

THE EFFECT OF COLORADO POTATO BEETLE CONTROL MEASURES ON NON-TARGET ARTHROPODS¹

G.L. Reed², K. Puls², A.S. Jensen², J. Feldman³, & R.E. Berry⁴

The recent development by HybriTech Seed International, Inc.⁵ of transgenic "Russet Burbank" potato clones with resistance to Colorado potato beetle (CPB) provides an opportunity to explore a new avenue for control of CPB. The resistance mechanism is from *Bacillus thuringiensis* var. *tenebrionis* which is effective in controlling CPB. Using this mechanism as a gene, rather than applying it as an insecticide has several significant advantages. First, it functions similar to a systemic insecticide except that Btt expresses nearly uniform levels throughout the growth of the plant and level of the expression does not recede as does an insecticide. Second, being expressed as a gene inside the plant reduces the impact that it has in the environment, making it ecologically "gentle".

Grower acceptance of the transgenic potato will be dependant on their ability to recover adequate reduction in insecticide costs or reduce crop losses to pay for the increased seed costs. Growers and scientists in the Pacific Northwest feel that CPB control occurs as a result of controlling green peach aphid (GPA) and thus no cost is expended to control CPB. In order to determine whether that was true, research was undertaken in 1992 to determine the impact of transgenic CPB resistant on non-target pest and beneficial species occurring in the potato crop.

Methods & Materials

Research was conducted at Hermiston Agricultural Research & Extension Center, Hermiston, Oregon. Crop production procedures were much the same as used locally except that the three acre plot was hand planted, seeded 14" apart on rows spaced 34" apart.

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² Oregon State University, Hermiston Ag Research & Extension Center and Department of Entomology.

³ HybriTech Seed International, Inc.

⁴ OSU, Department of Entomology.

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A Latin Square design was utilized to compare six treatments:

Unprotected plants - designated NONE in figures,
Systemic treated plants (Thimet followed by Di-Syston) - SYS,
Permethrin treated plants - PERM,
Transgenic "Russet Burbank" (RB) plants - TBtt,
Transgenic RB plants with systemic insecticide - TBtt&SYS, and
Insecticidal Btt (M-Trak) - IBtt.

Application dates included: Thimet 6/6; Di-Syston 7/9, permethrin every other week beginning 6/23, and M-Trak weekly beginning 6/23. Each experimental unit consisted of a 51'x51' (16 rows) area of treated plot bordered on all sides by unprotected plants. The center 20'x20' was used to collect research data. Plots, other than the transgenic, were planted to certified normal "Russet Burbank" seed.

Research parameters were measured twice each week from 6/19 to 8/27:

Colorado potato beetle and *Perillus* spp. were counted on one side of two rows (16 plants/row) without disturbing the plant vegetation. CPB adults, egg masses, 1st and 2nd instar larvae, and 3rd & 4th instar larvae were counted. All *Perillus* spp. stages were counted.

A beating cloth (28"x28") was inserted gently under one side of two plants to the stems, the plants were gently folded over the cloth, and the plants struck eight times with a beating stick. All arthropods on the beating cloth were counted.

A yellow water pan traps (8") was placed in each experimental unit. A small amount of copper sulfate was added to the water. Aphids, CPB, predators, and other arthropods were collected and counted.

Two pit-fall traps (3-1/2") containing copper sulfate were placed in each experimental unit. All arthropods were collected and counted.

Results

Colorado potato beetle:

Plants with transgenic resistance to CPB had fewer numbers of adults (Fig. 1) and egg masses (Fig. 2), and no CPB larvae survived (two freshly hatched 1st instars being the total observed on the two treatments season long) (Fig. 3 & 4). Permethrin, m-trak, and the systemic insecticides all provided adequate control of CPB. M-trak had similar numbers of adults and only one third the number of egg masses as the unprotected control, but provided adequate control of larvae. However, there were larvae that matured on the plants treated with m-trak, prompting concern about the potential for development of resistance to m-trak. Egg masses counts on insecticidal Btt plants was similar to those on permethrin. CPB has developed resistance to permethrin in the eastern U.S.

Exposure to egg masses and survival low numbers of larvae, create the potential for sub-lethal dosage resistance selection. Conversely, we have yet to observe surviving larvae on the Btt plants transgenic plants in two years of extensive research. In dynamic biological systems, it is likely that resistance to any mechanism will occur; however it seems more likely that resistance will occur more rapidly when selection occurs through sub-lethal dosage.

