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Cover Page Footnote

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Karshomyia caulicola (Diptera: Cecidomyiidae) Associated with *Sclerotinia*-Infected Soybean in the United States and Canada

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Abstract

The white-mold gall midge, *Karshomyia caulicola* (Coquillett), (Diptera: Cecidomyiidae) was documented in association with soybean, *Glycine max* (L.) Merr., infected with the fungus *Sclerotinia sclerotiorum* (Lib.) de Bary. This mycetophagous cecidomyiid appears widespread in the northern soybean producing region, with confirmed detections from Minnesota, North Dakota (U.S.) and Québec (Canada). Though likely not a pest of soybean plants, the presence of *K. caulicola* in soybean fields may complicate identification, population assessment and decision making for soybean gall midge, *Resseliella maxima* Gagné, which is a recently described pest of soybean. Here, we provide an overview of the known biology and distribution of *K. caulicola* and descriptions to aid in distinguishing these two cecidomyiids.

Keywords: White mold, *Sclerotinia* stem rot, Cecidomyiidae, gall midge

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This article documents the association of the white-mold gall midge, *Karshomyia caulicola* (Coquillett) (Diptera: Cecidomyiidae), with soybean, *Glycine max* (L.) Merr. (Fabales: Fabaceae), and other plants infected with the fungus *Sclerotinia sclerotiorum* (Lib.) de Bary (Helotiales: Sclerotiniaceae). This fungus causes the plant disease *Sclerotinia* stem rot (white mold), with infected plants exhibiting growth of white, cottony mycelia and sclerotia on and in above-ground tissues (Link and Johnson 2007). Of particular concern is that immature stages of *K. caulicola* on *Sclerotinia*-infected soybean plants or in soil of soybean fields could be confused for immatures of the recently described soybean gall midge, *Resseliella maxima* Gagné (Diptera: Cecidomyiidae) (Gagné et al. 2019). *Resseliella maxima* is an emerging pest of soybean in the Midwest Region of the U.S. (Gagné et al. 2019). *Resseliella maxima* infests the stems of soybean plants generally near the soil surface, where the larvae feed mainly on the phloem, but may eventually move to the xylem and pith (Gagné et al. 2019, Koch and Potter 2019).

During surveys to determine the geographic distribution of *R. maxima* and other scouting in soybean, larvae with a superficially similar appearance to *R. maxima* were found on or in *Sclerotinia*-infected tissues of soybean plants. Cecidomyiid adults were reared under ambient laboratory conditions from larvae in *Sclerotinia*-infected soybean stems collected in Val-Saint-François Municipality, Québec (Canada) (29 August 2018) and Ramsey County, Minnesota (U.S.) (2 August 2019). In addition, two cecidomyiid adults were collected in an emergence trap placed over the soil in a field in Stearns County, Minnesota (U.S.) (8 July 2019). In 2018, this field had a significant infestation of cecidomyiid larvae associated with *Sclerotinia*-infected soybean stems. Additional cecidomyiid larvae were collected from *Sclerotinia*-infected soybean stems Redwood (17 September 2019), Renville (16 September 2019) and Stearns (11 September 2019) counties in Minnesota, and in Barnes (17 September 2019), Cass (17 September 2019), LaMoure (17 September 2019), Richland (17, 20 and 29 September 2019) and Sargent (17 September 2019) counties in North Dakota

(U.S.). The above-mentioned cecidomyiid adults and larvae from Québec, Minnesota and North Dakota were determined to be *K. caulicola* by R. J. Gagné. Voucher specimens are deposited in the National Museum of Natural History (USNM), Smithsonian Institution, Washington, DC (U.S.).

Larvae matching the description of *K. caulicola* were reported (but not brought to the laboratory for identification) from *Sclerotinia*-infected tissues of soybean plants in 2018 in McLeod, Sibley, Stearns, Olmsted, Ottertail and Wadena counties in Minnesota (Potter unpublished data). In 2019, more larvae were reported from *Sclerotinia*-infected tissues of soybean plants in Benton, Brown, Chippewa, Dakota, Douglas, Goodhue, Kandiyohi, Meeker, Morrison, Nicollet, Ottertail, Pope, Redwood, Ramsey, Sherburne, Stearns, Stevens, Wadena, Waseca and Wright counties in Minnesota (Potter and Koch unpublished data), and from *Sclerotinia*-infected pepper, *Capsicum annuum* L. (Solenales: Solenaceae), in Val-Saint-François Municipality, Québec (Moisan-De Serres personal observation). Previously, observations were made of similar larvae in association with *Sclerotinia*-infected tissues of soybean, dry beans, *Phaseolus vulgaris* L. (Fabales: Fabaceae), and canola, *Brassica napus* L. (Brassicales: Brassicaceae), in Manitoba (Canada) in 2014 and 2016 (Gavloski and Bajracharya 2016, Gavloski unpublished data), and in association with *Sclerotinia*-infected soybean and sunflower, *Helianthus annuus* L. (Asterales: Asteraceae), in Minnesota dating back at least 25 years (Potter personal observation).

