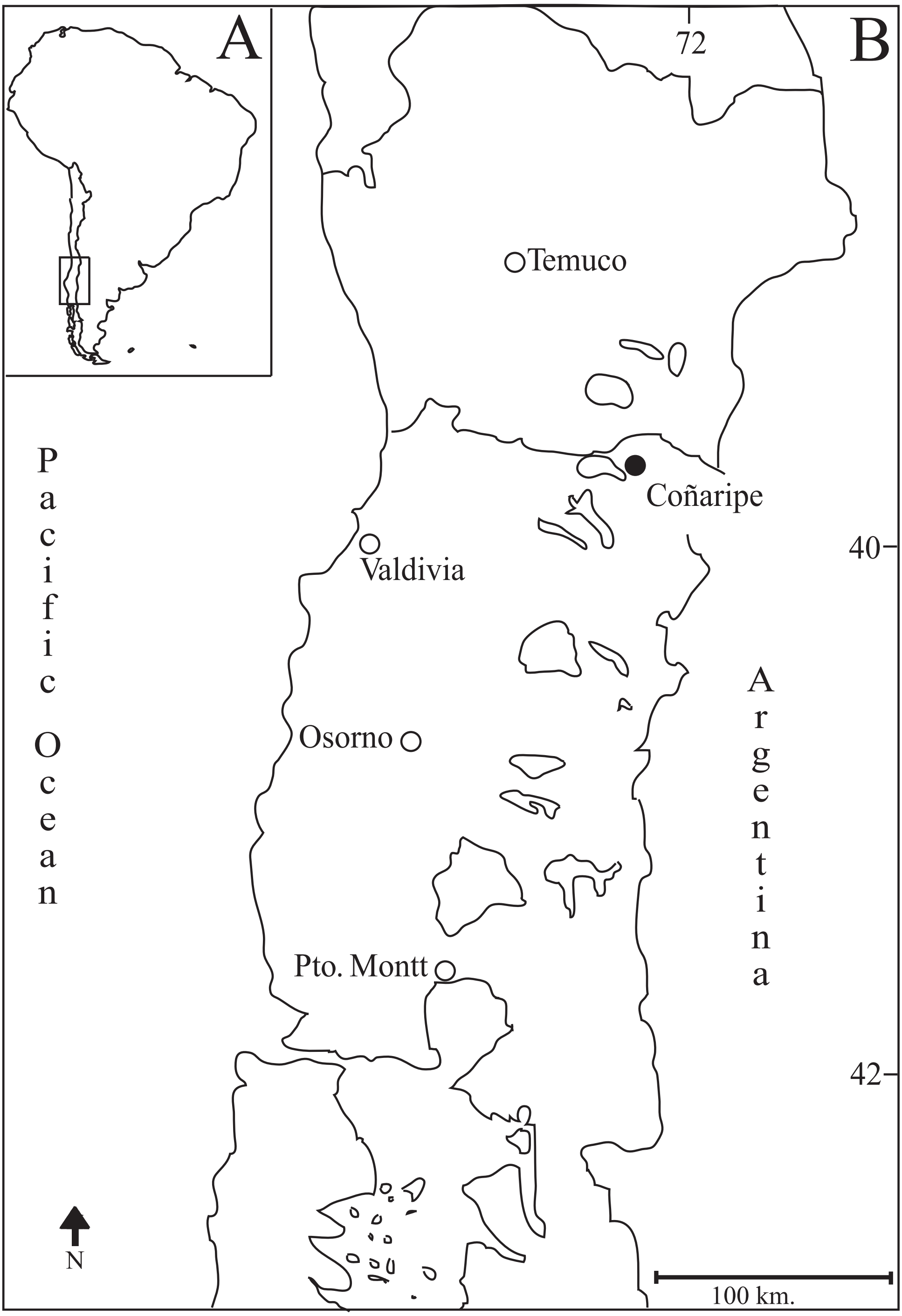


Figure 1. (A) Chile's location in South America. (B) Coñaripe's location in central Chile. Open circles, cities for geographical reference; filled circle, collection location.

Table 1. Colour change in *Rhinoderma darwinii* individuals from Concepción rearing facility.

