Diagnostics
Diagnostic

Rhagoletis completa

Specific scope

This standard describes a diagnostic protocol for *Rhagoletis completa*[1].

Specific approval and amendment

Approved in 2011-09.

Introduction

The genus *Rhagoletis* contains about 60 species throughout the world. Some of them are of economic importance. *Rhagoletis completa*, native of North America, is a pest of walnut (*Juglans* spp.). In the 1990s, the occurrence of this pest was reported for first time in Europe in Switzerland and Italy (Duso & Dal Lago, 2006). Since then this pest has spread to other countries of the EPPO region (EPPO Plant Quarantine data Retrieval system, EPPO, 2011). *Juglans regia* is the only host plant recorded in Europe. *Prunus persica* was reported as a host in the United States (Bush, 1966) although this has never been confirmed, and Yee & Goughnour (2008) reported English hawthorn *Crataegus laevigata* as a new host in the United States. Infested nuts are deformed and appear dirty due to the rotting husk (Fig. 1). *Rhagoletis completa* has only one generation per year (univoltine) and can be trapped from July until September in Central Europe. Additional information on the biology of the pest can be found in the datasheet on *R. completa* (EPPO/CABI, 1997).

Identity

Name: *Rhagoletis completa* Cresson, 1929
Synomyms: *Rhagoletis suavis* spp. *completa* Cresson, *Rhagoletis juglandis*: Boyce 1929 (misidentification)
Taxonomic position: Diptera Brachycera Tephritidae
EPPO code: RHAGCO
Phytosanitary categorization: EU Annex I/A1.

[1]Use of brand names of chemicals or equipment in these EPPO Standards implies no approval of them to the exclusion of others that may also be suitable.

Detection

Fruit flies may be detected as larvae in fruits or as adults caught in traps.

Detection on fruits

Usually, a single puncture is found per nut and the average number of eggs per puncture is 21.8. The three larval instars develop in the husk of nuts. Mature larvae leave the husks to pupate in the soil (Duso & Dal Lago, 2006).

Rearing larvae for morphological study of adults allows confirmation of identification. However, adult emergence only occurs after 10 or 11 months in the field. Infested fruits should be placed in a container that has a gauze or muslin top and dry medium at its base, such as sterilized sawdust or sand, in which emerging larvae can pupariate. Samples should be checked every 2 days for puparia and fruit from which larvae have emerged should be discarded. When all the larvae have emerged from the fruit or if any sign of mould appears the sawdust should be sieved and the puparia collected. As is the case for other *Rhagoletis* species, *R. completa* has a true winter diapause which is only broken after a certain period of cold temperatures. Although the diapause can last 10–11 months in the field, it has been shortened to 8 months in the laboratory (K. Koeppler, unpublished data). Exact data about the minimum time period are not available. After diapause, puparia can then be transferred to petri dishes and covered with a thin layer of moist heat-sterilized sawdust and then placed in a small emergence cage. It is important to provide sugar solution as food for the emerging adults and to keep the adults alive for at least 4 days after emergence, so that the flies develop their full body colouration and normal shape. Failure to feed the flies will result in specimens that have shrivelled abdomens and dull colours (White & Elson-Harris, 1992).

Fig. 1 Nuts with rotting husk.

Fig. 2 *Rhagoletis completa* larvae and larvae in a rotting husk.

Detection of adults

Different traps can be used: yellow sticky traps, yellow sticky traps with pherocon AM, the same baited with 3 g of ammonium carbonate and woody glued green spheres (Duso & Dal Lago, 2006). Additional information on trapping is available in EPPO/-CABI (1997).

Identification

Morphological identification to species level with a binocular microscope is the recommended diagnostic method. Magnification ×10 for adult to ×200 for larvae and aculeus.

A reliable identification can only be performed on an adult specimen. It is not possible to identify with certainty eggs, larvae and pupae with this protocol.

For morphological terminology refer to White & Elson-Harris (1992).

Description for egg, larval and pupal stages

Egg: No published description.

Larvae (3rd instar): (after White & Elson-Harris, 1992 and Carroll *et al.*, 2004) (Fig. 2).

For identification of the family Tephritidae see Stehr, 1991 and for identification of the genera and species *Rhagoletis completa* larvae see White & Elson-Harris, 1992.

