PLANT SCIENCES
ENTOMOLOGY (GENEVA) • 16

NEW YORK STATE AGRICULTURAL EXPERIMENT STATION, GENEVA, A DIVISION OF THE NEW YORK STATE COLLEGE OF AGRICULTURE AND LIFE SCIENCES, A STATUTORY COLLEGE OF THE STATE UNIVERSITY, CORNELL UNIVERSITY, ITHACA

Simplified rearing and bioassay for the seedcorn maggot, *Hylemya platura* (Meigen)

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Introduction.—Before 1926, Leach (7) was able to rear larvae of the seedcorn maggot (SCM), now known as *Hylemya platura* (Meigen), on potatoes or beef extract agar inoculated with the causal organism of potato blackleg, *Erwinia atroseptica* (van Hall) Jennison. Since then, attempts to maintain colonies in the laboratory have met with limited success because of low egg production until McLeod found that protein was essential for oviposition (8). Harris et al. (5), using the information of McLeod, was able to establish a productive laboratory colony of SCM. Although excellent results are obtained with this technique, procedures are somewhat time consuming. A simplified technique has been developed at the Geneva Experiment Station which produces large numbers of SCM for use in laboratory studies.

Adults.—Flies are maintained in screened cages (2 or 3 $ft.^3$) stocked with 1,000-2,000 individuals/cage. For maximum egg production, Brewer's yeast or yeast hydrolysate as the protein source is presented in a small open dish. Water is always available by means of a dental wick inserted in a narrow necked flask. In addition, a liquid diet (No. 116, General Biochemicals, Laboratory Park, Chagrin Falls, Ohio 44022) placed in a small vial with a dental wick or an inexpensive dry diet containing 10 parts skimmed milk powder, 10 parts sugar, 1 part Brewer's yeast, and 1 part soy peptone (11) is used (Fig. 1). Both diets have been equally effective.

To maintain colony vigor, 200 new individuals are added to the cage each week after oviposition begins. Flies are kept in a controlled environment of 16 hours of light, 50 per cent relative humidity, and a temperature between 21 -23 C.

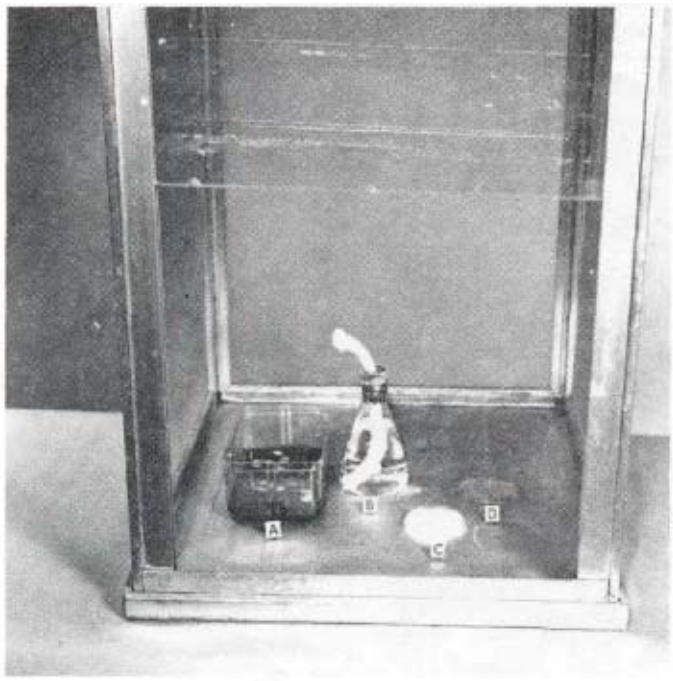

Figure 1.—Rearing cage equipped with: (A) oviposition dish containing squash seeds, (B) water flask, (C) dish of dry diet, and (D) dish of Brewer's yeast.

Under these conditions, adults emerge from pupae within 7 days and survive for ca. 30 days. Oviposition peaks between days 13 and 20.