Collection time	Individual number	Size at collection (SVL, TL) in mm	Sex	Colour at collection	Colour change
March 2009	32	25.9,13.2	male	brown	yes
	33	27.4,12.5	female	brown	no*
	34	26,12.8	male	brown	yes
	35	21.5,10	male	brown	yes
	36	21.5,11	male	brown	yes
	37	26.2,12.2	male	completely green	no
	38	26,13	female	brown	no*
	39	27,12	male	completely green	no
	41	29,12	female	brown	no
	42	26.2,12.2	male	brown	yes
	44	24.8,12.2	male	brown	yes
March 2010	1	25.2,11	male	green	no
	2	28.4,11.7	female	brown	no
	3	24.6,11.5	female	brown	no
	5	25.9,11.0	male	brown	no
	6	26.5,12.2	female	brown	no
	8	26.3,11.7	female	brown	no
	11	28.4,11.4	female	brown	no
	12	27.6,13	female	brown	no
	14	28.3,11	male	green	no

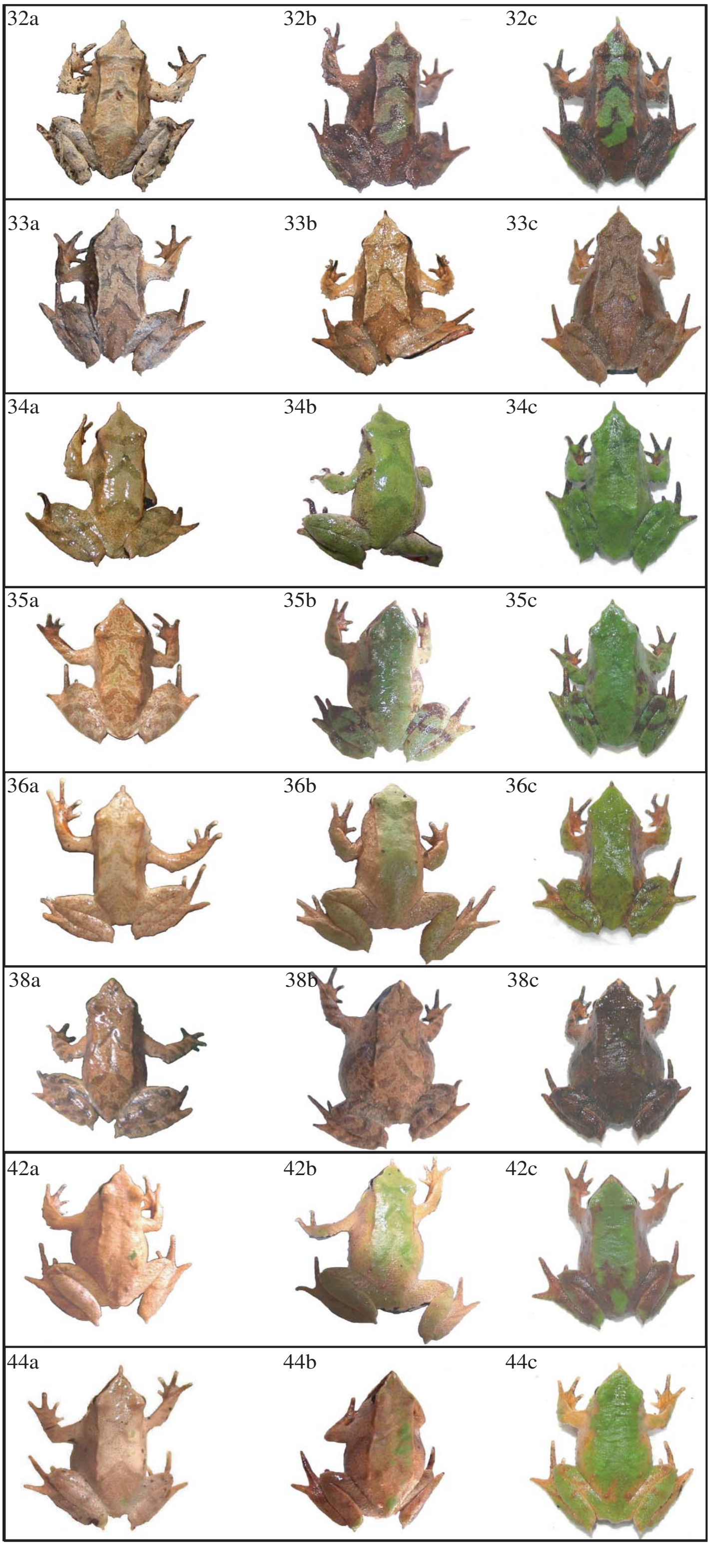
Notes: SVL, snout–vent length; TL, tibia length.
*Green colour spots enhanced in November 2010.

collected in 2010 changed colour, not the single brown male, the six females, or the two completely green males.

