Distribution and population size estimation of the moor frog Rana arvalis Nilsson, 1842 in Ljubljansko barje Nature Park, central Slovenia

David STANKOVIĆ^{1,2} & Maja CIPOT^{1,3}

¹ Herpetološko društvo – Societas herpetologica slovenica, Večna pot 111, SI-1000 Ljubljana, Slovenija;

Abstract. Until recently, the moor frog Rana arvalis has been recorded only from the Pannonian and Sava Basins of north-eastern and south-eastern Slovenia, respectively. Here we report on the occurrence of the species at Ljubljansko barje, central Slovenia, and give estimates of the species' abundance and distribution. In addition, we present some morphological characteristics of this population and offer insights into the present threats and the status of its habitats. The overall population size was estimated through egg mass count and is calculated to be more than 19,000 adults. All findings were distributed in two separate clusters. The northern cluster hosted more than 96% of all findings, while the smaller cluster in the south-eastern part of Ljubljansko barje accounted for only a fraction of the records. Even though the habitats in the northern cluster are relatively well preserved, the possibility of local extinction remains high owing to the immediate proximity of the capital and the municipal landfill site, which is planned for expansion in the near future.

Key words: moor frog, Rana arvalis, egg mass counts, population size, Ljubljansko barje Nature Park

Izvleček. Razširjenost in ocena velikosti populacije barske žabe Rana arvalis Nilsson, 1842 v Krajinskem parku Ljubljansko barje, osrednja Slovenija – Barska žaba (Rana arvalis) je evrazijska vrsta, za katero je do nedavnega veljalo, da živi zgolj v nižinah severovzhodne in jugovzhodne Slovenije. V prispevku poročamo o pojavljanju te vrste na Ljubljanskem barju, o njeni razširjenosti in o velikosti tamkajšnje populacije. Poleg tega podajamo še vpogled v stanje in ogroženost te populacije in predstavljamo nekaj njenih morfoloških značilnosti. Velikost populacije smo ocenili s štetjem mrestov na mrestiščih. Na Ljubljanskem barju živi več kot 19.000 odraslih barskih žab, ki so geografsko razporejeni v dve večji območji. Več kot 96 % vseh najdb prihaja iz večjega, severnega območja, medtem ko preostale najdbe prihajajo iz jugovzhodnega dela Barja. Čeprav so gozdni habitati na severnem območju razmeroma dobro ohranjeni, neposredna bližina mesta in deponije, ki se bo predvidoma širila, barske žabe močno ogrožata. Uničenje primernih habitatov in prekinitev povezav med mrestišči in kopenskimi habitati namreč vodi v lokalno izumrtje vrste.

Ključne besede: barska žaba, Rana arvalis, ocena velikosti populacije, štetje mrestov, KP Ljubljansko barje

E-mail: david.stankovic@guest.arnes.si

 ² Department of Animal Science, Biotechnical Faculty, University of Ljubljana, Groblje 3, SI-1230 Domžale, Slovenia
 ³ Center za kartografijo favne in flore, Antoličičeva 1, SI-2204 Miklavž na Dravskem polju, Slovenija;

E-mail: maja.cipot@ckff.si

Introduction

Borders of species distribution and identification of isolated populations have recently received considerable attention mostly due to the reappraisal of their conservation values. They possibly contain the gene pool that is best suited for colonisation of new habitats or recolonisation of adjunct areas where they have become extinct (Hamilton & Eckert 2007). Thus it is important and urgent to document the fine-scale distribution, habitat, life history and demography of peripheral populations. In the European Union, a great deal of new data on the borders of species distribution was collected during various activities related to implementations of Natura 2000 networks, especially in countries that have been under-explored in terms of recording and researching biodiversity (Jovanović 2009, Jelić et al. 2012, Bonk et al. 2012). This also applies to Slovenia, where knowledge of the border distribution of some amphibian species has increased significantly in recent years (Krofel 2005, Stanković & Delić 2012, Stanković & Poboljšaj 2013, Stanković et al. 2014).

The moor frog Rana arvalis is a Euroasiatic lowland species distributed through a vast area spreading all the way from northern France to Lake Baikal and beyond (Glandt 2008). Its distribution is continuous over most of its range, except in the western part, where a number of isolated populations occur. In most cases, fragmentation can be explained by recent human activities; however, fossil records from western Europe, the Balkans, southern Russia and the Caucasus, and an isolated population in southern Norway and by the Sea of Azov indicate an even broader species distribution at the end of the Pleistocene (Roček & Šandera 2008, Kuzmin et al. 2009). In Europe, two regions of species distribution can be distinguished. The larger area (i) spreads from above the Arctic Circle in Scandinavia over the European lowlands of central, western and eastern Europe and all the way to the coast of the Black Sea in Ukraine. The smaller area (ii) extends like a pocket from the central European lowlands into the Pannonian Basin along the River Danube. These two areas correspond with the two recognized geographic subspecies, whose status, however, is partly disputed by morphological and molecular data (Babik & Rafínski 2000, Babik et al. 2004): R. a. wolterstorffi inhabits the Pannonian Basin, whereas *R. a. arvalis* is found in the remainder of the distribution range. The species shows a great variety in the dorsal colouration pattern, which does not correspond to its taxonomic division, but is described as different colour morphs or varieties. The three most common morphs are striata, maculata and unicolor. While the maculata morph occurs practically everywhere, the unicolor and striata morphs have limited geographical distributions. In general, the unicolor morph is rare in the northern and north-western border of the species distribution and is commonest to the south and to the east - it is the most dominant in Croatia and in south-eastern Hungary, but is probably rare in Austria. On the other hand, the striata morph is more common towards the border areas of the species distribution range, including Austria and most of Hungary, but is rare in the central and south-eastern areas of the range. In addition, this form is absent in the Sava Basin (Fog 2008a, b).