Eggs.—Eggs are oviposited in a plastic container (5 x 3.5 x 2.3 in.) partially filled with moist washed greenhouse

sand with several grams of butternut squash seed placed on top. The squash seed produces a strong ovipositional stimulant (1) (Fig. 1). Eggs are collected three times weekly by washing the contents of the dishes into a 1,000-ml beaker. A modified Horsfall technique (6) is used to separate the eggs from the sand by stirring a saturated sodium chloride solution into the sand mixture in the beaker. Most of the eggs and seeds float to the surface and are then decanted onto a 100-mesh screen. These are washed into a crucible with fresh water, and then they are poured onto paper towels where the eggs are rinsed from seeds with water from a plastic wash bottle. If larvae are needed, the eggs are washed from the paper toweling onto a cloth which is then placed over moist paper towels in a covered plastic dish. A small amount of Corenco meat and bone meal is sprinkled over the eggs to provide food for the hatching larvae. The container is placed under the same environmental conditions described above. If they are kept in this manner for more than 4 days, additional meat and bone meal and water must be provided. Although larvae appear to develop normally on meat and bone meal (Fig. 2), we do not mass rear with this medium. We prefer to maintain the species in part on host plant material (lima beans) to reduce the pressure for selection of a strain which may prefer to feed only on the meal. Thus, we use this technique only to maintain larvae for short periods of time until they can be used in laboratory studies.

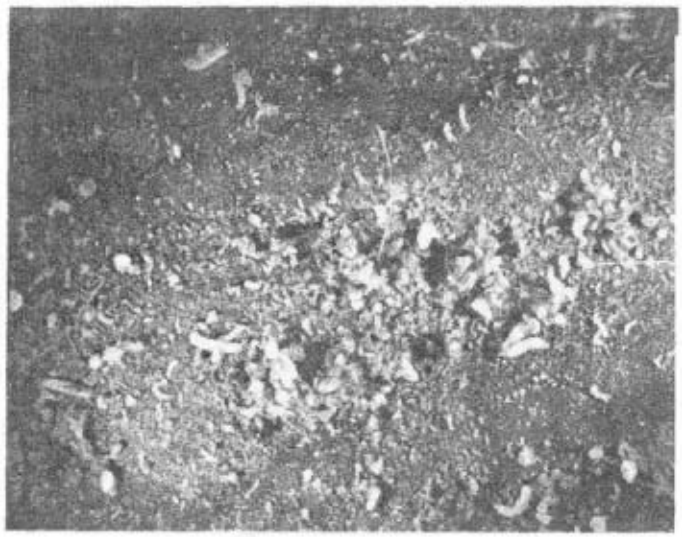

Figure 2.—Closeup of dish containing various instars of larvae feeding on a thin layer of meat and bone meal.

Larvae.—A plastic dish (7x5x2.3 in.) with a .3-inch hole in the bottom and a piece of dental wick extending through it is placed into another dish of the same dimensions which is filled with water. The top container is filled with greenhouse sand into which are pushed 50 lima bean seeds. Approximately 50 ml of Corenco meat and bone meal are sprinkled over the beans (Fig. 3). Apparently, the hatching larvae begin to feed on the meal and then move to the softening beans where they complete their larval development. The larval containers are removed twice a week from the fly cage and replaced with new ones. The boxes containing lima beans, meat and bone meal, and newly deposited eggs are placed under controlled conditions of environment (see above) for ensuing larval development. Extra lima beans and meat and bone meal may be added when needed.

Pupae.—After 14-16 days of development, most of the

Figure 3.—Larval dish containing greenhouse sand, meat and bone meal, and remnants of lima beans. A second dish underneath the container provides constant moisture to the sand by means of a dental wick.

larvae will have pupated. Since sudden changes in temperature may damage the pupae, they are collected by washing the contents of the box with lukewarm water into a 60-mesh sieve. After the remnants of the lima beans are removed by hand, the pupae are floated into a container of water for additional cleaning, and then they are poured onto a screen for separation and counting. Each box yields ca. 1,000-1,500 pupae when the colonies are properly maintained.

SCM pupae can be stored in moist sand in plastic containers (2 x 1.5 in.) at a temperature of 2-4 C. Per cent emergence and vigor of flies decreases gradually over a period of time, but at 6 months it drops rapidly. Also, cold storage of non-diapausing pupae prolongs emergence of the flies. It may take up to 3 weeks for flies to emerge from pupae which have been stored for 6 months. Techniques described under **Larvae** and **Pupae** sections are used primarily for colony maintenance.

