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Western Corn Rootworm Field-evolved Resistance to Bt Corn and Insect Resistance Management Practices

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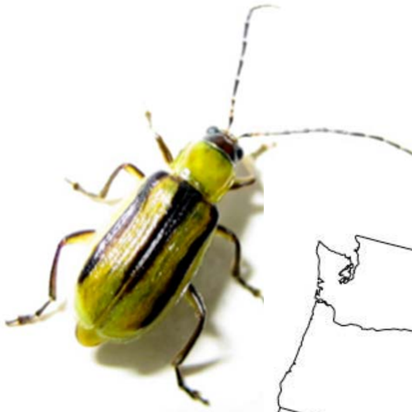
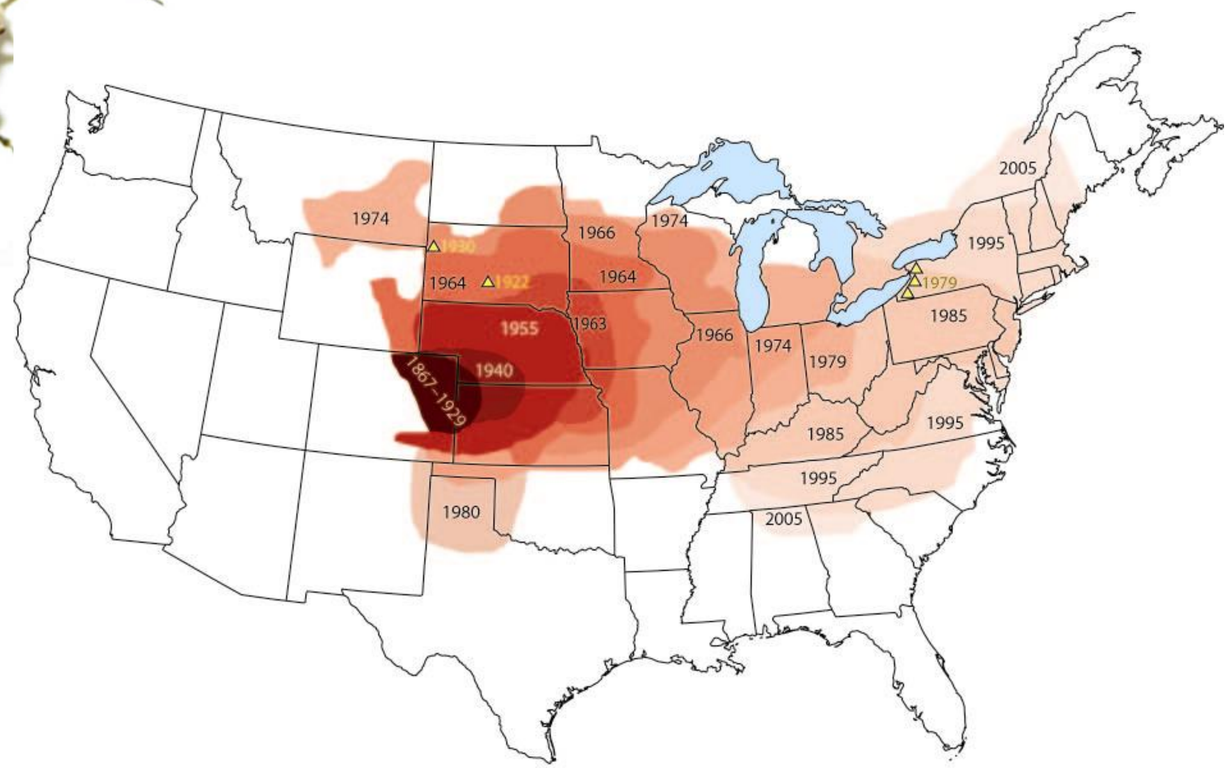
The U.S. Corn Crop (2012)

- Planted acreage 96.4 million acres, up 5 % from 2011 and highest in U.S. since 1937.
- More than one-third to livestock feed, 40% to ethanol (ethanol + distillers grains) and 13% exported. Remainder to food and beverage production.
- 2012: 67% of all corn planted contained a Bt trait (USDA ERS 2012)

