Original Article

Favourable areas for co-occurrence of parapatric species: niche conservatism and niche divergence in Iberian tree frogs and midwife toads

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ABSTRACT

Aim: The study of areas of sympatry of species with predominantly parapatric distributions can provide valuable insights into their evolutionary history and the factors shaping patterns of species co-occurrence. This information is key in biogeography, evolutionary biology and conservation planning. In this study we analyse the distributions of two pairs of partially co-occurring congeneric amphibian species: tree frogs (*Hyla molleri* and *H. meridionalis*) and midwife toads (*Alytes obstetricans* and *A. cisternasii*).

Location: Iberian Peninsula (SW Europe).

Methods: We obtained distribution data from the herpetological atlases of Portugal and Spain, consisting of presences and absences on UTM 10×10 km grid cells. We built an environmental favourability model for each species, using 24 potential predictor variables representative of physiography, climate and human activity. Variables were selected for each model using both information and significance criteria. Models were evaluated using both calibration and discrimination measures. Models were then combined using fuzzy intersection, and compared using correlation analysis (accounting for spatial autocorrelation), niche comparison metrics, and fuzzy similarity indices.

Results: We found significant dissimilarity between the favourability patterns for *A. obstetricans* and *A. cisternasii*, indicating environmental segregation of these two midwife toad species. In tree frogs, we found significant similarity between favourability for *H. meridionalis* and for its co-occurrence with *H. molleri* – i.e., sympatry occurs mainly in areas that are favourable for *H. meridionalis*.

Main conclusions: These results provide clues to understand the evolutionary history of these four species, including the evolution of reproductive isolation,

and suggest that conservation efforts for tree frogs may be focused on the areas that are favourable for both species, whereas midwife toads will require specific measures tailored for each species.

Keywords: amphibians; distributional overlap; environmental favourability; evolutionary biogeography; fuzzy logic; niche evolution; parapatry; species distribution modelling; sympatry.

INTRODUCTION

Many species share common distribution ranges, presenting considerable geographic areas of co-occurrence. Studying spatial and environmental patterns of overlap can provide valuable insights into the factors that determine the co-occurrence of species and shed light on aspects of their evolutionary history (Coyne & Orr, 2004). For instance, the characterization of spatial patterns of environmental favourability in species with partially overlapping ranges can provide clues about the geographical and ecological context of the speciation process (Fisher-Reid *et al.*, 2013; Wooten *et al.*, 2013). While ecological niche differentiation is not necessarily a pre-requisite for speciation, niche divergence can act as a pre-zygotic isolating mechanism in spatially continuous populations, reinforcing or consolidating reproductive isolation in nascent species (Nosil *et al.*, 2009). On the other hand, a lack of differentiation may result from phylogenetic niche conservatism (i.e., a tendency of species to retain their ancestral traits), which has been shown to be an important feature in allopatric speciation across a variety of taxa (Wiens *et al.*, 2010).

Based on this reasoning, we hypothesize that comparative analyses of the environmental favourability patterns of parapatric species can help to distinguish the dominant ecological/evolutionary forces shaping their distributions, in terms of independence, conservatism or divergence of niche (Fig. 1). We thus model and compare the distributions of two pairs of parapatric amphibian species in the Iberian Peninsula: tree frogs (*Hyla molleri* and *H. meridionalis*), with 11% of the occurrence area (based on UTM 10 x 10 km cells) shared by both species; and midwife toads (*Alytes obstetricans* and *A. cisternasii*), with an overlap of 4%

in their occurrence areas in this region (Loureiro *et al.*, 2010; MAGRAMA, 2015).

