sippiensis in the AMNH collection were loaned by Norman Platnick, and Henrik Enghoff advised me of the sample in the Zoologisk Museum Copenhagen. Specimens of *maculatus* and *C. hubrichti* from Cheaha and Oak Mountain State Parks, respectively, were taken with permission of the State Parks Division, Alabama Department of Conservation and Natural Resources. Renaldo G. Kuhler, NCSM scientific illustrator, prepared Fig. 1-2. This research was supported in part by NSF Grant No. DEB 7702596.

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BIOLOGY OF *DIAPREPES ABBREVIATUS* (COLEOPTERA: CURCULIONIDAE) REARED ON AN ARTIFICIAL DIET

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ABSTRACT

A method is described for rearing the so-called sugarcane rootstalk borer weevil, *Diaprepes abbreviatus* (L.), on an artificial diet, and its biology on this diet is reported. Females and males emerged after a mean of 381 ± 4.7 days and 382 ± 6.4 days, respectively. The mean number of eggs per mass was 69.2 ± 7.8; the mean number of eggs deposited per female was 6517 ± 931.1. Females lived a mean of 147 ± 17.1 days, and males 135 ± 21.5 days. Virgin females produced a mean of 1910 ± 349.3 eggs. Most larval hatch occurred 7-8 days after oviposition, with the longest incubation period being 15 days. Hatchling larvae were able to survive for 53 days with adequate moisture.

RESUMEN

Se describe un método de criar *Diaprepes abbreviatus* (L.) sobre una dieta artificial, y se relata su biología sobre esta dieta. Las hembras y los machos nacen después de un promedio de 381 ± 4.7 y 382 ± 6.4 dias, respectivamente. El promedio de huevos en cada masa fué 69.2 ± 7.8; el promedio de huevos depositados por cada hembra fué 6517 ± 931.1. Las hembras vivieron un promedio de 147 ± 17.1 dias, y los machos 135 ± 21.5 dias. Las hembras vírgenes producieron un promedio de 1910 ± 349.3 huevos. La mayoría de las larvas nacieron 7-8 dias después de oviposicion, y el período más largo de incubación duró 15 dias. Con suficiente humedad las larvas recién naicdas pudieron sobrevivir por 53 dias.

Diaprepes abbreviatus (L.) is a major pest of citrus and sugarcane in Puerto Rico and the West Indies. In the United States, it is presently found only in the State of Florida, infesting citrus and other host plants in Orange, Seminole, Lake, and Broward Counties. The adult weevils feed on the foliage of at least 41 plant species in Puerto Rico (Martorell 1945), and the larvae are root feeders. Although *D. abbreviatus* has not been found infesting Florida sugarcane, it is a threat to that industry.

Previous reports on the biology and development of *D. abbreviatus* have been based on observations and information obtained from a few specimens recovered from plant material. These indicated that the insect has a highly variable developmental period that ranges from ca. 6 mo to over 2 yrs (Watson 1904; Ballow 1912; Nowell 1913; Jones 1915; Barrow 1924; Wolcott 1933b, 1934, 1936; and Beavers and Selhime 1975). However, to facilitate studies related to the potential control of this insect, more detailed biological information was needed, including immature development and adult reproduction potential and longevity. An artificial diet and rearing procedures were therefore developed to facilitate the necessary laboratory studies.

REARING WITH ARTIFICIAL DIET

Initially several artificial insect diets were investigated, including diets for the boll weevil, *Anthonomus grandis* Boheman (Vanderzant and Davich 1961); the plum curculio, *Conotrachelus nenuphar* (Herbst) (Yonce et al. 1971); and *Dectes texanus* Le Conte (Hatchett et al. 1973). However, most were not accepted by the hatchling larvae. Therefore, a formulation was developed which provided the necessary physical characteristics acceptable to the hatchling larvae, and which could be easily prepared and stored for several weeks without spoilage. Ingredients of this diet were alphacel 307 g, cotton seed meal 250 g, soybean protein 104 g, sucrose 70 g, casein 70 g, wheat germ 60 g, cornstarch 44 g, Vanderzant's vitamin mix 31 g, Wesson® salts 16 g, methyl paraben 15 g, ascorbic acid 6 g, sorbic acid 5 g, cholesterol 3 g, choline chloride 2 g, agar 75 g, formalin (38.5%) 6 ml, and water 2550 ml. Batches of diet were prepared in amounts up to 38 liters in a gas-fired mixer by first weighing out the desired ingredients and placing them in the required volume of tap water while the agitators were revolving. Thus, the ingredients were thoroughly mixed before heat was applied. The medium was then heated to ca. 90-95° C during a period of 15-20 min. Finished diet was poured (ca. 18 ml) into 35-ml clear, plastic cups and allowed to solidify at

ambient temp. The open cups of diet were held 2-3 days until excess moisture had evaporated before introducing the hatchling larvae.

