

Occurrence of *Rhinella rubescens* (Lutz, 1925) (Anura: Bufonidae): update and comments on distribution

Alejandro Valencia-Zuleta^{1,2,*}, Paula Hanna Valdujo³ and Natan Medeiros Maciel²

Abstract. The rufous toad (*Rhinella rubescens*) is an endemic Brazilian amphibian known from the Cerrado Biome. In this paper we analyse the recorded distribution of this species based on registries deposited in collections and literature-based records. Here we estimate the habitat suitability of *R. rubescens* using Maxent. We found that although this species is predominately distributed within the Cerrado, occur also in the Atlantic Forest and Caatinga biomes. The distribution appears to be restricted to altitudinal ranges between 400 and 1400 m above the sea level. Moreover, its distribution is also strongly influenced by temperature and precipitation. Our analyses recovered a low support for the distribution of this species in the States of Pará, Piauí and Tocantins, as early indicated in the literature. However, high suitability was recovered between the limits of Tocantins and Goiás states, which should be verified. The hypothesized distribution provided here for *R. rubescens* differs in area from presented by the International Union for Conservation of Nature (IUCN). Therefore, results delivered here could improve knowledge for future research approaching new possible localities verification, reassessment conservation category, and scenarios with the climate change of the rufous toad.

Keywords: Rufous toad, Endemic species, Cerrado biome, Species Distribution Model.

Introduction

Rhinella Fitzinger, 1826 is a genus of toads distributed from southern United States of America to the southern part of South America. Currently, it comprises 91 species (Frost 2016) assigned to seven groups, including the *Rhinella marina* species group. Members of the *Rhinella marina* group are characterized by having large sizes, developed parotoid glands and presence of all cranial crests (Pramuk 2006). The group currently

comprises 11 species: *R. marina* (Linnaeus, 1758), *R. ictERICA* (Spix, 1824), *R. horribilis* (Wiegmann, 1833), *R. poeppigii* (Tschudi, 1845), *R. arenarum* (Hensel, 1867), *R. schneideri* (Werner, 1894), *R. rubescens* (Lutz, 1925), *R. jimi* (Stevaux, 2002), *R. achavali* (Maneyro, Arrieta, and de Sá, 2004), *R. veredas* (Brandão, Maciel, and Sebben, 2007) and *R. cerradensis* Maciel, Brandão, Campos, and Sebben, 2007.

Within the *Rhinella marina* species group, *R. rubescens* is characterized by the presence of (i) red brick coloration in the cranial crests, limbs and sometimes in the venter simulating erythema spots besides the (ii) long and narrow parotoid glands (Lutz 1925). *R. rubescens* can be confused with *R. veredas* in sympatry. However, unlike *R. veredas*, *R. rubescens* have a complete supraocular cranial crest, long and narrow parotoid gland, absence of glandular ridges extended from the posterior end of the parotoid gland to the groin, smaller size, dorsal coloration tan, brow or grey in males (yellow in *R. veredas*), greater extent of the foot webbing and also by the form of the head with round snout in dorsal view and obtuse in lateral view (Brandão et al. 2007).

Rhinella rubescens is an endemic species from Brazil, occurring in the biomes of Cerrado (Araújo et al. 2007,

¹ Programa de Pós-Graduação em Biodiversidade Animal, Instituto de Ciências Biológicas, Universidade Federal de Goiás, Cx. Postal 131, 74001-970 Goiânia, GO, Brazil.

² Laboratório de Herpetologia e Comportamento Animal, Departamento de Ecologia, Instituto de Ciências Biológicas, Universidade Federal de Goiás, Cx. Postal 131, 74001-970 Goiânia, GO, Brazil.

³ WWF-Brasil, Laboratório de Ecologia da Paisagem - Superintendência de Conservação. Entre Quadra SHIS EQL 6/8 Conjunto E, Setor de Habitações Individuais Sul, 71620430 - Brasília, DF - Brazil

* Corresponding author e-mail: alejandrovalencia08@gmail.com

2009, São-Pedro & Feio 2011, Valdujo *et al.* 2012, Magalhães *et al.* 2015) and Caatinga (Camardelli & Napolli 2012). This species is known to occur in the states of Pará, Piauí, Tocantins, Bahia, Goiás, Minas Gerais, São Paulo and Mato Grosso do Sul (Araujo *et al.* 2007, Bastos *et al.* 2010, Aoki *et al.* 2011, Frost 2016). Although based on specialist criterion, covering mainly the distribution of Brazilian Cerrado Biome, many new records for the *R. rubescens* appeared in the literature and a revision of them is needed. Herein we present a review of the sampled localities for *R. rubescens* by provide (i) an update distribution map for the species from the species distribution model (SDM) and (ii) present comments regarding distribution of *Rhinella rubescens*.

Materials and methods

Species occurrence data.—Occurrences for *Rhinella rubescens* (see Appendices I) were directly retrieved from collections or by confirmation of species with high resolution photographs. The following collections were included: Museum of Natural History, Division of Amphibians and Reptiles, Washington, D.C., USA (USNM); Museum of Comparative Zoology, Harvard University, Cambridge, Massachusetts, USA (MCZ); Museu de Zoologia da Universidade of São Paulo, São Paulo, Brazil (MZUSP); Museu Nacional, Rio de Janeiro, Brazil (MNRJ); Museu de História Natural, Universidade Estadual de Campinas, 13081-970 Campinas, São Paulo, Brazil (ZUEC); Célio F.B. Haddad Collection, deposited in the Departamento de Zoologia, Universidade Estadual Paulista, Campus de Rio Claro, São Paulo, Brazil. (CFBH); Coleção de Anfíbios, Universidade Estadual Paulista, São José do Rio Preto, Brazil (DZSJRP); Coleção Herpetológica da Universidade de Brasília, Brasília, Brazil (CHUNB); and Coleção Herpetológica da Universidade Federal de Goiás, Goiânia, Brazil (ZUFG). In addition, geographic coordinates for the species were retrieved from Leite *et al.* (2009), Aoki *et al.* (2011), and Medeiros-Magalhães *et al.* (2015).

