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STORAGE AND BEHAVIOR OF PLANT AND DIET-FED ADULT CEREAL LEAF BEETLE, *OULEMA MELANOPUS* (COLEOPTERA: CHRYSOMELIDAE)[1]

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The univoltine life cycle of the cereal leaf beetle *Oulema melanopus* (L.) in Michigan (Castro et al. 1965) is similar to that reported by Venturi (1942) in Europe. Adults emerge from pupal cells in the soil in mid-June to early July, feed voraciously for about three weeks, and enter aestivation sites. For the remainder of the summer and early autumn only a few adults can be found feeding on late-maturing native grasses. The beetles overwinter and usually emerge in late March to early April and resume feeding. Mating and oviposition occur, and larval development is usually completed by late June in southern Michigan.

Techniques for rearing the cereal leaf beetle on greenhouse-grown small grain seedlings have been developed by Connin, et al. (1968). Maintaining these cultures requires collecting field adults, growing host material, and handling the cultures to insure that all stages will be available for study.

In Michigan during July adults can be collected more economically and in greater numbers in the field than by rearing in the laboratory. A summary of collection techniques, laboratory feeding and storage conditions for large numbers of field-collected cereal leaf beetles is presented in this paper. In addition, the mortality during storage of newly emerged field collected beetles fed either barley seedlings or an artificial diet is compared.

MATERIALS AND METHODS

Collection and prestorage conditions. Cereal leaf beetles emerge from pupal cells in Michigan by late June or early July. A large number of adults can be collected from fields where larvae are abundant with a sweep net shortly after emergence, since the beetles may remain in the field where they emerged if the crop is succulent. More often they move to more attractive grasses or fields of oats, wheat or corn in adjacent areas.

Immediately after emergence the adults are photopositive and move upwards on their host. Since the beetles often drop from the plant to the ground as a net moves toward them or jars the plant, repeated collections in the same area at 15-minute intervals will yield additional specimens. As many as 500,000 beetles have been collected in early July in southwestern Michigan using 15-inch nets during 32 man-hours of sweeping.

Beetles were usually swept from oats or wheat, confined in numbers of approximately 5000 per 16x16x16 inch screened cage, and transported to the laboratory for further feeding before storage. Since cereal leaf beetles occupy an ecological niche on small grain leaves which was not fully utilized by other American insects, collections are relatively pure, with only a few species of other insects and spiders present.

Laboratory feeding prior to storage. During transit to the laboratory, beetles were provided with corn or potted barley seedlings. In the laboratory, 4 pots of greenhouse grown barley seedlings at least 3 inches tall were provided for each cage twice daily during the first 2 weeks of feeding. In 1966 and 1967, beetles were fed under natural daylengths in a greenhouse with daily temperatures from 22 to 32°C and relative humidities from 40 to 95%. The amount of food required was dependent on the age of the beetles at the time they were collected, because some of them may have fed for a week or more by that time.

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RESULTS AND DISCUSSION

Behavior of cereal leaf beetles prior to and during storage. The feeding period averages about 3 weeks, with the insects' behavior changing during this period. After feeding, they become photonegative and hide under the plastic pots of grain or in folded-paper towels tacked to the upper cage corners. Feeding beetles have their antennae spread at an angle of approximately 65° or more, but those in hiding usually have an angle of about 10° between their antennae (Fig. 1). Beetles may maintain the latter antennal position throughout their storage period in the laboratory at 2-7°C. If the beetles are held at 26.7°C after storage and not given food, they become active for a day or 2 and become inactive. They may remain in the same location with their antennae held forward in the nearly parallel position for a month or longer or until food becomes available. In addition, inactive beetles' legs are usually positioned near their body. Beetles removed from storage before diapause development is completed may remain inactive even if host plants are available; inactivity is often observed in lantern globes when population density is low, or when they enter a dark confining area. In the field, cereal leaf beetles seek overwintering quarters in straw, under bark (Venturi, 1942; Castro *et al.*, 1968) and in the soil (Hayward, J. T., personal communication). The advantage of the forward antennal position during overwintering is unknown. Adult asparagus beetles, *Crioceris asparagi* (L.), also hold their antennae in a similar parallel position when they are inactive or overwintering.

Storage, 1966 and 1967. Prior to storage about 300 cereal leaf beetles and 4 paper towels saturated with a ½% sodium hypochlorite solution were placed in each of a number of plastic pint boxes. The boxes were sealed with a tight-fitting cover and placed in a dark refrigerator at 3.3°C. Table 1 shows the mortality of beetles field-collected and laboratory-fed before storage. In these studies morality was determined when the boxes of beetles were used on each date. After 112 days of storage all dead beetles were removed from the remaining boxes, and new paper towels saturated in ½% sodium hypochlorite were added. The average survival time for the beetles was about 160 days with the maximum survival period being about 336 days. The percentage of cereal leaf beetles overwintering in the field is unknown, although by April 15 most of the beetles are probably active in the field. This date is 268 days after the date insects were placed in storage, and (Table 1) approximately 25% of the stored beetles were alive at this time.

