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ELECTRONIC RECORDINGS OF FEEDING MOTIONS AND RHYTHMS OF CORN EARWORM *(HELIOTHIS ZEA)* LARVAE

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ABSTRACT

Masticatory motions and feeding rhythms of corn earworm larvae *(Heliothis zea* (Boddie)) were monitored by means of automatic recordings obtained with a displacement sensor in contact with the feeding medium, Larvae fed on soybean leaves at a rate of 1.8 bites/sec., whereas larvae inside green bean pods fed at a rate of 1.5 bites/sec. Differences in masticatory rates were attributed to differences in moisture content of the two diets. Duration of meals usually Increased, and intervals between meals decreased in the afternoon hours; although not clearly defined, there seems to be a daily rhythmic pattern in the feeding activities of the com earworm larva. Feeding gradually increased after a molt to reach a peak prior to the following molt. The possible uses of feeding recordings in studies of host plant selection and feeding behavior are discussed.

INTRODUCTION

Contact chemoreceptors in the mouthparts of phytophagous insects mediate to a large extent the early stages of host-plant selection generally defined as host-plant recognition and acceptance (Dethier 1970). The probing behavior of insects during this phase has been used to bioassay and define specific biting stimuli (Hamamura 1970). As Hamamura observed, an accurate bioassay for biting stimuli should distinguish between biting and swallowing activities. Measurements of weight gain or fecal pellet counts indicate the amount of food ingested but are not suitable for the analysis of pure biting responses.

Detailed studies of the probing behavior of insects with piercing-sucking mouthparts were made possible by the development of techniques for the electronic recording of biting by aphids, mosquitoes, and hematophagous bugs (Kashin and Wakeley 1965, Kashin and Arneson 1969, McLean and Kinsey 1969, McLean and Weigt 1968).

Similar techniques adapted to insects with chewing mouth parts met with only partial success, possibly due to the inherent mobility of these insects (Kogan and Goeden 1971; Kogan, unpublished data, recording biting activities of the Mexican bean bettle, *Epilachna varivestis,* using an adaptation of McLean’s technique).

A different approach was attempted based on the detection of vibrations of the feeding medium caused by the chewing activities of the insect. A displacement sensor in contact with the medium transduced the feeding motions into an electrical signal that was amplified and displayed on a chart or on an oscilloscope screen. This technique was used to characterize masticatory motions of 5 species of leaf-feeding insects (Kogan 1972). I report here an adaptation of the same equipment to record the feeding motions and apparent feeding rhythms of 4th and 5th-stage larvae of the corn earworm, *Heliothis zea* (Boddie), feeding on soybean, *Glycine max*, leaves and inside green bean, *Phaseolus vulgaris*, pods. The application of this technique in host-selection and feeding-behavior studies is discussed.

MATERIALS AND METHODS

Recording system

The masticatory motions of the corn earworm were analyzed using a Model DB "Polysonic Detector and Bionic Sensor" (C.W. Dickey Assoc., State College, Pa., U.S.A.). The sensor responds in the range of 2 to 5,000 Hz, and the amplifier is provided with filters for two ranges of frequencies – the low pass position passes signals between 2 and 1000 Hz, and the band pass position passes signals above 200 Hz. In all recordings the band pass position was used and it was adequate to filter out most low-frequency environmental noises.

Recordings of leaf-feeding motions (Fig. 1A) were obtained with a leaf blade stretched over a rigid Plexiglas support (Fig. 2) in which the leaf acted upon by the insect behaved like a vibratory membrane. The tip of the sensor, bent to a 135° angle upwards, touched the center of the under surface of the leaf blade. Vibrations of the leaf caused by masticatory or locomotory motions of the insect produced signals that were detected by the sensor, were amplified, and were directly displayed on an oscilloscope screen or recorded on a chart or a magnetic tape for later playback, display, and analysis.