Environmental variables.—We used 20 environmental variables (resolution 30 arc-seconds: ~1 km) of the current climate conditions retrieved from WorldClim database (<http://www.worldclim.org/>) and Jarvis *et al.* (2008). The environmental layers were cropped between latitude 0° to -30° and longitude -35° to -65°, considering the IUCN distribution for *R. rubescens* and the area where the species most likely occur.

Statistical Analysis and Spatial Modeling.—Selection of the environmental variables follows Pearson correlations estimated in Hmisc package (Harrel 2015) from R software (R Development Core Team 2016). All correlations values (r) were significant ($p < 0.001$). Variables highly correlated with $|r| \geq 0.7$ (as threshold appropriated by Dormann *et al.* 2013), were excluded, considering the lower biological relevance to the species. Correlation analysis resulted in 13 variables for inclusion in the initial SDM: Altitude (Alt), Mean Diurnal Range (BIO2), Isothermality (BIO3), Max Temperature of Warmest Month (BIO5), Mean Temperature of Wettest Quarter (BIO8), Mean Temperature of Driest Quarter (BIO9), Mean Temperature of Coldest Quarter (BIO11), Annual Precipitation (BIO12), Precipitation Seasonality (BIO15), Precipitation of Wettest Quarter (BIO16), Precipitation of Driest Quarter (BIO17), Precipitation of Warmest Quarter (BIO18), Precipitation of Coldest Quarter (BIO19). These 13 variables were included in the initial analysis (model calibration) in Maxent 3.33k version (Phillips *et al.* 2004). The initial SDM were realized with one run (raw output setting) to calculate Akaike Information Criteria (AICc) in ENMTools v.1.3 (Warren *et al.* 2008). AICc was conducted to know performance model and computed based on jackknife analyses of variables, as follows: thirteen, twelve, eleven, ten, nine, eight, seven, six, five, four, three, two, and one of the variables, respectively (Table 1). The best model (lowest AICc value and delta AIC) presented

Table 1. Akaike's Information Criterion (AICc) values and delta AIC (Δ) for choose the best model for a Maxent model with differing numbers of variables.

Number of variables in the Maxent model	AICc	AIC (Δ)
13	1930.04	44.85
12	1912.52	27.33
11	1914.59	29.40
10	1921.42	36.23
9	1911.71	26.51
8	1894.59	9.39
7	1885.19	0.00
6	1923.79	38.60
5	1923.78	38.58
4	1949.85	64.66
3	1952.46	67.27
2	1947.45	62.26
1	1957.87	72.67

seven variables: Alt, BIO5, BIO8, BIO9, BIO11, BIO16 and BIO19.

For the final model, the seven variables and data occurrence were included in Maxent. Maxent analyses was set as follow: (i) logistic output format; (ii) created response curves; (iii) conducted jackknife analyses to measure variable importance; (iv) output file type ASCII; (v) select 10,000 background points; (vi) select a random seed; (vii) set random test percentage at 10%, (viii) set replicates at 100, (iv) replicated run-type was set as subsample, (x) chose that plot date be written, and (xi) set maximum iterations to 5,000. Using ArcGIS v.10 (ESRI 2011), ASCII files were converted to raster format, in which the values range 0 to 1, with values closer to 1 corresponding to more suitable habitat for the species. This classification was based on the maximum training sensitivity plus specificity logistic threshold (= 0.11) produced by the Maxent model.

Model evaluation.—To evaluate the final Maxent model with seven variables compared to random expectations we calculated a partial AUC (pAUC) as suggested by

Peterson et al. (2008). However, we used the AUC set random test percentage at 10% (train and test AUC) for supported the pAUC. A pAUC value of 1 indicates the model performed no better than random; values >1 indicate the model performed better than random. For the pAUC analysis, the suitability values of each testing point are used to calculate pAUC and we ran 1000 iterations with 50% of the points resampled with replacement for each bootstrap with software Partial ROC (Barve 2008). We also conducted a multiple linear regression with *lm* function from *stats* package in R software for testing relation between suitability with the seven variables.

Results

Rhinella rubescens occurs mainly along smooth to steep hillsides and uplands of Central and Southeastern Brazil, mostly ranging from 750 to 1,000 meters above sea level (m a.s.l.). Also few specimens were collected in the elevations as low as 430 m a.s.l. or as high as 1,400 m a.s.l. in the states of Bahia, Minas Gerais,

