



# Amphibian distribution in a traditionally managed rural landscape of Eastern Europe: Probing the effect of landscape composition

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## ABSTRACT

A massive decline of biodiversity is caused by land-use changes. Efforts must therefore be made to better understand the factors that govern organismal distribution, especially for countries where traditional management is about to be intensified such as in Romania. We here document the spatial distribution of amphibians from a Romanian rural landscape where land-use is still largely traditional. We related the occurrence of nine amphibian species and species richness to measures of composition and configuration of the landscape surrounding 54 ponds at three spatial scales: circular areas of 400, 600 and 800 m radii. Busy roads most severely impacted single species and amphibian richness whereas landscape composition measures, such as cover of urban areas, agricultural areas, pastures, forests and wetlands were of little importance. We suggest that the relative unimportance of landscape compositional measures on amphibians is a consequence of the traditional management of these landscapes that keep the environmental conditions favorable for most species.

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## 1. Introduction

One of the major drivers of biodiversity decline in Europe is habitat degradation, fragmentation and loss (Billeter et al., 2008; Hendrickx et al., 2007; Schweiger et al., 2005). The main underlying causes are the intensification of agriculture (i.e. the shift from low-intensity production systems to intensively managed farmlands – Tscharrntke et al., 2005; Kleijn et al., 2009) and the development of infrastructure (i.e. built-up areas due to urbanization and industrialization and the transport network – Lövenhaft et al., 2004). The impact of human development on biodiversity is, however, not uniform across the continent. Countries from Western Europe are more impacted than those from Central and Eastern Europe (CEE) (Pullin et al., 2009). Indeed, biodiversity in CEE landscapes is still high, yet largely unexplored. Calls are out for the integration of conservation science and conservation policy in order to minimize the negative effects of on-going socio-economic developments on protected and non-protected areas. This goal is

to be reached by sound management plans and environmental impact assessments (Pullin et al., 2009; Sutherland et al., 2009). As it stands, the integration of conservation science and policy making is hampered by the lack of data on biodiversity, and it appears that protected areas have been established more quickly than our capacity to manage them has grown (Sutherland et al., 2009). For example, the basic information required to gain protective status (such as inventories of species and habitats) will mostly be insufficient for long-term management planning, therewith jeopardizing local biodiversity on the long-term. One potential avenue for solving this problem is to identify those spatial elements of habitats and landscapes that directly influence the distribution of target species.

The recent joining of the European Union by many CEE countries has triggered additional conflicts between biodiversity conservation and human activities, with often important political, economic, and environmental consequences (Young et al., 2007). For instance, the rich biodiversity of Romania is threatened by some phases of the integration process, most of all by the Common Agricultural Policy. CEE countries still hold a wealth of semi-natural and natural habitats created and maintained by low intensity traditional farming. These landscapes are more diverse, both in space and in time, than most Western European ones (Palang et al., 2006). Human pressure has decreased considerably in

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Romania's rural area during the last two decades, leading to land abandonment. On the other hand, many cultural landscapes and their unique biodiversity might be lost in the near future because land-use intensification (Cremene et al., 2005; Kuemmerle et al., 2009).

Amphibians are declining worldwide (Stuart et al., 2004), but are still well represented in traditionally managed landscapes by stable populations and species rich communities (i.e. Crochet et al., 2004; Loman and Anderson, 2007; Hartel et al., 2010). Pond breeding amphibians from temperate areas are a suitable group for landscape-scale analysis (Hecnar and M'Closkey, 1998; Houlihan and Findlay, 2003; Pellet et al., 2004; Van Buskirk, 2005; Hartel et al., 2009; Zanini et al., 2009) because: (i) they require different habitats for breeding, feeding, and overwintering, (ii) have an obligate aquatic phase usually in ponds, and (iii) because of the seasonal migration between land and water habitats, amphibians are sensitive to the landscape configuration, including man-made structures such as roads that may cause high mortality (Fahrig and Rytwinski, 2009).