Length 8.0–10.0 mm, width 2.0 mm. Entirely whitish to yellowish.

- *Head*: Antenna 2-segmented. Each stomal sensory organ with a single preoral tooth at base. Oral ridges in 7 shorts rows. Mouthhooks heavily sclerotized, each with a stout bluntly rounded apical tooth (Fig. 3).
- *Thoracic and abdominal segments*: Anterior spiracles each with an irregular row of 21 [Anterior spiracular tubules 15–21, Carroll *et al.* (2004); approximately 15–20, Boyce 1934)] short tubules (Fig. 3). Minute body spinules confined to fusiform ventral areas, except for a narrow band on the anterior margin of T1 (or T1–T2, Carroll *et al.*, 2004). A8 with intermediate areas well developed, with 1 pair each of intermediate and ventral tubules obvious. Posterior spiracle slits about 3 times as long as broad; upper and lower slits at about 60° to each other; spiracular hairs short (about 1/3 the length of a spiracular slit), mainly branched; dorsal and ventral hair bundles of 8–11 hairs;

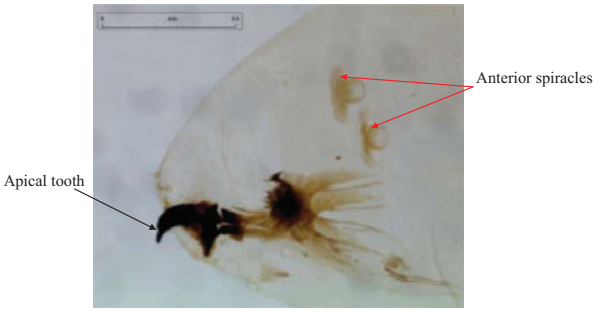

Fig. 3 *Rhagoletis completa* larvae – anterior part.

Fig. 4 *Rhagoletis completa* – female (length: 4.0–6.5 mm).

lateral hair bundles of 4–10 hairs; area between posterior spiracles smooth. Anal lobes large, protuberant, surrounded by a few discontinuous rows of small spinules.

When larvae are to be preserved, they should be placed in boiling water for a few seconds and then transferred to 70% ethanol. Other procedures can also be used.

- *Pupae*: No published description.

Description for adult (after Bush, 1966 and Foote, 1981).

Female (Fig. 4) and male (Fig. 5) about the same size; highly variable about 4.0–6.5 mm length.

Head: Entirely yellow. Genal, occipital, post-vertical, post-orbital and strong gular bristles yellow to yellowish orange; all other major bristles black. Gena about 0.30 times as high as eye.

Thorax: (Fig. 6)

- *Male*: Dorsum concolourous variable brown, uniformly covered with light yellow to golden yellow decumbent setae and

Fig. 5 *Rhagoletis completa* – male (length idem female).

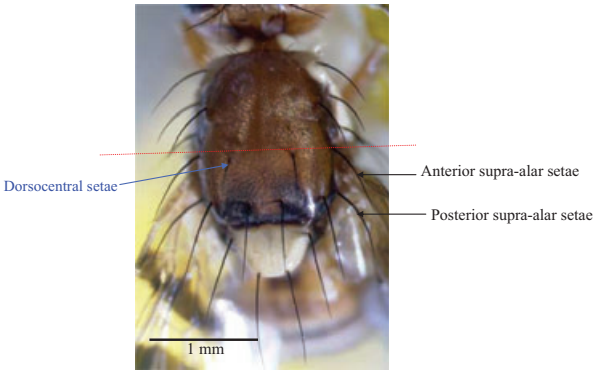

Fig. 6 *Rhagoletis completa* thorax.

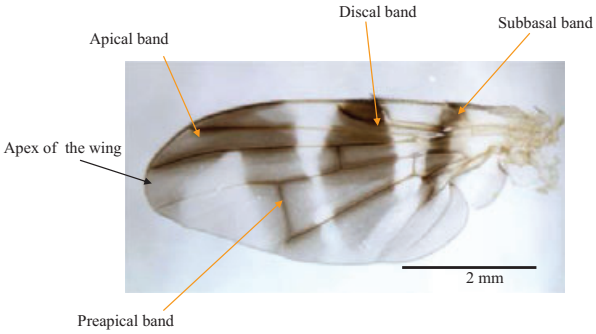

Fig. 7 *Rhagoletis completa*: wing.

microtrichia, without vertical banded pattern. Proepisternal bristles light yellow; all other major bristles black. Two anepisternal bristles of about equal size. Pleural and katepisternal regions brownish black. Notopleural stripe light yellow; scutellum light yellow grading to deeper golden yellow at base; halteres golden yellow. Base colour of postscutellum golden yellow with two broad vertical brownish black bands; some specimens with postscutellum concolorous brownish black.

