#### Thalassia Salentina Thalassia Sal. 42 (2020), 41-48 ISSN 0563-3745, e-ISSN 1591-0725

DOI 10.1285/i15910725v42p41 http: siba-ese.unisalento.it - © 2020 Università del Salento

# FRANCESCA OROSCOPI<sup>1</sup>, PIERO CARLINO<sup>1</sup>, OLIVIER S.G. PAUWELS<sup>2</sup>

 <sup>1</sup> Department of Herpetology, Museo di Storia naturale del Salento, s.p. Calimera-Borgagne km 1, 73021, Calimera, Italy
 <sup>2</sup> Département des Vertébrés Récents, Institut Royal des Sciences naturelles de Belgique, Rue Vautier 29, B-1000 Brussels, Belgium.
 e-mail: francescaoroscopi@yahoo.it

# AN UNUSUALLY PATTERNED ITALIAN TREE FROG HYLA INTERMEDIA FROM SALENTO PENINSULA, SOUTHERN ITALY (AMPHIBIA: HYLIDAE)

#### **SUMMARY**

Hyla intermedia is an endemic Italian tree frog species, distributed from the southern edge of the Alpine massif to the tip of Calabria and into Sicily. Previously referred as the widespread Hyla arborea, it has only recently been recognized as a distinct species. It shows a bright, uniformly green dorsal coloration. We report a previously unknown colour variant, displayed by an individual collected in Salento Peninsula. It differs from the typical form by its dark olive-brown background color with a contrasting marbled pattern on its dorsal and lateral surfaces. Similarly to typical individuals, this specimen was able to lighten its background color under the effect of stress, but its unusual marbled pattern was permanent. Marbled patterns are also known in other Hyla species, but were not so far recorded in H. intermedia.

#### INTRODUCTION

The Italian tree frog *Hyla intermedia* Boulenger, 1882 is an endemic species distributed from the southern edge of the Alpine massif to the tip of Calabria and into Sicily (Canestrelli *et al.*, 2007b). A thermophilic species, it prefers sunny and shallow water sites, and breeds in temporary ponds and puddles (Ebisuno and Gentilli, 2002). Because of the subtility of morphological and chromatic differentiation within the genus, the populations of this species were long attributed to the European tree frog *Hyla arborea* (Linnaeus, 1758), together with all other Palaearctic populations of tree frogs (Canestrelli *et al.*, 2007a). At least eight taxa of hylid frogs, remarkably similar in appear-

ance, are distributed around the Mediterranean Basin, providing an interesting case for the study of cryptic speciation (Dufresnes *et al.*, 2018, and references therein). The distinctiveness of *Hyla intermedia* was demonstrated based mostly on genetic distance and call differences, associated with a subtle combination of morphological differences (Nascetti *et al.*, 1995; Rosso *et al.*, 2004). The skin of *Hyla intermedia* is smooth and shiny, with a typically bright green and generally uniformly coloured dorsal surface; the ventral parts, except the throats of males, being white and translucent on the sides, grainy in the central portion (Boulenger, 1882; Lanza, 1983; Ancona and Gentilli, 2004; Piazzini *et al.*, 2005; Biasoli *et al.*, 2011).

In the region of Salento *Hyla intermedia* is very rare and the few known populations are located mainly along the Oriental side. These populations are progressively declining due to the strong alteration to the marshes during the last decades (Fattizzo and Marzano, 2002). Fortunately, four breeding sites were newly reported in Salento by Liuzzi and Scillitani (2010).

