

Incognito in Ticino : a novel species of lizard for the swiss herpetofauna

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Abstract

National species lists, especially red lists, must account for the latest taxonomic updates in order to best protect newly discovered biodiversity. Here we demonstrate the cryptic presence of a new species for the Swiss herpetofauna: Carnie's lizard *Zootoca carniolica*, an oviparous form of the viviparous common lizard *Z. vivipara*, which was recently elevated to species rank given their complete reproductive isolation. A few years ago, a range-wide phylogeographic study have genetically barcoded this subalpine species in Val Morobbia (Ticino), just a few hundred meters from the Italian border, but this finding has subsequently remained unnoticed. According to fine-scale distribution data, this population is presumably isolated, and the closest populations in Graubünden and northern Ticino all likely correspond to *Z. vivipara*. Since Val Morobbia is the only swiss record of *Z. carniolica*, the species rises as the most endangered reptile of the country. We confirmed the persistence of this population as of summer 2021 and call for immediate actions to document the geographic extent and life history of *Z. carniolica* in Switzerland, in order to implement conservation measures.

Key words : herpetology ; speciation ; viviparity ; *Zootoca carniolica* ; *Zootoca vivipara*.

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Résumé étendu

Les listes d'espèces, et notamment les listes rouges nationales, doivent prendre en compte les dernières avancées taxonomiques afin de protéger au mieux la biodiversité, y compris les taxons récemment découverts grâce à la génétique. L'herpétofaune suisse n'est pas épargnée par de tels changements, comme l'illustrent la rainette de Perrin ou encore l'orvet de Vêrone. Dans cet article, nous mettons en lumière la présence d'une nouvelle espèce passée complètement inaperçue des inventaires helvétiques: le lézard de Carnie *Zootoca carniolica*. Proche cousin du lézard vivipare (*Z. vivipara*), *Z. carniolica* est une forme ovipare restreinte au sud des Alpes, génétiquement et reproductivement complètement isolée de ce dernier. Ainsi, le Comité Taxonomique de la Société Européenne d'Herpétologie l'a unanimement érigé au rang d'espèce dans sa liste publiée en 2020. Il existe même des critères externes, bien que subtils, qui permettent de le différencier de *Z. vivipara* dans les Alpes. En 2018, une étude portant sur la phylogéographie du genre *Zootoca*

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au sein de sa vaste aire de répartition a analysé quelques échantillons helvétiques. Parmi eux, la nouvelle espèce ovipare *Z. carniolica* fut détectée dans une localité tessinoise: le Val Morobbia (46.16° N, 9.11° E), à quelques centaines de mètres de la frontière italienne. Les auteurs n'ayant pas étayé spécifiquement ce résultat remarquable pour notre herpétofaune, celui-ci est resté sans suite. D'après le patron de distribution, le Val Morobbia semble accueillir l'unique population suisse de l'espèce, à l'exception peut-être du Val Vedeggio situé non loin. Ces localités sont isolées par la basse vallée du fleuve Ticino, et les populations helvétiques voisines (Val Calanca dans les Grisons) appartiennent vraisemblablement à l'espèce *Z. vivipara*, connue dans les hautes vallées tessinoises de par ses mœurs vivipares (Valle Formazza and Valle Piumogna). Ainsi, le lézard de Carnie deviendrait le reptile le plus localisé et sans doute le plus menacé de Suisse. Nous avons pu constater la persistance de la population du Val Morobbia au cours de l'été 2021, et appelons désormais à des efforts de recherche visant à mieux connaître l'histoire de vie de cette espèce discrète et méconnue dans notre pays, ainsi qu'une campagne de barcoding génétique pour en établir sa distribution exacte. Une telle synthèse de connaissances permettra la mise en place de mesures de conservation efficaces, mesures plus que jamais nécessaires à la vue des fortes menaces qui pèsent sur l'espèce à l'échelle globale.

Mots-clés: hérapétologie; spéciation; viviparité; *Zootoca carniolica*; *Zootoca vivipara*.

MAIN TEXT

With the integration of molecular tools in taxonomy, species new to science are being discovered at a frenetic pace, keeping biodiversity lists under a constant state of revision. The issue does not spare the relatively well-known herpetofauna of Europe. Taxa once believed to be monotypic are now known to hide multiple, cryptic evolutionary lineages, representative of several genuine species or subspecies (reviewed by SPEYBROECK *et al.*, 2020). In Switzerland alone, taxonomic changes over the last few years have affected no less than three species of amphibians (*Hyla perrini*, DUFRESNES *et al.* 2018) and reptiles (*Natrix natrix / helvetica*, KINDLER *et al.* 2017; *Anguis fragilis / veronensis*, GVOŽDÍK *et al.* 2013). In this note, we emphasize a fourth case: the unnoticed presence of a neo-taxon allied to the common lizard *Zootoca vivipara*, at the eastern margin of the canton of Ticino.

