CONTROL OF PINK BOLLWORM, PECTINOPHORA GOSSYPIELLA, (SAUNDERS) (LEPIDOPTERA: GELECHIDAE) WITH BIOCONTROL AND BIORATIONAL AGENTS D. H. Gouge, L. L. Lee and J. R. Van Berkum, and T. J. Henneberry USDA-ARS Western Cotton Research Laboratory Phoenix, AZ K. A. Smith biosys, Inc. Columbia, MD C. Payne and D. Ortega Texas A & M University, Extention El Paso, TX

Abstract

At pre-sowing irrigation (mid-March), cotton fields were treated with two entomopathogenic nematode species; *Steinernema riobravis* and *S. carpocapsae* for control of diapausing *Pectinophora gossypiella* larvae. Pima S-6 cotton fields situated in Fort Hancock, Texas were treated at a rate of one billion nematodes per acre. Caged, diapausing larvae were buried in fields at a depth of one inch, in row tops and furrow bases. Nematodes were applied with a spray rig, fixed winged aircraft, or in furrow irrigation via a constant flow, battery box. Fields were irrigated after ground application, prior to aerial spraying and during furrow application. Caged larvae were recovered 48 hours after nematode application.

All application methods resulted in uniform distribution of nematodes over the treated fields. No significant differences in larval mortality between nematode species or application method could be determined. However, aerial and furrow application methods gave consistently better parasitism of larvae compared to ground rig delivery. Larval mortality in cassettes buried in furrow bases was significantly higher than in row tops. Larval mortality ranged from 53.26-79.14%. Both nematode species could be recovered 50 days post application.

At pin-head square Frustrate[®] PBW pheromone bands (biosys, Inc.) were applied at 100 bands per acre placement rate (16 g a.i./acre), giving a target release of 115 mg gossyplure/acre/day. Capillary gas chromatography was used to analyze bands throughout the growing season. A uniform release profile indicated sufficient release of pheromone for 144 days after placement.

Pink bollworm mating disruption was monitored in three ways: 1. Delta 2 traps were positioned throughout the farm, forming a continuous trap line. Significantly larger

numbers of moths were recovered form untreated zones. 2. Virgin female moths were placed in mating stations at dusk. At sun rise moths were collected and later dissected for spermatophores. Significantly higher mating activity occurred in untreated fields (p=0.000). 3. Green bolls were collected at random and examined for larvae. Significantly higher infestation levels existed in untreated zones.

At harvest (November), seed cotton yields were weighed using trailer scales. Higher yields were recovered from pheromone (1,864 lb/acre), and pheromone + nematode fields (1,712 lb/acre), than control fields (1,450 lb/acre). However, due to large variations between fields, the differences were not statistically significant (p=0.436).

Introduction

Pink bollworm, *Pectinophora gossypiella* (Saunders), is one of the most serious pests of cotton occurring throughout most of the tropical and subtropical regions of the world (Ingram, 1994). It is considered one of the most damaging cotton pests in Arizona and southern California. Heavy insecticide use often promotes resurgence of secondary pest species such as *Heliothis virescens* (tobacco budworm), *Helicoverpa zea* (bollworm), and *Bucculatrix thurberiella* (cotton leafperforator) (University of California, 1984).

Pink bollworm larvae feed on flower buds, flowers, bolls and the seeds within. Damage to developing seeds, and the termination of growth results in boll rotting, premature or partial boll opening, reduction of staple length, strength, and increases trash content in the lint. Estimated yield losses in the U.S.A. due to pink bollworm range from 9% when chemically controlled to 61% when uncontrolled (Schwartz, 1983), although 100% crop loss can occur with heavy infestations. Pink bollworms spend the winter as diapausing larvae, then pupate and emerge as adults in spring and early summer (Bariola & Henneberry, 1980). After eclosion, moths disperse widely over large areas primarily from the previous years cotton fields, to find susceptible cotton or wild plants (Flint & Merkle, 1981). Pima cotton is particularly susceptible to pink bollworm attack, due to the long growing season required before harvest.

