**Insecticide Resistance in Cotton Aphid (*Aphis gossypii*): Results and Management Options for after seasons 1999/2000, 2000 / 2001 and 2001/2002**

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**Abstract**
Strains of cotton aphid (*Aphis gossypii*, Glover) and the green peach aphid (*Myzus persicae*, Sulzer) were collected from cotton producing farms in eastern Australia. Green peach aphid was rarely collected but resistance to Curacron® (profenofos) and Pirimor® (pirimicarb) was documented. The abundance of cotton aphid populations showing Pirimor® resistance appears be increasing, however, in contrast the abundance of Curacron® may have reduced during 2001 / 2002. Talstar® (bifenthrin) resistance was detected although the product is not registered for aphid control. Endosulfan, Confidor® (imidacloprid) and Pegasus® (diafenthiuron) resistance was not detected. The results underpin the resistance management strategy for aphids in Australian cotton and are discussed in conjunction with management recommendations based on IPM principles.

**Introduction**
Resistance in both the cotton aphid (*Aphis gossypii*) and the green peach aphid (*Myzus persicae*) is a major new threat to Australian cotton production (Herron *et al.* 2001). Insecticide monitoring underpins the management of resistant populations and collections of aphids are made from fields where failures or problems have been experienced. These are tested against a range of the control options allowing us to identify emerging resistance problems as well as keeping track of existing problems. This information then contributes to the development of the aphid component of the Insecticide Resistance Management Strategy for cotton.

Here we report the results of the 1999/2000, 2000 / 2001 and 2001/2002 seasons testing on aphids and out line the practical implication of those findings for resistance management.

**Materials and methods**
*Insecticides*
Insecticides screened for resistance includes a range of control options as well as some products such as the pyrethroids bifenthrin and deltamethrin (Table 1). The pyrethroids are included so that issue of potential multiple or cross-resistance can be explored.

***Table 1*** Common name, trade name, chemical group, formulation[1], concentration and supplier for insecticides tested against aphids.

| Common Name | Trade Name | Chemical Group | Form | Conc. | Supplier |
|---|---|---|---|---|---|
| bifenthrin | Talstar | pyrethroid | EC | 100 g/L | Crop Care |
| CGA-140408 | carbodiimide of Pegasus | thiourea | EC | 250 g/L | Syngenta |
| endosulfan | Endosulfan | organochlorine | EC | 350 g/L | Crop Care |
| omethoate | Folimat | organophosphate | LC | 800 g/L | Bayer |
| pirimicarb | Pirimor | carbamate | WP | 500 g/Kg | Crop Care |
| deltamethrin | Decis | pyrethroid | EC | 27.5 g/L | Aventis |
| profenofos | Curacron | organophosphate | EC | 500 g/L | Syngenta |

[1] AC=aqueous concentrate, EC=emulsifiable concentrate, LC=liquid concentrate and WP=wettable powder.

*Strains tested*

Aphids are collected by researchers, CRC Industry Development Officers, consultants and growers from fields where there has been poor control of aphids following an application of aphicide. These are sent to our laboratory at Camden (EMAI) and each field strain is cultured separately on pesticide-free cotton (Deltapine 90) at 25±4 °C under natural light. Strain integrity was assured by maintaining populations in purpose built aphid proof cages.

*Bioassay method*

The method for testing is outlined in (Herron *et al.* 2000). Briefly, 35 mm Petri dishes were used and excised cotton plant leaf discs were placed onto 3 mL of liquid cooling agar within the Petri dish. When the agar had cooled and set, 5-6 batches (batch = 10) of adults female aphids were transferred onto the leaf discs and sprayed with the aid of a Potter spray tower producing an aqueous deposit of 1.6 ± 0.07 mg $cm^{-2}$ with a 2 mL aliquot. Aphids were exposed to a single discriminating concentration of chemical. Each test was replicated and included a water only sprayed control. After spraying, Petri dishes were covered with clear plastic film and perforated with a fine needle to prevent condensation. Tests were maintained at 25 ± 0.1 °C in constant light for 24 h after which mortality was assessed.

