

Morphological evidence for the presence of the Danube Crested Newt, *Triturus dobrogicus* (Kiritzescu, 1903), in Slovenia

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Abstract. We offer preliminary reports on the presence of the Danube Crested Newt, *Triturus dobrogicus*, in Slovenia. This species reaches its western border of distribution in Hungary, Austria and Croatia, but until recently it was not known from Slovenia. Here we report on the first record of this species in Slovenia, found in Dolinsko polje of the Prekmurje region during the herpetological surveys carried out in the spring of 2009 and 2010. The species identity was determined by coloration, body shape and the Wolterstorff Index values. Only with additional genetic analysis could we answer if this is a pure population or whether hybridisation with *T. carnifex* is present.

Key words: *Triturus dobrogicus*, Dolinsko polje, Slovenia, Wolterstorff Index

Izveček. PRVA NAJDBA PANONSKEGA PUPKA, TRITURUS DOBROGICUS (KIRITZESCU, 1903), V SLOVENIJI – V bližini Slovenije je panonski pupek poznan iz Avstrije, Hrvaške in Madžarske. Spomladi 2009 in 2010 smo na Dolinskem polju v Prekmurju na podlagi vzorca obarvanja, oblike telesa in Wolterstorffovega indeksa prvič določili in zabeležili to vrsto v Sloveniji. V prispevku so predstavljeni preliminarni rezultati, medtem ko bodo za ugotovitev, ali je populacija iz Dolinskega polja čista ali pa se križa z velikim pupkom, potrebne nadaljnje genetske analize.

Ključne besede: *Triturus dobrogicus*, Dolinsko polje, Slovenija, Wolterstorffov indeks

The Crested Newt (*Triturus cristatus*) superspecies consists of closely related parapatric taxa, which are characterized by narrow hybrid zones and share a complex evolutionary history (Arntzen et al. 2007, Espregueira Themudo et al. 2009). The large-bodied Crested Newts differ both in genetic and morphological features (Wallis & Arntzen 1989). The most reliable features for morphological distinction of the taxa are the number of rib bearing vertebrae (RBV) and the Wolterstorff index (WI): length of forelimb \times 100 / inter-limb length (Arntzen & Wallis 1999). Ivanović et al. (2007) showed that the size and the shape of the head are not reliable characters for this group, especially as the skull of *T. dobrogicus* undergoes significant post-metamorphic changes.

In Slovenia, the only Crested Newt species recorded so far is *T. carnifex* (Poboljšaj 2003, Veenvliet & Kus Veenvliet 2008). *T. dobrogicus* is spread throughout the Pannonian Basin and, after a gap in the Iron Gates area, along the Danube floodplain and its tributaries, where it is found in oxbows, along river margins and other permanent water bodies, and can coexist with fish. It was recorded in close vicinity of the Slovenian border, both on the Austrian and Hungarian sides (Mayer 2001, Puky et al. 2005); in Croatia, the nearest records were made near the confluence of the Mura and Drava Rivers and south-east of Zagreb (Kalezić et al. 1990, Trvtković 2006). However, there are examples in the literature where the presence of *T. dobrogicus* × *T. carnifex* hybrids has either been speculated or it could be concluded so from the data. First, »The IUCN Red List of Threatened Species« (Arntzen et al. 2008) states that individual hybrids of *T. carnifex* × *T. dobrogicus* occur in north-eastern Slovenia (Mura River). Secondly, Kalezić et al. (1990), Crnobrnja-Isailović et al. (1997) and Kalezić et al. (1997) found a Crested Newt population in vicinity of Radenci (Turjanci) in north-eastern Slovenia that showed both RBV number (Crnobrnja-Isailović et al. 1997) and WI values (Kalezić et al. 1990) that could be characteristic of either *T. dobrogicus* × *T. carnifex* hybrids or *T. cristatus* (Arntzen & Wallis 1999). However, it seems most improbable that the latter species would have been encountered in this area (Espregueira Themudo et al. 2009). Specifically for this population, the WI values for females span from just above the diagnostic threshold for *T. dobrogicus*, across the range characteristic for *T. cristatus* and into the lower half of the variation for *T. carnifex*, while WI values for males were well above the diagnostic threshold for *T. dobrogicus* and mostly in the range characteristic of *T. cristatus* and *T. carnifex* (the WI range for *T. dobrogicus* is < 54.0 for males and < 46.2 for females and for *T. carnifex* 63.7 – 67.09 for males and 53.9 – 59.19 for females - Arntzen & Wallis, 1999). Also, Kalezić et al. (1997) studied both qualitative traits and morphometric characteristics of Crested Newts in the western Balkans and identified Radenci (Turjanci) populations as *T. carnifex*; they did not find significant differences in regard to other studied *T. carnifex* populations. Although morphological characters, especially when backcrosses are involved, are not necessarily intermediate in *Triturus* hybrids (Kalezić et al. 1997, Arntzen & Wallis 1999), this appears to be the case at least with the RBV count in *T. carnifex* × *T. dobrogicus* hybrid populations in the adjacent areas (Vedropolje, Croatia and Donja Čadjavići, Bosna and Herzegovina; Arntzen & Wallis 1999). Furthermore, there are known hybrid and pure *T. dobrogicus* populations in Austria near to Radenci (Mayer 2001). Therefore, the deviation of RBV count and the WI in Radenci population is either a result of hybridisation or can be attributed to intraspecific biological variation. Similar conclusions can be made for Žumberak and Slavonski Brod populations from Croatia (Kalezić et al. 1990, Crnobrnja-Isailović et al. 1997, Kalezić et al. 1997).

