1997

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Ostlie, K. R.; Hutchison, W. D.; and Hellmich, R. L., "Bt corn and European corn borer" (1997). *Faculty Publications: Department of Entomology*. 597.
https://digitalcommons.unl.edu/entomologyfacpub/597

http://www.extension.umn.edu/agriculture/corn/pest-management/bt-corn-and-european-corn-borer/

North Central Regional Publication 602, 1997

Bt corn and European corn borer

Long-term success through resistance management

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The editors express their appreciation to J.F. Witkowski for hosting key discussions on publication content. We greatly acknowledge the encouragement and support provided by R.A. Higgins (current chair), and E.E. Ortman, (Administrative Advisor), NC-205.

Dedication

In recognition of his lifelong research contributions to improved understanding of European corn borer ecology and management, the authors respectfully dedicate this inter-regional bulletin to W.B. (Bill) Showers, emeritus member, NC-205 research committee.

"Bt Corn and European Corn Borer" was co-authored and sponsored by members of the North Central Regional Technical Committee on "Ecology and Management of the European Corn

Borer and Other Stalk-Boring Lepidop-tera," NC-205, including: Experiment Stations and Extension Services of Colorado, Delaware, Illinois, Indiana, Iowa, Kansas, Maryland, Massachusetts, Michigan, Minnesota, Missouri, Nebraska, New York, North Carolina, North Dakota, Ohio, Pennsylvania, South Carolina, South Dakota and Wisconsin, and the USDA Agricultural Research Service, Ames, Iowa.

Acknowledgments

Individuals from the following companies and U.S. government agencies provided invaluable input during multiple reviews of this publication's content and technical accuracy: AgrEvo (Plant Genetic Systems), Monsanto Co., DowElanco, DEKALB Genetics Corp., Garst Seeds, Mycogen Seeds, Novartis Seeds (Ciba Seeds, Northrup King), Pioneer Hi-Bred International, Inc., US Environmental Protection Agency and USDA Cooperative States Research, Education and Extension Service. Our sincere appreciation for their ongoing contributions.

This publication represents a collaborative inter-regional effort to provide timely guidelines for growers, crop consultants, cooperative extension educators and industry personnel about how best to use Bt corn technology. Because of the dynamic nature of this technology, and ongoing research, this publication will be updated frequently. Updated versions will be available on the World Wide Web at the Northern Plains Crop Base at http://www.mnipm.umn.edu/

Bt Corn & European Corn Borer

Seed companies are now marketing Bt corn, one of the first tangible fruits of biotechnology that has practical implications for U.S. and Canadian corn farmers. Bt corn hybrids produce an insecticidal protein derived from the bacterium Bacillus thuringiensis, commonly called Bt. These hybrids provide protection against the European corn borer equal to, and usually far greater than, optimally timed insecticides. Rapid introduction of Bt corn hybrids creates uncertainty about the technology and new questions about its use. What is Bt corn? How is it made? How does it work? What is the best way to use it? Is it worth the added cost? This publication provides an overview of Bt corn, an innovative technology for managing European corn borer, and discusses how to use this technology for long-term profitability.

Why manage European corn borer?

European corn borer, Ostrinia nubilalis, is the most damaging insect pest of corn throughout the United States and Canada (Figs. 1 and 2). Losses resulting from European corn borer damage and control costs exceed $1 billion each year. For example, losses during a 1995 outbreak in Minnesota alone exceeded $285 million. A recent four-year study in Iowa indicated average losses near 13 bushels per acre in both first and second generations of European corn borer, for total losses of about 25 bushels per acre.

Despite consistent losses to European corn borer, many growers are reluctant to use current integrated pest management (IPM) methods for this pest. Historically, this reluctance stems from several factors:

- larval damage is hidden,
- heavy infestations are unpredictable,
- scouting multiple times each summer takes time and requires skill,
- insecticides are expensive and raise health or environmental concerns, and
- benefits of European corn borer management are uncertain.

One geographical exception, to the prevailing attitude of "benign neglect" toward European corn borer, occurs in the intensively managed irrigated corn of the high-plains states, such as Texas, western Kansas, eastern Colorado, and Nebraska. Irrigated corn with its higher yields is monitored closely for insect pests, such as European corn borer and the southwestern corn borer, and is treated frequently with insecticides. These farmers have a history of aggressive management of European corn borer.

Bt corn provides a new management tool for all corn producers. Ciba Seeds (now Novartis Seeds) and Mycogen Seeds introduced the first Bt corn hybrids in 1996. Several seed companies have incorporated this technology into their best inbred lines. Availability of Bt corn hybrids will increase dramatically as additional companies receive registrations and Bt corn seed production increases. Bt corn hybrids have one common feature. They each have a gene from Bacillus thuringiensis. Because these hybrids contain an exotic gene, they are commonly called transgenic plants. The Bt gene in these plants produces a protein that kills European corn borer larvae. Most larvae die after taking only a few bites. Consequently, Bt corn provides high levels of yield protection even during heavy infestations of European corn borer (Fig. 3).

What is Bt?

Bt is a naturally-occurring soilborne bacterium that is found worldwide. A unique feature of this bacterium is its production of crystal-like proteins that selectively kill specific groups of insects. These crystal proteins (Cry proteins) are insect stomach poisons that must be eaten to kill the insect. Once eaten, an insect's own digestive enzymes activate the toxic form of the protein. The Cry proteins bind to specific "receptors" on the intestinal lining and rupture the cells. Insects stop feeding within two hours of a first bite and, if enough toxin is eaten, die within two or three days (Fig. 4). For more than 30 years, various liquid and granular formulations of Bt have been used successfully against European corn borer and other insect pests on a variety of crops.

There are several strains of Bt, each with differing Cry proteins. Scientists have identified more than 60 Cry proteins. Proteins have been found with insecticidal activity against the Colorado potato beetle (for example, Cry3A, Cry3C), corn earworm (Cry1Ac, Cry1Ab), tobacco budworm (Cry1Ab) and European corn borer (Cry1Ab, Cry1Ac, Cry9C). Most of the Bt corn hybrids, targeted against European corn borer, produce only the Cry1Ab protein; a few produce the Cry1Ac protein or the Cry9C protein.

Why create Bt corn?

Although conventional Bt insecticides may perform as well as synthetic insecticides, their performance is not always consistent. Erratic performance of Bt insecticides is attributed to:

- toxin sensitivity to UV radiation, heat and desiccation,
- incomplete coverage of feeding sites, or
- reduced toxicity against older larvae.

Modifying a corn plant to produce its own Bt protein overcomes these liabilities. The protein is protected from rapid environmental degradation. Plants produce the protein in tissues where larvae feed, so coverage is not an issue. Finally, the protein is present whenever newly-hatched larvae try to feed, so the timing of Bt application is not a problem. The result is an efficient and consistent built-in system to deliver Bt proteins to the target pest (Figs. 4 and 5).

How is Bt corn created?

Plant geneticists create Bt corn by inserting selected exotic DNA into the corn plant's own DNA. DNA is the genetic material that controls expression of a plant's or animal's traits. Seed companies select elite hybrids for the Bt transformation in order to retain important agronomic qualities for yield, harvestability and disease resistance. Three primary components of the genetic package inserted into corn include:

- **Protein gene**. Bt genes, modified for improved expression in corn, produce Cry proteins. Initial Bt hybrids in the United States and Canada include one of three Cry proteins, Cry1Ab, Cry1Ac or Cry9C. Future hybrids may produce other Cry proteins, or proteins from other sources.
- **Promoter**. A promoter controls where and how much of the Cry protein a plant produces. Some promoters limit protein production to specific parts of the plant (for example, leaves, green tissue and pollen) whereas others produce protein throughout the plant.
- **Genetic marker**. The presence of a genetic marker allows seed companies to identify successful transformations. Current examples of markers include genes for herbicide resistance or antibiotic resistance.

This genetic package is inserted into corn through a variety of plant transformation techniques (Fig. 6). Successful transformations, called "events," vary in the components of the genetic package and where this DNA is inserted into the corn DNA. The insertion site may affect Bt protein production and could affect other plant functions. Consequently, seed companies carefully scrutinize transformation events to ensure adequate production of Bt protein and no negative effects on agronomic traits.

As of September 1997, the Environmental Protection Agency (EPA) has registered four unique events for commercial use: 176 (Novartis Seeds and Mycogen Seeds), BT11 (Northrup King/Novartis Seeds), MON810 (Monsanto) and DBT418 (DEKALB Genetics Corp.). Event 176 is trademarked as "KnockOut" by Novartis and "NatureGard" by Mycogen. Both the BT11 and MON810 events are trademarked as "YieldGard" and the DBT418 event is trademarked as "Bt-Xtra." Various seed companies license each event, so when purchasing seed, note which trademark is present on the seed tag and bag. The number of events is likely to increase rapidly. Understanding these events and how they affect performance is critical to the wise selection of corn hybrids.

Does Bt corn work against European corn borer?

Bt corn has the potential to improve European corn borer control dramatically, compared with current IPM options. Chemical insecticides, if timed well, typically provide control from 60-95% of first generation larvae and 40-80% of second generation larvae. As indicated in Fig. 7, performance of Bt corn can be dramatic when compared with non-Bt versions of the same hybrid. In field tests against natural and supplemented European corn borer infestations, Bt corn hybrids (regardless of event) provide more than 99% control of first generation European corn borer larvae in whorl-stage corn. However, the level of European corn borer control against late-season European corn borer infestations differs between Bt events. Under heavy European corn borer pressure, events BT11 and MON810 provide a higher level of control than event 176. Why? Event 176 hybrids produce Bt protein only in green tissues and pollen, whereas BT11 and MON810 events produce Bt protein throughout the plant. Because some hatching larvae initially colonize ears to feed on silks and developing kernels (Fig. 8), these larvae may survive on event 176 and may tunnel later into stalks and ear shanks.