In Slovenia, *R. arvalis* reaches its south-eastern distribution limit. Until recently, it was believed to be limited to the Pannonian and Sava Basins in the north-eastern and south-eastern parts of the country, respectively (Poboljšaj et al. 2008). In the spring of 2010, however, the authors confirmed the species presence at Ljubljansko barje, central Slovenia (Stanković & Cipot 2013). Among the neighbouring countries, this species can be found in

Croatia, Austria and Hungary (Tvrtković & Kletečki 2008, Grillitsch & Cabela 2008, Puky & Schad 2008). Except for the populations along the River Kupa in Croatia and the River Drava in Austria, all other populations are continuous across the national borders of the adjacent countries.

Although *R. arvalis* is listed as a species of least concern (LC) by the IUCN (Kuzmin et al. 2009), it is strictly protected under the Bern Convention (Appendix II) and protected by the EU Habitats Directive 92/43/EEC (listed in Annex IV). In Slovenia, it is legally protected by the Decree on Protected Wild Animal Species (Ur. I. RS 2004a) and listed as an endangered species (E) in the »Slovenian Red Data List« (Ur. I. RS 2002).

The aim of this contribution is to give a precise description of the first indisputable record of *R. arvalis* in central Slovenia, to present data on the size and distribution of the subpopulations at Ljubljansko barje and to assess the status of the population and its habitat along with possible threats. By having extensively surveyed potential *R. arvalis* habitats over an area of 135 km² we have significantly expanded the knowledge of the distribution and abundance of this species in the south-eastern border of its range. In addition, we offer insights into the terrestrial and aquatic habitat selection and provide observations on some morphological characteristics of the populations studied. We conclude by placing these populations in the context of the known distribution in adjacent regions and countries and suggest two scenarios for the origin of this well isolated population.

Material and methods

Records of *R. arvalis* at Ljubljansko barje were compiled from a large-scale survey in 2011 (Cipot 2011), other herpetological surveys (Govedič et al. 2009) and unpublished data. All surveys were restricted to Ljubljansko barje Nature Park.

Study area

Ljubljansko barje is some 160 km² large lowland area located in central Slovenia south of the capital city of Ljubljana. This is one of the southernmost regions in Europe where raised bogs can be found. In the second half of the 18th century, large-scale draining operations were put in motion. By the turn of the 20th century, the marshes were drained to an extent that made the land exploitable for extensive farming (Brenčič 2008).

Despite the intensive degradation and urbanisation, Ljubljansko barje still hosts a few larger complexes of wet meadows, traces of lowland raised bogs and fragments of wet woodlands. Even though less than 10% of the area is covered by woodlands (Kotarac & Grobelnik 1999), some fragments are still in good condition and could present suitable habitats for *R. arvalis*.

Potential breeding sites of *R. arvalis* were preselected based on the species' ecological characteristics and breeding demands, taking all confirmed and possible sightings into account. The timing of the surveys coincided with the species breeding season in Slovenia (March through April) and was carried out in 2011. However, some sites were visited outside this timespan. Twelve areas were selected for the survey, including all larger lowland woodlands and marshes found in the region (Fig. 1). The surface of the preselected areas extended from 3.0 to 186.0 ha with an average of 64.2 ha, while the total combined surface area added up to 770.3 ha (Tab. 1). Most of the preselected areas are characterised by a relatively large proportion of woodlands (44% on average), followed by extensive agricultural land (24.9% on average) and finally by marshes (13.5% on average).

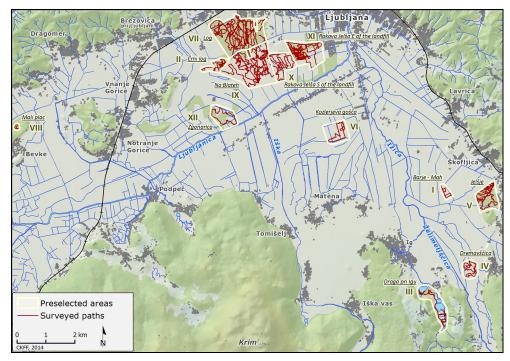


Figure 1. All preselected areas were checked on foot and all potential breeding grounds and habitats of *R. arvalis* were inspected.

Slika 1. Pregledanost izbranih območij. V vseh vnaprej izbranih območjih so bila potencialna mrestišča in bivališča barskih žab prehojena in pregledana.

Table 1. Land use and habitat characteristics in the preselected areas to be examined for *R. arvalis* breeding sites (after Kotarac & Grobelnik 1999). Each area is marked by a roman numeral. Areas where *R. arvalis* breeding was confirmed are in bold. The forest in Rakova Jelša E of the landfill (*) was completely cut down a few years prior to the 2011 survey.

Tabela 1. Pokritost pregledanih območij s skupinami habitatnih tipov v odstotkih glede na rabo pomembno za barske žabe (povzeto po Kotarac & Grobelnik 1999). Pregledana območja so označena z rimskimi številkami. Območja, kjer je bilo razmnoževanje barske žabe potrjeno, so odebeljena. Gozd v Rakovi Jelši vzhodno od deponije (*) je bil popolnoma posekan nekaj let pred inventarizacijo barskih žab v letu 2011.

