FACTA UNIVERSITATIS
Series: **Medicine and Biology** Vol. 24, No 1, 2022, pp. 27–29

UDC 582.651:632.76:591(497.11)
https://doi.org/10.22190/FUMB220303004Z

Original Scientific Article

AGRYPON POLYXENAE (SZÉPLIGETI, 1899) (HYMENOPTERA: ICHNEUMONIDAE: ANOMALONINAE); NEWLY RECORDED PARASITOID OF *ZERYNTHIA POLYXENA* (DENIS & SCHIFFERMÜLLER, 1775) (LEPIDOPTERA: PAPILIONIDAE: PARNASSIINAE) IN THE FAUNA OF SERBIA

Vladimir Žikić[1], Saša S. Stanković[1], Miloš Popović[1], Aleksandra Husarik[2], Marijana Ilić Milošević[1], Maja Lazarević[1], Aleksandra Trajković[1]

[1]Faculty of Sciences and Mathematics, Department of Biology and Ecology, University of Niš, Serbia
[2]Kovačica, Serbia

Abstract. *Ichneumonid Agrypon polyxenae is a solitary larval-pupal endoparasitoid developing within the caterpillars of the genus Zerynthia and distributed in the western Palearctic. Z. polyxena is the most common host, with its caterpillars feeding on highly poisonous Aristolochia species. Here we report A. polyxenae parasitising Z. polyxena for the first time in Serbia and provide a short species description. This parasitoid has also been recorded on Z. rumina in Spain. We suggest some evidence for its presence in the newly described Z. cassandra in Italy*.

Key words: *festoons, caterpillars, larval-pupal parasitoid, Aristolochia*

Introduction

Parasitic wasp *Agrypon polyxenae* (Szépligeti, 1899) is a slender body species and it is one of the larger members of Ichneumonidae family. This elegant wasp has been recorded as a solitary koinobiont endoparasitoid of caterpillars of the butterfly genus *Zerynthia* [1]. Its primarily host is the southern festoon, *Z. polyxena* (Denis & Schiffermüller, 1775). Since *A. polyxenae* parasitizes economically unimportant hosts, there is not much literature on its biology. Females are known to oviposit in mature *Zerynthia* caterpillars, where the parasitoid larva develops. The emergence of parasitoid imago occurs in the pupal stage of the host. Therefore, this kind of parasitism is defined as a larval-pupal parasitoid. Following the biology of its host, *A. polyxenae* has one generation per year and overwinters inside the host pupa. The integument of the pupa is known to be extremely strong and resistant to harsh weather conditions during a prolonged period, which can last over ten months [1]. Due to such strong skin, the adults of *A. polyxenae* chew the irregular exit hole on the integument of the host's pupa in its anterior dorsal part [1].

Being distributed in a large portion of the western Palearctic, *Z. polyxena* is the most common host of *A. polyxenae* [2]. However, there is solid evidence that it also parasitizes the Spanish festoon, *Z. rumina* (Linnaeus, 1758) [3, 4]. Although classified as LC (Least Concern), populations of *Z. polyxena* are undoubtedly in decline throughout Europe, and therefore strictly protected in Serbia and listed in the Habitats Directive 92/43 EEC of the European Union [5]. The taxonomy of the species is rather complex [6], as the great variability of the wing pattern triggered the description of over 30 subspecies and forms, where most of them belong to local populations. The very short flight period, and the specific food plant/microhabitat demands, make *Z. polyxena* convenient for further exploration of the post-glacial speciation [7]. It usually occurs in habitats lower than 900 m above sea level, but the altitude range may vary between 0 and 1700 m a.s.l. [8, 9]. The species occupies wetland areas close to water streams, riparian woodlands, abandoned agricultural land, roadsides, and urban areas, usually in patchy populations. This stenophagous caterpillar feeds on highly poisonous *Aristolochia* species (Piperales: Aristolochiaceae), namely *A. clematitis* L., *A. pallida* Willd., *A. pistolochia* L., *A. pontica* Lam., *A. rotunda* L. and *A. sicula* Tineo, with different local preferences [10]. Depending on the geographic region, the butterfly can be locally monophagous [11]. Along with several other plant genera from the family Aristolochiaceae, *Aristolochia* species are highly toxic and even carcinogenic to humans and responsible for the environmentally driven Balkan endemic nephropathy (BEN) [12].

