Pupae Busting is essential for resistance management

With more than 90 percent of Australian Cotton grown using bt technology, effective resistance management is vital to the longevity of bollgard ii and the yet to be released bollgard iii. d&d bt stewardship senior technical specialist sally ceeney reports.

CSIRO’s resistance monitoring research shows that in both of the target pests, H. armigera and H. punctigera, resistance genes to Cry2Ab are present, are higher than expected, and are probably increasing, making resistance management arguably one of the most important issues facing the industry.

While the introduction of Bollgard III and its additional Vip3A toxin may seem like the silver bullet for resistance, CSIRO screening of H. armigera and H. punctigera populations in 2009 and 2010 found not only the first examples worldwide of an insect with genes allowing resistance to a vegetative insecticidal protein, they also discovered that a larger than expected proportion of individuals in populations of both Helicoverpa species already carry a gene that allows them to tolerate Vip3A.

These frequencies were higher than expected, and they are greater than the initial frequencies of insects carrying a resistance gene to Cry2Ab when Bollgard II was first introduced. If Cry2Ab is ineffective when Bollgard III becomes available the Vip3A toxin may be exposed to selection in a similar fashion to what we assume currently occurs for Cry2Ab in Bollgard II.

Pupae busting is an essential component of the Bollgard II Resistance Management Plan.

Why pupae bust?

As autumn approaches in temperate regions day length decreases and temperatures begin to cool, triggering mature Helicoverpa larvae to enter a diapause phase in the soil. This dormancy strategy allows the pest to survive the winter months in temperate regions when host plants are scarce and temperatures are generally too low to allow successful development.

Cultivation of the soil between seasons, during the dormancy phase, is an effective way of preventing any moths that developed resistance in the previous year from contributing to the population in the following year. Although it is known that few larvae will survive in Bollgard II crops, those that do are more likely to be resistant
and so are precisely the ones that should be targeted to prevent them emerging and contributing resistant genes to the population the following spring.

So while the numbers of pupae killed may seem small, the proportions of resistant individuals are high, meaning that taking this group out of the overall population can have a big impact on the frequency of resistance in that population.

In Central Queensland, due to the warmer temperatures, *Helicoverpa* pupae produced late in the season do not remain in the soil but emerge within 15 days of pupating, making pupae busting ineffective.

Late season trap crops are used as an alternative. Trap crops of pigeon peas are timed to be at their most attractive after the cotton has cut out. Moths emerging from the Bollgard II fields late in the season should be attracted to the pigeon peas to lay their eggs. Once the cotton has been harvested the trap crops are destroyed and cultivated to kill the larvae and pupae.

History proof of success

In the early 1990s CSIRO researchers led by Gary Fitt showed that single operations reduced pupal survival by up to 90 percent, depending on timing and the method used, cementing the role of pupae busting in resistance management. Prior to this, pupae busting had become a mainstay in the cotton industry due to its success in helping to delay resistance to insecticides in conventional cotton. It became widely adopted as part of the industry’s voluntary IRMS. Because of the success of the pupae busting tactic in this era and the confirmation of the tactic’s validity through research, it followed that the industry adopted it as a tool for delaying resistance in Bt cotton from its introduction in 1996.

Pupae Busting Requirements

According to the RMP, pupae busting of all Bollgard II crops in NSW and Southern Qld should occur within four weeks of harvest and must be completed by July 31. Soil disturbance must occur to a depth of 10cm across the whole soil surface.

Soil disturbance of the unsprayed refuge associated with Bollgard II crops should not occur until all the pupae busting in the Bollgard II has been completed.

Ideally, unsprayed refuges should be left uncultivated until the following October. This ensures maximum emergence of late pupae from the refuges in the following spring. These emerging moths have not been exposed to the Bt toxins and can outcross with moths in the general population, thereby helping to dilute resistance.

Pupae busting in conventional sprayed cotton remains an important component of the IRMS however the use of the Helicoverpa Diapause Emergence Tool, available on the CotASSIST website, has meant there is now some flexibility for growers.

Models based on long term weather records show crops defoliated before March 9 are highly unlikely to harbour resistant diapausing *Helicoverpa* and do not need to
be pupae busted. It is recommended that fields defoliated after March 9 be pupae busted as soon as possible after harvest and no later than the end of August.

Pupae Busting Methods

There are a variety of implements that can be used to achieve soil disturbance of 10cm. The method chosen will depend on a range of factors including soil moisture and farming rotation (eg whether the field is going back into cotton, fallow or a winter crop). Effective pupae busting and crop destruction can also aid in the reduction of ratoon and volunteer cotton plants the following season.

