

Short Communication

The status of a rare phylogeographic lineage of the Vulnerable European souslik *Spermophilus citellus*, endemic to central Macedonia

BORIS KRYŠTUFEK, PETER GLASNOVIĆ and SVETOZAR PETKOVSKI

Abstract The conversion of grasslands for agriculture has triggered a serious decline of the European ground squirrel or souslik *Spermophilus citellus*, categorized as Vulnerable on the IUCN Red List since 1996. The Jakupica phylogeographic lineage of central Macedonia is the smallest of the three major evolutionary lines of the European souslik. This lineage is an important reservoir of within-species diversity and should be regarded as an independent unit for conservation management purposes. It is endemic to Mount Jakupica, where it lives in mountain pastures at 1,500–2,250 m altitude. The total area occupied by sousliks (88.4 ha) is fragmented and 94% of individuals occur in four colonies. Densities (0.8–5.5 adults ha⁻¹) are lower than those reported elsewhere for the species, with the total population probably < 2,000 adults. One large colony, reportedly of c. 1,000 sousliks, was decimated in 2007 by a catastrophic fire and had still not recovered by 2010. A steady decline in livestock grazing, together with the predicted advance of the tree line as a consequence of climate change, will probably reduce the optimal habitat for the souslik and negatively affect population fitness. Monitoring needs to be implemented, at least for the largest colonies, to provide early warning of any declines.

Keywords Density, distance sampling, endemic, European souslik, fragmentation, Macedonia, rarity, *Spermophilus citellus*

The European ground squirrel or souslik *Spermophilus citellus* is the western-most representative of the Palaearctic ground squirrels. This robust rodent occurs mostly in short-grass meadows up to c. 2,000 m altitude and, although well adapted to habitat disturbance, is vulnerable to both the anthropogenic conversion of grasslands for agriculture (Ružič, 1979) and urbanization

(Hoffmann et al., 2003). Formerly considered a major crop pest, the European souslik is now listed in the EU Habitats Directive (Council Directive 2006/105/EC of 20 November 2006) and has been categorized as Vulnerable on the IUCN Red List since 1996 (Coroiu et al., 2008).

Phylogeographic assessment based on a complete mitochondrial cytochrome *b* gene has shown that the European souslik comprises three historically isolated, independently evolving lineages. To preserve within-species diversity these deeply divergent lines should be regarded as independent units for conservation management purposes (Kryštufek et al., 2009). Although the conservation needs of the northern lineage of the European souslik, which occupies the largest range, have attracted attention among EU member states (Matějů et al., 2010), the status of the smallest of the European souslik lineages, endemic to Mount Jakupica in the Republic of Macedonia, is virtually unknown. Here we focus on the distribution and population status of this lineage.

Mount Jakupica is dominated by volcanic and metamorphic bedrock with Mesozoic evaporites of limestone folded over basal rocks. The Mount Jakupica complex consists of four major ridges that converge at Solunska Glava, the highest peak (2,540 m). The climate is alpine, with strong altitudinal shifts in precipitation and temperature (Reed et al., 2004). The western part of the area is managed as the Jasen Game Reserve. Grasslands are utilized as summer pastures by villagers from the foothills; however, there are no permanent settlements above 1,000 m. Sheep and cattle grazing is widespread and during our surveys the area of Gorno Begovo (Fig. 1) and its immediate vicinity were grazed by c. 2,600 sheep, 300 cows and 50 horses.

We searched Mount Jakupica for souslik colonies during the first half of July in both 2006 and 2010. Juvenile sousliks were starting to be weaned at that time but were still obviously smaller than the adults. Information on the locations of souslik colonies was provided by rangers, shepherds and mountaineers. Each colony was mapped by walking around its periphery and recording the geographical position of souslik burrows at the margins. The surface areas of the colonies were calculated using *ArcGIS v. 9.1* (ESRI, Redlands, USA). The number of adult sousliks was estimated by line transect sampling (Buckland et al., 2001).