PR	ESELECTED AREAS	Surface area [ha]	Urban area [%]	Intensive agriculture areas [%]	Extensive agriculture areas [%]	Woodlands [%]	Swamps [%]	Water bodies [%]
Ι	Barje – Mah	9.4	11.3	0.9	42.3	34.0	7.6	3.9
II	Črni Log	19.9	11.2	1.4	40.8	46.4	0	0.2
III	Draga pri Igu	64.1	3.6	1.3	12.7	52.9	8.9	20.6
IV	Dremavščica	26.3	9.2	19	33.3	33.8	0.5	4.2
V	Jelšje	55.8	6.0	1.8	6.9	84.2	0.4	0.7
VI	Kozlarjeva gošča	44.7	2.7	14.2	30.9	0.6	49.2	2.4
VII	Log	172.7	1.4	0.1	11.0	86.7	0	0.8
VIII	Mali plac	3.0	0	0	0	20.6	79.4	0
IX	Na blateh	186.0	2.7	14.6	37.8	41.9	2.6	0.4
x	Rakova Jelša S of the landfill	58.9	2.9	10.9	40.0	35.6	9.6	1.0
XI	Rakova Jelša E of the landfill*	80.5	16.9	6.2	29.8	*42.0	3.7	1.4
XII	Zgonarica	49.0	5.5	31.9	13.6	45.2	0.2	3.6
Areas combined		770.3	4.9	8.8	24.7	52.9	5.8	2.8
Average values		64.2	6.1	8.5	24.9	43.7	13.5	3.3

Species detection and estimation of census population size

During the survey, all wet meadows, flood-meadows, marshes and swamps, ponds and ditches in the preselected areas were inspected, searching for both egg clutches and adults. During the breading season, adult *R. arvalis* males were visually identified by their blue colouration. Otherwise, adults were identified based on the colouration of the belly, head mask pattern, the shape of the metatarsal tubercle and other morphological characters or by vocalisation. All caught specimens were released at the site of capture. Egg clutches were identified by the characteristic manner of egg deposition. Females of both *R. arvalis* and the common frog *Rana temporaria* deposit their eggs in aggregations in shallow water. By way of comparison, *R. temporaria* deposits somewhat larger egg clutches containing more eggs, while the clutches of *R. arvalis* are smaller and have a more compact globular shape (Nöllert & Nöllert 1992). An accurate distinction between the clutches of both species is possible only when they are relatively fresh. Therefore, our sightings were classified in two separate categories. The first category contained sightings of egg clutches that belonged to *R. arvalis* beyond doubt. On the other hand, clutches that were not indisputably identified and could belong to either of the two species were placed in the category *R. arvalis* temporaria group.

Although by no means representative of the true census population size, the number of clutches can give a rough indication of the number of breeding females and some idea on the abundance of frogs in a given area (Glandt 2008). To get a good estimate of the census size, egg clutches were counted in two independent runs, both conducted when most of the clutches had already been laid but were still fresh enough to allow reliable identification. Only the count with more clutches per locality was considered. The available data on the sex ratio in *R. arvalis* varies quite extensively. Some researchers report the ratio to be in favour of females (Günther & Nabrowsky 1996), while others claim the opposite (Hellbernd 1987). Because of these discrepancies, the proposition given by Glandt (2008) was followed, and 1:1 sex ratio for the estimation of the minimum number of males based on egg mass count was used.

Besides counts of egg mass, the census size was also estimated by counting adult blue males on the breeding sites. If possible, the males were counted from distance with the help of binoculars and camera. Because adult males can be very timid during breeding, the observer had to wait for the animals to habituate to his/her presence, which could last up to half an hour or even more. Preselected areas with no indisputable records of *R. arvalis* were processed as areas where this species is only probably present.

Morphometric analysis and analysis of colour pattern

A total of 33 adult males were collected at three Ljubljansko barje localities: the Jelšje (V), Log (VII) and Na blateh (IX) areas. Two morphometric measurements were taken on live individuals using callipers with an accuracy of 0.1 mm: snout-vent length (SVL) and tibia length (TL). The frogs were captured at their breeding sites during the mating season in April 2011, with males mostly in their blue colouration. The SVL/TL ratio was calculated.

For colouration analysis, a total of 35 adults from the Log (VII) area were inspected. Based on their colouration pattern and the distribution of skin tubercles, individuals were assigned to either striata, maculata, unicolor or the hemistriata morph as suggested by Fog (2008a, b). Hemistriata morph is a transitional form and can be conceived as a variant of other morphs. The main reason for defining it is to avoid specimens to be falsely classified as striata. Frogs were captured in August and September 2012. The prevalence of colour morphs was compared to that from the alluvial forest in Rački ribniki Nature Park in north-eastern Slovenia, where 20 adults were collected in July of 2013.

Results

Distribution of *R. arvalis* in the Ljubljansko barje area

On 25. 3. 2010, the authors confirmed the presence of *R. arvalis* at Ljubljansko barje. A small group of about twenty individuals, including blue males, was found on a flooded fallow land south of Ljubljana in an area known under the name of Na blateh (IX). A few days later another confirmation of the species presence in the area known as Log (VII) came from Davorin Tome. The closest known population at that time was located about 100 km SE in Krakovski gozd near Novo mesto.

During the spring of 2011, *R. arvalis* breeding sites were confirmed in five out of the twelve preselected areas, with altogether 6,112 egg clutches recorded, while additional 3,669 clutches could also belong to *R. temporaria*. Therefore, based on egg clutch counts, Ljubljansko barje hosts at least 12,224 *R. arvalis* adults and 7,338 adults that may alternatively belong to *R. temporaria*. This sums to 19,562 adults, when considering all *R. arvalis*/temporaria clutches as in fact belonging to *R. arvalis*. This conclusion can be drawn on the basis of the rareness or even absence of *R. temporaria* in preselected areas and generally in this lowland region. In fact, less than 3% of all our findings (egg clutches or adults) were assigned to *R. temporaria* beyond doubt.

Breeding of *R. arvalis* was confirmed in the following preselected areas - Črni Log (II), Log (VII), Na blateh (IX), Rakova Jelša south of the municipal landfill site (X) and Rakova Jelša east of the municipal landfill site (XI), while the Dremavščica (IV) and Jelšje (V) areas contained only clutches that could belong to either of the two species (Tab. 2). On the other hand, *R. arvalis* adults were observed at the following five preselected sites: Črni Log (II), Jelšje (V), Log (VII), Na blateh (IX) and Rakova Jelša east of the municipal landfill site (XI).

