INTEGRATING HERBICIDE USE WITH BIOLOGICAL CONTROL
OF BITOU BUSH

Nigel Ainsworth¹ and Royce Holtkamp²

¹ CRC for Weed Management Systems, KTRI, PO Box 48, Frankston Vic. 3199.
² NSW Agriculture, Weed Biological Control Unit and CRC for Weed Management Systems,
RMB 944 Calala Lane, Tamworth NSW 2340.

Abstract  Several insects have been released as biological control agents for bitou bush (Chrysanthe-
moides monilifera) and other potential agents are under investigation. Once established in the field the
agents are affected by herbicide treatments, mainly glyphosate, which are used against this weed on a large
scale. There have been no systematic assessments of what happens to biological control agents in sprayed
areas or whether the success of herbicide treatments is altered by the activities of biological control agents.
Integration of herbicide use with biological control to obtain the best possible outcome requires the devel-
opment of a working strategy based on use of current information, which should then be refined and updated
as field observations and experimental results are accumulated and new biological control agents are re-
leased. This paper outlines the current state of knowledge on biological control-herbicide interactions for
C. monilifera, including results of a recent experiment with the bitou seed fly, and identifies the most impor-
tant aspects needing further work.

INTRODUCTION

Bitou bush (Chrysanthemoides monilifera ssp. rotundata (L.) T. Norl.) is a serious environmental weed
in NSW and south Queensland and has been a target for biological control since 1987. Five insects have
been released as biological control agents for C. monilifera in Australia, three leaf-feeding beetles, a
moth and a fly. A further moth species is undergoing testing. Only the bitou tip moth (Comostolopsis
germana Prout) and more recently the bitou seed fly (Mesoecaris polana Munro) have established widely
and colonised large areas (Holtkamp 1996, Edwards et al. 1999). Chemical control of C. monilifera can
involve cut stump treatment or overall spraying from the ground or the air. Winter aerial application of
glyphosate has proven to be an effective means of control that is well tolerated by many native plants on the
NSW coast (Toth et al. 1996) and has recently been used to treat around 1000 ha per year. Chemical con-
trol of C. monilifera is proceeding in the presence of the biological control agents but there has so far been
no formal study of the interactions of the two techniques. Based on knowledge of herbicide effects and
the life histories of the current biological control agents best estimates of potential interactions are as follows:

Chrysolina spp. and Cassida sp.  Of the three species of leaf-feeding beetles released only Cassida sp.
appears to be persisting and reproducing but it is too early to say whether it has established and what its
impact might be. Herbicide treatment of sites where this agent has been released would be very unwise at
this stage and field experiments will be difficult until the range of the insect expands. On the basis of cur-
rent information all these beetles seem likely to react similarly to spraying. Glyphosate has very low direct
toxicity to insects so the main effect of spraying would be to deprive adult beetles and larvae of food. High
mortality of larvae on sprayed plants appears inevitable; adults might be able to move to C. monilifera that
was not killed by the spraying, perhaps increasing the effect of the spray by defoliating these surviving plants.
Although adults can fly they are seldom seen to do so and their ability to move in these circumstances is un-
known. If adult movement is ineffective they may still persist by feeding on seedlings that emerge after the
spraying, but it is not known how long they can survive without food. Under Australian field conditions
Cassida sp. has so far shown a low rate of increase, probably due to predation, and it seems likely that num-
bers would only recover slowly after spraying. If the beetles have a useful effect on C. monilifera then it
would be worthwhile to provide unsprayed areas as reserves, perhaps slashing the reserve plants to pre-
vent new seed production. Research on the beetles would be needed before appropriate size and spacing
of reserves could be determined. A final considera-
tion, common to all leaf-feeding biological control agents is that severe defoliation by the agent could re-
duce herbicide uptake so that the plant was not killed. Such severe defoliation over all the infestation would
in any case probably remove the need for herbicide treatment, but if occurring in patches it might increase
the incidence of mature *C. monilifera* surviving spraying.