Green peach aphid:

Alate GPA populations were very high in permethrin treated plots but remained low in all other treatments (Fig. 5). Flight of GPA into the research plots was rather uniform as shown by Fig. 6. Seasonal distribution of GPA alates (Fig. 7) indicated that populations; which were uniformly sampled with yellow water pan traps (YWPT), were significantly different when sampled on plants using a beating cloth. Alate populations in permethrin plots were significantly higher than populations in all other treatments. Alate populations in the systemic and combination transgenic-systemic plots were expectedly low because of treatment with the systemic insecticides. Reduction of the population that alighted in the unprotected check, Btt transgenic and insecticidal Btt plots was due to predation.

The early season alatae population of GPA measured with beating cloth was most likely the result of a flight that immigrated into the area about May 28 (YPT counts, Luther Fitch), prior to emergence of potato plants in this study. It left a residual population of GPA alatae in the region that migrated into the plots as potato plants emerged and were measured in the plots from 6/19 till 6/29 when a second much larger flight immigrated into the plots (Fig. 7). No additional immigration into the plots was measured during the season. However, significant alatae were trapped in YWPT during August (Fig. 8). These alatae were emigrating from the permethrin plots where the population of apterae were very large, and as it immigrated from the permethrin plots, it created a false image of emigrants in other treatments. Even though there were a limited number of GPA alatae emigrating from the other plots late in the season, the low population of GPA in these treatments could not explain the large number of alatae sampled during mid-August.

Apterous populations followed the same general pattern as alatae; populations were very high in permethrin plots and significantly lower in other treatments (Fig. 9). Counts of GPA apterae approached 4,000 per beating cloth sample during mid-August, the equivalent of over 26 million GPA apterae per acre. The seasonal distribution of apterae (Fig. 10) was not as expected. There were almost no apterae in beating cloth samples during the period from 6/19 until 6/29 when the population of apterae began increasing due to the 6/29 flight of alatae. Apterous population in the permethrin plot (which shows the normal development of GPA population when uncontrolled) declined from 7/2 until 7/16. We presume that this was the period necessary for GPA apterae, deposited from the 6/29 flight, to mature and become reproductive. Examination of the permethrin data in Fig. 10 might lead to a supposition that the majority of the apterae deposited in the plots occurred between 6/29 and 7/2.

Maturation of these apterae caused the GPA population in the permethrin plots to grow in an exponential manner between 7/16 and 8/13 (Fig. 11). During this period, apterous populations in the systemic insecticide and transgenic/systemic treatment were held in check by application of Di-Syston 7/9. GPA populations in the transgenic Btt plots increased to 35 per beating cloth sample or ca. 230,000 equivalent per acre during the period from 7/24 to 8/10 (Fig. 11). Though these populations were too small to have direct effect on potato yield or vigor, it remains to be determined whether they might have caused potato leaf roll plaques and increased frequency of infected plants. It is our hypothesis that the principal factor restraining GPA population growth in the transgenic and insecticidal Btt treatments were predators, and that these predators also contributed to low GPA late season population in the systemic and transgenic/systemic plots also.

Predators:

The primary predator populations in the plots were generalist hemipterans and spiders (Fig. 12). *Geocoris* spp., spiders, nabidae, and anthocoridae were most common and comprised over 97% of the predators observed during 1992. These predators were most common in the transgenic and insecticidal Btt plots. Contrary to our expectation, populations of Coccinellidae (lady beetle), Hemerobiidae (brown lacewing), Syrphidae, and *Perillus* spp. were very low. Concern that we might be missing these populations with our sampling procedures resulted in significant observation of the plots in order to verify it. Comparison of treatments in Fig. 12 shows the severe reduction in predator populations caused by the biweekly applications of permethrin.

Geocoris spp. was the most common predator during the study. Populations of *Geocoris* spp. were significantly higher in the transgenic Btt treatment than in all other treatments (Fig. 13). All insecticide treatments reduced *Geocoris* spp. population, although in a commercial planting Di-syston would have been applied earlier in the season and may have impacted the predators less. *Geocoris* adult populations were highest during late June (Fig. 14). Nymphs were observed in high numbers between 7/16 and 7/20 (Fig. 15) suggesting that reproduction began in early July. Adults were reduced in permethrin plots season long. Adults were reduced in plots treated with systemic and transgenic/systemic insecticides and in transgenic and insecticidal Btt treatments after application of Di-Syston on 7/9. The number of predators in samples taken 7/13 was reduced, which we assumed was caused by the Di-Syston application. Adult populations never recovered in the systemic insecticide plots. However, it is important to note that these plots were planted much later than normal because transgenic Btt plants were received late and planted after the normal planting date of north-central Oregon. Di-Syston would most likely be applied in May which may help avoid impacting *Geocoris* spp.. Adult populations in general were highest in the transgenic and insecticidal Btt treatments.

Populations of *Geocoris* nymphs were substantially higher in the transgenic and insecticidal Btt plots and substantially lower in other treatments.

Nymphs feed on sap during the first two which may account for the reduction of nymphs in plots treated with systemic insecticides.