# **RESULTS**

An adult female *Hyla intermedia* was caught in San Donato di Lecce (GPS: 40°16′56.03″N, 18°10′46.84″E), Lecce Province, in mid-May 2019. It was found near an artificial pond surrounded by green meadow, reeds, and rock outcrops, at approximately 150 m from a mixed wood forest. It showed a slender body (snout-vent length -SVL, 40 mm); a head narrower than the body (head width 13.2 mm), a snout rounded in dorsal and lateral view, slightly protruding over upper jaw in lateral view. Nostrils not protruding, with anterolateral openings closer to the tip of snout than to the eye; an eye diameter (5.00 mm) more than twice as long as tympanum diameter (2.3 mm). Tympanum brownish, round with weakly developed supratympanic fold extending from posterior corner of eye to near the shoulder. Hindlimbs long and slender (hind leg length 61.0 mm) and relatively robust; tibia 47.7% of SVL and tarsus 27.5% of SVL, with fingers bearing enlarged and round discs. Ventral skin granular, dorsal skin smooth. Dorsum dark olive-green with diffuse lighter marbling on the head, body and limbs. Flanks pale green, ventral surfaces of throat, chest, belly, and limbs beige; black and with lateral lines extending from nostrils along canthus rostralis to the groin, without forming a well-defined inguinal loop.

We identified it as a *Hyla intermedia* based on its morphometrics and on comparison with voucher material (see Appendix). However this individual differs notably in color and pattern from typical *Hyla intermedia*. Its coloration was indeed much darker, and noticeably it was marbled and not uniform. We observed other individuals of this species in the same locality,

some of them calling from the adjacent wood, but none of them showed these atypical coloration and pattern. Figure 1 compares the marbled specimen with another adult (snout-vent length 43 mm), typically coloured and patterned, from the same locality.

Wassef et al. (2019) provided a nomenclature for the different states of the white and black stripes composing the *linea marginalis* in *Hyla arborea*. Following this nomenclature, which is also applicable to other *Hyla* species, our marbled individual showed a black stripe which was "narrow with diffuse pigmentation", and a white stripe which was "very thin" (Fig. 2b). By comparison, the syntopic green individual shown on Figure 2a showed a "well-defined but narrow" white stripe and a "narrow with diffuse pigmentation" black stripe.

We observed a rapid background color change from darker to lighter olive green with a decreasing brightness in a span of a few minutes, with the handling of the marbled individual. After the individual had been handled, it returned to its previous coloration (Fig. 3). The specimen retained its darker and marbled colour after placement in a terrarium. It is currently kept alive in the vivarium of the *Museo di Storia Naturale del Salento*, where it will be preserved upon its death.

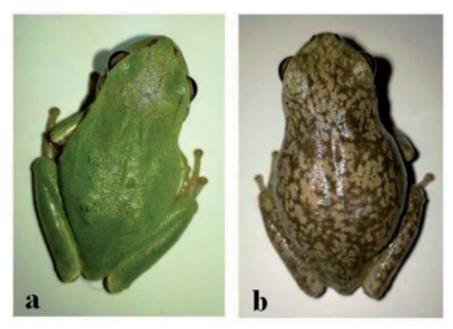


Fig. 1. Dorsal view of two differently patterned adult *Hyla intermedia* from San Donato di Lecce: typical bright green and uniform (a) and dark olive-green and marbled (b).

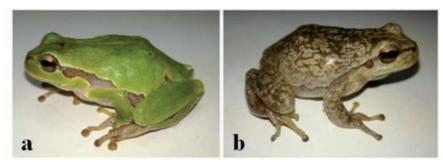


Fig. 2. Linea marginalis in two differently patterned adult *Hyla intermedia* from San Donato di Lecce: white stripe "narrow and well-defined" (a) or "very thin" (b), and black stripe "narrow with diffused pigmentation" (a, b).



Fig. 3. Change in colour observed in a marbled *Hyla intermedia* from San Donato di Lecce due to the stress of handling (compare with Fig. 1b and Fig. 2b).

#### **DISCUSSION**

Our presently reported marbled specimen represents a newly recorded colour pattern in *Hyla intermedia*, but variants to the typical uniformly green pattern have been described in various *Hyla* species, sometimes due to physiological changes (King *et al.*, 1994; our material examined). Among the known cases, in *Hyla arborea*, Zimić and Agović (2018) identified several colour types among which the prevailing light green, and numerous different colour gradients and marbled colourations. Koren and Jelić (2011) also distinguished different colorations within the green form of *Hyla arborea* (from green-grey to brown). A marbled pattern for *Hyla japonica* was illustrated by Kang *et al.* (2016).

