

Description of the new species Coptera tonic (Hymenoptera, Diapriidae), a pupal parasitoid of Rhagoletis juniperina Marcovitch (Diptera, Tephritidae), and revised partial keys to Nearctic Coptera Say

Hannah C. Ericson¹, Andrew A. Forbes¹

I University of Iowa, Department of Biology, Iowa City, IA 52242, USA

Corresponding author: Andrew A. Forbes (andrew-forbes@uiowa.edu)

Academic editor: A. Köhler | Received 27 July 2020 | Accepted 23 September 2020 | Published 5 November 2020

http://zoobank.org/BAE9B72C-8155-4DCB-865C-505E15BCAB01

Citation: Ericson HC, Forbes AA (2020) Description of the new species *Coptera tonic* (Hymenoptera, Diapriidae), a pupal parasitoid of *Rhagoletis juniperina* Marcovitch (Diptera, Tephritidae), and revised partial keys to Nearctic *Coptera* Say. ZooKeys 985: 49–60. https://doi.org/10.3897/zookeys.985.56974

Abstract

A new species of the parasitic wasp *Coptera* Say was previously distinguished from other species via correspondence between ecological (host) differences and DNA barcodes. A description and figures for *Coptera tonic* **sp. nov.**, along with revisions to existing keys that allow it to be distinguished from other Nearctic species without the aid of molecular characters, is provided in this work.

Keywords

Coptera cingulatae, Coptera pomonellae, Eastern red cedar, Psilini

Introduction

Coptera Say, 1836 is a genus of parasitic wasps in family Diapriidae with a near-worldwide distribution. Muesebeck (1980) recognized 29 Nearctic *Coptera* species, representing a fraction of the more than 150 species estimated to occupy this region (Masner and Garcia 2002). *Coptera* females search for hosts, usually Dipteran pupae buried shallowly in soils, by keying in on chemical signals left by the host before pupation (Granchetti et al. 2012). Females use their heads to dig up loose soil around buried pu-

Copyright Hannah C. Ericson, Andrew A. Forbes. This is an open access article distributed under the terms of the Creative Commons Attribution License (CC BY 4.0), which permits unrestricted use, distribution, and reproduction in any medium, provided the original author and source are credited.

pae, then drag the host to the surface and oviposit (Buckingham 1975). Hosts, when known, are primarily true fruit flies (Diptera: Tephritidae), and parasitism rates of pupae can exceed 10% (Cameron and Morrison 1977; Maier 1981), such that species in this genus have been explored as potential biological control organisms (Silvestri 1914; Hagen et al. 1980; Sivinski et al. 1998; Baeza-Larios et al. 2002; Guillén et al. 2002; Cancino et al. 2019). Further, though some *Coptera* species may be flexible in their host associations (e.g., *Coptera occidentalis* Muesebeck, 1980; Kazimírová and Vallo 1992), others are apparently limited to single fly host species and have garnered interest from evolutionary biologists interested in co-speciation (Hamerlinck et al. 2016).

Coptera species delimitation and ascertainment of host breadths have both proved challenging. These issues can be especially problematic when identifying potential biocontrol species if apparent oligiphagous species are actually complexes of cryptic specialists (e.g., *Coptera silvestrii* (Kieffer, 1913); Yoder and Wharton 2002). *Coptera* are common in Malaise and pan trap collections, but they have little color variation and limited sculpturing on their sclerites, offering few landmarks for species-level identification. The last revision of the Nearctic *Coptera* (Muesebeck 1980) relied heavily on relative lengths and shapes of body parts, such that some species, as described, have much intraspecific variation. Host associations are perhaps even more challenging, as they are known only from studies where parasitized pupae have been extracted from soils – an uncommon collection technique except when specifically targeting pupal parasitoids (e.g., Buckingham 1975; Maier 1981; Hamerlinck et al. 2016).

