

## GIS analysis to assess the groundwater habitat pollution of black proteus

### GIS-analiza za oceno onesnaženosti podzemne vode v habitatu črnega proteusa

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Karst areas are among the world's most vulnerable landscapes to environmental impacts (Veni 2004). Most of pollution in karst areas is due to unsustainable anthropogenic activities (intensive agriculture and industry, unregulated urbanization) that are reflected in the decline of subterranean biodiversity and in the loss of drinking water resources. Slovenia may be particularly vulnerable in this respect, as it is a prime hotspot of subterranean biodiversity (Culver & Sket 2000), and its ground waters constitute the drinking water supply for 97% of its residents (Kranjc et al. 2009). Slovenia is thus facing urgent conservation challenges, and among the critically affected organisms is *Proteus anguinus*, a global symbol of subterranean biodiversity.

Since the 1986 discovery of a unique, darkly pigmented population of proteus, the black proteus (*Proteus anguinus parkelj*) within a single cave system of less than 30 km<sup>2</sup> in Southeast Slovenia, the protection of karst underground habitats has become even more important. This most distinct and rare of all proteus populations is highly endangered; because of its extremely limited distribution, even a small local pollution could have a devastating impact on the entire population and

could also destroy the overlapping regional drinking water supply.

The main threats for the black proteus relate to agriculture, particularly the overuse of pesticides and fertilizers on farming land and vineyards, and after 2009, the production of biogas slurry distributed as free fertilizer (Hudoklin 2011). Industry's side products, likewise, pollute the groundwater, e.g. foundry sand contaminated with heavy metals dumped into a doline 700 m from the Jelševnik Springs (a local system comprising the permanent spring, Jezero, and two groups of temporary springs, Na trati, and Jamnice), harbouring the black proteus. After alarming accumulation of zinc in the tissues of the black proteus from Jelševnik Springs (i.e., Na trati 2) was shown (Bulog et al. 2002), the washing out of the waste disposal was minimized by a clay cover; however, this remained *in situ* as a long-lasting threat. Furthermore, villages in the catchment area lack a central sewage system and continue to use primitive individual household septic tanks, from which waste waters enter the karst groundwater. Researchers have detected increased levels of phosphates and nitrates in the Jelševnik Springs (i.e., Na trati 1 and 2; Bulog et al. 2002).

Recently, detection of proteus environmental DNA (eDNA) traces (Gorički et al. 2017) has helped obtain a more precise information on the distribution of the black proteus. Although the number of its known localities was doubled, the expected distribution area of the black proteus (i.e., the area between confirmed localities of the black proteus, without the plausible continuation under the high plateau of Kočevski Rog) is now estimated at less than 3 km<sup>2</sup> (Gorički et al. 2017), extending into the near shallow karst plain of the Črnomelj area. Unfortunately, this newly discovered part of the black proteus habitat lies under the most intensive agriculture areas (about 2/3 of land use type above the black proteus habitat comprise fragmented cultivated fields and vineyards), adding to our inference that the black proteus is severely endangered.

Based on proteus eDNA survey, we employed GIS both to draw the new distribution of the black proteus as well as to test whether any long-term pollution trends within its habitat may be assessed despite the general scarcity of prior data on groundwater quality. Recent data are available at the Dobljčica Spring (drinking water control) and

the Jelševnik Springs (monitoring of the black proteus habitat since 2000; Bizjak Mali & Bulog 2016). We additionally employed historic data on physical and chemical parameters of groundwater in Bela krajina from an extensive survey of the Karst Research Institute ZRC SAZU (Habič et al. 1990), which also led to the unexpected discovery of the black proteus.

Limited by only a basic set of pollutants measured by Habič et al. (1990), we analyzed concentrations of nitrates and orthophosphates in groundwater in order to reconstruct their quantity during the time of black proteus discovery, and thus to assess the trends in the threat to the black proteus habitat (1987 vs. 2014).

We employed the IDW (Inverse Distance Weight) interpolation tool from ArcGIS 10.3.1 (Esri 2015) to visualize the distribution of nitrates and orthophosphates in 1987 within the expected distribution area of the black proteus and the adjacent white proteus localities (i.e., measurements belong to 7 karst springs, of which 5 are already confirmed localities of proteus; hereafter area of interest, AOI). Unfortunately, most of these springs were not included in subsequent investigations; hence we were able to illustrate pollution trends in the 1987–2014 period only for two of the springs: Dobljčica and Jelševnik (i.e., Jezero; ARSO 2014). Three main categories were used in IDW analysis to assess the pollution with nitrates and orthophosphates in AOI, defined as: *low* ( $< 2.22 \text{ mg NO}_3^- / \text{l}$ ,  $< 0.045 \text{ mg PO}_4^{3-} / \text{l}$ ), *medium* ( $2.22\text{--}13.3 \text{ mg NO}_3^- / \text{l}$ ;  $0.045\text{--}0.25 \text{ mg PO}_4^{3-} / \text{l}$ ) and *high* ( $> 13.3 \text{ mg NO}_3^- / \text{l}$ ;  $> 0.25 \text{ mg PO}_4^{3-} / \text{l}$ ) (USGS 1999).