To appreciate this case, let us overview the molecular ecology and taxonomy of *Zootoca*. For long, *Z. vivipara* was considered the only species of the genus. Widespread across the temperate, boreal, alpine, Atlantic and continental climates of the Eurosiberian region, it is the most largely distributed terrestrial reptile on Earth, ranging from northern Spain to the Russian Pacific coast (HOREO *et al.* 2018). The species is present from sea level to mountaintops (~2400 m) and spends the winter buried in the ground, where it can survive subzero temperatures (BERMAN *et al.* 2016). Most populations are viviparous, a reproductive strategy often viewed as an adaptation to harsh and unpredictable ecological conditions (but see HOREO *et al.* 2021), as also seen in our alpine salamanders (*Salamandra atra*), slow worms (*Anguis* sp.) and adders (*Vipera* sp.). In this mode, soft-shell eggs are retained and developed within the mother's oviduct, and hatch within hours after parturition (HEULIN & GUILLAUME 2010). There are, however, some oviparous populations in the Pyrenean and the sub-alpine regions (Northern Italy, Slovenia and Southern Austria), where females lay regular eggs that incubate and hatch in the outside environment. Such intraspecific variation of reproductive mode is nearly unique in reptiles and makes *Zootoca* a valuable model to study the evolution of viviparity.

Recent phylogeographic studies have demonstrated that the oviparous populations consist of independent genetic lineages (HOREO *et al.* 2018). The oviparous Pyrenean populations (clade B) have remained isolated from the viviparous populations (clades C-F) since the Pleistocene (~2.0 Mya). Even more diverged, the oviparous subalpine populations (clade A) previously described as the subspecies *Z. v. carniolica* (MAIER *et al.* 2000) have a Pliocene origin (~4.4 My) and are thus basal to all other lineages. As barcoding studies unveiled lineage distributions around the Italian and Austrian Alps, researchers discovered secondary contact zones between the oviparous (*carniolica*) and viviparous (*vivipara*) forms. Hybridization was found to be very rare (LINDTKE *et al.* 2010) and genetic studies showed a complete absence of gene exchange despite close parapatry and even local syntopy of individuals (CORNETTI *et al.* 2015a, b). Given their full reproductive isolation, the two are thus now considered valid distinct species (SPEYBROECK *et al.*, 2020), as the subalpine oviparous Carnie’s Lizard *Z. carniolica*, and the widespread, mostly viviparous Common Lizard *Z. vivipara*. Although phenotypically very similar, it is possible to distinguish these species without molecular tools in the Alpine region, namely by counting the ventral scales and the femoral pores to compute a diagnostic ratio (RODRIGUEZ-PRIETO *et al.* 2017).

Only *Z. vivipara* is listed among the Swiss herpetofauna (karch.ch), since all populations of the country are thought to be viviparous (HEULIN & GUILLAUME 2010). However, a close inspection of the extensive mitochondrial barcoding data from HOREO *et al.* (2018) unambiguously show that clade A, i.e. the oviparous species *Z. carniolica*, also occurs in Switzerland (Fig. 1), namely at a single locality labelled “Morobia, 46.15° N, 9.12° E” (Appendix S1 in HOREO *et al.* 2018). This locality corresponds to Val Morobbia in eastern Ticino. One can assume mitochondrial markers to offer a reliable barcoding tool between these species, since

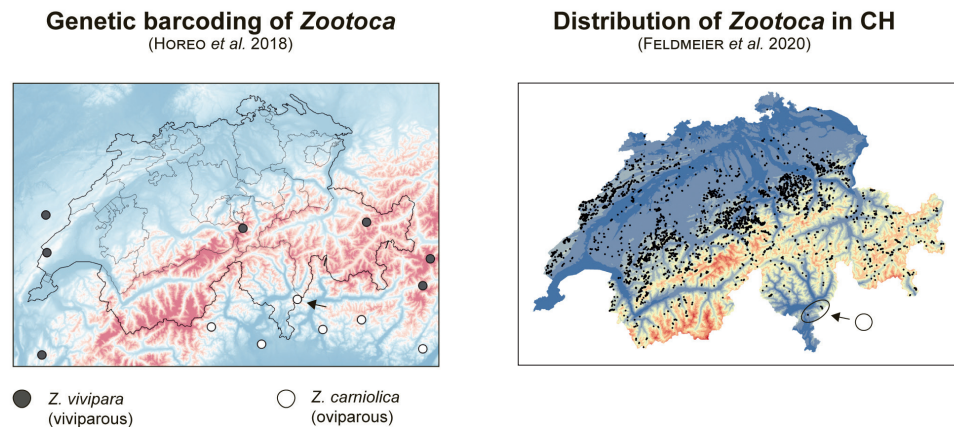


Figure 1. Molecular evidence for the presence of *Zootoca carniolica* in Switzerland (arrows), according to the genetic barcoding of HOREO *et al.* (2018) (left), with the corresponding isolated populations from the occurrence dataset of *Zootoca* in CH by FELDMEIERS *et al.* (2020) (right). Topography is illustrated by the blue-red background gradient.