Are Bt expression differences important? Presence of late-season European corn borer in ears in event 176 can be unsettling to growers, who expect complete control of European corn borer, and is a topic of debate in resistance management discussions. Control with event 176, nonetheless, is better than insecticide options.

Survival differences between events may be more critical where other late-season caterpillars (for example, southwestern corn borer and corn earworm) are serious pests. A case in point: Kansas State University trials during 1996 showed that BT11 and MON810 events provided 93% control of southwestern corn borer, while event 176 afforded only 19% control. In this situation the full-season Bt expression provided by the YieldGard™ events gave better control of southwestern corn borer.

The Bt gene is only one of several thousand genes that affect a hybrid's yield. Growers should carefully consider all the hybrid's characteristics, particularly its yield performance. Primarily, growers should select hybrids that have consistently performed well in yield trials, especially when corn borer pressure is low. Maturity, standability, harvest moisture and disease resistance are just a few examples of other traits that growers should consider when selecting hybrids.

How safe is Bt and Bt corn?

The EPA considered 20 years of human and animal safety data before registering Bt corn. Bt proteins are not toxic to people, domestic animals, fish, or wildlife; and they have no negative impacts on the environment. The Food and Drug Administration (FDA) exempts Bt Cry proteins from residue analyses because of Bt's history of safety and because these proteins degrade rapidly.

Are other insect pests or mites controlled by Bt corn?

European corn borer is not the only insect pest attacking corn (Fig. 9). Will Bt control other corn insects? The following summary reflects known toxicity of current Bt proteins plus field and laboratory studies of Bt corn in Iowa and Kansas. Current Bt corn hybrids have virtually no activity on the following pests: aphids, spider mites, black cutworm, western bean cutworm and soil insects such as corn rootworms, wireworms, white grubs, seedcorn maggots and seedcorn beetles. Bt corn might suppress, but not control, foliar feeding pests, such as stalk borers and armyworm. Although some events have good activity on armyworm, fall armyworm and corn earworm, field studies on these pests are limited. Finally, Bt corn protection from the southwestern corn borer differs markedly between events (see discussion on Bt events). If southwestern corn borer, armyworm or corn earworm are consistent pests, ask about the availability of hybrids with specific activity on these pests. Future Bt corn hybrids may include new Bt proteins or other novel toxins that will control more pests.

Indirect impacts of Bt corn on pests might occur. For example, Bt corn does not have activity on spider mites. Yet a reduction in pyrethroid use for corn borers might minimize outbreaks of spider mites. Although puzzling at first, this makes sense when one considers that natural enemies of mites are not reduced by insecticide applications in Bt corn fields. In contrast, minor pests may become more predominant, such as the western bean cutworm in western Kansas, eastern Colorado and Nebraska. Beginning with the widespread use of foliar insecticides, this insect has been a minor pest of corn. Pest status of this insect could change, though, when foliar insecticides are reduced and if this insect is not controlled by current Bt events or natural enemies.

Does Bt corn affect beneficial insects and natural enemies?

Many studies have shown that Bt Cry proteins are highly selective in killing larvae of moths. Bt corn, however, does not affect beneficial insects including honey bees, lady beetles, green lacewing larvae, spiders, pirate bugs or parasitic wasps (Fig. 10). Indirect effects on natural enemies of European corn borer, however, could occur. Predators, parasites and pathogens of the corn borer might decline as corn borer populations decline. Refuge areas, discussed below, may moderate these indirect effects. Unfortunately, little data on the subject exists. Bt corn fits into and complements an integrated pest management approach to farming that includes conservation of biological control agents.

What are the economical benefits of Bt corn?

Bt corn technology is so new that performance data from research and extension entomologists are limited. Also, few studies have compared Bt corn with other management options. Many questions remain concerning benefits of Bt corn, and whether these benefits are worth the extra cost for seed.

An economic analysis of historical corn borer infestation and yield-loss data provides insight into the potential benefits of Bt corn. The following analysis examines the comparative performance of four different management strategies: do nothing, use insecticides based on scouting and economic thresholds (Fig. 11), plant Bt corn event 176, or plant Bt corn events BT11 or MON810. Net gain for each management strategy is calculated by subtracting management costs from expected benefits.

Expected benefits are based on the following assumptions:

- Bt corn (MON810, BT11) provides 96% average control of European corn borer larvae,
- Bt corn (event 176) provides 96% control of first generation corn borers and 75% control of second generation corn borers.
- Insecticides provide 80% and 67% control of first and second generation borers, respectively.
- Physiological losses average 5.5% and 2.8% for first and second generation tunnels, respectively.
- Corn yields average 123 bushels per acre nationally.
- Market price averaged $2.33 per bushel from 1991-1995.
- Bt hybrids have no yield penalties.
- Improved corn production will not affect corn price.

This study includes data on insecticide and Bt corn performance from several states.

Table 1. Projected value ($ per acre) of yield protection provided by Bt corn in southern Minnesota during endemic and outbreak infestations of European corn borer (K. Ostlie, B. Potter & D. Sreenivasam).

Corn Price ($/bushel)	**Infestation Level**	**Expected Yield (bushels per acre)**				
		100	**120**	**140**	**160**	**180**
3.40	Endemic	$8.74*	$11.22	$12.23	$14.96	$16.77
	Outbreak	$42.74	$47.88	$59.83	$68.37	$76.91
3.00	Endemic	$7.71	$9.90	$10.79	$13.20	$14.20
	Outbreak	$37.71	$42.25	$52.79	$60.33	$67.87
2.60	Endemic	$7.15	$8.58	$10.01	$11.44	$12.87
	Outbreak	$32.68	$39.21	$45.79	$52.29	$58.82
2.20	Endemic	$6.05	$7.26	$8.47	$9.68	$10.89
	Outbreak	$27.65	$33.18	$38.17	$44.25	$49.77
1.80	Endemic	$4.95	$5.94	$6.93	$7.92	$8.91
	Outbreak	$22.62	$27.15	$31.67	$37.20	$40.72

*Bt corn yield protection will differ among hybrids because hybrids vary in their tolerance to European corn borer infestations

Benign neglect of European corn borer costs U.S. growers about $6.57 and $12.90 per acre for first and second generation borers, respectively. An IPM approach, basing insecticide use on scouting and economic thresholds, was profitable against both first and second generation borers, $0.38 and $4.07 per acre, respectively (Fig. 12). Bt corn, however, offered much better economic advantage with returns of $2.79 per acre for all events against first generation borers. The 176 and BT11/MON810 events returned $5.74 and $8.72 per acre, respectively, against second generation borers.

Analysis of historical European corn borer damage in Minnesota from 1988 to 1995 gave similar results. Estimated yield protection by Bt corn was $5.61 and $11.63 for first and second generation European corn borer control, respectively. The projected benefits, totaling $17.24 per acre, significantly exceed the current price premium for Bt corn of $7 to $10 per acre. Both the national and Minnesota projections clearly suggest that Bt corn offers a sound economic return, under the assumptions listed above.

European corn borer populations fluctuate over the years and from one field to the next. Similarly, corn yields and market prices often are volatile. This variability raises concerns about fluctuations in yearly economic benefits of Bt corn. To illustrate this point, the risk of investing in Bt corn was scrutinized for southern Minnesota over an eight-year period 1988-1995. This period included three outbreak (high) years for European corn borer and five endemic (low) years. The average benefit for this period, $17.24 per acre, was very close to the national estimate of Bt value, but returns varied considerably between endemic and outbreak years (Table 1). During the endemic years, the yield protection offered by Bt corn barely covered the price premium for seed, currently $7 to $10 per acre. During outbreak years, yield savings were four to five times the added seed cost ($28 to $50 per acre). The bottom line: Do not expect an economic return every year or in every field. As with any type of natural resistance, Bt corn only delivers an economic benefit when European corn borer outbreaks occur. Unfortunately, no predictive tools for European corn borer outbreaks are currently available.

Yield data on Bt corn are available from university entomologists and agronomists, and seed companies, who are conducting studies in nearly every state that grows corn (Fig. 13). As mentioned previously, yield results will depend on Bt events, specific hybrids and European corn borer infestation levels.

Are there indirect benefits from Bt corn?

Bt corn reduces the European corn borer population in a field and, depending on prevalence of Bt corn in the area, influences the local European corn borer population. For example, if 50% of the corn acreage is planted to Bt corn, then the corn borer population in the area could be reduced by 50%. Conceptually, this population suppression should be greatest nearer Bt corn than farther away. Neighboring corn fields could experience reduced attack by European corn borer. Movement of adult moths during each generation will influence the area and magnitude of this neighborhood effect. Conceivably, planting non-Bt corn near Bt corn could be beneficial because European corn borer populations near Bt corn fields should be suppressed.

Indirect benefits may also occur through decreased incidence of corn disease. Bt corn reduces European corn borer tunnels that provide entryways for plant pathogens. Thus, stem rots and ear rots could be reduced along with mycotoxin production.

Fewer dropped ears with Bt corn will mean less volunteer corn in the following year's crop.

Can European corn borer develop resistance to Bt corn?