Spider populations were more abundant than anticipated. Most of the spiders that we collected in beating cloth samples were immature, suggesting that the population was initiated by individuals emigrating "ballooning" into the plots. Permethrin drastically reduced spider populations (Fig. 16) and stable populations never developed, although new migrants were entering the plots routinely. Spider populations were highest in the transgenic Btt plots. Spider populations were significantly reduced in the systemic and transgenic/systemic plots after application of Di-Syston on 7/9 (Fig. 17). However, spider populations recovered in these plots between 8/3 and 8/6, contrary to the response of *Geocoris* which never recovered in these plots. Spiders dispersed into plots treated with Di-Syston as it ceased to be efficacious against them; and importantly, before the insecticide ceased to kill GPA. The Btt plots had significantly more spiders than all other treatments from 7/9 to 8/3. Btt plots had more spiders than permethrin treated plots season-long.

Populations of Nabidae were highest in the transgenic Btt plots (Fig. 18). Permethrin dramatically reduced populations of adult and nymph nabids. Nabid adults are very migratory and we were unable to clearly determine seasonal distribution. Nabid adults were present in all plots season-long, although never as numerous as *Geocoris* spp., particularly during the first half of the season. Even though nabid nymphs were observed all season, (Fig. 19), significant populations began 7/16 and continued through 8/20. Like *Geocoris* spp. and spiders, populations of nabid nymphs were reduced by the 7/9 application of Di-Syston. Similar to spiders, the nabid nymph population recovered after about 3 weeks. The Btt treatments had the highest numbers of nymphs compared with the Di-Syston treated plots from 7/13 to 8/3, but were similar during the remainder of the season. Permethrin plots had very few nabid nymphs season-long.

Anthocoridae populations were relatively low season-long. Adult populations were higher than expected in the permethrin plots (Fig. 20). Observations indicate that adult pirate bug is very effective in searching for large populations of aphids. Our data indicate that permethrin is highly toxic to anthocorid adults and nymphs. The populations of adults in permethrin plots were probably new migrants. Adult population in permethrin plots crashed after every application. Adults were very migratory and we did not establish seasonal distribution.

Conclusions:

Transgenic Btt "Russet Burbank" potato were very effective in controlling Colorado potato beetle. The number of adults, egg masses, and larvae were significantly reduced. The Btt insecticide M-Trak effectively controlled all stages of Colorado potato beetle. Exposure to large populations of adults, moderate populations of egg masses and limited numbers of larvae through maturity cause concern that the potential exists for development of resistance in CPB.

Green peach aphid population development in 1992 can be summarized as follows:

- 1) a small population of alatae dispersed through the plots 6/19 to 6/29.
- 2) there was a large influx of alatae into the plots on 6/29;
- 3) the first generation of apterae were deposited in the plots between 6/29 and 7/2.
- 4) the first generation apterae matured about 7/16 and deposition of second field generation apterae began,
- 5) exponential growth of GPA apterae occurred between 7/16 and 8/13, and
- 6) alatae dispersed from permethrin plots beginning 7/30 as plants over-populated.

The population of GPA was significantly higher in plots treated with permethrin than in all other plots due to the effect of permethrin on predators. GPA populations were reduced in plots treated with thimet and Di-Syston. GPA populations were reduced in transgenic and insecticidal Btt plots by high levels of predators. Whether the level of aphid reduction by predators will suppress virus transmission remains to be determined.

Predator populations were generalists with *Geocoris* spp. and spiders being most common, followed by Nabidae and Anthocoridae. Populations of all predators were reduced significantly by permethrin. Di-Syston reduced populations of all of these predators, populations of spiders and nabids recovered after three weeks - prior to recovery of GPA populations. *Geocoris* spp. populations did not recover after application of Di-Syston but perhaps an earlier application of Di-Syston would not have such a drastic effect.

Most of the predators observed in the potato field were immature stages. Observations indicated that most predation against green peach aphid occurred against newly deposited 1st instar nymphs, which is consistent considering the size and maturity of predators. The predators were much more mobile than we anticipated, they quickly infested an area when prey populations existed and quickly dispersed when prey were absent. Research is needed to develop techniques which will (1) foster early season populations of generalist predators in the potato crop and (2) maintain predators in the crop when green peach aphid populations are low.

It appears from the 1992 research that transgenic resistance to Colorado potato beetle may make Integrated Pest Management of insects in the potatoes a real possibility. Obviously an additional year of research is needed to validate these results. If resistance to potato leaf roll virus and root knot nematodes is combined with the Btt transgenic varieties; it could have wide ranging effects by reducing the cost of potato production and by reducing the impact of farming on the environment. If other transgenic resistance sources prove to be nondisruptive in the agroecosystem as the transgenic Btt Colorado potato beetle plants, a new age of INTEGRATED PEST MANAGEMENT may be realized.