In the present paper we analyze the relationship between single species occurrence and species richness of amphibians and land-use patterns around permanent ponds in a traditional rural landscape in southern Transylvania, Romania. To explore this relationship, we applied the fragmentation landscape model approach (see Fischer et al., 2004; Hartel et al., 2008). This is a widely used approach in animal landscape research (Mazerolle and Villard, 1999) including amphibians (e.g., Van Buskirk, 2005; Zanini et al., 2008; Ficetola et al., 2009; Hartel and Öllerer, 2009) and considers the landscape as the sum of various patches, representing various land uses. Patches are represented by relatively homogeneous land covers (i.e. forest or grassland cover and other clearly discernible elements such as roads, wetlands, human settlements, arable lands etc.; Fischer and Lindenmayer, 2007). The conservation implication of this approach is that landscape elements (patches) positively influencing amphibian occurrence are seen

as habitats and are supposed to act as terrestrial buffers that protect pond breeding amphibians (Ficetola et al., 2009). To our knowledge, landscape approaches based on terrestrial buffers around the breeding habitats were rarely used to predict amphibian distribution in landscapes from the CEE (see e.g., Babik and Rafinski, 2001). One important feature of CEE countries such as Romania is that traditionally managed rural landscapes are still well represented, including extensively used semi-natural meadows and pastures, forests and low urban and infrastructural development (Schmitt and Rákósy, 2007).

Testing the importance of fragment based landscape models as determinants of amphibian distribution in traditional farmlands of CEE may yield insights into the usefulness of these models for biodiversity conservation. In this study we used concentric buffers at three spatial scales around the permanent ponds, with the aim to identify the spatial scale at which land-use influences amphibian occurrence.

## 2. Methods

### 2.1. Study area

The study area is situated in the Târnava Mare river valley in central Romania (Fig. 1). Altitude ranges from 250 m to 800 m a.s.l. in an area of c. 4000 km<sup>2</sup>. Agricultural practices are largely traditional. Land-use is dominated by meadows and pastures (40%), deciduous forests (30%) and arable land (15%). About 5% of the area is urban or industrial and other land uses such as traditionally managed or abandoned orchards and vineyards make up for the remainder. About 850 km<sup>2</sup> (21%) is part of the Natura 2000 network of protected areas (Hartel et al., 2010). Large-scale developments include the regularization of 35 km of the Târnava Mare river and its tributaries, following the severe floods of 1970 and 1975. This work includes the creation of oxbow-like ponds. It also