European corn borer may have the potential to develop resistance to Bt Cry proteins. Insects are known for their ability to rapidly develop resistance to certain insecticides. Resistance occurs particularly when insecticides are used repeatedly and at high concentrations. More than 500 species of insects and mites have developed resistance to insecticides and miticides. A recent Midwestern example in corn includes adult western corn rootworm resistance to Penncap-M in Nebraska. In addition, laboratory colonies of more than 15 different insect pests have developed resistance to Bt proteins, including Indian meal moth, tobacco budworm, beet armyworm, pink bollworm and Colorado potato beetle. Moreover, the diamondback moth, a worldwide pest of cole crops, has developed high levels of resistance to Bt insecticide in field populations in Hawaii and Florida.

Many factors contribute to the development of resistance. Some of these factors for the European corn borer include: predictions for widespread use of Bt corn, high season-long mortality, and two or more generations per year. Recent laboratory studies in Minnesota, Kansas and Delaware confirm that European corn borers (collected from Minnesota, Iowa and Kansas corn fields) can develop moderate levels of resistance to Bt insecticides or Bt Cry proteins (Fig. 14). Resistant European corn borer strains in these studies require 30-60 times more toxin (resistance ratio) to kill 50% of a test population of young borers compared with nonresistant European corn borer strains. This modest level of Bt resistance developed in relatively small lab populations after seven to nine generations of exposure. Although these results confirm the genetic potential of European corn borer to develop resistance, laboratory studies do not prove resistance will develop under field conditions. Bt corn and European corn borers in the field pose a dramatically different situation than larvae feeding on Bt insecticides in laboratory diet.

How does resistance develop?

Scenarios of resistance development by European corn borer are suggested by studies of insecticide resistance in many insects and by resistance to Bt insecticides by tobacco budworm and diamondback moth. In any population of European corn borers, a few of the borers will have two copies of genes for resistance (rr), some will have one copy of the gene (rs) and most will have none (ss). Resistance genes are likely to be rare. On Bt corn, European corn borer with one or more copies of resistance genes (rr or rs) could survive and produce more offspring (Fig. 15). Improved survival or reproductive success results in a "selective advantage." As the Bt corn acreage increases, and with it the proportion of the European corn borer population exposed to Bt corn, more larvae carrying resistance genes could survive to adulthood. The overall population of Bt-resistant individuals increases with each generation. At some point, control failure could occur with resistant larvae reaching infestation levels in Bt corn fields similar to levels found in non-Bt corn fields.

What are the implications of European corn borer developing resistance to Bt corn?

Growers and seed companies will face the primary impacts of European corn borer resistance to Bt corn. Initially, while seed companies and entomologists develop strategies for countering European corn borer resistance, producers in problem areas might lose the option to use Bt corn. Organic growers who rely on Bt insecticides also could lose a valuable management option in these areas. Resistance effects could be minor, though, if hybrids that express alternative Cry proteins are effective and if they are introduced rapidly into problem areas. European corn borers, however, could develop cross resistance to two or more of the Cry proteins. If entire groups of Cry proteins are neutralized by resistance development, growers could permanently lose Bt corn and Bt insecticides as valuable management tools. This loss would be unfortunate for organic growers and other producers who rely on Bt insecticides. In addition, the failure of a voluntary, proactive resistance management plan could create more regulatory pressure for future transgenic crop technologies. This could limit the use of a transgenic Bt approach for other high-value crops, such as sweet corn.

Can development of European corn borer resistance to Bt corn be managed?

The potential threat of resistance by European corn borer to Bt corn necessitates a management plan to delay or avoid the risk of resistance. Resistance management is a key element of good IPM practices. Consequently, the EPA has issued conditional registrations that require companies selling Bt corn to develop and carry out resistance management plans by 2001.

Resistance management in Bt corn is currently based on two complementary principles: high dose and refuge. Plant geneticists designed Bt corn to produce very high levels of Bt Cry proteins, much higher than levels found on corn treated with Bt insecticides. The intent is to kill all European corn borer larvae with no genes for resistance (ss), plus those with one copy of a resistance gene (rs). The assumption inherent in this resistance management approach is that Bt hybrids have achieved this high-dose objective. If a high-dose objective is not achieved, then corn borer larvae with one copy of a resistance gene may survive to adulthood and mate with other resistant moths. Most of the offspring from these matings would be resistant to Bt corn.

The second principle of the resistance management plan is the use of refuges. The purpose of the refuge is to provide a source of European corn borers, not exposed to Bt corn or Bt insecticides, that could mate with potential resistant moths emerging from nearby Bt corn. The goal is to produce an overwhelming number of susceptible moths to every resistant moth (Fig. 16). A refuge is any non-Bt host of European corn borer, including non-Bt corn, potatoes, sweet corn, cotton or native weeds that occur near Bt corn (within the same 1/2 section, 320 acres). The question is, "How large a refuge is needed to provide enough susceptible moths?" In any given year, approximately 20-30% of European corn borer larvae should not be exposed to Bt Cry proteins. This estimate is based on current knowledge of European corn borer biology, pesticide resistance studies and computer simulation models. To be effective, European corn borer moths must emerge from the refuge at the same time as resistant moths and be close enough to mate with resistant moths. Although some European corn borer moths can fly substantial distances, many moths fly less than a mile from their emergence site. Consequently, each farm should have one or more refuge areas next to Bt corn. Examples of possible refuge configurations are illustrated in Fig. 17.

The actual amount of refuge required will vary among regions, farms, and corn production systems. Always the goal is to prevent Bt protein exposure to 20-30% of the larval population. In continuous corn and corn-soybean rotations, the primary available refuge is non-Bt corn, so 20-30% of the corn acreage should be non-Bt corn. In continuous corn areas where European corn borers are typically sprayed with insecticides, the refuge should be increased to 40% to compensate for larval mortality. Where the total corn acreage is small and much of the local European corn borer population is associated with alternative hosts that do not contain Bt proteins, a smaller refuge may be suitable. This reduction in refuge size assumes that corn borers from alternative hosts emerge at similar times as corn borers from corn. When the proportion of the local European corn borer population that flows through non-Bt hosts is unknown, a refuge of 20-30% non-Bt corn may be the simplest and best insurance to delay resistance. For specific refuge recommendations, contact local extension entomologists.

Key Steps to Implementing a Resistance Management Plan for European Corn Borers

The following summary is based upon the principles outlined throughout this publication and assumes that a voluntary, proactive approach by growers will provide product stewardship for long-term yield benefits and profitability. Key steps toward implementing a resistance management plan include:

1. Use Bt corn hybrids in fields where the risk of severe European corn borer infestations warrants the price premium for seed.
2. Carefully record and mark where Bt and non-Bt corn hybrids are planted, so Bt corn performance can be monitored and non-Bt corn can be scouted, and if needed, treated with a non-Bt insecticide.
3. Plant non-Bt corn refuge(s) to protect 20-30% of the European corn borer larval populations from exposure to Bt Cry proteins. Plant non-Bt corn at a similar time and in close proximity to Bt corn. In corn-soybean production areas, where corn is the primary refuge, at least 20-30% of the corn acreage should be non-Bt corn. Where spraying of non-Bt corn is anticipated, increase the refuge size to 40%.
4. Continue to use an IPM approach for all pests, as Bt corn is just one tool for European corn borer management. Other insect pests such as cutworms, wireworms, white grubs, seedcorn maggots, seedcorn beetles, corn rootworms, aphids and mites are unaffected by Bt corn. When designing scouting priorities, consider that Bt corn performance against other caterpillars, such as southwestern corn borer, stalk borer, armyworm, corn earworm and fall armyworm, may vary among events.
5. Monitor Bt corn to verify European corn borer control for both first and later-season generations. Do not wait until harvest. All events should provide nearly complete control of European corn borer in whorl-stage corn whereas BT11 and MON810 events should provide high levels of late-season control. Normally seed lots contain a small percentage (typically less than 4%) of off-types that may produce less or no Cry protein. Considering variability between seed lots, investigate fields with leaf feeding damage in more than 4% of the plants. In BT11 and MON810 hybrids, investigate fields after silking where larvae, stalk tunnels or ear damage are found on more than 4% of the plants. If feeding damage occurs, investigate the cause. If needed, get help to identify feeding caterpillars. If European corn borer larvae or excessive damage are discovered, resistance to Bt corn is a possibility and the situation should be investigated. Verify from field records that Bt corn was planted where excessive damage or larvae are observed. Consult your grower's guide for the seed company's procedure for investigating suspected resistance cases. Notify seed company representatives and/or extension agents immediately if evidence indicates a performance problem.

Can resistance development be monitored?

Monitoring for the development of resistance to transgenic plants provides information essential to managing European corn borer resistance (Fig. 18). Monitoring is necessary to learn whether a field control failure resulted from resistance or other factors that might inhibit expression of the Bt Cry protein. The extent and distribution of resistant populations can be mapped so that alternative control strategies can be adopted in areas where resistance has become prevalent. Finally, detecting resistance may be possible before control failures occur, if monitoring techniques are sensitive enough to provide complete discrimination between resistant and susceptible individuals (See sidebar).

What is the best strategy for using Bt corn?

Insect control by Bt expression is only one trait that farmers need to consider in their selection of hybrids. Bt genes only protect the yield potential inherent in the hybrid. Because corn borer populations fluctuate, looking at hybrid performance over several years is important (Fig. 19). Expect yield protection with Bt hybrids when European corn borer infestations are heavy, and little to no yield protection when infestations are light. Sound preliminary choices might be Bt versions of commercial hybrids that are proven performers.