Species in each of these pairs differ in aspects of their natural history and ecological preferences at the local scale. For instance, in *Alytes* there seems to be little overlap in the breeding seasons of the two species (Malkmus, 2004), which limits the chances for hybridization. Also, *A. cisternasii* usually breeds in more temporary habitats, compared to the generally permanent water bodies selected by *A. obstetricans*. With respect to the terrestrial habitat, *A. cisternasii* usually occurs in open areas and avoids limestone or littoral dunes, whereas *A. obstetricans* can be found in a wider range of habitats, including dense forests, and in many types of geological substrates, including limestone or gypsum terrains (García-París *et al.*, 2004; Malkmus, 2004). There are also differences in their preferred altitudes (*A. cisternasii* populations are mostly found below 500 m, whereas *A. obstetricans* prefers altitudes between 400 and 1000 m and can reach much higher, up to >2000 m). Finally, *A. cisternasii* is present in meso- or thermo-Mediterranean areas, with annual precipitation <900 mm, whereas *A. obstetricans* is present in a wider variety of bioclimatic regions, but generally where annual precipitation is >900 mm (Malkmus, 2004).

Conversely, the two *Hyla* species show fewer ecological differences. For instance, their terrestrial habitats are very similar (although *H. meridionalis* has a broader range of habitat types), and they seem to have synchronized their breeding seasons, thus probably increasing the chances for hybridization. Ecological differences among *Hyla* include their preferred elevation ranges (wider in *H. molleri*, but rarely >500 m in *H. meridionalis*) and precipitation levels

(high annual precipitation in *H. molleri*, but <1000 mm in *H. meridionalis*) (Malkmus, 2004).

These local-scale ecological differences among *Hyla* and *Alytes* species were also identified in previous attempts to model their distributions (Arntzen, 2006) and suggest the existence of niche segregation. The two species pairs have similarly old divergence times (Miocene; see Smith *et al.* 2005; Maia-Carvalho *et al.* 2014), but the temporal scale of their overlap is markedly different: *H. meridionalis* is inferred to have entered the Iberian Peninsula in recent times, perhaps even in historical times, via human-mediated introduction (Recuero *et al.*, 2007), whereas *A. obstetricans* and *A. cisternasii* have both evolved *in situ* and coexisted for millions of years (Martínez-Solano *et al.*, 2004; Maia-Carvalho *et al.*, 2014).

In amphibians, vicariance is the most common mode of speciation (Vences & Wake, 2007), and therefore the study of areas of secondary contact is key to assess patterns of reproductive isolation and hybridization. Previous studies have shown that our target species pairs are reproductively isolated, with no viable and fertile hybrids reported to date (Oliveira *et al.*, 1991; Barbadillo & Lapeña, 2003). However, little is known about the intrinsic and extrinsic factors involved in the maintenance of species boundaries in areas where their geographical distributions overlap. Here we address the role of extrinsic (environmental) factors in patterns of spatial co-occurrence between Iberian *Alytes* and *Hyla* species, and discuss the implications of our findings in an evolutionary context.

METHODS

Study area and data

The study area was the Iberian Peninsula, the south-western tip of Europe (Figure 1). With a total area of nearly 600,000 km², it is a physiographically heterogeneous region, comprising the mainland territories of Portugal and Spain, and linked to the European continent by a narrow and mountainous isthmus. It thus constitutes a discrete biogeographical unit appropriate for modelling species distributions (Real *et al.*, 2009; Barbosa *et al.*, 2010).

The species distribution data, consisting of presences and absences on 10×10 km grid cells with a Universal Transverse Mercator (UTM) projection (Fig. 2), were compiled from Loureiro *et al.* (2010) for Portugal and from MAGRAMA (2015; including Pleguezuelos *et al.*, 2002) for Spain. Maps of the 10×10 km grids and the boundary of the study area were downloaded from the EDIT Geoplatform (Sastre *et al.*, 2009). We used QGIS 2.4 (QGIS Development Team, 2014) for spatial data processing and R 2.11 (R Core Team, 2014) for data management and analysis.

We used 24 potential predictor variables representative of Iberian physiography, climate and human activity (Table 1). Details on how these variables were digitised and interpolated were described elsewhere (Barbosa *et al.*, 2003, 2009). The values of solar radiation were corrected following (Barbosa *et al.*, 2011). Variables were then scaled by subtracting their mean values and dividing by their standard deviations, so that their coefficients (i.e., their effects on the models) could be directly compared.

