Final Project Report to the NYS IPM Program, Agricultural IPM 2003-2004

1. Title:
Choosing the Best Refuge Hybrids for Planting with Corn Rootworm Resistant Bt Corn

2. Project Leader(s):
Margaret Smith, Department of Plant Breeding, Cornell University
Laraine Ericson, Keith Payne, Department of Plant Breeding, Cornell University

3. Cooperator(s):
John Losey, Department of Entomology, Cornell University
Leslie Allee, Department of Entomology, Cornell University

4. Type of Grant:
Pest resistant crops.

5. Project Location(s):
Throughout the Northeast.

6. Abstract:
Corn hybrids that have been genetically engineered with the Bt gene for resistance to corn rootworm offer growers in New York an interesting option for control of this pest. These hybrids would eliminate the need for soil-applied insecticides in many corn fields, but also require planting 20% of the area of the field with a non-genetically engineered variety (called a "refuge") to ensure that Bt-susceptible rootworms continue to predominate in the rootworm population. Finding corn hybrids that are well adapted for refuge plantings will reduce the need for insecticide use in refuges, increase profitability from refuge plantings, and make it more likely that farmers will comply with the refuge requirement. Ideal refuge hybrids would suffer relatively little from corn rootworm damage (either because larvae do not damage them much or because they can regrow roots well and thus recover from the damage) and yet produce many adult rootworms to ensure that the rootworm population remains mostly susceptible to Bt. Our research was designed to identify such ideal refuge hybrids. Nine corn hybrids that are widely sold in New York were grown in plots that were infested with corn rootworm eggs and in plots that were not infested. Ratings of root damage and root regrowth after damage were made, and cages were installed in each plot to collect and count how many adult rootworms emerged. Based on just this initial year's data, one hybrid appeared to be especially promising as a refuge hybrid, because it had excellent root regrowth and large numbers of adults emerged. It will take several more years of collecting data to confirm this result, and we will need to collect yield data (which is essential to evaluating damage to hybrids from rootworms, but could not be collected in 2003 because circumstances forced a very late planting date). In future years, we will be collecting data not only on rootworm damage, root regrowth, and adult emergence, but also on hybrid maturity, standability, and yield. This will provide a data set that will allow us to identify highly productive hybrids that serve as excellent refuge hybrids, thus providing growers with options to reduce or eliminate pesticide use in refuges, obtain more yield from them, and promote the effectiveness of genetically engineered Bt rootworm resistant corn in the future.

7. Background and Justification:

The population of corn rootworm (CRW) in New York formerly consisted primarily of northern CRW (*Diabrotica barberi*). Over the past decade, the more competitive and more aggressive western CRW (*D. virgifera virgifera*) has invaded and now comprises a significant proportion of the population. Western CRW populations appear to build up more rapidly and cause more severe damage to corn than did those of northern CRW. The cost of CRW to the corn crop nationally is estimated at one billion dollars, including costs of insecticide applications to soil and corn silks and of yield losses to the insect. Insecticide use in New York has increased over the last decade, and now more than half of corn acres are treated with insecticide, mostly for CRW control. Annual rotations, which can control CRW in New York, is not an option for many dairy farmers, as they must fit corn into a dairy rotation that includes multi-year forage crops. The potential damage from CRW and the cost, handling, and labor involved in using insecticides to prevent it will make many farmers view genetically engineered CRW-resistant hybrids as an excellent option.

This resistance, which is based on a Bt gene, brings with it the potential for evolution of Bt-resistant CRW. This would destroy the effectiveness of the Bt crops themselves. To minimize selection pressure towards resistance to Bt in CRW populations, farmers who grow the new hybrids will be expected to plant 20% of their acreage as a non-Bt refuge. The refuge is designed to produce Bt-susceptible rootworm beetles that will mate with any resistant beetles emerging from the Bt hybrid, thus producing progeny that are likely to be susceptible and limiting the evolutionary success of genes for Bt resistance in CRW populations.

The refuge strategy relies on production of significant numbers of Bt-susceptible adult beetles from the refuge planting, hence the beetles are beneficial insects in this system. At the same time, farmers have an interest in minimizing yield losses due to CRW larval damage in the refuge planting, thus the larvae are still pests. Consequently, the ideal profile of a corn hybrid for use as a refuge would be a hybrid that maximizes production of CRW beetles (beneficials) and minimizes yield loss to CRW larvae (pests).

Past evaluations of resistance to CRW within corn revealed that most variation found was due to tolerance – the ability to sustain relatively high levels of larval damage but then regrow roots, thus avoiding significant yield loss. Tolerant hybrids would be ideal for refuge plantings since they would support a relatively large CRW population to adulthood (thus producing numerous "beneficials" or CRW beetles) but would not suffer major yield losses (thus effectively resisting damage from "pests" or CRW larvae). Identifying desirable hybrids for refugia would improve the effectiveness of the refuge strategy for Bt-based CRW management and would provide farmers with the information to choose hybrids that are highly productive in refuge plantings.

8. Objectives:

1. Evaluate corn hybrids that are widely grown in New York to assess larval damage by CRW, adult beetle emergence, and yield under CRW-infested and non-infested conditions.
2. Identify hybrids that fit the desired profile for refuge plantings and share this information with seed industry personnel and farmers.
3. Project evaluation.