Collections of *Coptera* from known hosts in soil, coupled with DNA barcoding (sequencing of short segments of the mitochondrial COI gene), have proved useful in distinguishing among species, determining host associations, and identifying possible new species. Collections and barcoding of *Coptera* differentiated a new species associated with the juniper maggot fly, *Rhagoletis juniperina* Marcovitch, 1915 from the apparently cryptic species *Coptera pomonellae* Muesebeck, 1980 that attacks *Rhagoletis pomonella* (Walsh, 1867) and *Rhagoletis suavis* (Loew, 1862) flies in hawthorns and walnuts, respectively (Forbes et al. 2012; Hamerlinck et al. 2016). The argument that this was a new species and not just *C. pomonellae* wasps with two divergent COI haplotype families was bolstered by ecological data: while pan trap collections underneath juniper and hawthorns included both *C. pomonellae* and the new species, *C. pomonellae* was only reared from pupae of *R. pomonella*, and the new species was only reared from *R. juniperina* pupae (Forbes et al. 2012).

Though the combination of ecological and genetic data is useful for identification of reproductively isolated groups for taxonomically-challenging groups like *Coptera* (and see: Smith et al. 2005, 2008; Condon et al. 2014; Shashank et al. 2014; Ward et al. 2020), genetic evidence of apparently cryptic species is also an opportunity to determine taxonomically informative, but previously overlooked, morphological characters (Lukhtanov et al. 2016). Further, naming species based only on DNA barcodes is unacceptable (though see Brower 2010) and morphological characters remain the cheapest and most accessible means for most researchers and naturalists to differentiate species. Here, we describe a new species of *Coptera* associated with junipers, which was discov-

ered in Forbes et al. (2012). We also provide an amendment to the existing Nearctic *Coptera* species keys such that other researchers can distinguish this species from other similar species, including *C. pomonellae*, a species with promise for biological control of the apple maggot fly (Cameron and Morrison 1977; Maier 1981). We do not attempt a full revision of the Nearctic *Coptera* at this time because – as this example shows – such an effort would be premature without additional ecological and molecular work.

Materials and methods

Study material

Collections used for study are described in Forbes et al. (2012). As part of that work, *Coptera* DNA was sampled non-destructively, such that most individuals were preserved for morphological study. Samples of the new juniper-associated *Coptera* species and *C. pomonellae* were collected via both soil pupal collections and in yellow pan traps in East Lansing, MI and Iowa City, IA in 2011. Samples of *Coptera cingulatae* Muesebeck, 1980 were collected in yellow pan traps under black cherry trees (host of *Rhagoletis cingulata*) in Rose Lake, MI and Iowa City, IA also in 2011.

Morphological descriptions and photography

We developed a character matrix of all previously described Nearctic *Coptera* based on Muesebeck (1980) and then used a Leica M125 stereomicroscope (Leica Inc., Switzerland) to record morphological characters of males and females identified via DNA barcodes as belonging to the new juniper-associated *Coptera*. Because females of the new species keyed to *Coptera pomonellae* and males of the new species keyed to *Coptera cingulatae* in the Muesebeck (1980) key, we placed particular emphasis on searching for characters that differentiated them from these two species. Terminology in the description of the new species follows Muesebeck (1980).

We used a Hitachi S-3400N (Hitachi High-Tech Corp., Tokyo, Japan) to perform scanning electron microscopy (SEM) of males and females of *C. pomonellae* and the new species. Color photographs of the same two species were photographed using a Canon EOS 60D camera with a Canon MP-E 65 mm macro lens and a Canon Macro Ring Lite MR-14EX (Canon USA, Melville, NY), mounted on a StackShot Automated Focus Stacking Macro Rail (Cognysis Inc., Traverse City, MI). Stacked images were processed using Zerene Stacker (Zerene Systems LLC., Richland, WA) and Adobe Photoshop (Adobe, San Jose, CA, USA). Measurements of relevant body parts (in mm) were made using a Leica M125 stereomicroscope (Leica Inc., Switzerland) and Leica Application Suite v4.13. Holotypes, paratypes of the new species, and additional study specimens of *Coptera pomonellae* and *Coptera cingulatae* were deposited into the collection of the University of Iowa Museum of Natural History (**UIMNH**; ID#s: SUI:INS:04567 – SUI:INS:04588).

