PARASITES PATHOGENS and PREDATORS of some Lepidopterous Pests in Alabama

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PARASITES, PATHOGENS and PREDATORS of some Lepidopterous Pests in Alabama

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SEVERAL LEPIDOPTEROUS pests are economically important in Alabama. Some of the more important ones are corn earworm, *Heliothis zea* (Boddie), also known as the bollworm and the tomato fruitworm; tobacco budworm, *H. virescens* (F.); cabbage looper, *Trichoplusia ni* (Hubner); and false looper, *Pseudoplusia includens* (Wlk.). Brazzel, et al., (*4*) noted that the bollworm has been recognized as a pest of cotton since 1820. By 1841 the bollworm had become prominent as an enemy of cotton and corn in the Southeastern United States *(10)*. Folsom *(6)* reported that in 1934 the tobacco budworm, long recognized as a tobacco pest *(9)*, occurred on cotton at Tallulah, Louisiana, in almost as great numbers as the bollworm. The cabbage looper is a serious pest of crucifers and many other cultivated crops. At least one other looper species, *P. includens*, attacks practically the same hosts as the cabbage looper, and is often found in mixed populations with *T. ni (5, 8)*.

Synthetic organic insecticides used during the late 1940's temporarily provided effective controls of these pests. However, the bollworm became difficult to control with DDT in certain areas of Louisiana during 1956, and Graves et al. *(7)* determined that the bollworm had developed resistance to some of the chlorinated hydrocarbon insecticides. The tobacco budworm has since developed resistance to the same class of insecticides *(3)*. Subsequent widening of geographic areas having resistant populations of bollworm and tobacco budworm and difficulty encountered in controlling looper populations have caused increased concern. These circumstances provided stimulus for more research in the area of biological control, especially since many natural enemies of this group of pests were already known.

This paper reports the parasites, pathogens, and predators found on bollworm, tobacco budworm, cabbage looper, and false looper in Alabama during 1965.

Methods and Materials

Parasites. Egg and larval parasites of *Heliothis* spp., *T. ni*, and *P. includens* were obtained by collecting eggs and larvae from the field and holding them in the laboratory until parasites emerged or until development was completed. Eggs were collected by removing the small section of plant tissue upon which the egg had been placed. Larvae were collected individually by hand or swept from the foliage with a 15-inch insect net. Eggs were held in half-ounce coffee creamers until parasite emergence or hatching occurred. Larvae were placed in individual, 1-ounce, plastic containers one-third to one-half full with artificial diets [modified from Berger *(2)* and Shorey *(11)*].

Predators. Presence of predaceous species known to attack *Heliothis* spp. was determined from whole-plant examinations and samples taken with sweep nets or D-Vac suction sampling machine.

Pathogens. Larvae, both dead and those suspected of being diseased, of *H. virescens, H. zea, P. includens,* and *T. ni* were collected from various crops. Host, date, and location as well as any disease symptoms were recorded for each larva collected.

Larvae were examined to determine if fungi, bacteria, or viruses were present. Where microbial growth was apparent, direct transfers to isolation media (potato dextrose agar, Sabouraud's dextrose agar, or egg-yolk agar) in petri dishes were made from diseased larvae. In the absence of conspicuous microbial growth, oral and anal openings of larvae were sealed with paraffin and larvae were routinely surface sterilized for 2 to 3 minutes in aqueous mercuric chloride (diluted 1:1000), followed by 3 rinses in sterile, demineralized water. Following the final rinse, larvae were bisected with a flamed scalpel and one-half of each larva was aseptically cut into smaller pieces, which were then plated on isolation media. The remaining half was aseptically macerated in sterile water and the suspension streaked on the isolation media. A portion of the larval suspension was retained for microscopic examination and bioassay to determine if viruses were present. Isolation plates were held at room temperature and at 30° C., and examined daily for microbial growth.

Pathogenicity Tests. Healthy, 5 to 7-day old larvae of *H. virescens, H. zea, P. includens,* or *T. ni* that had been reared in the lab were used for pathogenicity tests. Each larva was kept in a 1-ounce plastic carton containing an artificial diet. All larvae were held at 25° C. and 75 to 85 per cent relative humidity. Mortality counts were made daily for 15 days. Observed mortalities were corrected according to Abbott's formula *(1)*.

Fungal isolates were increased on agar media in petri dishes at room temperature. Heavily sporulating cultures were used as inoculum sources and inoculations were made by: (1) stroking a loopful of spores and mycelia on the dorsum of the integument, (2) spraying the larva with a suspension of spores in sterile, demineralized water containing 0.01 per cent Triton X-100, or (3) allowing a larva to crawl for 15 to 30 minutes over a fungal culture in an agar plate. Control larvae were handled similarly using a sterile loop, water alone, or fungus-free agar plates.