Entomopathogenic nematodes in the family Steinernematidae are established biocontrol agents of a broad spectrum of insect pests occupying cryptic habitats (Begley, 1990, Klein, 1990). Steinernematid (Rhabditida: Steinernematidae) nematodes are obligate insect parasites (Poinar, 1979) associated with a bacterial symbiont, *Xenorhabdus* spp. (Akhurst & Boemare, 1990). The infective juvenile stage of the nematode seek potential host insects within the soil. Having entered through natural body openings, the bacteria is released in the insect hemocoel causing septicemia and death of the insect (Kaya & Gaugler, 1993). Nematodes feed on the bacterial cells and certain components of host tissues. The nematodes may pass through several generations, and once host reserves are

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depleted a new generation of infective juveniles exit the cadaver (Kung *et al.* 1991).

Interest in developing steinernematids as biological control agents has arisen due to their safety for vertebrates and nontarget organisms, mass production and commercial availability, and exemption from Environmental Protection Agency registration (Gaugler, 1981). Steinernematids have been particularly successful bio-insecticides when used against soil dwelling lepidopterous larvae (Cabinillas & Raulston, 1996). Entomopathogenic nematodes are commercially used in mint, citrus and turfgrass.

Steinernematids have been applied using a variety of standard agrochemical application equipment. Nematodes have been applied using aircraft (Lindegren *et al.*, 1981), through tractor spray booms (Georgis, 1990), back-pac sprayers (Gouge & Hague, 1995), soil injection and shanking equipment (Smith, personal communication), and in-furrow irrigation in corn (Cabanillas & Raulston, 1996) and cotton (Forlow-Jech & Henneberry, 1996).

Robbins & Brown (1996), discussed that nematode application methods appear to influence effectiveness of *S. riobravis*. Abiotic conditions such as desiccation and ultraviolate light affect nematode mortality (Gaugler & Boush, 1978). Nematodes appear to be most effective when applied to moist soils after irrigation (Downing, 1994, Shetlar, *et al.*, 1988), or via furrow irrigation (Cabanillas & Raulston, 1996). Cabanillas and Raulston (1996), concluded that application success was dependent upon a system that provides a uniform nematode distribution in the cotton bed, and that *S. riobravis* is effective at suppressing corn earworm (*H. zea*) under field conditions of high temperature with irrigation.

Another limiting factor affecting efficiency of biocontrol agents is coordinating application with the phenology of susceptible insect stages. Application of entomopathogenic nematodes through irrigation systems may introduce nematodes to the insect pest species whilst reducing the effects of desiccation. Wright *et al.* (1993) obtained good control of *Diabrotica* spp. by applying *S. carpocapsae* through a center pivot irrigation system.

Grewal *et al.* (1993), reported the optimum temperature for *S. carpocapsae* activity to be 75.2° F. Grewal *et al.* (1994) also established that *S. riobravis* is able to infect over a wide range of temperatures ($50.0-102.2^{\circ}$ F). Soil temperatures in cotton rarely exceed 104.0° F, thus the nematode activity in the cotton field should not be adversely affected for much of the year.

Gossyplure, (Z)-7(Z,E)-11-hexadecadien-l-ol, acetate, a sex attractant for *P. gossypiella* males (Hummel *et al.* 1973), is commercially used to disrupt mating in the U.S.A., Egypt and Greece. In theory male moths fail to locate and mate with females, thus protecting susceptible bolls from infestation. The present experimental field trial investigates the efficacy of *S. carpocapsae* and *S. riobravis* combined with pheromone induced mating disruption as a biocontrol system for *P. gossypiella* in cotton.

Materials and Methods

455 acres of Pima S-6 cotton was treated with either S. carpocapsae (biosys, Inc. Strain 25) or S. riobravis (biosys, Inc. Strain 355) nematodes, at pre-sowing irrigation (mid-March). The crop was sown at 38 inch spacing in sandy loam soil. Nematodes were applied at a rate of 1 billion per acre using one of three methods: 1) Infective juvenile nematodes were applied using a standard spray rig, delivering 40 gallons per acre at 40 PSI, through course VS 80-O8 nozzles (Tee Jet). The tractor advanced at 6 mph, and broadcast the nematodes over 8 rows using 18 nozzles (20 inch nozzle spacing). Fields were irrigated immediately after application. 2) Aerial application, utilized an Ag Husky fixed wing aircraft. Nematodes were applied in 10 gallons per acre. 31 nozzles were used fitted with D-12 cores and sprayed at 32 PSI. Whirl plates and strainers removed from the nozzles, and orientated directly backwards producing a 35 foot swath. A Satellite Locking GPS guidance system and a prototype flow control unit aided accurate field application. Plane ground speed was 120 mph. Fields were irrigated prior to aerial spraying. 3) Application of nematodes through the irrigation water involved mixing nematodes in an agitation nurse tank, they were then fed into a constant flow battery box situated over an irrigation ditch at the water inlet. Fields received water through base gates.