Hatchling larvae were obtained from egg masses of field-collected adult weevils. The procedure was as follows: the adult weevils were held in 60 x 60 x 74-cm screen cages and provided with citrus foliage for food and with folded kitchen wax-paper strips suspended from the top of the cage as oviposition sites (Wolcott 1933a). These strips with the egg masses were removed daily, attached to 12 x 25-cm paper cards, and placed in a black 6 x 10 x 18-cm wooden frame with spacers that held the cards 2.5 cm apart. This unit was positioned over a 6 x 30 x 53-cm plastic collection tray that had a barrier glued around the inside top edge, forming a 90° angle with the sides of the tray. This barrier prevented the larvae that dropped into the tray from escaping. Predetermined numbers of the larvae were subsequently transferred to the plastic cups of diet, and foil-lined paper lids were heat sealed onto the cups to prevent escape of the very active larvae. After 60 days, the larvae were transferred to fresh diet, and fresh diet was supplied at ca. 45-day intervals until the insects pupated. The infested cups were held throughout in a dark room at ca. 25 C and 60-70% RH. Since ca. 18 ml of prepared diet/cup was used, a weevil was provided with ca. 150 ml of diet during the average 1-year life cycle.

Biological Studies and Results

Egg Incubation and Neonate Larval Longevity.—Wolcott (1936) indicated that *D. abbreviatus* eggs hatched 7 days after oviposition and that the larvae could survive a week or longer without food. To confirm this, we collected 10 egg masses 24 h after adult females were put in a cage, placed the masses in individual vials, held the vials at 25° C and 80 ± 5% RH, and observed the eggs daily. Eclosion began on the 7th day, and within 24 h 90.0% of the 1,061 eggs contained in the 10 egg masses had hatched. However, eclosion in one egg mass did not start until the 10th day when 78.4% hatched. By the 10th day, 91.3% of total eclosion had occurred. In all, 93.6% of the eggs hatched. A single larva hatched on the 15th day, the longest incubation period. Thus, most eclosion occurs 7-8 days after oviposition.

After eclosion the larvae continued to be held in the vials to determine the length of survival without food. All vials contained live larvae 25 days after eclosion began, and after 31 days, 80% contained live larvae. After 53 days, 40% of the vials contained live larvae. All were dead at 60 days.

Larval, Pupal and Adult Development.—The developmental stages of *D. abbreviatus* are shown in Fig. 1. Hatching larvae (0.1 mg/larva) were placed onto diet at the density of 1, 5, 10, or 25 larvae cup. Larvae were introduced into 50 cups at each density. Most larval growth occurred during the first 3-4 mo, when a maximum weight of ca. 0.55 g was reached. Wolcott (1933b) reported the occurrence of up to 16 instars in *D. abbreviatus* larvae, but he considered the 8th instar equivalent to full growth because maximum weight was attained at that time. Later, Beavers and Selhime (1975) recovered larvae on a weekly basis from potted citrus seedlings, but because of individual variation in growth, were unable to determine the number of instars from the frequency distribution curve of head capsule measurements. Under the test conditions, a few larvae survived for 29 mo, although in other