**C. germana** This agent is widely established in NSW and spreading. Feeding on shoot tips by larvae of this agent has reduced flowering and seed production of *C. monilifera* at some sites (Holtkamp 1996). All life-stages of the moth are present throughout the year so, as with the beetles, it is not possible to time herbicide application to minimise losses of the agent. Starvation of larvae is again the most likely effect of spraying but *C. germana* adults seem to be reasonably mobile and therefore likely to locate surviving *C. monilifera* to lay eggs on. Adult *C. germana* feeding specificity is unknown; it is a multivoltine species with total lifecycle being six to nine weeks and adult lifespan approximately 5.5 days (Adair and Scott 1989). There is one report that following aerial spraying of *C. monilifera* at Port Kembla P in 1994 with glyphosate the subsequent seedlings were heavily attacked by *C. germana*; (J. Toth pers. comm.); possibly eggs were laid on these seedlings by moths emerging from pupae present at spraying, although movement into the sprayed area by adult moths could also be the explanation. On balance it appears that *C. germana* is unlikely to be lost from a site due to spraying and if present in large numbers before spraying may make a useful contribution to control of seedling regeneration. Field studies are needed to find out what is happening and to determine whether reserves would make any useful contribution when large areas are sprayed. The contribution of *C. germana* to success of chemical control by reducing the seedbank would be the same as *M. polana* below.

**M. polana** *M. polana* has spread and increased in numbers at a phenomenal rate since the first introduction in 1996 and is now present from Fraser Island, Queensland to Tathra, NSW and is reducing seed production substantially. *C. monilifera* seeds are relatively short-lived; only about 2% are ungerminated and still viable after three years (Weiss, 1984). *M. polana* should thus reduce the seedbank within a few years so that *C. monilifera* will be less invasive and control of seedling regeneration will be easier. Nevertheless, it is not likely that *M. polana* alone will greatly reduce existing infestations and chemical control will remain useful. Control by aerial glyphosate spraying typically involves three applications in alternate years at the end of which only a low rate of further seedling emergence is expected. A much smaller seedbank due to *M. polana* could allow control with two aerial treatments rather than one. One option is to monitor the seedbank over the next few years and if it seems to be decreasing at a site delay large-scale chemical control programmes until the maximum effect has been obtained. A delay might only be justifiable where *C. monilifera* is already dominant and little further damage to conservation values would result from postponing control. Due to a very high rate of population increase and highly mobile adults, major harmful effects of glyphosate spraying on *M. polana* seem unlikely. A simple field experiment was however conducted, to determine whether *M. polana* females would continue to lay eggs on glyphosate sprayed plants, thus wasting reproductive effort because the eggs would fail to develop into adults. Avoidance of sprayed plants would be the optimum outcome both to maintain the *M. polana* population and to place the maximum pressure on adjacent unsprayed *C. monilifera*. The question of whether herbicide-treated weeds can be detected and avoided at an early stage is central to the integration of many insect biological control agents with chemical control.

**MATERIALS AND METHODS**

The field experiment was carried out in September/October 1998 at Wilson’s Headland on the north coast of NSW in an area of dunes densely covered by *C. monilifera*. Ten pairs of 2 × 2 m plots were established and one of each pair was designated at random to receive the glyphosate treatment. One week before glyphosate application 10 shoots with developing flower buds in each glyphosate plot and 10 in each of the control plots were covered with fine gauze to prevent egg-laying by *M. polana*. Immediately before spraying all the bags were removed; thus all eggs on these shoots were known to have been laid post-spraying. Spraying was with glyphosate at 1:100 applied by backpack sprayer to fully wet all leaves, using approximately 0.25 litres of spray per plot. Flowers were sampled from each plot on the day of spraying and three, seven and fourteen days later. Samples on the day of spraying and three days later were taken by use of a crown sampler; all material within the sampler was used. Seven and fourteen days after spraying the number of flowers in glyphosate-sprayed plots was so low that few were obtained within the crown sampler and additional flowering shoots were collected to provide a sufficient number for estimating egg densities. The previously bagged material was removed at day seven. All material was preserved in vinegar for transport to KTRI then stored in alcohol. The number of eggs on each flower was counted by examining the preserved material under a binocular microscope.
RESULTS

Table 1 shows the mean number of eggs per flower or flower bud on each of the sample dates. Samples that had been bagged until the date of spray application showed a large and highly significant (P<.001) difference in density of eggs between sprayed and unsprayed plots, with the sprayed plots having far fewer eggs. The samples of unbagged shoots showed an initial difference between glyphosate and control plots before any herbicide was applied and this continued to the third day after treatment. Samples taken seven days after treatment showed the reverse, with approximately three times more eggs per flower from glyphosate-sprayed plots, and there was little change after a further seven days. The data for unbagged samples were analysed by analysis of repeated measures using the Systat 7 package. There was a highly significant (mainly linear) change in density of eggs over time and highly significant interactions between time and spray treatment, confirming the impression from Table 1 that the glyphosate and control plot egg density changes over time were different.

