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DIPHYUS QUADRIPUNCTORIUS (MÜLLER, 1776) (HYMENOPTERA: ICHNEUMONIDAE: ICHNEUMONINAE): THE FIRST RECORDS FROM SERBIAN CAVES

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Abstract

Here we present new findings of the hymenopteran *Diphyus quadripunctorius* (Müller, 1776) from three cave localities in Serbia. We examined a total of five overwintering female specimens collected during 2016, 2019 and 2020 in caves – Devojačka Pećina Cave, Sisevačka Pećina Cave in eastern Serbia and Ogorelička Pećina Cave in southeastern Serbia. Additionally, a short description of *D. quadripunctorius* females is provided. Its phenology, tentative host association and adaptations to cave life are also discussed.

KEY WORDS: Cave-dwelling insects, trogloxenes, parasitoids, overwintering

Introduction

Diphyus quadripunctorius (Müller, 1776) is a solitary parasitoid wasp that is widespread in the western Palaearctic region. It is present in most of Europe (van Achterberg, 2013). According to literature data (Yu *et al.*, 2012), it is reported from the following countries bordering Serbia: Bulgaria, Croatia, Hungary and Romania. There are no published sources of its presence in Serbia, but we have found data on two findings of *D. quadripunctorius* in Serbia on the internet forum "Insects of Serbia" on Facebook, posted on 20.04.2016 from the town of Zrenjanin (https://www.facebook.com/ groups/insectserbia/permalink/507737822762635), and on 05.03.2019 from the village of Aradac, near Zrenjanin (https://www.facebook.com/ groups/insectserbia/permalink/1206342092902201), where the presence of the wasp was indicated with

photos. An interesting fact about this parasitoid is that it overwinters in caves (hence the attribution of the trivial name of four-spotted cave ichneumon wasp) and in mines in temperate regions, rarely in cellars or similar buildings (Constantineanu, 1959; Novak *et al.*, 2010). This species encounters winter as an adult, so the wasp must find shelter in which to spend the cold winter days. Only females overwinter, while males die shortly after mating (Broad *et al.*, 2018). The behavior of *D. quadripunctorius* was studied in detail by Baird & Shaw (2019). After overwintering, females leave the caves in spring and search for nectar, but also for hosts into which they lay their eggs. The host range of *D. quadripunctorius* includes primarily caterpillars belonging to the moth families Geometridae and Noctuidae, but to a lesser extent tortricid and yponomeutid caterpillars as well (Constantineanu, 1959). *Diphyus quadripunctorius* is a univoltine larval-pupal parasitoid, whose adults emerge from the pupal stage of the host (Baird & Shaw, 2019).

Although caves in Serbia have been relatively well-explored biospeleologically (Ćurčić *et al.*, 2014), no precise data on Serbian cave-dwelling Hymenoptera exist and therefore no correlative publications. The presence of hymenopterans has been previously recorded in certain Serbian caves (e.g., Žanel & Stanković, 1924; Remy, 1953), but their accurate identification was not conducted. The aims of this study were to: (i) report the first findings of the trogloxenic hymenopteran species *D. quadripunctorius* from Serbian caves; (ii) provide a short description of its females; (iii) present our observations on its biology and ecology.

Material and Methods

Localities explored

We investigated the Hymenoptera fauna of three Serbian caves: the Devojačka Pećina Cave (Gaura Fećilor), the Sisevačka Pećina Cave and the Ogorelička Pećina Cave (Fig. 1). Devojačka Pećina Cave is located in the village of Podgorac, in the municipality of Boljevac (43°57'8.12" N, 21°55'38.64" E). It is located in the Velika Reka (Valja Mare) Gorge (the eastern ridge of the Kučajske Planine Mts.). The cave's entrance sits at an altitude of 462 m and the cave has 209 m of explored channels so far (Nešić & Pavićević, 2009). Sisevačka Pećina Cave is located east of the city of Kragujevac, near the town of Paraćin, in the village of Sisevac (43°57'18.7" N, 21°35'25.6" E). The cave's entrance is at an altitude of 370 m. The cave is in the form of a simple hydrologically-active horizontal channel in limestone, with one small chamber at the end, about 50 m away from the entrance (Žanel & Stanković, 1924). Ogorelička Pećina Cave is part of the transitional fluviokarstic cave system that belongs to the mountain system of the Svrljiške Planine Mts. (Nešić *et al.*, 2008). This cave is located 1 km northeast of the village of Sićevo, in the vicinity of the city of Niš (43°20'52.29" N, 22°5'38.41" E). It is about 140 m long and is composed of five small chambers connected by passages. The entrance of the cave is located at an altitude of about 645 m.