To record larval feeding inside green bean pods, an incision, ca. 1 cm^2, was made on one lateral wall of the pod. Enough of the endocarp was removed with a number 1 cork borer to permit introduction of a caterpillar through the lateral incision. The square piece of pod wall material was returned to its original position and held in place with a strip of transparent tape. The caterpillar was thus confined inside the pod, which offered an adequate fit for its body. The pod was suspended from the top of a plastic box and the probe of the sensor, passing through a hole in the wall of the box, was inserted about 1 cm deep into the tip of the pod (Fig. 1B). In this case the vibrations were transmitted through the whole pod to the sensor, and the signals obtained were recorded as above.

Recordings were made at a chart speed of 25.0 cm/min (Dohrmann recorder) or 24.0 cm/min (Coleman 105 recorder) to study rates of biting and biting mechanisms. Feeding rhythms were recorded at chart speeds of 25.0 cm/hr. Recordings of rhythms were also obtained with the use of a Chauvin-Arnoux "Detecta", an attachment applied to the front panel of the microammeter of the amplifier. This attachment uses a photocell to sense the passage of the pointer above a set reading. When triggered, the

"Detecta" gives a signal of uniform amplitude which registers as bipolar peaks on the chart. This attachment makes it possible to screen out, over extended periods of time, interfering sub-threshold signals resulting from environmental noise. Eliminating environmental noise by this means made the recordings easier to measure and interpret. Fig. 3 shows a double recording (obtained with a dual channel Dohrmann Model DC recorder) with the lower tracing registering the direct output of the amplifier and the upper tracing registering the output of the "Detecta" attachment. The correspondence of the "Detecta" records to certain amplitude "amplifier" records is quite obvious from this illustration.

RESULTS AND DISCUSSION

Masticatory motions

High-speed recordings show individual biting motions. Fig. 4 illustrates typical recordings obtained with a 5th-stage larva feeding on a soybean leaf of the variety Wayne (Fig. 4A) and a larva of the same stage feeding inside a green bean pod (Fig. 4B). As was observed previously (Kogan 1973), the corn earworm usually bites continuously during each meal. The biting rate is apparently higher when the larva is feeding on soybean leaves (ca. 1.8 bites/sec) than when it feeds inside a green bean pod (ca. 1.5 bites/sec). This observation, however, was based on rates of feeding of different animals, and the number of replications was not sufficient to eliminate the influence of individual variability among larvae.

In both cases larvae were removed from a general culture maintained on a wheat-germ medium. Those larvae implanted into a pod started feeding immediately or after a short (10-15 min.) period of adaptation to the new food and environment. A lag period of one to several hours occurred when larvae were placed on soybean leaves. As older corn earworm larvae are essentially feeders on fruiting structures (fruit borers), soybean foliage was accepted only reluctantly, although the larvae could complete development on a pure soybean leaf diet (Kogan unpublished data).

Duration of meals

Low-speed recordings of larvae feeding inside a green bean pod (Fig. 5) indicate that, following a molt, meals are of short duration (3-15 min) with somewhat long intervals of rest between successive meals (10-20 min). Meals become increasingly long towards the mid-point of a larval stage (here the 5th-stage); they reach maximum durations of 45-50 min, sharply falling thereafter 8-12 hours prior to prepupation.

Feeding rhythms

Fig. 6 is a summary of recordings of one larva feeding in a green bean pod; the number of minutes of active feeding is plotted against time (in hours). Recordings of the feeding activity of a 2-day old, 4th-stage larva began at 17:00 hours (day 1). On day 2 the larva molted. In general, the larva's feeding activity, measured as the sum of meal duration (in minutes) per hour, was highest between 11:00 hours and 18:00 hours. During the remaining 17 hours of each day, feeding activity oscillated at rather lower levels but never came to a prolonged halt except prior to the molt. The larva became increasingly active in successive days after molting until day 6 (4th day of 5-th stage) when activity was generally high during the 24-hour period. Feeding ceased at the beginning of day 7 shortly before prepupation started. From this recording there is reason to suspect that the feeding activity in the corn earworm larva follows a daily rhythmic pattern.