To avoid a spurious effect of surface area on the probability of each species being present, only the 5464 UTM cells whose area was not altered by

intersections with the coastline, study area borders or the unions between UTM zones were used for model building (Barbosa *et al.*, 2009; Real *et al.*, 2009). The models were then applied to the 6180 Iberian grid cells for which the variable values were available.

Model building

We built, for each species, a generalized linear model with binomial error distribution (adequate for the presence-absence response) and the favourability function link, which replaces 1 in the *logit* link (used in logistic regression) with the number of presences divided by the number of absences. It thus assesses local variations in presence probability with respect to the overall species prevalence (Real *et al.*, 2006; Acevedo & Real, 2012). This makes model predictions independent of each species' presence to absence ratio in the study area, enabling direct model comparison and combination when two or more taxa are analysed (Real *et al.*, 2009; Estrada *et al.*, 2011; Barbosa & Real, 2012).

The selection of variables for each model was made in four phases, all included in the *multGLM* function of the *fuzzySim* R package (Barbosa, 2015a). First, we pre-selected variables that had a significant bivariate relationship with the target species distribution, accounting for the false discovery rate by correcting the significance of variables under repeated testing (Benjamini & Hochberg, 1995; García, 2003). Then, to avoid multicollinearity and its effects on coefficient estimates (Legendre & Legendre, 2012), among each pair of selected variables that were visibly correlated (Pearson's $r > 0.8$) we excluded the one that had the weakest relationship with the response, considering both p -

value and Akaike's information criterion (AIC; Akaike, 1973). Using the remaining variables, we built multivariate models with a forward-backward stepwise selection, using AIC to include or exclude variables (Burnham & Anderson, 2002) as is implemented in R. Finally, if there were any variables with non-significant coefficients left in the model (chi-squared test, $p > 0.05$), they were eliminated step by step until all coefficients were significantly different from zero (Crawley, 2007).

We used the *fuzzyOverlay* function of the *fuzzySim* package to calculate a fuzzy intersection (Zadeh, 1965) between the model predictions for the species in each congeneric pair. This intersection represents favourability for the co-occurrence of both species, and has been used previously in comparative distribution modelling (Barbosa & Real, 2010, 2012).

Model analysis and evaluation

We assessed the two different components of performance in each model: discrimination capacity (i.e., the ability of the model to distinguish presences from absences); and calibration or reliability (i.e., the magnitude of the overall deviations of predictions from observed values, and the relationship between predicted probabilities and observed occurrence frequencies). Although the latter component is often neglected, both are equally important in the evaluation of species distribution models (Pearce & Ferrier, 2000; Wintle *et al.*, 2005; Jiménez-Valverde *et al.*, 2013).

The discrimination capacity of the models was measured with several widely used indices, including e.g. the area under the receiver operating characteristic curve (AUC) and the true skill statistic (TSS). The AUC

summarises the discrimination performance of a model over the whole range of prediction thresholds. TSS weights sensitivity (correct classification of presences) and specificity (correct classification of absences) at a given threshold; we chose 0.5 as the threshold for discrimination measures. With favourability models, this equates to using prevalence as a threshold in logistic regression (Real *et al.*, 2006; Acevedo & Real, 2012).

Model reliability was measured with the Hosmer-Lemeshow goodness-of-fit test (Hosmer & Lemeshow, 1980), as well as with D^2 (proportion of deviance explained by each model) and the pseudo R^2 measures of Cox-Snell, McFadden, Nagelkerke and Tjur. All metrics are implemented in a unified framework in the *modEvA* R package (Barbosa *et al.*, 2013), including functions *AUC*, *threshMeasures*, *HLfit*, *Dsquared*, and *RsqGLM*.

Model (environmental niche) comparison

We first analysed the correlations between model predictions, checking for significant similarities or dissimilarities between environmental favourability values for the analysed species and for the co-occurrence of congeneric species. We calculated the significance of these correlations after accounting for spatial autocorrelation with the *modified.ttest* function of the *SpatialPack* R package (Osorio *et al.*, 2012), which assesses the correlation between two spatial processes, estimating an effective sample size that takes into account their spatial association (Clifford *et al.*, 1989).

