


IRAC Susceptibility Test Methods Series

Method No: 026

Version: 1

Details:

Method:	No: 026	 <p>♀</p> <p>Photograph Courtesy of: Bayer CropScience</p>
Status:	Approved	
Species:	<i>Musca domestica</i>	
Species Stage	Adult female	
Product Class:	Neonicotinoids	
<p>Comments: This method is suitable for use with commercial formulations of insecticides. This method was developed by Bayer CropScience AG and validated for endorsement as an IRAC approved method</p>		

Objectives:

Susceptibility Baseline:

Resistance Monitoring:

Description:

Materials:

Sucrose (Merck, Nr.: m.1.07651.1000), dental wicks (orbis dental Handels GmbH, Nr.: 107562), plastic container (polystyrene containers 175 ml, PS, Greiner BioOne, Nr.: 960177), foam plugs (ceaprene stopper, GreinerBioOne, Nr.: 332070), acetone (technical grade preferred, but analytical also acceptable), syringes/pipettes for liquids or weighing balance for solids, syringes/pipettes for making dilutions.

Methods:

Collect representative house fly samples. The insects should not be subjected to high temperature, humidity or starvation stress after collection.

If there are insufficient insects available, or further tests are desirable, housefly may be reared in the laboratory and the F1 generation is used. The sex of the flies is determined after anaesthetization with CO₂.

Preparation of insecticide dilutions:

The required serial dilutions of insecticide solution are prepared in 20% w/v sucrose using formulated insecticide. Approximately 5ml of insecticide solution is required for each concentration. 20% Sucrose is used as a control solution.

Example of imidacloprid dilutions:

1000 µg a.i./ml
200 µg a.i./ml
40 µg a.i./ml

Note: When using a sucrose stock solution insure there is no fungal contamination!

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Application:

A piece of dental wick (2cm) is placed in a 175ml plastic container and treated with 1.2 ml 20% aqueous sucrose with or without insecticide.

Introduction of test flies:

Ten uniform size females (fly weight: WHO-N $\approx 11,5 \pm 1,7$ mg RW $\approx 16,6 \pm 1,0$ mg) are introduced to each container (3 replicates per concentration) and containers are then covered with ceaprene foam plugs.

Assessment:

Mortality is scored after 48h at $23 \pm 2^\circ\text{C}$, 50% RH and 12L:12D photoperiod. At assessment, flies are classed as either: (a) unaffected, giving a normal response (making a coordinated move) when gently stimulated by dropping down the containers, or (b) dead or affected, the latter giving an abnormal response to stimulation or showing abnormal movement. Thus, % response or mortality will include both dead and affected flies. Mortality of untreated flies should be recorded.

Express results as percent mortality and correct for untreated mortality using Abbott's formula.



Photographs Courtesy of: Bayer CropScience

Precautions & Notes:

1. Disposable plastic equipment is preferred provided that it is not affected by the formulation constituents; glass equipment may be used but must be adequately cleaned with an appropriate organic solvent before use. To avoid higher control mortality it is necessary to use containers with adequate volume as recommended in materials. Smaller containers lead to higher control mortality.
2. Insecticide products (formulations) may contain varying concentrations of active ingredient(s). Ensure insecticide dilutions are based on active ingredient content.
3. Where possible, bioassays to measure the variation in insecticide susceptibility should run in parallel with a bioassay to measure the susceptibility of a known susceptible standard population of houseflies (e.g. WHO-N).

References & Acknowledgements:

This IRAC method is based on a method developed by Bayer CropScience.