Figure 1. Seasonal average CPB adults per treatment.

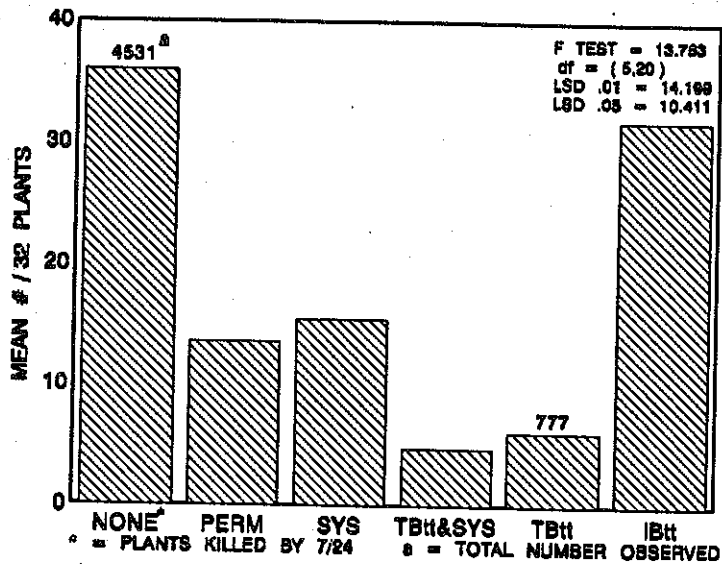


Figure 2. Seasonal average CPB egg masses per treatment.

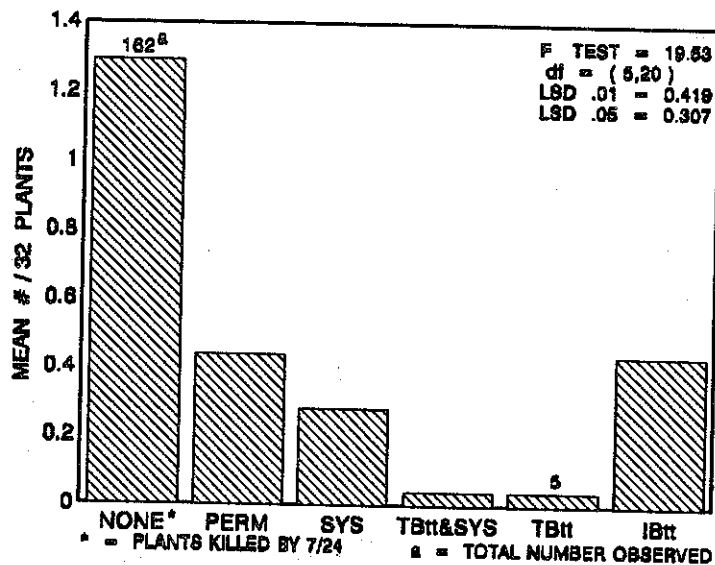


Figure 3. Average CPB 1st & 2nd instar larvae per treatment

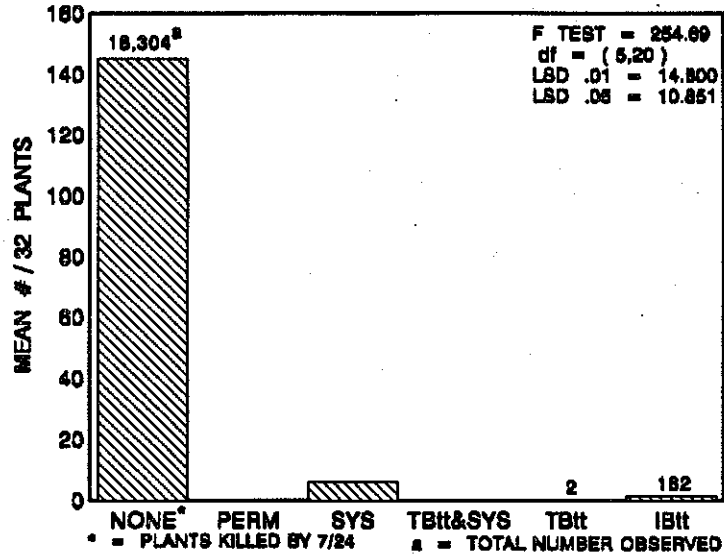


Figure 4. Average CPB 3rd & 4th instar larvae per treatment.

