A Worker-Like Female of *Myrmica sabuleti* Meinert, 1861 (Hymenoptera: Formicidae: Myrmicinae) in a Pitfall Trap with Five Mermithids (Nematoda: Mermithidae) Protruding from the Gaster

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**Abstract**

A worker-like female of *Myrmica sabuleti* Meinert, 1861, pitfall-trapped near Jena, Germany, in late summer 2016, was infested by five postparasitic juvenile mermithids. They poked out of the ant’s gaster as a trail of seven filaments of various lengths. Apart from its swollen gaster, the ant differed from conspecifics in several morphometric parameters. Using both morphological and molecular techniques, the parasite family Mermithidae was confirmed. Our stray find raises multiple questions concerning the genus and species identity of the parasite, its biology, and the infestation rate of the host ant population. More mermithid awareness by the various researchers working with *Myrmica* will help, but directed fieldwork, experimental life-history research, and molecular studies are needed to emancipate progress in ant-mermithid research from serendipity.

**Introduction**

Nematodes of 10 families are known as ant parasites worldwide, with Mermithidae as the largest and most conspicuous one (summarized by Poinar, 1975; Poinar, 2012). They occasionally can be found either by dissecting the gaster of a host ant or because they attract attention when leaving their host. Sometimes, mermithids considerably change the morphology of the (female or male) hosts compared with unparasitized conspecifics, forming so-called parasitogenic phenotypes (e.g. Kloft, 1949; Czechowski et al., 2007a; Csősz & Majoros, 2009; Csősz, 2012; Poinar, 2012; de Bekker et al., 2018). Wheeler (1928) distinguished three discrete aberrant forms of female infested ants, mermithergates (resembling workers), gynaecoid mermithergates (intermorphs), and mermithogynes (resembling gynes). Csősz and Majoros (2009) hypothesized that all infested adult females originate from gyne-destined larvae even if resembling workers.

Mermithids in ants have been noticed for a long time, with the first report dating back to 1747 (Poinar, 2012). Nevertheless, knowledge is still scarce, and the number of reliably documented instances of mermithids in ants was estimated at just “> 50” (Lachaud & Perez-Lachaud, 2015). Particularly, the life history of most ant mermithids has remained mysterious, and even for the best analyzed systems, crucial questions remain open (e.g., Kaiser, 1986; Poinar et al., 2007). All resolved instances have in common that the parasite enters its host as parasitic juvenile, grows in the gaster, and leaves the gaster as postparasitic juvenile, causing its host’s death (Poinar, 2012). Most frequently, a single mermithid per ant occurs, densely coiled up within the gaster.
and invisible from outside; the highest number of individuals in a single ant was nine (Poinar et al., 2007).

Several species of the myrmicine genus *Myrmica* have been reported as hosts of mermithids: *M. gallienii* Bondroit, 1920, *M. rubra* (Linnaeus, 1758), *M. ruginodis* Nylander, 1846, *M. rugulosa* Nylander, 1846, *M. sabuleti* Meinert, 1861, *M. scabrinodis* Nylander, 1846, and *M. schencki* Vierreck, 1903 (Czechowski et al., 2007a; b and references therein). Interpreting these host records, of which several date back to the first half of the 20th century, needs be done with a grain of salt, though, because the morphological changes of ants parasitized by mermithids often make species identification difficult (and have even resulted in erroneous species descriptions; Csősz, 2012; Borowiec & Salata, 2015) and because the species-level identification of *Myrmica* has remained difficult, even when not parasitized (Radchenko & Elmes, 2010; Seifert, 2018).

Here, we describe a case of gregarious parasitism with five mermithids protruding from the gaster of a pitfall-trapped worker-like *Myrmica* ant. Based on morphometric and molecular-genetic analyses of the host and the parasite, respectively, we discuss questions about parasite taxonomy, parasite biology, and the host’s infestation rate. Like in many other instances of mermithids in ants, many questions remain open – more directed research efforts from various directions will be needed to solve the many exciting questions in this field.