- *Female*: Dorsum, humeral stripe and scutellum as in male. Sternal and katepisternal regions lighter with variable brownish black markings on sternal regions, rarely slight brownish black markings on dorsal margin of katepisternum. Vertical stripes present on postscutellum but frequently much narrower than in male.

Legs:

- *Male*: Colouration highly variable. Coxae I and II generally brownish black; coxa III golden yellow. Trochanters golden yellow to tan. Femur I brownish black with dorsal rows of bristles golden yellow to brownish black; ventral row black. Femur II tan with single row of weak semi-erect stout setae on anterior surface. Femur III golden yellow or tan grading to brownish black ventrally. Tibia I and II straw coloured. Tibia III golden yellow grading to brownish black ventrally; outer surface with single row of about eight yellowish orange semi erect setae.
- *Female*: All legs segments golden yellow without black markings; last tarsal segment sometimes dark yellowish brown. All setae and bristles as in male.

Fig. 8 *Rhagoletis completa* abdomen.

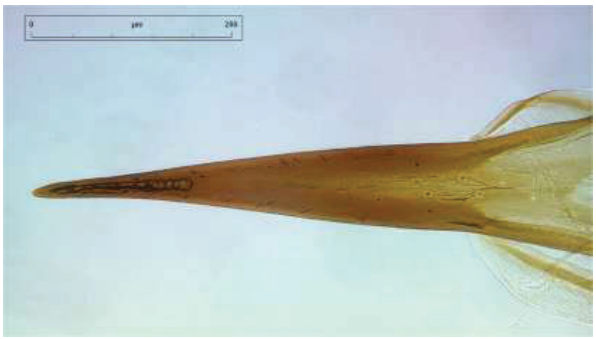

Fig. 9 *Rhagoletis completa* aculeus.

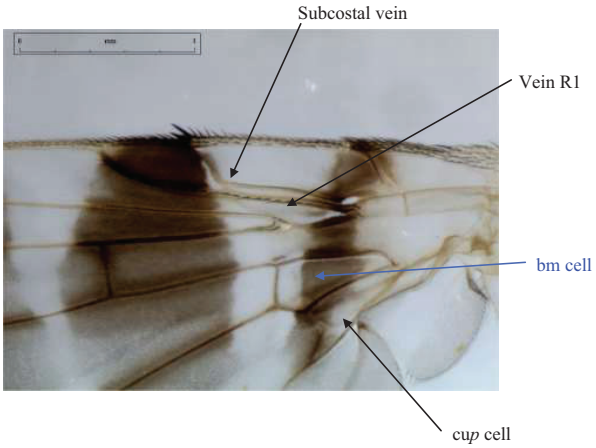

Fig. 10 *Rhagoletis completa* wing – detail.

Wings: (Fig. 7) Apical band complete and joined to preapical band. Discal band not joined to subbasal and rarely joined to preapical band. Junction of R2 + 3 and R4 + 5 with 0–2 setae dorsally, 0–2 ventrally; R4 + 5 bare.

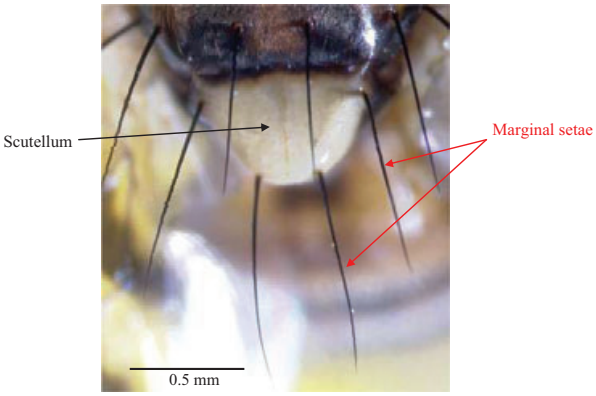

Fig. 11 *Rhagoletis completa* scutellum.