Sources: USDA National Agricultural Statistics Service and Reuters

Western corn rootworm, *Diabrotica virgifera virgifera* (Coleoptera: Chrysomelidae)





Gray ME, et al. 2009.
Annu. Rev. Entomol. 54:303–21

Impressive history of adaptation

- Resistance to the organochlorine insecticide aldrin (Ball and Weekman 1963)

- Resistance to adult control measures methyl-parathion (organophosphate) and carbaryl (carbamate) (Meinke et al. 1998)
- Resistance to crop rotation in the western corn rootworm attributed to loss of ovipositional fidelity to corn (Levine et al. 2002, Gray et al. 2009)

Rotation-resistant western corn rootworm populations in Wisconsin

Fig. 1(a). Severely lodged first-year corn field (2004) following soybeans (2003) as a result of variant western corn rootworm, Walworth County, WI. **(b).** Economic corn rootworm feeding damage to corn roots. (a) Photograph by A. Peltier. (b) Photograph by UW Madison Entomology Dept.

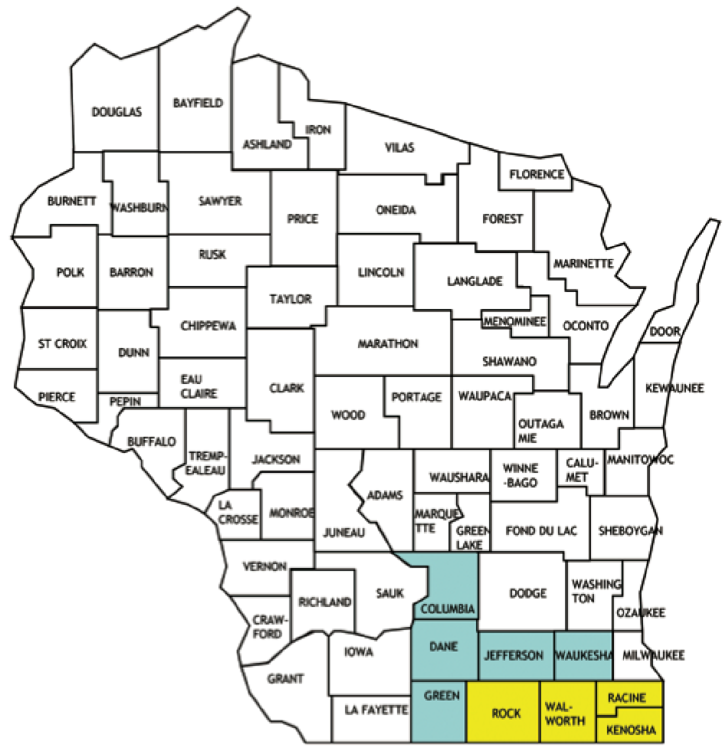


Fig. 4. Map showing the confirmed range of variant western corn rootworm in Wisconsin, October 2004. Affected counties (yellow), Unaffected counties (blue).

