

First report of *Cotesia vanessae* (Hymenoptera: Braconidae) in North America

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Abstract—We report for the first time the occurrence of the well-known Eurasian and north African parasitoid *Cotesia vanessae* (Reinhard) (Hymenoptera: Braconidae) in North America. Specimens were reared from *Chrysodeixis chalcites* (Esper) and *Trichoplusia ni* (Hübner) (Lepidoptera: Noctuidae) recovered from several locations in southwestern Ontario, Canada, and detected by DNA sequencing from one *Autographa californica* (Speyer) (Lepidoptera: Noctuidae) in southern Alberta, Canada.

Résumé—Nous rapportons pour la première fois la présence du parasitoïde *Cotesia vanessae* (Reinhard) (Hymenoptera: Braconidae) en Amérique du Nord. Des spécimens ont été élevés à partir de *Chrysodeixis chalcites* (Esper) et *Trichoplusia ni* (Hübner) (Lepidoptera: Noctuidae) collectés à plusieurs localités au sud-est de l'Ontario, Canada, et détecté par une séquence d'ADN provenant d'un *Autographa californica* (Speyer) (Lepidoptera: Noctuidae) collecté à une localité au sud de l'Alberta, Canada. Ce parasitoïde n'a été reporté auparavant qu'en Europe, Asie et Afrique.

Cotesia Cameron (Hymenoptera: Braconidae) is a genus of braconid wasps that are parasitoids of Lepidoptera larvae. Several species are of interest as biological control agents of pest caterpillars. For example, *Cotesia urabae* Austin and Allen was released into New Zealand in 2011 for the biological control of the eucalyptus pest *Uraba lugens* Walker (Lepidoptera: Nolidae) (Avila and Berndt 2011). *Cotesia flavipes* Cameron has been studied for many years and has been introduced in many countries as a biological control agent for the sugarcane borer *Diatraea saccharalis* (Fabricius) (Lepidoptera: Crambidae) on cereal crops (Poaceae) (Overholt *et al.* 1997; Jiang *et al.* 2004). *Cotesia rubecula* (Marshall) has been released in the United States of America as a natural enemy of *Pieris rapae* (Linnaeus) (Lepidoptera: Pieridae) on cabbages,

Brassica Linnaeus species (Brassicaceae) (Driesche and Nunn 2002).

In the most recent checklist of *Cotesia* species in Canada and Alaska, Fernández-Triana (2010) reported 55 species (51 described) with the expectation that many more species in the genus remained unreported. We add to this list the first report of *Cotesia vanessae* (Reinhard) in Canada and, more broadly, North America (Whitfield 1995).

Cotesia vanessae is a gregarious endoparasitoid that primarily attacks species of Vanessini (Nymphalidae) and certain Noctuidae, with records from Notodontidae, Lasiocampidae, Crambidae, and Pterophoridae (Yu *et al.* 2012) here considered doubtful. Host records from the Old World that we consider reliable include the vanessines *Vanessa cardui* (Linnaeus), *Vanessa*

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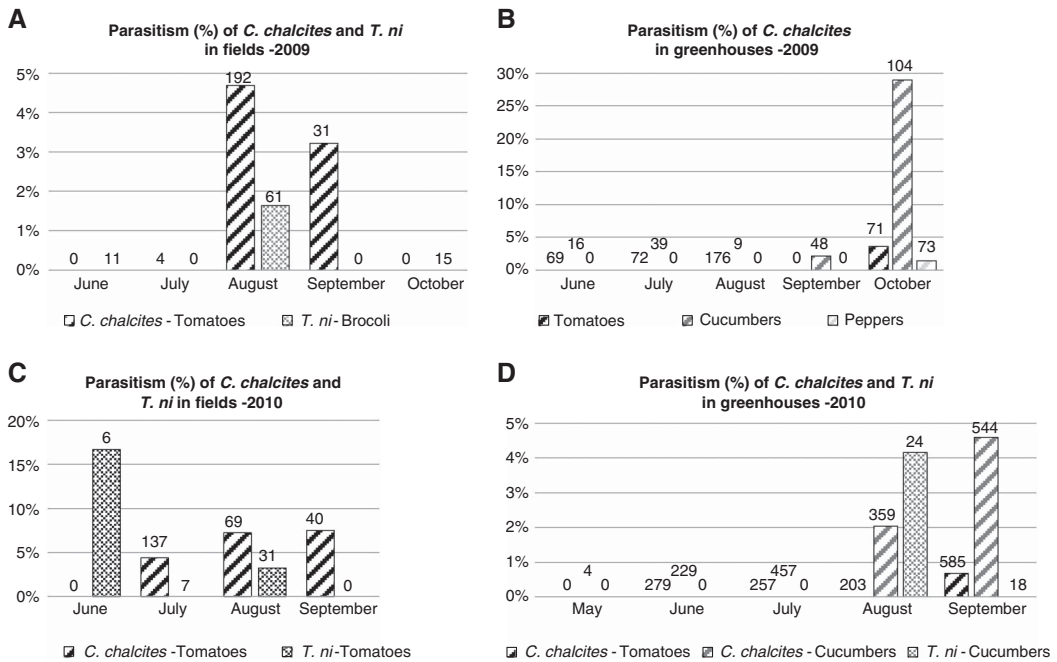
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Fig. 1. Parasitism (%) by *Cotesia vanessae* on *Chrysodeixis chalcites* and *Trichoplusia ni* collected from fields and commercial greenhouses in southwestern Ontario, Canada in 2009 and 2010. The numbers represent the total number of caterpillars collected each month on the indicated plants.