Figure 1. Barcoding génétique indiquant la présence de *Zootoca carniolica* en Suisse (flèches), d’après HOREO *et al.* (2018) (carte de gauche), avec les populations correspondantes annotées d’après les données de distribution de *Zootoca* en Suisse publiée par FELDMEIERS *et al.* (2020) (carte de droite). Le gradient bleu-rouge de fond illustre la topographie.

they were explicitly shown to lack genetic introgression (CORNETTI *et al.* 2015a, b). Yet, such key finding has remained overlooked because the authors themselves did not report it in their Table 2 (which summarizes the national occurrences of each clade), and the close proximity of Italy (< 500 m) makes the Swiss record of *Z. carniolica* imperceptible on their map (see figure 1 in HOREO *et al.* 2018).

Carnie's lizard *Zootoca carniolica* thus consists of a novel species of the Swiss herpetofauna, with a presumably narrow distribution. In Ticino, *Zootoca* is essentially restricted to the northern parts of the canton and Val Morobbia consists of an isolated patch (figure 1 ; FELDMIEIER *et al.* 2020, see also MAIER *et al.* 2009). This patch is disconnected from the rest of the Swiss range by the Ticino River : the closest populations are located in Val Calancasca (Graubünden), about 20 km north (FELDMIEIER *et al.* 2020). Val Calancasca appears relatively well connected to the Ticinese northern valleys, where some populations were confirmed as viviparous by direct observations (e.g. Valle Formazza and Valle Piumogna, GHIELMI *et al.* 2001). Hence, these valleys would correspond to the southern limit of *Z. vivipara* in Switzerland, while the few records south of Ticino River correspond to *Z. carniolica*. This biogeographic pattern in which species bypassed the Alps during their post-glacial colonization is unusual, yet it is shared by a few terrestrial vertebrates similarly adapted to a mountainous lifestyle. For instance, the north-alpine *A. fragilis* crossed the Graubünden Alps and might come into contact with *A. veronensis* in Italy (URSENBACHER & ZWAHLEN 2015). The blind mole (*Talpa caeca*) naturally reached the Upper Rhône valley by Simplonpass (MARCHESI *et al.* 2000).

Because it has not been integrated in conservation programs as per its recent discovery (2015), and since only a single population is known at present (with a putative range of less than 100 km²), *Z. carniolica* is undoubtedly the most endangered reptile of Switzerland. We have visited Val Morobbia twice in the summer of 2021 (on July 18th and July 29th), and could observe (without capture) two individuals during the second visit around 2 000 m a.s.l., hence confirming the persistence of the Swiss population. We call for immediate actions to improve knowledge of the ecology and exact distribution of this novel species for the country, in order to implement adapted protective measures. Refining species boundaries will require genetic barcoding and phenotypic assessments throughout the subalpine Swiss ranges. In particular, new prospections should target the areas adjacent to Val Morobbia, such as Val Vedeggio, where an isolated record in FELDMIEIER *et al.* (2020) might also correspond to *Z. carniolica* (figure 1). Although not yet assessed by the IUCN, *Z. carniolica* is highly threatened at the species level. In Italy, where most of its range is situated, many lowland populations have gone extinct due to high anthropization combined with climate changes, which have led to the destruction of its humid habitat in the last century (SINDACO *et al.* 2016). The presence of both viviparous and oviparous *Zootoca* species highlights the remarkable herpetological biodiversity of Switzerland, a small country yet situated at a major biogeographic crossroad.

