Why stewardship of Bt cotton is still crucial: Insights from the US

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Bt cotton was initially commercialised largely for use outside of Australia in countries like the US where key lepidopteran pests were causing substantial economic losses over larger areas. Even so, this technology is valuable to Australian growers because its key target, *Helicoverpa armigera*, rapidly evolved significant levels of resistance to virtually every class of chemistry used to treat it.

Exposing pests to a high dose of the toxin and generating susceptible moths from refuge crops is the recommended strategy for slowing resistance to Bt crops. Although the toxins in Bt cotton can control *H. armigera* they have never been considered a ‘high dose’ for the Australian situation. As a result, and because *H. armigera* is so good at becoming resistant to insecticides, a conservative Resistance Management Plan (RMP) was developed for the first generation Bt cotton released in Australia in 1996.

The regulators of this technology (Australian Pesticides and Veterinary Medicines Authority) worked with Cotton Australia (who merged with the Australian Cotton Growers Research Association) and Bayer (formerly Monsanto) to ensure its appropriate resistance management. Even with the current third generation of Bt cotton and Bayer (formerly Monsanto) to ensure its appropriate resistance management, the performance of the new product. The other important component of management in Australia is that good baseline information on resistance levels in the target pest species means that quick action can be taken when larval survival rates in Bt cotton are determined as needing mitigation.

Despite Australian *H. armigera* carrying resistance genes that could overcome the toxins in Bollgard 3 cotton, and the lack of a truly high dose product against this pest, there is no evidence of emerging field resistance. This contrasts the situation in the US where *H. zea*, which is closely related to *H. armigera*, is evolving resistance to Bt cotton. Below we describe the background to the resistance situation in the US to serve as a reminder to Australian growers of the continued importance of good stewardship of Bt cotton in Australia.

The situation in the US

Bt maize and Bt cotton were first commercialised in the US during 1996. The original pests targeted by Bt cotton were *Chloridina virescens* (tobacco budworm), *Helicoverpa zea* (corn earworm or bollworm or tomato fruitworm), and *Pectinophora gossypiella* (pink bollworm: an invasive pest from South Asia and present in the US for about 100 years). *Pectinophora gossypiella* was a pest in the western US Cotton Belt, while the native species of *C. virescens* and *H. zea* are primarily eastern pests. Resistance management plans differed for these insects.

The RMPs for *C. virescens* and *H. zea* were reliant on the plant delivering a high dose of Bt toxin to the insect and areas planted to non-Bt cotton as structured refuge. Stalk destruction and burial following harvest were not used as part of the RMP tactics. While the Bt toxin dose killed nearly all of the individuals in the field, resistance for both *C. virescens* and *H. zea* could quickly be selected in the laboratory. Because of this, there was concern that resistance would quickly evolve in the field. This was even more apprehension for *H. zea*, since the Bt toxin dose provided by the cotton plant was not high enough and allowed significant survival to the point that, for example, many Bt cotton acres required foliar insecticide treatment during 1996. *Helicoverpa zea* also uses maize as a host.

Growers rapidly began planting more and more Bt maize and Bt cotton varieties, which meant that Bt maize functioned as a selective filter for Bt cotton. Later, Bt cotton varieties were introduced with multiple Bt toxins (pyramids) across the US Cotton Belt, but single-toxin Bt cotton was not immediately phased out like it was in Australia, and the introduction of multi-toxin Bt cotton therefore overlapped with plantings of single toxin Bt cotton for many years.

The RMP for *P. gossypiella* relied on the plant delivering a high dose of Bt toxin to the insect, using areas planted to non-Bt cotton as refuge, and stalk destruction and burial following harvest to support host-free periods. But unlike the RMP for the other insects, the use of Bt cotton was part of a broader strategy.
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using mass releases of sterile moths and other tactics to eradicate *P. gossypiella*. So even though resistance could be quickly selected in the laboratory, the use of Bt cotton and other tactics meant that *P. gossypiella* was eradicated from the US.

**How is RMP compliance enforced in the US?**

Current RMP compliance efforts in maize focus on the planting of non-Bt refuges. Furthermore, for *C. virescens* and *H. zea*, the requirement for non-Bt cotton refuges was removed during 2010, since by then:

1. All Bt cotton varieties planted were pyramids; and,
2. Cabbage was determined to be a relatively unimportant host that did not contribute significantly to overall population sizes, relative to maize, soybean, and uncultivated hosts.