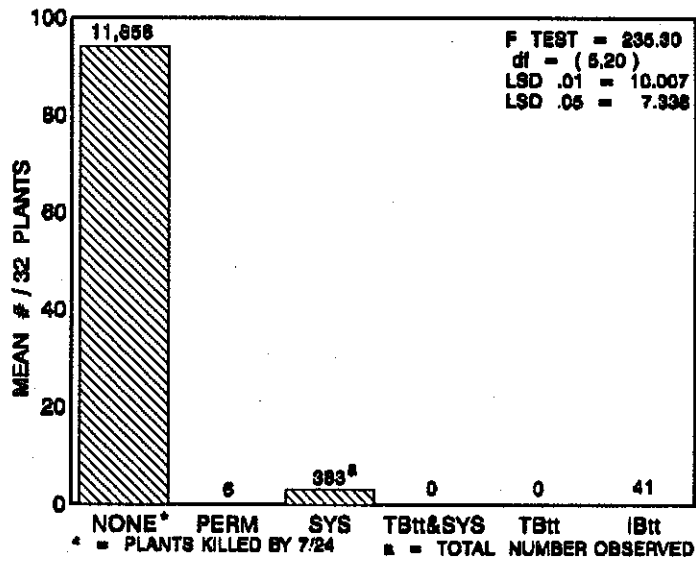


Figure 5. Seasonal average GPA alatae (winged) per treatment.

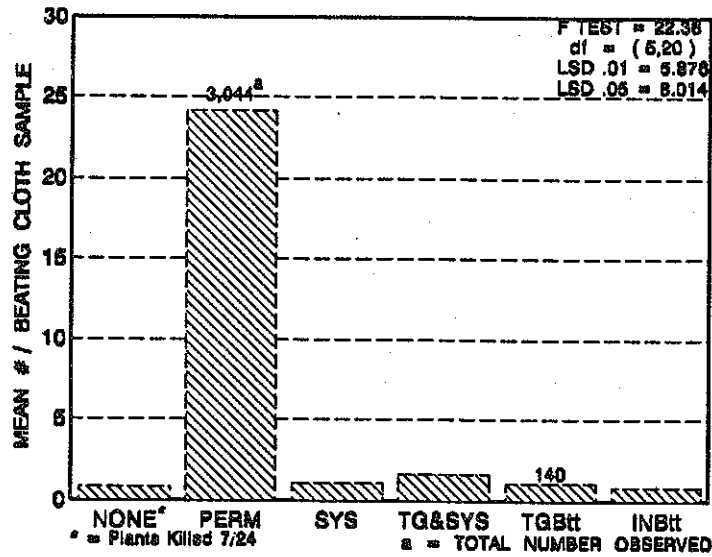


Figure 6. Early season distribution GPA alatae (winged) - yellow pan trap data.

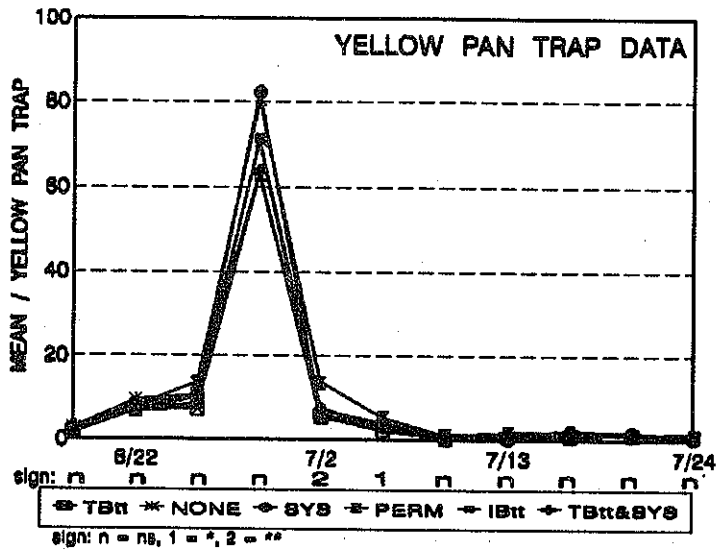


Figure 7. Early season distribution GPA alatae (winged) - beating cloth data.

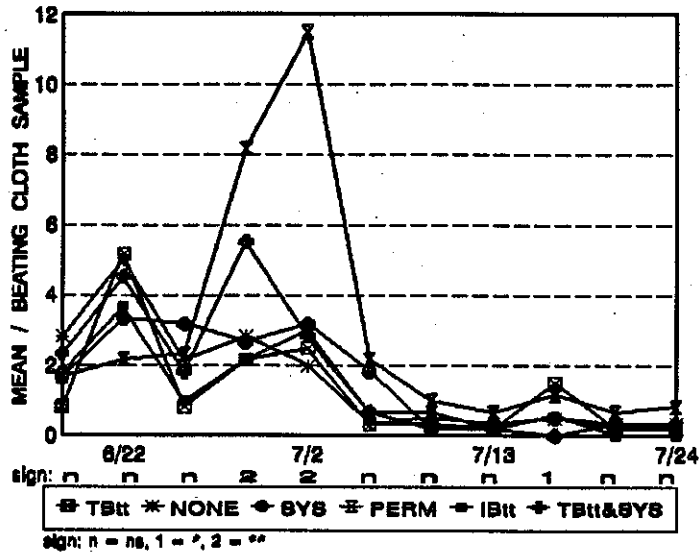


Figure 8. Seasonal distribution of GPA alatae (winged) - beating cloth data.