In this paper, we report *A. polyxenae* for the first time in Serbia and give a more detailed species description and notes on its record. Furthermore, we discussed *Z. cassandra* (Geyer, 1828) as a potential host of *A. polyxenae*.

Correspondence to: Vladimir Žikić
Faculty of Sciences and Mathematics, Department of Biology and Ecology, University of Niš, Višegradska 33, 18000 Niš, Serbia
E-mail: zikicvladimir@gmail.com
Received March 3rd, 2022 / Accepted July 13th, 2022

Materials and Methods

Caterpillars of *Z. polyxena*, mainly 4th and 5th instar, were found on *A. clematitis*, usually on the leaf underside. In total, 56 caterpillars were collected. Individuals were put into plastic containers of approximately 0.5 litres of volume. The caterpillars were fed with birthwort leaves *ad libitum* in expectation of parasitoid appearance. All obtained parasitoid individuals were pinned and mounted on cardboards and prepared specimens were photographed using Nikon stereomicroscope (SMZ 745T). The distribution of *A. polyxenae* was compiled according to TaxaPad databases [13].

Results

Only four parasitoids emerged from *Z. polyxena* pupae. All of them belonging to *A. polyxenae*. The analysed material includes: 3♂ Idvor (45°11′18″ N; 20°30′47″ E), 15.09.2019, emerged on 04.05.2020, leg. A. Husarik; 1♀ Kladovo (44°36′24″ N; 22°36′47″ E), 05.05.2021, leg A. Trajković. Photographs of both male and female *A. polyxenae* habitus as well as selected body details were presented in Figure 1 (A-H). Also, the mummified pupa of *Z. polyxena* from which the parasitoid emerged was presented (Fig. 1I). The exit hole is partly visible on the dorsal anterior part of the empty mummy.

Short description

Female: body length 13.5 mm; head black with yellow frons; mandible yellow; ocelii black; antennae black, 51-segmented, first flagellar segment 2.5 x longer than the second; mesosoma entirely black covered with white dense setae; mesoscutum shiny, punctuated; both pair of wings hyaline; coxae and trochanters of all legs black, first and second pair of legs yellowish, hind legs brown; metasoma elongated, twice as long as the wings; petiole brown, slender rod-shaped; metasomal segments 2-4 brown, the rest black; proximal part of ovipositor black, distal part brown, length of ovipositor almost equal to hind tibia.
Male: body length 15-17 mm; general morphology and coloration as in female. All legs brown.