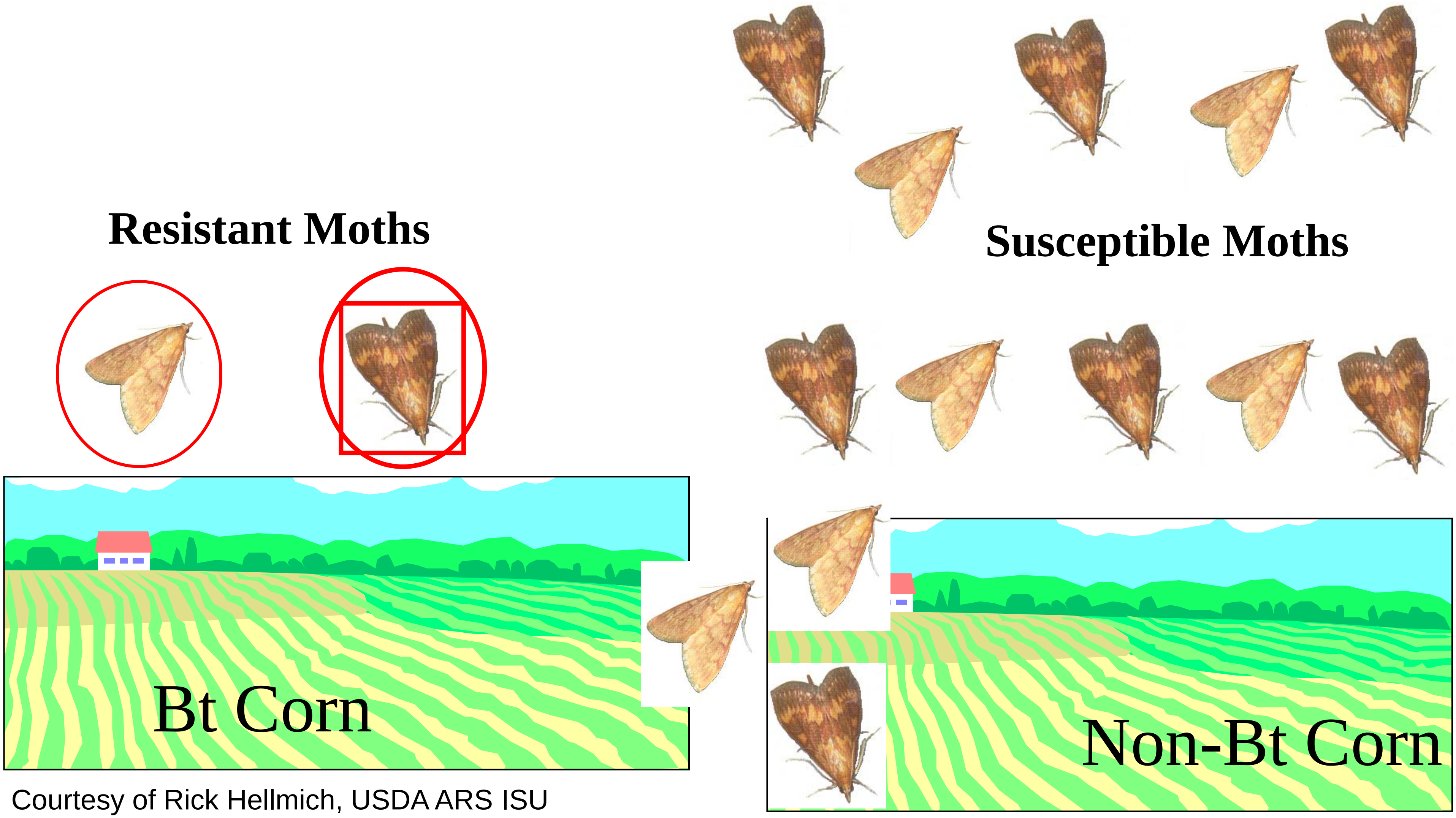
Cullen et al. 2008: *American Entomologist* 54: 170-178.

Insect Resistance Management

- IRM plan required by EPA for Bt corn registration to preserve long-term viability of plant incorporated protectants (PIPs) and microbial Bt sprays.
- Refuge (areas within or close to a field of the Bt corn where non-Bt corn is planted).
- Refuge percentage and configuration
- Historically a 20% structured refuge

Insect Resistance Management (IRM) Refuge Strategy:

Reduce chances that resistant moths mate with each other by providing large numbers of susceptible moths from the refuge, non-Bt corn.



Courtesy of Rick Hellmich, USDA ARS ISU

High Dose/Refuge Strategy

- IRM assumptions as modeled for Bt European corn borer-protected corn.

1. Protein expressed in plant tissue at high dose
2. Resistance is inherited recessively
3. Random mating occurs
4. Initial resistance alleles are rare
5. Fitness costs are associated with resistance

Sources: Gould 1998; Glaser and Matten 2003

Bt Corn Rootworm Traits

- Cry3Bb1 (MON863) in 2003
- Cry34/35Ab1 (DAS-591227-7) in 2005
- mCry3A (MIR604) in 2006

- Registered first as single traits with a 20% structured refuge.

- 'Stacks' and 'pyramids' registered with reduced refuge percentage and seed blend (refuge-in-the-bag) options.

- By 2011 different Bt corn products from different seed companies have different refuge requirements.