**Material and Methods**

**Sampling**

As part of a 2012-2017 monitoring project for studying arthropod communities by the Institute of Ecology and Evolution (Friedrich-Schiller-University of Jena), a pitfall-trapping campaign was conducted in the Leutra valley near Jena/Thuringia, Germany; for details, see Köhler (2018) and Autorenkollektiv (2019). Briefly, a total of 72 pitfall traps were exposed in groups of four traps (at the corners of 1.4 x 1.4 m squares) along three transects from mid April to mid October. The traps used were hard plastic tubes with an opening diameter of 4.7 cm and a depth of 9 cm; the trapping liquid was a 1,2-propandiol:water = 3:1 (v/v) mixture plus 1.4 × 1.4 m squares

The thermal profile for 18S was 10 min initial denaturation at 95 °C, followed by 35 cycles of 30 s at 95 °C, 30 s at 58 °C, 1 min at 72 °C, and a final elongation of 10 min at 72 °C; the profile for COI was as in Malysheva (Bioline, UK), 0.2 µM NEM_COI_F primer, 0.2 µM NEM_COI_R primer (both Porazinska et al., 2009), and 2 µl DNA extract in 10 µl total volume. COI PCR reactions contained 1× QuantiTect Probe PCR mastermix containing 0.2 µM NF1 primer, 0.2 µM 18Sr2b primer (both Malysheva et al., 2016), and 1 µl DNA extract in 10 µl total volume.

The externally visible mermithid filaments were measured with a Zeiss stereomicroscope (SM XX) with measuring ocular using 12.5× and 50× magnification. Photographs were taken using a Zeiss stereomicroscope (Stemi 305) with integrated HD IP Wi-Fi camera and transmitting light (Institute of Ecology and Evolution, University of Jena).

Later, the ant was dissected in tap water in a wax bowl under a Wild M5 dissecting microscope at 25× magnification using two watchmaker forceps Dumont No. 5. After dissection, the entire mermithids were measured and photographs taken under 50× magnification. During further handling, one worm got lost.

**Identifications**

Host species identification was done by (a) using the characters in the key of Seifert (2018) to exclude less similar species of the *Myrmica scabrinodis* and *M. sabuleti* species complexes, (b) running the infected specimen as wild-card in a two-class linear discriminant analysis (LDA) comparing nine *M. bibikoffi* Kutter, 1963 workers from three sites in Germany and Switzerland and 77 *M. sabuleti* workers from 34 sites in the whole Westpalaeartic range, and (c) running the same individuals in a principal component analysis (PCA).

The stereomicroscopic investigation methods, the removal of allometric variance (RAV), and the set of 19 investigated characters used in LDA and PCA are described in Seifert et al. (2014). LDA and PCA were run using the SPSS 16.0 software package (IBM, USA).

For morphological identification, one nematode was analyzed via the 18S ribosomal DNA (18SrDNA) and the cytochrome c oxidase I (COI) marker. Nematode DNA was extracted using the QIAamp DNA Micro Kit (Qiagen, Germany) following the instructions of the manufacturer. 18S PCR was performed in 1× QuantiTect Probe PCR mastermix containing 0.2 µM NFI primer, 0.2 µM 18Sr2b primer (both Porazinska et al., 2009), and 2 µl DNA extract in 10 µl total volume. COI PCR reactions contained 1× MyTaq buffer (Bioline, UK), 0.2 µM NEM_COI_F primer, 0.2 µM NEM_COI_R primer (Malyshcheva et al., 2016), 0.25 U MyTaq (Bioline), and 1 µl DNA extract in 10 µl total volume.