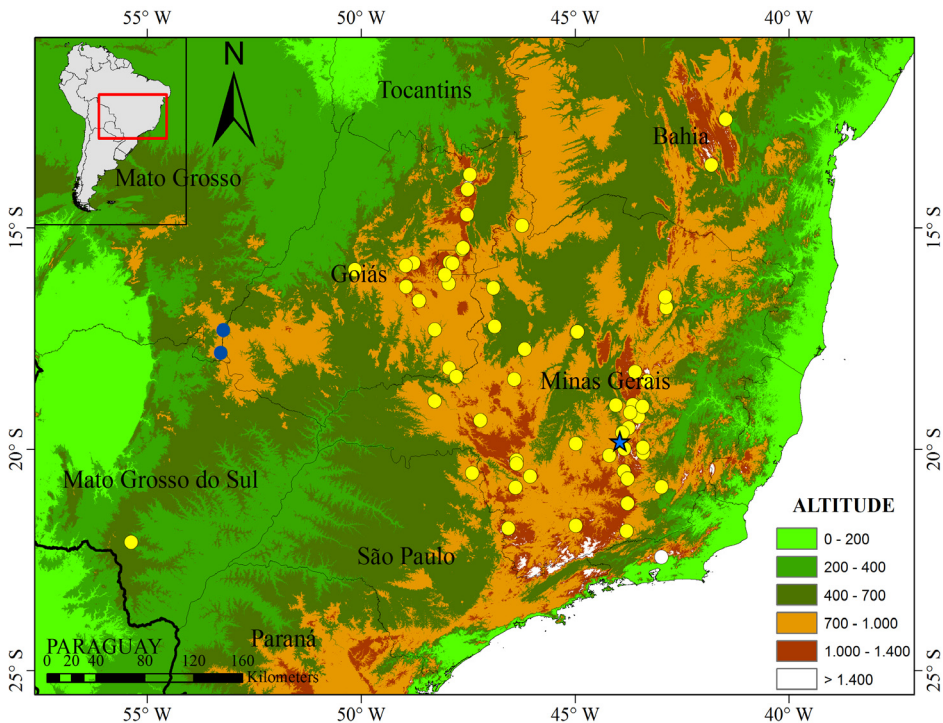


Figure 1. Distribution map of *Rhinella rubescens* based on museums/collections records and the literature. Type locality: Belo Horizonte (blue star) and new localities for the states of Mato Grosso (blue circles) and Rio de Janeiro (white circle).

Rio de Janeiro, São Paulo, Goiás, Distrito Federal (Brasília), Mato Grosso and Mato Grosso do Sul (Fig. 1). We registered three new records for the species in both eastern and western extremes of currently known



Figure 2. *Rhinella rubescens* (MZUSP 152084) collected in Alto Taquari, Mato Grosso state, Brazil.

species distribution across the states of Mato Grosso and Rio de Janeiro, in Brazil (Fig. 1). (i) An adult female (USNM 97656) was collected in March, 1935 in Teresópolis, Rio de Janeiro state ($22^{\circ}25'59.88''\text{S}$, $42^{\circ}58'59.88''\text{O}$; 967 m a.s.l.). This new locality corresponds to the easternmost record for this species, being 103 kilometers in straight line from Lima Duarte (Minas Gerais state, Brazil), the nearest distribution. (ii) Two adult males of *R. rubescens* (MZUSP 66752-3) collected in reproductive condition with keratinization in dorsal tubercles and fingers I, II and III. Specimens were collected on May 8, 1989 in Coqueiro Farm, Alto Araguaia ($17^{\circ}18'56.81''\text{S}$, $53^{\circ}13'6.96''\text{O}$; 670 m a.s.l.) in Mato Grosso state by J.P. Caldwell. (iii) One specimen (MZUSP 152084; Fig. 2) collected in Garrote Farm, Alto Taquari ($17^{\circ}49'33.60''\text{S}$, $53^{\circ}16'55.20''\text{O}$; 879 m a.s.l.) by P.H. Valdujo. These last localities are 357 (Alto Araguaia) and 390 (Alto Taquari) kilometres in straight line from the westernmost currently known (Goiás, Goiás state, Brazil) distribution for *R. rubescens*.

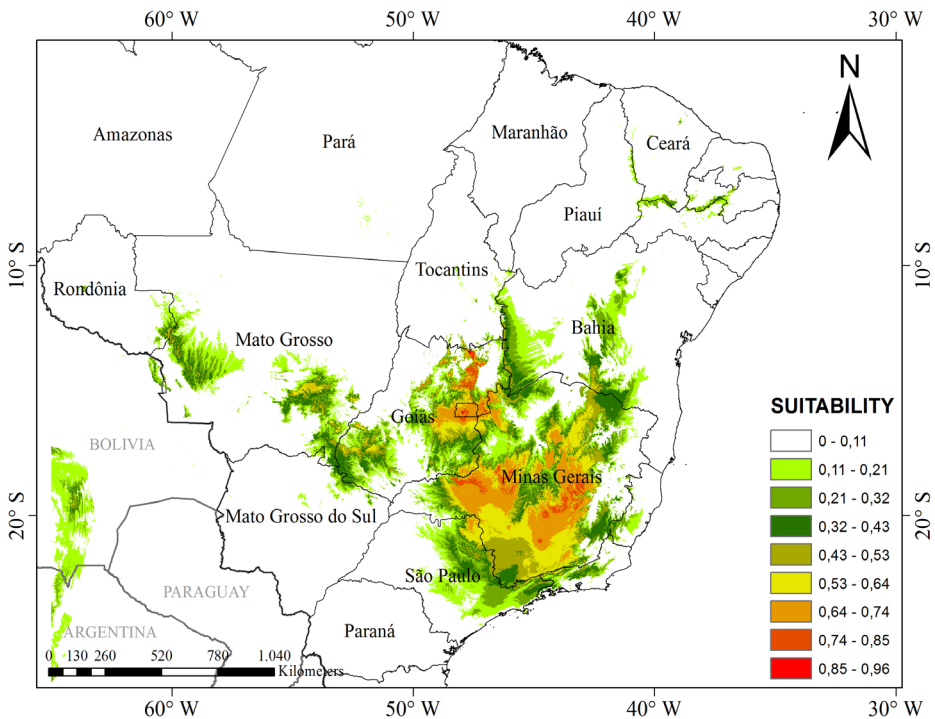


Figure 3. Suitable habitat map of *Rhinella rubescens* modelling in Maxent based on records of occurrence obtained in museums/collections and literature. The "SUITABILITY" predicted the suitability of habitat for this species.