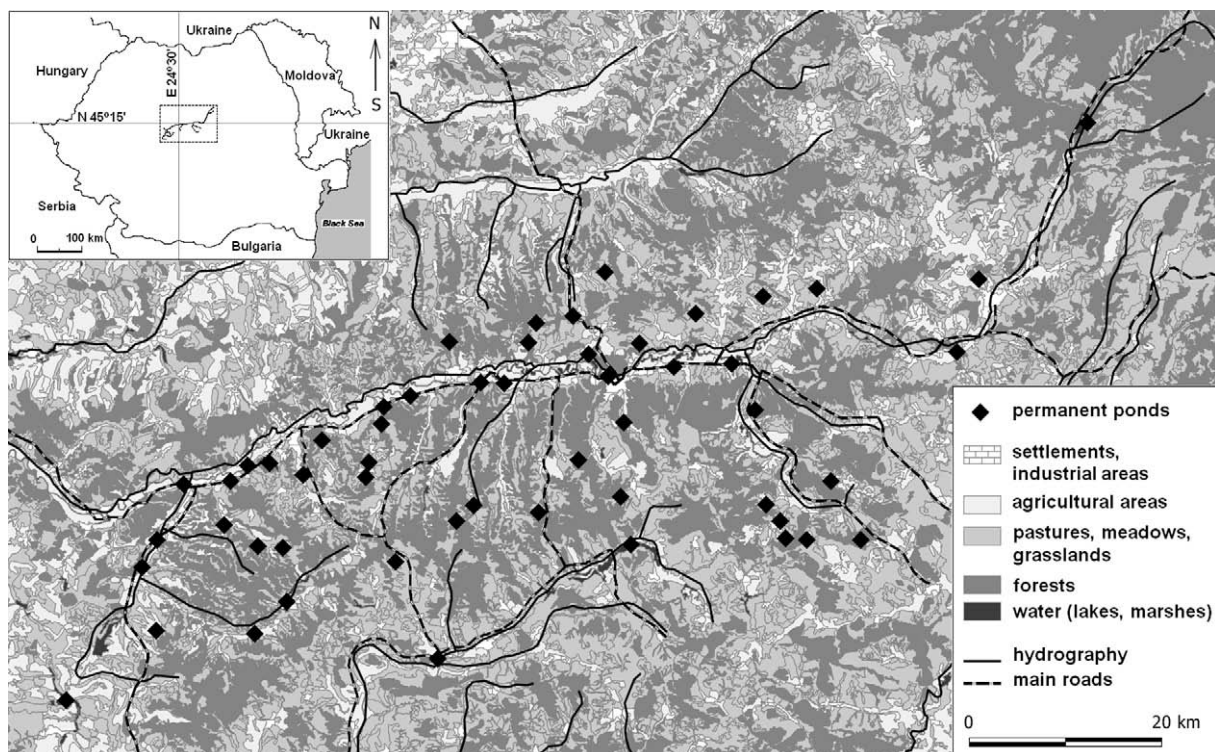


Fig. 1. Location of the sample ponds in Târnava Mare basin and major land-use categories.

triggered the intensification of agriculture and associated fertilizer input in the floodplain, and the planning of a motorway that would complement two main roads and a railway. These developments constitute the most obvious current threats to the area as does the land abandonment, mostly around the more remote villages.

## 2.2. Amphibian survey

Pond surveys were made from 2000 to 2008. Surveys were started at the end of February and lasted till August. Each pond was surveyed in two major seasons: (i) surveys carried out in February–May aimed to detect breeding adults, eggs and larvae of some species. Ponds were surveyed three–four times in each season in the mentioned period. At least one night survey was also carried out in each pond in this period. (ii) Additional two–three surveys were carried out in June–August period to identify adults of potential prolonged breeders, larvae and metamorphosis. In all but two species (the exceptions being *Bombina variegata* and *Bufo viridis*) at least one life stage denoting reproduction was detected (eggs and/or larvae). Further details in sampling methodology and effort are presented in Hartel et al. (2007, 2010).

## 2.3. Environmental data

The environmental variables were: (i) land cover data (% cover) from CORINE 2000 (EEA 2006) grouped into five land-use classes: ‘forest’, ‘pasture’, ‘arable land’, ‘wetland’ and ‘settlement’, (ii) three clearly distinguishable road categories were used according to the nature of the roads: (a) no road present, (b) low volume traffic roads connecting remote villages with main roads (with an average of 12 cars per hour at night, range 0–25), and (c) high volume traffic roads connecting larger cities as national and international roads (with an average number of 163 cars per hour at night, range 36–300), (iii) distance to the nearest forest (m), and (iv) the binary variable connectivity of ponds to adjacent forests (hereafter ‘corridor’). A corridor was considered present on the basis of one or more landscape elements that are thought to promote amphibian movement (grasslands, spinneys, hedgerows and fish free streams and springs) and absent if no such landscape element was present. Landscape composition and configuration were measured over areas of different size that were determined by radii of 400, 600 and 800 m, with the Manifold GIS software (CDA International Ltd. 2006). To ensure independence of the data we selected 54 ponds (out of 96 inventoried) for which the largest circular areas were non-overlapping while maximizing the number of ponds (Fig. 1). The remaining ponds were additionally considered with the “wetland cover” variable (see above).