atalanta (Linnaeus), and *Aglais urticae* (Linnaeus), and the noctuids *Actebia praecox* (Linnaeus) and *Mythimna litoralis* (Curtis) (specimens in the British Museum of Natural History (London, United Kingdom) and the National Museum of Scotland (Edinburgh, United Kingdom)). Nixon (1974) recorded *Spodoptera exigua* (Hübner) as a host, but we have been unable to rear *C. vanessae* on this host in laboratory experiments (V.A.D.H., personal observation). In Britain, *C. vanessae* necessarily passes the winter within overwintering noctuid host larvae, because its vanessine hosts are available as larvae only in the summer and the parasitoid adults always emerge from the cocoons in the year of their formation.

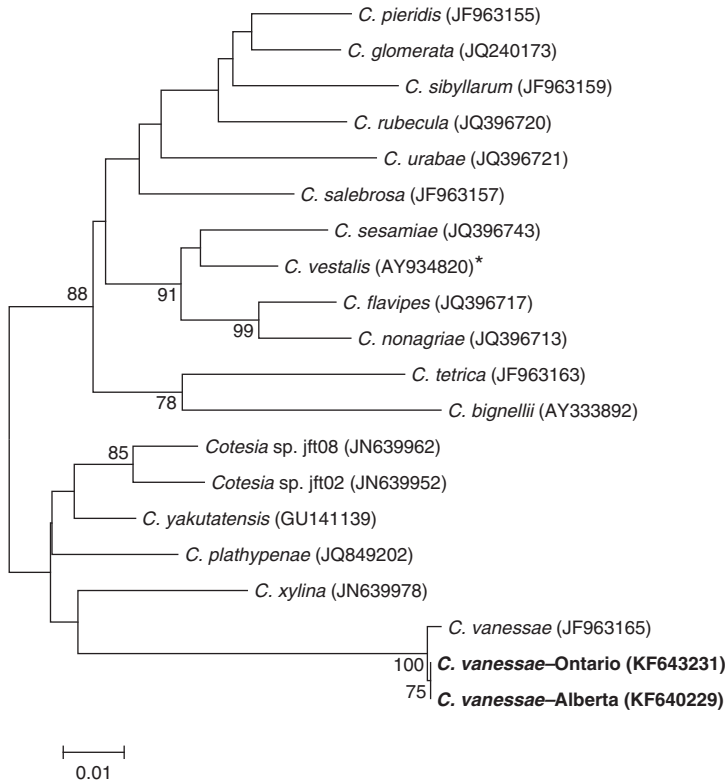
European populations of *C. vanessae* may be bisexual or, in the Mediterranean region, comprise only thelytokous females (Stefanescu *et al.* 2012).

Using genetic analyses as well as morphological and biological observations, we document populations of *C. vanessae* in Ontario and Alberta, Canada. This parasitoid species has previously been reported throughout Europe and scattered locations in Asia and North Africa (Stefanescu *et al.* 2012; Yu *et al.* 2012).

Collections from Ontario: *Cotesia vanessae* was first reared from the tomato looper, *Chrysodeixis chalcites* (Esper) and cabbage looper, *Trichoplusia ni* (Hübner), collected in 2009 in southwestern Ontario. *Chrysodeixis chalcites* has already been reported as a host of *C. vanessae* (Messelink 2002), but *T. ni* is a new host record for *C. vanessae*.

