NOTE

A Caging, Handling, and Bioassay Procedure for Eggplant Flea Beetle, *Epitrix fuscula* Crotch (Coleoptera: Chrysomelidae)[1]

Paul McLeod,[2,3] Tahir Rashid,[4] and Don Steinkraus[2]

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The eggplant flea beetle, *Epitrix fuscula* Crotch (Coleoptera: Chrysomelidae), is a serious pest of eggplant, *Solanum melongena* L. var. *esculentum* (Solanaceae), at time of transplanting. Eggplant seedlings, both commercial and for home gardens, are grown in greenhouses and moved to the field as close to the local frost-free date as possible in the spring. At this stage of plant development, plants are susceptible to injury from flea beetles. In the Mid-South of the U.S.A., eggplant flea beetles overwinter as adults. Flea beetles can locate the new seedlings within hours of transplanting and begin to feed on both the upper and lower leaf surfaces (McLeod 2006). The beetles initially produce small "shot-holes" and as feeding continues, the small holes can coalesce into large areas of necrotic leaf tissue. Commercial and home garden eggplants often require flea beetle management with foliar applied insecticides. Without this management, heavily infested transplants can be killed. Injury is common along the edges of large commercial eggplant fields (Diaz et al. 2004). If transplants survive flea beetle attack for two or three weeks and initiate rapid growth, the eggplant flea beetle appears to have little additional impact on the plant (Sorensen & Baker 1994).

Several effective insecticides are currently labeled for eggplant flea beetle management on eggplant. However, interest exists in the development of alternative insecticides, especially for the organic market. Although field testing of candidate insecticides generally provides the best information for new insecticide selection, this can be expensive and may require several seasons of testing. Laboratory bioassays can be utilized to rapidly screen new insecticides for toxicity to various insect species. This information can be used to rapidly narrow the list of insecticides for further testing in field trials. With the eggplant flea beetle, laboratory bioassays have been difficult due to a lack of an available rearing technique, the minute size of the insect, and difficulties with the jumping behavior of the flea beetle. This report describes a procedure for eggplant flea beetle collection from the field, handling, and determination of insecticide toxicity with a laboratory bioassay.

The flea beetles used in the study were collected from field produced "Black Beauty" eggplant grown at the University of Arkansas Main Experimental

[1]Accepted for publication 2 September 2011.
[2]Department of Entomology, University of Arkansas, Fayetteville, Arkansas 72701.
[3]Corresponding author (pjmcleod@uark.edu).
[4]Alcorn State University, 294 Grainger Dorsey Road, Mound Bayou, Mississippi 38762.

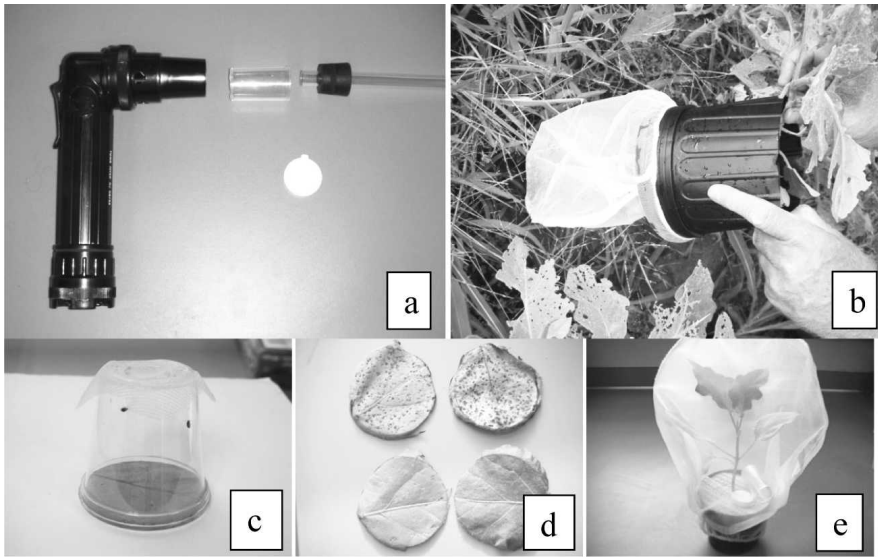

Fig. 1. Materials for eggplant flea beetle collection and bioassays; a. Battery powered aspirator with collection vial; b. paint strainer with top half of 3.8-L plastic pot for field collection; c. test chamber for leaf disk bioassay; d. flea beetle feeding on non-treated (upper) and insecticide treated leaf disks; e. test chamber for whole plant bioassay.

Station, Fayetteville, AR. Transplants were produced in 5.5 × 5.5 × 7.6 cm peat cups in late winter in a greenhouse and set into the field each year in late April. Plants were produced following local recommendations including fertilization, weed management, and irrigation practices. No insecticides were applied. Plants grew and produced fruit throughout the production season until freezing temperatures occurred in late October. The same field was used from 2008 through 2010.