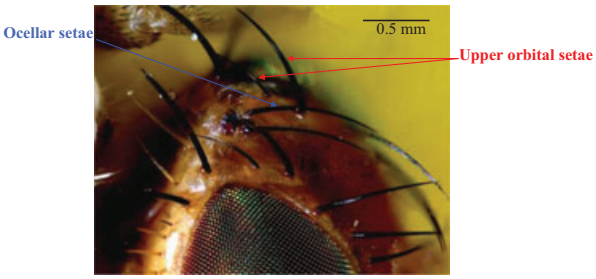

Fig. 12 *Rhagoletis completa* top of the head.

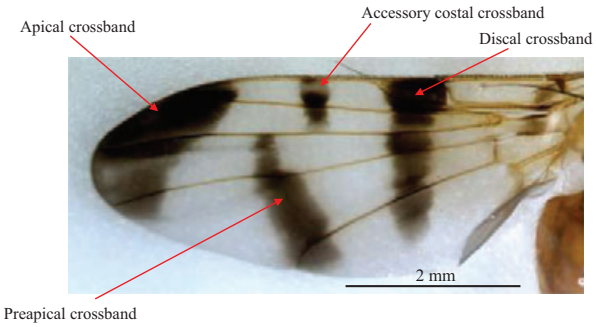

Fig. 13 *Rhagoletis meigenii* (Loew) – wing.

Abdomen: (Fig. 8)

- *Male*: Colouration highly variable. Normally tergites I and II golden yellow with some brownish black shading on either side of median line. Tegites III–V generally golden yellow but becoming progressively more heavily marked with brownish black; tergite V usually almost entirely brownish black. Tergites II–IV with pollinose cream coloured band along posterior margin.
- *Female*: As in male colouration variable, but all segments substantially more golden yellow than male. Tergites III–V usually marked with some dark shading on either side of medial line and with cream colored pollinose band along posterior margin of tergites III–IV.

Aculeus: (Fig. 9) Length 0.8–1.1 mm, pointed (see the different picture of *Rhagoletis*'s aculeus in White & Elson-Harris, 1992). This comparison is not enough for identification but can confirm if there is some doubt.

Reference material

ANSES Plant Health Laboratory (LSV) – Entomology and Invasive Plants Unit Montpellier, CBGP, Campus International de Baillarguet, CS 30016, 34988 Montferrier-sur-Lez Cedex.

Acknowledgements

This protocol was originally drafted by V. Balmès. ANSES Plant Health Laboratory (LSV) – Entomology and Invasive Plants Unit Montpellier, CBGP, Campus International de Baillarguet, CS 30016, 34988 Montferrier-sur-Lez Cedex, France. E-mail: valerie.balmes@anses.fr.

Feedback on this Diagnostic Protocol

If you have any feedback concerning this Diagnostic Protocol, or any of the tests included, or if you can provide additional validation data for tests included in this protocol that you wish to share please contact diagnostics@eppo.fr.

References

Boyce AM (1934) Bionomics of the walnut husk fly, *Rhagoletis completa*. *Hilgardia* **8**, 363–579.

Bush GL (1966) The taxonomy, cytology and evolution of the genus *Rhagoletis* in North America (Diptera: Tephritidae). *Bulletin of the Museum of Comparative Zoology* **134**(11), 431–562.

Carroll LE, Norrbom AL, Dallwitz MJ & Thompson FC (2004) Pest fruit flies of the world –larvae. http://delta-intley.com [accessed on 13 April 2005].

Duso C & Dal Lago G (2006) Life cycle, phenology and economic importance of the walnut husk fly *Rhagoletis completa* Cresson (Diptera: Tephritidae) in northern Italy. *Annales de la Societé Entomologique de France (n.s.)* **42**(2), 245–254.

EPPO (2011) PQR - EPPO database on quarantine pests (available online). http://www.eppo.int

EPPO/CABI (1997) *Rhagoletis completa* In: *Quarantine Pests for Europe*, 2nd edn, Eds. Smith IM, McNamara DG, Scott PR, Holderness M, pp. 475–479. CAB International, Wallingford (GB).

