AN EVALUATION OF A PORTABLE GEIGER COUNTER TO TRACE RADIOLABELED *HELIOTHIS ZEA* (BODDIE) (LEPIDOPTERA: NOCTUIDAE) LARVAE IN CORN AND COTTON[1]

Robert R. Farrar, Jr. and Douglas A. Landis
Department of Entomology
North Carolina State University
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ABSTRACT

A portable Geiger counter was evaluated for tracing the movements of radiolabeled *Heliothis zea* (Boddie) larvae in cotton and corn. In cotton, larvae used were labeled by injection of the parent females and phosphorus-32. These could easily be located using the Geiger counter. In corn, larvae were labeled topically so that pupation sites in the soil could be labeled. The percentage of these that could be located proved variable.

Key Words: Geiger counter, radiolabeling, *Heliothis,* larvae, cotton, corn.

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INTRODUCTION

In studies of the biology of insect pests, it is frequently necessary to release marked insects in the field and follow their development apart from native insects. There are various techniques for doing this, including fluorescent powders, paints and dyes, and radiolabeling. Radiolabeling has the advantage that it can be retained through molts and concealed insects can be located in the field with a portable Geiger-Muller counter or survey meter.

Geiger counters have been used to trace a variety of insects and other arthropods labeled with several different radioisotopes. Cobalt-60 has been employed with wireworms (Arnason et al. 1950) and the European pine shoot moth (Green et al. 1957). Iridium-192 tags were utilized with honeybees by Nelson and Baldwin (1977). Branagan (1971) made use of cerium-144 in labeling ticks. Geiger counters have been used to trace several insects labeled with phosphorus-32, including a coccinellid (Ali and Azam 1977), planthoppers (Lee et al. 1981), mosquitoes (Papierok et al. 1973), flies (Ryu et al. 1974), two species of armyworm larvae (Persson 1975), and larvae of a pyralid species (Cheshire et al. 1977).

In studies on the behavior of *Heliothis zea* (Boddie) in cotton (Farrar 1984) and corn (Landis 1984), methods of locating radiolabeled larvae using a Geiger counter were evaluated. Phosophorus-32 was chosen for the label because it is less likely to be lost through metabolism than ^{14}C. Its half-life of 14.2 days is long enough to permit following larvae through their developmental period but short enought that it will not persist in the environment. It is a much stronger beta emitter than ^{14}C. Additionally, it has been successfully used to label *Heliothis* larvae for predation studies (Hines et al. 1973; McDaniel et al. 1978). The purpose of this paper is to discuss the usefulness and limitations of a Geiger counter for tracing labeled *Heliothis* larvae in two different crops, cotton and corn.

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MATERIALS AND METHODS

In 1981 through 1983, near Clayton, North Carolina, *Heliothis zea* larvae feeding on cotton were monitored to study survival and movement (Farrar 1984). Cotton plants were infested with radiolabeled eggs and individual insects were monitored until they became large larvae. The method described by McDaniel et al. (1978) for labeling eggs of *H. virescens* by injection of adult females was used. *Heliothis zea* moths were either obtained from the stock culture maintained at North Carolina State University, or were collected from light traps when possible. Each moth was injected with 7μl of ^{32}P as orthophophoric acid in 0.02 N hydrochloric acid at a concentration of 5 mCi/ml for a dose of 35 uCi per moth. Eggs laid between 72 and 120 hr after injection were selected for use because they contained the most label. Eggs were obtained on paper using a cage similar to that described by Knott et al. (1966).

A preliminary test in the lab and greenhouse determined how well larvae retained radioactivity. Two Eberline model E-120 Geiger counters with speakers were evaluated initially, one with a side-window probe and a second with an end-window probe. The latter was selected as it was more sensitive and directional. Ten labeled eggs were placed on each of two cotton plants; 20 labeled eggs, on artificial diet. The resulting larvae, frass, and exuviae were tested periodically with the Geiger counter.