The distribution model for *Rhinella rubescens* (Fig. 3) predicted by environmental variables presented a mean pAUC ratio of 1.81 (sd = 0.07), indicating that the model overcomes random expectations. This is confirmed with the mean test-AUC (0.94, sd = 0.02). A mean train AUC of 0.96 (sd = 0.003) suggests good performance of the model for identifying potential occurrences with low levels of commission and omission errors. Only one occurrence in the Dourados (Mato Grosso do Sul state) was omitted by the model results. The distribution model predicts high habitat suitability of *R. rubescens* mainly associated with plateaus and mountain chains in central and east-central Brazil, existent in the states of Minas Gerais and Goiás. However, interesting localities could be found along the border between Goiás and Tocantins, where a suitability percentage was presented.

The high suitability area (above the 75th percentile values in the model) is associated with altitude (Fig. 4). This is reflected in the percentage of importance in the jackknife analyses for the Alt (gain: train = 1.48, sd = 0.04; test = 1.48, sd = 0.33). Three additional important variables for the species are associated with

the temperature. The first was the BIO5 (gain: train = 1.11, sd = 0.04; test = 1.14, sd = 0.33) meaning the mean temperature of Warmest Month that varied for the species between 25.7 and 32.3°C (mean = 29°C). The second was the BIO8 (gain: train = 1.04, sd = 0.03; test = 1.10, sd = 0.30), which is the mean temperature of Wettest Quarter that showed temperatures between 19.2 and 25.2°C (mean = 22.4). The third important BIO11 (gain: train = 0.73, sd = 0.02; test = 0.78, sd = 0.22) was the mean of Colder Quarter with temperatures between 14.7 and 23.3°C (mean = 18.8). Three variables, BIO9 and the two precipitation variables (BIO19 and BIO16) did not contribute in the same level for the model. BIO16 was the variable with the lowest contribution when it was considered alone in the model (Fig. 5). In contrast to the importance index, only one temperature (BIO5) and two precipitation variables (BIO16 and BIO19) explained 74% of *R. rubescens* suitability by multiple linear regression ($r^2=0.74, p<0.01$). The variables BIO5 (coefficient = -0.001; $p<0.01$) and BIO19 (coefficient = -0.0034; $p<0.01$) indicated that 1°C or 1mm of rain, the suitability decreases in -0.001 or -0.0034, respectively.

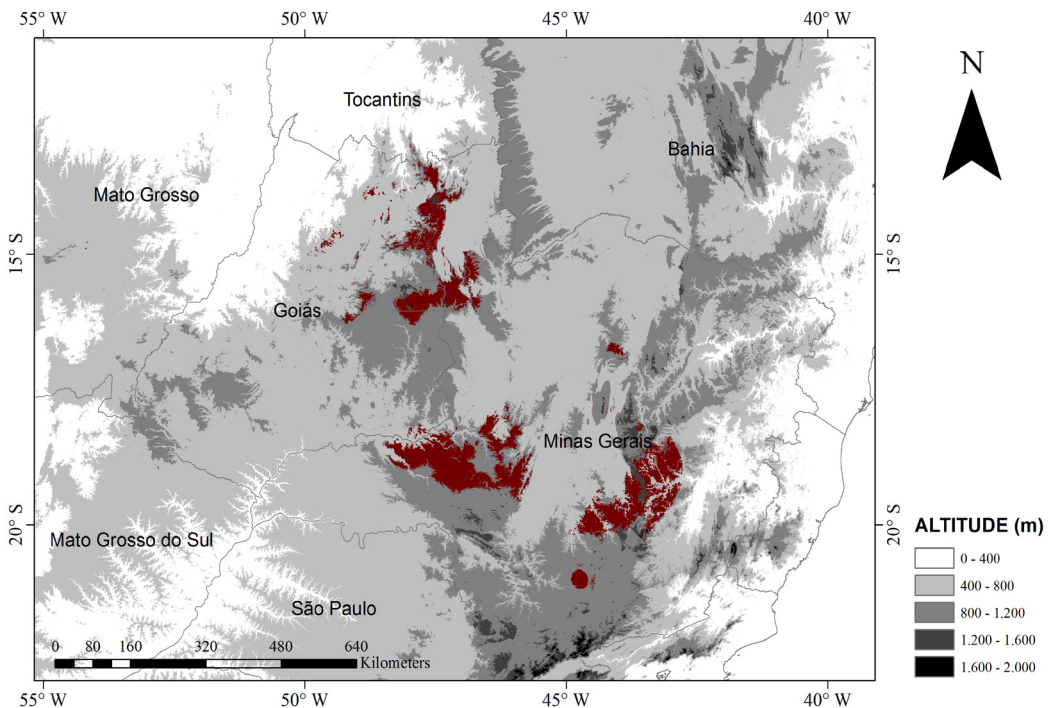


Figure 4. Overlap between Brazilian altitude and last range of the maximum training sensitivity plus specificity logistic threshold for model distribution of *Rhinella rubescens*.