These larvae associated with *Sclerotinia*-infected tissues displayed a distinctly different biology than those of typical *R. maxima* infestations (Gagné et al. 2019, Koch and Potter 2019). Infestations of *K. caulicola* were seen later in the season (after soybean flowering and the onset of *Sclerotinia* stem rot), whereas *R. maxima* infestations are typically found as early as the third trifoliate stage growth stage of soybean. *Karshomyia caulicola* larvae were found inside and outside of *Sclerotinia*-infected soybean stems, pods and other tissues (Fig. 1), whereas *R. maxima* larvae are typically found under the epidermis of the stem at the base of soybean plants. *Karshomyia caulicola* infestations were encountered throughout soybean fields wherever *Sclerotinia*-infected stems are present, whereas *R. maxima* infestations have typically been most abundant near field edges that are adjacent to fields that had soybean the previous year.

In the field, late instar larvae of *K. caulicola* displayed a generally less intense orange coloration (Fig. 1) than similarly sized larvae of *R. maxima* (Gagné et al. 2019). Under close examination, larvae of these two species can be distinguished. The larval integument of *K. caulicola* is generally smooth with minor sculpturing (Fig. 2a). The terminal abdominal segment of *K. caulicola* ends in two blunt lobes, one on each side, and each with four inconspicuous apical setae (Fig. 2a, b). The spatula of *K. caulicola* is deeply incised anteriorly and

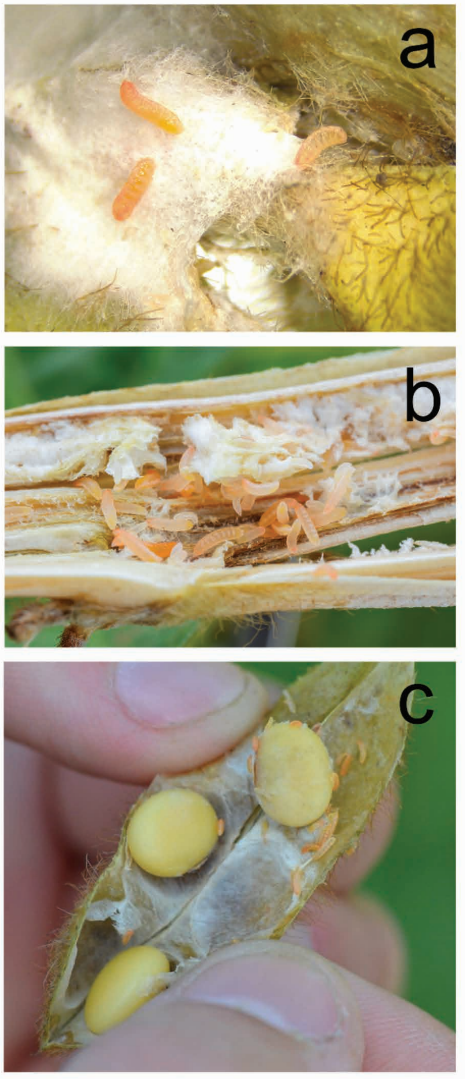

Figure 1: Larvae of *Karshomyia caulicola* associated with *Sclerotinia*-infected soybean: larvae on mycelia on exterior of soybean pods [a], larvae inside a *Sclerotinia*-infected stem [b], and larvae inside a *Sclerotinia*-infected pod [c]. Image by T. Cira & R. Aita [a], J. Moisan-De Serres [b] and B.D. Potter [c].