For field tests, the paper with labeled eggs was cut into pieces about 5 mm square, each with a single egg. Only eggs which had developed a brown ring, indicating fertility, were used. Each egg was also tested with the Geiger counter for adequate levels of label (at least 600 counts per minute). Each paper square was then attached to a desired location on a small cotton plant using a straight pin. Labeled larvae were located by systematically checking each feeding site (terminals, squares, flowers, fruit) with the Geiger counter. When surviving larvae were late fifth instars, they were collected and disposed of as radioactive waste. Over the course of three seasons, 2835 eggs were used. Farrar (1984) gives further details on these tests.

During 1982, field experiments were conducted near Clayton, NC, to study the movement of *Heliothis zea* larvae from corn to site of pupation in the soil (Landis 1984). Mature fifth instar larvae were obtained from the stock culture or obtained from the ears of corn in the field. Each insect was topically labeled with 2 uCi of the above solution of ^{32}P mixed with an equal volume of acetone. In preliminary tests, this dosage produced no noticeable side effects in the larvae or subsequent stages. The stock solution was applied to the dorsum of the insect, just behind the head region, using a Pipetman® micropipeting device.

Labeled insects were placed on host plants and monitored daily with the Geiger counter. When the insect left the release site, surrounding plants and soil were systematically searched with the Geiger counter to determine the new location of the insect. The exact site was located by holding the probe 1 cm from the soil surface and moving it until the area of highest activity was pinpointed. When an individual had entered the soil, the site of entry was recorded and the insect excavated. A 5 mm screen was used to sift the insect out of the soil. The insect and any contaminated soil was returned to the lab for disposal.

RESULTS AND DISCUSSION

The method of labeling larvae by injection of female moths provided good levels of radioactivity in the eggs and larvae. Readings for eggs ranged from 2400 to 15,000 counts per minute, with a mean of 8567 counts per minute. In the lab, labeled larvae reared on both cotton and artificial diet all registered well above background levels with the Geiger counter (at least 1500 counts per minute), through the last instar. Frass and exuviae did not give readings above background.

In the field, labeled larvae were readily detected with the Geiger counter at a distance of 2 to 3 cm. This distance is preferable to a longer one in this case as it allows one to pinpoint the location of a larva to the particular square or boll being tested. Larvae completely concealed within bolls or flowers were easily detected, as plant matter does not appreciably screen the beta particles emitted by the radioisotope. Labeled predacious insects and spiders were also sometimes found, so positive readings were checked visually to confirm the presence of a live larva. Generally, about 20 minutes were required to search a 1 m tall flowering cotton plant.

The percentage of pupation sites successfully located using the dermal labeling technique proved to be variable. Over nine releases of 10 to 20 larvae each, the highest recovery rate was 100%, the lowest, 13.3%. The mean recovery rate for the entire experiment was 40.5%.

The variation in the success of this technique was dependent upon the amount of ^{32}P deposited on the soil surface. Beta particles emitted by ^{32}P are blocked by soil; as little as 0.5 cm of soil over a labeled larva reduced emissions to virtually undetectable levels. Since *H. zea* larvae typically pupate at depths greater than 2.5 cm (Neunzig 1960), deposition of ^{32}P on the soil surface is necessary for detection of a site of pupation. During larval burrowing, a certain amount of the labeling solution was transferred to the excavated soil and remained on the surface. This could have been due either to abrasion, solution, or a combination of both. A labeled site could typically be located at a distance of 15 - 20 cm. Most labeled sites could be located in 1 to 4 minutes.

In some cases, burrow sites were not adequately labeled. Two factors were noted which could have contributed to insufficient site labeling. One was the water soluble nature of the orthophosphoric acid which allowed some portion of the label to be transferred onto the host plant before the larva left to enter the soil. This reduced the amount of label available to be transferred to the soil, particularly if the larva had been feeding in a moist whorl or ear. The moisture content of the upper 1 - 2 cm of soil also appeared to be important. When this soil was moist, more insects were recovered than when it was very dry. Moist soil conditions may have aided the transfer of the water-soluble orthophosphoric acid from the insect to the soil.

In conclusion, tracing radiolabeled larvae with a Geiger counter can be a useful technique for the study of the behavior of *Heliothis*. It can greatly improve efficiency in locating released insects. However, this technique is not without its limitations. Factors affecting its usefulness include the methods available for labeling the insects, the distance at which they must be detected, and whether they must be detected in soil. Thus, these and other factors should be considered before using a Geiger counter to study the behavior of labeled insects.

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