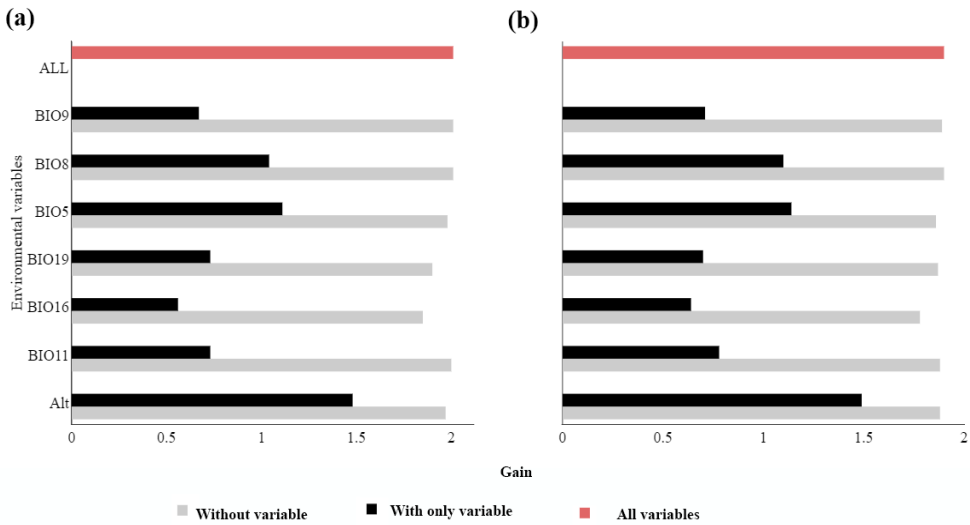


Figure 5. Variable contribution to training gain (a) and testing gain (b).

While BIO16 (coefficient= 0.001; $p < 0.01$) each 1 mm of rain the suitability increase in 0.001.

Discussion

The toad *Rhinella rubescens* is present in the States of Minas Gerais, Goiás, Distrito Federal (Brasília), north of São Paulo, south central and southeastern Bahia, central highlands of Rio de Janeiro, southeastern Mato Grosso and northeast of Mato Grosso do Sul. These were confirmed areas where the specimens were evaluated and properly identified. In particular, two doubts are present by the model (Maxent): (i) we cannot rule out the possibility of an error in the registry data of the specimen of State Rio de Janeiro, as this area is historically well documented. By considering this record, it means that *Rhinella rubescens* has not been collected or observed in State of Rio de Janeiro during the last 90 years. A second area indicated in the model, where low habitat suitability predicted (omission error), is the recent record in southern registration for State of Mato Grosso do Sul by Aoki *et al.* (2011). It is a confirmed record for *R. rubescens*, however the model not identified as high suitable habitat for the species. We think that the elevation and climate conditions of the registry area with respect to other localities did not possible recognition as an area suitability for its occurrence. Additionally, the model also predicts possible new locations that could harbor the presence of the species in the southeastern of

Tocantins in the limits with Goiás states, which would merit collect efforts in these locations to confirm those records.

The altitude, temperature and precipitation were the variables with high important for the species occurrence. These generated restrictions in the distribution are related to the patterns of the Biomes where the species occurs. Predominantly in the Cerrado Biome, but also in the Atlantic Forest where populations are submitted to the several elevation and weather changing conditions. Interesting, it was currently known that *Rhinella rubescens* had a strongly relation to temperature and rainfall, where it's breeding patterns is associated with dry months (Vasconcellos & Colli 2009). We found strong relationships with the temperature, mainly in the warmest month, wettest and colder quarter. The precipitation variables did not contributing in the same way for the model based in Jackknife index. But if it has an effect in the species suitability in the multiple linear regression. BIO19 (Precipitation of Coldest Quarter) and BIO16 (Precipitation of Wettest Quarter) are possibly influencing pattern activities throughout these periods (e.g. reproduction).

We found no authentic information associated in collections and literature for support the presence of this species in the states of Pará, Tocantins and Piauí. These states have been used in the IUCN assessments (Bastos *et al.* 2010). Most of the localities are within the

Cerrado biome, but there are also few collecting sites in high elevation areas within the Atlantic Forest and Caatinga biomes. This is evidenced by Roberto et al. (2013), who cannot find records for municipalities in the State of Piauí. These authors took in consideration the distribution assumed by Frost (2016) through IUCN evaluation. Based in this evidence, these states should be excluded of the distribution of *R. rubescens* until new information be provided.

In conclusion, the known distribution of *Rhinella rubescens* were only found in the states of Bahia, Minas Gerais, São Paulo, Goiás, Distrito Federal (Brasília), Mato Grosso do Sul, Mato Grosso and Rio de Janeiro. This result offers evidence for possible reassessment in *R. rubescens* IUCN category and the opportunity to evaluate potential effects of climate change in suitability areas of the species. The distribution model provided interesting data in the limits of Tocantins and Goiás states where this species can occur and requires to be confirmed. Regarding the mismatch on occurrence in the states of Pará and Piauí, we could infer that the presence of this species seems improbable, mainly because that habitat conditions (altitude) limit the distribution of this species.