During the herpetology surveys of the Prekmurje region, Slovenia, in the spring of 2009 and 2010, *T. dobrogicus*-like specimens (Fig. 1) were recorded for the first time. The research area was limited to the eastern part of the Dolinsko polje, consisting of a floodplain forest, Murska Šuma, and adjacent swampy meadows. In this area, there are a few large and a number of smaller and shallower oxbows (Fig. 2). The area is typical sparsely inhabited Pannonian lowland. Newts were captured with dip nets and floating water traps and with torches at night. All specimens were released to the original sites of capture after measurements were taken. Material was identified based on chromatic and morphological characters indicated, mainly upon WI; inter-limb distance was measured excluding the extremities as recommended by Arntzen & Wallis (1994). The newts were anesthetized with MS 222 diluted to 0.3 % (Sigma-Aldrich, Vienna), photographed and measured on the site with 0.1 mm precision vernier calliper.



Figure 1. Male (left) and female (right) specimens of the Danube Crested Newt, *T. dobrogicus*, from a small oxbow east of the Muriša oxbow (Photo: Teo Delić).

Slika 1. Samec (levo) in samica (desno) panonskega pupka, *T. dobrogicus*, iz manjše mrtvice vzhodno od Muriše (Foto: Teo Delić).



Figure 2. A typical habitat of the Danube Crested Newt, *T. dobrogicus* (Photo: David Stanković).

Slika 2. Habitat, značilen za panonskega pupka *T. dobrogicus* (Foto: David Stanković).

The body shape and the chromatic features taken into account were more characteristic of *T. dobrogicus* than *T. carnifex*. All specimens were lean built with short legs and had white-stippled sides, black to dark brown gular coloration with angular white spots and deep orange ventral surface with many small to medium sized sharply delineated black roundish black spots, which showed a tendency to unite longitudinally. By way of comparison, *T. carnifex* is a larger medium built newt with large legs and usually has yellow ventral colouration with few large roundish blurred and ill-defined blotches and spots (Arntzen & Wallis, 1999, Jehle et al. 2011). Measurements of WI and localities are presented in Tab. 1. In total, fifteen adult newt specimens from five sampling localities were collected. For thirteen specimens, the WI is within the known range of variation for *T. dobrogicus*, while two specimens had WI values just above and were identified as possible *T. carnifex* × *T. dobrogicus* hybrids; none of the individuals was classified as *T. carnifex*. All newts were either captured in floodplain woodland or oxbows, where they coexist with fish. *T. carnifex* populations in Slovenia rarely coexist with fish and are therefore almost never found in oxbows. *T. carnifex* populations from other parts of Prekmurje and the rest of Slovenia were studied quite intensely in the recent years in accordance with Natura 2000 implementation, and there are no reports on populations that were morphologically as distinct as this population from Dolinsko polje (Poboljšaj & Lešnik 2003, Cipot & Lešnik 2007, Lešnik & Cipot 2007).

Table 1. List of known localities inhabited by the Danube Crested Newt, *Triturus dobrogicus* (Tdob), and hybrids, *Triturus dobrogicus* × *Triturus carnifex* (Tdob x Tcor) from NE Slovenia with geographic coordinates, applied survey method and biometric data. WI diagnostic range for *T. dobrogicus* was taken from Arntzen & Wallis (1999).

Tabela 1. Seznam najdišč panonskega pupka, *Triturus dobrogicus* (Tdob), in križancev panonskega in velikega pupka, *Triturus dobrogicus* × *Triturus carnifex* (Tdob x Tcar) iz SV Slovenije z geografskimi koordinatami, metodo lova in biometričnimi podatki. Značilen razpon WI za *T. dobrogicus* je povzet po Arntzen & Wallis (1999).