Results

Taxonomy

Coptera Say, 1836

Coptera Say, 1836: 281.

Type-species. Coptera polita Say. By monotypy.

Coptera tonic sp. nov.

http://zoobank.org/92F47ACC-5957-41E6-B297-80BE71905189 Figures 1–8

Type material. *Holotype*: USA • ♀; Ingham Co., East Lansing, MI; 42.7274, -84.4777; 3 Jul. 2011; Serdar Satar; reared from soil-collected pupa of *Rhagoletis juniperina*; UIMNH ID: SUI:INS:04567.

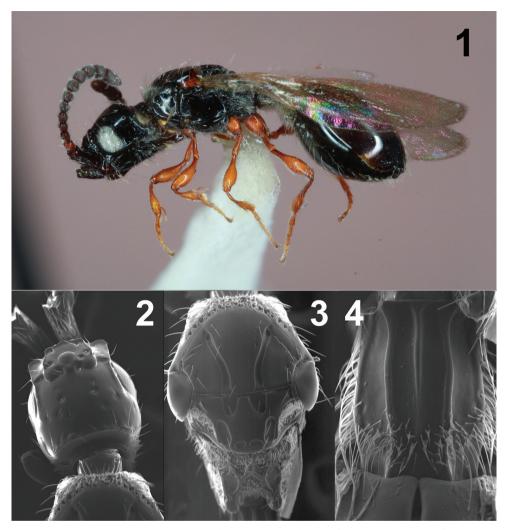
Paratypes: USA • \bigcirc ; Ingham Co., East Lansing, MI; 42.7274, -84.4777, 21 Aug. 2011; Serdar Satar; reared from pupa of *R. juniperina*, SUI:INS:04568 • 6 \bigcirc ; ibid; 8–9 Aug. 2011; SUI:INS:04569-04573, 04576 • \bigcirc ; ibid; 13 Aug. 2011, yellow pan trap; SUI:INS:04577 • \bigcirc ; Johnson Co., Iowa City, IA, 41.6509, -91.5603, 11 Sep. 2011, Andrew Forbes; yellow pan trap; SUI:INS:04574 • \bigcirc ; ibid; 10 Sep. 2011; SUI:INS:04565.

Diagnosis. Coptera tonic females (Figs 1–4) may be distinguished from female *C. pomonellae* (Figs 9–12) most readily by the distance between the apical punctures on the scutellum. In *C. tonic*, this distance is small, less than 1/2 of the shortest diameter of either puncture (Fig. 3), while in *C. pomonellae* the inter-puncture distance is subequal to the shortest diameter of each puncture (Fig. 11). Male *C. tonic* (Fig. 5) have each apical puncture partially or completely divided into two, such that there are indeterminately four apical punctures (Fig. 7), compared to the two standard punctures in male *C. pomonellae* (Fig. 15). Most flagellomeres of male *C. tonic* are 2–2.5 × longer than wide, with the apical segment $2.7-3.3 \times \text{longer than wide}$ (Fig. 6), while the antennal segments of male *C. pomonellae* are shorter, less than $2 \times \text{as long as wide}$ (final segment may approach $2.5 \times \text{as long as wide}$; Fig. 14). *Coptera tonic* of both sexes differ from *C. cingulatae* by the color of their antennae, which are dark brown to black in *C. tonic* and yellow to light brown in *C. cingulatae* (at least the first 3–4 flagellomeres; Figs 17, 18).

Description. Female. Length 3.0 - 3.1 mm; wing length 2.1 - 2.2 mm. Holotype length 3.0 mm; Holotype wing length 2.1 mm.