The map of interpolated values of pollution in 1987 (Fig. 1) shows medium concentrations of nitrates (Fig. 1a) and orthophosphates (Fig. 1b) in the majority of the AOI, while high concentrations of nitrates are exhibited only in the buffer areas of two springs (Obršec and Pački Brežiček – both verified localities of the black or white proteus, respectively). Low concentrations of nitrates are present in a small area around the Dobljčica Spring, while low concentrations of orthophosphates covered less than half of the expected habitat of the black proteus.

The comparison of pollution in two analyzed springs (Dobljčica and Jelševnik/Jezero) in 1987 vs. 2014 revealed a concerning growing trend for both

nitrates and orthophosphates (Tab. 1). The critical increases of nitrates (280% for Dobljčica Spring and 272% for Jelševnik Springs/Jezero) and orthophosphates (150% for Dobljčica Spring and 111% for Jelševnik Springs/Jezero) implicate intensified use of fertilizers in agriculture and unregulated sewage disposal in the settlements within AOI.

The results of this pilot GIS analysis indicate an urgent need for implementation of a monitoring scheme for the black proteus and its habitat. Immediate action is needed to reverse the pollution trends and to prevent population declines. Moreover, the highly increased levels of nitrates and orthophosphates in the Dobljčica Spring do not only affect subterranean biodiversity and the unique black proteus, but also raise public health concerns.

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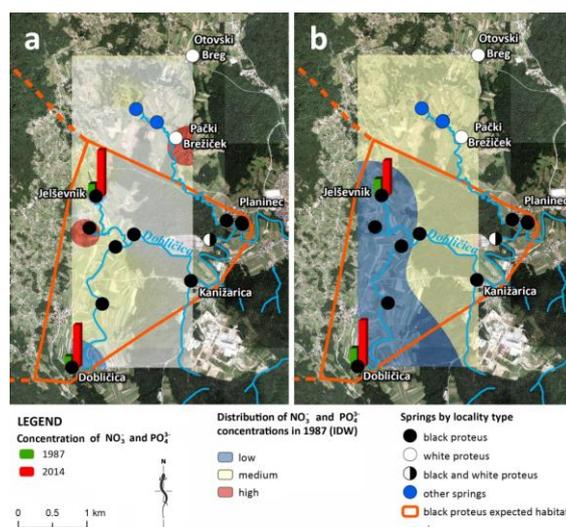
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**Table 1.** Concentrations of nitrates and orthophosphates from Dobljica and Jelševnik Springs (i.e., Jezero) in 1987 and 2014, with their corresponding percentage of increase.

**Tabela 1.** Koncentracije nitrata in ortofosfatov iz izvira Dobljice ter izvira v Jelševniku (Jezero) v letu 1987 in 2014, z odgovarjajočim odstotkom povečanja.

Spring	Nitrates mg NO <sub>3</sub> <sup>-</sup> /l		NO <sub>3</sub> <sup>-</sup> increase 1987 vs. 2014 (%)	Orthophosphates mg PO <sub>4</sub> <sup>3-</sup> /l		PO <sub>4</sub> <sup>3-</sup> increase 1987 vs. 2014 (%)
	1987	2014		1987	2014	
Dobljica	0.9	3.42	280	0.01	0.025	150
Jelševnik (Jezero)	1	3.72	272	0.009	0.019	111



**Figure 1.** Pollution of *Proteus* habitat indicated by the distribution of interpolated concentration ranges of pollutants (NO<sub>3</sub><sup>-</sup> (a) and PO<sub>4</sub><sup>3-</sup> (b)) for the year 1987 within the AOI in Bela krajina, Slovenia; the bar charts show the concentration of pollutant in analyzed springs for 1987 and 2014.

**Slika 1.** Onesnaženost habitata človeške ribice, prikazana z interpolacijo porazdelitve koncentracije onesnažil (NO<sub>3</sub><sup>-</sup> (a) in PO<sub>4</sub><sup>3-</sup> (b)) za leto 1987 na preučevanem območju Bele krajine; stolpci prikazujejo koncentracije onesnažil v analiziranih izvirihih v letu 1987 in 2014.