The inspected breeding sites can be divided into two clusters. Of a total of 18 breeding aggregations with >100 *R. arvalis* egg clutches of recorded at Ljubljansko barje, all but one belong to the northern cluster (A) (Figs. 2–3). This cluster spans four preselected areas, which are spread in the close vicinity of the municipal landfill site. In the Log area (VII) alone, four breeding-aggregation sites with >500 clutches and four with >200 *R. arvalis* clutches were registered, while three breeding aggregations with a high density of clutches (>200) were also recorded in the neighbouring Črni Log (II) and Na blateh areas (IX). On the other hand, Rakova Jelša east and south of the municipal landfill site (X, XI) holds fewer small breeding sites. In other words, the northern cluster includes more than 96% of all findings and has the highest ecological densities (Tab. 3). On the other hand, the smaller cluster (B) is comprised of only one or possibly two preselected areas, as Dremavščica (IV) contains only *R. arvalis/temporaria* group (Fig. 2). During the survey, 2,019 clutches of *R. dalmatina* and 334 clutches that belong to *R. temporaria* frog beyond doubt were also recorded (Tab. 2).

PR	ESELECTED	R. arv	alis	R. arv tempo		R. dalm	atina	R. temp	oraria	<i>R.</i> sp.	(comb	ined)
AREAS		No.	No.	No.	No.	No.	No.	No.	No.	No.	No.	No.
		clutches	adults	clutches	adults	clutches	adults	clutches	adults	adults	clutches	adults
Ι	Barje – Mah	0	0	0	0	13	0	0	0	0	13	0
II	Črni Log	258	180	254	0	56	0	0	0	0	568	180
III	Draga pri Igu	0	0	0	0	96	0	192	633	8	288	641
IV	Dremavščica	0	0	59	0	19	0	0	0	0	78	0
V	Jelšje	0	8	277	0	949	64	8	0	0	1234	72
VI	Kozlarjeva gošča	0	0	0	0	2	0	0	0	0	2	0
VII	Log	1923	4347	2245	0	165	1	5	11	1	4338	4360
VIII	Mali plac	0	0	0	0	178	4	16	0	2	194	6
IX	Na blateh	1984	321	482	0	245	0	0	0	4	2711	325
Х	Rakova Jelša S of the landfill	12	0	10	0	46	0	0	0	0	68	0
XI	Rakova Jelša E of the landfill	1935	19	342	0	239	0	113	0	2	2629	21
XII	Zgonarica	0	0	0	0	11	0	0	0	0	11	0
Α	All together		4875	3669	0	2019	69	334	644	17	12134	5605

Table 2. Adult brown frogs and egg clutches counted in preselected areas during the survey in 2011.
Tabela 2. Število preštetih odraslih osebkov in mrestov rjavih žab na posameznem pregledanem območju v letu 2011.

Table 3. Population and ecological density of *R. arvalis* in preselected areas at Ljubljansko barje. Number of adults was estimated by egg counts and considering 1:1 sex ratio (Glandt 2008b). Preselected areas marked with an asterisk (*) belong to the southern cluster (B).
Tabela 3. Gostota in ekološka gostota odraslih osebkov barskih žab in skupine *R. arvalis*/*temporaria* v letu 2011, izračunana iz preštetih mrestov obeh taksonov na podlagi razmerja med spoloma 1:1 (Glandt 2008b) na posameznem raziskovalnem območju. Območja označena z zvezdico (*) so del južne zgostitve (B).

	PRESELECTED AREAS	Surface area [ha]	Density	Ecological density
II	Črni Log	19.9	51.46	58.94
IV	Dremavščica*	26.3	4.48	6.63
٧	Jelšje*	55.8	9.92	10.85
VII	Log	172.7	48.27	49.44
IX	Na blateh	186.0	26.52	32.221
Х	Rakova Jelša S of the landfill	58.9	0.75	0.88
XI	Rakova Jelša E of the landfill	80.5	56.57	74.94

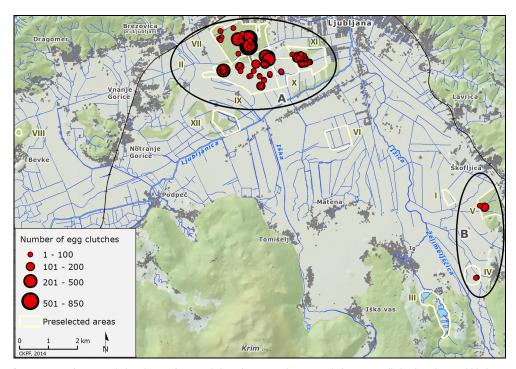


Figure 2. Distribution and abundance of *R. arvalis* breeding grounds as recorded in 2011. All clutches that could belong to *R. arvalis* were considered, including those that could not be identified beyond any doubt and were assigned to the *R. arvalis/temporaria* group. Dots represent either individual clutches or aggregations thereof rather than separate breeding grounds; a single breeding ground could thus be represented by more than one dot.
 Slika 2. Razporejenost in razpršenost seštevka mrestov barskih žab in nadrejene skupine *Rana arvalis/temporaria* na

Slika 2. Razporejenost in razpršenost seštevka mrestov barskih žab in nadrejene skupine Rana arvalis/temporaria na Ljubljanskem barju v letu 2011. Pike ponazarjajo najdišča skupine (blazine) mrestov ali posameznih mrestov in ne posameznega mrestišča (eno mrestišče lahko ponazarja več pik).

Morphometric analysis and colour pattern morphs

The mean body length of inspected males (SVL) was 70.2 mm, while the overall SVL/TL ratio was 1.65 (Tab. 4).