Material of *D. quadripunctorius* examined

1 ♀, Devojačka Pećina Cave, village of Podgorac, near Boljevac, eastern Serbia, 22.05.2016, leg. N. Vesović; 2 ♀♀, Sisevačka Pećina Cave, village of Sisevac, near Paraćin, eastern Serbia, 03.11.2019, leg. N. Vesović; 2 ♀♀, Ogorelička Pećina Cave, village of Sićevo, Sićevo Gorge, near Niš, southeastern Serbia, 26.09.2020, leg. N. Vesović.

The specimens are deposited in the Entomological collection of the Faculty of Sciences and Mathematics, University of Niš, Serbia.

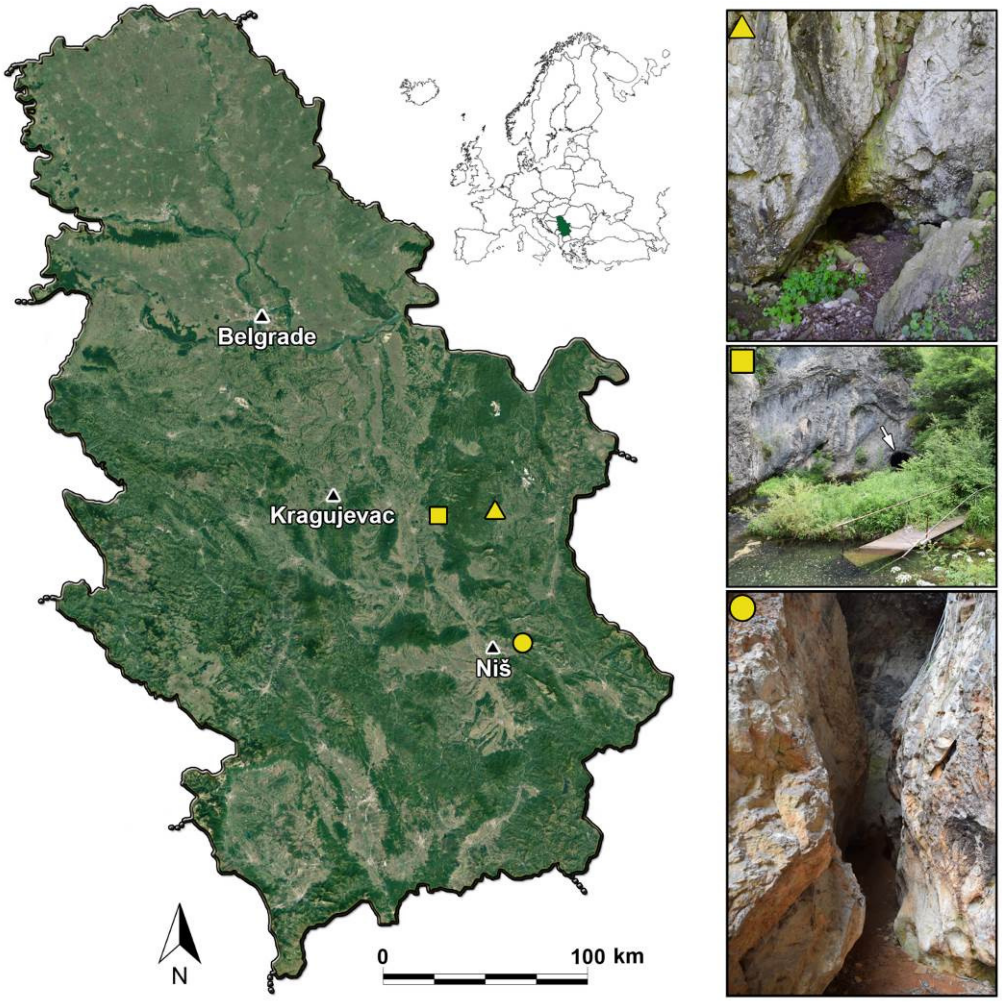

Figure 1. A map of three cave locations where the four-spotted cave ichneumon wasp has been found in Serbia (left) and the entrances of the visited caves (right). Yellow triangle – Devojačka Pećina Cave; yellow square – Sisevačka Pećina Cave; yellow circle – Ogorelička Pećina Cave.