## 2.4. Data analysis

The first step of the analysis was to determine the relevant scale for each variable per species and for species richness data (Houlihan and Findlay, 2003). We calculated separate generalized linear models (GLMs) with binomial error distribution for presence/absence data, with a Poisson error distribution for richness data for each variable at each of the three spatial scales. The most relevant spatial scale was selected according to the Akaike Information Criterion (AIC). The selected variables on the selected scale were subjected to multiple regression analyses and hierarchical variance partitioning to explain patterns in species occurrence (GLM with binomial error distribution) and species richness (GLM with Poisson error distribution). We controlled for potential effects of pond size and altitude by including both as explanatory variables in the initial models but did not force them into the minimal adequate model when their effects were not significant. Model performance was assessed with the area under the receiver operating characteristic curve (AUC; Fielding and Bell, 1997; Jesus and Angel, 2004). To compare the relative importance of the different environmental variables, we applied hierarchical variation partitioning (Chevan and Sutherland, 1991; MacNally, 2002). Since usually not all explanatory variables are orthogonal, a certain degree of collinearity appears in most multiple regression analyses (Table 1). Hierarchical variation partitioning calculates model fits according to all possible combinations of explanatory variables and thus allows disentangling independent effects, that can be exclusively attributed to a particular variable, and joint effects, that are equally well explained by any variable, as a fraction of total variation explained (MacNally, 2002). Hierarchical variation partitioning was performed on the variables remaining in the simplified models. Analyses were performed with R software (R Development Core Team, 2007). Species richness was compared with ANOVA with roads and corridor as grouping variables. *B. viridis* and *Rana arvalis* were excluded from the single species modeling because of their low prevalence in the studied ponds.

## 3. Results

### 3.1. Single species analysis

Ten amphibian species and a species complex were observed in 54 ponds: *Triturus cristatus* 28 ponds, *Triturus vulgaris ampelensis* 23 ponds, *Bufo bufo* 44 ponds, *B. viridis* five ponds, *R. arvalis* one pond, *Rana dalmatina* 45 ponds, *Rana esculenta* complex (i.e. the European waterfrog complex) 40 ponds, *Rana temporaria* 29 ponds,

**Table 1**  
Pearson  $r$  correlation coefficients among environmental variables used for modelling amphibian species richness. Note that for single species modelling the spatial scale of the variables differed but effects on collinearities were minimal. Analysis based on 54 ponds.

	Pond altitude (m)	Pond size (ha)	Forest distance (m)	Corridor presence (400 m radius)	Road presence (800 m radius)	Settlement cover (400 m radius)	Arable land cover (800 m radius)	Pasture cover (400 m radius)	Forest cover (800 m radius)
Pond size (ha)	-0.17								
Forest distance (m)	-0.30	0.11							
Corridor presence (400 m radius)	0.45	-0.15	-0.65						
Road presence (800 m radius)	-0.39	0.07	0.27	-0.55					
Settlement cover (400 m radius)	-0.26	0.20	0.05	-0.33	0.10				
Arable land cover (800 m radius)	-0.37	0.17	0.21	-0.30	0.47	-0.08			
Pasture cover (400 m radius)	-0.06	0.01	0.06	0.07	-0.25	-0.22	-0.43		
Forest cover (800 m radius)	0.44	-0.24	-0.41	0.31	-0.26	-0.25	-0.49	-0.15	
Wetland cover (800 m radius)	0.01	-0.06	0.33	-0.04	0.05	0.12	-0.05	-0.33	-0.10

**Table 2**Relationship between landscape variables and amphibian pond occupancy investigated by logistic regression analysis.  $\beta$ , coefficient estimate; SE, standard error.