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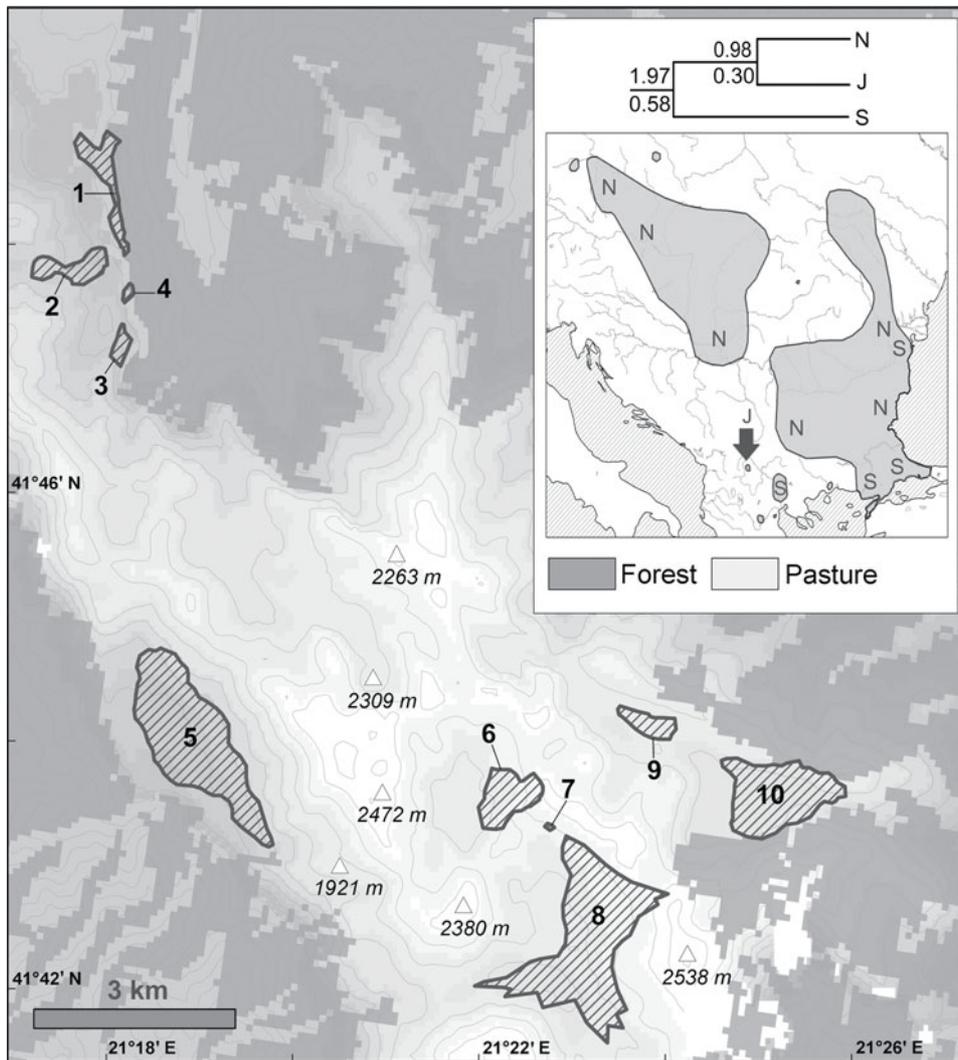


FIG. 1 The occurrence of the 10 colonies (numbers as in Table 1) of the European souslik *Spermophilus citellus* on Mount Jakupica, central Macedonia. The shaded areas on the inset show the likely range of the European souslik in south-eastern Europe and the distribution of phylogeographic lineages (N, Northern; S, Southern; J, Jakupica). Phylogenetic relations between lineages are summarized in a simplified phylogenetic tree; numbers denote mean genetic distance (in %, above branch) and time (million years ago, below branch) elapsed since the divergence of two lineages (modified from Kryštufek et al., 2009).