Artificial lab and greenhouse selection for CRW resistance

- Acquisition of Bt Cry3Bb1 resistance by western corn rootworm over a small number of generations (3 to 11) (Meihls et al. 2008, Oswald et al. 2011).
- Acquisition of Bt Cry34/35Ab1 and mCry3A resistance in lab-selected colonies (Lefko et al. 2008; Meihls et al. 2011).

Field-Evolved Resistance to Bt Maize by Western Corn Rootworm

- Researchers collected beetles from four NE Iowa corn fields in 2009 with unexpected damage to Bt CRW corn expressing the Cry3Bb1 protein.
- Severe root feeding reported by farmers to ISU Extension. Bt toxin confirmed by ELISA. (Avg. node-injury score of 1.8).
- Populations collected from five control fields not associated with unexpected feeding damage by corn rootworm on Bt corn. Four of these fields had no history of Bt corn, one did but no performance failures.

Gassmann et al. 2011: *PLoS ONE* 6: 1-7.

Field-Evolved Resistance to Bt Maize by Western Corn Rootworm

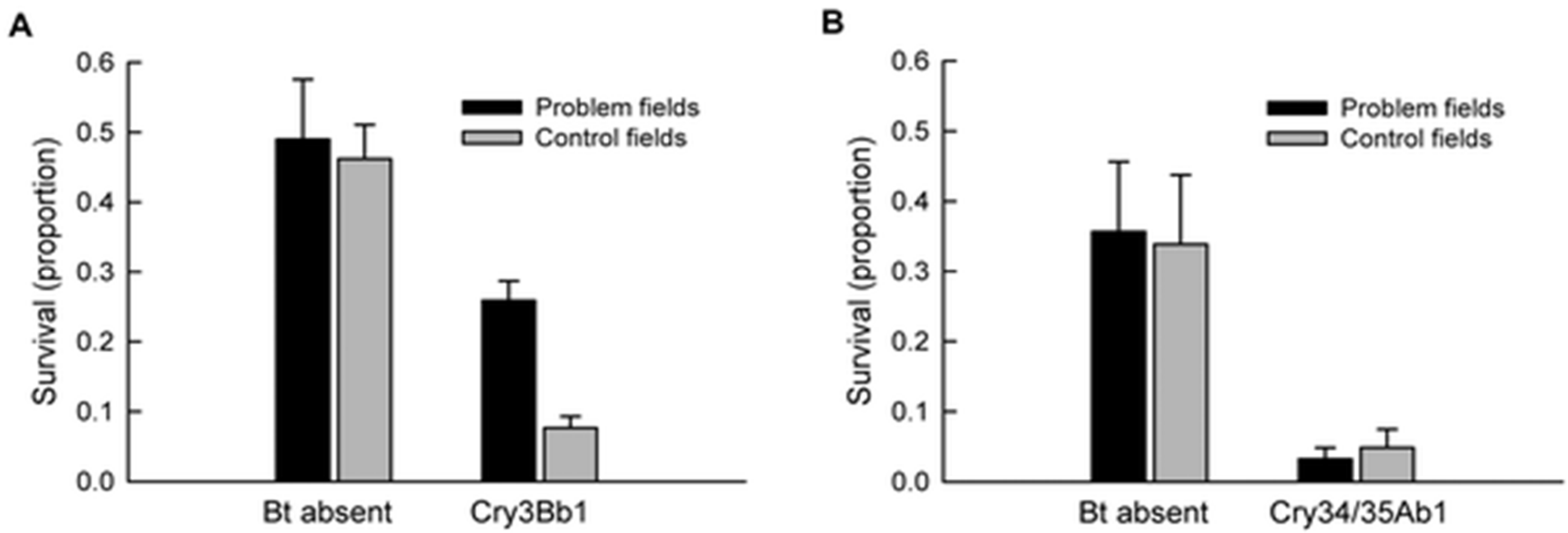
- Farmer interviews on field history 2003-2009 (crop rotation, type of Bt corn). No questions asked about refuge compliance.

- Progeny from field-collected beetles reared in laboratory. Neonate larvae used in on-plant bioassays to assess survival on Bt corn producing Cry3Bb1 and a near isogenic hybrid lacking the Bt trait.

