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Standard Specification for *Aphidoletes aphidimyza* (Rondani) (*Diptera: Cecidomyiidae*)[1]

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1. Scope

1.1 This specification covers information on and the test method for determining the purity and number of adults released in commercial containers of the aphid predatory midge, *Aphidoletes aphidimyza* (Rondani).

1.2 The values stated in SI units are to be regarded as the standard. The values given in parentheses are for information only.

2. Referenced Documents

2.1 *ASTM Standards:*[2]

E2200 Specification for Information Included with Packaging of Multi-Cellular Biological Control Organisms (Withdrawn 2010)[3]

3. Terminology

3.1 *name of product—Aphidoletes aphidimyza* Rondani

3.2 *preferred host prey*—most species of true aphids (*Aphidoidea*).

3.3 *life stage when shipped*—pupal.

4. Classification

4.1 *Phylum*—Arthropoda.

4.2 *Class*—Insecta.

4.3 *Order*—Diptera.

4.4 *Family*—Cecidomyiidae.

4.5 *Genus*—Aphidoletes.

4.6 *Species*—aphidimyza.

5. Summary of Test Method (Determining the Purity, Sex Ratio, and Number of Adults Released from Containers of *A. aphidimyza*)

5.1 This test method describes a method of determining the purity, sex ratio, and number of adult *A. aphidimyza* released in containers.

5.2 This test method is for a standard commercial package of 250 to 1000 midges packaged in vermiculite carrier in trays or bottles. Adult midges and any parasites are allowed to emerge over a 21 day period and are counted and the sex ratio determined. Any live parasite or arthropod product contaminants are identified and recorded.

6. Significance and Use

6.1 The biological control of aphids by *A. aphidimyza* depends on accurate release numbers, presence of male and female adults, and absence of live product contaminants such as parasites and other insects and mites. This test will be typically employed by producers and users of the product to assess the number of live biological control agents delivered. It is complementary to the quality guidelines for *A. aphidimyza* that were developed by the International Organization of Biological Control (IOBC) by J. van Schelt in 1998.

7. Materials

7.1 Clear (translucent) or white plastic container, with a tight fitting lid (35 to 40 L capacity) with a bottom surface area of approximately 1500 cm^2), parallel lines marked 2.5 cm apart across the entire bottom of the bin with permanent marker to aid in counting. One container is required for each test unit.

7.2 A dissecting microscope or magnifier (7 to 20×).

7.3 A fine brush.

7.4 A hand tally counter.

8. Test Unit

8.1 A single container of *A. aphidimyza* is considered a test unit. This test is designed for standard shipments of 250 to 1000 midges. In large shipments, a minimum of three containers should be chosen at random for testing.

9. Pre-Test and Test Conditions

9.1 Samples should be held in a warm place at 22 to 25°C, and the vermiculite should be slightly moist and maintained at

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[2] For referenced ASTM standards, visit the ASTM website, www.astm.org, or contact ASTM Customer Service at service@astm.org. For *Annual Book of ASTM Standards* volume information, refer to the standard's Document Summary page on the ASTM website.

[3] The last approved version of this historical standard is referenced on www.astm.org.

high relative humidity of 70 to 80 % and a light regime of 16L:8D (out of direct sunlight) during these tests.

10. Sampling

10.1 Sex Ratio test requires a minimum of 150 midges per batch randomly selected from the emergence test.

11. Sample Preparation and Treatment

11.1 For each test unit, place the open bottle or tray containing the midges in the center of the emergence container and fasten the lid tightly. The majority of the adult midges will emerge over a period of 14 days, but to check for the presence of parasites, the sample should be held for a total of 21 days.

12. Counting Procedure

12.1 Remove the sample from the emergence bin, and count all of the dead adult midges on the bottom of the container using the reference lines to aid counting.

12.2 *Purity*—To check for purity, examine the inside of the container and surface of vermiculite for live aphids or other insect or mite contaminants during the quantity and emergence test above and while counting. If live contaminants are found, identify and record these. Parasites are not commonly found, but the parasites *Aphanogmus fulmeki* (Ashmead) and *Synopeas rhanis* (Walker) have been recorded on *A. aphidimyza*.[4]

13. Sex Ratio

13.1 The sex ratio may be determined by examining a sample of 150 dead adults from the quantity and emergence test under magnification. The female midges are easily distinguished from the males as females have short antennae without hairs while males have long curved hairy antennae (see Fig. 1).

14. Interpretation of Data

14.1 The total number of emerged adults should be equal to or greater than the number specified on the package.

14.2 No live contaminants should be found in this test.

14.3 The sex ratio should be equal to or greater than 45 % females (see 6.1).

14.4 If any of the above conditions are not met, the shipment is considered below standard.

15. Precision and Bias

15.1 The precision and bias of these methods have not been determined.

[4] Gilkeson, L.A., et al , "Aphanogmus fulmeki Ashmead (Hymenoptera: Ceraphronidae), a parasitoid of Aphidoletes aphidimyza Rondani (Diptera: Cecidomyiidae)," *Can. Entomol*, Vol 125, No. 1, 1993, pp. 161–162.

FIG. 1 Male Adult Aphidoletes aphidimyza Showing Long Curved Hairy Antennae

16. Keywords

16.1 *Aphidoletes aphidimyza*; quantity; purity; sex ratio

BIBLIOGRAPHY

(1) van Lenteren, et al., "Guidelines for Quality Control of Commercially Produced Natural Enemies," *Quality Control and Production for Biological Control Agents—Theory and Testing Procedures*, J.C. van Lenteren, Ed., CABI Publishing, 2003, pp.278 –279.