Color. Body (Fig. 1) black; legs, including coxae, honey yellow; antennal scape black; flagellum testaceous; eyes and 3 ocelli yellow to white; wings slightly infuscated.

Head. Head about as long as broad; dorsum of head normally with several large punctures (Fig. 2); distance from lateral ocelli to posterior margin of occiput longer

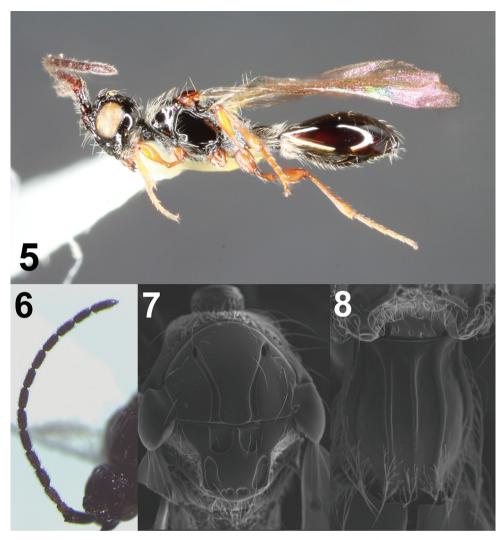


Figures 1–4. Female *Coptera tonic* 1 lateral habitus 2 dorsal view of head 3 dorsal view of mesosoma 4 dorsal view of petiole.

than eyes; temples weakly round, in lateral view nearly as wide as eyes; malar space nearly half as long as eye; antennae strongly clavate and 12-segmented; first flagellomere twice as long as wide; second and third flagellomeres less than twice as long as wide but still longer than wide; all remaining flagellomeres wider than long.

Mesosoma. Pronotum smooth. Notaulices on mesoscutum fine and slightly broadened posteriorly; scutellum weakly convex; paired punctures at apex of scutellum moderately large and separated by less than the shortest diameter of either puncture (Fig. 3); mesopleuron not impressed medially; metapleuron not impressed medially; metapleuron densely hairy.

Metasoma. Petiole of abdomen about 1.5 times as long as wide; petiole with all three dorsal longitudinal carinae strong but median one reduced on some specimens



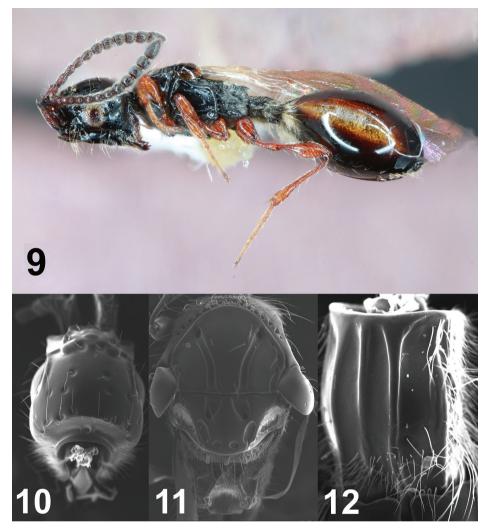
Figures 5–8. Male *Coptera tonic* 5 lateral habitus 6 antenna 7 dorsal view of mesosoma 8 dorsal view of petiole.

(Fig. 4); median sulcus of large tergite not reaching or extending beyond middle of segment; basal lateral sulci not developed.

Male. Length 2.5–3.0 mm; wing length 2.1–2.3 mm.

Color. Body black; legs (including coxae) honey yellow; antennal scape black; flagellum testaceous; eyes and 3 ocelli tan; wings slightly infuscated.