Discussion

We demonstrated for the first time individual colour change in *R. darwinii*. Colour change had been mentioned by Janvier (1935) and in some developing individuals by Busse (2003). Also, previously some individuals exhibited colour change during the spring, changing from some green spots to completely green, at the Zoologisches Forschungsmuseum Alexander Koenig (ZFMK) rearing facility in Bonn, Germany (Busse personal communication). This process agrees with the colour change observed in the present study at the Concepción rearing facility. The colour change was probably induced by the breeding season, light and the terrarium's swamp vegetation, which was mainly green (moss and ferns), in both captive rearing facilities. The frequency of

Figure 2. *Rhinoderma darwinii* individuals that exhibited colour change. Individual number is indicated in each case. (A) Individuals in March 2009, (B, C) the same individuals in (B) February 2010 and (C) October 2010.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38b
38c
42a
42b
42c
44a
44b
44c

the different colour states probably depends on the area in which the individuals are living (Wilhelm 1927; Janvier 1935; Kilian 1965).

In the present study colour change was limited to the male frogs. At the ZFMK rearing facility, some females also turned green (K.B., personal observation) and in the field population at Coñaripe females with green spots have also been observed (J.B., personal observation). Two of the three females from the 2009 collection exhibited a green spot by the end of November 2010 (Table 1; C.B., personal observation), which is the beginning of the summer season in the southern hemisphere. For this reason we suggest that colour change is related to sex, being more prevalent in males, but also possibly occurring in females. Further studies are clearly needed.

Individuals collected in 2010 did not change colour. Possible reasons for this could be the following. (1) Most of the 2010 individuals were females and green males, which did not change colour in the 2009 individuals. (2) The 2010 frogs may not yet have been sufficiently stimulated by the breeding season, light and substrate colour to change colour. (3) Although all individuals were collected at Coñaripe, they were collected from different areas in 2009 and 2010, that were located at different altitudes, and had a different land relief and vegetation. (4) In 2009, male–male competition was possible and may have enhanced the colour change.

Our results agree with the previously described colour changes in amphibians that were also related to sex, habitat and breeding season. In *Mannophryne trinitatis*, males assume a cryptic brown colouration when not calling or defending calling sites, but turn black and attack other males when calling (Wells 2007). In *Scinax elaechrous* and *Isthmohyla psedopuma*, males turn brighter or adopt a different colour during the breeding season (Savage 2002). In red spotted newts (*Notophthalmus v. viridescens*), background colouration changes with habitat, as adults migrating to the forest for the non-breeding season (winter) become browner, suggesting that background colour serves a camouflage function in the adult stage (Davis and Grayson 2007).

In summary, we prove for the first time that *R. darwinii* can undergo colour change. This colour change is probably induced by the breeding season and microhabitat change, as suggested by Janvier (1935). Colour change may benefit frogs by better camouflaging them in their microhabitat and thereby increasing their survival. Males particularly, which care for the offspring, would have a greater reproductive success if matched to greener and warmer microhabitats. Lighter coloured frogs have a higher metabolic rate (Wente and Phillips 2003) and the resulting higher temperatures accelerate tadpoles' metamorphosis (Werner and Glennmeier 1999; Skelly et al. 2002).

Further studies are obviously needed, which focus on season-related and sex-related colouration and colour change. The ability to change colour is an advantage for this species because it uses different microhabitats. In addition to natural selection (crypsis and decreased predation risk), male colouration may also be under sexual selection.

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References

Bagnara JT, Fernandez PJ, Fuiji R. 2007. On the blue coloration of vertebrates. Pigment Cell Res. 20:14–26.

Bagnara JT, Matsumoto J. 2006. The pigmentary system: physiology and pathophysiology. 2nd edition. Oxford: Blackwell Publishing. Chapter I, Comparative anatomy and physiology of pigment cells in nonmammalian tissues, p. 11–59.

Bourke J, Mutschmann F, Ohst T, Ulmer P, Gutsche A, Busse K, Werning H, Böhme W. 2010. *Batrachochytrium dendrobatidis* in Darwin's frog *Rhinoderma* spp. in Chile. Dis Aqua Org. 92:217–221.

Busse K. 2003. Fortpflanzungsbiologie von *Rhinoderma darwinii* (Anura: Rhinodermatidae) und die stammesgeschichtliche und funktionelle Verkettung der einzelnen Verhaltensabläufe. Bonn Zool Beitr. 51:3–34.

Crump ML. 2002. Natural History of Darwin's frog, *Rhinoderma darwinii*. Herpetol Nat Hist. 9:21–30.