*Bioassay analysis*

Mortality at the discriminating concentration was corrected for control mortality (Abbott 1925) which did not exceed 10%.

**Results and Discussion**

*Green peach aphid*

Two strains associated with control failure were collected in 2000/2001 though only one could be tested for resistance, due to parasites destroying the other strain. The strain tested was profenofos (18%) (Curacron, Sabre) and pirimicarb (24%) (Pirimor, Aphidex) resistant but was susceptible to endosulfan and deltamethrin (Decis). Despite there being 24% of aphids with resistance to pirimicarb, the resistance level of the aphids is relatively low, < 5 fold. For comparison, a cotton aphid strain with a similar 24% of individuals resistant to pirimicarb would have a high (> 1000 fold) resistance level (Herron *et al.* 2001). The practical outcome is that it is important not to mix up the two species when planning insecticide control. These differences are likely due to differences in the resistance mechanisms between the aphid species.

*Cotton aphid*

High-level organophosphate (OP) and carbamate resistance has been repeatedly confirmed in a number of regions, with hotspots in the lower Namoi Valley and Emerald. There is continued evidence of cross-resistance between the older OPs dimethoate/omethoate and the carbamate, pirimicarb. However, there is also resistance to profenofos.

Preliminary work by Herron, Gunning (NSW Agriculture) and Moores (Rothamstead) shows that profenofos resistance is probably due to a different primary mechanism to the primary mechanism causing high-level pirimicarb/dimethoate/omethoate resistance. However, there may be some cross-resistance as profenofos resistance (probably due to esterases) may also result in some degree of resistance to pirimicarb/dimethoate/omethoate. Clearly, the underlying resistance mechanisms are very complex. A Post Doctoral fellow, Emma Cottage (NSW Agriculture), has been appointed to work with Drs Herron and Gunning to study resistance mechanisms in aphids and develop a 'test kit' for pirimicarb/dimethoate/omethoate resistance detection in cotton aphid.

Comparison with earlier data suggests the proportion of strains showing resistance to pirimicarb may be rising as there were 59 % resistant in 1999-2000, 79 % resistant in 2000-2001and 80% in 2001-2002 (Table 2). However, confusingly, the proportion of profenofos

(Curacron) resistant strains rose from 64 to 79% and then fell to 70% in 2001-2002. Although bifenthrin is not registered to control aphids resistance was detected in 37% of the strains sampled. Clearly, aphids have been selected for resistance with sprays targeting other species, such as *Helicoverpa*. Interestingly, endosulfan resistance was not detected nor was it detected to imidacloprid (Confidor) or diafenthiuron (Pegasus). Consequently, any control problems experienced with these chemicals are due to factors other than resistance.

***Table 2*** Number and percentage of cotton aphid strains from cotton showing resistance during the 1999-2000, 2000-2001and 2001-2002cotton season.

| Chemical | Number of strains tested* (2001/02) | Number of strains resistant (2001/02) | Percent of strains with resistance 2001/02 | Percent of strains with resistance 2001 | Percent of strains with resistance 2000 |
|---|---|---|---|---|---|
| Pirimor | 10 | 8 | 80 | 79 | 59 |
| Folimat | Not tested | Not tested | Not tested | 79 | 65 |
| Curacron | 10 | 7 | 70 | 76 | 64 |
| Pegasus | 10 | 0 | 0 | 0 | 0 |
| Confidor | 10 | 0 | 0 | 0 | 0 |
| Talstar | 8 | 3 | 37 | Not tested | Not tested |
| Endosulfan | 8 | 0 | 0 | Not tested | Not tested |

* testing only 30% complete at time of writing.

Improving management of aphids requires and understanding of the carry-over of resistant populations from one season to the next. During the 2000 / 2001 season we investigated this by re-sampling a proportion of fields sampled in the previous season. In some instances resistance persisted from season to season, a scenario seen at "Glendale" (Table 3). This is consistent with our limited understanding of the resistance mechanism where resistance can be maintained without selection. It seems reasonable to speculate that resistant aphids could overwinter on farms causing ongoing and worsening control problems season to season.