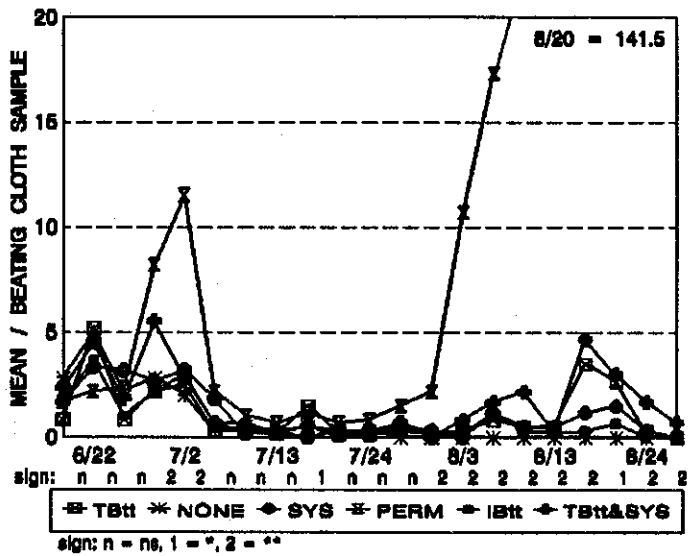


Figure 9. Seasonal average GPA apterae (wingless) per treatment.

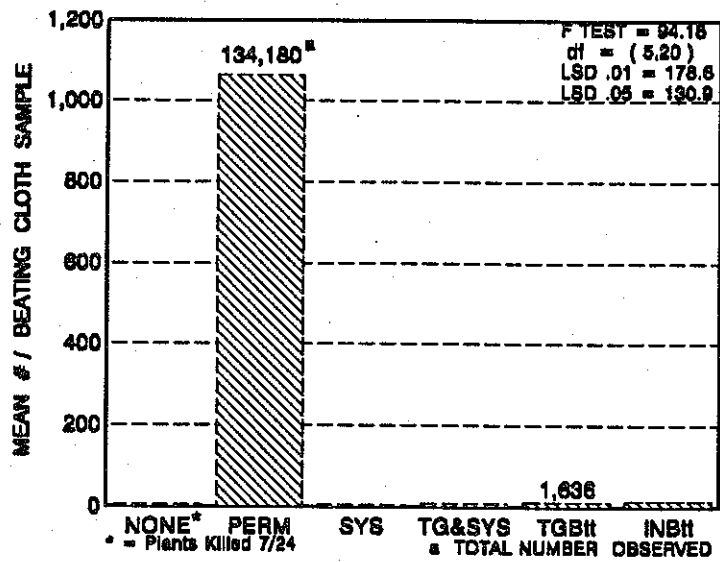


Figure 10. Early season distribution of apterae (wingless) - beating cloth data.

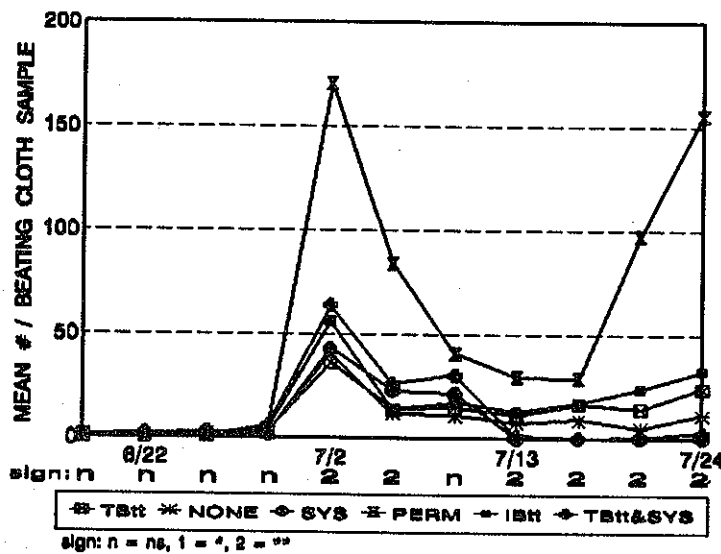


Figure 13. Seasonal average Geocoris individuals per treatment.