The thermal profile for 18S was 10 min initial denaturation at 95 °C, followed by 35 cycles of 30 s at 95 °C, 30 s at 58 °C, 1 min at 72 °C, and a final elongation of 10 min at 72 °C; the profile for COI was as in Malyshcheva et al. (2016). Presence of PCR product was verified by gel electrophoresis, and 1 µl ampiclon was cloned with the CloneJET PCR Cloning Kit (Thermo) following the instructions of the manufacturer. Insert size was determined by PCR with the vector primers provided with the kit. Plasmid DNA of three colonies with
correct insert size was extracted by alkaline lysis (Sambrook et al., 2001) and Sanger sequenced using the forward vector primer. The obtained sequence was analyzed with the Basic Local Alignment Search Tool (BLAST), which searches for regions of similarity between sequences of a query sequence and the NCBI GenBank database. Because for COI, default settings did not yield any plausible result, “optimize for more dissimilar sequences (discontiguous megablast)” was chosen.

Results

Host ant

There are six Myrmica species known from Central Europe (Seifert, 2018) to which the nematode host might possibly belong: M. sabuleti, M. bibikoffi, M. scabrinodis, M. lonae Finzi, 1926, M. vandeli Bondroit, 1920, and M. curvitiorax Bondroit, 1920. The latter four species can be clearly excluded by simple eye inspection based on structure of basal scape and head sculpture as described in Seifert (2018). Regarding the former two species, normal workers of Myrmica sabuleti without teratological or parasite-induced changes of morphology are easily separable from its temporary social parasite Myrmica bibikoffi by different RAV-corrected body ratios. These discriminators are, according to an LDA with stepwise character reduction, a smaller postpetiole width index PpW/CS, a smaller postpetiolar hair length index PpHL/CS, a lower postocular distance index PoOc/CS, a larger frontal carinae index FL/FR, and a shorter spine length index SP/CS. However, nematode infection is known in Myrmica to result in more massive petiole and postpetiole, a smaller frontal carinae index, and more diverging propodeal spines (Czechowski et al., 2007a; Csősz & Majoros, 2009). This could make the identification of the host specimen problematic. Considering the five diagnostic characters mentioned above and running the Leutra host specimen as wild card in a two-class LDA comparing M. bibikoffi and M. sabuleti, the host was allocated to Myrmica sabuleti with p=0.611. Furthermore, the infested specimen was clearly allocated to M. sabuleti in a PCA using these five characters, but it was the most M. bibikoffi-like specimen within the M. sabuleti cluster (Fig 1) due to its increased width and height of waist segments and the lower frontal carinae index (Table 1). The just low likelihood of belonging to M. bibikoffi is also supported by the rarity of this species in general and the fact that it has never been found in the Leutra valley, which has been intensively studied myrmecologically since the 1970s (Seifert, 1982).

Table 1. Morphometric data (average ± standard deviations, minimum and maximum values in square brackets) of workers of Myrmica bibikoffi, of the nematode-infested M. sabuleti worker from Leutra, and of normal M. sabuleti workers from its whole Palaearctic range. The data are corrected in the allometric space. The data for PpHL, MetSp, and MetL are based on a reduced sample of only 77 specimens. For definitions of the morphometric characters, see Seifert et al. (2014).