***Table 3*** Percentage of cotton aphids showing resistance, collected from "Glendale" during the 1999-2000 / 2000-2001 cotton seasons

| Season | Pirimicarb | Profenofos | Imidacloprid | Omethoate | Diafenthiuron |
|---|---|---|---|---|---|
| 99/00 | 34 | 29 | 0 | 40 | 0 |
| 00/01 | 62 | 49 | 0 | 27 | 0 |

As expected there were also examples where resistance was absent during 1999 / 2000 but detected during 2000 / 2001. The result for "Pasadena" (Table 4) is typical of such populations. In this instance resistance has gone from undetectable to a high frequency and likely control failure within a season. This is probably due to the immigration of a resistant aphid strain.

***Table 4*** Percentage of cotton aphids showing resistance from "Pasadena" collected during the 1999-2000 / 2000-2001 cotton seasons

| Season | Pirimor | Curacron | Confidor | Folimat | Pegasus |
|---|---|---|---|---|---|
| 99/00 | 0 | 0 | 0 | 0 | 0 |
| 00/01 | 53 | 23 | 0 | 68 | 0 |

Once resistance is selected and established it will usually persist through the growing season unless the resistant clone is killed. At "Waverley" (Table 5) resistance was detected early in the season and then remained at a high frequency for the remainder of the season.

*Table 5* Percentage of cotton aphids showing resistance from "Waverley" collected during the 2000-2001 cotton season.

| Season | Pirimor | Curacron | Confidor | Folimat | Pegasus |
|---|---|---|---|---|---|
| 30/11/00 | 91 | 82 | 0 | 98 | 0 |
| 04/01/01 | 66 | 34 | 0 | 64 | 0 |

Interestingly, at "Parker Farms" (Table 6), monitoring detected a high frequency of resistant aphids" during 1999 / 2000. However, collections of aphids from the same field early in 2000 / 2001 showed no resistance. A subsequent sample later in the 2000 / 2001 again showed resistance. This apparent overwinter loss of resistance between seasons is not an isolated instance, we have two other examples. The results for "Mirrabooka" (Table 7) are particularly fascinating as adjacent fields had aphids with completely different resistance profiles. "Mirrabooka" field 2 contained only susceptible aphids while field 5 contained highly resistant aphids. This confirms anecdotal grower and consultant feed back that aphids separated by metres might have completely different resistance profiles.

*Table 6* Percentage of cotton aphids showing resistance from "Parker Farms" collected during the 1999-2000 / 2000-2001 cotton season

| Season | Pirimor | Curacron | Confidor | Folimat | Pegasus |
|---|---|---|---|---|---|
| 1999/00 | 96 | 65 | 0 | 100 | 0 |
| 2000/01 (28 Nov) | 0 | 0 | 0 | 0 | 0 |
| 2000/01 (16 Jan) | 5 | 34 | 0 | 85 | 0 |

*Table 7* Percentage of cotton aphids showing resistance from "Mirrabooka" collected during the 1999-2000 / 2000-2001 cotton season.

| Strain | Season | Pirimor | Curacron | Confidor | Folimat | Pegasus |
|---|---|---|---|---|---|---|
| Field 1 | 1999/00 | 70 | 58 | 0 | 89 | 0 |
| Field 2 | 2000/01 (18 Dec) | 0 | 0 | 0 | 0 | 0 |
| Field 5 | 2000/01 (8 Dec) | 95 | 54 | 0 | 93 | 0 |

If resistant aphids do not always persist season to season at a given location then the aphid strain must either die out at that location, emigrate or be diluted (by immigration). Aphid control problems in one season do not necessarily mean control problems next season making a 'test kit' for resistance very useful. Some of these findings can be explained by the 'clonal' nature of aphids.