Davis AK, Grayson KL. 2007. Improving natural history research with image analysis: the relationship between skin color, sex, size, and stage in adult red-spotted newts (*Notophthalmus viridescens viridescens*). Herpetol Conserv Biol. 2:67–72.

Endler JA. 1988. Frequency-dependent predation, crypsis and aposematic coloration. Phil Trans R Soc Lond B. 319:505–523.

Fellers GM, Green DE, Longcore JE. 2001. Oral chytridiomycosis in the mountain yellow-legged (*Rana muscosa*). Copeia. 2011:945–953.

Günther R. 1996. Die Amphibien und Reptilien Deutschlands. Jena: Gustav Fisher.

Haddad CFB, Toledo LF, Prado CPA. 2008. Anfíbios da MataAtlântica, São Pablo: editora neotropica.

Haugen L. 1992. Background color selection and color change in cryptic hylid frog, *Pseudacris regilla*. [Masters thesis]. [Northridge (CA)]: California State University.

Henle K. 2010. A hitherto overlooked type of colour change in frogs and its significance for the identification of species in the Australian genus *Neobatrachus*. Amph-Rep. 31:282–286.

Hoffman EA, Blouin MS. 2000. A review of color and pattern polymorphisms in anuran. Biol J Linn Soc. 70:633–665.

Janvier H. 1935. Observations biologiques sur les *Rhinoderma darwinii*. Ann Sci Nat Paris. 10:197–204.

Jiggins CD, Naisbit RE, Coe RL, Mallet J. 2001. Reproductive isolation caused by colour pattern mimicry. Nature. 411:302–305.

Kilian E. 1965. Das Farbkleid von *Rhinoderma darwinii* D. & B., seine Zeichnungsmuster und Variabilität. Beitr Neotrop Fauna. IV:180–190.

Maan ME, Cummings ME. 2009. Sexual dimorphism and directional sexual selection on aposematic signals in a poison frog. Proc Natl Acad Sci USA. 106:19072–19077.

Mackey JP. 1958. Morphological variation among populations of the pacific tree frog (*Hyla regilla*, Baird and Girard) at several elevations in western Oregon. [Ph. D. Thesis]. [Eugene (OR)]: University of Oregon.

Neira E, Verscheure H, Revenga C. 2002. Chile's Frontier Forests: Conserving a Global Treasure. World Resources Institute, Comite´ Nacional Pro Defensa de la Fauna y Flora, University Austral of Chile.

Pyburn WF. 1961. Vertebrate speciation. Austin: University of Texas Press. Chapter IV, The inheritance and distribution of the vertebral stripe color in the cricket frog, p. 235–261.

Ries C, Sztatecsny M, Hödl W. 2008. Geschlechtspezifischer Farbwechsel beim Moorfrosch (*Rana arvalis*) während der Paarungszeit. Z. Feldherpetol Suppl. 13:127–134.

Savage JM. 2002. The Amphibian and Reptilians of Costa Rica: A Herpetofauna between Two Continents, between Two Seas. Chicago and London: University of Chicago Press.

Skelly DK, Freidenburg LK, Kiesecker JM. 2002. Forest canopy and the performance of larval amphibians. Ecology. 83:983–992.

Stevens M, Cuthill IC. 2006. Disruptive coloration, crypsis and edge detection in early visual processing. Proc R Soc Lond B. 273:2141–2147.

Summers K, Symula R, Clough M, Cronin T. 1999. Visual mate choice in poison frogs. Proc R Soc Lond B. 266:2141–2145.

Tordoff W. 1980. Selective predation of grey jays, *Perisoreus canadensis*, upon boreal chorus frogs, *Pseudacris triseriata*. Evolution. 34:1004–1008.

Wells KD. 2007. Ecology and Behavior of Amphibians. Chicago and London: The University of Chicago Press. Chapter VIII, Mating systems and sexual selection in anurans, p. 338–402.

Wente WH, Phillips JB. 2003. Fixed green and brown color morphs and a novel color-changing morph of the pacific tree frog *Hyla regilla*. Am Nat. 162:461–473.

Werner EE, Glennmeier KS. 1999. Influence of forest canopy cover on the breeding pond distributions of several amphibian species. Copeia. 1999:1–12.

Wilhelm O. 1927. *La Rhinoderma darwinii* D. y B. Bol Soc Biol Concepción, Chile. I:11–39.

Young BE, Lips KR, Reaser JK, Ibánez R, Salas AW, Cedeño JR, Coloma LA, Ron S, La Marca E, Meyer JR, et al. 2001. Population declines and priorities for amphibian Conservation in Latin America. Conserv Biol. 15(5):1213–1223.