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REFERENCES

- BERMAN D.L., BULAKHOVA N.A., ALFIMOV A.V. & MESHCHERYAKOVA E.N., 2016. How the most northern lizard, *Zootoca vivipara*, overwinters in Siberia. *Polar Biology* 39: 2411-2425.
- CORNETTI L., BELLUARDO F., GHIELMI S., GIOVINE G., FICETOLA G.F., BERTORELLE G., VERNESI C. & HAUFFE H.C., 2015a. Reproductive isolation between oviparous and viviparous lineages of the Eurasian common lizard *Zootoca vivipara* in a contact zone. *Biological Journal of the Linnean Society* 114: 566-573.
- CORNETTI L., FICETOLA G.F., HOBAN S. & VERNESI C., 2015b. Genetic and ecological data reveal species boundaries between viviparous and oviparous lizard lineages. *Heredity* 115: 517-526.
- DUFRESNES C., MAZEPA G., RODRIGUES R., BRELSFORD A., LITVINCHUK S.N., SERMIER R., BETTO-COLLARD C., BLASER O., BORZÉE A., CAVOTO E., FABRE G., GHALI K., GROSSEN C., HORN A., LAVANCHY G., LEUENBERGER J., PHILLIPS B.C., SAUNDERS P.A., SAVARY R., MADDALENA T., STÖCK M., DUBEY S., CANESTRELLI D. & JEFFRIES D.L., 2018. Formal description of the new tree frog species inhabiting Northern Italy and Southern Switzerland. *Bulletin de la Société Vaudoise des Sciences Naturelles* 97: 5-11.
- FELDMER S., SCHMIDT B.R., ZIMMERMANN N.E., VEITH M., FICETOLA G.F. & LÖTTERS S., 2020. Shifting aspect or elevation? The climate change response of ectotherms in a complex mountain topography. *Diversity & Distributions* 26: 1483-1495.
- HEULIN B. & GUILLAUME C.-P., 2010. Le lézard vivipare. Pp. 394 -401 in VACHET J-P & GENIEZ P (Eds): *Les reptiles de France, Belgique, Luxembourg et Suisse*. Mèze: Biotope.
- HORREO J.L., PELAEZ M.L., SUAREZ T., BREEDVELD M.C., HEULIN B., SURGET-GROBA Y., OKSANEN T.A. & FITZE P.S., 2018. Phylogeography, evolutionary history and effects of glaciations in a species (*Zootoca vivipara*) inhabiting multiple biogeographic regions. *Journal of Biogeography* 45: 1616-1627.
- HORREO J.L., JIMENEZ-VALVERDE A. & FITZE P.S., 2021. Climatic niche differences among *Zootoca vivipara* clades with different parity modes: implications for the evolution and maintenance of viviparity. *Frontiers in Zoology* 18: 32.
- GHIELMI S., HEULIN B., SURGET-GROBA Y. & GUILLAUME C.P., 2001. Identification de populations ovipares de *Lacerta (Zootoca) vivipara* en Italie. *Bulletin de la Société Herpétologique de France* 9: 19-29.
- GVOŽDÍK V., BENKOVSKY N., CROTTINI A., BELLATI A., MORAVEC J., ROMANO A., SACCHI R. & JANDZIK D., 2013. An ancient lineage of slow worms, genus *Anguis* (Squamata: Anguillidae), survived in the Italian Peninsula. *Molecular Phylogenetics & Evolution* 69: 1077-1092.
- KINDLER C., CHÈVRE M., URSENBACHER S., BÖHME W., HILLE A., JABLONSKI D., VAMBERGER M. & FRITZ U., 2017. Hybridization patterns in two contact zones of grass snakes reveal a new central European snake species. *Scientific Reports* 7: 7378.
- MARCHESI P., BLANT M., HOLZGANG O. & MADDALENA T., 2000. Aperçu de la richesse en petits mammifères du Simplon et découverte de la taupe aveugle *Talpa caeca* (Savi, 1822) en Valais. *Bulletin de la Murithienne* 118: 27-38.
- MAYER W., BÖHME W., TIEDEMANN F. & BISCHOFF W., 2000. On oviparous populations of *Zootoca vivipara* (Jacquin, 1787) in south-eastern Central Europe and their phylogenetic relationship to neighbouring viviparous and South-west European oviparous populations (Squamata: Sauria: Lacertidae). *Herpetozoa* 13: 59-69.
- RODRIGUEZ-PRIETO A., GIOVINE G., LADDAGA L., GHIELMI S. & CORNETTI L., 2017. Very similar, but not identical: morphological taxonomic identification to improve the resolution of fine-scale distribution of *Zootoca (vivipara) carniolica*. *Amphibia-Reptilia* 38: 533-539.
- SINDACO R., DORIA G., RAZZETTI E. & BERNINI F., 2006. *Atlas of Italian amphibians and reptiles*. Firenze: Societas Herpetologica Italica, Edizioni Polistampa.
- SPEYBROECK J., BEUKEMA W., DUFRESNES C., FRITZ U., JABLONSKI D., LYMBERAKIS P., MARTÍNEZ-SOLANO I., RAZZETTI E., VAMBERGER M., VENCES M., VÖRÖS J. & CROCHET P.-A., 2020. Species list of the European herpetofauna - 2020 update by the Taxonomic Committee of the Societas Europaea Herpetologica. *Amphibia-Reptilia* 41: 139-189.
- URSENBACHER S. & ZWAHLEN V., 2015. Les orvets: une ou deux espèces en Suisse? In : *Résumés des communications présentées lors du 22ème Colloque herpétologique du Centre de Coordination pour la Protection des Amphibiens et des Reptiles de Suisse (karch)*. 5th December 2015.