<table>
<thead>
<tr>
<th>Character</th>
<th>bibikoffi (n=9)</th>
<th>sabuleti Leutra (n=1)</th>
<th>sabuleti (n=313)</th>
</tr>
</thead>
<tbody>
<tr>
<td>CS [µm]</td>
<td>1154 ± 76 [1062,1257]</td>
<td>1165 [942,1322]</td>
<td>1158 ± 65 [990,1074]</td>
</tr>
<tr>
<td>CL/CW (1150)</td>
<td>1.042 ± 0.020 [0.996,1.064]</td>
<td>1.055 [0.761,1.081]</td>
<td>1.031 ± 0.015 [0.761,1.081]</td>
</tr>
<tr>
<td>SL/CS (1150)</td>
<td>0.810 ± 0.013 [0.795,0.833]</td>
<td>0.848 [0.137,0.179]</td>
<td>0.802 ± 0.017 [0.137,0.179]</td>
</tr>
<tr>
<td>SW/SL (1150)</td>
<td>0.179 ± 0.008 [0.167,0.191]</td>
<td>0.173 [0.159,0.245]</td>
<td>0.199 ± 0.017 [0.159,0.245]</td>
</tr>
<tr>
<td>PoOc/CL (1150)</td>
<td>0.444 ± 0.004 [0.440,0.450]</td>
<td>0.424 [0.408,0.456]</td>
<td>0.432 ± 0.009 [0.408,0.456]</td>
</tr>
<tr>
<td>EYE/CS (1150)</td>
<td>0.194 ± 0.007 [0.183,0.202]</td>
<td>0.200 [0.181,0.220]</td>
<td>0.195 ± 0.006 [0.181,0.220]</td>
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<tr>
<td>FL/CS (1150)</td>
<td>0.459 ± 0.013 [0.443,0.484]</td>
<td>0.464 [0.421,0.489]</td>
<td>0.457 ± 0.012 [0.421,0.489]</td>
</tr>
<tr>
<td>FR/CS (1150)</td>
<td>0.327 ± 0.017 [0.312,0.357]</td>
<td>0.325 [0.262,0.356]</td>
<td>0.300 ± 0.014 [0.262,0.356]</td>
</tr>
<tr>
<td>FL/FR (1150)</td>
<td>1.405 ± 0.040 [1.333,1.457]</td>
<td>1.423 [1.306,1.739]</td>
<td>1.530 ± 0.076 [1.306,1.739]</td>
</tr>
<tr>
<td>PeW/CS (1150)</td>
<td>0.321 ± 0.025 [0.300,0.370]</td>
<td>0.321 [0.253,0.318]</td>
<td>0.280 ± 0.011 [0.253,0.318]</td>
</tr>
<tr>
<td>PpW/CS (1150)</td>
<td>0.461 ± 0.031 [0.430,0.520]</td>
<td>0.482 [0.351,0.453]</td>
<td>0.399 ± 0.016 [0.351,0.453]</td>
</tr>
<tr>
<td>PeH/CS (1150)</td>
<td>0.364 ± 0.015 [0.345,0.391]</td>
<td>0.365 [0.307,0.373]</td>
<td>0.335 ± 0.012 [0.307,0.373]</td>
</tr>
<tr>
<td>PeL/CS (1150)</td>
<td>0.499 ± 0.024 [0.465,0.528]</td>
<td>0.533 [0.441,0.523]</td>
<td>0.479 ± 0.014 [0.441,0.523]</td>
</tr>
<tr>
<td>SP/CS (1150)</td>
<td>0.386 ± 0.021 [0.345,0.419]</td>
<td>0.445 [0.303,0.455]</td>
<td>0.398 ± 0.023 [0.303,0.455]</td>
</tr>
<tr>
<td>PpHL/CS (1150)</td>
<td>0.211 ± 0.010 [0.198,0.227]</td>
<td>0.164 [0.115,0.197]</td>
<td>0.168 ± 0.013 [0.115,0.197]</td>
</tr>
<tr>
<td>MetL/CS (1150)</td>
<td>0.239 ± 0.016 [0.214,0.259]</td>
<td>0.245 [0.194,0.256]</td>
<td>0.228 ± 0.010 [0.194,0.256]</td>
</tr>
<tr>
<td>MetSp/CS (1150)</td>
<td>0.207 ± 0.029 [0.147,0.243]</td>
<td>0.187 [0.147,0.250]</td>
<td>0.180 ± 0.018 [0.147,0.250]</td>
</tr>
</tbody>
</table>
The infested female *Myrmica* ant was a mermithergate according to the categorisation by Wheeler (1928) and had no ocelli. It possessed a slightly thickened and longer gaster (~2.5 mm) compared with unparasitized workers from the same trap (~2.0 mm).