BIOASSAY

Introduction.—Field evaluations using candidate pesticides are time consuming, particularly when several dosage levels, methods of application, soil types, etc. are evaluated. In view of rising costs of time and labor, it is essential to conduct most of the preliminary experiments in

the laboratory. Thus, only those materials with proven laboratory efficacy are tested in the field where the final decision on rate and application method should be made. Commercial control of SCM is usually achieved by coating host seeds with small amounts of toxicant. However, bands of insecticide either in or over the seed furrow will also provide satisfactory protection, although more active ingredient per acre is needed than when using the seed economical treatments.

Several excellent laboratory bioassay techniques for root and SCM flies have been developed (3,4). These are useful when testing contact insecticides on the adults, but most damage from root maggots occurs underground by larvae. We feel that studies with insecticides to control seed and root larvae are complicated by the complex environments in soils. Furthermore, SCM larvae may be present in the soil **before** a crop is planted. A technique with either rutabaga slices or other plant tissue has been used to assay pesticide efficacy on the closely related root maggots (9,10) but not on SCM larvae.

Recently a technique for evaluation of seedcorn maggot larval control was developed at the New York State Agricultural Experiment Station, Geneva. Although it was briefly discussed in previous reports (2,12), we have never reported all of the details. Interest from other workers has prompted this report.

In the laboratory tests, SCM injury is rated about 2 weeks after seeds are infested with first instar maggots. This is the time when the first leaves on the emerged seedlings have unfurled to expose maggot injury (13). Another indicator of insecticidal effectiveness may be determined by washing out surviving larvae or pupae onto a 60-mesh screen for a live form recovery count. A treatment may kill the young larvae when they attempt to penetrate a seed, or the maggots may survive until after the seeds are injured, thus necessitating an increased dose of toxicant. Phytotoxicity to the seedlings may be assessed at any time after the seedlings have appeared.

Seed Coating.—-To test the efficacy of various seed coatings, treated seeds and first instar maggots are placed on a 1 -inch layer of moist washed greenhouse sand in a plastic box (5 x 3.5 x 2.3 in.). We usually replicate four times with each replicate containing five seeds/box. To maximize establishment of larvae, hard-coated seeds (such as beans) can be pre-softened in moist paper toweling for 24 hours before test initiation or a pinch of meat and bone meal may be placed over the seeds before they are completely covered with moist sand so that a food supply for the larvae is available while the seeds are softening. Maggots (maintained in the egg container described above) are transferred with a camel's hair brush next to the seeds before covering with sand. Ten maggots/seed will injure all untreated seeds severely. Smaller numbers of larvae may be used if lesser degrees of injury are desired. Finally, an additional layer of sand (1 in.) is placed over the seeds and maggots, the entire contents of the box are moistened, and a cover is added to maintain high humidity and to prevent escape of the larvae. The containers are stored in the controlled environment described above.

In-Furrow.—When testing a treatment of pesticide applied in the seed furrow, 2 inches of sand are placed in the plastic boxes and a 1 -inch deep furrow is made. The seeds, pretreated with a recommended commercial fungicide, are placed in the furrow. If a granular formulation is to be tested, a pre-weighed amount is mixed with dry greenhouse sand, and then this mixture is distributed evenly over the seeds. Because wild populations of SCM seldom come in contact with furrow applications immediately after planting in the field, a thin layer of dry sand is placed over the granules and the seeds in the furrow to insure that the larvae are not placed on the chemical band. Then the maggots are placed in the furrow and covered with sand to fill the furrow. If an emulsifiable formulation is to be tested, it is mixed with water and then applied with a pipette; however, careful calibration must be made to insure that proper rates are used. The amount of water may vary, but we compute our rates of pesticide on the basis of material/foot of row based on a 36-inch row width.

Over-Furrow.—Sprays or treatments of granules over the furrow would require a larger (7 x 5 x 2.3 in.) container than for seed or in-furrow treatments since they are generally put on in the field as 2-inch or larger bands. For spray application, we use a Jet Pak Power unit (Wards Natural Science Establishment, Rochester, New York 14603). Considerable care should be used to achieve the desired band width. The materials can be worked into the sand by any small hand implement if incorporation is desired.

After one week, the box covers are permanently removed until assessment of damage. During this 1 -week period after the removal of the covers, the sand must be remoistened several times. Because surviving maggots are actively feeding by this time, they are less inclined to escape from the boxes. When insecticides are tested, the sand should not be reused because it may retain chemical residues.

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