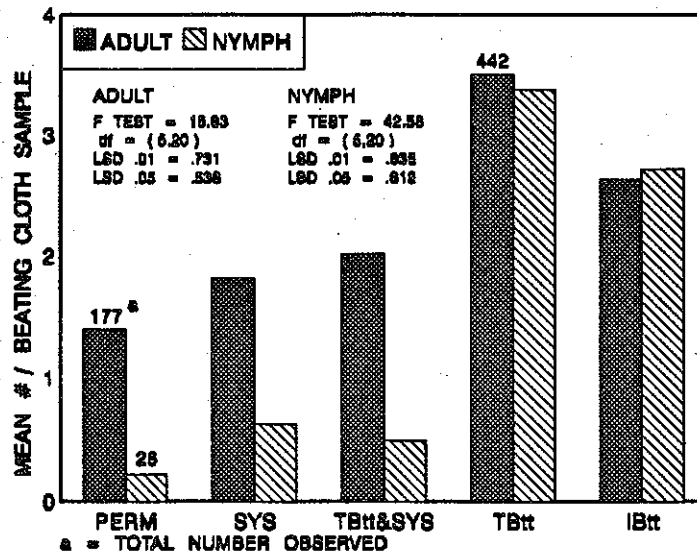


Figure 14. Seasonal distribution Geocoris spp. adults.

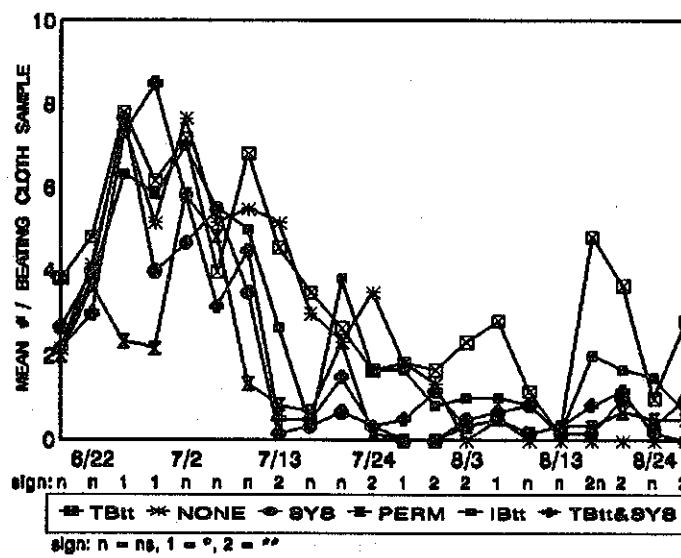


Figure 15. Seasonal distribution *Geocoris* spp. nymphs.

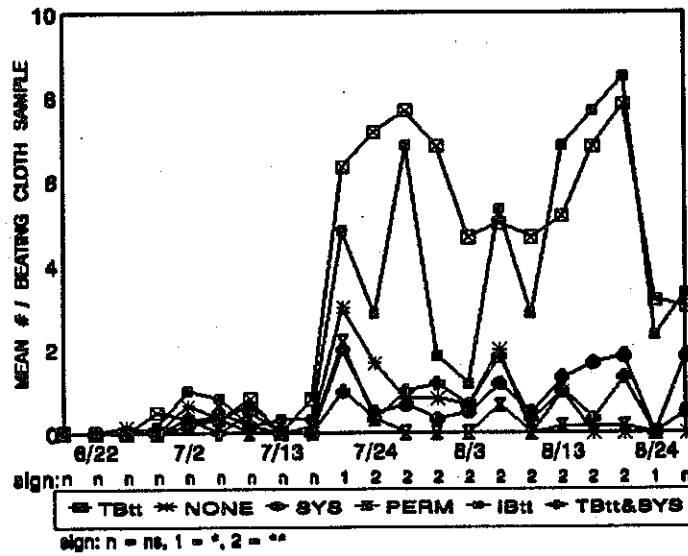


Figure 16. Average seasonal spider population.

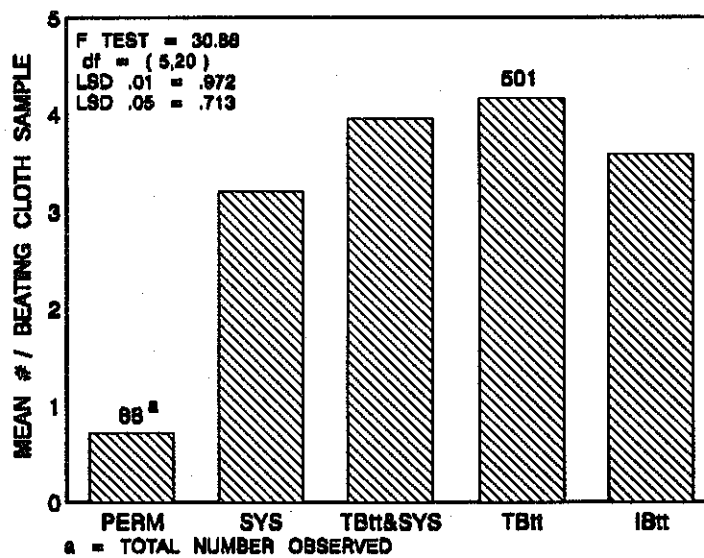


Figure 17. Seasonal distribution of spiders.