For *C. chalcites*, parasitised loopers were collected from tomato plants, *Solanum lycopersicum* Linnaeus (Solanaceae), in fields around the cities of Harrow and Leamington in the summers of 2009 and 2010 (42°05'07.25"N, 82°36'08.65"W; 42°02'11.54"N, 82°57'03.78"W; 42°02'36.45"N, 82°38'05.60"W; 42°05'00.78"N, 82°37'07.26"W; 42°02'00.82"N, 82°53'58.49"W). Parasitised *C. chalcites* were also collected from tomato, pepper, *Capsicum annum* Linnaeus (Solanaceae), and cucumbers, *Cucumis sativus* Linnaeus (Cucurbitaceae), in greenhouses around Leamington and Chatham-Kent in the fall of 2009, and in the late summer and fall of 2010. For *T. ni*, a parasitised larva was collected from a broccoli plant in a field west of Leamington in August 2009 (42°02'36.45"N, 82°38'05.60"W), two from tomato

Fig. 2. Neighbour-joining tree, K2P distance model for CO1 sequences for *Cotesia* species generated using Mega 5.2. Sequences obtained from GenBank. Bootstrap values <70 are not reported. *Although the name *Cotesia plutellae* (Kurdjumov) is used for this sequence in GenBank, the valid name for this species is now *Cotesia vestalis* (Haliday) (see Shaw 2003).



plants in fields around Harrow and Chatham-Kent in the summer of 2010 (42°02'11.54"N, 82°57'03.78"W; 42°23'26.93"N, 82°09'27.13"W), and one from a cucumber plant in a commercial greenhouse northeast of Leamington in August 2010 (42°04'59.21"N, 82°34'10.93"W) (Fig. 1).

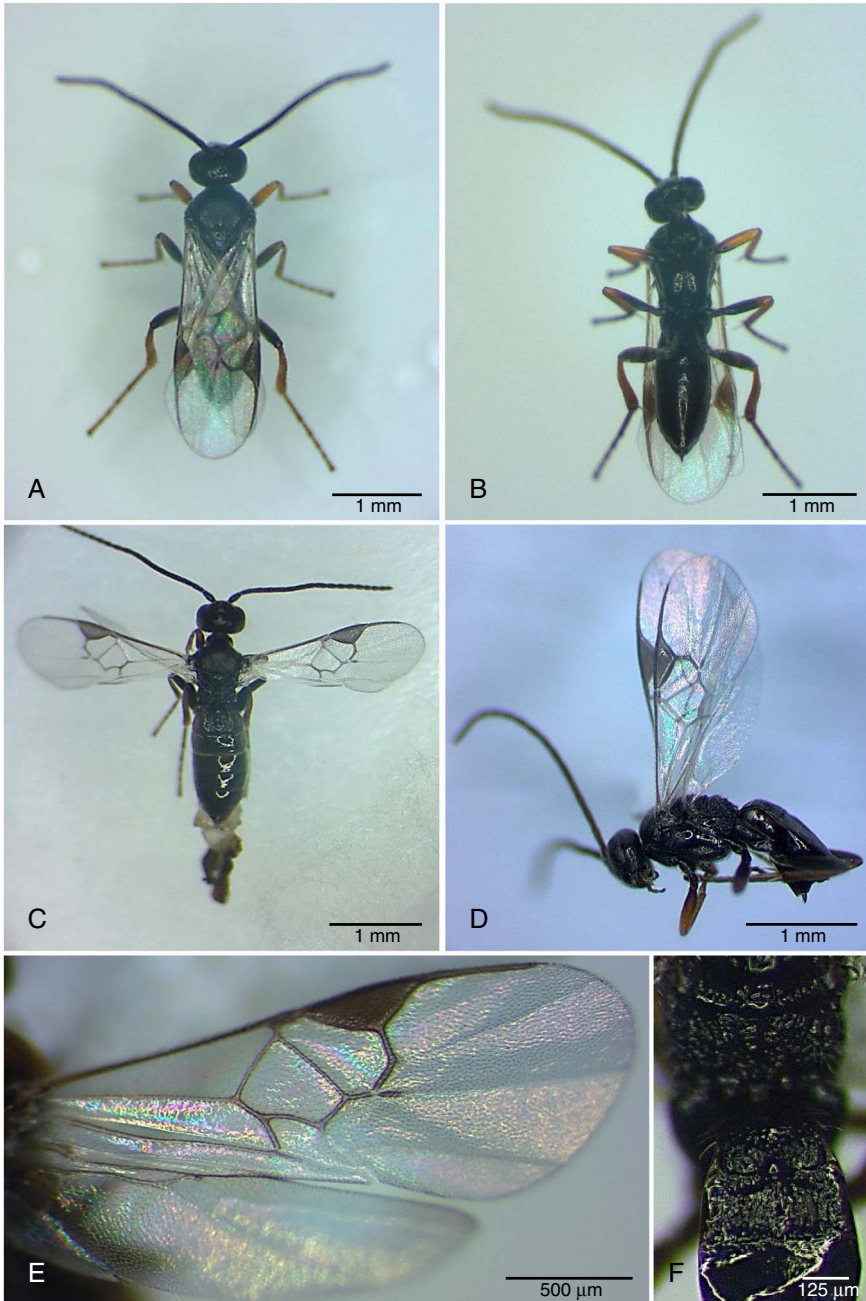
Parasitoids from these original field collections initially were reared at the Sunrite Greenhouses Biocontrol laboratory, Ontario (by H.M.), and subsequently at the Lethbridge Research Centre, Alberta (by V.A.D.H.). Colonies were maintained on *T. ni* purchased from the Insect Production Services of the Great Lakes Forestry Centre, Sault Ste. Marie, Ontario, which were reared on McMorran diet (McMorran 1965) (recipe modified by the Great Lakes Forestry Centre). Only females have been observed in our laboratory colony, reproducing by thelytokous parthenogenesis over multiple generations. From a subsample of 10 *T. ni*, each exposed to a single