Pairs of models whose predictions were correlated (either positively or negatively) after accounting for spatial autocorrelation were then analysed for niche overlap using Schoener's *D* and Warren *et al.*'s indices (Warren *et al.*,

2008), and for overall similarity based on fuzzy versions (Barbosa, 2015a) of classic binary similarity indices, accounting for shared presence areas (Jaccard, 1901) and for both shared presence and shared absence areas (Baroni-Urbani & Buser, 1976). The latter two indices have associated tables of significant values (Baroni-Urbani & Buser, 1976; Real & Vargas, 1996; Real, 1999). These two indices are traditionally based on categorical presence-absence data, but the *fuzSim* function of the *fuzzySim* package adapts them to work directly with continuous data such as those provided by favourability model predictions, without any loss of quantitative information (Barbosa, 2015a).

RESULTS

Model performance and variables

The four models achieved good overall prediction accuracy, with e.g. AUC values generally above 0.8, which is considered “good”, or 0.9, which is considered “excellent” (Swets, 1988); and McFadden’s pseudo- R^2 values generally above 0.2, which is considered “excellent fit” (McFadden, 1978) (Fig. 3). The variables included in each model and their coefficient estimates are shown in Table 1. For more detailed statistics on model parameters, see Appendix S1 in Supporting Information.

Favourability for midwife toads was higher in grid cells with higher values of maximum precipitation within a day (*MP24*) for both species. Relative maximum precipitation (*RMP*) and the annual air humidity range (*HRan*) had opposite effects on the favourability for these two species. Favourability for *A. obstetricans* was also associated with higher values of relative maximum precipitation (*RMP*) and lower values of potential evapotranspiration (*PET*),

January temperature (*TJan*) and annual variation in air humidity (*HRan*), among other variables (Table 1). Favourability of *A. cisternasii* was associated with lower values of annual precipitation and of how concentrated it is in time (*Prec* and *RMP*), and with a smaller annual number of frost days (*DFro*), in this multivariate context (Table 1; see also Appendix S2).

Among the tree frogs, the annual number of snow days (*DSno*), January air humidity (*HJan*) and annual insolation (*Inso*) had opposite effects on environmental favourability for the two, while both species were associated with higher actual evapotranspiration (*PET*) and with lower maximum daily precipitation (*MP24*). Favourability for *H. molleri* was also higher in grid cells with more time-concentrated precipitation (*RMP*) and lower values of slope (*Slop*). Favourability of *H. meridionalis* was associated with lower altitudes (*Alti*), among other variables in the multivariate context of Table 1 (see also Appendix S1).

Relationships between models

After accounting for spatial autocorrelation, there was a significant negative correlation between the environmental favourability patterns of the analysed midwife toad species (Table 2), indicating environmental segregation between the two (see also Fig. 4). These results were supported by small values of niche overlap and by significantly low similarity (i.e., less than expected by chance) between the two environmental favourability models (Table 2).

In contrast, although there was no correlation between the models of the two *Hyla* species (when accounting for spatial autocorrelation), we found a strong positive correlation between environmental favourability for *H.*

meridionalis and for its co-occurrence with *H. molleri*. This was corroborated by large values of niche overlap and significantly high similarity between these two models (Table 2). There is thus a high environmental potential for sympatry among these two species across the south-western quarter of the Iberian Peninsula, i.e., within the range of *H. meridionalis* (Fig. 4).

DISCUSSION

Differences between observed and potential occurrence

Species distribution models can produce valuable information about the variables that shape current species ranges. When applied to pairs of partially co-occurring (parapatric) species, such models can reveal aspects of their evolutionary histories and interactions (e.g., Barbosa & Real, 2012; Pollock *et al.*, 2014), which may be difficult to infer based on other lines of evidence such as molecular data. In this sense, our models captured the present distribution of the four studied species, while showing additional favourable but currently unoccupied areas. This information complements previous and ongoing studies about the evolutionary history of these species (Recuero *et al.*, 2007; Gonçalves *et al.*, 2009, 2015; I. Martínez-Solano, unpublished data), although our methodological framework may be applied to other *taxa* as well.