**Cotton Aphid Management Options**

Cotton aphids in Australia reproduces asexually with female aphids giving birth to live female young. The result is aphid 'clones' exactly the same as the mother, including their resistance status. As there is no 'sexual' reproduction there is no dilution of resistance through crossing between susceptible and resistant parents. Dilution can only occur if there is an influx of susceptible aphid clones.

The clonal structure of the aphids therefore explains why resistance to aphids can increase so quickly and why it can vary from field to field. The problem is compounded by the fast rate of

aphid population growth; a female aphid can produce 4-6 live young per day (which are all female) and these will mature to adults in about a week under ideal conditions.

There are no silver bullets for managing aphids at present, instead a number of integrated controls tactics are recommended to help reduce aphid numbers and manage resistance. Our current resistance management strategy for aphids hinges on these following four main points:

- A maximum of 2 sprays of any registered aphicide against aphids, unless the product is otherwise restricted
- Rotation of chemistry, that is, do not use chemicals from the same mode of action group consecutively
- The first aphicide spray should not be from the same chemical group as any seed treatment or at planting insecticide that also controls aphids.
- There is cross-resistance between carbamates (Group 1A) and organophosphates (Group 1B) and therefore they should be considered as the same group for aphid control.

The chemical control rotation based resistance management strategy is then further augmented with a number of IPM based control strategies to additionally reduce the fitness of the resistant clone. These include:

- *Reduction of on-farm overwintering hosts for aphids (i.e have a host free period).* Cotton aphids survive through winter on suitable hosts either on-farm and in uncultivated areas. Growers should aim to reduce the availability of on-farm overwinter hosts. If farm hygiene is adequate through winter it is possible that localised aphid survival will be low and that immigrant colonising aphid clones will be susceptible. Growers on farms where resistance problems occurred could consider planting a 'non-host' rotation crop, such as a winter cereal and ensuring that on-farm control of weeds, cotton stubble and cotton volunteers is excellent. The relative importance of particular host species in overwintering is poorly understood but is the subject of a major research effort.

- *Consider an at-planting insecticide treatment BUT be wary of potential resistance implications.* Some of the 'at-planting' insecticides or seed treatments will control aphids reducing early season aphid numbers and delaying population development. This will reduce or postpone the need for foliar aphicides allowing beneficials to survive and may also reduce the risk of CBT (cotton bunchy top). However, do not follow a seed treatment or at-planting insecticide with a foliar spray from the same group. Appropriate chemical alternation is still necessary late in the season even if aphids have not been targeted for control since the use of the 'at planting' insecticide.

- *Sample effectively for aphids.* Aphid sampling should begin from seedling emergence and be done at least weekly looking carefully at younger growth. If a high proportion of plants have winged forms then resample within a few days to check if they have settled and produced young. If aphids are found it is important to confirm the species, as it will dictate control options.

- *Rotate aphicides.* Select from the full range of aphicides available at the time. The four key insecticide rotation groups are (i) pirimicarb and organophosphates, (ii) Confidor (iii) Pegasus and, (iv) endosulfan.

- *Don't follow a failure with another product from the same group.*

- *Maintain beneficial insects by using the most selective option when pests need to be controlled.* A range of parasitoids and predators will help to reduce aphid survival.

- *Thresholds.* Data from overseas suggests that cotton will generally recover from aphid damage provided infestations do not persist at high levels (> 90 % plants infested) for too long (< 10 days). The conventional threshold for aphids through the growing period (90% of plants infested) is still therefore appropriate in terms of cotton's capacity to tolerate aphid damage. However the recent advent of the CBT syndrome (for which aphids are the suspected vector) means that some growers are unwilling to tolerate conventional aphid thresholds. It is important to weigh-up the risk of CBT and frequent spraying to control aphids against the risk of resistance when deciding if aphids need to be controlled.

- *Varieties.* Selection of varieties may also be an important consideration. There is a range of CBT resistance in commercial varieties. For growers or consultants concerned with the risk of CBT, select from the more resistant varieties. This will reduce concerns about managing aphids and provide growers with the confidence to tolerate higher numbers, allow beneficials to survive thereby helping to reduce insecticide resistance selection.

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