Like other plant-resistance strategies, the decision to purchase Bt corn seed is made before pest population levels are known. Unfortunately, predicting when and where heavy European corn borer infestations will occur is not possible. Producers should consider using Bt corn only in areas where the economic risk from European or southwestern corn borer justifies the price premium for Bt corn. Watch Bt corn fields closely. Learn about the level of European or southwestern corn borer protection achieved with Bt corn.

The best strategy for using Bt corn may be to protect fields likely to bear the heaviest brunt of European corn borer attack or fields with the highest yield potential. For example, in northern areas, the best choice may be earlier planted fields with their higher yield potential and heavier first generation European corn borer attack. Conversely, using Bt corn in later-planted, later-pollinated fields provides optimal protection against second-generation or late-season infestations. In southern areas where multiple generations of European corn borer can contribute to yield loss, protecting hybrids throughout the season may be desirable. Regional and local strategies for using Bt corn will become more refined as producers and agricultural scientists gain experience with the product. Future versions of this publication will bring together this knowledge.

Transgenic crops, such as Bt corn, are at the forefront of a revolution in pest management. The concept of managing insects by a simple seed choice is a powerful one. As with any new technology, Bt corn brings mixed feelings: excitement of using new technology, desire to know more about it, apprehension about its wise use, and uncertainty about its value. In the next few years, much will be learned about how to use this powerful new tool wisely. Hopefully this new approach toward corn borer management will not falter because of resistance problems. By working together, producers, seed companies, scientists and regulators can better ensure the longevity of Bt corn.

Glossary

Bacillus thuringiensis (Bt):

A naturally-occurring soil bacterium that occurs worldwide and produces a toxin specific to certain insects (e.g. moths, beetles, blackflies or mosquitoes).

Biotechnology:

The science and art of genetically modifying an organism's DNA, such that the transformed individuals can express new traits that enhance survival (e.g., insect or disease resistance, herbicide resistance) or modify quality (e.g., oil, amino acids).

Cry Proteins:

Any of several proteins that comprise the crystal found in spores of Bacillus thuringiensis. Activated by enzymes in the insect's midgut, these proteins attack the cells lining the gut, cause gut paralysis and subsequently kill the insect.

Deoxyribonucleic Acid (DNA):

Double-stranded molecule, consisting of paired nucleotide units grouped into genes and associated regulatory sequences. These genes serve as blueprints for protein construction from amino-acid building blocks.

Event:

Successful transformation of an organism by insertion of exotic genetic material (DNA). Events vary in the genetic package inserted into the organism and the particular place where this genetic package is inserted into the host DNA.

Expression:

Production of the desired trait (e.g., protein concentration) in a transgenic plant. Expression varies with the gene, its promoter and its insertion point in the host DNA.

Gene:

The basic unit of inheritance and diversity; a section of DNA that codes for a specific product (e.g., protein) or trait.

High-Dose Strategy:

An approach for minimizing the rapid selection for resistance to transgenic plants by using plants that produce Cry proteins at a concentration sufficient to kill all but the most resistant insects.

Host Plant Resistance:

Ability of a plant to avoid insect attack, kill attacking insects or tolerate their damage.

Integrated Pest Management (IPM):

A management approach that integrates multiple, complementary control tactics (e.g., biological control, crop rotation, host plant resistance, insecticides) to manage pests in a profitable, yet environmentally sound manner.

Lethal Concentration (LC):

Concentration at which a toxin kills a given percentage of the insect test group, e.g., the LC50 refers to the concentration necessary to kill 50% of the insect test group.

Marker:

A genetic flag or trait used to verify successful transformation, and to indirectly measure expression of inserted genes. For example, a gene used as a marker in BT11 confers tolerance to the herbicide Liberty[TM].

Mode-of-Action:

Mechanism by which a toxin kills an insect. For example, the mode-of-action of Bt is ingestion and disruption of cells lining the midgut.

Promoter:

A DNA sequence that regulates where, when, and to what degree, an associated gene is expressed.

Refuge:

An area planted to non-transgenic plants, e.g., non-Bt corn or alternative hosts for European corn borer, where susceptible pests can survive and produce a local population capable of inter-mating with any possible resistant survivors from Bt corn.

Registration:

Legal approval of pesticides and transgenic crops for use in the U.S. by EPA, after extensive review of toxicology to mammals, birds, fish and other non-target organisms, environmental fate, and health/safety issues and precautions.

Resistance:

The capacity of an organism to survive exposure to a toxin.

Resistance Management:

A proactive process of limiting or delaying resistance development in a pest population with a focus on preserving susceptible genes (individuals).

Resistance Ratio:

A measure of an insect population's resistance to a toxin, typically calculated by dividing the LC50 of the resistant population by the LC50 of a susceptible population.

Selection:

A natural or artificial process that results in survival and better reproductive success of some individuals over others. Selection results in genetic shifts if survivors are more likely to have particular inherited traits.

Transgenic:

An organism genetically altered by addition of foreign genetic material (DNA) from another organism into their own DNA.

Figure 1: Tunneling by European corn borer reduces ear size and test weight: the major cause of yield loss (K. Ostlie).

Figure 2: Stalk breakage from severe infestation of European corn borer; an obvious symptom (K. Ostlie).

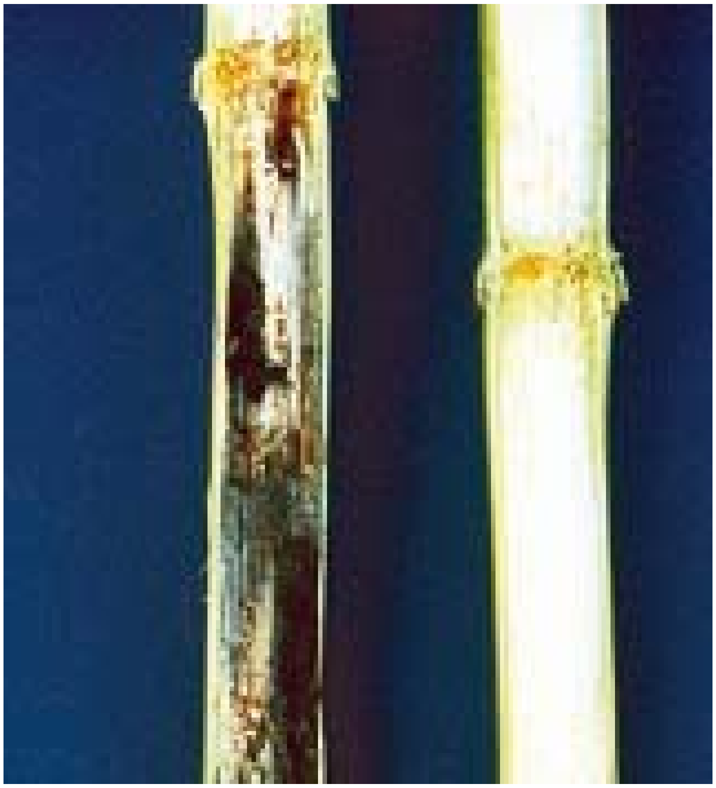

Figure 3: Stalk tunnels after European corn borer infestation: conventional hybrid (left) and Bt corn (right) (M. Rice).

Figure 4: Corn borers may eat a toxic dose of Cry proteins in just a few bites from a Bt corn leaf (K. Ostlie).

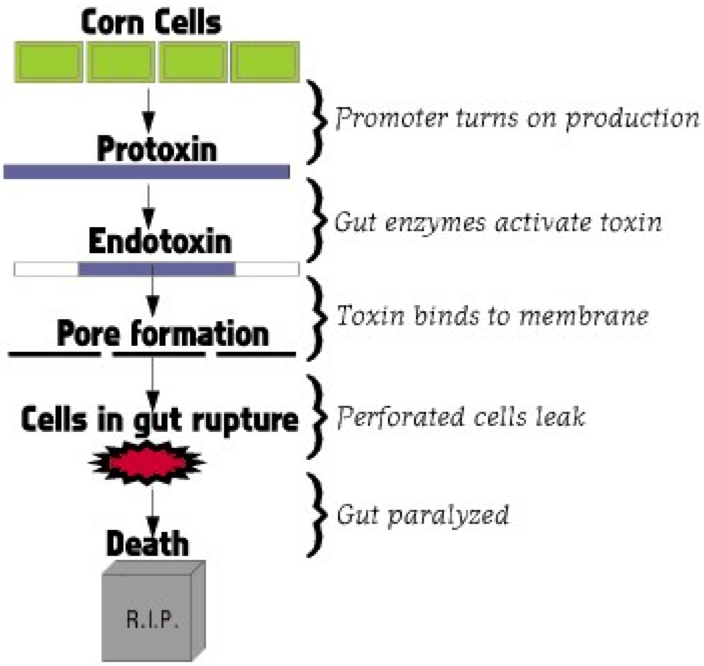

Figure 5: Bt mode of action after eaten by a European corn borer larva (K. Ostlie).

Figure 6: The "gene gun", one tool used to transform corn into Bt corn (Monsanto).

