Bt Hybrids and Resistance Management Considerations – Experimental Evaluation of Multi-Gene Insect Control Products

Illinois Corn Breeders’ School
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University of Illinois, Urbana-Champaign
We have a transgenic landscape in the corn and soybean production system.
USDA – Economic Research Service Estimates of Stacked Gene Varieties – IL and USA

Average U.S. Corn Seed Costs – Projected for 2010 to be $82.50 per acre: NPK Fertilizer Advisory Service

% of all Corn Planted

Years

2000 2001 2002 2003 2004 2005 2006 2007 2008 2009

Illinois
USA
USDA – Economic Research Service Estimates of Genetically Engineered Corn Plantings – IL and USA

% of all Corn Planted

<table>
<thead>
<tr>
<th>Years</th>
<th>Illinois</th>
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<td>2009</td>
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Did you plant a Bt hybrid in 2009?
- Yes – 97.06% (n=132), No – 2.94% (n=4)

In 2009, did you plant a Bt hybrid for CRW or ECB control knowing that anticipated damage levels were low?
- Yes – 80.33% (n=98), No – 19.67% (n=24)

Did you have access to elite (high yield potential) non-Bt corn germplasm in 2009?
- Yes – 59.26% (n=80), No – 40.74% (n=55)

Would you be willing to use a seed blend (Bt and non-Bt) as a refuge?
- Yes – 79.14% (n=129), No – 20.86% (n=34)

If you answered “Yes,” would you be willing to use a seed blend that contains non-Bt seed in the 2 to 5% range?
- Yes – 88.41% (n=122), No – 11.59% (n=16)

If you answered “Yes,” would you be willing to use a seed blend that contains non-Bt seed in the 6 to 10% range?
- Yes – 57.86% (n=81), No – 42.14% (n=59)
Transgenic Bt Corn Rootworm Products

- Agrisure RW – mCry3A
- Agrisure ECB/RW – Cry1Ab + mCry3A
- Herculex RW – Cry34Ab1/Cry35Ab1
- Herculex XTRA – Cry1F + Cry34Ab1/Cry35Ab1
- YieldGard RW – Cry3Bb1
- YieldGard Plus – Cry1Ab + Cry3Bb1
- YieldGard VT RW – Cry3Bb1
- YieldGard VT Triple – Cry1Ab + Cry3Bb1
- YieldGard VT Triple Pro – Cry3Bb1 + Cry1A.105 + Cry2Ab2
SmartStax™ Hybrids

- Event MON88017 – Cry3Bb1
- Event MON89034 – Cry1A.105 + Cry2Ab2
- Event DAS-59122-7 – Cry34/35Ab1
- Event TC 1507 – Cry1F
- Glyphosate tolerance
- Glufosinate tolerance
Gene Pyramiding with Seed Mixtures – The Future of IRM Programs and Insect Management?

Plants expressing insecticidal proteins

Insecticidal proteins kill insect larva by distinct mechanisms

Reduced selection pressure

doi:10.1038/nbt1003-1152
**SmartStax (Monsanto Company) Trial**

**Urbana, Illinois, 2008**

Planting 5/22  
Root evaluation 8/1

*Avg. node injury rating significantly greater than avg. node-injury ratings for all other treatments.*
SmartStax (Monsanto Company) Trial
Urbana, Illinois, 2008

*Avg. node injury ratings with the same lower-case letter are not significantly different.
Gene Pyramiding: Will this Sustain Bt Durability?

  - Greenhouse experiment with transgenic broccoli plants and diamondback moths with resistance genes to Cry1Ac and Cry1C proteins
  - After 24 generations of exposure to Bt broccoli, resistance to pyramided plants was delayed compared with plants expressing a single Cry protein

“Our experiments showed that allowing the concurrent release of cultivars with the two Bt genes in separate plants, each with one Bt gene, is not the best way to delay resistance. Even sequential release would result in control failure of at least one cultivar sooner than if pyramided varieties were used.”
Gene Pyramiding: Will this Sustain Bt Durability?

- In 2002, pyramided cotton (Bollgard II®) that express Cry1Ac and Cry2Ab2 was approved in Australia and the United States.
  - These “stacked” cotton plants have proven very effective.
  - The Cry proteins have different binding sites in the insect midgut.
  - Key to longterm durability of pyramided transgenic plants is the absence of cross resistance.
Gene Pyramiding: Will this Sustain Bt Durability?