Possible uses of feeding activity recordings

The main purpose in the development of this technique has been to bioassay specific biting stimuli. Whether the feeding process of most phytophagous insects follows the scheme described by Hamamura for the silkworm still remains to be proved. This bioassay will permit the quantitation of biting as a clearly independent response from continuous feeding. In this case plant fractions are applied to a neutral substrate and recordings are obtained as described for fresh leaves.

The complexity of the interactions of insects with physical and chemical plant factors is apparent in the response of the corn earworm larvae feeding on soybean leaves and in green bean pods. The slower biting rate registered with the green bean may be related to its higher water content. The green bean pod had ca. 8% dry matter, whereas the soybean leaves had 16–18% dry matter. Without considering the balance of nutrients, just to achieve similar water intake the larva had to consume about twice as much leaf material as pod material. The accelerated biting rate may conceivably be a response to the drier diet. This response also poses the question of energy budget, i.e., how much energy taken in by the larva as food is wasted in the feeding process? The hydric balance and the energy budget may explain why the corn earworm is essentially a feeder on fruiting structures. These questions, however, are merely raised by this biting analysis. Complex experiments will have to be designed specifically to test these hypotheses.

Finally, the technique described opens a new way to approach the study of feeding rhythms in phytophagous insects. With feeding responses being used in many laboratories as a method to test plant resistance to insect attack, host-selection behavior, the effects of antifeedants, and other questions of fundamental and practical interest, it is necessary to define the periods of peak activity so that experiments can be performed when maximum response can be obtained in the minimum time, thus optimizing the conditions of the experiments. Knowledge of feeding rhythms may also have practical importance in scheduling and timing of control measures, mainly of chemical controls that require ingestion by the insect.

The technique described here is in its first steps of development. New advances in technology will certainly permit improvement of the sensitivity of the detector and the clarity of the recorded response. The applications of the method, however, seem to open certain opportunities for the elucidation of basic problems of host selection and feeding behavior of phytophagous insects.

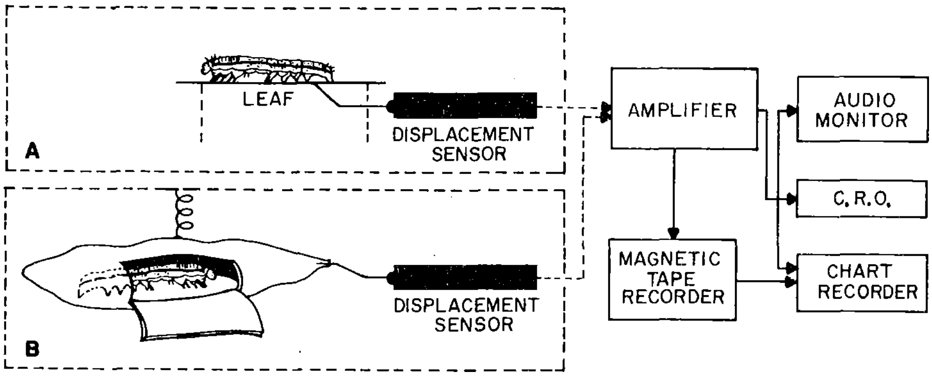

Fig. 1 Schematic of the components used in the recording of feeding activities of phytophagous insects with chewing mouthparts. A. system used with leaf feeders; B. system used with fruit borers.