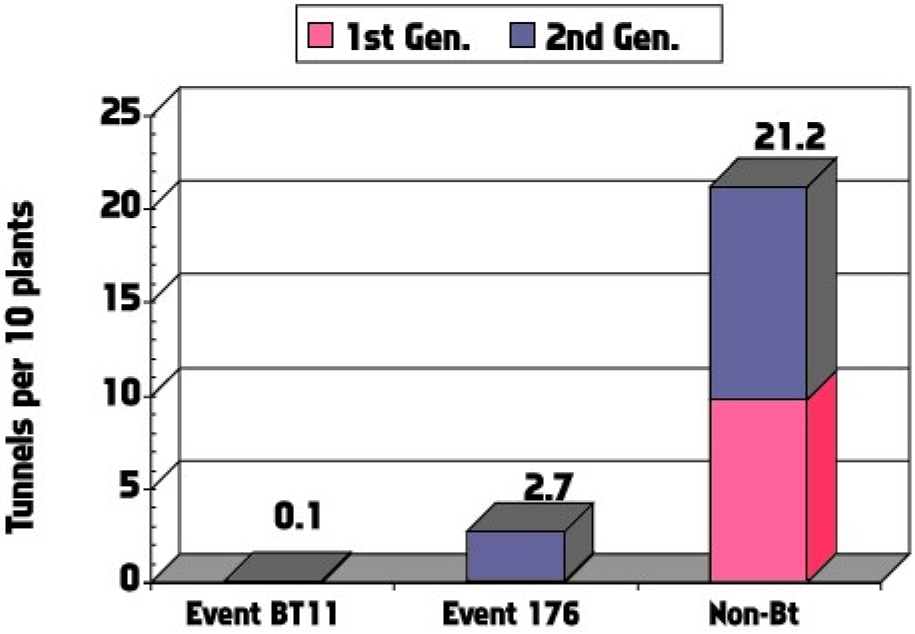

Figure 7: Representative performance of Bt corn hybrids, events 176 (KnockOut™), and BT11 (YieldGard™) (K. Ostlie, Minnesota, 1996).

Figure 8: Some late-season larvae may survive by feeding in the ear tips of Bt corn (event 176) (K. Ostlie).

Figure 9: Do not assume Bt corn controls all caterpillars that feed on corn! Scouting may be needed for these pests (from left to right):

- Black cutworm (M. Rice),
- Stalk borer (M. Rice),
- Armyworm (K. Ostlie),
- Corn earworm (W. Cranshaw).

Figure 10: Bt corn does not directly affect natural enemies of corn borers, such as the twelvespotted lady beetle (M. Rice).

Figure 11: Bt corn offers a management alternative to insecticide application with fewer environmental and safety concerns (K. Ostlie).

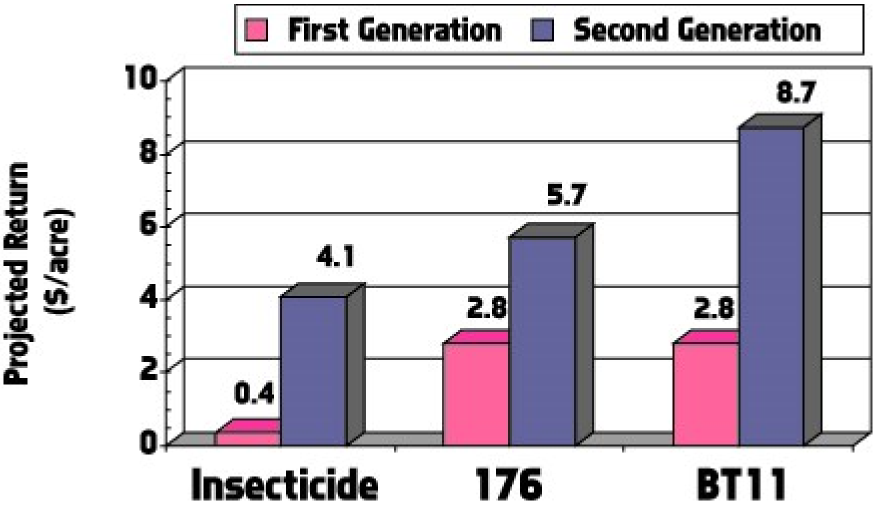

Figure 12: Expected net return of Bt corn and insecticide-based management strategies for European corn borer compared with a "do nothing" strategy (based on calculations by D. Calvin, 1995).

Figure 13: Yield performance of Bt corn will vary among hybrids, events. (A. EKhart).

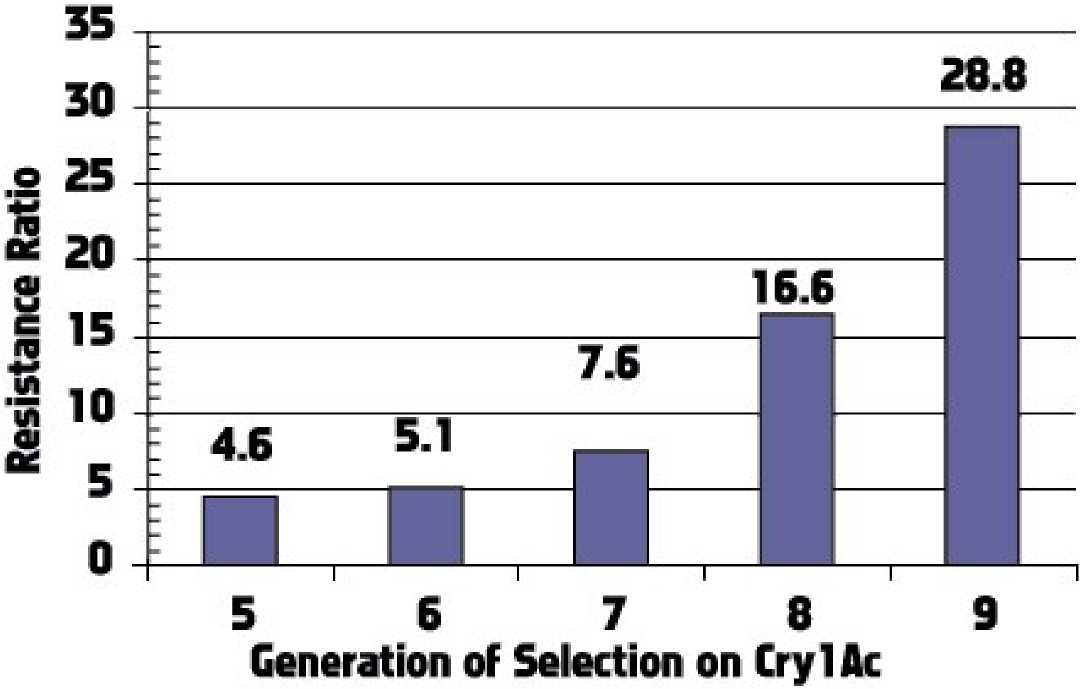

Figure 14: Resistance development to Bt in a Minnesota lab population of European corn borer (P. Bolin and W. Hutchison).

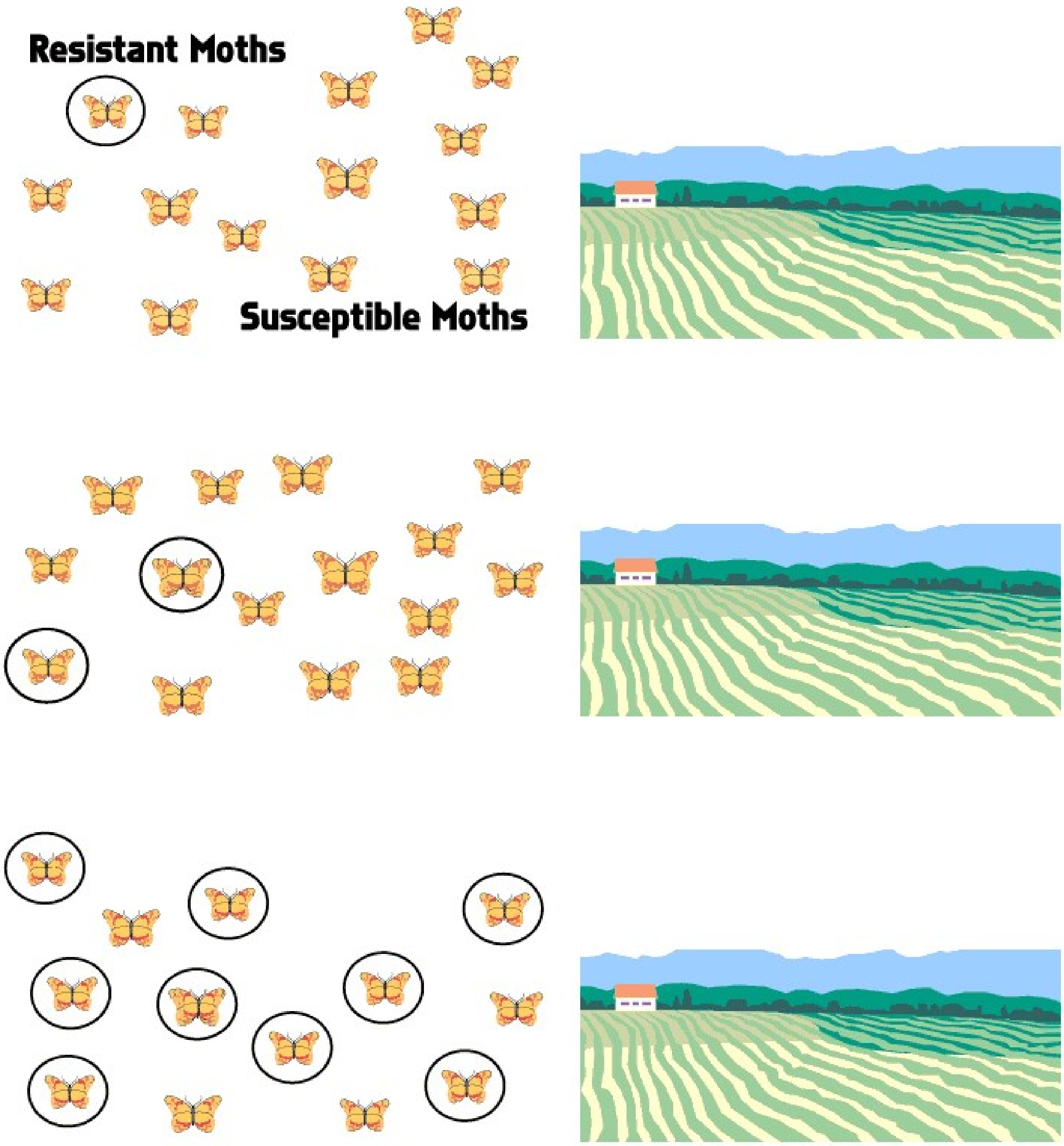

Figure 15: Schematic on resistance development (D. Bartels and W. Hutchison). From top to bottom:

1. Bt Corn Not Yet Available
2. Introduction of Bt Corn
3. Prevalent Use of Bt Corn

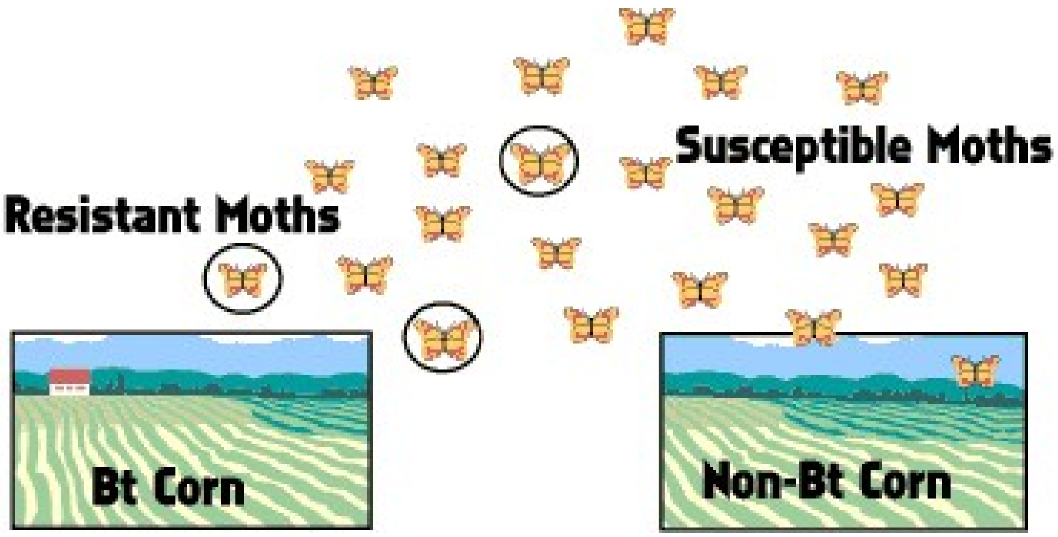

Figure 16: Mixing of resistant survivors from Bt corn with susceptible moths from refuges delays resistance (R. Hellmich).

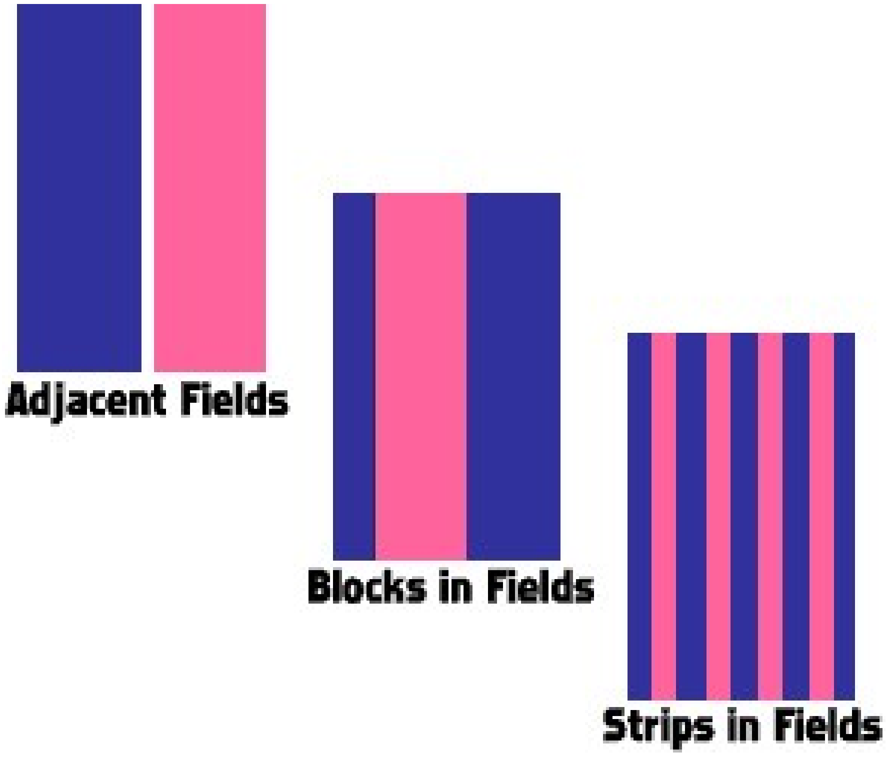

Figure 17: Options for refuge placement in proximity to Bt corn (K. Ostlie).

Figure 18: Monitoring performance in Bt corn fields provides an early warning of possible resistance problems (K. Ostlie).

Figure 19: Sound decisions on whether or not to invest in Bt corn require homework on hybrid performance and local risk from corn borer (Iowa State University Extension IPM program).

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This publication was coordinated by IDEA - Information Development * Expanding Awareness, a collaborative effort initiated by the North Central Cooperative Extension Services to increase the efficiency and effectiveness of developing, producing, and/or marketing educational products nationwide. Publications are subject to peer review and prepared as a part of Cooperative Extension Activities in cooperation with the Cooperative State Research Education and Extension Service (CSREES) - US Department of Agriculture, Washington, D.C.

Issued in furtherance of Cooperative Extension Work, Acts of May 8 and June 30, 1914, in cooperation with the U.S. Department of Agriculture. Katherine Fennelly, Director, University of Minnesota Extension Service, St. Paul, MN.

Supplement to: Bt Corn & European Corn Borer: Long-Term Success Through Resistance Management, NCR-602

Executive Summary

1. NC-205 is a regional research committee supported by Land Grant Universities, USDA-CSREES and ARS. It is comprised of scientists from 20 states, Mexico and Canada who have conducted research on stalk-boring pests since 1954.
2. The Committee re-examined many of the assumptions upon which our previous scientific assessments were based. This update http://ent.agri.umn.edu/ecb/nc205doc.htm summarizes our scientific understanding and recommendations for resistance management of Bt corn. Our initial recommendations were published in North Central Regional Publication 602 during 1997. An electronic version of NCR-602 is located at http://www.extension.umn.edu/Documents/D/C/DC7055.html.
3. The Committee reaffirmed, as a premise, the importance of prolonging the durability of Bt corn technology. Bt corn provides more effective and consistent control of European corn borer than insecticides, with less cost and fewer logistical, health, or environmental concerns. Bt corn has insurance value by reducing risk of yield loss from European corn borer.
4. We believe that resistance management using the high-dose/refuge strategy is possible. Recent data based on samples from three localities support a key assumption that major resistance genes are rare. Survival of resistant heterozygotes is assumed to be low. Additional data are needed to confirm both assumptions.
5. Providing susceptible mates for resistant survivors in the Bt crop is a crucial component of resistance management. A refuge of 20-30% of the larval population of European corn borer should be protected from exposure to Bt toxin on each farm. Recent data on non-random mating and regional genetic structure of European corn borer, coupled with new theoretical models, suggest that a 20% refuge is the minimum needed for resistance management. A 30% refuge provides a hedge for uncertainty in biological and operational assumptions.
6. Economic analyses suggest corn growers can benefit from planting refuges. Under plausible biological, genetic and economic conditions, and a 10-20 year planning horizon, economic models indicate that farmers capture most, if not all, of the benefits of Bt technology by planting 20-30% refuge.
7. A refuge of 20-30% of the larval population of European corn borer can be achieved by planting 20-30% of the corn on a farm to unsprayed non-Bt corn. This area should increase to 40% if the refuge is sprayed with insecticides. The non-Bt corn refuge should be planted within each 320-acre area that has Bt corn, at a similar time and with similar maturity characteristics as the nearby Bt corn.
8. Possible biological threats to successful resistance management are declines in toxin concentration early in the growing season, interactions between minor and major resistance genes, non-random mating or inbreeding of resistant individuals, and the effects of Bt corn on natural enemies of pests and other non-target organisms.
9. Until additional data are obtained, we suggest that these recommendations for European corn borer be applied in areas where other stalk-boring pests of corn occur.
10. Growers are key partners in managing insect resistance to Bt corn. Dissemination of consistent information to growers is essential.

Regional Research Committee, NC 205
October 1998

Introduction

NC-205 is a North Central Regional Research Committee supported by the Land Grant Universities and USDA-CSREES and ARS. This Committee has a long history (beginning in 1954) of addressing research issues on the "Ecology and Management of European Corn Borer and Other Stalk-Boring Lepidoptera." Participants in the program include representatives from 20 states, USDA-ARS, Mexico, and Canada. Collaborative efforts among members of the Committee have produced several hundred publications, including practical pest management guidelines for corn stalk-boring insects (Mason et al. 1996).