Head. Head wider than long; dorsum of head normally with several large punctures; distances from lateral ocelli to posterior margin of occiput slightly longer than eyes, temples roundly receding, in lateral view slightly narrower than eyes; malar space nearly half as long as eyes; antennae slender with uniform thickness throughout,

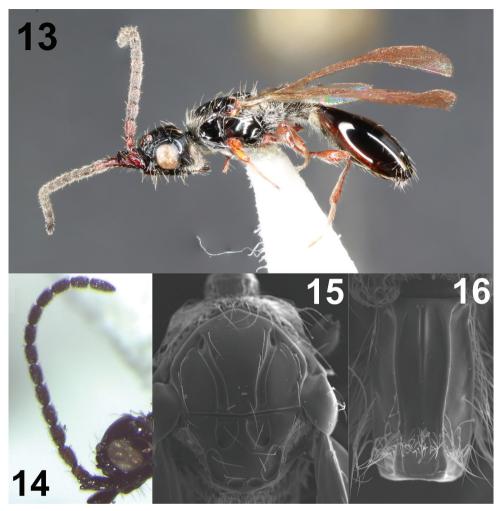


Figures 9–12. Female *Coptera pomonellae* 9 lateral habitus 10 dorsal view of head 11 dorsal view of mesosoma 12 dorsal view of petiole.

14-segmented; all flagellomeres at least twice as long as wide with apical segment about three times as long as wide (Fig. 6).

Mesosoma. Pronotum smooth. Notaulices on mesoscutum fine, slightly broadened posteriorly; scutellum flat; paired punctures at apex of scutellum each subdivided into two smaller punctures (Fig. 7), though sometimes indistinctly; mesopleuron flat, not impressed medially; metapleuron densely hairy.

Metasoma. Petiole about 1.5 times as long as wide; petiole with all three dorsal longitudinal carinae strong and complete; median sulcus of large tergite not reaching the middle of the segment; basal lateral sulci not defined.



Figures 13–16. Male *Coptera pomonellae* 13 lateral habitus 14 antenna 15 dorsal view of mesosoma 16 dorsal view of petiole.

Etymology. The species name is a noun in apposition and refers to tonic water; this parasitic wasp and tonic water are both at their best when in close association with products of *Juniperus* cones.

Ecology. Coptera tonic is a parasitoid of the juniper maggot fly, *Rhagoletis juniperina*, a parasite of the female cones of Eastern red cedar (*Juniperus virginiana*) and other members of genus *Juniperus*. Though oviposition has not been directly observed in *C. tonic*, these wasps have only been reared from pupae floated from soils, and not from larvae extracted from juniper cones, suggesting that attack likely occurs during the fly's pupal stage after it has left the cone. Some pan trap collections of *C. tonic* (e.g., the female paratype labeled "Crab Apple") were made under or near male *Juniperus*, suggesting that these wasps may use plant volatiles as an indicator for host searching.



Figures 17, 18. Coptera cingulatae male and female; lateral habitus.

All known adults were captured or emerged from pupae between late July and early October (Forbes et al. 2012), consistent with the phenology of *R. juniperina* pupation.

Distribution. Existing collections of *C. tonic* are limited to Iowa and Michigan. However, *Rhagoletis juniperina* is distributed across the continental United States and into southern Canada (Bush 1966, Frayer et al. 2015), so a wider distribution for *C. tonic* is possible, if not likely.

Revised partial key to Nearctic Coptera species

Muesebeck (1980) supplied keys to both male and female *Coptera* in the Nearctic, such that changes to both keys are necessary. We propose the following revisions to the Muesebeck (1980) key to *Coptera* females:

- more than ½ of breadth of either puncture (Fig. 11)pomonellae Muesebeck
 Metapleuron densely hairy; paired punctures at apex of scutellum separated by less than ½ of breadth of either puncture (Fig. 3)......tonic, new species

We also propose the following revisions to Muesebeck's (1980) key to Coptera males:

Acknowledgements

Serdar Satar (Çukurova University) and Jim Smith (Michigan State University) collected most of the *C. tonic* material. SEM imaging support was provided by Phil Pagano and Kenny Horkley at the Iowa Center for Research, Exploration, and Advanced Technology in Engineering and Sciences (Iowa CREATES). Comments and questions from Ali Asghar Talebi and an anonymous reviewer substantially improved the manuscript from its original form.