Table 1. Effect of glyphosate on density of *M. polana* eggs (mean eggs per flower with s.e. from 10 plots per treatment).

<table>
<thead>
<tr>
<th></th>
<th>Control</th>
<th>Glyphosate</th>
</tr>
</thead>
<tbody>
<tr>
<td>Pre-spraying</td>
<td>2.4 (.51)</td>
<td>1.2 (.20)</td>
</tr>
<tr>
<td>3 days</td>
<td>1.8 (.18)</td>
<td>1.3 (.29)</td>
</tr>
<tr>
<td>7 days</td>
<td>0.35 (.10)</td>
<td>0.99 (.99)</td>
</tr>
<tr>
<td>7 days (bagged)</td>
<td>1.58 (.14)</td>
<td>0.38 (.05)</td>
</tr>
<tr>
<td>14 days</td>
<td>0.45 (.09)</td>
<td>0.86 (.17)</td>
</tr>
</tbody>
</table>

DISCUSSION

Given the random allocation of plots to treatments the pre-spraying difference in egg densities can only be due to chance. Three days later there was little change; the drop in the control mean was mainly due to one plot with very high numbers of eggs in the pre-spraying sample having a lower number in the sample taken on day three. Seven days after spraying there appeared to be a dramatic switch, with far fewer eggs per flower from control plots and far more from glyphosate plots. Some of the possible explanations are: glyphosate could have affected the flowers or the sprayed bushes as a whole in some way that made them more attractive to the flies; glyphosate could have impaired egg development so that the number hatching was reduced, so more remained to be counted; or the sampling methods could have been biased in favour of collecting flowers with more eggs from the glyphosate plots.

Results from the samples that had been bagged until the day of spraying may provide some clue to what happened. Bagged shoots were chosen to be at the same stage of development seven days before the spraying and were selected before allocation of plots to different treatments. A reduced number of eggs from bagged shoots in the glyphosate plots tends to suggest that these plots or the flowers in them were actually less attractive to *M. polana* after spraying. None of the eggs on the bagged shoots were present at the time of spraying, so the possibility of delayed egg development due to glyphosate in the unbagged samples cannot be ruled out, but this is an unlikely explanation as there is no previous record of glyphosate acting in this way or any reason to expect that it would. Restricting the comparison to the bagged shoot samples may provide the best measure of *M. polana* oviposition response to glyphosate i.e. a strong avoidance, probably due to an ability to detect impaired health of the plant or maybe because a decreased density of flowers made the plant as a whole less attractive. Many buds and flowers had died in the glyphosate plots by day seven, and in making up the minimum sample size of unbagged material, flowers that had more eggs appear to have been collected. The surviving flowers in the glyphosate plots could have been at a different developmental stage than the majority of the ones collected in control plots. Most obviously, one would expect that buds might accumulate more eggs over the time from when they first become attractive to flies. So if older flowers survive glyphosate better than younger ones and the samples are therefore of older flowers than the control plot sample, then it might explain the higher density of eggs in the glyphosate plot samples. There could be other reasons for these surviving buds being the ones with most eggs e.g. a position on the plant that both avoided the spray and was favoured by the flies. More detailed work to record exact age, developmental stage and position on the plant of all the flowers sampled and the effect of glyphosate on different ages of flowers would be needed to resolve these questions.

The conclusions that can be drawn from this study are that (i) quite large differences in egg densities can exist between adjacent and apparently similar bushes, so greater replication or stratification by pre-treatment egg numbers would be helpful; (ii) for flowering shoots of the same age glyphosate treatment seems to decrease *M. polana* oviposition over the following seven days; (iii) for reasons not yet understood the flowers that persist after glyphosate treatment have on average a relatively high density of eggs.
This study illustrates the complexities involved in attempting to answer one apparently simple question regarding a plant-herbicide-biological control agent interaction. All the recent information on *M. polana* in Australia suggests that its abilities to locate *C. monilifera* and to breed quickly make it highly unlikely that diminished biological control would result from herbicide treatment. Other *C. monilifera* biological control agents, particularly *C. germana* would repay further study to determine whether altered herbicide strategies could obtain a better combined effect.

REFERENCES


DISCLAIMER

The advice provided in this publication is intended as a source of information only. Always read the label before using any of the products mentioned. The State of Victoria and its employees do not guarantee that the publication is without flaw of any kind or is wholly appropriate for your particular purposes and therefore disclaims all liability for any error, loss or other consequence which may arise from you relying on any information in this publication.