The Great Lakes Entomologist

Volume 16
Number 4 - Winter 1983 *Number 4 - Winter 1983*

Article 7

December 1983

Mass Rearing of the Greater Wax Moth, *Galleria Mellonella* (Lepidoptera: Pyralidae), for Small-Scale Laboratory Studies

M. A. Mohamed
University of Wisconsin

H. C. Coppel
University of Wisconsin

MASS REARING OF THE GREATER WAX MOTH, *GALLERIA MELLONELLA* (LEPIDOPTERA: PYRALIDAE), FOR SMALL-SCALE LABORATORY STUDIES[1]

M. A. Mohamed and H. C. Coppel[2]

ABSTRACT

A technique was developed to mass rear the greater wax moth, *Galleria mellonella*, economically (ca. 0.2 cents per larva exclusive of 3–5 h of labor costs per week). Mortality in the egg and early larval stages was ca. 48% whereas in later larval and pupal stages it was ca. 10% and 27% respectively. With a fecundity of 650–1120 eggs per female, and notwithstanding the high egg and early larval mortality, the procedure easily provides a self-sustaining culture with high yields of all stages.

The greater wax moth, *Galleria mellonella* (L.), is extensively cultured as a laboratory test animal for basic studies in many disciplines (physiology, biochemistry, toxicology, pathology, etc.). Additionally its egg, larval, and pupal stages are often used as hosts or prey for rearing parasitic and predaceous insects for both laboratory studies and field releases. Beck (1960) and Dadd (1964, 1966) originally formulated diets to culture *G. mellonella* in the laboratory. However, without modifications, we encountered variable survival rates among the various stages under mass rearing conditions. By manipulation of both the basic diet suggested and the procedures for rearing the greater wax moth we were able to establish an efficient and reproducible system for providing large numbers at a relatively low cost. Our system is herein described.

MATERIALS AND METHODS

The following ingredients comprise our diet:

Water, deionized, preboiled, 100 ml
Honey, raw or commercial brand, 150 ml
Glycerine, 50 ml
Beeswax, 3 g
Cholesterol, 1 g
Polyvisol Multivitamin supplement, 4 ml
Babyfood cereal, Gerber's Hi-Protein 454 g

Heat the deionized water to 80°C and separately add the honey, glycerine, beeswax, and cholesterol while swirling vigorously. When all ingredients are in solution allow to cool to ca. 50°C. Add the multivitamin supplement.

Place the cereal in a container and manually pulverize it to a fine dusty texture (a blender may be used). Add the above solution, while still warm, to the cereal in 50 ml increments, manually mixing it into the cereal after each addition. The resulting mixture should have a loose, friable consistency. The medium (diet) may be stored indefinitely at room temperature in a tightly closed container.

Adult wax moths from our stock cultures were placed into 1-liter mason jars where mating took place. Also placed into the jars were folded sheets of wax paper held together

[1]Research supported by the College of Agricultural and Life Sciences, University of Wisconsin—Madison, in part by USDA, SEA, AR Agreement #58-519-B-1-991 and in part by Federal Hatch Support, project no. 2116.

[2]Department of Entomology, University of Wisconsin, Madison, WI 53706.

with paper clips and on which the moths deposited their eggs. The eggs were removed from the jars and used to initiate the next generation of wax moths. We placed 115 g of the medium into each 500 ml sterilized mason jar, added 1000 wax moth eggs to the surface of the medium, and 0.5 ml of deionized preboiled water on and around the eggs. We then sealed the jar with aluminum foil. The jars were incubated in the dark at 28.5°C. Second instar larvae are readily observed in 7–10 days by the extensive tunnelling at the periphery of the medium. The foil is then removed and replaced with wire screening covered with cheesecloth. This allows adequate gaseous exchange and prevents excessive moisture buildup. The larvae remain in the medium for an additional 8–10 days (28.5°C, 60–70% RH, total darkness). By this time the medium is reduced to a fine, dusty, dark powder and the larvae are readily separated from it by screening. The larvae are transferred into a 1-liter mason jar to which 175 g of the medium are added. Within 4–5 days, fourth instar larvae become available, and in 7–8 additional days fifth instars are present. If pupae are desired another 7–10 days are required in incubation.

Those larvae which are not in the prepupal stage in the mason jar may be transferred to fresh medium (100 g) either in another jar or in aluminum baking pans covered with metal screens. The latter are incubated at 26.5 ± 0.5°C, 80–90% RH, and 24 h darkness to provide either final instar larvae in 2–3 days or a cocoon mat on the screen in 5 days.

As a measure of the efficiency of our system the following data were obtained: the percentage survival of eggs and instars 2 and 3, instars 4 and 5, and pupae, from 12 replicates each seeded with 1000 eggs; the percentage of adults eclosing from pupae and their ratio from 25 replicates of 50 randomly collected pupae obtained from the 12 replicate cultures originally used; the number of eggs deposited when one male was placed with one female (12 replicates) and when 25 males were placed with 25 females (eight replicates); and, the weight distribution of the pupae.