- To test for cross-resistance, a Cry34/35Ab1 corn hybrid and its isoline hybrid were included in the bioassay experiment.

Gassmann et al. 2011: *PLoS ONE* 6: 1-7.

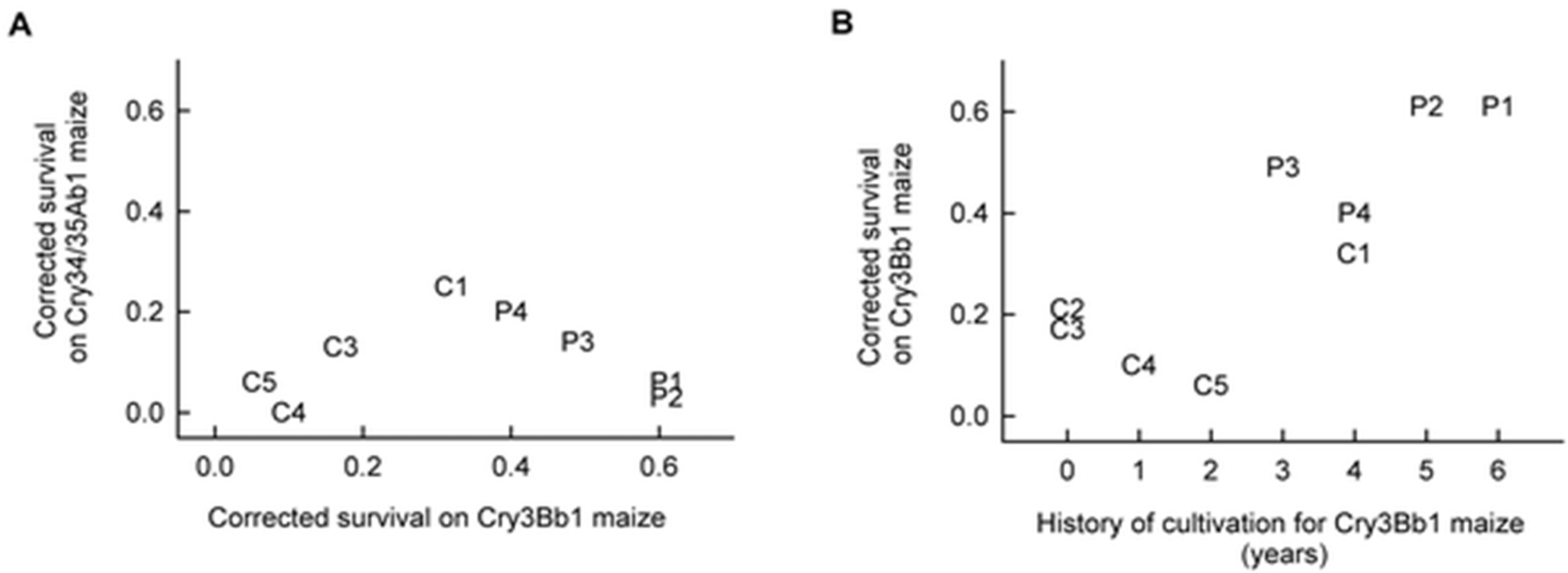
Figure 2. Survival of western corn rootworm on Bt and non-Bt maize.



Gassmann AJ, Petzold-Maxwell JL, Keweshan RS, Dunbar MW (2011) Field-Evolved Resistance to Bt Maize by Western Corn Rootworm. PLoS ONE 6(7): e22629. doi:10.1371/journal.pone.0022629
http://www.plosone.org/article/info:doi/10.1371/journal.pone.0022629

PLoS one

Figure 3. Correlation analysis for corrected survival of western corn rootworm.