- Most instances of resistance to Cry proteins is related to changes in receptor binding sites within the midgut of insects
- Resistance also can potentially develop if rare individuals are able to alter the metabolism of Cry proteins
- An integration of resistance management tactics (mosaic of refuges in the landscape) with pyramided plants is necessary
- Implementation of IPM tactics along with Bt hybrids is recommended (use of scouting and economic thresholds to influence the temporal and spatial deployment of Bt hybrids)
Corn and soybean producers in the North Central Region of the United States are increasingly relying upon the use of two neonicotinoid insecticides applied as seed treatments: thiamethoxam (Cruiser ®) and clothianidin (Poncho®).
All Bt (transgenic corn hybrids) have seed treated with a low rate (0.25 mg a.i./seed) of a neonicotinoid product (clothianidin or thiamethoxam). The refuge (non-Bt hybrid) may be planted to seed treated with the higher rate (1.25 mg a.i./seed) of the same neonicotinoid insecticide.
Percentage of Plants Infested in Farmers’ Fields Over a 10-Year Period

Bigger and Petty (1965)
Corn Rootworm Control Trial
Perry, Illinois, 2009

Average node-injury rating

<table>
<thead>
<tr>
<th>Planting 4/23</th>
<th>Root evaluation 7/20</th>
<th>LSD($P=0.05$) = 0.15</th>
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Corn Rootworm Control Trial
Perry, Illinois, 2009

Planting 4/23
Root evaluation 7/20

LSD(P=0.10) = 19
Standard Corn Rootworm Trial, Perry, 2009

\[ R^2 = 0.003 \]
Corn Rootworm Control Trial
Urbana, Illinois, 2009

Average node-injury rating

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Corn Rootworm Control Trial
Urbana, Illinois, 2009

Planting 4/18
Root evaluation 7/22

LSD(\(P=0.10\)) = 25
Standard Corn Rootworm Trial, Urbana, 2009

\[ R^2 = 0.72 \]
Are Bt hybrids for corn rootworm control bullet proof?

What does resistance look like?
Significant rootworm injury to the roots of a YieldGard VT hybrid from the DeKalb site in 2008. (University of Illinois)
Significant rootworm injury to the roots of an HxXTRA hybrid from the DeKalb site in 2008. (University of Illinois)

- Hybrid 1 – V4 (60.21 ppm); V9 (49.34 ppm)
- Hybrid 2 – V4 (61.98 ppm); V9 (40.5 ppm)
- Hybrid 3 – V4 (69.67 ppm); V9 (42.88 ppm)
- Hybrid 4 – V4 (75.17 ppm); V9 (42.33 ppm)
- Hybrid 5 – V4 (80.67 ppm); V9 (45.81 ppm)
YGRW Hybrid Evaluation Trial
Planting Date (May 5, 2006) Urbana, IL 2006

Average Root Rating

Evaluation Dates

control

Failed to meet commercial standards

20-Jul
7-Aug

Gray et al. 2007. J. Appl. Entomol. 131: 386-390
Field Evolved Resistance to Bt Crops

- Resistance to Cry1Ac – some field populations of cotton bollworms (*Helicoverpa zea*) in Arkansas and Mississippi

- Bt cotton producing the Cry1Ac protein did not meet the high-dose standard for *H. zea*.

- “… dominant inheritance of resistance to Cry1Ac appears to have hastened the evolution of resistance in *H. zea*.”

- “The hybrid progeny produced by matings between a laboratory-selected resistant strain and a susceptible strain of *H. zea* were resistant to Cry1Ac ..”

Field Evolved Resistance to Bt Corn

- Maize stalk borer (Busseola fusca) resistance to the Cry1Ab protein in transgenic corn grown in South Africa
- Fall armyworm (Spodoptera frugiperda) resistance to the Cry1F protein in transgenic corn produced in Puerto Rico

“Although published data are limited for these instances, failure to achieve the high-dose standard and to implement adequate refuges may have hastened resistance. To maximize knowledge gained from these and other cases, it is important for scientists in academia, industry, and government to make publicly available the relevant information on the efficacy of transgenic crops and refuge abundance.”

Evolution of resistance to Cry3Bb1 reported within three generations of greenhouse selection – larval survival of 25%.

After three generations, LC$_{50}$ of constant-exposure colony was 22-fold greater than LC$_{50}$ of control colony.

After six generations – percent survival on Bt corn compared with isoline was 11.7-fold greater in the field experiment for constant-exposure colony vs. control colony.
Summary Comments

- The use of neonicotinoids will continue to escalate in the corn and soybean agroecosystem as the demand for transgenic crops increases.
- At present no IRM plan is in place for the use of neonicotinoids in corn or soybeans – no plan is anticipated.
- Resistance to this class of chemistry has been documented previously.
- The prospects for resistance development to neonicotinoids and/or Bt, particularly by western corn rootworms, seems likely.
- Producers will increasingly manage insect pests of corn using an “insurance” based approach (use of Bt hybrids) rather than rely on traditional (scouting and use of economic thresholds) IPM approaches.
Summary Comments

Integration will increasingly come to mean use of multiple pyramided genes in transgenic plants that express unique Cry proteins (or other novel proteins) with different binding sites within the insect gut.

Ultimately it is very likely that seed blends (transgenic and non-transgenic seed) will form the foundation of resistance management programs. This will ensure some baseline level of compliance.

Based upon current trends – seed costs will very likely continue to increase.