- **Table 4.** Mean body length values (X) of 33 *R. arvalis* males from Ljubljansko barje, with standard deviations (SD) and minimum (MIN) and maximum (MAX) values for snout-vent length (SVL), tibia length (TL) and for the ratio between snout-vent and tibia length (SVL/TL).
- Tabela 4. Povprečni podatki morfometrijskih meritev (X) 33 samcev barske žabe z Ljubljanskega barja, standardne deviacije (SD) ter najnižja (MIN) in najvišja (MAX) vrednost za dolžino telesa (SVL), dolžino goleni (TL) in izračunano razmerje med dolžino telesa in dolžino goleni (SVL/TL).

	SVL [mm]	TL [mm]	SVL/TL
Х	70.2	42.5	1.65
MIN	54.1	31.4	1.47
MAX	79.3	47.7	1.84
SD	6.3	3.6	0.09

The analysis of colour patterns revealed that 25 of the 35 specimens (71.4%) belonged to the unicolor morph, only three specimens (8.6%) to the striata morph, while the rest (20.0%) belonged to the transitional hemistriata morph and none was assigned to the maculata morph. The prevalence of the unicolor morph is even more striking when compared to colour morph distribution of a *R. arvalis* population from the alluvial forest in Rački ribniki Nature Park in north-eastern Slovenia (River Drava watershed). Of 20 inspected specimens only four (20%) belonged to the unicolor morph, while 9 belonged to the striata morph (45%) and six to the transitional hemistriata morph (30%). In addition, one specimen was recognised to belong to the maculata morph (5%).

Discussion

Distribution of *R. arvalis* in the Ljubljansko barje area

During the short time frame since the first indisputable records of *R. arvalis* at Ljubljansko barje in 2010, our knowledge of its distribution has increased extensively. This survey confirms that it is not limited only to the Na blateh (IX) and Log (VII) areas, but is also present in adjunct woodlands in the northern part of Ljubljansko barje and in two isolated areas approximately 10 km away in the eastern and south-eastern part of Ljubljansko Barje – Jelšje (V) and Dremavščica (IV).

Based on egg clutch counts, Ljubljansko barje could hold more than 19,500 R. arvalis adults. For comparison, the entire Ecologically important area Mura - Radmožanci (EPO Mura -ID. Nr. 42100) (185 km²) contains only five large breeding areas with more than 500 clutches: Polanski log (~1,800), Muriša (~700), Bobri - Spodnja Bistrica (~600), Bunčani (~800) and Petanjci. The latter location used to hold the biggest R. arvalis population in north-eastern Slovenia - in 2006, more than 1,700 and in 2008 more than 2,800 clutches had been counted there, while herpetological surveys in 2013 and 2014 reported no more than 50 clutches counted here (unpublished data). As follows, Ljubljansko barje holds the largest and best preserved R. arvalis population in Slovenia. However, data presented here can serve only as a rough quide, as the majority of the River Mura floodplain has never been systematically investigated for amphibians and could still hold some yet unrecorded larger populations of R. arvalis. Our field work has revealed that R. arvalis distribution in central Slovenia is also closely related to the alluvial alder forest fragments at Ljubljansko barje. Indeed, all the findings were located no more than 500 m from the closest woodland, R. arvalis is nearly always associated with deciduous forest in Slovenia and adjunct countries, and usually chooses its breeding sites within or adjacent to the woodland (Pobolišaj et al. 2008). Of all the breeding aggregations, the Log alder forest (VII), within the northern cluster (A), represents the most important breeding area for R. arvalis at the national scale, which can be ascribed to the large number of man-made water bodies in this relatively extensive woodland habitat. These depressions of different sizes and depths used to be part of a clay pit. Today, they represent the only larger system of permanent lentic waters in the wider area of the northern part of Ljubljansko barje. While the area east of the municipal landfill site contains many small breeding sites with a couple of clutches in each, the southern area contains only two such sites. Even though the area south of the municipal landfill site contains a favourable share of

woodlands, it is not an appropriate breeding ground for *R. arvalis*, as its forest is more fragmented by meadows and intensive agricultural land. In addition, this area is somewhat elevated and contains only a few scarcely distributed smaller depressions, which hold water for a limited period of time. The area east of the landfill site depicts a special situation. It used to be covered with a rather large share of woodland, which was completely cut down a few years before the survey. As most of the depressions are filled with refuse, only smaller breeding sites were recorded. In general, Ljubljansko barje holds only a few forest habitats outside the northern cluster (A) suitable and large enough to sustain *R. arvalis*.

The entire *R. arvalis* distribution lies within the borders of Ljubljansko barje Nature Park and, furthermore, the majority of findings fall within SCI SI3000271 Ljubljansko barje Natura 2000 site. However, fragmentation and habitat destruction due to urbanization still present a major threat to *R. arvalis* in this area. Even though the forest habitats in the northern cluster (A) can be qualified as good, the possibility of local extinction remains high, as there are no other suitable habitats in the vicinity (Fig. 3). Moreover, the northern cluster lies in the immediate proximity of the capital city and the municipal landfill site. The planned expansion of the landfill will most probably separate the eastern and western breeding sites and seriously affect this sub-population. In addition, this area is under strong influence of human recreational activities. Numerous visitors frequent this site, especially during the spawning time, when they come to observe the »blue frogs«; they often approach the animals up close and frighten them.

The origin of *R. arvalis* in central Slovenia

Extensive search of historical literature on natural history and faunistics of Ljubljansko barje revealed only one vague record that could be related to *R. arvalis* presence of in this part of Slovenia. In his description of Slovenian batrachofauna, Bevk (1951) stated that inhabitants of Ljubljansko barje know this species under the common name »plavček«, meaning the blue one; while in other parts of Slovenia this species was at that time known under the common name »barska žaba«, meaning the moor frog. However, Bevk does not clarify any further whether this species was indeed present here. Nevertheless, the high ecological densities and the presence of at least two clusters separated by almost ten kilometres of inappropriate habitats contradict the hypothesis of man-mediated origin. For such a scenario to be plausible, it is at least highly unlikely that the translocation was mediated in recent decades. In addition, some unconfirmed and possible sightings of *R. arvalis* have been made at Ljubljansko barje in previous decades.