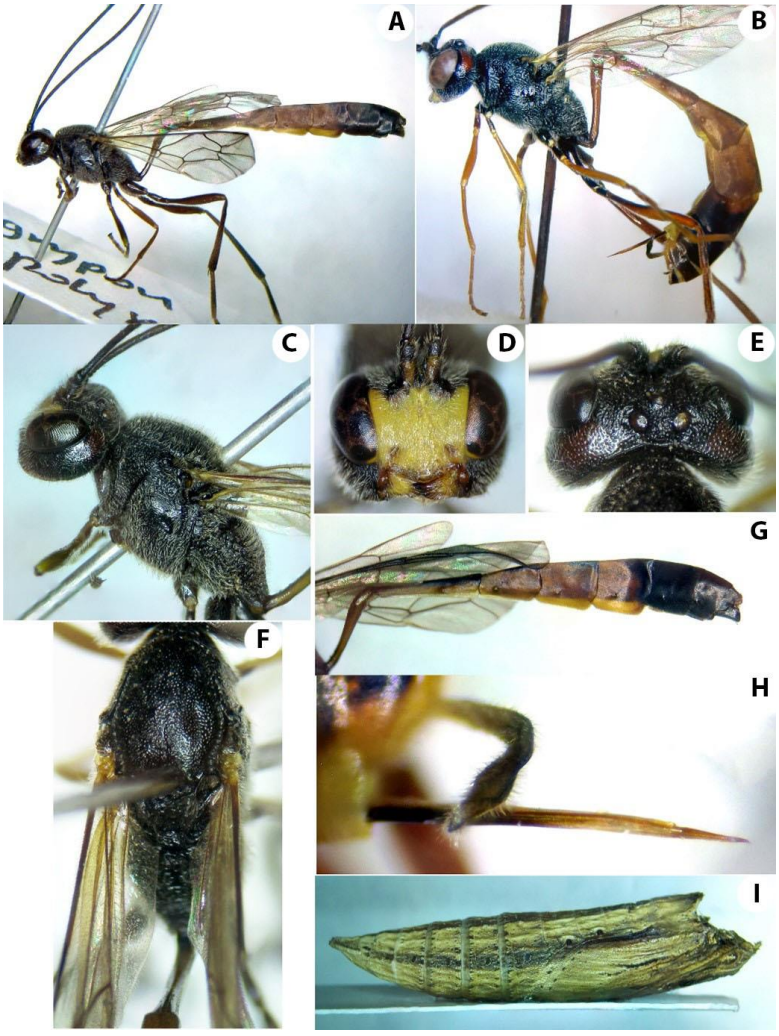

Fig. 1 General morphology of *Agrypon polyxenae*: A – male, B – female, C – lateral aspect of head and mesosoma (male), D – frontal aspect of the head (male), E – temporal aspect of the head (male), F – dorsal aspect of mesosoma (male), G – lateral aspect of metasoma (male), H – ovipositor, I – mummified pupa of *Zerynthia polyxena/*

Distribution: Austria, Bulgaria, Czech Republic, Germany, Hungary, Italy, Romania, Russia (Astrakhanskaya Oblast, Dagestanskaya Respublika, Samarskaya Oblast), Serbia, Spain, Ukraine.

Discussion

The flight period of *Zygaena polyxena* is only a few weeks long, in Serbia it occurs usually between late-March and mid-May, depending on the altitude (Biologer Community in Serbia 2018; Popović et al., 2020). On the territory of Serbia, the primary host plant for this butterfly is *Aristolochia clematitis*, a very abundant species, especially in ruderal areas. *A. pallida* has also been reported sporadically as a feeding plant, primarily in the regions with higher altitudes [14, 15]. Since *A. pallida* and *A. lutea* are morphologically very similar and given the distribution of *A. lutea* on the territory of Serbia [16], it is more likely that *Z. polyxena* was found on *A. lutea* in some reports due to misidentification of *Aristolochia* species. It should be noted here that the taxon *Z. cassandra* was separated from the *Z. polyxena* complex and raised to the species level. This species inhabits Italy [17] and it is associated with *Aristolochia rotunda* and *A. lutea* [18, 19]. Overlapping distribution of *Z. cassandra* and *A. polyxenae* indicates that *A. polyxenae* parasitizes a total of three species of *Zerynthia*, for which additional research is needed.

It is important to mention that in the literature, *Euproctis chrysorrhoea* (L.) (Erebidae) is also registered as a host for *A. polyxenae* [20]. However, this information is quite unreliable because it is unaccompanied by other sources and very likely to be an erroneous, less possible misidentification.

Conclusion

Analysed material in this study comes from the caterpillar samples collected exclusively from *A. clematitis*. Despite dozens of collected specimens, the percentage of observed parasitism was apparently very low. However, we cannot state this as a fact merely due to statistical reasons. The scarce literature data on *A. polyxenae* has impaired comparison of our data and no significant conclusion can be extrapolated. Also, surveying online databases did not show any data on *A. polyxenae* occurrence in the territory of Serbia.