In midwife toads, the model for *A. obstetricans* indicated the existence of favourable conditions south of its current distribution range, in the Betic mountains of SE Iberia (Fig. 4). This area has long been occupied by a possible competitor, the morphologically similar, small-range endemic Betic midwife toad *A. dickhilleni* (Pleguezuelos *et al.* 2002), which was not included in this study because it does not share noticeable co-occurrence areas with the other *Alytes*

species. For *A. cisternasii*, we also found favourable conditions outside of its current distribution range in the Iberian south, center and north-east, where the current absence of this species may reflect dispersal limitations. Favourable areas in the south are separated from this species' current range by the Guadalquivir river basin (S Spain), which has acted as a major biogeographic barrier for several amphibian taxa (García-París *et al.*, 1998; García-París & Jockusch, 1999; Carranza & Arnold, 2004; but see Real *et al.*, 2005). On the other hand, genetic data show the existence of several independent glacial refugia for this species in western Portugal during the Pleistocene, from which the present range was colonized through demographic expansions to the north and east, as revealed by a progressive loss of genetic variation along these geographic axes (Gonçalves *et al.*, 2009).

This interpretation probably applies also to *H. molleri*, whose favourable but unoccupied regions in the Iberian south and east probably represent areas to which this species has not yet been able to expand from its western refugia (I. Martínez-Solano, unpublished data). The presence of the congeneric *H. meridionalis* in these areas may represent an additional obstacle to its potential expansion, through inter-specific competition. The two Iberian nuclei of *H. meridionalis*, in the south-west and north-east (Fig. 2), proceed from two different genetic sources and have probably reached Iberia in different historical times as well (Recuero *et al.*, 2007). In this case, the discrepancies between model predictions and the actual distribution suggest a great expansion potential for the north-eastern nucleus, along the Iberian Mediterranean coast and inland along the Ebro river basin (NE Spain). In central Iberia, however, the

Sistema Central mountains seem to present a major dispersal barrier for this species, possibly limiting its expansion (e.g., Sillero, 2009).

Contrasting co-occurrence patterns in *Alytes* and *Hyla*

Apart from the historical insights obtained from individual species models, the fuzzy intersections of these models (revealing favourability for co-occurrence; Barbosa & Real, 2010, 2012) among each species pair revealed contrasting results that are informative about the mechanisms maintaining species borders in strict sympatry. In midwife toads, we found evidence of niche divergence between *A. obstetricans* and *A. cisternasii*, with very little observed overlap (Fig. 2), despite some environmental potential for co-occurrence as inferred from our models (Fig. 4). These species diverged at least 10 million years ago (Gonçalves *et al.*, 2015), and thus it is reasonable to assume that there have been opportunities for secondary contact and hybridization at different times in their evolutionary history, at least in some parts of their ranges. Thus, given that these two species are currently reproductively isolated (hybridization is extremely rare; Rosa, 1995), and considering their small distributional overlap (Fig. 2, Table 2), it follows that ecological factors may have played a major role in speciation in this group. In anurans, species differences in call characteristics act as a powerful pre-zygotic reproductive isolation mechanism, also leading to assortative mating and thus promoting species divergence (Lemmon, 2009). In *Alytes*, the calls of the different species are very simple and similar, although playback tests showed differences in female preference between sympatry and allopatry areas, which are consistent with reproductive character displacement under co-occurrence (Márquez & Bosch, 1997). Also, temporal overlap in their

breeding seasons is limited (Malkmus, 2004), restricting the opportunities for hybridization where the two species spatially overlap. External factors, here represented by the environmental variables in our models, can also contribute to promote or maintain species differentiation.