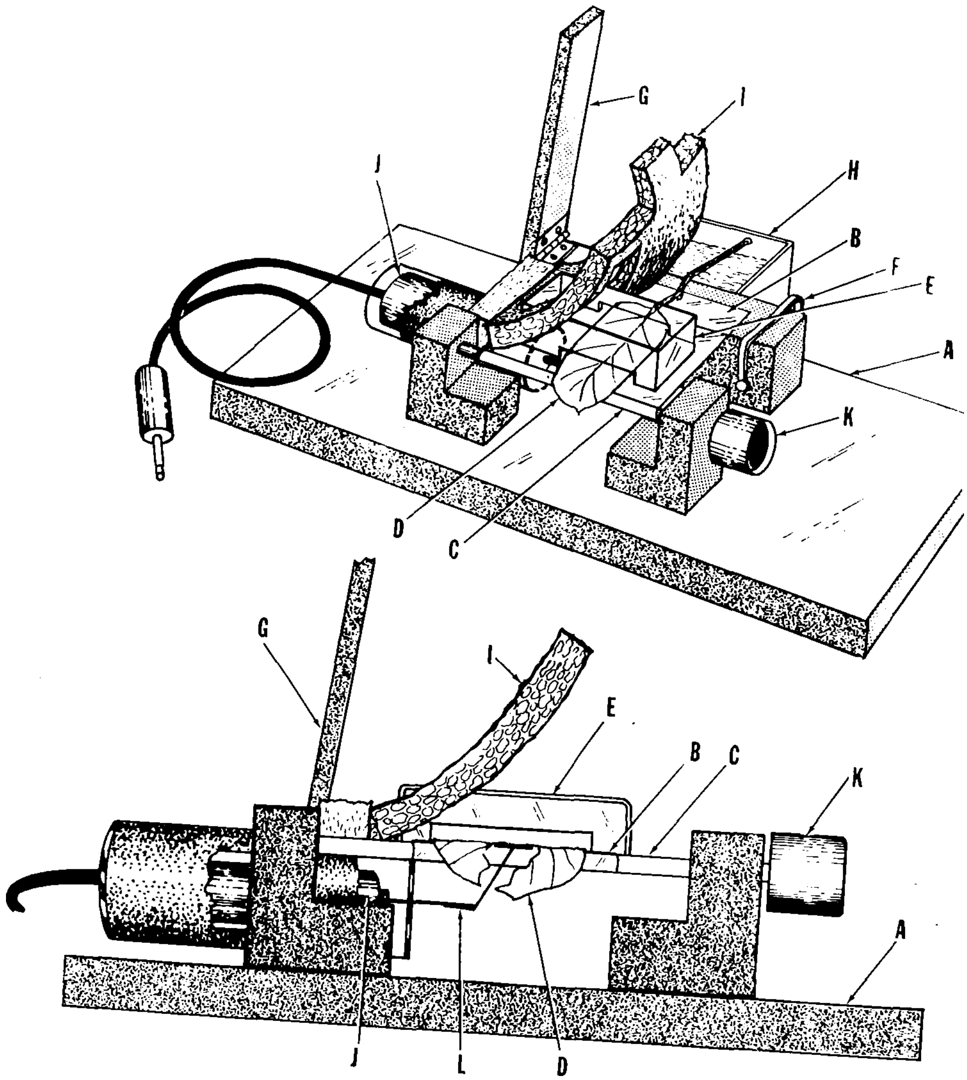

Fig. 2. Perspective (top) and front view of leaf-holder and sensor. A. Plexiglas base; B. Mylar membrane; C. Plastic rod to which Mylar membrane (B) is attached; tension of the membrane is adjusted by turning rod counter-clockwise by means of knob (K); D. Test leaf strip with petiole extending to moist reservoir (H); E. Half box enclosure of test insect; F. Rubber band used to fasten the hinged plate (G) when lowered to hold the base of the leaf strip; G. Hinged plate in open position; H. Moist reservoir partially filled with wet cellulose sponge; I. Cellulose sponge pad that extends from moist reservoir and around the animal enclosure half box (H) to maintain leaf turgidity; J. Bionic Sensor enclosed in foam-padded plastic cylinder; K. Knob that permits control of tension rod (C). L. Sensor probe with bent insect pin attached and touching underside of leaf strip. (A bulldog clip is attached to tip of leaf blade to add tension – the clip is not shown in the picture).