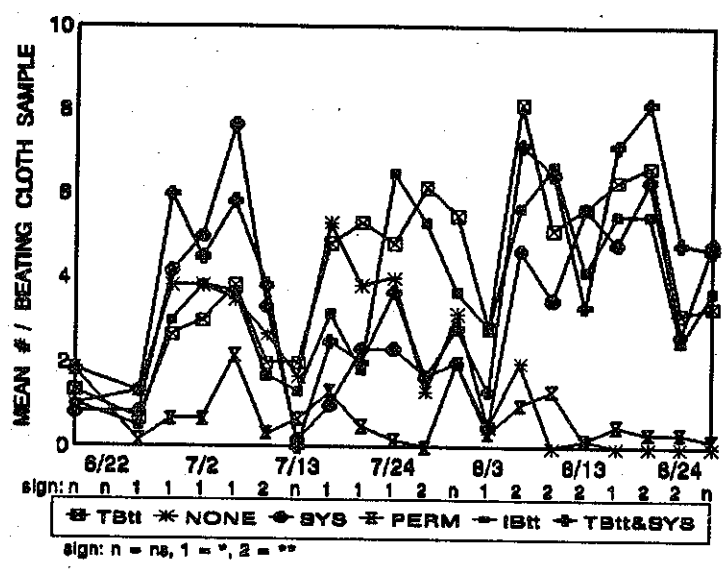


Figure 18. Average seasonal population of Nabidae.

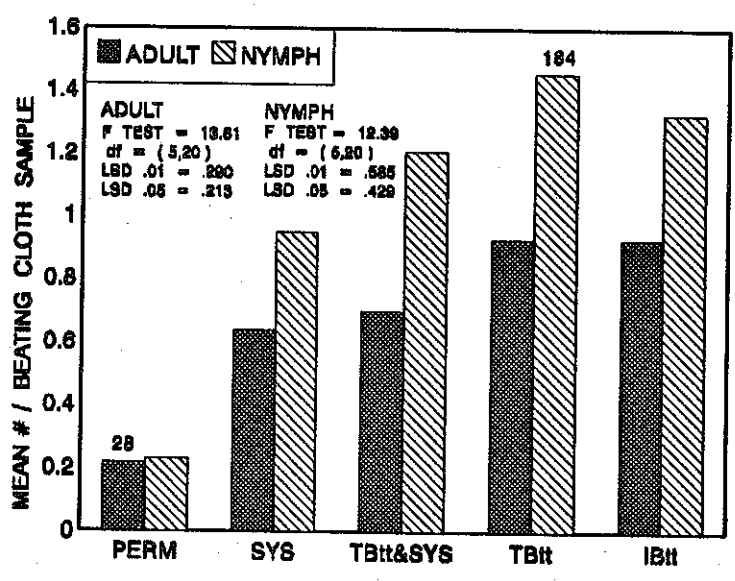


Figure 19. Seasonal distribution of Nabidae nymphs.

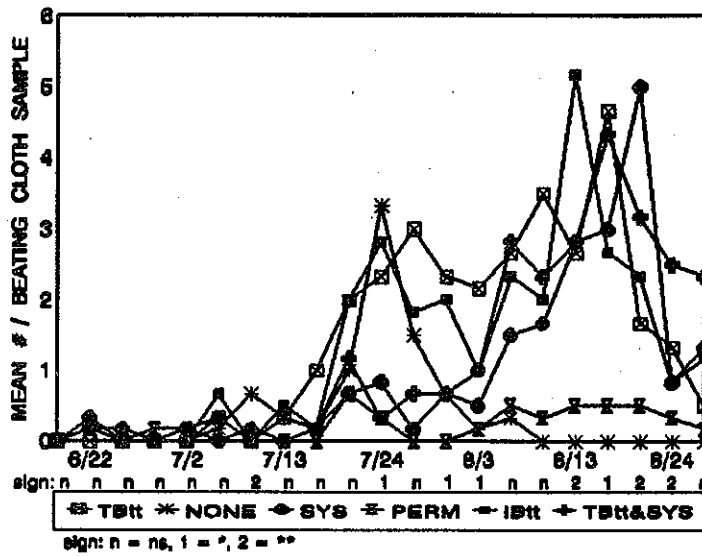


Figure 20. Average seasonal population of Anthocoridae.