In tree frogs, in contrast, we found no evidence of niche divergence and a better match between observed and predicted overlap (Figs. 2 and 4), with most favourable areas for the co-occurrence of both species located within the environmental range of *H. meridionalis* – i.e., *H. meridionalis*, the most recent Iberian resident (Recuero *et al.*, 2007), is occupying a subset of the areas that were also favourable for *H. molleri*. In this case, assortative mating through audible differences in male calls between the two species acts as an efficient barrier to hybridization (Barbadillo & Lapeña, 2003). Additional post-zygotic barriers, such as reduced fitness or sterility of hybrids, have also been shown to play a role: histological examination of the gonads of hybrid individuals revealed the absence of viable spermatogonia (Barbadillo & Lapeña, 2003). Clearly, environmental factors like those included in our models play a minor role in shaping patterns of *Hyla* co-occurrence at a finer spatial scale. In fact, in areas where the two species co-occur (for instance, in southern Portugal and along the southern slopes of the Sistema Central mountains in central Spain) it is common to find breeding aggregations of both species in syntopy (e.g., Oliveira *et al.*, 1991; Barbadillo & Lapeña, 2003), with *H. meridionalis* often occurring in higher abundance (Ferreira, 2006).

We should remark that broad-scale species occurrence patterns are not necessarily matched by patterns on finer, more local scales (Segurado *et al.*, 2012). While the analysed species partially co-occur at broad spatial scales

(Fig. 2; Sillero *et al.*, 2014; IUCN, 2015), microhabitat differences may preclude strict sympatry on a local scale (see Introduction). Finer-scale distribution data from these co-occurrence areas can thus be used, in the future, to analyse these patterns in more detail.

Evolutionary and conservation implications

Our study showed how species distribution models can provide relevant information to help reconstruct the evolutionary history of species and of their interactions with other taxa, as well as identify the relative roles of intrinsic versus extrinsic factors in maintaining species boundaries in areas of secondary contact. In addition, from a conservation perspective, our results suggest that conservation efforts for tree frogs may be focused on the areas that are favourable for both species, regarding significant similarity between their favourability patterns; whereas efforts aimed at the conservation of midwife toads will require specific measures tailored separately for each species because of their environmental segregation.

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Supporting Information:

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

Appendix S1 Model parameters and associated statistics.

BIOSKETCHES:

Luís Reino (PhD) and **Mário Ferreira** (MSc) contributed equally to this work and are considered joint first authors. They are post- and pre-doctoral researchers at CIBIO/InBIO, working mainly on the distribution and ecology of birds and amphibians. The team have a common interest in the interactions between climate and biogeographical patterns. Author contributions: L.R. and P.S. conceived the idea; A.M.B., M.F. and L.R. gathered and analysed the data and produced the results; A.M.B., L.R., I.M.S. and M.F. wrote the paper; I.M.S. and C.X. provided ideas for additional analyses and improved writing and interpretation.

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Table 1. Variables used for modelling the distributions of midwife toads (*Alytes* spp.) and tree frogs (*Hyla* spp.) in the Iberian Peninsula, and the coefficient estimates of the variables finally included in each model. Variables were scaled and sorted in alphabetical order of their codes. The statistics associated to the coefficients are shown in Appendix S1.