The following table shows some general guidelines of the adequacy of typical cultivation equipment for effective pupae busting:

<table>
<thead>
<tr>
<th>Generally satisfactory</th>
<th>Inadequate alone</th>
<th>Unsatisfactory</th>
</tr>
</thead>
<tbody>
<tr>
<td>(more than one of these options may be required)</td>
<td></td>
<td></td>
</tr>
<tr>
<td>chisel, disc or blade plough</td>
<td>centre busting</td>
<td>stalk pull (wet)</td>
</tr>
<tr>
<td>stalk pull and go – devils or lillistons plus alabamas</td>
<td>stalk pull (dry), rake and burn</td>
<td>phoenix harrows</td>
</tr>
<tr>
<td>cultivators with wide sweeps</td>
<td>go-devils</td>
<td>drag harrows</td>
</tr>
<tr>
<td>planters with cultivating tines</td>
<td>stubble mulchers</td>
<td>direct drill planters</td>
</tr>
</tbody>
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Source: Machinepak

Luke Sampson, Monsanto Regional Business Manager for Bourke, Macquarie and Southern NSW, has shared some top tips for effective pupae busting:

- Work with your Technology Service Provider (TSP) to ensure your pupae busting operation is effective from the start
- Regularly check the level of soil disturbance achieved, particularly when moving into new fields or variable soil types
- Unsprayed refuge areas do not need pupae busting
- Achieving effective pupae busting can be more difficult in dry conditions. Again, work with your TSP to ensure soil disturbance is adequate. This may help prevent having to perform multiple passes.

Luke also points out that effective crop destruction can not only assist with pupae busting, but can also help reduce volunteer and ratoon cotton the following season:

- Root-cutting is highly recommended to prevent regrowth of ratoon cotton and improve soil disturbance in the plant line.
- Early incorporation aids in the breakdown of any residual seed cotton which helps in reducing volunteer numbers the following summer

THE FUTURE OF PUPAE BUSTING
With Monsanto currently working to introduce a third generation Bt technology, it is important to ensure that future RMPs are as robust as possible in managing resistance, whilst still being practical and achievable for growers. The industry is investing in a number of research projects to ensure that the development of new RMPs for this and other potential Bt technologies in the future can be well informed by locally relevant science.

Pupae busting is a proven effective means of destroying over-wintering pupae which are the section of the population with the highest probability of carrying resistance into the next season.

However, from a farming perspective there are some drawbacks.

Firstly, pupae busting is incompatible with minimum tillage, which is widely adopted bringing many benefits including reducing soil erosion, conserving soil moisture and improving carbon sequestration. These issues are most pressing for dryland cotton growers, since irrigated growers often have to perform tillage operations in normal ground preparation, especially for back-to-back cotton crops

Yet even for irrigated growers, the need to pupae bust can often restrict rotation crop options, and prolonged wet weather can expose growers to the risk of being non-compliant with their RMP. There is also an expense involved, growers estimate pupae busting costs between $40-55/ha, depending on field rotation and the operation performed.

Research underway for alternatives

CRDC is currently funding research projects investigating the potential of novel ways to reduce the resistance risk associated with pupal survival and late season moths that may emerge prior to pupae busting occurring, which is a potential risk particularly in early maturing crops.

Professor Gregg and his team are researching the potential of using moth attractants and insecticides to control moths emerging late in the season. The use of Magnet, a registered moth attractant, combined with low rates of insecticide applied in small bands (every 70-140m) across a field has shown some potential in being a cost effective, IPM-compatible means of removing these late emerging moths.

Some research is also being conducted by CSIRO and collaborators to investigate the application of certain chemicals late season that have the potential to disrupt the pupal development stage of Helicoverpa sp. as a potential alternative to pupae busting.

It is not expected that these methods will fully replace pupae busting in all future RMPs but hoped that the techniques may improve the robustness of RMPs and that there may be some potential to increase the flexibility in RMPs for growers while offering protection for the next generation of Bt cotton. Such changes to the RMP would be determined by the response of the government regulators to
submissions by technology providers, advised by the TIMS Committee of Cotton Australia.

More information

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Pupae busting is an essential component of the Bollgard II Resistance Management Plan. Pupae busting is a proven effective means of destroying over-wintering pupae which are the section of the population with the highest probability of carrying resistance into the next season.