The presence of *R. arvalis* at Ljubljansko barje is more likely a result of natural colonization. Assuming the origin of *R. arvalis* in central Slovenia is not man-mediated, two mutually non-exclusive hypotheses can be put forward to explain the presence of this species in central Slovenia. Isolated populations from southern Norway and by the Sea of Azov and fossil records bear witness to the species' broader historical distribution at the end of the Pleistocene (Roček & Šandera 2008, Kuzmin et al. 2009). A similar course of events can also explain the presence of this amphibian at Ljubljansko barje. *R. arvalis* probably colonized central Slovenia after the last glacial maximum (about 14,000 years ago). This amphibian could have reached central Slovenia from its putative source population in south-eastern

Slovenia (i) from the east along the River Sava and/or (ii) from the south over flooded karst poljes (Radensko polje, Dobrepolje) and along the River Krka. Both scenarios predict an existence of a period when potential habitats in southern Slovenia were more abundant and the species had a more extensive distribution along at least one of these two possible routes.

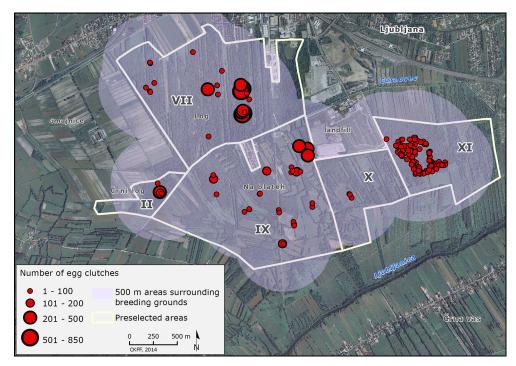


Figure 3. Distribution and abundance of *R. arvalis* breeding grounds in the immediate vicinity of Ljubljana. All clutches that could belong to *R. arvalis* were considered, including those that could not be identified beyond any doubt and were assigned to the *R. arvalis/temporaria* group. Dots represent either individual clutches or aggregations thereof rather than separate breeding grounds; a single breeding ground could thus be represented by more than one dot. Shaded area represents *R. arvalis* home range, if we take into the consideration that adults migrate less than 500 m (Günther & Nabrowsky 1996).

Slika 3. Razporejenost in razpršenost seštevka mrestov barskih žab in nadrejene skupine *R. arvalis/temporaria* v neposredni bližini mesta Ljubljana. Pike ponazarjajo najdišča skupine (blazine) mrestov ali posameznih mrestov in ne posameznega mrestišča (eno mrestišče lahko ponazarja več pik). Osenčeni pas okoli mrestišč ponazarja predvideni domači okoliš barskih žab, če upoštevamo, da se odrasli osebki ne selijo več kot 500 m (Günther & Nabrowsky 1996).

Because variation of colour morphs is due to genetic rather than environmental factors, the occurrence of forms can also shed some light on the origin of *R. arvalis* in central Slovenia. Prevalence of the unicolor morph indicates a connection with the populations from the River Sava, as this is the most dominant form in Croatia (Fog 2008a). Unfortunately, no data are available for populations from south-eastern Slovenia, while based on the photographs deposited at the Centre for Cartography of Fauna and Flora database (CKFF 2014), striata morph is also common elsewhere in north-eastern Slovenia and not just in Rački ribniki Nature Park.

Nevertheless, variation in distribution of forms is not only due to non-selective forces, but is also connected with adaptations to different habitats. For example, non-striata morphs thrive better in woody habitats, while the striata morph is better suited for sandy soil and less eutrophic habitats in general. This inclination towards more oligotrophic habitats is probably also related to higher sensitivity for competition from other arvalis tadpoles in striata morph, as more tadpoles in a breeding ground inevitably leads to eutrophication of water (Fog 2008a, b). As follows, intraspecific competition could also explain lower local densities of *R. arvalis* in north-eastern Slovenia, where striata form is common.

Furthermore, the individuals from Ljubljansko barje are in general bigger and have relatively longer legs, when compared to those from north-eastern Slovenia, Hungary, Romania, Czech Republic, Poland and Germany (Dély 1964, Günther & Nabrowsky 1996, Babik & Rafínski 2000, Poboljšaj et al. 2008). On the basis of their relatively longer legs and larger bodies, frogs from the Pannonian and Sava Basins were traditionally described as the *R. a. wolterstorffi* subspecies, which is supposed to have the SVL/TL ratio below 1.75 (Dély 1964); at Ljubljansko barje, the ratio is 1.65. However, Babik & Rafínski (2000) found that morphometric characters, including SVL/TL ratio, are not reliable for discriminating between the subspecies, while Babik et al. (2004) even showed that molecular data do not reflect the partition into two subspecies.

The case of *R. arvalis* at Ljubljansko barje is a good example that this species can be overlooked in nature or mistaken for other brown frog species if one is not careful. Even though chances of finding possible relic populations of *R. arvalis* in the remaining suitable habitats along the two suggested corridors are small at best, future field surveys should not neglect these areas. Other potential areas that could hold yet unrecorded *R. arvalis* populations in Slovenia are along the Drava and Sotla Rivers. The contribution of the outlined scenarios could also be elucidated through the application of quickly evolving molecular markers such as microsatellites.