C. vanessae for 48 hours, an average of 103 adult parasitoids emerged (range: 56–165).

To identify the parasitoid species, we obtained CO1 sequences for specimens from the laboratory colony using primers and general methods as described in Hebert *et al.* (2003). All the sequences were identical, one of which (KF640231) was deposited in GenBank (<http://www.ncbi.nlm.nih.gov/genbank/>). This sequence was compared with existing sequences from GenBank, using similarity analyses that identified our specimens as *C. vanessae* (Fig. 2).

To characterise the morphology of this species, photographs of adult and immature *C. vanessae* are provided in Figures 3, 4. There are many similar *Cotesia* species, thus these pictures are not meant to be used to identify an unknown specimen. Reinhard's descriptions of *C. vanessae* (Reinhard 1880, 1881) and Nixon's (1974) diagnosis correspond with our observations. However, the Ontario specimens tend to have lighter metafemur, metatibia, and

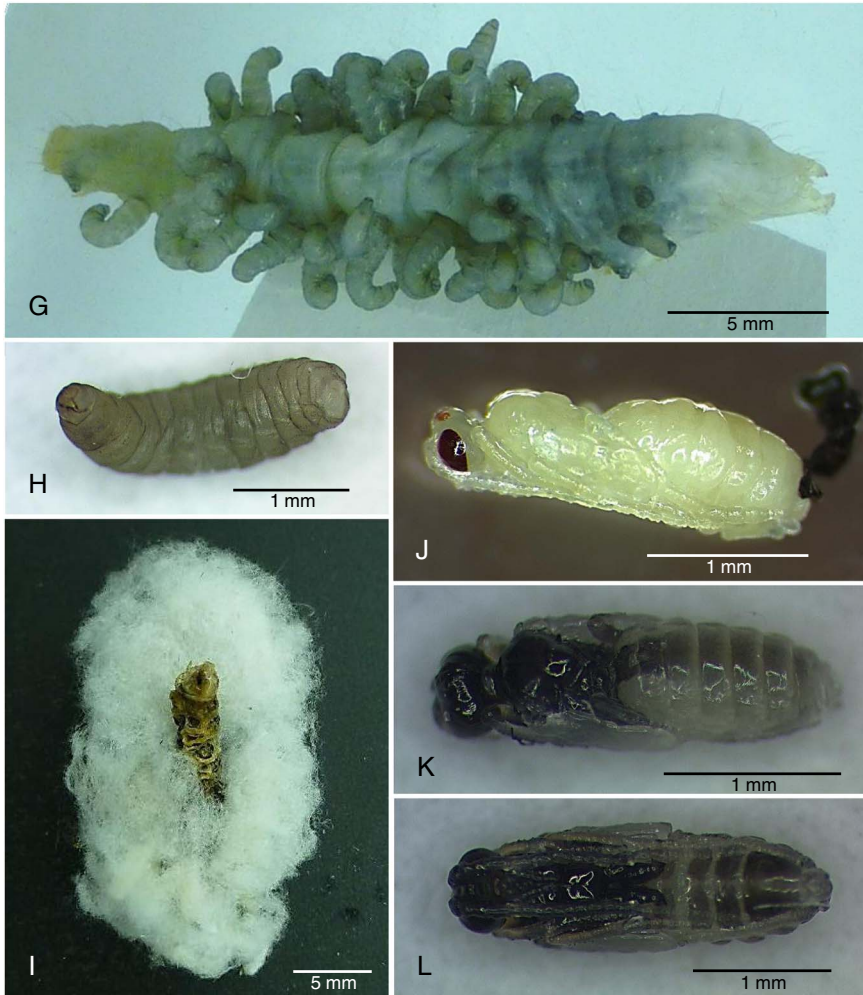
Fig. 3. *Cotesia vanessae* adults (all females): (A) live, dorsal side; (B) live, ventral side; (C) live, emerging from pupal exuvia, dorsal side; (D) dead, pointed, left lateral side; (E) dead, upper surface of right wings; (F) dead, dorsal side of propodeum and basal three tergites of metasoma.