Acknowledgments. We thanks to C.F.B. Haddad (CFBH), G.R. Colli (CHUNB), D. Rossa-Feres (DZSJRP), J. Rosado (MCZ), J.P. Pombal Jr. (MNRJ), T. Grant (MZUSP), R.W. McDiarmid (USNM), and L.F. Toledo (ZUEC) gave access, loaned or sent information regarding specimens in their care. J.P. Pombal Jr. and C. Roman-Palacios for help in the pre-peer review. AVZ and NMM acknowledge the Coordenação de Aperfeiçoamento de Pessoal de Nível Superior (CAPES) and Conselho Nacional de Desenvolvimento Científico e Tecnológico (CNPq), respectively for the fellowships. This work was support by Programa de Apoio a Núcleos de Excelência (PRONEX) in the project “Biomonitoramento, Modelagem e Marcadores Genéticos no Cerrado” (Processo #201210767000812).

References

- Aoki, C., Landgraf, P., Oda, F.H., Gamarra, R.M. (2011): Geographic distribution: *Rhinella rubescens*. Herpetological Review, **42**: 108.
- Araújo, C.O., Condez T.H., Haddad, C.F.B. (2007): Amphibia, Anura, *Barycholos ternetzi*, *Chaunus rubescens*, and *Scinax canastrensis*: Distribution extension, new state record. Check List, **3** (2): 153 – 157.
- Araújo, O.G.S., Toledo, L.F., Garcia, P.C.A., Haddad, C.F.B. (2009): The amphibians of São Paulo State. Biota Neotropica, **9** (4): 197 – 209.
- Barve, N. (2008): Tool for Partial-ROC, version 1.0. Biodiversity Institute, Lawrence, KS.
- Bastos, R., Pavan, D., Cabral, P., Silvano, D. (2010): *Rhinella rubescens*. The IUCN Red List of Threatened Species. Version 2015.2. <www.iucnredlist.org>. Downloaded on 25 August 2015.
- Brandão, R. A., Maciel, N.M., Sebben, A. (2007): A new species of *Chaunus* from central Brazil (Anura: Bufonidae). Journal of Herpetology, **41**: 309 – 316.
- Camardelli, M., Napoli, M.F. (2012): Amphibian conservation in the Caatinga biome and semiarid region of Brazil. Herpetologica, **68** (1): 31 – 47.
- Dormann, C.F., Elith, J., Bacher, S., Buchmann, C., Carl, G., et al. (2013): Collinearity: a review of methods to deal with it and a simulation study evaluating their performance. Ecology, **36**: 27 – 46.
- ESRI (2011): ArcGIS Desktop: Release 10. Redlands, CA: Environmental Systems Research Institute.
- Fitzinger, L. J. F. J. (1826): Neue Classification der Reptilien nach ihren Natürlichen Verwandtschaften nebst einer Verwandtschafts-Tafel und einem Verzeichnisse der Reptilien-Sammlung des K. K. Zoologisch Museum's zu Wien. Wien: J. G. Heubner.
- Frost, D.R. (2016): Amphibian Species of the World: an Online Reference. Version 6.0 (07/07/2016). Electronic Database accessible at <http://research.amnh.org/herpetology/amphibia/index.html>. American Museum of Natural History, New York, USA.
- Harrel, F.E. (2016): Package ‘Hmisc’, version 3.17-3, Harrell Miscellaneous. Cran.r-project. <https://cran.r-project.org/web/packages/Hmisc/Hmisc.pdf>. Downloaded on 05 April 2016
- Hensel, R. (1867): Beiträge zur Kenntnis der Wirbelthiere Südbrasilens. Archiv für Naturgeschichte (Berlin), **33**: 120–162.
- Jarvis, A., Reuter, H.I., Nelson, A., Guevara, E. (2008): Hole-filled SRTM for the globe Version 4, available from the CGIAR-CSI SRTM 90m Database (<http://srtm.csi.cgiar.org>).
- Linnaeus, C. (1758): Systema Naturae per Regna Tria Naturae, Secundum Classes, Ordines, Genera, Species, cum Characteribus, Differentiis, Synonymis, Locis. 10th Edition. Volume 1. Stockholm, Sweden: L. Salvii.
- Leite, F. S. F., Juncá, F. A., Eterovick, P. C. (2008): Status do conhecimento, endemismo e conservação de anfíbios anuros da Cadeia do Espinhaço, Brasil. Megadiversidade, **4**: 182 – 200.
- Lutz, A. (1925): Batraciens du Brésil. Comptes Rendus et Mémoires Hebdomadaires des Séances de la Société de Biologie et des ses Filiales (Paris), **93** (2): 211 – 214.
- Maciel, N. M., Brandão, R. A., Campos, L. A., Sebben, A. (2007): A large new species of *Rhinella* (Anura: Bufonidae) from Cerrado of Brazil. Zootaxa, **1627**: 23 – 39.
- Magalhães, F.M., Laranjeiras, D.O., Costa, T.B, Juncá, F.A., Mesquita, D.O, Röhr, D.L., da Silva, W.P., Vieira, G.H.C., Garda, A.A. (2015): Herpetofauna of protected areas in the Caatinga IV: Chapada Diamantina National Park, Bahia, Brazil. Herpetology Notes, **8**: 243 – 261.
- Maneyro, R., Arrieta, D., de Sá, R.O. (2004): A new toad (Anura: Bufonidae) from Uruguay. Journal of Herpetology, **38**: 161 – 165.
- Medeiros-Magalhães, F., Laranjeiras, D.O., Costa, T.B., Juncá, F.A., Mesquita, D.O., Röhr, D.L., Silva, W.P., Calazans-Vieira,