Because the population survey was incomplete in 2006 we used only the 2010 counts.

When animals occur singly the estimator of density (D) is $n/2L\mu_w$, where n is the number of detections, L is the total transect length, and μ_w is the estimated effective strip half-width. The constant μ_w was estimated for each colony from the distribution of distances (r) of observed animals perpendicular to the transect. The distance was calculated for each individual as $r = d \sin(\theta)$. The sighting distance (d) was measured from the observer to the souslik when first detected and the sighting angle (θ) between the transect line and the line of sight was measured to the nearest 5° . Two observers walked slowly ($< 1 \text{ km h}^{-1}$) within a souslik colony in a predetermined straight line, searching for sousliks using range-finding binoculars. A total of 30.71 km of transects were surveyed, of which only 10.76 km were used for density estimates. We omitted transects walked in weather conditions suboptimal for souslik activity (midday heat, rain and strong wind) as they yielded an obviously

lower number of observations. The density of sousliks was calculated using *Distance v. 6.0* (Thomas et al., 2009).

We recorded eight isolated colonies of sousliks, in areas of 20–280 ha, and two small groups, in 2.1 and 0.4 ha (Fig. 1, Table 1). These colonies were at altitudes of 1,500–2,250 m over c. 15 km of rugged mountain topography. The majority of souslik habitats were heavily grazed mountain pastures of short ($< 10 \text{ cm}$) grasses (*Poa*, *Nardus*, *Bromus*, *Festuca*) and herbs (*Plantago*, *Thymus*). Within 197 m perpendicular to the transects 419 sousliks were observed, c. 50% of which were seen within 33 m of the transect. The effective strip width was c. 60 m (range 59.8–63.7 m) for most of the estimates. Using *Distance* we estimated mean population densities in seven colonies to be 0.8–5.5 ha^{-1} (Table 1). We could not estimate densities in the other three colonies (locations 4, 5 and 7 in Fig. 1) because sightings were too few. High coefficients of variation (0.16–0.74), which imply wide confidence intervals, mean these estimates must be interpreted with caution. Published densities for European

TABLE 1 The 10 colonies of the European souslik *Spermophilus citellus* on Mount Jakupica (Fig. 1) surveyed in this study. Population densities were estimated in 2010 by line transect sampling. Density is given as adult sousliks per ha, with the coefficient of variation (CV) in parentheses.

Location	Coordinates	Altitude (m)	Area (ha)	Density (CV)	Population
1, Karlica	41°48 N, 21°18 E	1,500–1,697	80.0	5.49 (0.28)	439
2, Ajdučka Rupa	41°48 N, 21°17 E	1,682–1,781	30.8	0.80 (0.43)	25
3, Rada	41°47 N, 21°18 E	1,708–1,754	20.1	1.01 (0.26)	20
4, Unnamed	41°48 N, 21°18 E	2,154–2,172	2.1		< 10*
5, Boro Pole	41°44 N, 21°18 E	1,645–1,735	255.0		< 100*
6, Šilegarnik	41°43 N, 21°21 E	2,052–2,143	59.0	0.77 (0.37)	45
7, Unnamed	41°43 N, 21°22 E	2,112	0.4		< 10*
8, Solunsko Pole	41°43 N, 21°23 E	2,027–2,250	280.0	1.05 (0.74)	294
9, Vraca	41°44 N, 21°23 E	2,019–2,095	33.0	4.06 (0.40)	134
10, Gorno Begovo	41°43 N, 21°25 E	1,945–2,045	123.7	4.77 (0.16)	590
<i>Total</i>			884.1		< 1,667

*Tentative estimate (density could not be calculated; see text for details)

souslik populations are almost invariably higher than the densities estimated on Mount Jakupica; for example, 5–15 adults ha⁻¹ in high mountain pastures in eastern Serbia (Ružić-Petrov, 1950) and 6.5–110 adults ha⁻¹ in anthropogenic habitats in Austria (Hoffmann et al., 2003, 2008). The results from our study are possibly underestimates because it is unlikely that all sousliks were seen at zero distance from the line transect.