Acknowledgement. *This investigation is supported by the Ministry of Education, Science and Technological Development of Serbia, contract number: 451-03-68/2022-14/200124.*

References

1. Shaw M, Kan P, Kan-van Limburg Stirum B. Emergence behaviour of adult Trogus lapidator (Fabricius) (Hymenoptera, Ichneumonidae, Ichneumoninae, Heresiarchini) from pupa of its host Papilio machaon L. (Lepidoptera, Papilionidae), with a comparative overview of emergence of Ichneumonidae from Lepidoptera pupae in Europe. J. Hymenopt. Res. 2015; 47:65.
2. Tshikolovets VV. Butterflies of Europe & the Mediterranean area. Tshikolovets Publications, Chezch Republic, 2011.
3. Stefanescu C. Zerynthia rumina, un esclat de color entre les brolles i els erms. Cynthia, butlletí del Butterfly Monitoring Scheme a Catalunya. 2006; 16-8.
4. van Nouhuys S, Shaw MR, Stefanescu C. Parasitoids of European butterflies. In Ecology of butterflies of Europe (pp. 130-156). Cambridge University Press, 2009.
5. van Swaay C, Collins S, Dušej G et al. Dos and Don'ts for butterflies of the Habitats Directive of the European Union. Nature Conservation. 2012; 1:73.
6. Nardelli U, Hirschfeld G. Aberrations, formes et sous-especes de *Zerynthia polyxena* Denis & Schiffermüller, 1775 (Lepidoptera: Papilionidae). Lambillionea. 2002; 102:223-40.
7. Dapporto L, Dennis RL. Conservation biogeography of large Mediterranean islands. Butterfly impoverishment, conservation priorities and inferences for an ecologica "island paradigm". Ecography. 2009; 32(1):169-79.
8. Örvössy N, Kőrösi Á, Vozár Á, Batáry P, Peregovits L. Microhabitat preference of the Southern Festoon (Zerynthia polyxena). Studies on the ecology and conservation of butterflies in Europe. 2005; 1:24.
9. Čelik T. Adult demography, spatial distribution and movements of Zerynthia polyxena (Lepidoptera: Papilionidae) in a dense network of permanent habitats. Eur. J. Entomol. 2012; 109(2).
10. Slancarova J, Vrba P, Platek M, Zapletal M, Spitzer L, Konvicka M. Co-occurrence of three Aristolochia-feeding Papilionids (Archon apollinus, Zerynthia polyxena and Zerynthia cerisy) in Greek Thrace. J. Nat. Hist. 2015; 49(29-30):1825-48.
11. Piccini I, Di Pietro V, Bonelli S. Zerynthia polyxena locally monophagous on Aristolochia pallida in the Susa Valley. Environ. Entomol. 2021; 50(6):1425-31.
12. Pavlović M, Maksimović M, Dragišić J et al. Possible health impacts of naturally occurring uptake of aristolochic acids by maize and cucumber roots: links to the etiology of endemic (Balkan) nephropathy. Environ. Geochem. Health. 2013; 35.2: 215-226.
13. Yu S, van Achterberg K, Horstmann K. Taxapad 2012. Database on flash drive. www.taxapad.com, Ottawa, Ontario.
14. Biologer Community in Serbia 2018. Biologer, a simple and free software for collecting data on biodiversity. https://biologer.rs. Accessed on 12th February 2022.
15. Popović M, Vasić N, Koren T et al. Biologer: an open platform for collecting biodiversity data. Biodivers. 2020; 8, e53014.
16. Bogosavljević S, Zlatković B. Report on the new floristic data from Serbia II. Biologica Nyssana. 2018; 9(2):63-75.
17. Zinetti F, Dapporto L, Vovlas A et al. When the rule becomes the exception. No evidence of gene flow between two Zerynthia cryptic butterflies suggests the emergence of a new model group. PLoS One. 2013; 8(6):e65746.
18. Ghesini S, Magagnoli S, Marini M. Biology and conservation of Zerynthia cassandra (Lepidoptera, Papilionidae) in semi-natural environments and agricultural landscapes. J. Insect Conserv. 2018; 22(1):151-61.
19. Cini, A., Bordoni, A., Ghisolfi et al. Host plant selection and differential survival on two *Aristolochia* L. species in an insular population of *Zerynthia cassandra*. *Journal of Insect Conservation*, 2019; *23*(2), 239-246.
20. Petcu P. Species of Ichneumonidae (Hym.) obtained from cultures of different injurious insects. II. (in Romanian with English summary) Al. I. Cuza din Iasi. Sect. II a. 1978; 24:79-80.