Cereal leaf beetle adults do not become active until late March or early April; hence storage under these conditions insures that large numbers of beetles are available in mid-winter when they are difficult to collect in the field.

Storage, 1968. Cereal leaf beetles collected at Galien, Michigan in early July 1968 were placed in a 6x12x6 ft. screened cage in East Lansing. They were provided barley and corn seedlings during their feeding period. On August 2 and 4 they were brought to the laboratory and prepared for storage as previously described. 200 beetles were stored in each box with or without a cover and under 0, 8, or 16 hour daily photophases. They were maintained in bioclimatic cabinets programed at 5.5 ± 2°C and 10 ± 10% RH (DD 0:24) and 60 ± 10% RH (LD 8:16 and 16:8). The accumulative percent mortality for these beetles is presented in Table 2. Every 30 days the dead beetles in each carton were removed, and the papers were changed and sprayed with a ½% sodium hypochlorite solution. Beetles stored in boxes with open tops under lower relative humidity had a higher mortality than beetles stored in closed boxes. For example, 50% of the beetles in open cartons died in about 90 days while 50% died in closed cartons in 210 days. Perhaps the beetles lived longer in these experiments than the previous one, because some boxes may have lost all of their moisture or were more contaminated in the previous experiment. There did not appear to be any notable difference in storing beetles under the 8 hour rather than the 16 hour photophase. Beetles stored in the dark in closed cartons at 10% RH died earlier than beetles stored under comparable conditions at 60% RH. For example, after 150 days 98.0% of the beetles fed barley seedlings stored under the low humidity had died while only about 31% of the beetles in closed boxes were dead. The earlier mortality is assumed to be due to the loss of moisture from the closed plastic cartons under the lower relative humidity, as the paper towels were dry when they were changed every 30 days while the towels in boxes in the 60% RH cabinet remained damp.

Storage after being fed an artificial diet. Cereal leaf beetle adults collected at Galien, Michigan on 5 July were provided barley seedlings and small corn plants in a 6x6x12 ft screened cage outdoors at East Lansing, Mich. On 8 July they were transferred to bioclimatic cabinets programmed at 16 hours of light daily, 27.6°C, and 60 ± 10% RH, and held there throughout the remainder of their feeding period. They were confined in screen-topped lantern globes with a Petri dish filled with artificial diet as the base. About 150 adults were confined in each of 13 lantern globes, and provided with a diet change every 2nd or 3rd day from 8 July to 2 August.

The ingredients of the artificial diet are listed in Table 3. This is the boll weevil diet of Sterling *et al.* (1965), modified by replacing the cottonseed meal with wheat germ. In addition, chlorophyllin, gelatin, and formaldehyde were added, and the relative amounts of many other ingredients were changed. The diet was dispensed into Petri dishes, stored in a refrigerator in plastic bags until used, and used within two weeks after preparation.

The beetles on the diet surface in each globe were counted prior to changing the diet. In this way the relative numbers feeding on the diet was obtained. On 29 July corrugated cardboard strips were placed within each globe as possible aestivation sites. Throughout the feeding period there was a gradual decrease in the average number of beetles on the diet surface. Thus, the average number of beetles on diet in the 13 globes was 61 beetles on 10 July, the first time the diet was changed, and 13 beetles per globe on 2 August, the date they were placed in storage. During the diet feeding period, 14.5% of the beetles died. This figure does not represent the entire feeding period, as all of these beetles had fed previously in the field. Comparable data is not available on the mortality for plant-fed beetles in the field or laboratory for this feeding period.

The diet-fed beetles were stored in closed plastic boxes in the same manner mentioned previously. Mortality in each carton was checked every 30 days (Table 2).

Diet-fed beetles did not live as long on the average as those fed on plants and maintained under the same conditions. By the 90th day of storage 57.8% of the diet-fed beetles were dead while only 15% of the plant-fed beetles maintained in the same bioclimatic cabinet had died. By the 210th day of storage 52.9% of the plant-fed beetles had died while 91.1% of the diet-fed beetles were dead. There were beetles in both groups which lived nearly 330 days. Perhaps the few long-lived beetles in the diet-fed experiment had accumulated sufficient food reserves in the field prior to being placed on diet, enabling them to live as long as plant-fed beetles.