Numerous bacterial isolates obtained from diseased larvae were also tested for pathogenicity. Bacterial cells suspended in sterile saline (0.85% NaCl) were used as inoculum. Suspensions were prepared by adding saline to

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24-hour-old cultures on agar slants followed by a gentle stroking of the slant with a rubber-tipped glass rod. Approximately 0.1 ml. of suspension was pipetted on the diet in a carton, and inoculation was accomplished through larval ingestion of bacterial-contaminated diet. Control larvae ingested diet upon which only sterile saline was pipetted.

Several isolates of a virus, apparently of the nuclear polyhedrosis type, were tested for pathogenicity. The integument of each larva from which these isolates were obtained was dark colored and easily ruptured. The highly liquified contents of the larvae contained numerous particles morphologically characteristic of nuclear polyhedrosis. A suspension of the body contents from a diseased larva in sterile, demineralized water was used as inoculum. Inoculation was accomplished by pipetting 0.1 ml. of the inoculum onto the diet and allowing the larva to ingest the contaminated medium. Control larva ingested diet upon which 0.1 ml. of sterile, demineralized water was pipetted.

Results and Discussion

Parasites. *Heliothis* eggs collected from corn and tomatoes were heavily parasitized with *Trichogramma minutum* Riley, whereas those collected from cotton showed little parasitism. The combined results from both 1964 and 1965 collections, involving several hundred *Heliothis* eggs, showed 66.5, 72.7, and 5.2 per cent of the eggs from corn, tomatoes, and cotton, respectively, were parasitized.

Larval parasites encountered in *Heliothis* spp. included members of two families of Hymenoptera (Braconidae and Ichneumonidae) and one of Diptera (Tachinidae). These parasites and their *Heliothis* hosts were: *Microplitis croceipes* (Cress.) (both *H. zea* and *H. virescens*); *Cardiochiles nigriceps* Vierick (*H. virescens*), *Apanteles marginiventris* (Cress.) (*H. zea*), and *Meteorus autographae* Mues. (*Heliothis* spp., undetermined) of the family Braconidae; *Archytas marmoratus* (Tns.) (*H. zea* and *H. virescens*) and *Lespesia* sp. (*H. zea*) among the Diptera; and *Netelia* sp. (tentative identification) (*H. zea* and *H. virescens*) which belongs to the family Ichneumonidae.

The amount of larval parasitism in *Heliothis* spp. during the 1965 growing season was relatively low on most crops from which collections were made (Table 1). Parasitism was much greater on *H. virescens* collected from beggar's ticks, a weed of the genus *Desmodium*. The parasite involved was the Ichneumonid, *Netelia* sp.

A certain amount of larval parasitism may have been overlooked since many of the larvae died of diseases. This would tend to mask the effects of the parasite.

Two parasites appeared to be more important than all others on late season populations of *Heliothis* larvae. A large fly, *Archytas marmoratus*, was more prevalent on *H. zea* on grain sorghum in September and early October, and a large Ichneumonid wasp, probably *Netelia* sp., was found on *H. virescens* collected from beggar's ticks throughout the fall months. The last collection of live larvae was made December 3 following a period of sub-freezing temperature. This parasitism undoubtedly influenced the number of *Heliothis* that successfully entered hibernation.

Table 1. Fate of *Heliothis* Larvae Collected from Various Crops in 1965

Host	Larvae collected	Pupating	Parasitized	Diseased
	No.	*Pct.*	*Pct.*	*Pct.*
Cotton	550	65.2	4.0	39.8
Corn	607	58.2	0.8	40.9
Peanuts	150	31.3	1.3	68.0
Sorghum	168	56.5	4.8	36.0
Beggar's ticks	479	23.6	23.8	54.7

Fewer parasitic species were collected from the loopers; however, less emphasis was placed on this group. Parasites obtained from field-collected larvae included an egg/larval parasite, *Copidosoma truncatellum* (Dalm.) (family, Encyrtidae) and the tachinid fly, *Eucelatoria rubentis* (Coq.). Parasites also successful against the cabbage looper in the laboratory were the egg parasite, *Trichogramma minutum*, and the larval parasite, *Netelia* sp. These may occur on loopers in the field.

Predators. Although the predators observed or collected in this study were generally widespread and prevalent on many crops, those reported in this publication were primarily those found in cotton at some time during the growing season. These predators are listed in Table 2. Actually, several species of many of the groups listed are known to occur in cotton fields. Whitcomb and Bell (*12*) reported findings of approximately 600 species of predators representing 45 families of insects, 19 families of spiders, and 4 families of mites that were associated with cotton in Arkansas.

