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New record of *Lissotriton vulgaris meridionalis* (Boulenger, 1882) at the southernmost edge of its distribution in Italy

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The smooth newt *Lissotriton vulgaris* (Linnaeus, 1758) is a polytypic taxon considered as a species complex with several subspecies (Wielstra et al., 2018; Speybroeck et al., 2020). Its wide range includes most of Europe, except Portugal and Spain, and extends eastward to Ukraine and Russia. In Italy there is an endemic subspecies, the Italian smooth newt *L. v. meridionalis* (Boulenger, 1882). This urodele is mainly distributed in northern and central regions in Italy, reaching its southern limit in the Campania region where it is known from a limited number of locations (Scillitani et al., 2012; Iannella et al., 2017; Di Nicola et al., 2019). *Lissotriton vulgaris meridionalis* inhabits several types of terrestrial environments and lentic freshwater habitats (both perennial and temporary, natural or artificial), with philopatric habits and low dispersal abilities (Razzetti et al., 2007). The nominal species is not considered as threatened (Arntzen et al., 2009) because of abundant and stable populations across most of the range and is not protected by the EU Habitats Directive (92/43/EEC). However, the conservation status of some subspecies with limited distribution ranges should be revised (Wielstra et al., 2018; AmphibiaWeb, 2021). In particular, the Italian smooth newt deserves special concern and protection since some populations have declined in recent years due to habitat loss, destruction, and fragmentation (Razzetti et al., 2007; Scillitani, 2012). This newt is now classified as Near Threatened at the national level (Rondinini et al., 2013), and as Critically Endangered in the Campania region (Scillitani, 2012).

To our knowledge, during recent years, no systematic surveys have been conducted to document the distribution and abundance of this newt species at its southernmost distributional limit in the Campania region. In this note, we report the presence of *L. v. meridionalis* in a new locality in the province of Caserta (Table 1). This finding improves the knowledge on the distribution of this newt in southern Italy. As a part of the BIO.FOR. POLIS project (https://www.esperienzeconilsud.it/bioforpolis), we visited and surveyed for amphibians in a protected pond in Riserva Naturale di Castel Volturno during spring 2017 (Fig. 1A). This small water body is the only freshwater habitat in the protected area and was probably part of the Volturno river floodplain's wetlands before the land reclamations occurred since 1600. The pond is completely isolated from other freshwater habitats by the main road SP303 and some buildings, and by a concrete enclosure delimiting the protected area. This aquatic habitat can be defined as a Mediterranean temporary pond for its characteristics (*sensu* Zacharias and Zamparas, 2010). It's formed in a depression with a sandy ground in the retrodunal area, and is fed by rainfall and probably by shallow water table. Its hydroperiod is variable, but the pond is usually water-filled between November and June/July, depending on the rainfall's annual variation. The maximum wetted surface is about 850 m^2, and the maximum water depth is about 1 m in the centre. The pond is located in a littoral pinewood plantation and is bordered by holm oaks (*Quercus ilex*) with the presence of field elm (*Ulmus minor*) and grey willow (*Salix cinerea*). The aquatic and amphibious vegetation includes plants such as *Lemna minor*, the fool's-water-cress *Helosciadum nodiflorum*, the yellow flag *Iris pseudacorus*, the common reed *Phragmites australis* and the Cyperacaea *Carex pendula* (Esposito et al., 2012). A rich community of invertebrates inhabits this small water body, such as microcrustaceans (mainly Cladocera), crustacean amphipods, aquatic snails (*Lymnaea stagnalis*, *Planorbis* spp.), larvae of Odonata and Diptera (Stratiomidae and other families), whirligig beetles (Gyrinidae), and the water beetle *Hidaticus*

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Table 1. The new record of *Lissotriton vulgaris meridionalis* at the southernmost edge of the range in Italy.

Region	Locality (Province)	Coordinates	Elevation	Dates
Campania	Riserva Naturale di Castel Volturno, Ischitella Lido, Castel Volturno (CE)	40.9279° N 14.0235° E	6 m a.s.l.	4, 8.III.2017 26.IV.2017

(*Hydaticus*) *seminiger* (Fusillo, 2019).