To check the reliability of our density estimates for the largest colony on Mount Jakupica, at Gorno Begovo, sousliks were counted on 0.25-ha plots in 2010. Each plot was observed with binoculars for at least 3 hours during the morning activity of the sousliks, with their burrows temporarily closed to avoid multiple counts of the same individuals. Care was taken to avoid recounting the same individuals on different plots. On 17 plots (total area = 4.25 ha) 1–10 adults were recorded (mean = 4.1 ± SD 2.76). These results suggest a mean souslik density of 16.5 ha⁻¹, c. 3.5-fold higher than for the entire colony. These 0.25-ha plots were located on the western side of the colony where the number of sousliks was greatest.

Estimates for the total number of sousliks were 20–590 per colony (Table 1). The total area of the seven colonies (626.6 ha) for which densities could be estimated therefore contained c. 1,547 adult sousliks. The majority of these (c. 94%) were in four colonies (Gorno Begovo, Karlica, Solunsko Pole and Vraca) in two areas. Tentative population estimates for the other three colonies do not significantly modify the estimated total (Table 1). These data suggest that the overall number of sousliks within the Jakupica phylogeographic lineage is most likely < 2,000 adults.

Boro Pole, with the second largest colony (Table 1), was only surveyed in 2010. Because sousliks were scarce there, with none seen < 200 m from the transect, the density could not be estimated. We assume, from a scattering of occupied burrows, that the total number of sousliks in this colony is < 100. Rangers employed by the Jasen Game Reserve

indicated that the low density is the result of a catastrophic forest fire in 2007 that also burned the meadow. Sheep grazing was abandoned in Boro Pole c. 40 years ago when the Reserve management began to utilize hay from this pasture as supplementary feed for game. As a result, grasses (*Festuca*, *Bromus* and *Stipa*) now grow tall, although sparsely, and the presence of a relatively large number of umbellifers (species of *Apiaceae*) on the margin of the area suggests that it is overgrown. Rangers claimed that prior to the 2007 fire sousliks were as abundant in Boro Pole as they were in Karlica in 2010. This suggests there were formerly c. 1,000 sousliks in Boro Pole, about half the total estimate for the Jakupica lineage in 2010.

Our results show that the Jakupica lineage of the European souslik occupies a small, fragmented area. Colony densities are lower than those reported elsewhere for the species and the total population is probably < 2,000 adults. Although souslik colonies in remote Mount Jakupica are not threatened by large-scale agricultural activities they are likely to become increasingly scarce. Livestock grazing is declining on Mount Jakupica (e.g. the number of sheep raised by villagers from Gorno Jabolchiste, in the eastern foothills of Mount Jakupica, had declined from 20,000 in 1980s to c. 10,000 at the time of our survey; pers. comms, local villagers). Fewer sheep means taller grass and more shrubs, which in turn increase the risk of high-intensity fires. In addition, the predicted advance of the tree line in response to climate change (Grace et al., 2002) will most likely cause a further reduction in the area of habitat optimal for the souslik. The largest souslik colonies (Gorno Begovo and Karlica) are already fringed on one side by forest (Fig. 1).

A decline in souslik populations, together with increasing fragmentation, is likely to have a negative impact on fitness. In the small fragmented populations of the European souslik genetic drift could outpace the role of balancing selection and result in inbreeding depression (Řičanová et al., 2011). A decline in fitness is therefore the

most likely scenario for the Jakupica lineage. Monitoring needs to be implemented, at least for the largest colonies, to provide early warning of any declines. The management plan for Jasen Reserve for 2012–2021 (which, at the time of writing, has still not been approved) stipulates an action plan and monitoring for souslik colonies 6–10 (Fig. 1) but not for the Karlica colony, which had the second largest estimated population in 2010. The implementation of these measures will be the responsibility of the Reserve and the Ministry of Environment and Physical Planning.

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Biographical sketches

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