**Mermithid parasites**

At the *Myrmica* worker’s back end, seven filaments of various lengths protruded from the gaster, of the same yellow-brown color as the ant itself, presumably because of storage in the same trap liquid (Fig 2a). The nematodes had penetrated the intersegmental membranes between gastral segments three and four (= abdominal segments six and seven), rupturing both tergites and sternites distinctly. The worm parts were more or less straight and rod-like, and of two worms, both anterior and posterior ends were visible outside the ant’s gaster, whereas of three worms, only one end (one front and two rear ends) were visible before dissection. The coiled-up parts of the worms were partially visible through the cuticle up to the second gaster segment (Fig 2b). Three filaments were rising more ventrally and rather short (0.9/1.2/2.0 mm), whereas four filaments were rising more dorsally and much longer (3.5/4.8/5.1/6.5 mm), with the longest exceeding the ant size (~5.5 mm) (Fig 2a). Their diameters were between 0.13-0.19 mm at the base and 0.11-0.15 mm at the ends, narrowing only slightly.

After removal of the tergites and sternites, the parts of the worms inside the ant were found coiled and had remained pale, contrasting the yellow-brown parts exposed to the pitfall fluid (Fig 2c). A total of five mermithids could be confirmed, with the largest reaching 12.0 mm total length (Fig 2d). Only small remains of the ant’s internal organs were discernible and not clearly identifiable.

Morphologically, the nematodes were identified as postparasitic juvenile mermithids (family Mermithidae). The genus can only be determined in adults (G. Poinar, unpubl., using the key in Poinar, 1975).

This morphological identification was corroborated by sequencing 363 basepairs of the 18Sr DNA (GenBank Accession Number MH793873) and 563 basepairs of COI (GenBank Accession Number MK256737). The 18S BLAST search yielded several hits with 100% query cover, 99% identity match, and e=0 to the genera *Agamermis* and *Hexamermis*; the COI BLAST search yielded hits with 98% query cover, 73% identity match, and e=10^-41 to the same genera.

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**Fig 2.** The infested *Myrmica* female and its parasites. (a) Two workers of *Myrmica sabuleti*, on the left infested by mermithids (total ant body length ~5.5 mm, gaster length ~2.5 mm, length of visible part of longest mermithid filament ~6.5 mm, diameters at base 0.13-0.19 mm and at the ends 0.11-0.15 mm), on the right an uninfested conspecific. (b) Through the *Myrmica* gaster, parts of the mermithids are visible. (c) The dissected *Myrmica* gaster shows the compact juvenile mermithids. (d) The five juvenile mermithids of various lengths found in the infested *Myrmica* (length of longest mermithid 12.0 mm). (Photograph (a) by A. Ebeling & I. Wolf; photographs (b-d) by A. Buschinger).
Discussion

Our record of *Myrmica sabuleti*, identified using morphometrics, as host of five postparasitic mermithids, identified using morphology and molecular genetics, was a stray find during a pitfall-trapping campaign with research aims other than ant mermithid infection. Multiple questions about parasite taxonomy, parasite biology, and the infestation rate of the host population can now be addressed, albeit mostly without definitively answering them.

What is the genus and species identity of the parasite?

The morphological identification of postparasitic juveniles, at which stage mermithids leave their ant hosts, is not possible to species nor to genus (Poinar, 1975). Mermithid species from ants have thus often remained unidentified on morphological grounds (e.g., McInnes & Tschinkel, 1996; Csősz & Majoros, 2009; O’Grady & Breen, 2011; Lacinety et al., 2017), and the genus and species identity of Mermithidae in *Myrmica* has never been resolved so far (Poinar, 2012).