Species	Landscape variables	Statistics			
		$\beta$	SE	P	AUC
<i>Triturus cristatus</i>	Forest distance	−0.001	0.0009	0.03	0.67
<i>Triturus vulgaris</i>	High traffic road (400 m)	−2.91	1.38	0.03	0.80
<i>Rana dalmatina</i>	High traffic road (400 m)	−3.14	1.21	0.009	0.84
<i>Rana temporaria</i>	High traffic road (800 m)	−3.37	1.34	0.01	0.95
<i>Rana esculenta</i> complex	Forest (800 m)	0.07	0.03	0.03	
	Elevation	−0.02	0.01	0.01	0.92
	Low traffic road (400 m)	3.96	1.77	0.02	
	Arable lands (400 m)	−0.11	0.05	0.04	
<i>Bufo bufo</i>	Forest (800 m)	−0.10	0.05	0.04	
	Green corridor (400 m)	3.73	1.01	<0.001	0.85
<i>Hyla arborea</i>	High traffic road (600 m)	−2.73	1.34	0.04	0.81
<i>Pelobates fuscus</i>	Grassland (600 m)	−0.15	0.05	0.007	0.80
	Arable lands (600)	−0.12	0.05	0.01	
	Forest (400)	−0.15	0.05	<0.01	
	Settlement (400 m)	−0.12	0.05	0.01	
<i>Bombina variegata</i>	Low traffic road (600 m)	−1.44	0.73	0.04	0.64

*B. variegata* 30 ponds, *Pelobates fuscus* 19 ponds and *Hyla arborea* 37 ponds.

Single species regression models showed a model fit of AUC > 0.8 for all species except *T. cristatus* and *B. variegata* (Table 2). Four species were negatively associated with the presence of high traffic roads at various scales (*T. vulgaris* and *R. dalmatina* at 400 m spatial scale, *H. arborea* at 600 m and *R. temporaria* at 800 m) whereas the presence of the *R. esculenta* complex was positively associated with low traffic roads (400 m; Table 2). Significant associations were found for: (i) forest and *R. temporaria* (positive, 800 m) and the *R. esculenta* complex and *P. fuscus* (negative, 800 m), (ii) corridor and *B. bufo* (positive, 400 m), and (iii) arable land and the *R. esculenta* complex (negative, 400 m). The presence of *P. fuscus* was also negatively associated with urbanization and forest (400 m) and grassland and arable land (600 m; Table 2).

According to the hierarchical partitioning analysis, the percentage of explained deviance by roads was the small (<30%) in *T. cristatus*, *T. vulgaris*, *Pelobates fuscus*, *Rana esculenta* complex, *R. temporaria* and *B. bufo*, medium (<45%) in *H. arborea* and *R. dalmatina* and highest (50%) in *B. variegata* (Fig. 2). The independent component of the road effect was larger than the joint component for *R. dalmatina* (Fig. 2). In *T. vulgaris* the independent effect of roads explained 1.2% of deviance whereas the joint effect was 19.5% (Fig. 2). Forest cover explained most of the deviance in *P. fuscus* and *R. temporaria*, with a large joint component in the latter species (Fig. 2). The hierarchical partitioning analysis suggests that landscape elements such as cover of grassland, forest, arable land, settlements and wetlands have a minor effect on the occurrence of the majority of amphibian species (low I and J effects) (Fig. 2).

### 3.2. Species richness

Amphibian species richness was best explained by high volume traffic roads at the 800 m spatial scale and by forest distance (Table 3). Species richness was smallest in ponds with high volume traffic roads in the surrounding landscape and the largest in ponds with no roads (Tukey HSD,  $P < 0.001$ ; Fig. 3A). The species richness in ponds with low volume traffic road was intermediate (Fig. 3A). Ponds connected to a forest by corridor have higher species richness on average than ponds with no corridor ( $t = 3.33$ ,  $df = 52$ ,  $P < 0.05$ ; Fig. 3B).

According to the hierarchical partitioning analysis, roads (% deviance >30%), corridors (% deviance >20%) and forest distance (% deviance >15%) had the highest effect on species richness

(Fig. 2). The independent effect of road was higher than those of the other two variables. However, the joint components were larger than the independent effects in all three variables. The percentage of deviance was small in settlement, agriculture, pasture, forest and wetland (<5%) (Fig. 2).