References

- Baeza-Larios G, Sivinski J, Holler T, Aluja M (2002) The ability of *Coptera haywardi* (Ogloblin) (Hymenoptera: Diapriidae) to locate and attack the pupae of the Mediterranean fruit fly, *Ceratitis capitata* (Wiedemann) (Diptera: Tephritidae), under seminatural conditions. Biological Control 23(3): 213–218. https://doi.org/10.1006/bcon.2001.1010
- Brower AV (2010) Alleviating the taxonomic impediment of DNA barcoding and setting a bad precedent: names for ten species of 'Astraptes fulgerator' (Lepidoptera: Hesperiidae: Eudaminae) with DNA-based diagnoses. Systematics and Biodiversity 8(4): 485–491. https://doi. org/10.1080/14772000.2010.534512
- Buckingham GR (1975) The parasites of walnut husk flies (Diptera: Tephritidae: *Rhagoletis*) including comparative studies on the biology of *Biosteres juglandis* Mues. (Hymenoptera: Braconidae) and on the male tergal glands of the Braconidae (Hymenoptera). PhD thesis, University of California, Berkeley.
- Bush GL (1966) The taxonomy, cytology, and evolution of the genus *Rhagoletis* in North America (Diptera, Tephritidae). Bulletin of the Museum of Comparative Zoology, Harvard University 134: 431–562.
- Cameron PJ, Morrison FO (1977) Analysis of mortality in the apple maggot, *Rhagoletis po-monella* (Diptera: Tephritidae), in Quebec. The Canadian Entomologist 109: 769–788. https://doi.org/10.4039/Ent109769-6
- Cancino J, Pérez B, Johnson AC, Reynolds OL (2019) Parasitoids are choosy: increase in the capacity to discriminate parasitised tephritid pupae by *Coptera haywardi*. BioControl 64: 357–366. https://doi.org/10.1007/s10526-019-09941-5
- Condon MA, Scheffer SJ, Lewis ML, Wharton R, Adams DC, Forbes AA (2014) Lethal interactions between parasites and prey increase niche diversity in a tropical community. Science 343(6176): 1240–1244. https://doi.org/10.1126/science.1245007
- Forbes AA, Satar S, Hamerlinck G, Nelson AE, Smith JJ (2012) DNA barcodes and targeted sampling methods identify a new species and cryptic patterns of host specialization among North American *Coptera* (Hymenoptera: Diapriidae). Annals of the Entomological Society of America 105(4): 608–612. https://doi.org/10.1603/AN12012
- Frayer M, Hulbert D, Satar S, Smith JJ (2015) Phenological attributes and phylogenetic relationships of *Rhagoletis juniperina* Marcovitch (Diptera: Tephritidae) in the Great Lakes region. The Great Lakes Entomologist 48(1 & 2): 67–78. https://scholar.valpo.edu/tgle/vol48/iss1/5
- Granchietti A, Sacchetti P, Rosi MC, Belcari A (2012) Fruit fly larval trail acts as a cue in the host location process of the pupal parasitoid *Coptera occidentalis*. Biological Control 61(1): 7–14. https://doi.org/10.1016/j.biocontrol.2011.10.015
- Guillén L, Aluja M, Equihua M, Sivinski J (2002) Performance of two fruit fly (Diptera: Tephritidae) pupal parasitoids (*Coptera haywardi* [Hymenoptera: Diapriidae] and *Pachycrepoideus vindemiae* [Hymenoptera: Pteromalidae]) under different environmental soil conditions. Biological Control 23(3): 219–227. https://doi.org/10.1006/bcon.2001.1011
- Hagen KS, Tassan RL, Fong M (1980) Biological control of the walnut husk fly. University of California Walnut Research Board Report. http://walnutresearch.ucdavis. edu/1980/1980_9.pdf