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http://www.plosone.org/article/info:doi/10.1371/journal.pone.0022629

Field-Evolved Resistance to Bt Maize by Western Corn Rootworm – **Why?**

- For western corn rootworm, none of the three Bt CRW traits are expressed at high dose (EPA 2002; Storer et al. 2006; Hibbard et al. 2010)
- Non-recessive inheritance (Gassmann et al. 2011)
- Evidence of non-random mating (Kang &Krupke 2009)
- Initial resistance allele frequencies may be much higher than assumed (Onstad & Meinke 2010)
- Fitness costs of resistance may be small (Meihls et al. 2008; Gassmann et al. 2009)
- Insufficient planting of refuge plays a role (Gassmann et al. 2011)

Summary of findings to date

- A second study published in 2012 reported similar results for 7 additional populations of WCR collected in 2010 from Iowa corn fields expressing the Cry3Bb1 trait with root node-injury score greater than 1.0 (Gassmann et al. 2012).
- Two populations with confirmed resistance in bioassays on CRW collected in 2011 from NW and NC Illinois. Severe lodging, root pruning, continuous corn, reliance on single trait (Cry3Bb1) over many years (Gray, 2012).
- North Central Region entomologist project underway screening 2012 beetle populations from 40+ fields across 6 states and all three CRW traits.

March 5, 2012 letter to EPA IRM docket from 22 members and participants of NCCC46

regulations.gov
Your Voice in Federal Decision-Making

FARM JOURNAL

Resistance Reality

JULY 30, 2012

npr
the SALT
NPR'S FOOD BLOG

HIGH PLAINS/MIDWEST AG JOURNAL

Experts' letter sparks Bt corn refuge discussion

Insect Experts Issue 'Urgent' Warning On Using Biotech Seeds

Experts suggest steps to stop spread of resistant corn rootworms

By David Tenenbaum, University Communications

Friday, April 20th, 2012

Entomologists Letter to EPA

- EPA-HQ-OPP-2011-0922-0013. Porter, P., E. Cullen, T. Sappington, A. Schaafsma, S. Peuppke, D. Andow, J. Bradshaw, L. Buschman, Y. Cardoza, C. DiFonzo, et al. 2012. Comment submitted by Patrick Porter, North Central Coordinating Committee NCCC46 http://www.regulations.gov/#!documentDetail;D=EPA-HQ-OPP-2011-0922-0013

- Recommendations on IPM and IRM to preserve Bt CRW technology, while acknowledging the challenges.

Summary points of EPA letter

- Resistance to Cry3Bb1 threatens hybrids carrying two toxins (SmartStax corn hybrids with Cry3Bb1 and Cry34/35Ab1)
- Particularly concerning given decreased refuge requirements (20% to 5%) for these hybrids
- Crucial that susceptibility to Cry34/35Ab1 be preserved. It is now the common toxin in two different pyramids from two registrants. A third is seeking to register it with mCry3A.
- Insecticide use can mask trait performance issues and select for further resistance. Beginning to reverse the trend of reduced soil insecticide on Bt CRW corn realized over the past several years.

NCCC46 Letter to EPA, 2012

Challenges

- Bt CRW traits incorporated into elite germplasm with highest yield potential.
- Growers report increasing difficulty obtaining non-transgenic seed with high yield potential.
- Bt CRW hybrids planted prophylactically in areas with little or no CRW pressure.
- Widespread adoption of Bt technology has left many growers without the equipment necessary to apply soil insecticides (to non-Bt corn) at planting if necessary.

NCCC46 Letter to EPA, 2012

Questions to ask on your farms

- What is the crop rotation history?
- Planting one Bt CRW trait over consecutive years?
- Performance issues observed? (lodging, root feeding, yield impact)
- Is refuge compliance where it should be?
- Adult beetle density. (were beetle numbers high in the field the previous year in corn? Are you seeing greater numbers emerging in current year?)

Current Recommendations

- Consider crop rotation to non-host crop
- Soil insecticide at-planting with a non-Bt hybrid
- Plant a Bt hybrid with a different CRW protein than one which may have performed poorly
- Use a pyramided Bt hybrid that expresses multiple Cry proteins targeted against CRW.
- Adhere to IPM principles of crop rotation and insect scouting to apply control measures when and where necessary.
- Develop a long-term integrated approach relying on multiple tactics.

NCCC46 Letter to EPA, 2012

"Effectively dealing with the challenge of field-evolved resistance to Bt corn by western corn rootworm will require better adherence to the principles of integrated pest management."

Photo: Mark Steil, Minnesota Public Radio, Quote: Gassmann et al. 2012

Acknowledgements

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