The pond was surveyed for amphibians on 4 and 8 March, and 26 April, which should be within the breeding season of *L. v. meridionalis*. The wetted area was at a maximum in March, whereas the pond resulted fragmented in two small wetted surfaces of a few square meters each at the end of April. The water temperature ranged between 14 and 15.9° C during the three visits. We performed Visual Encounter Surveys (VES, following Heyer et al., 1994) and dip-netting techniques to collect newts and amphibian larvae. The pond was sampled by dip-netting several times along a transect (about 2 m) to cover most pond shoreline. After capture, newts were identified, measured and then released. To prevent the transmission of diseases, we observed strict biosecurity guidelines according to the Conservation Committee of the *Societas Herpetologica Italica*. During surveys, we also recorded the presence of the common edible frog *Pelophylax* kl. *esculentus*. On average we captured 1.9 newts/hour of searching among three sampling occasions. Overall, we identified 11 individuals (4 females and 7 males) and one larva of *L. v. meridionalis* (Fig. 1B, C). Considering sampling effort, we hypothesised a low to moderate population density. The mean total length (distance from the tip of the snout to the tip of the tail) of captured newts was 73.25 mm (± 4.7 SD) whereas the mean weight was 1.65 g (± 0.43 SD). These values are similar to those recorded in other populations in Central Italy (e.g., Razzetti et al., 2007; Di Giuseppe, 2012). The presence of Italian smooth newts in the pond has been further confirmed during occasional observations in spring-early summer 2018 and 2019.

Our distributional record of *L. v. meridionalis* represents the most recent one of the scattered occurrences of this newt at the southernmost edge of its range. Balletto (2005) and Scillitani (2012) reported 10 and 11 records of the species in the Campania region, respectively. Iannella et al. (2017) listed only 6 localities of presence in Campania, of which 4 are in the Caserta province. However, these reported sites were mostly (90%) recorded between 1977 and 1998 and not verified in recent years. Besides our new record, the presence of

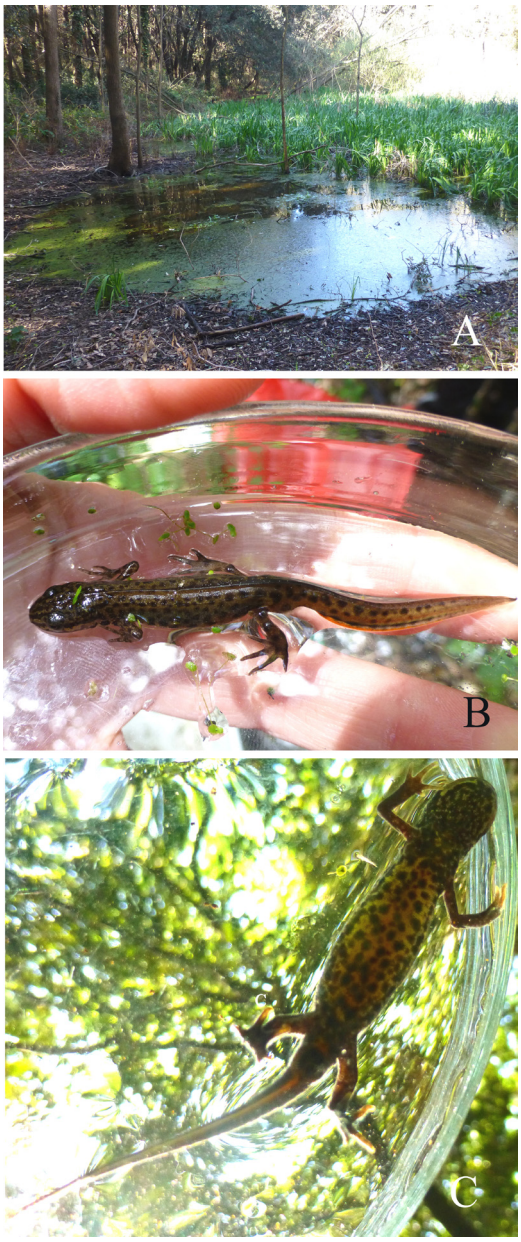

Figure 1. The temporary pond in the Riserva Naturale di Castel Volturno (A); dorsal (B) and ventral (C) view of an adult individual of *Lissotriton vulgaris meridionalis*. Photos by Manlio Marcelli.