The color change under stress conditions, observed in our marbled individual, is well known in *Hyla* and other amphibians. Studies in *Hyla arborea* showed that it was the result of the dispersion (darkening) or aggregation (lightening) of melanosome pigment within melanophores (Nielsen, 1978a-b).

The colour variant of *Hyla intermedia* here described has been detected in a locality where usually typical bright green individual occur. Syntopic occurrences of various patterns have been shown in other *Hyla* species; for example Dordević *et al.* (2016) observed four colour variants of tree frog *Hyla arborea* in two close water bodies and under the same stable weather conditions. The ability of *Hyla* to change color for camouflage is an important adaptation, particularly in xeric and seasonally changing environments (Degani and Biton, 2013), with differently coloured substrates according to the seasons.

The locality in which our marbled tree frog was collected is a patchy environment offering different and seasonally changing microhabitats, from green vegetation to dark rocky and unvegetated soil, where a marbled pattern could favour a better crypsis while moving between different substrates. Further field studies in this locality will allow to see in what proportions this pattern is found and to better evaluate its adaptation to micro-environmental and substrate conditions.

# **ACKNOWLEDGMENTS**

We are grateful to Antonio Durante (*Museo di Storia Naturale del Salento*) for providing working facilities.

# **REFERENCES**

- Ancona N., Gentilli A., 2004 *Raganella Italiana*. *Hyla Intermedia* Boulenger, 1882. In: Bernini F., Bonini L., Ferri V., Gentilli A., Razzetti E., Scali S. (Eds.) *Atlante degli Anfibi* e dei *Rettili* della *Lombardia*. Monografie di Pianura 5, Cremona: 93-95.
- BIASIOLI M., GENOVESE S., MONTI A., 2011 Gestione e conservazione della fauna minore: esperienze e tecniche di gestione delle specie d'interesse conservazionistico e dei loro habitat. Fondazione Cariplo, Parco del Lura: 334 pp.
- BOULENGER G. A., 1882 Catalogue of the Batrachia Salientia s. Ecaudata in the collection of the British Museum. Second edition, Taylor and Francis, London: 503 pp.
- Canestrelli D., Cimmaruta R., Nascetti G., 2007b Phylogeography and historical demography of the Italian treefrog, *Hyla intermedia*, reveals multiple refugia, population expansions and secondary contacts within peninsular Italy. *Molecular Ecology* **16**: 4808-4821.
- Canestrelli D., Verardi A., Nascetti G., 2007a Genetic differentiation and history of populations of the Italian treefrog Hyla intermedia: lack of concordance between mitochondrial and nuclear markers. Genetica **130** (3): 241-255.
- Degani G., Biton E., 2013 *Tree frog (Hyla savygnyi*) color and substrate preference. *American Open Animal Science Journal* **1** (3): 31-39.