Results and Discussion

Short description of *D. quadripunctorius* females: body length 15-16 mm (Fig. 2). Head black, punctate, genae with pale dense hairs, compound eyes large, black, hairless, each delimited by a pale line from vertex (Figs. 2A and 2B). Ocelli large, in a triangular position (Fig. 2A). Thorax, metasoma, coxae and hind tibiae black, remaining leg segments orange (Figs. 2C and 2D). Antennae black, except for flagellomeres 9-12, which are pale (Fig. 2C). Dorsally, scutellum white, abdominal tergites II and III each with two characteristic white spots (Fig. 2E).

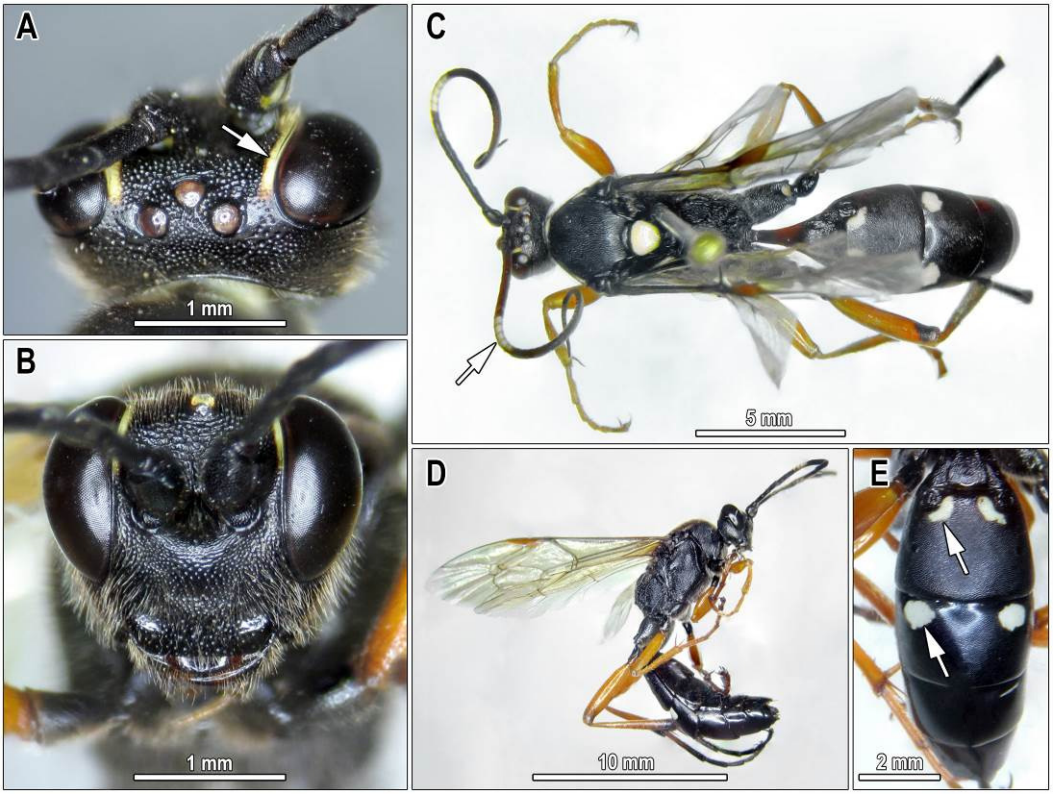

Figure 2. A female four-spotted cave ichneumon wasp, *D. quadripunctorius*, from Ogorelička Pećina Cave: A – head (dorsal view); B – head (frontal view); C – habitus (dorsal view); D – habitus (right lateral view); E – metasoma (dorsal view). Taxonomically important morphological features are indicated by white arrows.

In both the Sisevačka Pećina and the Ogorelička Pećina caves, we found two female specimens of *D. quadripunctorius*, one beside the other, in the same groove on the cave walls. This behavior is most likely a consequence (response) of the release of chemical cues by the wasps intended for intraspecific communication (aggregating pheromones), which is known in many insects that form swarms at certain times of the year [e.g., the very well-known harlequin ladybird, *Harmonia axyridis* (Pallas, 1773)] (Legrand *et al.*, 2019). In other ichneumonids, aggregation is observed in some members of the subfamily Rhyssinae (Quicke *et al.*, 2015). Namely, some males of certain European species, such as *Megarhyssa vagatoria* (Fabricius, 1793) and *M. rixator* (Schellenberg, 1802), form aggregations attracted by the pheromones of a female that is about to eclose. Aggregating in this case does not occur for the same reason as in *D. quadripunctorius*, but it has been shown that female pheromones in some parasitic wasps have a role in gathering, as in social Hymenoptera (Vander Meer *et al.*, 2019).