DISCUSSION OF RESULTS

Under the conditions of the cultural procedures discussed in the previous section, the highest mortality (ca. 48%) occurred from the egg stage to larval instars 2 and 3 (Table 1). As no cadavers were observed during handling of the stock, it is possible that low egg viability contributed significantly to mortality. Beyond larval instars 2 and 3, however, survival increased significantly to the pupal stage (90%). Survival of the pupae to the adult stage was approximately 73 ± 5.6% providing slightly over 300 adults from an initial seeding of 1000 eggs. The sex ratio of the emerging adults was approximately 1:1.

Egg production per female using a single female to a single male ratio was 1120 ± 57 (n = 12), and in a 25 female to 25 male ratio it was 16.3 ± 1.6 x 10^3 (n = 8). From the relatively high egg production obtained in either situation it becomes apparent that wax moth cultures may rapidly become self-sustaining in an almost exponential manner.

As an index of the relative homogeneity of the culture, a histogram (Fig. 1) shows a normal distribution of pupal weights (determined from a probability plot) where $\bar{x}$ ± S.D. was 171.4 ± 35.0/mg (n = 550).

Table 1. Survival (%) of the larval and pupal stages of the greater wax moth, *Galleria mellonella* (L.) from the egg stage.

Stage	Survival (%)[a]
2nd–3rd larval	52 ± 1.6
4th–5th larval	90 ± 1.3
Pupal	90 ± 0.9

[a]The percentage survival was calculated on the basis of the numbers of the stage listed as a ratio of the preceding stage. The initial number of eggs was 1000 for each of 12 replicates.

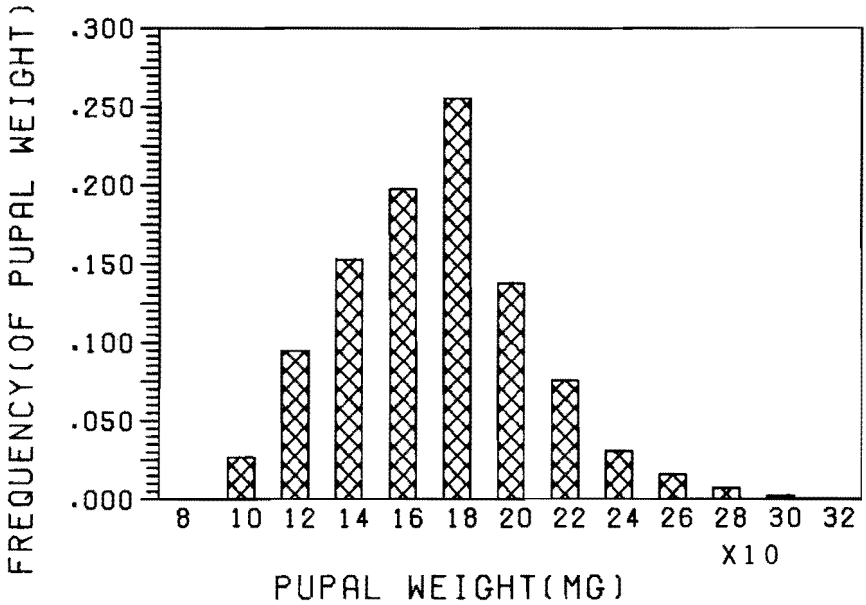

Fig. 1. Pupal weight distribution of the greater wax moth, *Galleria mellonella* (L.)

We have used this procedure for 1.5 years and have had no problems of reproducibility. Waste does accumulate on the bottom of the mason jars but it can be removed readily and separated from the intact upper layer in which most of the larvae aggregate. The waste layer can then be dried in the aluminum pans and the remaining larvae harvested.

The ingredients used are relatively inexpensive resulting in a cost per larva (exclusive of labor) of 0.2–0.3 cents. Approximately 3–5 h per week (labor) is required to maintain and harvest 5000–10,000 individuals of any stage of the greater wax moth per week.

ACKNOWLEDGMENT

The authors gratefully acknowledge the technical assistance of Jennifer Birdsall, Jean Adams, Steve Kratz and John Haanstad.

LITERATURE CITED

Beck, Stanley D. 1960. Growth and development of the greater wax moth, *Galleria mellonella* (L.). Trans. Wisconsin Acad. Sci. Arts, Letts. 49:137–148.

Dadd, R. H. 1964. A study of carbohydrate and lipid nutrition in the wax moth, *Galleria mellonella* (L.), using partially synthetic diets. J. Insect Physiol. 10:161–178.

_____.1966. Beeswax in the nutrition of the wax moth, *Galleria mellonella* (L.). J. Insect Physiol 12:1479–1492.