DNA-sequence-based identification can be a valuable tool (Poinar et al., 2007). The sequences we established are similar to ones from *Agammermis* and *Hexamermis*, which build a monophyly within the Mermithidae (Park et al., 2011; Kubo et al., 2016). Both *Agammermis* and *Hexamermis* have been reported from a broad range of insect hosts (e.g., Achinelly & Camino, 2008; Stubbins et al., 2016; and references therein) but, as far as we know, not from ants (Poinar, 2012 and Web of Science queries on 20 November 2018). However, despite the high similarity to *Agammermis* and *Hexamermis*, uncertainty remains, in that of the safely identified recent mermithid genera found in ants (Poinar, 2012), two (*Allomermis*, *Pheromermis*) are represented in GenBank (Nucleotide query of 21 November 2018) but not the other four (*Agamomermis*, *Camponotomermis*, *Comanimermis*, *Meximermis*).

This uncertainty is typical of current mermithid research – a lack of sequence data from safely identified specimens of the mermithid species at question and thus a lack of comparison has often impeded taxonomic resolution even when molecular analyses were undertaken (hornets: Villemant et al., 2015; ants: Lacinety, 2017; moths: Kumar et al., 2018; bumble bees: Tripodi & Strange, 2018). A comprehensive reference DNA sequence database of the mermithid species identified so far in ants would allow improved taxonomic resolution for postparasitic individuals and thus access to the knowledge established for those parasites.

At which developmental stage of the ant did the parasite infest it?

The female *M. sabuleti* presented here showed similar differences from nonparasitized conspecifics to those described by Csősz and Majoros (2009) for mermithogenic *Myrmica gallienii* and to those described by Czechowski et al. (2007a) for mermithogenic *M. sabuleti*. These differences (i.e., not the enlarged gaster) allow us to conclude that the parasites entered the ant before the ant was adult, in line with findings that mermithids that enter ant larvae can be carried over to the adult stage (Wheeler, 1901). Likely, the ovipositing mermithid was close to the ant nest, thus resulting in multiple infections – *Hexamermis* adults, for example, produce eggs in the soil, and from these eggs hatch pre-infective stages that search the surroundings for hosts (Poinar & Gyrisco, 1962).

What is the relevance of the high number of five parasites in the same host?

In most ants parasitized, a single mermithid is found. The highest available numbers of mermithids per ant individual are four in *Myrmica* (Csősz & Majoros, 2009), five in *Solenopsis* and *Lasius* (McInnes & Tschinkel, 1996; O’Grady & Breen, 2011), and nine in *Solenopsis* (Poinar et al., 2007). In all these instances, large numbers of parasitized ants were screened and the higher the number of individuals per ant, the rarer it was detected. Our find is among the highest numbers of mermithid per ant host recorded, but just a single infected worker was found. This combination is improbable (but obviously not impossible) if the frequency distribution of multiple infections (2, 3, 4, etc. nematodes per ant) in the *M. sabuleti* population Leutra is the same as in those other studies. Series of infected *M. sabuleti* from the population analyzed here will be needed to interpret our current find of five parasites as typical or atypical of this particular relationship.

Did the parasite start emerging from the host before or after the host was pitfall-trapped?

Our find of a pitfall-trapped host with parasites protruding from its gaster could mean that the parasites started emerging before the ant got caught (pre-trapping) or just after contact with the trapping liquid (post-trapping). Pre-trapping emergence may be possible, in that the process of emerging from the ant was reported to take, for example, one hour (after which the ant hosts lived for a further hour; O’Grady & Breen, 2011). In support of pre-trapping emergence may be seen that the worms were shaped rod-like and did not, as normally the case, form a tangle – the rod-like posture may have resulted from death in the trapping liquid. This aspect could be tested by killing living mermithids in diluted 1,2-propandiol. Walking with such a voluminous trail appears difficult, but before contact with the liquid, the worms may have been much thinner – mermithids were reported to gain volume once in contact with water by imbibing it (Poinar et al., 2007). Not in line with pre-trapping emergence is the report by Kaiser (1986) that the ants remain still during the emergence – the conclusion is impossible if the frequency distribution of multiple infections (2, 3, 4, etc. nematodes per ant) in the *M. sabuleti* population is skewed. The rate of the host population can now be addressed, albeit aims other than ant mermithid infection. Multiple questions about parasite biology and the infestation rate of the host population can now be addressed, albeit mostly without definitively answering them.
was necessary to trigger emergence of the parasite (Crawley & Baylis, 1921; Kaiser, 1986; Maeyama et al., 1994; Poinar et al., 2007; O’Grady & Breen, 2011). From many parasitized grasshoppers pitfall-trapped in the same liquid used here, not a single emergence of the mermithids was observed (G. Köhler, unpubl.), but this may not be in conflict with post-trapping emergence in this ant case because the mermithids of the two hosts may have different emergence triggers. Experiments with living, infected *M. sabuleti* from the population analyzed here will be needed to solve this question.