Range of temperatures at application March $12-19^{\text{th}}$: 62.6-82.4°F four feet above bare soil, 55.4-75.2°F one inch below the surface, the nematode suspension temperature never exceeded 78.8°F during application. Nematode viability assessments were made visually with the aid of a microscope as the nematodes were sprayed from the nozzles. Nematode viability never decreased below 93%.

Prior to nematode application, caged diapausing pink bollworm larvae were caged within biopsy cassettes and buried in fields one inch below the soil surface (Diagram 1). Cassettes were recovered 48 hours later. Cassettes were positioned in five strip plots within each field. Each strip consisted of five cassettes, in row tops and five in furrow bases. The cassette cadavers were dissected in 1/4 strength Ringer's solution and the presence of nematodes recorded.

Ten, 100 ml soil samples, were collected randomly from row tops and furrow bases. The samples were baited with eight late instar *Galleria mellonella* larvae in large Petridishes, from each field. Dry soil was moistened with distilled water until the sample was moist but not wet. After three days incubation at 80.6^oF, larvae were collected from the soil samples and washed in distilled water. Larvae were then dissected in ${}^{1}/_{4}$ strength Ringer's solution under a stereo dissecting microscope and number of infected insects recorded. Weekly soil samples were taken from all fields until entomopathogenic nematodes could no longer be detected.

A delta 2 trap line was arranged across treated and untreated zones. Traps were examined every 3-4 days and numbers of male moths caught recorded.

At pin-head square, Frustrate[®] PBW (biosys, Inc.) pheromone bands were hand applied at 100 per acre placement rate (160 mg gossyplure/band), the target release of 115 mg gossyplure/acre/day is reported to disrupt mating. Capillary gas chromatography was used to analyze gossyplure release from bands throughout the growing season. All nematode treated fields received pheromone bands along with an additional 50 acres of cotton situated north of the nematode treated fields. A 50 acre block of fields, east of the nematode treatments was selected as the control zone and cotton in this area received neither nematodes nor pheromone bands.

Weekly mating stations (Lingren et al., 1982) were placed in five fields treated with nematodes + pheromone. pheromone only, or control fields. The stations were used to assess the impact of treatments on mating of virgin laboratory reared female moths and native males. Mating stations were made from one gallon buckets attached to wooden stakes which were adjusted so that the buckets remained at the top of the cotton plants as they grew. Laboratory pupae were sexed and separated. Two-three days after emergence, females were anaesthetized by chilling and their wings clipped using fine scissors to prevent them from flying out of the mating stations. After eclosion, moths were allowed to adjust to the natural photoperiod and temperature. Two wingless virgin female moths were placed in mating stations with an excised cotton terminal for cover at dusk. The inner rim of the bucket was sprayed with belt grease to prevent the moths from crawling out of the station. At sun rise moths were collected and later dissected for spermatophores, indicating successful mating had taken place.

Fifty green bolls were collected at random from each field at 7-14 day intervals. Bolls were cracked open by hand, and number of pink bollworm larvae and exit holes counted.

Harvesting

Fields were harvested mid-November using four and five row commercial cotton pickers. Cotton from all experimental fields was weighed by dumping from pickers into a boll buggy situated on digital trailer scales next to a module maker. 0.5 lb samples of cotton were collected from the buggy for each field. These samples were ginned using a 20 saw laboratory cotton gin (Porter Morrison & Son) and the seed x-rayed for damage evaluation. Lint samples were also graded using HVI standards of quality. In each of the experimental fields, 0.5 lb samples of cotton were hand picked from each of the following plant nodes: 10, 13, 18, and 20. These samples were also were ginned and subject to seed x-ray evaluation and lint grading. Once the seed cotton samples had been ginned the lint and seed was weighed separately to establish the percent lint from the total seed cotton weight.

Analysis of variance was used to test for significance and difference among treatment means. Duncan's multiple range test was used to partition means into significant ranges when a significant F value was determined by analysis of variance. The Student-Newman-Keuls test was used to established range differences when an uneven number of replicates exist. 5% level of probability was used in all statistical tests. Percentage data was transformed using an arc sine transformation before analysis.