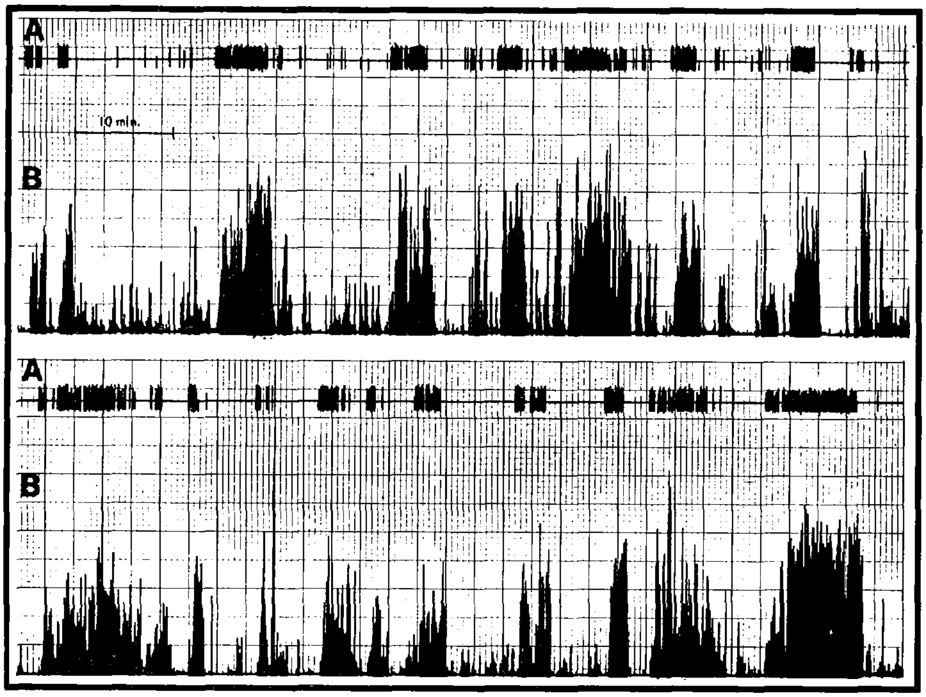

Fig. 3. Double recording using a dual channel Dohrmann recorder showing the correspondence of the Detecta (A) and the direct amplifier recordings (B). Corn earworm larva, 9 d. old, 5th stage, feeding inside green bean pod. Chart speed 25 cm/hr; full scale sensitivity-channel 1 (Detecta) 100 mv; channel 2 (amplifier) 500 mv. Lower pair of recordings is the continuation of the upper pair of recordings.

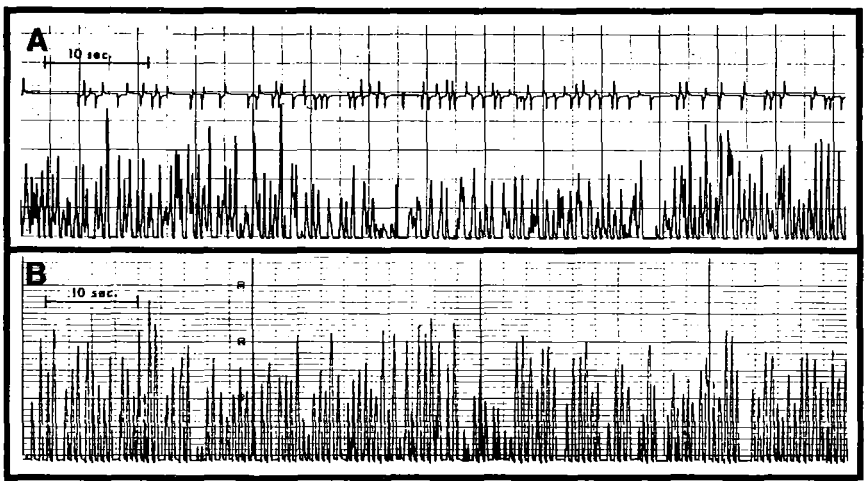

Fig. 4. Recordings of feeding motions of a corn earworm on a soybean leaf (A) and in a green bean pod (B). Chart speed of Dohrmann recorder (A) 25 cm/min; chart speed of Coleman recorder (B) 24 cu/min. Both at full scale sensitivity of 500 mv.