Povzetek

Barska žaba (*Rana arvalis*) je evrazijska nižinska vrsta, za katero je do nedavnega veljalo, da je razširjena zgolj v severovzhodni in jugovzhodni Sloveniji. Pojavljanje vrste na Ljubljanskem barju je bilo prvič zanesljivo potrjeno spomladi leta 2010, ko sta avtorja 25. marca na območju »Na blateh« našla nekaj deset barskih žab, vključno z modro obarvanimi samci. Le nekaj dni kasneje je Davorin Tome odkril veliko mrestišče barskih žab v poplavnem jelševem gozdu »Log« na severnem delu Barja. Za boljši vpogled v razširjenost te vrste je Krajinski park Ljubljansko barje pri Centru za kartografijo favne in flore naročil obširno raziskavo, katere glavni namen je bil opredelitev najpomembnejših območij za varstvo barske žabe na območju parka.

Na podlagi terenskih raziskav iz marca in aprila 2011 in drugih raziskav podajamo karto razširjenosti in oceno številčnosti te vrste na Ljubljanskem barju. Barske žabe smo z gotovostjo potrdili na šestih raziskovalnih območjih (Črni Log, Jelšje, Log, Na blateh, Rakova Jelša južno od odlagališča in Rakova Jelša vzhodno od odlagališča), medtem ko smo na območju »Dremavščice« zabeležili le najdbe, ki jih nismo mogli določiti do nivoja vrste. Iz tega razloga »Dremavščico« obravnavamo zgolj kot potencialno območje za barsko žabo. Pred začetkom terenskega dela smo na podlagi kartiranih habitatnih tipov izbrali dvanajst

območij, ki bi lahko gostile to dvoživko. Velikost populacije smo ocenili s štetjem mrestov na mrestišču. Ocenili smo, da na Ljubljanskem barju živi več kot 19.000 odraslih barskih žab, ki se geografsko razporejajo v dve območji. Na širšem območju gozdnega kompleksa Log, v neposredni bližini mesta in deponije, živi in se razmnožuje več kot 96 % barskih žab v osrednji Sloveniji, medtem ko preostale najdbe prihajajo iz jugovzhodnega dela barja, predvsem iz območja Jelšje, morda pa tudi iz območja Dremavščice. Med terenskimi raziskavami v letu 2011 smo poleg mrestov barskih žab prešteli še 2.019 mrestov rosnice (*Rana dalmatina*) in 334 mrestov sekulj (*Rana temporaria*).

Vsa območja z barskimi žabami so v celoti del Krajinskega parka Ljubljansko barje in večina jih je del SCI-območja Ljubljansko barje (SI3000271). Barska žaba je kot strogo zavarovana vrsta uvrščena na Prilogo IV Direktive o habitatih in je tako varstvo vrste in njenih habitatov z Naturo 2000 možno vsaj posredno. Dolgoročni obstoj vrste na Ljubljanskem barju je odvisen predvsem od načina upravljanja območij barske žabe v prihodnje, pri čemer je največja nevarnost načrtovana širitev deponije.

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Literatura

- Babik W., Branicki W., Sandera M., Litvinchuk S., Borkin L.J. Irwin J.T., Rafínski J. (2004): Mitochondrial phylogeography of the moor frog, *Rana arvalis*. Mol. Ecol. 13(6): 1469-1480.
- Babik W., Rafínski J. (2000): Morphometric differentiation of the moor frog (*Rana arvalis* Nilss.) in Central Europe. J. Zool. Syst. Evol. Research 38: 239-247.
- Bonk M., Bury S., Hofman S., Szymura J.M., Pabijan M. (2012): A reassessment of the northeastern distribution of *Rana dalmatina* (Bonaparte, 1840). Herp. Notes 5: 345–354.
- Bevk S. (1951): Naše žabe. Ribič 10: 103-105.
- Cipot M. (2011): Inventarizacija plavčka (*Rana arvalis*) v Krajinskem parku Ljubljansko barje. Center za kartografijo favne in flore, Miklavž na Dravskem polju, 26 pp.
- Dély O.G. (1964): Contribution a l'etude systematique, zoogeographique et genetique de *Rana arvalis arvalis Nilss* et *Rana arvalis wolterstorffi* Fejérváry. Acta Zool. Acad. Scient. Hungaricae 10(3–4): 309-361.
- Fog K. (2008a): Colour pattern morphs of the moor frog (*Rana arvalis*) in Europe. In: Glandt D., Jehle R. (Eds.), Der Moorfrosch/The Moor Frog, Der Zeitschrift Für Feldherpetologie, Supplement 13. Laurenti-Verlag, Bielefeld, pp. 35-56.