Results

The highest parasitism of caged pink bollworm larvae occurred in fields treated with *S. riobravis* applied by air (79.14 %, Table 1). No significant differences between application method or nematode species occurred. However, infection of larvae caged in the base of furrows was significantly higher than those caged at the row tops (p=0.013) and the Student-Newman-Keuls test established statistical range differences.

Capillary gas chromatography (Perkin Elmer Model 8600 chromatograph) was used to analyze pheromone bands throughout the growing season. The linear gossyplure release profile indicated sufficient release of pheromone for 144 days after placement (Figure 1). Regression analysis (R=0.916) indicates an average daily release of 1.31 mg gossyplure per band, daily (a release value above the target of 1.15 mg per band, daily). 144 days after placement 91.49% of the total gossyplure load had been released (Figure 1). Of the 160 mg a.i. load, 13.6 mg remained.

Entomopathogenic nematodes extracted from soil samples indicted both nematode species persisting until the end of April (approximately 6 weeks). The percentage of G. *mellonella* parasitised fell below 20% and soil sampling was discontinued at this time.

Male moth counts collected from Delta 2 traps (Diagram 2) from March through May suggest few adults emerging from nematode treated zones (Figure 2). Throughout the season, four points along the trap line can be identified as areas where moth counts are larger: traps 1-10 (control fields west of the farm), traps 42-50 (control fields east of the farm), traps 22-23, and 29-30 were situated on southern perimeter next to farm boundaries. Finally, traps 39-40 and 32-34 were situated in the northern control fields.

Moths trapped June though November (Figure 3 and 4), indicate large differences between the numbers of male moths caught in fields treated with pheromone bands (fields 1-26, 32-39) and control fields (fields 27-31). Larger numbers were recovered from traps 1-10 traps (control west of the farm), 42-50 traps (control east of the farm), the control fields north of the treated fields show higher average numbers caught (Figure 4), but there is obviously a pheromone influence depressing trap counts due to the close positioning of treated and untreated fields. Figure 5 indicates the largest moth populations occurring in the months of May and November.

Mating stations showed significant differences in mating activity between pheromone treated and untreated fields (p=0.000). However, the percentage of females mated by native males in control fields was abnormally low (<25%, Figure 6). Normally you could expect to find up to 62% of females mated in non-pheromone treated areas (Flint & Merkle, 1984). Perhaps due to the close proximity of treated and untreated fields, the reduced frequency of mating in untreated fields was due to the influence of gossyplure movement from treated fields.

Similarly, the percentage of bolls infested with pink bollworm or with exit holes was less than expected in control fields north of pheromone treated fields (Figure 7). However, boll infestation counts taken from the Cook Farm (approximately half a mile east of the Miller Farm) showed significantly higher infestation levels (p=0.000). Infestation levels in treated fields remained below 6% until September 24, and below 12% until October 7. However, by October 25, infestation levels in all fields had risen above 40% (Figure 7.)

<u>Harvest</u>

Despite higher mean seed cotton yields from pheromone and pheromone + nematode treated fields (Table 2), significant differences were not observed due to large variations between fields. However, treated fields positioned in certain peripheral areas sustained higher levels of pink bollworm attack throughout the year (e.g. Fields 35 and 36, Figure 2). Compared to mean control lint yield, pheromone only treated fields produced an average of 25.70% higher lint yield, while Pheromone + nematode treated fields produced an average of 18.92% lint yield increase.

Percent lint values of total seed cotton weight were determined to be significantly higher in pheromone and pheromone + nematode treated fields (p=0.039, Table 2).

Control fields having larger pink bollworm infestation experience seed damage due to larval feeding. A single larva may attack several seeds. Damaged seeds produce less fiber and decreased percentage lint. Cotton seed xrayed for insect damage, showed significant differences between treated and untreated seed (p=0.018) (Table 3). Analysis of seeds taken from specific nodes indicated no significant differences in damage to lower nodes N10 and N13, but significant differences between treated and untreated seed from higher nodes N18 and N20. The latter two nodes offer susceptible bolls later in the season when pink bollworm populations are increasing. Mean seed damage values taken directly from the cotton pickers also shows significant differences (p=0.018) between treated and untreated cotton (Table 3).