Foote RH (1981) *The Genus Rhagoletis* Loew South of the United States (Diptera: Tephritidae). Technical bulletin of the U.S. Department of Agriculture 1607, 75 pp.

Merz B (1994) Diptera: Tephritidae. Insecta. *Helvetica Fauna* **10**, 198.

Papp L & Darvas B (2000) *Contribution to a Manual of Palaearctic Diptera*, Vol. 1. Science Herald, Bupapest, 978 pp.

Stehr FW (1991) *Immature Insects*, Vol. 2. Kendall/Hunt Publishing Company, Dubuque (IA), 975 pp.

White IM & Elson-Harris M (1992) *Fruit Flies of Economic Significance: Their Identification and Bionomics*. International Institute of Entomology/CAB International, Wallingford (UK), 601 pp.

Yee WL & Goughnour RB (2008) Host plant use by and new host records of apple maggot, western cherry fruit fly, and other *Rhagoletis* species (Diptera: Tephritidae) in western Washington state. *The Pan-Pacific Entomologist* **84**, 179–193.

Appendix 1

For identification of the family Tephritidae see Papp & Darvas, 2000.

Identification of the genera *Rhagoletis*

After White & Elson-Harris (1992). [Note that this key adapted from White & Elson-Harris (1992) is not exhaustive. It will only separate *Rhagoletis* from the four other major pest genera; users should ensure that the specimens match the species description given].

1	Subcostal vein abruptly bent and dorsal side of vein R1 with setulae (Fig. 10)	Tephritidae 2
	Subcostal vein not abruptly bent or dorsal side of vein R1 lacks setulae	Other families
2	Cell cup broad, always considerably broader than half depth of cell bm, and usually about as deep as cell bm (Fig. 10). Apex of vein M meeting C with a distinct angle. Cell cup extension short, never more than one-fifth as long as vein A_1 + CuA_2	3
	Not with this combination of characters. Cell cup different. Apex of vein M turned anteriorly to merge with C without a distinct angle	Other genera
3	Scutellum with 4 marginal setae and usualy entirely cream to yellow (Fig. 11). Scutum with dorsocentral setae based close to a line between the anterior supra-alar setae (Fig. 6)	4
	Scutellum with a different number of marginal setae or scutum with dorsocentral setae based close to a line between the posterior supra-alar setae	Other genera
4	Head with 2 pairs of orbital setae. Ocellar setae long, usually similar in length and strength to orbital setae. (Fig. 12) Scutellum usually entirely cream to yellow; if marked with black, the black areas are confined to the base and lateral areas	*Rhagoletis*
	Chaetotaxy different or large black patches on scutellum at the base of each setae and near the centre of the dorsal area	Other genera

Appendix 2

Identification of the adult of *Rhagoletis completa*

After Foote (1981), White & Elson-Harris (1992) and Merz (1994).

1	Thorax and abdomen predominantly brown to yellow (Figs 6 and 8)	2
	Thorax and abdomen predominantly black	Other species
2	Accessory costal crossband absent	3
	Accessory costal crossband present (Fig. 13 – see the illustration for *Rhagoletis meigenii*)	Other species

3	Apical band of wing pattern complete, not at all narrowed at its junction with preapical band (Fig. 7). Discal and preapical crossbands usually separated	4
	Apical band of wing pattern narrowed or broken at its junction with preapical band (Fig. 13). Discal and preapical crossbands joined	Other species
4	Apex of the wing black, covered by the pattern of apical band (as shown in Fig. 7)	*R. completa*
	Apex of the wing hyaline, not covered by the pattern of apical band	Other species

Appendix 3

Preparation of aculeus for observation under a binocular microscope with ×200 or ×400 magnification

Break off the abdomen of the female and place it in a 10% potassium hydroxide solution, 1 h at room temperature or 20–30 min below boiling temperature.

When the abdominal sclerites are smooth enough, remove them leaving only the aculeus. Use a pin to separate aculeus and take care to not damage the tip of the aculeus.

Transfer the aculeus into distilled water for approximately 20 min and mount on a glass slide in a drop of glycerol with a cover slip.

This preparation method produces a temporary mount. There are also published permanent methods available (e.g. White & Elson-Harris, 1992).