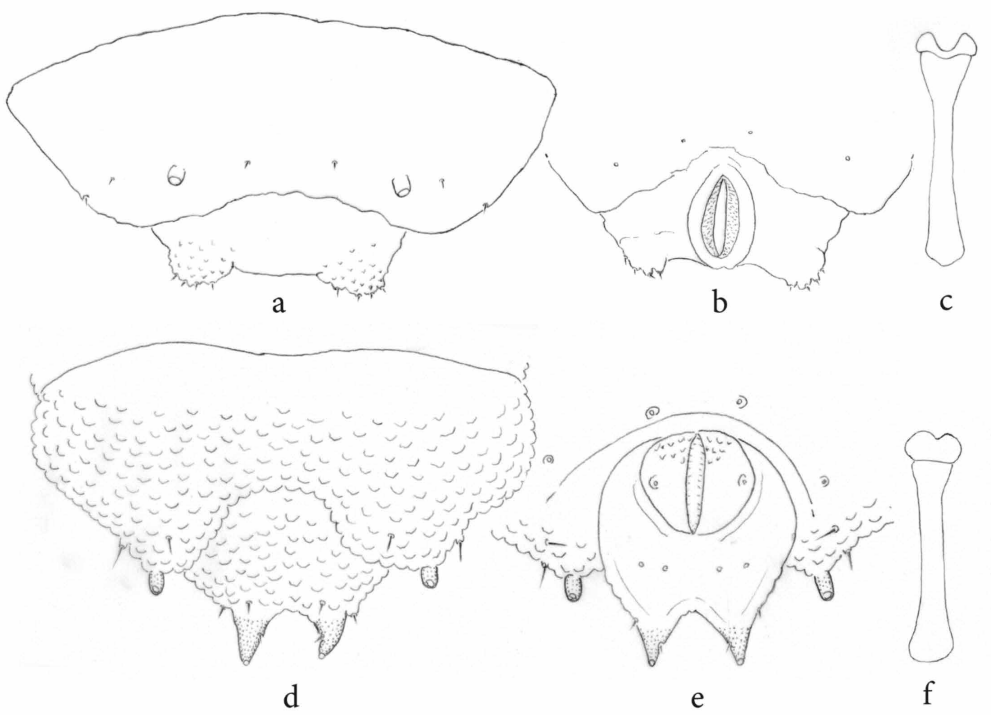

Figure 2: Distinguishing characters of *Karshomyia caulicola* larva (dorsal [a] and ventral [b] views of larval eighth and terminal abdominal segments and spatula [c]) and *Resseliella maxima* larva (dorsal [d] and ventral [e] views of larval eighth and terminal abdominal segments and spatula [f]). Drawings by R.J. Gagné.

the lobes rounded (Fig. 2c). In contrast, the larva of *R. maxima* has a uniformly pebbled integument, and its terminal segment is more tapered and ends in a conspicuous pair of conical projections (Fig. 2d, e). The spatula of *R. maxima* is only weakly incised apically (Fig. 2f).

Adults of the two species are strikingly different. Adults of *K. caulicola* have uniformly grayish antennae, wings and legs, and the abdominal tergites and sternites are very definitely subdivided horizontally (Fig. 3). In contrast, adults of *R. maxima* have striped antennae and legs, mottled wings, and the abdominal tergites and sternites are uniformly sclerotized (Gagné et al. 2019). The male sexual apparatus of both are distinct also; that of *K. caulicola* was illustrated in Gagné (1973) and that of *R. maxima* in Gagné et al. (2019). A key to genera of adult Cecidomyiinae of North America is available (Gagné 2018) to identify other possible gall midges that might be encountered in work on soybean.

The genus *Karshomyia*, which is comprised of 51 species worldwide, is in the tribe Karshomyiini in the subfamily Cecidomyiinae (Gagné and Jaschhof 2017). *Karshomyia caulicola* has also been referred to as: *Diplosis caulicola* Coquillett, *Hiastatus concinnus* Marikovskij, and *Karschomyia concinna* (Marikovskij) (Pakalniškis et al. 2000, Gagné and Jaschhof 2017). *Karshomyia caulicola* has a Holarctic distribution. It is widespread in Europe, with reports from the United Kingdom (Harris 2004), Netherlands (Nijveldt 1985), Germany (Meyer 1984), Russia (Marikovskij 1956), Latvia (Spungis 2003), Lithuania (Pakalniškis et al. 2000) and Ukraine (Gagné and Jaschhof 2017). In Germany, *K. caulicola* is reported as univoltine with adult flight activity occurring over a period of five months (Meyer 1984). In North America, *K. caulicola* was previously reported from only the U.S. state of New Hampshire (Coquillett 1895). The detections reported above, expand considerably the known North American range of *K. caulicola*.

Karshomyia caulicola is reported as being mycetophagous (Meyer 1984), which explains the observed association with *Sclerotinia*-stem rot of plants. In Europe, larvae of this species have been reported from *Sclerotinia*-infected "stems of beans and potatoes and in the stems and pods of

Figure 3: Adult of *Karshomyia caulicola*. Image by J. Moisan-De Serres.

winter rape" and to feed on the mycelia of such fungi (Nijveldt 1985). Similarly, the detections reported here of *K. caulicola* in North America were also associated with *Sclerotinia*-infected stems and reproductive tissues of plants. Therefore, we suspect this species is not a plant pest. However, the report of *K. caulicola* larvae from stems of Icelandic poppy, *Papaver nudicaule* L. (Ranunculales: Papaveraceae), in New Hampshire (Coquillett 1895) and "oil poppy" in the United Kingdom (Harris 2004) had no mention of *Sclerotinia*-stem rot or other fungal infection of the plants.

Here, we provided the first documentation of *K. caulicola* associated with *Sclerotinia*-infected soybean and other plants in North America, which corroborates reports of its biology from Europe. The geographic extent of the reports presented here and a report from Pierce County, Wisconsin (U.S.) (Hamilton 2019) confirm that *K. caulicola* is widespread. Furthermore, knowledge of the presence of this cecidomyiid in soybean refines our understanding of the geographic distribution of *R. maxima*, which was previously confused with *K. caulicola* in some earlier distribution maps. The detection of adult *K. caulicola* in emergence cages placed in a field in 2019 that had numerous orange-colored cecidomyiid larvae in association with *Sclerotinia*-infected soybean in 2018, suggests *K. caulicola* likely overwinters in the soil or plant debris in the field. Additionally, we clarify the morphological differences between *K. caulicola* and *R. maxima* to facilitate identification of these species for efforts to understand the biology of both these species in soybean fields. The presence of both cecidomyiid species in soybean may complicate identification, population assessment and decision making for the plant pest, *R. maxima*.

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