For the past three years, the NC-205 Committee has sponsored meetings with EPA and industry to discuss Bt corn, resistance management and associated issues. Recognizing the need of corn growers for more information, a publication entitled, ***Bt Corn & European Corn Borer: Long-Term Success Through Resistance Management*,** (North Central Regional Publication 602) was produced in 1997 with more than 35,000 copies distributed. An electronic version is located at http://www.extension.umn.edu/Documents/D/C/DC7055.html. The NC-205 Committee met September 24-25, 1998 reviewed NCR-602, shared current research results, and developed this statement. These findings will be periodically reviewed and updated as new data become available.

Bt corn provides more effective and consistent control of European corn borer than insecticides, with less cost than an insecticide application and fewer logistical, health, or environmental concerns. Bt corn has shown to farmers what years of educational efforts could not; local evidence that European corn borer significantly reduces yield. Furthermore, this technology has insurance value by reducing risk of European corn borer infestations, thereby improving yield stability. However, Bt corn involves season-long expression of a control measure that can be expected to produce intense selection for resistance to Bt toxin in the corn borer population. Left unmanaged, this evolutionary pressure could limit the value of Bt corn technology as a pest-management tool, just as it has frequently limited the value of chemical pesticides applied by traditional means.

NC-205 members recognize the benefit of prolonging the commercial usefulness of Bt toxins, including their traditional use as an organic pesticide, and the growers' need for information on the stewardship of Bt corn. As farmers gain experience with Bt corn, we believe they will have strong motivation to preserve the benefits of Bt technology. Grower surveys indicate a willingness to embrace resistance management recommendations that are logistically feasible. Incentive-based programs and consistent educational messages from academic, extension, industry, and regulatory sources should enhance the acceptance of sound management practices, including the adoption of non-Bt corn refuges.

In the NCR-602 document, our basic goal was to communicate information about Bt corn and resistance management. We recognized the need to provide susceptible mates for resistant survivors in the Bt crop as a crucial component of resistance management. We recommended protecting 20 to 30% of each local European corn borer larval population from exposure to Bt toxins. Since the publication of NCR-602, additional research results have refined our understanding of the factors that affect management of resistance to Bt corn.

The information used to develop our initial recommendations in NCR-602 included the best available biological data and theoretical models assessing the interaction between European corn borer and Bt corn. We have re-examined many of the assumptions on which our previous scientific assessment was based. In the following paragraphs, we summarize current information relative to the high-dose/refuge resistance management strategy.

High-Dose/Refuge Strategy

The high-dose/refuge strategy, which the Environmental Protection Agency (EPA) (1997, 1998) and industry (Fishhoff 1996) also have advocated, involves exposing one portion of the pest population to Bt plants with an extremely high concentration of toxin, while maintaining another part of the population in a refuge where the pests do not encounter any Bt toxin. By maintaining the refuges in close proximity to the Bt corn, susceptible pests that survive in the refuge are expected to intermingle and mate with any toxin-resistant pests that survive on the Bt corn plants. The offspring from these matings are assumed to be unable to survive on Bt corn. Population genetic theory (e.g., Tabashnik and Croft 1982; Gould 1986; Mallet and Porter 1992; Alstad and Andow 1995; Onstad and Gould 1998b; Caprio 1998) and experiments (Tabashnik 1994) predict that this approach will substantially delay resistance, if it is appropriately implemented and its assumptions are met.

High-Dose/Refuge Strategy Has Three Essential Assumptions.

1. Major resistance genes must be sufficiently rare so that nearly all such genes will be in heterozygous individuals. (A heterozygous individual has only one copy of the resistance gene and is referred to as a RS heterozygote). A gene frequency of less than one in 1,000 for major resistance genes is needed for the high-dose/refuge strategy to be successful.
2. Resistance genes must be nearly recessive. In other words, the RS heterozygotes should have very low survival on the Bt crop. RS survival rates that are less than 5% of the expected survival of homozygous RR resistant individuals on Bt corn are needed for the high-dose/refuge strategy to be successful. (For an operational definition of 'high-dose' refer to EPA 1998 at http://www.epa.gov/pesticides/SAP/finalfeb.pdf).
3. Non-Bt refuges are needed to provide a source of susceptible pests to mate with the resistant ones so that their offspring will be RS heterozygotes. This requires random mating within the typical dispersal distances of the adults.

While these are the three critical assumptions, most of the theoretical models also have assumed that the pest population exhibits local random mating and no regional genetic isolation. Recent data and models that have been used to evaluate these assumptions are described below. In summary, the high-dose/refuge strategy can substantially delay resistance if (1) the frequency of major resistance genes is low, (2) RS heterozygote survival is low, and (3) there is random mating of adults within typical dispersal distances.

Insect Resistance Management: Current Issues

- **Frequency of Resistance Genes Suggests that Resistance Management is Possible.** Successful resistance management requires that resistance genes be rare in the insect population. When we made our first recommendation, the initial frequency of resistance to Bt toxins in the European corn borer population was unknown. Based on estimates from other insects, we assumed that the initial frequency would be low and initial results of empirical studies support this assumption. We now know that the initial frequency of resistance genes is probably less than 10^{-3} in parts of Minnesota, Iowa, and Illinois (Andow et al. 1998; unpublished; Andow and Hutchison 1998; Hutchison et al., unpublished; Pierce et al. 1998;). Statistical techniques (Andow and Alstad 1998) applied to samples from Iowa and Minnesota give an expected frequency of major resistance alleles of 8.93×10^{-4}, and a 95% confidence interval of $[0, 4.38 \times 10^{-3}]$. Collectively, this information suggests that resistance management is still possible if effective refuges are employed. Estimates of resistance gene frequency may be needed from other corn-producing areas.

- **Will RS Heterozygote Survival Be Low Enough to Enable Resistance Management?** Survival of RS heterozygotes is still unknown because major resistance genes have not yet been found and characterized. This lack of knowledge requires us to make a critical assumption: that RS heterozygote survival is low enough to enable resistance management (i.e., is almost fully recessive). Two lines of indirect evidence suggest that RS survival is low despite this absence of direct evidence. Work on resistance to Bt toxin in other organisms has shown RS survival to be low, ranging from ~0 to 0.025 (Tabashnik et al. 1992; Gould et al. 1997; McGaughey 1985; McGaughey and Beeman 1988). In addition, because several searches for resistance in the field have yet to confirm resistant individuals in European corn borer populations (Pierce et al. 1998; Hutchison et al., unpublished), dominant major resistance genes may be rare. However, these searches have included only a minuscule part of the approximately 80 million acres of corn grown annually in the U.S. Additional research is needed to evaluate the assumption of low RS survival.

- **Significant Numbers of European Corn Borers Move Only Short Distances.** For the high-dose refuge strategy to be effective, refuge insects must mate with resistant insects surviving in the Bt corn. For this to occur, European corn borer moths must emerge from the refuge at the same time as resistant moths and be close enough to mate with resistant moths. Data from MN and NE indicate that significant numbers of European corn borers move only short distances under some conditions. Refuge corn adjacent to Bt corn sustains less borer damage up to 100 meters from the Bt corn, suggesting limited dispersal during the second flight (Alstad and Andow, unpublished). In a recent mark-release-recapture experiment conducted in Nebraska, almost all recaptures of unmated females were made within ca. 500 meters of the release point (Hunt et al., unpublished). Collectively, these data suggest limited adult European corn borer movement. To improve the probability of desired matings, NC-205 recommends that refuges should occur on each farm where Bt corn is planted and within each 320-acre area. In other corn production regions, where the landscape patterns differ, movement patterns of adult European corn borers also may be quite different.

- **European Corn Borer Populations Exhibit Local Non-Random Mating.** Non-random mating in local populations can lead to more rapid evolution of resistance, because RR homozygotes are more likely to mate with each other, fewer RS heterozygotes will be produced, and fewer resistance genes will be killed by Bt corn. Local non-random mating is measured by the ***Fis*** statistic (Wright 1965). When ***Fis*** = 0, local mating is random, and ***Fis*** > 0 implies that local populations contain fewer RS heterozygotes than expected under random mating. Electrophoretic analysis of three genetic markers revealed 15 of 45 European corn borer samples collected from 40 North American localities by NC-205 scientists to have ***Fis*** > 0.27 (NC-205, unpublished). With an initial R gene frequency of 10^{-4}, SS survival of 0, RS survival of 0.005, 20% refuge, 100% random mating, and no inbreeding, resistance is projected to evolve in 206 generations (Caprio 1998; Hutchison and Andow, in press). Holding the other parameters constant and either decreasing the population mating at random to 65% or increasing the inbreeding to 7%, and resistance evolves in <31 generations. A 30% refuge under these same conditions would extend the life of the technology to >45 generations. The observed values of ***Fis*** and ***Fst*** (below) suggest that the evolution of resistance will be faster than these simulations. Other studies have concluded that non-random mating can occur in local populations of European corn borer (Ni 1995, Mason unpublished).