Several fiber characteristics are graded using HVI analysis. Strength, reflectance, spottiness, color grade length, length uniformity, elongation, and micronaire. Significant differences between fiber reflectance (p=0.000) and spottiness (p=0.000) due to cotton node placement showed upper node fibers with higher reflectance values and lower node boll fibers with higher spottiness values. No treatment effects were apparent.

Discussion

Infective juveniles of S. riobravis were capable of detecting, infecting and killing larvae of P. gossypiella during the period that fields retained suitable soil moisture levels after irrigation. All three application methods employed had no harmful effects on the infective juvenile nematodes, and provided uniform releases. Application to dry soil (as in the case of tractor boom application) may account for the reduction in mortality of caged pink bollworm larvae. Irrigation of the ground rig treated fields took up to 8 hours to be completed. A percentage of nematodes applied to the dry soil surface will probably be lost to desiccation and exposure to UV (Gaugler & Boush, 1978). Cabanillas and Raulston (1996) describe similar trends spraving S. riobravis in corn. Application to the dry soil surface produced lower mortality levels of H. zea compared to application of nematodes in the furrow irrigation water or by air. after irrigation.

Nematode recovery was consistently better in the furrow bases. Nematodes will avoid dry areas where their movement is restricted. Infective juvenile nematodes of *S. carpocapsae* and S. *riobravis* appear to be highly attracted to pink bollworm larvae under field conditions. The presence of buried, caged larvae in the row tops may have improved migration of nematodes into this area. Georgis and Poinar (1983), described enhanced lateral dispersal of several nematode species due to the presence of a suitable host.

The furrow base remains moist for longer periods and nematodes may naturally desiccate gradually as fields dry. Steinernematid nematodes can survive extended periods in an anhydrobiotic state if the drying process is gradual (Simons & Poinar, 1973). Although nematodes are immobile and thus non-pathogenic in the desiccated state, it is a survival strategy that allows infective juveniles to persist until more favorable environmental conditions return. Consequently, when fields are irrigated, nematodes rehydrate and are once again mobile, infective and pathogenic to pink bollworm.

Certain nematode species are known to be ecologically adapted with respect to soil temperature and humidity requirements. Kung *et al.* (1991), attributed the subtropical origin of *S. glaseri*, to be one adapted factor allowing it to tolerate higher soil temperatures ($59.0-95.0^{\circ}F$). However, survival under low soil temperatures ($41.0^{\circ}F$) was poor.

Cabanillas and Raulston (1996), report successful parasitism of *H. zea* by *S. riobravis* in soil temperatures between 71.6-98.6°F. Grewal *et al.* (1993) determined the optimum temperature for nematode penetration and establishment of *S. carpocapsae* in *G. mellonella* as 75.2° F. However, Grewal *et al.* (1994) established the temperature range for infection of *G. mellonella* by *S. riobravis* as 50.0-102.2°F. Additionally, Gouge *et al.* (1996) reports successful control of pink bollworm larvae using *S. riobravis* in mid-season cotton where soil temperatures reached 128° F.

Diapausing *P. gossypiella* larvae are highly susceptible to *S. carpocapsae* and *S. riobravis* under field conditions. Nematodes may be applied using standard pesticide spray equipment or added to furrow irrigation water during irrigation of laser level fields. Since nematodes are compatible with most chemical pesticides and fertilizers (Georgis, 1990), and have little impact on beneficial insects (Georgis *et al.*, 1991, Poinar, 1989), they can be considered a convenient tool for use in cotton IPM systems.

Gossyplure rates below 4 g a.i./acre/season may be applied to initiate "false trail following" (Cardé, 1981) by male moths. Higher rates of gossyplure (in the case of the present experiment, 18.86 g a.i./acre/season) are sufficient to induce adaptation of attennal receptor sites or habituation of the central nervous system (Shorey *et al.* 1976). With larger doses of gossyplure, the mode of action is different and the number of point sources of pheromone is not as critical (Flint *et al.* 1985).

In this experiment placement of pheromone bands at cotton pin head square, resulted in bands placed between the 5th and 8th node. It is unlikely that gossyplure moves through the canopy to the aerial part of the plant. However, moths spend their days on the lower third of the plant or in the soil below (Flint *et al.* 1975), and are exposed to an ambient atmosphere containing large amounts of gossyplure. Male moths emerging from the soil are disorientated sufficiently. Late season infestation of the top susceptible bolls may have a resulted from large populations emerging in surrounding areas and entering the treated fields. In the upper plant canopy, the arriving ingress, may not be affected by the gossyplure, or females may already be fertilized.