- Fog K. (2008b): Colour pattern morphs of the moor frog (*Rana arvalis*) in Denmark. In: Glandt D., Jehle R. (Eds.), Der Moorfrosch/The Moor Frog, Der Zeitschrift Für Feldherpetologie, Supplement 13. Laurenti-Verlag, Bielefeld, pp. 37-76.
- Glandt D. (2008): Methoden der Beobachtung und Bestandserfassung von Moorfroschen (*Rana arvalis*) als Grundlage fur Schutzmafinahmen/Methods of observation and population census of moor frogs (*Rana arvalis*) as a basis of conservation measures. In: Glandt D., Jehle R. (Eds.), Der Moorfrosch/The Moor Frog, Der Zeitschrift Für Feldherpetologie, Supplement 13. Laurenti-Verlag, Bielefeld, pp. 431-442.
- Brenčič M. (2008): Vode Ljubljanskega barja in njegovega obrobja. In: Pavšič J. (Ed.), Ljubljansko barje: neživi svet, rastlinstvo, živalstvo, zgodovina in naravovarstvo. Društvo Slovenska matica, Ljubljana, pp. 17-39.
- Govedič M., Vamberger M., Sopotnik M., Cipot M., Lešnik A., Šalamun A., Poboljšaj K. (2009): Inventarizacija močvirske sklednice, hribskega urha in velikega pupka na Ljubljanskem barju. Center za kartografijo favne in flore, Miklavž na Dravskem polju, 62 pp.
- Grillitsch H., Cabela A. (2008): Verbreitung und Lebensraumansprüche des Moorfrosches (*Rana arvalis*) in Österreich. In: Glandt D., Jehle R. (Eds.), Der Moorfrosch/The Moor Frog, Der Zeitschrift Für Feldherpetologie, Supplement 13. Laurenti-Verlag, Bielefeld, pp. 291-308.
- Günther R., Nabrowsky H. (1996): Moorfrosch *Rana arvalis* Nilsson, 1842. In: Günther R. (Ed.), Die Amphibien und Reptilien Deutschlands. Gustav Fischer Verlag Jena, Jena, pp. 364-388.
- Hamilton J.A., Eckert C.G. (2007): Population genetic consequences of geographic disjunction: a prairie plant isolated on Great Lakes alvars. Mol. Ecol. 16: 1649-1660.
- Hellbernd L. (1987): Zweijährige Untersuchungen am Laichplatz vom Moorfrosch (*Rana arvalis* NILSSON) und vom Grasfrosch (*Rana temporaria* L.). Beiheft der Schriftenreihe Naturschutz und Landschaftspflege in Niedersachsen 19: 119-130.
- Jelić D., Kuljerić M, Koren T., Treer D., Šalamon D., Lončar M., Podnar Lešić M., Janev Hutinec B., Bogdanović T., Mekinić S., Jelić K. (2012): Red book of amphibians and reptiles of Croatia. Ministry of Environmental and Nature Protection, State Institute for Nature Protection, Zagreb, pp. 54-61.
- Jovanović M. (2009): Amphibia and reptilia of Štoj Plain (Ulcinj, Montenegro). Bulletin of the Natural History Museum 2: 137-152.
- Kotarac M., Grobelnik V. (1999): Kartiranje habitatnih tipov na Ljubljanskem barju. Center za kartografijo favne in flore, Miklavž na Dravskem polju, 11 pp.
- Kuzmin S., Tarkhnishvili D., Ishchenko V., Tuniyev B., Beebee T., Anthony B., Schmidt B., Ogrodowczyk A., Ogielska M., Babik W., Vogrin M., Loman J., Cogalniceanu D., Kovács T., Kiss I. (2009): *Rana arvalis*. In: IUCN 2011. IUCN Red List of Threatened Species. Version 2013.1. www.iucnredlist.org [accessed on 16. 6. 2013]
- Krofel M. (2005): Record of an Alpine Salamander (*Salamandra atra*) on Menišija plateau, Central Slovenia. Nat. Slo. 7(1): 45.
- Nöllert A., Nöllert C. (1992): Die Amphibien Europas: Bestimmung, Gefahrdung, Schutz. Franckh-Kosmos, Stuttgart, 382 pp.

- Poboljšaj K., Cipot M., Lešnik A. (2008): Distribution and conservation status of the moor frog (*Rana arvalis*) in Slovenia. In: Glandt D., Jehle R. (Eds.), Der Moorfrosch/The Moor Frog, Der Zeitschrift Für Feldherpetologie, Supplement 13. Laurenti-Verlag, Bielefeld, pp. 317-328.
- Puky M., Schád P. (2008): Distribution and status of the moor frog (*Rana arvalis*) in Hungary. In: Glandt D., Jehle R. (Eds.), Der Moorfrosch/The Moor Frog, Der Zeitschrift Für Feldherpetologie, Supplement 13. Laurenti-Verlag, Bielefeld, pp. 309-316.
- Roček Z., Šandera M. (2008): Distribution of *Rana arvalis* in Europe: a historical perspective. In: Glandt D., Jehle R. (Eds.), Der Moorfrosch/The Moor Frog, Der Zeitschrift Für Feldherpetologie, Supplement 13. Laurenti-Verlag, Bielefeld, pp. 135-150.
- Stanković D., Delić T. (2012): Morphological evidence for the presence of the Danube Crested Newt, *Triturus dobrogicus* (Kiritzescu, 1903), in Slovenia. Nat. Slo. 14(1): 23-29.
- Stanković D., Cipot M. (2013): Distribution and status of *Rana arvalis* in Central Slovenia. In: 17th European Congress of Herpetology, Veszprém, Hungary, University of Pannonia (22.–27. 8. 2013). Programme & abstracts. Societas Europe Herpetologica, Hungarian Ornithological and Nature Conservation Society, University of Pannonia and Hungarian Natural History Museum, Budapest, Veszprém, p. 294.
- Stanković D., Poboljšaj K. (2013): New data on the distribution of the Italian agile frog *Rana latastei* Boulenger, 1879 in Slovenian Istra. Nat. Slo. 15(2): 51-55.
- Stanković D., Lužnik M., Poboljšaj K. (2014): Conservation and declines of amphibians in Slovenia. In: Heatwole H., Wilkinson J. W. (Eds.), Amphibian Biology, Status of Conservation and Decline of Amphibians: Eastern Hemisphere: Western Europe, Vol. 11, pt. 4, Pelagic Publishing, Exeter, pp. 41-52.
- Tvrtković N., Kletečki E. (2008): Distribution of *Rana arvalis* in Croatia with remarks on habitats and phenology. In: Glandt D., Jehle R. (Eds.), Der Moorfrosch/The Moor Frog, Der Zeitschrift Für Feldherpetologie, Supplement 13. Laurenti-Verlag, Bielefeld, pp. 329-336.
- CKFF (2014): Bioportal. Center za kartografijo favne in flore, Miklavž na Dravskem polju. http://www.bioportal.si/fotoarhiv.php [accessed on 16. 6. 2014]
- Ur. I. RS (2002): Pravilnik o uvrstitvi ogroženih rastlinskih in živalskih vrst v Rdeči seznam. Uradni list RS 12(82): 8893-8975.
- Ur. I. RS (2004a): Uredba o zavarovanih prostoživečih živalskih vrstah. Uradni list RS 14(46): 5963-6016.