Code	Variable	<i>Alytes obstetricans</i>	<i>Alytes cisternasii</i>	<i>Hyla molleri</i>	<i>Hyla meridionalis</i>
	(Intercept)	-0.903	-2.768	-1.717	-2.677
<i>AET</i>	Mean annual actual evapotranspiration (mm) (=minimum(<i>PET</i> , <i>Prec</i>))		0.496	0.365	0.878
<i>Alti</i>	Mean altitude (m) (U. S. Geological Survey, 1996)				-0.928
<i>DFro</i>	Mean annual number of frost days (min. temperature $\leq 0^{\circ}\text{C}$) (Font, 1983, 2000)		-0.696		
<i>DHi</i>	Distance to the nearest motorway (km) (I.G.N., 1999)		0.406		0.356
<i>DPre</i>	Mean annual number of days with precipitation $\geq 0,1$ mm (Font, 1983, 2000)			0.286	
<i>DSno</i>	Mean annual number of days with snow (Font, 1983, 2000)	-0.407	-0.508	0.286	-1.041
<i>HJan</i>	Mean relative air humidity in January at 07:00 hours (%) (Font, 1983, 2000)	-0.137	0.180	0.736	-0.517
<i>HJul</i>	Mean relative air humidity in July at 07:00 hours (%) (Font, 1983, 2000)			-0.496	
<i>HRan</i>	Annual relative air humidity range (%) ($= HJan-HJul $)	-0.689	0.587		
<i>Icon</i>	Index of continentality (Font, 2000)			-0.223	
<i>IHum</i>	Index of humidity (Font, 2000)			0.595	0.439
<i>Inso</i>	Mean annual insolation (hours/year) (Font, 1983, 2000)	-0.329	0.443	-0.319	1.866
<i>MP24</i>	Maximum precipitation in 24 hours (mm) (Font, 1983, 2000)	0.267	1.370	-0.313	-0.285
<i>PET</i>	Mean annual potential evapotranspiration (mm) (Font, 1983, 2000)	-0.883		-0.442	
<i>Prec</i>	Mean annual precipitation (mm) (Font, 1983, 2000)		-2.472		
<i>RMP</i>	Relative maximum precipitation ($=MP24/Prec$)	0.166	-2.235	0.502	0.326
<i>Slop</i>	Slope (degrees) (calculated from <i>Alti</i>)			-0.663	
<i>SRad</i>	Mean annual solar radiation (kwh/m ₂ /day) (Font, 1983, 2000)				
<i>Temp</i>	Mean annual temperature ($^{\circ}\text{C}$) (Font, 1983, 2000)				
<i>TJan</i>	Mean temperature in January ($^{\circ}\text{C}$) (Font, 1983, 2000)	-0.742	-0.281		
<i>TJul</i>	Mean temperature in July ($^{\circ}\text{C}$) (Font, 1983, 2000)				
<i>TRan</i>	Annual temperature range ($^{\circ}\text{C}$) ($=TJul-TJan$)				
<i>U100</i>	Distance to the nearest town over 100,000 inhabitants (km) (I.G.N., 1999)		0.257		
<i>U500</i>	Distance to the nearest town over 500,000 inhabitants (km) (I.G.N., 1999)	-0.619	-0.126	0.078	

Table 2. Coefficients of Pearson's correlation (varying between -1 and 1, with negative values indicating inverse relationships), Schoener's and Warren *et al.*'s indices of niche overlap, and fuzzy Jaccard's and Baroni-Urbani & Buser's indices of similarity between environmental favourability values (the latter four indices varying between 0 and 1; $n = 6180$). p_{SA} : significance after accounting for spatial autocorrelation; $p_{(-)}$, $p_{(+)}$: significance for the value being, respectively, smaller or larger than expected by chance (when available).

	<i>Alytes obstetricans</i>	<i>Hyla meridionalis</i>
	<i>Alytes cisternasii</i>	<i>Hyla co-occurrence</i>
Correlation	-0.726, $p_{SA} = 0.006$	0.628, $p_{SA} = 0.018$
Schoener <i>D</i>	0.379	0.700
Warren <i>I</i>	0.669	0.922
Jaccard	0.232, $p_{(-)} < 0.05$	0.513, $p_{(+)} < 0.001$
Baroni	0.430, $p_{(-)} < 0.01$	0.749, $p_{(+)} < 0.01$

FIGURE LEGENDS

Figure 1. Flowchart of the hypothesis-testing approach followed in this study.

Figure 2. Distribution of the studied species based on recorded occurrences on UTM 10 x 10 km cells of the Iberian Peninsula (after Loureiro *et al.*, 2010; MAGRAMA, 2015).

Figure 3. Discrimination and calibration performance of the environmental favourability models. Measures and plots were obtained with the *modEvA* R package. *AUC*: area under the curve. *CCR*: correct classification rate. *TSS* (true skill statistic) and Cohen's *kappa* were standardized (s) to vary between 0 and 1, so that they can be directly compared with the other measures (Barbosa, 2015b).

Figure 4. Environmental favourability values obtained for the studied species and their fuzzy intersection (favourability for co-occurrence) within each congeneric pair.

Figure 1.

Figure 2.

Figure 3.

Figure 4.