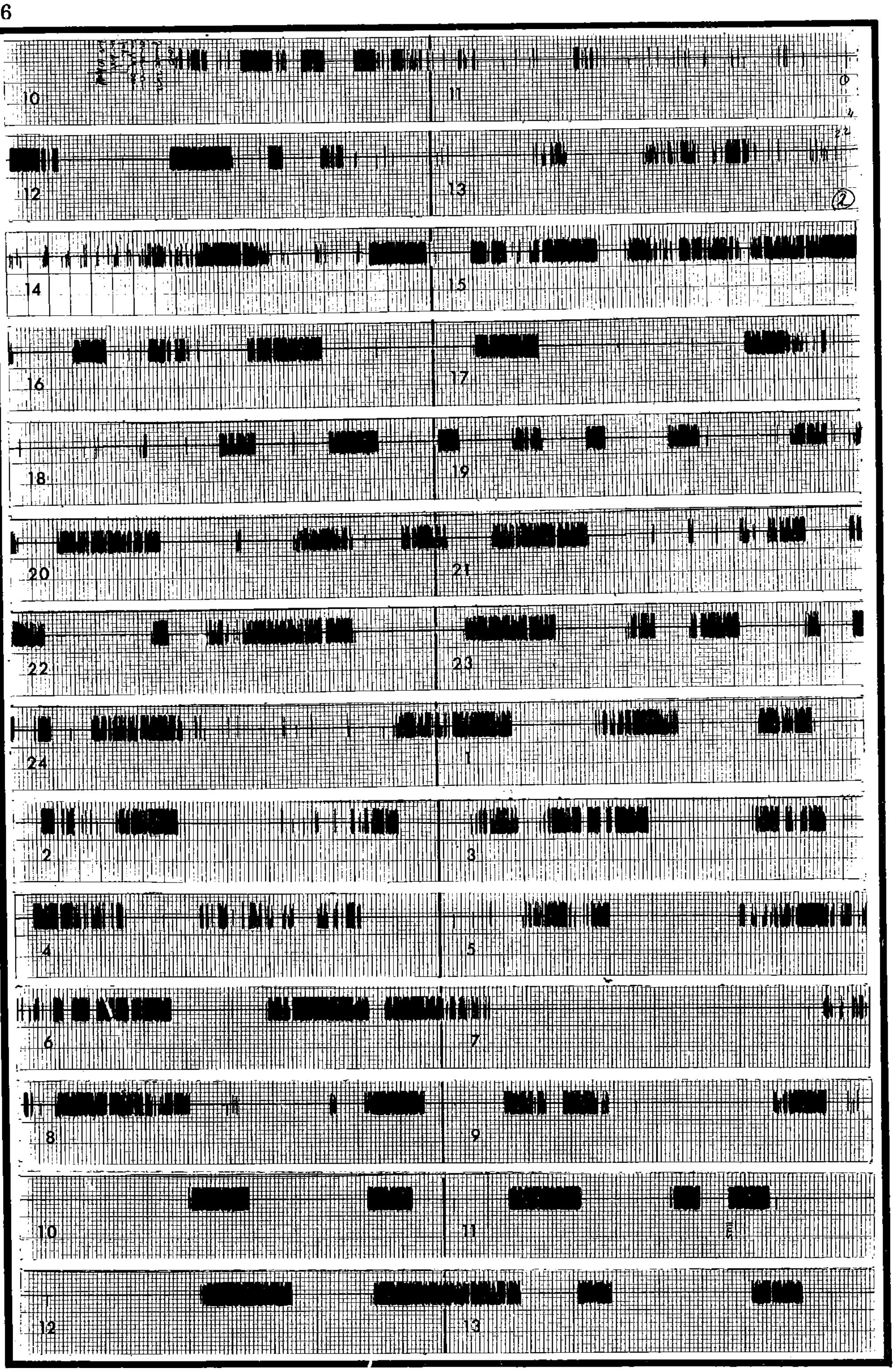

Fig. 5. Slow speed (25 cm/hr) recording of the output of the Detecta attachment showing duration of meals and intervals of rest. Corn earworm larva, 9 d. old, 5th stage, feeding inside a green bean pod.

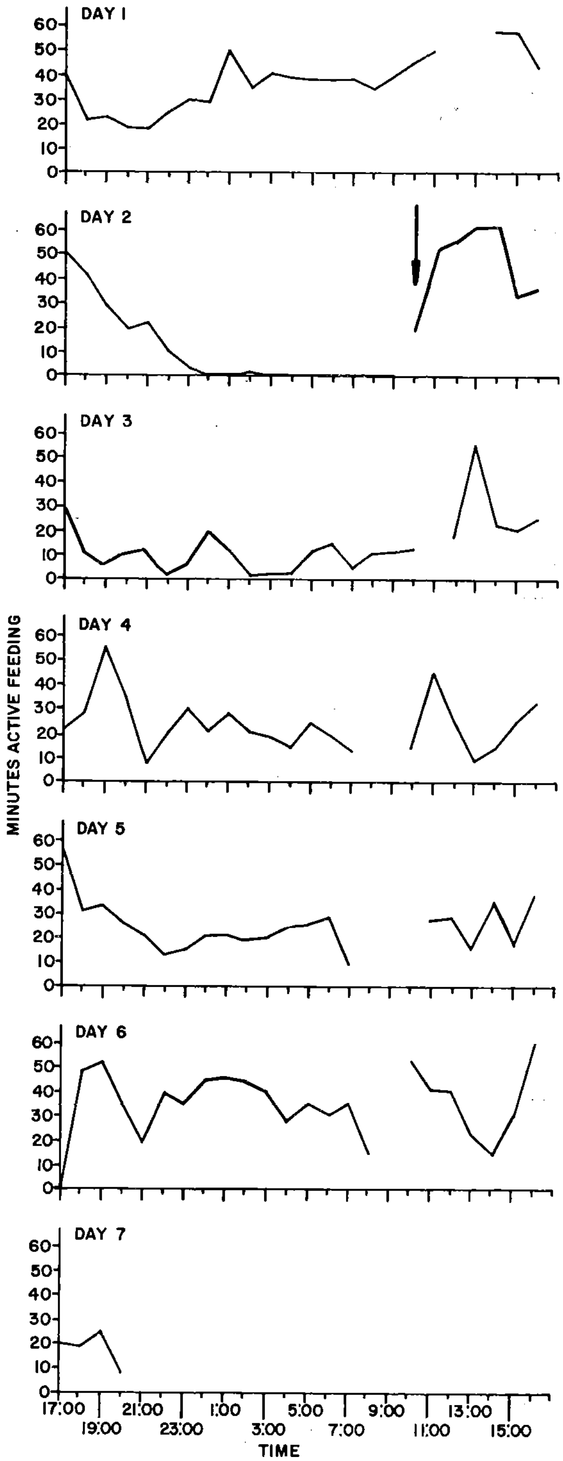

Fig. 6.

Summary of a 6 day recording sequence of a corn earworm larva feeding inside a green bean pod. Activity measured in minutes of active feeding per hour. Discontinuities in the graph correspond to interruptions necessary to transfer larva to new pod and recharge the battery of the amplifier. Vertical arrow at 10:00 a.m. of day 2 indicates end of molt (4th to 5th stage).

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