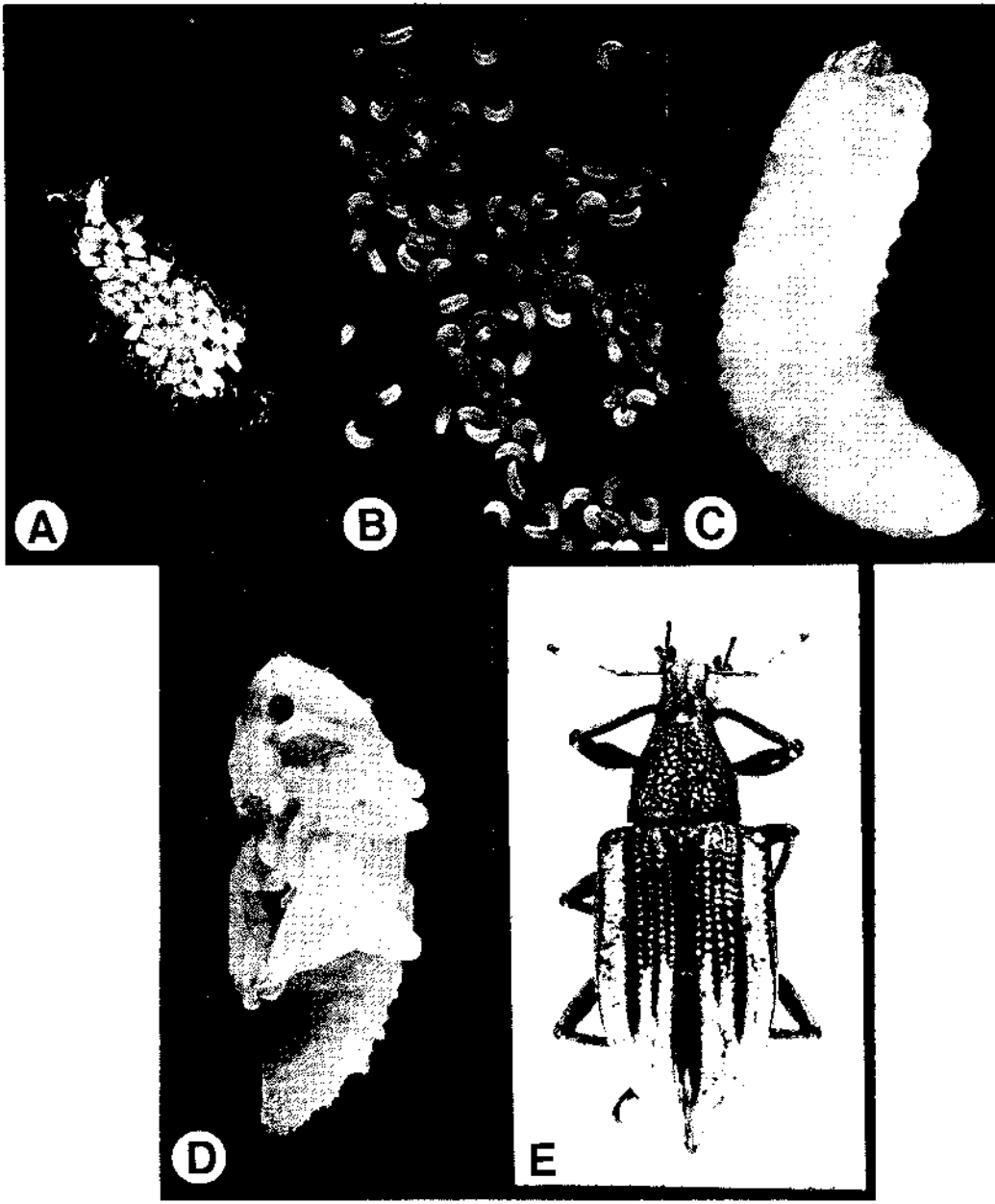

Fig. 1. Stages in the life cycle of *D. abbreviatus*. A, egg mass (5X); B, hatchling larvae (2X); C, 2-month-old larvae (6X); D, pupa (3.7X); and E, adult (3.5X).

tests larvae have required up to 4 yr to complete development. The larval period of *D. abbreviatus* entails a period of rapid growth for ca. 3-4 mo and then a period when the weight tends to decrease at pupation. Therefore, the most active and injurious period would be the first 3-4 mo after hatch, when growth is most rapid.

The percentage larvae, pupae, and adults recovered when cups were infested with 1, 5, 10 or 25 larvae/cup are indicated in Table 1. Mortality was greatest during the first 60 days after hatch, and increased when cups contained more than 1 larva, perhaps because of crowding or injury to each other. However, the greatest number of adults were recovered from the diet infested with 10 larvae/cup. Also, the exarate pupae proved to be extremely fragile and highly susceptible to injury when disturbed, and this contributed significantly to the overall mortality of the immature stages. The diet maintained good quality and texture with all densities of larvae infested.

Although the maximum yield of adults from the artificial diet appears low relative to production of other insects with shorter developmental periods, it was greater than has been found on plant hosts. When Beavers and Selhime (1975) infested 1200 citrus trees in 12-cm-diam pots with a total of 16,000 larvae, they obtained only 90 adults after 1-1/2 yrs, (0.5% yield). Also, when Hamlen et al. (1979) tested chemicals for control of the weevil on potted citrus, only 12-18% of the larvae in the controls were recovered 8-10 weeks after infestation.