- Dordević S., Simović A., Krizmanić I., Tomović L., 2016 *Colour variations in the European tree frog, Hyla arborea* (Linnaeus, 1758), from two small adjacent ponds in the Vojvodina province, Serbia. *Ecologica Montenegrina* **5**: 18-21.
- Dufresnes C., Mazepa G., Rodrigues N., Brelsford A., Litvinchuk S. N., Sermier R., Lavanchy G., Betto-Colliard C., Blaser O., Borzée A., Cavoto E., Fabre G., Ghali K., Grossen C., Horn A., Leuenberger J., Phillips B. C., Saunders P. A., Savary R., Maddalena T., Stöck M., Dubey S., Canestrelli D., Jeffries D. L., 2018 *Genomic evidence for cryptic speciation in tree frogs from the Apennine Peninsula, with description* of *Hyla perrini* sp. nov. *Frontiers in Ecology and Evolution* **6**: 144. doi: 10.3389/fevo.2018.00144.
- EBISUNO M., GENTILLI A., 2002 Reproductive site selection and characteristics of sources and sinks in an Italian tree frog metapopulation (Hyla intermedia, Boulenger 1882). Revue d'écologie 57 (3-4): 263-278.
- Fattizzo T., Marzano G., 2002 Dati distributivi sull'erpetofauna del Salento. *Thalassia Salentina* **26**: 113-132.
- Kang C., Kim Y. E., Jang Y., 2016 Colour and pattern change against visually heterogeneous backgrounds in the tree frog Hyla japonica. Scientific reports **6**: 22601. doi:10.1038/srep22601.
- KING R. B., HAUFF S., PHILLIPS J. B., 1994 Physiological color change in the Green treefrog: responses to background brightness and temperature. Copeia 2: 422-432.
- Koren T., Jeuć D., 2011 Interesting color forms of the European tree frog, Hyla arborea (Linnaeus, 1758) (Amphibia: Ranidae) from Croatia. Hyla 2: 27-29.
- Lanza B., 1983 Anfibi, Rettili (Amphibia, Reptilia). Guide per il riconoscimento delle specie animali delle acque interne italiane 27. Consiglio Nazionale delle ricerche, Roma: 196 pp.
- Liuzzi C., Scillitani G., 2010 L'Erpetofauna della Puglia; aggiornamenti e integrazioni. In: Di Tizio L., Di Cerbo A. R., Di Francesco N., Cameli A. (Eds.) Atti VIII Congresso Nazionale Societas Herpetologica Italica, 22-26 settembre 2010, Chieti. Ianieri Edizioni, Pescara, 31-36.
- NASCETTI G., LANZA B., BULLINI L., 1995 Genetic data support the specific status of the Italian treefrog (Amphibia: Anura: Hylidae). Amphibia-Reptilia 16 (3): 215-227.
- NIELSEN H. I., 1978a Ultrastructural changes in the dermal chromatophore unit of Hyla arborea during color change. Cell Tissue Research 194 (3): 405-418.
- NIELSEN H. I., 1978b The effect of stress and adrenaline on the color of Hyla cinerea and Hyla arborea. General and Comparative Endocrinology **36** (4): 543-552.
- Piazzini S., Favilli L., Manganelli G., 2005 Atlante degli Anfibi della Provincia di Siena (1999-2004). Sistema delle Riserve Naturali della Provincia di Siena. Quaderni Naturalistici 1: 112 pp.
- Rosso A., Castellano S., Giacoma C., 2004 The advertisement call of Hyla intermedia and H. sarda. Italian Journal of Zoology **71** (S2): 169-173. doi: 10.1080/11250000409356629.
- Wassef J., Savary R., Ghali K., Pasteur B., Dufresnes C., 2019 Biogeographic patterns of color and size polymorphisms in the European tree frog (Hyla arborea). Bulletin de la Société Vaudoise des Sciences Naturelles **98:** 93-103.

ZIMIĆ A., AGOVIĆ V., 2018 – Colour variations in the European tree frog, Hyla arborea (Linnaeus, 1758). 2nd Balkan Herpetological Symposium within 13th Croatian Biological Congress with International Participation, 19-23 September 2018, Poreč, Croatia.

# Appendix: Comparative voucher material examined (Spec. = specimen(s))

- Hyla arborea: RBINS 123 D (1 spec.; Dresden, Saxony, Germany); RBINS 123 E (2 spec.; Forro, Hungary); RBINS 7300 (2 spec.; Château-Salins, Moselle, France);
  RBINS 15128 (8 spec.; along the road between Vrsar and Brajkovic, 1 km before Klostar, Istria, Croatia); RBINS 15129 (2 spec.; Polykastro, Kilkis, Greece).
- Hyla meridionalis: RBINS 15123 (7 spec.; La Môle, Campaux, Maures, Var, France); RBINS 15126 (2 spec.; Forêt de Palayson, Maures, Var, France).
- Hyla molleri: RBINS 123 F (2 spec.; Spain); RBINS 15127 (1 spec.; E of RN 268, between Vila do Bispo and Carrapateira, Algarve, Portugal).
- Hyla orientalis: RBINS 7433 (1 spec.; Cernica Forest near Bucharest, Romania).
- Hyla perrini: RBINS 123 B (1 spec.; Milan, Italy); RBINS 2042 (10 spec. including a marbled one; Sasso, Bologna, Italy).