metasoma (especially the laterotergites) compared with the specimens that we examined from north-western Europe. Specimens from Libya and Ethiopia (perhaps really Eritrea) also have been reported to be lighter in colour than European specimens

(Nixon 1974; Papp 1987). Nixon (1974) also drew attention to minor differences, that he considered to be of little significance, between European series reared from vanessines and noctuids – presumably merely season- and/or host-related.

Fig. 4. *Cotesia vanessae* immature stages: (G) larvae (prepupae) erupting from a *Trichoplusia ni* larva before pupating; (H) prepupa just emerged from a Noctuidae host; (I) cocoon mass of about 80 cocoons around dead host; (J) one-day-old pupa, left lateral side; (K) three-day-old pupa, dorsolateral side; (L) five-day-old pupa, ventral side.



Collections from Alberta: A DNA sequence for *C. vanessae*, but not specimens, was recovered from a final instar larva of alfalfa looper, *Autographa californica* (Speyer) (Lepidoptera: Noctuidae), which had also produced a brood of a different species of *Cotesia*. The larva was one of about 30 *A. californica* collected for an unrelated study on 9 and 10 July 2012 from three canola fields; that is, 12 km NE of Claresholm (50°04'45"N, 113°26'21"W), 17 km NE of Claresholm (50°07'23"N, 113°26'21"W), 30 km E of Calgary (50°57'27"N, 113°31'42"W). Larvae were held indoors in a Plexiglas cage (30 × 30 × 30 cm) at

22 °C and fed canola leaves, *Brassica napus* Linnaeus (Brassicaceae) for three days after which an unidentified gregarious species of *Cotesia* emerged from most individuals. Based on morphology and CO1 sequences (KF640233), the parasitoid was not *C. vanessae*. However, in sequencing parasitised caterpillars to verify their identity, we obtained a CO1 sequence (KF640229) that was identical to that for *C. vanessae* from Ontario (Fig. 2).

We speculate that this *A. californica* was parasitised by both the unknown *Cotesia* species and by *C. vanessae*, but only the former emerged from the host. Cases of caterpillars being parasitised by

Fig. 5. Reported distribution of *Cotesia vanessae*. The North American reports are from this study. The Old World distribution includes unpublished data (specimens in the Natural Museum of Scotland). Figure generated using SimpleMapppr.



two *Cotesia* species and progeny of both species successfully developing within the host have previously been documented (Ngi-Song *et al.* 2001).

Current known range: *Cotesia vanessae* is widely distributed throughout the Palearctic (Fig. 5). Nixon (1974) recorded it from Eritrea (Asmara), Libya (Benghazi), Turkey, Italy, and “northwest Europe”. Stefanescu *et al.* (2012) reported it from locations in central-western Morocco and north-east Spain. Specimens in the Natural Museum of Scotland also include records from: Israel (Negev Mountains), Bulgaria (East Rhodope Mountains), Greece (N. Peloponnese), Crete, Canary Islands (Lanzarote, Fuerteventura), Spain (Girona, Lleida, Barcelona, Córdoba, Aragon), France (Charente), Finland (Åland), and most of the counties in the southern half of England. Additional records (Fig. 5) are from Yu *et al.* (2012).

We provide morphological, biological and genetic evidence that documents the first record of *C. vanessae* in North America. Where, when, and how this parasitoid may have been introduced to North America is unknown. We speculate that it may have been introduced into North America developing inside a host in a shipment of imported plants.

Recovery of this parasitoid from three pest species thus far (*C. chalcites*, *T. ni*, and *A. californica*) warrants further study of *C. vanessae* as a potential biological control agent against noctuid crop pest caterpillars. However, this parasitoid is also

reported to parasitise nymphalid butterflies such that non-pest species are likely to be at risk. Benson *et al.*, (2003) showed that the deliberate introduction of *C. rubecula* (Marshall) for the biological control of pest *Pieris* butterflies had a negative impact on a native nonpest congener. Hence, monitoring the establishment and spread of *C. vanessae* in North America and its possible colonisation of native vanessines and noctuids is advised, before any deliberate manipulations in the interests of crop protection are attempted.

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