### 4. Discussion

While landscape elements such as distance to forest and presence of green corridors and wetlands differ in their importance among amphibian species, our study shows that roads are the prime landscape elements influencing amphibian occurrence and overall species richness. Negative effects of roads on biodiversity, including amphibians, are well documented (reviewed by Cushman (2006), Eigenbrod et al. (2008a,b, 2009), Fahrig and Rytwinski (2009)). Roads may impact amphibians directly by massive road kills when their migration routes cross roads, aggravated by the absence of car avoidance behavior and low migration speed. The impact can also be indirect, by pollution, by disturbance affecting the individual behavior and through isolation from neighboring populations and the disruption of connectivity between critical habitats that are required to complete their year-round life history (see Fahrig and Rytwinski (2009) for a review). In addition to the dominant independent effect of roads, we often observed a large joint component. These joint components cannot be exclusively attributed to roads but can be equally well explained by other variables that may represent further reduction of terrestrial habitat quality, such as the degree of cover and connectivity with forests and wetlands. As an example, *R. temporaria* is negatively associated with roads at the 800 m spatial scale (Table 1). As the joint component of this variable was high, the actual extent to which roads affect this species remains unknown, but a combination of negative effects of roads, reduced connectivity and increasing distance to forests, decreasing amount of forest and neighbouring wetland habitats and increasing arable land, whose joint effects are also comparably high, is likely (see also Van Buskirk, 2005, who found positive relationship between *R. temporaria* and forest cover; Hartel et al., 2009).

The effect of landscape structure on amphibian pond occupancy at various spatial scales was tested in Europe (e.g., Pellet et al., 2004; Denoël and Ficetola, 2007; Zanini et al., 2008; Ficetola et al., 2009) and in North America (e.g., Lehtinen et al., 1999; Houlihan and Findlay, 2003; Price et al., 2006; Eigenbrod et al., 2009). In our study, the landscape effect was more prominent at the