- G.H., Garda, A.A. (2015): Herpetofauna of protected areas in the Caatinga IV: Chapada Diamantina National Park, Bahia, Brazil. *Herpetology Notes*, **8**: 243 – 261.
- Peterson, A.T., Papes, M., Soberón, J. (2008): Rethinking receiver operating characteristic analysis applications in ecological niche modeling. *Ecological Modelling*, **213** (1): p. 63 – 72.
- Phillips, S.J., Dudík, M., Schapire, R.E. (2004): A maximum entropy approach to species distribution modeling. *Proceedings of the Twenty-First International Conference on Machine Learning*: 655 – 662.
- Pramuk, J. B. (2006): Phylogeny of South American Bufo (Anura: Bufonidae) inferred from combined evidence. *Zoological Journal of the Linnean Society*, **146**: 407 – 452.
- R Development Core Team (2016): R: A Language and Environment for Statistical Computing. R Foundation for Statistical Computing, Vienna, Austria.
- Roberto, I.G., Ribeiro, S.C., Loebmann, D. (2013): Amphibians of the state of Piauí, Northeastern Brazil: a preliminary assessment. *Biota Neotropica*, **13**(1): 322 – 330.
- São-Pedro, V.A., Feio, R.N. (2011): Anuran species composition from Serra do Ouro Branco, southernmost Espinhaço Mountain Range, state of Minas Gerais, Brazil. *Check List*, **7** (5): 671 – 680.
- Spix, J. B. (1824): *Animalia nova sive Species novae Testudinum et Ranarum quas in itinere per Brasiliam annis MDCCCXVII–MDCCCXX jussu et auspiciis Maximiliani Josephi I. Bavariae Regis*. München: F. S. Hübschmann.
- Stevaux, M. N. (2002): A new species of *Bufo* Laurenti (Anura, Bufonidae) from northeastern Brazil. *Revista Brasileira de Zoologia*, **19**: 235–242.
- Tschudi, J. J. (1845): Reptilium conspectus quae in Republica Peruana reperiuntur et pleraquae observata vel collecta sunt in itinere a Dr. J. J. de Tschudi. *Archiv für Naturgeschichte*, **11**: 150 – 170.
- Valdujo, P.H., Silvano, D.L., Colli, G., Martins, M. (2012): Anuran species composition and distribution patterns in Brazilian Cerrado, a Neotropical hotspot. *South American Journal of Herpetology*, **7**(2): 63 – 78.
- Vasconcellos, M.M., Colli, G.R. (2009): Factors Affecting the Population Dynamics of Two Toads (Anura: Bufonidae) in a Seasonal Neotropical Savanna. *Copeia*, **2009**(2): 266 – 276.
- Warren, D.L., Glor, R.E., Turelli, M.N. (2008): Environmental niche equivalency versus conservatism: quantitative approaches to niche evolution. *Evolution*, **62**: 2868 – 2883.
- Wiegmann, A.F.A. (1833): *Herpetologischen Beyträge. I. Ueber die mexicanischen Kröten nebst bemerkungen über ihren verwandte Arten anderer Weltgegenden*. *Isis von Oken* **26**: 651–662.
- Werner, F. (1894): *Herpetologische Nova*. *Zoologischer Anzeiger*, **17**: 410 – 415.