Table 2. Predaceous Arthropods Known to Prey on *Heliothis* spp. That Were Observed in Alabama Cotton Fields, 1965

Order	Family	Group or species
Odonata	Aeshnidae	Dragonflies
Neuroptera	Chrysopidae	Lacewings
Hemiptera	Anthocoridae	Flower bugs
Hemiptera	Phymatidae	Ambush bugs
Hemiptera	Reduviidae	Assassin bugs
Hemiptera	Nabidae	Damsel bugs
Hemiptera	Lygaeidae	Big-eyed bugs
Hemiptera	Pentatomidae	Stink bugs
Coleoptera	Carabidae	Ground beetles
Coleoptera	Cicindelidae	Tiger beetles
Coleoptera	Melyridae	Flower beetles
Coleoptera	Coccinellidae	Lady beetles
Diptera	Asilidae	Robber flies
Diptera	Syrphidae	Syrphid flies
Hymenoptera	Formicidae	Ants
Hymenoptera	Pompilidae	Spider wasps
Hymenoptera	Vespidae	Vespid wasps
Hymenoptera	Sphecidae	Sphecid wasps
Araneida	----------	Spiders

Pathogens. More than 500 diseased larvae were collected during 1965. *H. zea* larvae comprised almost 90 per cent of the total, with greatest numbers of this insect being found on corn. The microorganisms associated with diseased *H. zea* larvae are tabulated by crop in Table 3. *Aspergillus flavus*, *A. niger*, *Rhizopus* sp., *Fusidium* sp., *Penicillium* sp., and *Spicaria* sp. were some of the fungi commonly isolated from diseased larvae on most crops. Isolation frequency for bacteria was about as high as that for fungi from insects on each host. Numerous isolates were made from larvae suspected of having viruses, but only approximately 20 per cent contained recognizable polyhedral bodies.

The kinds and numbers of microorganisms isolated from diseased *H. virescens* larvae collected on various crops were: *A. flavus* 2 (cotton), 7 (rearing room); *Penicillium*

TABLE 3. MICROORGANISMS ASSOCIATED WITH DISEASED *Heliothis zea* COLLECTED FROM VARIOUS CROPS DURING 1965

Microorganism	Number isolates by crop						
	Corn	Cotton	Crimson clover	Peanuts	Sorghum	Tomatoes	None[1]
Alternaria sp.	0	0	0	1	0	0	0
Aspergillus flavus	28	17	22	7	0	1	3
A. niger	10	7	6	4	5	0	6
Cephalosporium sp.	2	0	0	1	1	0	0
Cladosporium sp.	0	0	0	1	0	0	4
Fusarium spp.	3	3	0	5	0	5	1
Fusidium sp.	17	1	0	1	0	0	1
Monilia sp.	1	0	0	0	0	0	0
Penicillium spp.	8	7	1	3	0	0	4
Rhizopus sp.	15	6	0	0	0	0	0
Spicaria sp.	1	6	0	11	2	0	0
Unknown (fungi)	6	2	0	5	4	1	2
Bacteria	127	38	0	12	4	2	27
Viruses[2]	107(19)	20(0)	0	2(0)	5(1)	0	32(16)
None	12	23	0	5	0	0	3

[1] Rearing room.
[2] Suspected virus; number of larvae containing polyhedra indicated in parentheses.

sp. 1 (crimson clover), 3 (cotton); *Rhizopus* sp. 3 (cotton); *Spicaria* sp. 1 (cotton); bacteria 4 (cotton); suspect virus 2 (cotton), no polyhedra found; and no microorganisms were isolated from 16 other larvae collected or cotton.

The fungus *Spicaria rileyi* was isolated from *Pseudoplusia includens* larvae collected on cotton, peanuts, and soybeans. A bacterium, *Streptococcus* sp., was isolated from diseased *P. includens* and another looper, *Rachiplusia ou* (Gn.), reared in the laboratory. One isolate of *S. rileyi* was obtained from a *T. ni* larva on cotton.

Numerous isolates of a nuclear polyhedrosis virus were obtained from *T. ni* larvae throughout the season. Epizootics of this virus among *T. ni* larvae have frequently been observed in Alabama fields.

Eighteen fungal, 71 bacterial, and 118 suspect-viral isolates were tested for pathogenicity. Numerous isolates in each group were found to cause some mortality among test larvae. Those microorganisms that induced highest and most rapid mortalities and thereby considered the most promising as control agents are listed in Table 4.

Pathogenicity tests revealed many potential control agents for *H. virescens, H. zea, P. includens,* and *T. ni.* Several factors influencing the activity of the pathogens and disease development must be investigated before promise of field control can be anticipated.

TABLE 4. MICROORGANISMS PATHOGENIC TO CERTAIN LEPIDOPTEROUS INSECTS

Insect	Pathogen
	Fungi
Heliothis zea	*Alternaria* sp.
H. virescens, H. zea	*Aspergillus flavus*
H. zea	*Cladosporium* sp.
H. zea	*Fusarium* spp.
H. zea	*Fusidium* sp.
H. zea	*Monilia* sp.
H. zea	*Penicillium* spp.
Pseudoplusia includens, Trichoplusia ni	*Spicaria rileyi*
	Bacteria
H. zea	*Achromobacter* sp.
H. zea	*Aerobacter* sp.
H. zea	*Pseudomonas* sp.
H. zea	*Serratia* sp.
P. includens, Rachiplusia ou	*Streptococcus* sp.
	Viruses
H. virescens, H. zea, T. ni	Nuclear polyhedrosis virus

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