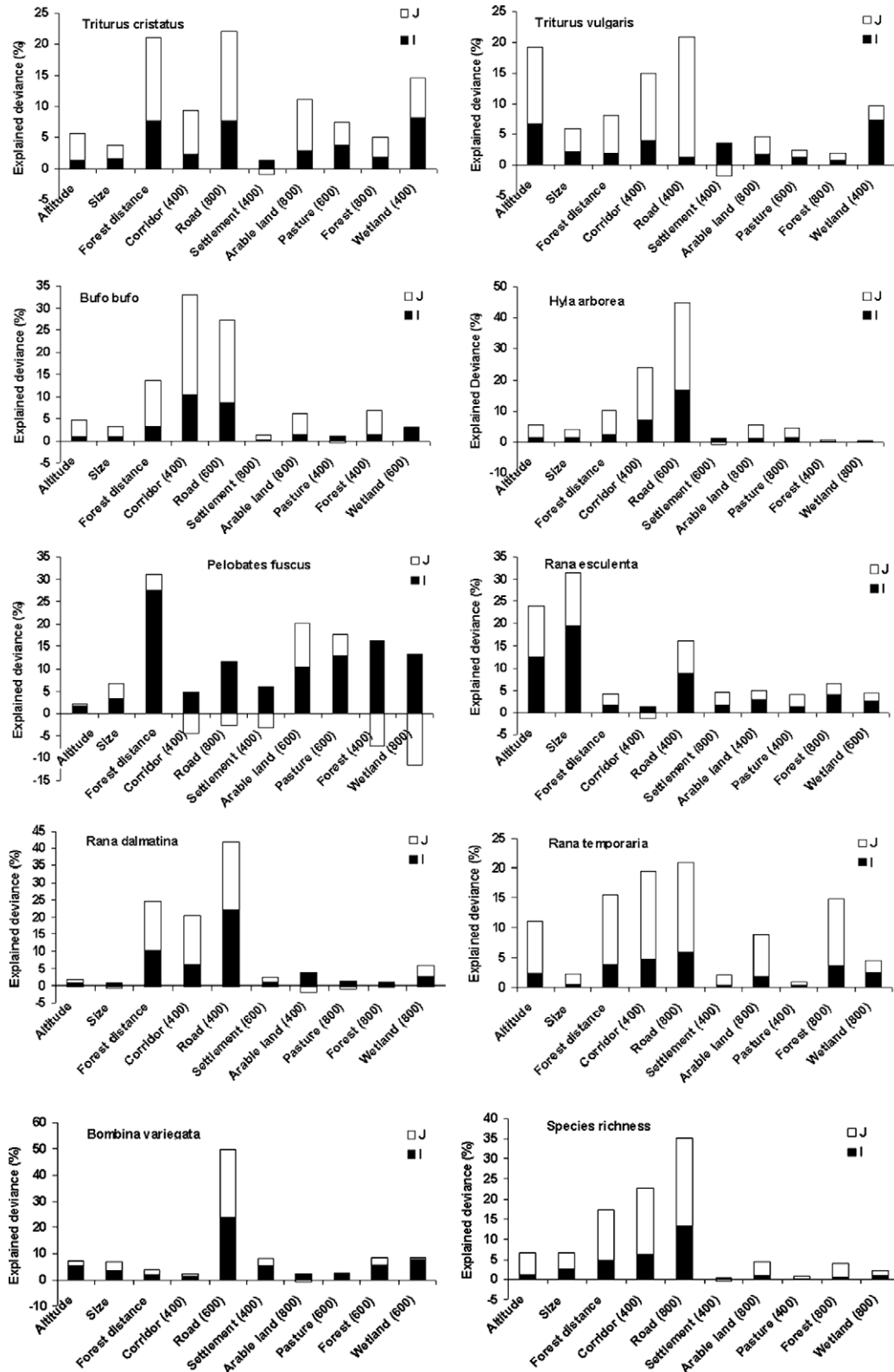


Fig. 2. Hierarchical partitioning analysis of the proportion of deviance in pond occupancy of individual species and overall species richness explained by landscape variables. Independent (I) and joint (J) effects are shown.

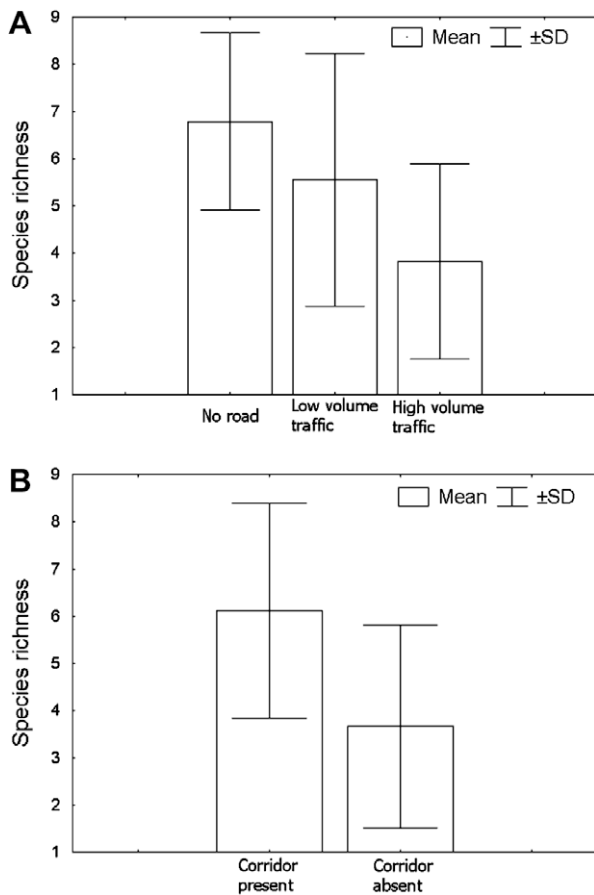
400 m and 600 m radius, with the exception of *R. temporaria* and *R. esculenta* complex to forests and roads at the 800 m scale. Corridors were important for *B. bufo* and for the overall amphibian species richness. Several studies have highlighted forests as important terrestrial habitats for amphibians (e.g., Houlahan and Findlay, 2003;

Van Buskirk, 2005; Eigenbrod et al., 2008a,b). However, the negative effect of forest cover at the 800 m scale on *R. esculenta* complex indicates that forest ponds are not amenable to this taxon (Van Buskirk, 2005; Ficetola et al., 2009), or that such ponds are less likely to be colonized. Instead, the presence of the *R. esculenta* com-

**Table 3**

Coefficient estimates ( $\beta$ ) of the minimal adequate generalized linear regression model relating amphibian species richness to landscape variables. SE, standard error.