Male and Female Development Time on Artificial Diet.—The first 500 adults reared on the artificial diet were used to determine the developmental periods for *D. abbreviatus* males and females.

The mean developmental periods (± SE) for larvae and pupae, respectively, were 377 ± 4.8 days and 15.2 ± 0.43 days for females and 378 ± 6.4 days and 15.4 ± 0.77 days for males. Adult females emerged after a mean of 381 ± 47 days; adult males required 382 ± 6.4 days. Thus, development of *D. abbreviatus* on artificial diet compared favorably with development on plant hosts (Ballow 1912; Jones 1915; Wolcott 1936; Beavers and Selhime 1975). The ratio of females to males for adults from the diet was 61.8:38.2; in Florida in 1972 and 1973, field-collected adults had ratios of 55.6:44.4 and 59.1:40.9, respectively (Beavers and Selhime 1976).

Reproductive Potential and Adult Longevity.—Twelve pairs of adults emerging from artificial diet were individually coded with enamel paint

TABLE 1. PERCENTAGE OF *Diaprepes abbreviatus* LARVAE, PUPAE, AND ADULTS RECOVERED FROM ARTIFICIAL DIET INFESTED WITH THE INDICATED DENSITY OF HATCHLING LARVAE (50 CUPS/TREATMENT).

No. larvae/ cup	% larvae recovered after 60 days[1]	% pupae obtained[1]	% adults obtained[1]
1	30.0 (15.0)	10.0 (5.0)	6.0 (3.0)
5	12.8 (32.0)	5.2 (13.0)	2.8 (7.0)
10	10.6 (53.0)	5.8 (29.0)	3.8 (19.0)
25	2.5 (31.0)	1.7 (21.0)	1.1 (14.0)

[1]Numbers in parentheses indicate actual numbers of insects obtained.

(Cross and Mitchell 1964), and placed in 1-liter cages with citrus foliage as the food source. Wax-paper strips were placed in each cage as oviposition sites, but some egg masses were deposited in the foliage. Dead adults were recorded and removed, and fresh foliage was provided 3 times a week. Adults were observed and egg masses were removed daily to determine the reproductive potential and adult longevity. The preoviposition period of females reared on the artificial diet was 21.8 ± 3.2 days; the mean number of eggs per mass was 69.2 ± 7.8; the mean number of eggs deposited per female was 6517 ± 931.1; mean hatch was 89.4% ± 27.2; mean female longevity was 147 ± 17.1 days; and mean male longevity was 135 ± 21.5 days.

Wolcott (1936) determined that the reproductive potential of 12 females obtained from larvae reared on plant material ranged from 3000 to 7000 eggs with a mean of ca. 5000. Thus the 3 to 7-day preoviposition period reported by Wolcott (1936) vs. the 21.8 ± 3.2-day mean we obtained indicates that females are probably more mature when they emerge from the pupal cell in the soil.

Although virgin females did oviposit, none of these eggs hatched. For example, 10 unmated females, maintained as previously described, produced a mean of 31.8 ± 7.7 egg masses and a mean of 1910 ± 349.3 eggs/♀. However, the mean number of eggs per mass was about the same for virgin and mated females (64.1 ± 10.2 vs. 69.2 ± 7.8).

Discussion

The results of these tests confirm that *D. abbreviatus* has a mean developmental period of ca. 1 year. The long subterranean period of the immature stages combined with the ability of hatchling larvae to survive under optimum conditions for long periods indicates the potential for movement of this pest to uninfested areas with plant hosts shipped in containers of soil. The adult weevils are present in Florida from May through November with the peak population period occurring from July through October (Beavers and Selhime 1976). This indicates the economic problem presented in developing control and containment strategies using conventional or presently approved techniques. The long developmental period of *D. abbreviatus* precludes mass production, although with adequate planning, adults can be produced in limited numbers for research purposes. This diet has been beneficial in the production of adult weevils for biological studies, and large numbers of 4- to 6-mo-old larvae for chemical and biological control studies.

Endnote

Mention of a trademark, or proprietary product does not constitute a guarantee or warranty of the product by the USDA and does not imply its approval to the exclusion of other products that may also be suitable.

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