Habitat type	Lat	Lon	Survey method	Sex	Length of forelimb [mm]	Inter-limb length [mm]	Wolter-storff index [%]	Determination based on WI
oxbow	46°28'56"	16°32'27"	water trap	juvenile	11	24	45,83	Tdob
oxbow	46°29'14"	16°32'6"	water trap	male	16,5	31,9	51,72	Tdob
oxbow	46°29'14"	16°32'6"	water trap	male	14,9	28,5	50,17	Tdob
oxbow	46°29'40"	16°32'6"	water trap	female	15,4	38,5	40	Tdob
forest	46°30'1"	16°30'49"	torching	male	16,1	32,4	49,69	Tdob
forest	46°30'0"	16°30'41"	torching	male	15,3	29,7	51,52	Tdob
forest	46°30'0"	16°30'41"	torching	female	19,3	45,1	42,8	Tdob
oxbow	46°28'37"	16°34'23"	water trap	male	17,3	32	54,06	Tdob x Tcar
oxbow	46°28'37"	16°34'23"	water trap	male	17,3	35,1	49	Tdob
oxbow	46°28'37"	16°34'23"	water trap	male	23,1	45,4	50,88	Tdob
oxbow	46°28'39	16°34'52"	water trap	female	18,8	44,7	42,06	Tdob
oxbow	46°28'39	16°34'52"	water trap	female	20	49,1	40,73	Tdob
oxbow	46°28'39	16°34'52"	water trap	female	19,3	41,6	46,39	Tdob x Tcar
oxbow	46°28'39	16°34'52"	water trap	female	15,5	40,4	38,37	Tdob
oxbow	46°28'39	16°34'52"	water trap	female	20	48,5	41,24	Tdob

Based on chromatic and morphological characters, geographical characteristics and habitat type, we can conclude that the range of *T. dobrogicus* also extends into Slovenia, at least into Dolinsko polje. Further morphological (RBV) and genetic analyses on mitochondrial and independent nuclear loci are required to undisputedly determine if this population is a pure *T. dobrogicus* population or whether hybridisation with *T. carnifex* is present.

The Danube Crested Newt is a strictly protected species under the Bern Convention and furthermore listed as globally Near Threatened by the IUCN (2008) with an additional comment of being close to qualifying for Vulnerable. In Slovenia, *T. dobrogicus* is protected by the Decree on Protected Wild Animal Species Annex II Chapter B. This chapter lists animals that are not considered to be indigenous to Slovenia and their habitats are protected by law only, if they appear or expand to Slovenia without human interference. This is clearly the case with *T. dobrogicus*. The absence of records for *T. dobrogicus* in Slovenia does not indicate recent colonization or introduction by humans, but is more likely to be a consequence of a high degree of similarity with *T. carnifex* and inappropriate survey methods for this species (dip nets). *T. dobrogicus* is also protected by the EU Habitats Directive, where it is listed on Annex II and therefore requires designation of Special Areas of Conservation. This means that it is protected by the Directive only in designated Nature 2000 areas, while *T. carnifex* is also protected by Annex IV, which additionally provides protection outside those areas. Protection of their hybrids presents an interesting question and cannot be addressed similarly to the protection of *Bombina bombina* × *B. variegata* hybrids, where both parent species are listed only on Annex IV.

The importance of the first record of *T. dobrogicus* in Slovenia is even greater as this species inhabits habitats that are different from those of *T. carnifex*, which leads to different requirements for its successful conservation, protection and management. Apart from genetic analysis, we have to proceed with further investigation of the distribution of this amphibian and also re-evaluation of previous studies to determine whether its presence has been overlooked elsewhere in the Slovenian lowlands. If genetic analysis reveals that Dolinsko polje population is a hybrid population, it should still be protected by more strict measures of conservation and considered as Annex II species.

Povzetek

Spomladi leta 2009 in 2010 smo v Sloveniji prvič potrdili pojavljanje panonskega pupka (*Triturus doborogicus*). Panonski pupek je razširjen po Panonski nižini in poplavnem pasu Donave od Djerdjapa dolvodno, kjer naseljuje mrtvice, rečne rokave, struge počasi tekočih voda ter druga večja in trajnejša vodna telesa. V vodi se zadržuje dlje kot veliki pupek (*Triturus carnifex*) in pogosteje sobiva z ribami. Od velikega pupka, ki je v Sloveniji sicer splošno razširjen, ga ločimo po relativno krajši dolžini nog, vitkejšemu telesu, manjši velikosti, večjem številu reber in vzorcu obarvanosti. Prve osebkte te dvoživke smo našli v Murski Šumi in njeni bližini. Določili smo jih po vzorcu obarvanosti in obliki telesa - predvsem na podlagi Wolterstorffovega indeksa, to je razmerja med dolžino nog in dolžino trupa. Ker se veliki in donavski pupek lahko križata, zgoj na podlagi morfoloških podatkov ne moremo z gotovostjo trditi, da gre za populacijo čistih donavskih pupkov in da ne prihaja vsaj do občasnega križanja. Panonski pupek je zaščiten po Bernski konvenciji, na rdečem seznamu IUCN pa je označen kot potencialno ogrožena vrsta. V EU je zaščiten s Habitatno direktivo, kjer je uvrščen v Prilogo II, in nacionalno z Uredbo o zavarovanih prostoživečih vrstah. V tej Uredbi je uvrščen v Prilogo 2 Poglavlja B, to je seznam v Sloveniji alohtonih vrst, katerih habitati pa postanejo zaščiteni, če se ta vrsta k nam razširi brez človeške pomoči. Če se izkaže, da v populaciji prihaja do križanja z velikim pupkom, jo vendarle velja zaščiti v skladu s Prilogo II Habitatne direktive.

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