In the Devojačka Pećina Cave, we found one dead female on the cave wall (Fig. 3). All wasps were located in a place about 20 m from the cave's entrance, in complete darkness. Novak *et al.* (2010) reported that the females can stain the flowstone of hypogean habitats reddish with acid secretions, which we observed in the Devojačka Pećina Cave (Fig. 3).

Figure 3. A dead female of the four-spotted cave ichneumon wasp, *D. quadripunctorius*, found in Devojačka Pećina Cave. A white arrow indicates the stained cave wall, probably a result of the aggregation of conspecific specimens during the winter.

In the chamber of Ogorelička Pećina where we found the wasp specimens, the measured air temperature and air relative humidity were 13°C and 60%, respectively. Close to the collecting place in the cave we saw the remnants of adult lepidopterans, suggesting that they also enter the cave close to the entrance. As all our findings of *D. quadripunctorius* were accidental (we were not searching for them in a targeted manner), we do not know when this species entered the caves from the open environment. According to Baird & Shaw (2019), its presence in caves and mines was recorded from the end of July to late April. It is most likely that this parasitoid harmonizes its life cycle with that of its hosts due to the diapause of the latter, which are also looking for overwintering shelter.

The faunas of the Devojačka Pećina, Sisevačka Pećina and Ogorelička Pećina caves have been explored sporadically (Žanel & Stanković, 1924; Nešić *et al.*, 2008; Nešić & Pavićević, 2009), but no data on hymenopterans were reported. Concerning the findings of hymenopterans in other Serbian caves, only presence/absence is reported for certain caves, with no data provided on the identity of the occurring species (Žanel & Stanković, 1924; Remy, 1953). Chappuis & Jeannel (1951) found five large specimens of ichneumonids, four of which were aggregated, at the bottom of a deep depression in the Tmušnica Cave in the village of Boljare, close to the town of Sjenica, southwestern Serbia, but we cannot claim with certainty that this taxon was *D. quadripunctorius*.

Only 220 species of Hymenoptera worldwide are partly hypogean (Moldovan, 2004). No troglobitic hymenopteran species is recorded, although some trogloxenic and troglophilic hymenopterans regularly

occur in caves (Decu *et al.*, 1998). The four-spotted cave ichneumon wasp is the commonest hymenopteran in caves over most of Europe (Novak *et al.*, 2010).

It should be noted that *D. quadripunctorius* is not the only ichneumonid species that overwinters in caves. There are several more trogloxenes found in caves and other similar shelters among Hymenoptera, mostly within the Ichneumonidae and Proctrotrupidae families (Novak *et al.*, 2010). Concerning ichneumonids, these examples are from the subfamilies Ichneumoninae – *Amblyteles armatorius* (Förster, 1771) (Novak *et al.*, 2010) and *Exephanes occupator* (Gravenhorst, 1829) (Baird & Shaw, 2019), Phygadeuontinae – *Xenolytus bitinctus* (Gmelin, 1790) (Gauld, 1984), and Cryptinae – *Aclastus longicauda* Horstmann, 1980 (Vas & Kutasi, 2016).

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DIPHYUS QUADRIPUNCTORIUS (MÜLLER, 1776) (HYMENOPTERA: ICHNEUMONIDAE: ICHNEUMONINAE): ПРВИ НАЛАЗИ ИЗ ПЕЋИНА СРБИЈЕ

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Извод

У овом раду наведени су подаци о новим налазима опнокрилца *Diphyus quadripunctorius* (Müller, 1776) из три пећине Србије. Врста је регистрована током 2016., 2019. и 2020. године на следећа три локалитета у Србији: у Девојачкој пећини и Сисевачкој пећини у источној Србији и Огореличкој пећини у југоисточној Србији. Прикупљено је и анализирано укупно пет женки, које су презимљавале у пећинама. Такође, приложен је и кратак опис женки *D. quadripunctorius*. Додатно, изнети су и подаци о њиховој фенологији, могућој асоцијацији са домаћинима, као и прилагођеностима на боравак у пећинама.

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