9. Procedures:

Nine corn hybrids that are widely grown in central New York were identified in consultation with seed industry representatives. These hybrids and a known susceptible check hybrid were planted in a three-rep split-plot trial on 17 June 2003 (notification of project funding was not received until early June, and a few weeks were then required to identify appropriate hybrids, find appropriate field space, and line up insects for use in infestations, resulting in the late planting date). Main plots consisted of CRW-infested and non-infested treatments (a field not planted to corn in the previous year was used, to avoid natural CRW infestation of the non-infested treatment). Hybrids were planted in sub-plots consisting of two 17.5′ rows at 30″ row spacing, and thinned to a final density of 28,000 plants/acre. On 26 June 2003, the CRW-infested treatments were artificially infested with 600 CRW eggs per foot of row, injected on either side of the row. The injectors were driven through non-infested plots also without applying any CRW eggs, to control for any effects of the injection process itself. In infested treatments, three competitive plants from each plot were rated for CRW root feeding damage at the late whorl stage using a standard 1-6 rating scale (1 = no damage, 6 = three or more nodes of roots entirely eaten off). Another three plants were rated for root regrowth in late September using a standard 0-4 scale (0 = no new growth on the node above the damaged node, 4 = en entire node of roots grown above the damaged node and more roots or nodes grown above this node). Cages were installed and counts made of CRW adult emergence from all plots throughout the adult emergence period (14 August through 26 September). Harvest and yield data could not be collected due to the very late planting date during this first project year.

CRW root damage and root regrowth ratings and adult beetle emergence were analyzed and summarized for each hybrid to obtain first year information on hybrids that combine: (a) relatively high emerging adult beetle populations and (b) minimal CRW root damage and/or excellent root regrowth. These hybrids may be good choices for refuge plantings.

10. Results and Discussion:

Due to the late start date for this project, we were searching for seed late in the season and the only seed we could obtain of our susceptible check, Cornell 281, was extremely old and did not germinate. Thus we do not have comparisons with a susceptible check for this first project year. Also, as noted above, harvest and yield data could not be collected as the hybrids did not dry down in time due to late planting. Despite these complications, some interesting results were obtained.

Analysis of variance for root damage ratings, root regrowth, and number of adults emerged for infested plots showed significant variation among entries for regrowth ($P \leq 0.05$) and number of adults emerged ($P \leq 0.10$) (Table 1). The hybrids in the top group for regrowth ratings were Pioneer 36B08 and Away 4717 (Table 2). Mean numbers of corn rootworm adults emerged in the infested plots were particularly high for H7298RR and intermediate for Pioneer 36B08 (Table 2). This single year of data suggests that Pioneer 36B08 might be particularly good as a corn rootworm refuge hybrid due to the combination of relatively high numbers of adults emerging from this hybrid and strong root regrowth that it exhibited. Additional years of evaluation are required before drawing any firm conclusion about this or the other hybrids tested.

Table 1. Results from analysis of variance for rootworm damage ratings, root regrowth ratings, and number of adult rootworms emerged for nine corn hybrids infested with western corn rootworm at Aurora, NY, 2003.

		Mean Squares from Analysis of Variance		
Source of Variation	DF	Rootworm Damage Rating (1-6 scale)	Root Regrowth Rating (0-4 scale)	No. Adults Emerged (no. per cage)
Replications	2	1.56	1.02	1036 **
Varieties	8	0.39	0.64 **	476 *
Error	16	0.49	0.25	223

*, ** Significant at $P \leq 0.10$ and $P \leq 0.05$, respectively.

Table 2. Means for rootworm damage ratings, root regrowth ratings, and number of adult rootworms emerged from nine corn hybrids infested with western corn rootworm at Aurora, NY, 2003.

Variety	Rootworm Damage Rating (1-6 scale)	Root Regrowth Rating (0-4 scale)	No. Adults Emerged (no. per cage)
Pioneer 36B08	2.7	3.6	19.7
Agway 4717	3.1	3.0	7.0
Pioneer 38A24	2.2	2.6	8.7
DKC-46-28-RR	2.9	2.4	11.7
H7298RR	2.8	2.4	44.3
Agway 5206	3.3	2.3	18.0
EX26621Bt	2.7	2.3	1.0
H7900	2.3	2.1	7.3
DKC-53-33-RR	3.2	2.1	18.0
CV (%)	25.0	19.6	99.0
LSD (0.05)	1.2	0.9	25.8

Numbers of adults emerged were actually counted on both infested and non-infested plots, and this entire data set was analyzed as a split plot design. Results showed no significant differences due to infestation or variety (data not shown). This was not expected, but probably reflected the surprising fact that rootworm adults emerged in the non-infested plots. This may have resulted from some leaking of eggs after the infester was turned off and as non-infested plots were driven through. Alternatively, these adults may have emerged from rootworm eggs laid in the field during the previous season (although the field used was previously planted to small grains so this seems unlikely). Finally, it is possible that rootworm adults entered the cages after emerging from the soil outside the area of the cage. The seal between the cage and the soil was very good when the cages were installed, but with the extreme changes in soil moisture during the sampling period the soil may have dried and pulled away from the base of the cage in some cases. We monitored this and repaired the seal as soon as we noticed it happening. This latter explanation also seems unlikely, as the adults in non-infested treatments emerged throughout the period of sampling, rather than all on the early sampling dates when we might not have realized that some cages had poor seals. For subsequent years, we are considering several measures to better control rootworm presence in non-infested plots, including possible application of soil insecticide and thoroughly cleaning out the infesting equipment between infested and non-infested treatments.

Information from this first project year provides some interesting preliminary data suggesting that the corn hybrids tested may vary in their utility as corn rootworm refuges. It will require additional years of testing to verify the results from this year's rootworm screening, and yield data will be needed to complete this initial data set. Optimal refuge hybrids could increase farmers' profitability from their refuges, reduce the incentive to use insecticide in corn rootworm refuge plantings, and promote the stability of genetically engineered rootworm resistance in corn.