**Did the parasite trigger water-seeking behavior in the host?**

Whenever the parasites started emerging from the host, the ant may have been trapped because it fell into the pitfall trap by chance or because the parasite triggered its host to seek a moist environment. Such behavioral manipulation has been postulated for several mermithids in ants (Kaiser, 1986; Mayeama et al., 1994; Poinar et al., 2007). A potential adaptive value of seeking a moist environment appears somewhat problematic in a dry habitat during a dry period (total rainfall measured by a climate station 6 km from the habitat was 16.1 mm during the trapping period 19.IX.-05.X.2016; Jena University of Applied Science, unpubl.) but cannot be refuted entirely. Avoiding exposure to the dry conditions above surface may actually be more adaptive for the parasites. Experimental evidence is needed.

**What is the infestation rate of the host population?**

The literature record holds both high infestation rates in some ant nests of selected host populations (McInnes & Tschinkel, 1996; Czechowski et al., 2007b; Csősz & Majoros, 2009) and difficulties in finding mermithids in ant populations infested in a previous year (reviewed by O’Grady & Breen, 2011). At the population level, this means clumped spatial distribution with temporal variation. Our find of a single parasitized worker out of thousands *Myrmica* workers trapped in the habitat analyzed mid April to mid October 2016 (G. Köhler, unpubl.) may thus mean that the *M. sabuleti* population has a low infestation rate. Alternatively, it might have a high infestation rate but no trap may have been close to a strongly infested nest or infested ants may be influenced by their parasites to rather stay in their nests during dry periods (see previous section). To characterize the infestation rate of the analyzed population, but also those of other *Myrmica* populations, dedicated screening of many living individuals from many nests will be needed, ideally from multiple years, as done recently in a single-year survey of thousands of bumble bees at the continent scale (Tripodi & Strange, 2018) and in a multiple-year survey of tens of thousands of hornets at the district scale (Villemant et al., 2015). If combined with genetic approaches, such studies would also help to shed some light on the entirely understudied population genetics of nematode parasites in general (Cole & Viney, 2018).

**Conclusion**

While a lot of research effort has been directed at many of the parasitic organisms infesting *Myrmica* species (Witek et al., 2014), data on mermithids are scarce. As *Myrmica* ants are important study systems for various disciplines (e.g., Radchenko & Elmes, 2010; Seifert, 2018), including the study of lycaenid butterflies and socially parasitic ants (e.g., Barbero et al., 2012; Tartally et al., 2019), awareness of this phenomenon by the various researchers working with *Myrmica* may bring to light further instances of mermithid infections, possibly with living individuals. However, additionally to mermithid awareness when analyzing other aspects of *Myrmica* ants, directed and dedicated efforts in fieldwork, experimental life-history research, and molecular-genetic analyses will be needed to reduce the dependence of mermithid research in *Myrmica* and ants generally on chance.

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**Authors’ contributions**

GK noticed the host ant when sorting pitfall-trapped insects. GK and AB measured and photodocumented the nematodes. AB dissected the host ant. BS analyzed the host ant and other *Myrmica* ants using morphometrics and ran statistics on the data. FMS, WA, and BCS-S designed the molecular analyses of the nematodes and interpreted the data. All authors wrote the paper.

**References**


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