Flea beetle adults were collected using two methods. Initially, individual beetles were vacuumed from the leaf surface with the aid of a hand-held "flashlight" aspirator (Hausherr's Machine Works, Toms River, NJ). The collection tube consisted of a 7-dram (26 ml) plastic vial with the bottom removed and covered with screen (Fig. 1a). Once the desired number of beetles was aspirated into the tube, the aspirator neck was removed and a cap was placed over the opening. This was completed with the aspirator pump running to prevent beetle escape. Capped vials were held in a cooled ice chest until the required number was collected. At times, when greater numbers of flea beetles were needed, an alternate collection method was used. Attempts at sweeping flea beetles from eggplant foliage similar to the method used for rape flea beetle, *Phyllotreta cruciferae* (Goeze), on canola (McLeod & Weiss 1992) proved difficult because sweeping broke the petioles of the eggplant leaves. To reduce this damage, infested eggplant leaves were shaken over a 3.8-L elastic top paint strainer (Trimaco, Durham, NC). The strainer was held open by inserting the upper half of a 3.8-L black plastic pot (Fig. 1b). When sufficient flea beetles were

Table 1. Percent mortality and feeding of eggplant flea beetles on eggplant (SE in parentheses).

	No. days in test arena	Percent mortality	No. feeding pits
Leaf disk bioassay	1	0 (0.00)	81.4 (3.66)
	2	0 (0.00)	134.9 (5.90)
	3	0 (0.00)	162.5 (5.32)
	5	2 (1.38)	226.0 (4.81)
Whole plant bioassay	1	0 (0.00)	32.6 (2.40)
	2	0 (0.00)	63.1 (3.11)
	3	0 (0.00)	102.0 (5.16)
	5	2 (1.38)	147.0 (3.53)

collected the net was twisted shut and closed with a rubber band. Once in the laboratory, beetles were aspirated into test chambers.

Two types of bioassays were developed. The first was similar to the procedure described by McLeod & Weiss (1992) for testing insecticide efficacy on horseradish, *Armoracia rusticana* P. Gaertn., B. Mey & Scherb., foliage against the rape flea beetle. Eggplant foliage was collected from greenhouse-produced plants and sprayed with various concentrations of insecticide with a hand operated sprayer (Gilmour Manufacturing Co., Somerset, PA). After drying, leaves were placed over a 28-ml plastic cup (Solo Cup Co., Lake Forest, IL) with the treated side to the inside of the cup (Fig. 1c and d). Cup bottoms had been removed and replaced by hot gluing a piece of a paint strainer described above. A loose fitting cardboard cap was placed over the leaf and five field-collected eggplant flea beetle adults were aspirated into the cup. The cap was quickly pushed into the cup rim. Leaf edges were trimmed with a razor blade and the cup was inverted over moist blotter paper in a plastic tray.

To check flea beetle feeding and survival in the test arena, five field-collected adult eggplant flea beetles were aspirated into each of 20 cups containing non-treated eggplant foliage. Cups were inverted on moist blotter paper in an environmental chamber set at 23°C and 12:12 h day:night lighting. Counts of feeding punctures and beetle mortality were made 1, 2, 3, and 5 days after beetles were enclosed. No mortality was detected on the 1, 2, and 3 day observations (Table 1). On day 5, a single dead flea beetle was observed in each of two cups. The mean number of feeding pits was 81.4, 134.9, 162.5, and 226.0 for day 1, 2, 3, and 5, respectively. Although no additional beetle mortality occurred for the next two days and feeding continued, leaf tissue began to desiccate and feeding pits overlapped and became difficult to count. The testing procedure proved effective. However, care was needed to avoid eggplant foliage with large leaf veins. If the leaf vein was pressed into the cup, the leaf tissue sometimes tore and beetles were able to move into the space between the cap and leaf, which made counting difficult.

An alternate bioassay was developed using eggplant seedlings from the greenhouse. Plants were produced in a greenhouse in 15-cm diameter plastic pots

to the 4-leaf stage. After spraying plants with the desired insecticide concentration, they were fitted with two 15-cm diameter disks made from Sill Seal Foam Gasket (Dow Chemical Co., Midland, MI). Each disk was slit from the outer edge to the center to enable it to fit around the plant stem. Foam disks were turned to prevent overlap of the slits. The foam disks prevented beetle movement into the potting soil. A capped 7-dram (26 ml) vial containing five eggplant flea beetles was placed on the foam. Plants and vials containing beetles were next enclosed in a 3.8-L paint strainer described above (Fig. 1e). Although the elastic band was sufficient to prevent beetle escape, an additional rubber band was added. Finally, the vial was lifted within the paint strainer and the cap was carefully removed to release the beetles. Both cap and vial were left on the foam disk with the enclosed plant. Pots were placed in plastic trays and watered daily from the bottom. A study similar to the leaf disk study was completed with the whole plant test arena.

A vial containing five adult eggplant flea beetles was placed in each of 20 whole plant cages containing eggplant seedlings. Vial caps were removed and flea beetle mortality and feeding on the uppermost two unfolded leaves were determined 1, 2, 3, and 5 days after beetle release. Beetles moved from the vial to the plant within minutes and readily fed on both top and bottom of the eggplant foliage. No flea beetle mortality was observed until five days after release when one dead beetle was detected in each of two cages (Table 1). The mean number of feeding pits per leaf was 32.6, 63.1, 102.0, and 147.0 for day 1, 2, 3, and 5, respectively. Although mortality and feeding were not determined after 5 days, eggplant flea beetles remained active on non-treated control plants and easily survived for a two week period. The procedures described herein provide a usable method for eggplant flea beetle collection and bioassay on eggplant.

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