In cotton growing regions Bt maize dominates plantings and growers are legally required to plant 20 per cent of their total maize acres to non-Bt maize as a refuge. This stipulation is attached to the registration of the particular Bt trait in maize, although very few growers plant refuge maize in the US Cotton Belt.

A few reasons for this have been identified. First, marketing and breeding efforts for refuge hybrids have fallen over time. Many growers complain that non-Bt seed availability is an issue and patents allow Bt trait providers to charge a premium for Bt seed at a greater profit than non-Bt seed. Seed producers complain that growers don’t demand enough non-Bt seed. Furthermore, the herbicide tolerance traits might not be available in a non-Bt hybrid to fill a particular grower’s needs.

As a condition of registration in the US, Bt trait providers participate in a coalition organisation, the Agricultural Biotechnology Stewardship Technical Committee (ABSTC). This organisation, among other functions, reports surveys (done through a third party) of non-Bt refuge compliance yearly to the governmental regulatory agency.

Most of these surveys are done by phone; usually less than 150 growers (probably less than 0.1 per cent) are surveyed across the US Cotton Belt and the registrants also perform an even more limited number of on-farm checks. Note that these phone surveys are designed to assess compliance, while on-farm visits are meant to be a deterrent. The government does not enforce refuge compliance with growers.

Rather, if a grower is found to be non-compliant with refuge practices twice in a five-year period, the company must deny the grower access to Bt maize. Therefore, Bt trait providers are disincentivised to find growers out of compliance, although they have, in limited cases, denied sales to some growers. Growers found out of compliance from one Bt trait provider can still purchase seed from another competing trait provider and very few growers are ever reported as non-compliant by ABSTC.

**RMP outcomes and resistance in the US to Bt cotton**

As mentioned earlier, *P. gossypiella* was successfully eroded in the US. Prior to eradication, although Bt resistance genes could be detected, refuge compliance associated with Bt cotton was good, and resistance levels never increased. Eradication was a grower-run program in concert with an extensive university extension education effort across a large irrigated desert cotton-growing region. They used a combination of factors, including:

- Stalk destruction after harvest to support host-free periods;
- Pheromone mating disruption;
- Chemical control if thresholds were exceeded;
- Use of pyramided cotton varieties; and,
- Mass release of sterile male pink bollworm moths as a replacement for non-Bt cotton structured refuges and to support maximum adoption of Bt cottons (over 98 per cent of acreage).

Similarly, no Bt resistance has been detected for *C. virescens*. This insect undergoes multiple generations a year and is likely only exposed to a Bt crop, cotton, for one to three generations since it does not use maize as a host. But the most likely explanation for why resistance hasn’t yet evolved is because Cry1Ac, expressed in all commercially planted varieties, provides a very high dose for *C. virescens*, killing most of the potentially resistant insects.

In contrast, *H. zea* has evolved resistance to two-toxin (Cry1 and Cry2) cotton varieties (known in Australia as Bollgard II). Early claims for resistance were made during 2008, but were difficult to confirm without good baseline resistance monitoring efforts, gaps in monitoring efforts over time, high natural variability in susceptibility to Cry toxins, and inconsistent crop losses.

Arguably, widespread resistance was not evident in the field until 2016, likely a function of a heavy pressure year combined with increasing levels of resistance.

The consensus is that *H. zea* Bt resistance evolved more quickly than other target pests in cotton because of problems in the RMP including:

- Maize and cotton plants providing too low of a Bt dose;
- Cross-resistance from selection on similar toxins in maize and cotton;
- Overlapping times in the release of pyramided Bt maize and cotton varieties with single Bt toxin varieties;
- Too few growers planting non-Bt refuge maize or cotton; and,
- A lack of crop diversity.

University extension trials indicated that chlorantraniliprole (marketed in Australia as Altacor and a component of Voltan Flexi), an insecticide with an extremely long residual, applications made prior to larvae hatching from the eggs could provide protection. Growers nearly immediately adopted this technology to tackle *H. zea* resistance. In some geographies, only a single additional application is needed, while in others two additional applications are sometimes required when pressure is heavy.

Therefore, these growers are still paying a premium for two-toxin Bt seed (to control *C. virescens*) and are paying more for foliar insecticide applications. Increasingly, three-toxin Bt varieties (that express the Cry1, Cry2, and Vip toxins; e.g., Bollgard 3 cotton) are being planted that generally do not require foliar insecticides for *H. zea* management. But without changes in RMP tactics, *H. zea* will soon evolve resistance to Vip or chlorantraniliprole.

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