Delta 2 traps use gossyplure loaded rubber septa to attract male moths. Septa are placed on a platform of sticky glue

that traps moths as they arrive. Trap counts were used to assess populations throughout the growing season. Pheromone baited traps are commonly used for detection (Foster *et al.* 1977), and a tool for determining if control action is necessary (Toscano *et al.* 1979). Delta 2 trap population estimates along with boll infestation data, and virgin female mating frequency (in mating stations), were all used together to assess the potential of crop damage. In this study there was a strong correlation between Delta 2 trap counts, and crop infestation. However, when using pheromone bands to disrupt mating, Delta 2 trap counts alone should not be used as reliable population indicators.

The Delta 2 trap line across the Miller farm did indicate large numbers of moths in the peripheral fields treated with pheromone bands. The heaviest infestation of treated fields occurred along the southern farm limits which bordered the boundary between the U.S. and Mexico (a cotton growing area where pink bollworm control methods are unknown). Observing these affects, reinforces the necessity to treat large areas if biological and bio-rational agents of migratory pests are to be studied.

The late season breakdown in pheromone control was reflected by high Delta 2 trap counts (Figure 5) and high boll infestation levels (Figure 7). However, virgin female moth mating rates in control fields (Figure 6) remained low. This is another indication that top susceptible bolls are attractive to fertilized female moths arriving from surrounding fields.

Harvest data showed consistent but non-significant increases in lint yield. An 18.92% and 25.70% lint increase for nematode + pheromone and pheromone treated cotton fields was observed respectively. Similarly, Gouge *et al.* (1996) reports a 19% seed cotton yield increase between mid-season cotton treated with *S. riobravis* and untreated cotton.

Seed damage due to insect feeding showed consistent differences between treated and untreated cotton. But specifically, upper bolls offered statistically significant differences when bolls were under heavier attack from larger moth populations.

Results of this study suggest that combining entomopathogenic nematodes and pheromone induced mating disruption, offers a control strategy designed to reduce reliance on chemical control of pink bollworm. Harvestable bolls were protected season long, but top boll infestation prior to harvest, would have required chemical treatment to maintain infestation levels below 6%.

Disclaimer

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

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Cross section of cotton bed



Diagram 1. Cassette positioning.

Table	1.	Mortality	of	caged	pink	bollworm	larvae,
parasitised by entomopathogenic nematodes.							

Treatment	% PBW parasitised			
	Furrow base	Row top		
Air- S. carpocapsae	73.27	60.68		
Air- S. riobravis	79.14	73.71		
Boom- S.	64.54	63.45		
carpocapsae				
Boom- S. riobravis	68.40	60.90		
Gate- S. carpocapsae	63.08	53.26		
Gate- S. riobravis	68.56	58.67		



Figure 1. Release rate of gossyplure from PBW bands.



Diagram 2. Miller farm field map.



Figure 2. Total male moths caught in Delta 2 traps throughout the farm.



Figure 3. Total moths caught in Delta 2 traps throughout the farm, June-November.



Figure 4. Mean moth capture per trap for the growing season.



Figure 5. Mean number of male moths caught in Delta 2 Traps each month.



Figure 6. Percent virgin female moths mated by native male moths.



Figure 7. Percent boll infestation, both the presence of pink bollworm larvae and exit holes was considered

Table 2. Cotton harvest.

Treatment	Mean seed- cotton (lb/acre)	% lint	Lint (lb/acre)
Control	1,450.05	42.02	609.32
Pheromone	1,863.97	44.00	820.14
Pheromone + nematodes	1,711.88	43.90	751.52

Table 3. Percent insect seed damage from hand picked cotton samples, samples were taken from different nodes along the main stem. **Mean - cotton picker**, refers to samples taken directly from cotton pickers.

	Nodes along main stem					
Treatme nt	N10	N13	N18	N20	Mean - Cotton picker	
Control	11.3 5	14.59	21.15 a	20.05 a	17.17 a	
Pheromo ne	11.9 9	11.74	14.31 b	13.52 b	12.92 b	
Pheromo ne + nematod es	11.6 0	10.58	14.68 a b	15.61 a b	13.23 b	