SUMMARY

On the basis of these experiments, it is apparent that the cereal leaf beetles can be stored for long periods in tightly closed plastic containers containing damp paper towels. Changing the paper towels monthly and spraying with ½% sodium hypochlorite reduces contamination, ensures adequate moisture and increases the length of survival. In addition, the duration of the survival can be used as an indirect method to determine the nutritional adequacy of an adult artificial diet.

LITERATURE CITED

Castro, T. R., R. F. Ruppel, and M. S. Gomulinski. 1965. Natural history of the cereal leaf beetle in Michigan. Mich. Agr. Expt. Sta. Quart. Bull. 47:623-653.

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Table 1. Per cent of mortality for 32,300 (1966) and 116,100 (1967) adult cereal leaf beetles stored at 3.3°C. in plastic boxes.

	PER CENT MORTALITY	
Days in storage	*1966*	*1967*
28	4	3
56	4	4
84	18	13
112	32	23
140	41	20
168	62	49
196	62	59
224	73	65
280	–	87
308	77	100
336	100	–
364	–	–

Table 2. Accumulative per cent mortality of adult cereal leaf beetles fed either plants or an artificial diet, and stored in open and closed plastic boxes at 5.5°C under 0, 8, or 16 hour daily photophases.

		ACCUMULATIVE PER CENT MORTALITY DAYS IN STORAGE											
CONDITIONS	Number of Beetles	30	60	90	120	150	180	210	240	270	300	300	360
Plant Fed Open boxes													
LD 8:16[1]	1206	15.1	32.3	62.2	92.0	96.9	99.4	99.9	100				
LD 16:8	1220	6.2	23.4	53.1	78.0	84.2	99.7	99.9	100				
Closed boxes													
DD 0:24	1210	2.5	8.0	58.8	79.3	98.0	99.9	100					
LD 8:16	990	5.9	11.7	16.5	22.9	30.7	35.4	48.4	71.0	77.7	92.9	98.4	100
LD 16:8	907	6.5	9.6	17.0	25.8	32.4	40.5	52.9	75.3	97.4	99.9	100	
Diet Fed Closed boxes													
LD 16.8	1901	4.8	21.5	57.8	67.1	73.7	86.6	91.1	96.6	99.4	99.9	100	

[1]LD 8:16 means 8 hours of light and 16 hours of dark daily.
DD 0.24 is continuous darkness. The relative humidities within the bioclimatic cabinets were 10 ± 10% (DD 24:0) and 60 ± 10% (LD 8:16 and 16:8).

Table 3. Ingredients in an adult cereal leaf beetle artificial diet.

DRY INGREDIENTS IN GRAMS:	
Chlorophllin Chlorophyll	0.1
Wheat Germ	19.6
Casein, Vitamin Free	22.8
D-Sucrose	32.8
Salt Mixture-W[1]	6.5
Brewer's Yeast	7.5
Alphacel	10.8
Egg Albumen	5.0
Cholesterol	1.5
Agar	5.0
Gelatin	3.5
LIQUID INGREDIENTS IN MILLILITERS:	
Water	430.0
Wesson Oil	5.6
Wheat Germ Oil	2.5
Inhibitor[2]	5.0
10% Potassium Hydroxide	10.0
10% Formaldehyde	5.0
Vitamin Solution[1]	8.0

[1] Salt mixture and Vanderzant vitamin mixture sold by Nutritional Biochemical. 26201 Miles Ave., Cleveland, Ohio.

[2] Inhibitor solution contained 100 ml 95% ethyl alcohol, 10 g. sorbic acid and 10 g methyl parahydroxybenzoate.

BRIEF NOTICES

AQUATIC DIPTERA (1934-7). O. A. Johannsen; Part V by L. C. Thomsen. Los Angeles: Entomological Reprint Specialists, 1969. [iv], 95, 74, 102, 98 pp. Hardbound, $11.25; paper (1970), $5.00.

The reprinting of this still very useful work covering the North American genera of aquatic Diptera, with its copious and excellent illustrations, is indeed welcome. The reprint is every bit as good as the original, with original pagination maintained, the paper perhaps a little better than the original, and in a very good buckram binding. It is unfortunate that reprints cost so much, but then, the original edition does not even appear in recent antiquarian catalogs.

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NEARCTIC WALSHIDAE: NOTES AND NEW TAXA (LEPIDOPTERA: GELECHIOIDEA). Ronald W. Hodges. Smithsonian Contributions to Zoology, 18. Washington: Smithsonian Institution Press, 1969. [ii], 30 pp. $0.40 (from the Superintendent of Documents, Washington, D.C. 20402).

Former Michigan microlepidopterist Hodges furnishes new information regarding distribution and food plants of the nearctic Walshiidae. 14 new species and one genus are described; there are keys and a list of known species. Illustrations of some adults and genitalia are included.

R. S. Wilkinson