Appendices I. Specimens examined

BRAZIL: GOIÁS: CHUNB 67539 (Cavalcante: 47° 27' 25.2'' W, 13° 47' 52.8'' S); CHUNB 69135 (Catalão: 47° 56' 38.4'' W, 18° 9' 57.6'' S); CFBH 7696 (Cocalzinho de Goiás: 48° 46' 33.6'' W, 15° 47' 38.4'' S); CHUNB 743, 58765 (Alto Paraíso de Goiás: 47° 30' 35.999'' W, 14° 7' 58.8'' S); CHUNB 6977 (Planaltina: 47° 36' 50.399'' W, 15° 27' 10.8'' S); CHUNB 23846, ZUFG 6181 (Pirenópolis: 48° 57' 32.4'' W, 15° 51' 7.2'' S); CHUNB 38728, 38733-34 (São João d'Aliança: 47° 31' 29.999'' W, 14° 42' 21.6'' S); CHUNB 43400 (Pires do Rio: 48° 16' 48'' W, 17° 18' 3.6'' S); CHUNB 43421 (Luziânia: 47° 57' 0'' W, 16° 15' 10.8'' S); CHUNB 65274-75, 65277-78, 65280-81 (Novo Gama: 48° 2' 31.2'' W, 16° 3' 32.4'' S); MNRJ 56498, DZSJRP 593, 663-65 (Anápolis: 48° 57' 10.8'' W, 16° 19' 37.2'' S); MCZ 367-8 (Syntypes, Goiás Velho: 50° 8' 42'' W, 15° 56' 31.2'' S); CHUNB 44736 (Três Ranchos: 47° 46' 44.4'' W, 18° 21' 36'' S); ZUFG 4008-9, 6181 (Silvânia: 48° 38' 52.8'' W, 16° 38' 34.8'' S). **BRASILIA (Distrito Federal):** CFBH 1910, 2587-88, CHUNB 739, 744, 12915, 12950, 14957-60, 23619, 29387, 32239, 32249-50, 32650, 34008, 39969-70, 43290 (Distrito Federal: 47° 51' 50.04'' W, 15° 48' 0'' S); CHUNB 6976, 6978, 125810, 12915, 12952 (Planaltina: 47° 38' 20.4'' W, 15° 28' 58.8'' S); CHUNB 30839, 33793-94 (Aeroporto: 47° 55' 47.999'' W, 15° 46' 48'' S); CHUNB 43679 (Jardim Botânico: 47° 55' 47.999'' W, 15° 46' 48'' S). **MATO GROSSO:** MZUSP 66752-53 (Fazenda Coqueiro, Alto Araguaia: 53° 13' 8.4'' W, 17° 18' 57.6'' S); MZUSP 152084 (Fazenda Garrote, Alto Taquari: 53° 16' 55.199'' W, 17° 49' 33.6'' S). **MINAS GERAIS:** CFBH 283, 769, DZSJRP 5425, MNRJ 38472, 38714-5, 39024, ZUEC 2204-5 (Santana do Riacho: 43° 42' 50.399'' W, 19° 10' 8.4'' S); CHUNB 57467-68, CFBH 4451, 5834-37, MNRJ 51795-96, 87824, MZUSP 132393-96, 132425-32, 134461-66, ZUEC 8396 (Poços de Caldas: 46° 33' 39.599'' W, 21° 47' 16.8'' S); CHUNB 24733, MZUSP 28433 (Unai: 46° 54' 21.599'' W, 16° 21' 28.8'' S); CHUNB 24733, 25172, 25175-82, 25385-6, 25392, 25397-400, 25466-7, 25477, 25480, 26468-9, 26477, 26480 (Paracatú: 46° 52' 29.999'' W, 17° 13' 19.2'' S); CHUNB 44342, 44344 (Fazenda Jatobá, Buritizinho: 44° 57' 43.2'' W, 17° 21' 3.6'' S); DZSJRP 138, MNRJ AL1409, 89290, MZUSP 21553 (Lagoa Santa: 43° 53' 24'' W, 19° 37' 37.2'' S); DZSJRP 5363, 5366, 5425, 5507-09, MZUSP 36947-54, 37543, 134632 (Santa Barbara: 43° 24' 53.999'' W, 19° 57' 32.4'' S); MNRJ 623 (Nova Serrana: 44° 59' 2.4'' W, 19° 52' 33.6'' S); MNRJ 993, 39045-6, MZUSP 15875, USNM 70606, 97236-7*Syntypes (Belo Horizonte: 43° 57' 21.6'' W, 19° 49' 1.2'' S); MNRJ 2414, AL1409 (Pirapora: 44° 56' 31.2'' W, 17° 20' 42'' S); MNRJ 22470, 34766 (Presidente Olegário: 46° 25' 4.799'' W, 18° 25' 4.8'' S); MNRJ 41804-07, 51797 (Lima Duarte: 43° 47' 34.799'' W, 21° 50' 34.8'' S); MNRJ 49470 (Capitólio: 46° 2' 59.999'' W, 20° 38' 60'' S); CHUNB 57615-19, MNRJ 51339, MZUSP 104318, 126187, ZUEC 1640, 1885, 1947, 2506, 2573, 2815-7, 3043, 3358, 3362-3, 3537-41 (Serra do Cipó, Jaboticatubas: 43° 44' 41.999'' W, 19° 30' 50.4'' S); MNRJ 77544-47,

MZUSP 28422-3 (Serra do Caraça, Catas Altas: 43° 24' 28.8'' W, 20° 4' 30'' S); MZUSP 28422-28, 28430 (Congonhas do Campo: 43° 51' 28.799'' W, 20° 30' 0'' S); MZUSP 132992-95 (São Roque de Minas: 46° 21' 57.599'' W, 20° 14' 42'' S); USNM 218063 (Chapéu do Sol: 43° 38' 59.999'' W, 19° 0' 0'' S); ZUEC 1133 (São Tomé das Letras: 44° 59' 5.999'' W, 21° 43' 19.2'' S); ZUEC 4333 (Alpinópolis: 46° 23' 16.799'' W, 20° 51' 50.4'' S); ZUEC 4353 (Vargem Bonita: 46° 21' 57.599'' W, 20° 19' 37.2'' S); ZUFG 4780 (Alto Palácio: 43° 31' 1.2'' W, 19° 15' 39.6'' S); ZUFG 6438 (Diamantina: 43° 45' 25.199'' W, 17° 55' 40.8'' S); ZUFG 8205-11, 8214-7 (Uberlândia: 48° 16' 29.999'' W, 18° 54' 54'' S); CHUNB 71611, 73274-76, 73291 (Formoso: 46° 14' 13.2'' W, 14° 56' 42'' S); MNRJ 3844-45, 57651 (João Pinheiro: 46° 10' 26.399'' W, 17° 44' 24'' S); MNRJ 59172 (Barbacena: 43° 46' 12'' W, 21° 13' 15.6'' S); MNRJ 60312 (Paula Cândido: 42° 58' 47.999'' W, 20° 51' 0'' S); MNRJ 60806 (Conselheiro Lafaiete: 43° 46' 29.999'' W, 20° 39' 57.6'' S); MNRJ 88983-84 (Grão Mogol: 42° 53' 24'' S, 16° 33' 32.4'' S); CHUNB 49416 (Perdizes: 47° 12' 50.399'' W, 19° 20' 49.2'' S). **RIO DE JANEIRO:** USNM 97656 (Teresópolis: 42° 58' 58.799'' W, 22° 25' 58.8'' S). **SÃO PAULO:** MNRJ 715 (Franca: 47° 24' 3.6'' W, 20° 32' 20.4'' S).