- **European Corn Borer Populations Exhibit Regional Genetic Isolation.** Regional genetic isolation can lead to more rapid evolution of resistance (Peck et al. 1998; Caprio 1998). Resistance will develop faster in a subdivided population because resistance genes can become common in a sub-population by chance (random drift). Resistance genes from these isolated populations then could spread to other populations. Subdivision of a population is measured by ***Fst***. When ***Fst*** is 0, there is no subdivision, and when it is 0.2 there is substantial subdivision in the population. Model results suggest that when ***Fst*** > 0.05, resistance evolution occurs much more rapidly than when ***Fst*** < 0.02 (Caprio 1998). Recent empirical studies of the genetic structure (via electrophoretic analysis of enzymes) of European corn borer in North America found substantial regional genetic isolation. Forty-five samples provided by NC-205 cooperators from 40 North American localities show ***Fst*** values above 0.175 at three concordant genetic markers. These values demonstrate very high levels of genetic isolation (less than 1 migrant exchange per generation) between locations separated on average by 300 kilometers (NC-205, unpublished). This regional isolation could accelerate the rate of resistance evolution. Such a finding supports larger, rather than smaller, refuge proportions.

Economic Assessment

Economic analysis of refuge size can provide additional insight into resistance management considerations. The refuge size depends on the biological and genetic information discussed previously, along with the planning horizon. Under these conditions (initial r allele frequency = 0.0001, RS survival rate = 0.025, random mating, and no inbreeding) and a 10-20 year planning horizon, economic models suggest that farmers capture most, if not all, of the benefits of Bt technology by planting 20-30% refuge (Hurley et al., submitted). This model is sensitive to underlying biological and genetic uncertainties at low levels of refuge. Onstad and Guse (unpublished) found that with an initial r allele frequency of 0.0001-0.001, RS survival rates of 0.0-0.025, and a 15-20 year time horizon, a 20% refuge level was usually superior economically. In extreme cases of pest density and crop value one could project an effective refuge ranging from 10 to 30%.

Risk analysis shows that the cost to farmers of planting too much refuge is less than the cost of planting too little refuge. For example, under the conditions stated previously for the Hurley et al. model, and a 15-year planning horizon, increasing refuge from 10% to 20% is expected to decrease the value of the Bt technology by less than 1%, while reducing the probability of resistance developing from 47% to less than 1%. However, reducing refuge from 10% to 5% is expected to increase the value of the technology by less than 1%, while increasing the probability of resistance developing from 47% to 79% (Hurley et al., unpublished). Therefore, economics and uncertainties about important model parameters suggest larger rather than smaller refuges.

Refuge Recommendations

The scientific evidence suggests that sufficient refuges, properly placed in space and time, have high potential to delay European corn borer resistance to Bt corn. After considering the implications of this research, the Committee unanimously reaffirmed its previous recommendation that refuges should prevent Bt protein exposure to 20-30% of the European corn borer larval population.

Implementation Issues

- **Non-Bt Corn Refuge Size.** A refuge of 20-30% of the larval population of European corn borer can be achieved by planting 20-30% of the corn on a farm to unsprayed non-Bt corn. This non-Bt corn refuge should increase to 40% if the refuge will be sprayed with insecticides. The non-Bt corn should be planted within each 320-acre area that has Bt corn. The non-Bt corn refuge should be planted at a similar time and should exhibit similar maturity characteristics as the nearby Bt corn.

- **Bt Corn Protection Extends Into Adjacent Non-Bt Corn Refuges.** Damage from European corn borer was reduced in non-Bt corn adjacent to Bt corn. Under high borer pressure, up to 50% reduction in damage occurred in refuge corn within 5-10 meters of Bt corn. Damage increased gradually with distance from the Bt corn. Some reduction of damage continued out to 80 meters from the Bt corn, but was undetectable beyond 80 meters (Andow & Alstad, unpublished). Theoretical simulation models also predict this phenomenon (Alstad and Andow 1995; Onstad and Guse, unpublished). Refuge corn planted in narrow strips within a field of Bt-corn experiences less damage than blocks of refuge (Andow and Alstad, unpublished). Simulations show that refuge strips 6-12 rows wide are effective at delaying resistance and can provide similar economic return as a separate block refuge established adjacent to the Bt corn field (Onstad and Guse, unpublished). Consequently, by positioning the refuges near Bt corn, producers could extend the protection benefits of Bt corn into the refuge.

- **Non-Field Corn Refuges.**
  Sacrificial Refuges. High-density popcorn can produce substantial numbers of European corn borer, which could considerably reduce the percentage of land required to produce refuge insects (Hellmich, unpublished). The logistics and feasibility of this strategy have not been investigated.

  Weeds, natural vegetation, and alternative crops. Many plants serve as aggregation areas and hosts for European corn borer and may provide refuges to conserve susceptibility in certain geographic areas (Hellmich et al. 1998). However, it is unknown whether these habitats will produce enough unselected individuals at the right time and whether their proximity to Bt corn allows for random mating. Until the contributions of these alternative hosts as refuges are known, refuge recommendations are being based solely on non-Bt corn (Hellmich, unpublished; Whalen et al., unpublished; Dively, unpublished; Losey et al., unpublished).

- **Impacts on Natural Enemies and Other Non-Target Organisms.** Recommendations regarding the size and distribution of non-Bt corn refuges have been made primarily to preserve susceptibility of the pest insects to Bt-toxins. Less attention has been paid to the potential effects of Bt corn on natural enemies in agricultural ecosystems (Orr and Landis 1997, Pilcher et al. 1997) and of the effects on other non-target organisms.

  Because of the extensive acreage that may be planted to Bt corn in the near future, this technology has potential to have widespread and lasting impacts on beneficial insects. One concern involves the effect of substantial local or regional declines in the natural enemy prey base that could result from widespread adoption of Bt corn. Additionally, direct Bt toxicity to natural enemies has recently been suggested (Hilbeck et al. 1998a, 1998b). These effects could ripple through other crops and habitats in unpredictable ways. While it is unclear if 20-30% refuge is sufficient to mitigate negative impacts on natural enemies in the long term, in the short-term, refuges of at least this size are prudent. A significant refuge should minimize negative impacts on beneficial insects that control other pests.

- **New Genes and Gene Combinations.** New transgenic technologies, including gene stacks, introduction of other Bt toxins, and registration of novel toxins are under development and their resistance management implications need to be evaluated. When based on other Bt toxins, cross-resistance is an important issue (Bolin 1998). Cross-resistance among Bt toxins that share a common binding receptor is well documented, especially among those classed as Cry1 toxins (McGaughey and Oppert 1998). Novel non-Bt toxins may interact with Bt receptors, and each technology will need examination for resistance management implications. Gene stacks with other pest management traits (e.g., herbicide resistance) need to be examined for impacts on European corn borer resistance management. The commercial availability and viability of these new technologies is unknown, which reinforces concerns about durability of existing strategies.

- **Incentive-Based Options.** A voluntary insurance program or discounts on the purchase of non-Bt seed might help "level" the perceived differences in economic returns associated with Bt-corn net revenue and non-Bt corn refuge acres.

- **Southwestern Corn Borer and Other Stalk Borers.** Southwestern corn borer, southern cornstalk borer, and (common) stalk borer also attack corn in parts of the U.S. Our biological information on these borers is limited. We recognize that there could be many differences between other borers and European corn borer that will influence insect resistance management strategies. Until we have additional information on how the relevant parameters are affected, we suggest that recommendations developed for European corn borer should be employed in areas where these other borers occur. We recognize that the sprayed refuge option is more likely to be used in areas infested with southwestern corn borer because of the serious losses associated with this insect.

- **Education.** Growers are key partners in resistance management. Dissemination of information to growers is essential for effective implementation and extending the durability of the technology. Economic and scientific reasons must be combined with practical deployment strategies for this educational message (particularly the refuge component) to be embraced widely by growers (Rice and Ostlie 1997; Rice and Pilcher 1998). Revision of NCR-602 is underway to reflect new events, and new information on Bt corn performance and resistance management.

Potential Threats to the High-dose Strategy.

- **Changes in toxin concentration throughout the growing season.** All current and future transgenic hybrids should be measured for toxin concentration throughout the season under a wide range of environmental conditions (e.g., soil, weather, irrigation). Toxin concentrations can decline after pollen shed in some Bt hybrids (Walker 1998), jeopardizing the high-dose strategy (Onstad and Gould 1998a).

- **Minor Bt-Resistance Traits are Common in European Corn Borer.** Laboratory selection programs for Bt resistance have shown increases in Bt tolerance of 20 to 80 fold (Huang et al. 1997; Keil et al. 1997; Bolin 1998; Keil and Mason, unpublished). These results demonstrate that minor resistance genes are common enough to be included in all of the original selection stocks, and there is substantial genetic variability for resistance in wild European corn borer populations. To date, however, survival of selected strains has not been documented on transgenic Bt corn hybrids. When major resistance genes are found, they are likely to occur in populations and genotypic combinations with minor traits that may increase their relative dominance, threatening the high-dose strategy (Alstad and Andow 1996).

Conclusions

Collectively, the new scientific information reinforces the basic principles of our 1997 resistance management statement. The premise of the NC-205 position is to prolong the practical benefits associated with Bt transgenic corn technology. We support a high-dose/refuge strategy for management of resistance to Bt corn. Based on current data, modeling, and scientific interpretation, these recommendations are that under a high-dose/refuge strategy, refuges should protect 20 to 30% of the European corn borer larval population from exposure to Bt toxins. In a practical sense, this suggests 20 to 30% of the corn acreage should be planted to non-Bt corn. This non-Bt percentage should be increased to 40% if the refuge will be sprayed. We also recommend refuges and Bt plantings be established in close proximity, such that the refuge always occurs within the same half-section (320 acres) wherein Bt corn is planted.

In summary, we find the scientific evidence leads us to reaffirm our 1997 recommendations. As additional information becomes available, we will continue to reassess these recommendations.

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