Variable	Statistics			
	$\beta$	SE	Z	P
High traffic road (800 m)	-0.49	0.15	-3.29	<0.001
Forest distance	-0.0003	0.0001	-1.82	0.06



**Fig. 3.** Average species richness in relation to (A) road categories and (B) presence/absence of a corridor in between breeding ponds and forest (see text for details). (A) species richness significantly differed between ponds having no roads in their vicinity (highest species richness) and ponds with high traffic roads (lowest species richness) (Tukey HSD,  $P < 0.001$ ).

plex appears to be associated with unshaded, well vegetated pond with or without predatory fish (Hartel et al., 2007, 2009). The hierarchical partitioning analysis indicated the importance of pond size and altitude for this taxon.

In areas of intensive agriculture, amphibians usually benefit from forests, wetlands and grasslands (e.g., “rough pasture”, Scribner et al., 2001), while they suffer from cropland and urban areas (e.g., Joly et al., 2001; Scribner et al., 2001; Pellet et al., 2004; Van Buskirk, 2005; Denoël and Ficetola, 2008; Greenwald et al., 2009). Our study confirms these findings, but the effects size of negative and positive impacts of these different landscape structures is low compared to that of roads. We suggest that the low level of agricultural intensity in the study region, characterized by small-scale traditionally managed landscapes and the absence of heavy machinery and chemical pollutants, still provides viable conditions for amphibian species. Moreover, the low cover of settlements (see methods) also can explain the lack of settlement cover on amphibians.

For three species (*T. cristatus*, *B. variegata* and *P. fuscus*) the logistic regression models remained unconvincing, perhaps due to explanatory variables that we failed to consider. In *T. cristatus* for example it is likely that pond related variables such as predatory fish and vegetation cover are more relevant than parameters of the landscape (Hartel et al., 2007). *B. variegata* prefers temporary ponds for breeding and permanent ponds considered here represents a sub-optimal habitat for this species (Hartel and Moga, 2007). The logistic regression model for *P. fuscus* shows a high fit but considering that just negative associations were found, interpretation of the model is not straightforward. The CORINE data base did not allow us to include the parameter ‘soil type’ that is potentially the most relevant to explain the distribution of this species (Nyström et al., 2007).

In conclusion, this study showed that high volume traffic roads were the most important landscape element influencing the pond occupancy by amphibians. In contrast, the effects of landscape compositional elements which are usually attributed with immense negative effects on amphibians such as cover of settlements and agricultural land were largely negligible. Similarly, the importance of land-cover types that are thought to have positive effects on amphibians such as forests, pastures and wetlands was only marginally higher. This leads to the conclusion that landscape composition generally is of little importance for amphibian species in the study area. We suggest that this is a consequence of the mainly traditional, extensive management of the land where agriculture represents no threat to amphibians and refuges are thus not needed. These results have potentially important consequences for conservation management of amphibians in the traditionally managed rural landscapes of CEE. Maintaining traditional (extensive) land management would be a key factor in the protection of amphibians in CEE. This will be a real challenge especially because the adherence to the European Union will result in land-use intensification and infrastructural and urbanistic development in many areas. These will lead to increased fragmentation of landscapes. A balance has to be sought between the legitimate desire to develop infrastructure and increase agricultural revenue and the beneficial effects of low intensity land-use. The environmental richness as found in many CEE countries is at stake. This presents a challenge that may be seen as such an opportunity to not repeat mistakes from the past as to find new approaches in conservation biology.

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