- Hamerlinck G, Hulbert D, Hood GR, Smith JJ, Forbes AA (2016) Histories of host shifts and cospeciation among free-living parasitoids of *Rhagoletis* flies. Journal of evolutionary biology 29(9): 1766–1779. https://doi.org/10.1111/jeb.12909
- Kazimírová M, Vallo V (1999) Larval morphology and development of *Coptera occidentalis*. BioControl 44(3): 263–280. https://doi.org/10.1023/A:1009907927554
- Lukhtanov VA, Sourakov A, Zakharov EV (2016) DNA barcodes as a tool in biodiversity research: testing pre-existing taxonomic hypotheses in Delphic Apollo butterflies (Lepidoptera, Papilionidae). Systematics and Biodiversity 14(6): 599–613. https://doi.org/10.1080 /14772000.2016.1203371
- Masner L (2002) The genera of Diapriinae (Hymenoptera: Diapriidae) in the new world. Bulletin of the American Museum of Natural History 2002(268): 1–138. https://doi. org/10.1206/0003-0090(2002)268<0001:TGODHD>2.0.CO;2
- Muesebeck CFW (1980) The Nearctic Parasitic Wasps of the Genera *Psilus* Panzer and *Coptera* Say (Hymenoptera, Proctotrupoidea, Diapriidae) U.S. Department of Agriculture Technical Bulletin 1617: 1–71.
- Maier CT (1981) Parasitoids emerging from puparia of *Rhagoletis pomonella* (Diptera: Tephritidae) infesting hawthorn and apple in Connecticut. The Canadian Entomologist 113(9): 867–870. https://doi.org/10.4039/Ent113867-9
- Shashank PR, Chakravarthy AK, Raju BR, Bhanu KRM (2014) DNA barcoding reveals the occurrence of cryptic species in host-associated population of *Conogethes punctiferalis* (Lepidoptera: Crambidae). Applied Entomology and Zoology 49: 283–295. https://doi. org/10.1007/s13355-014-0248-0
- Silvestri F (1914) Report of an expedition to Africa in search of the natural enemies of fruit flies (Trypaneidae) with descriptions, observations and biological notes. Bulletin of the Division of Entomology, Board of Commissioners of Agriculture and Forestry, Hawaii 3: 1–176.
- Sivinski J, Vulinec K, Menezes E, Aluja M (1998) The bionomics of *Coptera haywardi* (Ogloblin) (Hymenoptera: Diapriidae) and other pupal parasitoids of tephritid fruit flies (Diptera). Biological Control 11(3): 193–202. https://doi.org/10.1006/bcon.1997.0597
- Smith MA, Fisher BL, Hebert PDN (2005) DNA barcoding for effective biodiversity assessment of a hyperdiverse arthropod group: the ants of Madagascar. Philosophical Transactions of the Royal Society B: Biological Sciences 360(1462): 1825–1834. https://doi.org/10.1098/rstb.2005.1714
- Smith MA, Rodriguez JJ, Whitfield JB, Deans AR, Janzen DH, Hallwachs W, Hebert PDN (2008) Extreme diversity of tropical parasitoid wasps exposed by iterative integration of natural history, DNA barcoding, morphology, and collections. Proceedings of the National Academy of Sciences 105(34): 12359–12364. https://doi.org/10.1073/pnas.0805319105
- Yoder MJ, Wharton RA (2002) Nomenclature of African Psilini (Hymenoptera: Diapriidae) and status of *Coptera robustior*, a parasitoid of Mediterranean fruit fly (Diptera: Tephritidae). The Canadian Entomologist 134(5): 561–576. https://doi.org/10.4039/Ent134561-5
- Ward AKG, Sheikh SI, Forbes AA (2020) Diversity, host ranges, and potential drivers of speciation among the inquiline enemies of oak gall wasps. BioRxiv. https://doi. org/10.1101/2020.05.08.084699