L. v. meridionalis in Campania is documented in only two other localities lately (one in Caserta province and one around Napoli; Basile et al., 2014). Thus, future research is necessary to update the actual distribution and estimate population abundance of *L. v. meridionalis* at the range limit in northern Campania.

Most of the occurrence records that have been previously reported in the Caserta province lie in a zone that has been declared a National Concern Site by the Italian Government due to high contamination potential (Fig. 2). The entire area is severely polluted by untreated discharge of wastewater from settlements, illegal dumping of industrial wastes as well as by intensification of livestock farming (buffalo farms) and agriculture (Maresca et al., 2018). Considering these degraded environmental conditions, we hypothesise that *L. v. meridionalis* populations recorded before 2000 might have been declined or even disappeared completely.

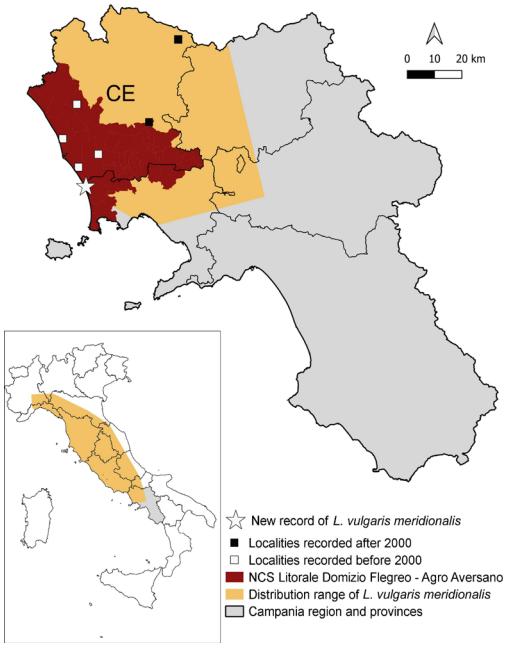

Figure 2. Map of the new distributional record of *Lissotriton vulgaris meridionalis*. The range of the Italian smooth newt (based on Iannella et al., 2017), the Caserta province (CE) of the Campania region, and the polluted area named National Concern Site (NCS) Litorale Domizio Flegreo-Agro Aversano, are shown. White squares: records of *L. v. meridionalis* reported before 2000 (1986-1996) in the Caserta province (Balletto, 2005; Scillitani, 2012; Iannella et al., 2017); black squares: localities of newt presence recorded in 2008 and 2014 (Basile et al., 2014, Iannella et al., 2017).

The isolated breeding site of the Riserva Naturale di Castel Volturno could be therefore significant for the conservation of *L. v. meridionalis* in this area, being potentially unpolluted and inhabited by one of the few remnant populations. Moreover, according to Buono and colleagues (2018), the isolation of this population could have conservation genetic implications that deserve to be further investigated at local spatial scale.

The temporary ponds are considered important breeding sites for amphibians in Mediterranean climates (e.g. Gomez-Rodriguez et al., 2009). However, because of their small size and intrinsic fragmentation, these habitats are considered highly vulnerable. Their vulnerability could increase under future scenarios of climate changes in the Mediterranean region. A strong decrease in rainfall could reduce the hydroperiod and determine the disappearance of temporary ponds in several cases (Zacharias and Zamparas, 2010). For these reasons, it is essential to protect ephemeral ponds and amphibian species that breed in these freshwater habitats. In this sense, we believe that the temporary pond and the Italian smooth newt population discovered in the Riserva Naturale di Castel Volturno are worthy of further investigations and conservation efforts. Considering the rarity of the species in the southernmost range, demographic and genetic surveys are equally necessary to better assess the newt population size, viability and genetic diversity.

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