

Herbicides compromise biological control of ragwort

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Abstract

In spite of ragwort flea beetle (RFB) being present on a Dannevirke dairy farm, pastures were sprayed each winter to reduce ragwort density and limit the risk of ragwort poisoning of stock. The trial on this farm from June 1999 – October 2001, aimed to determine whether herbicide (H) impacted on RFB and how H and RFB each impacted on ragwort growth and persistence. RFB-free areas were created by spraying with insecticide (I). Effects of ragwort on animal health are also reported.

High ester 2,4-D (H) boom-sprayed once only, in June 1999, killed most ragwort plants and reduced RFB larvae densities to low levels before the plants died. Once new ragwort established in treatment H, the plants became infested with RFB larvae. RFB larvae were suppressed by I resulting in ragwort density declining more slowly than in treatments where RFB were not suppressed. Insecticide treatments were stopped after 15 months and, at 24 months, ragwort could not be found within the trial area. Ragwort control was attributed to the cessation of herbicide spraying allowing the RFB population to reach a sufficient density to kill both small and large ragwort plants. Sub-clinical ragwort poisoning was found in livers of culled cows that had grazed on ragwort-dense pastures.

Keywords: animal health, biological control, *Longitarsus jacobaeae*, pyrrolizidine alkaloids, ragwort, ragwort flea beetle, *Senecio jacobaea*

Introduction

Ragwort (*Senecio jacobaea* L.) has been a wide-spread and toxic weed on dairy, beef and deer farms in most areas of New Zealand, and has been the target of intense efforts by farmers to remove it from pastures. Regional Council Plant Pest Officers report that ragwort flea beetle (RFB) has resulted in excellent ragwort control in many areas of New Zealand but in others and/or on some individual farms, RFB control has been negligible.

Ragwort control has traditionally been achieved with herbicides or by sheep grazing. Herbicide control is expensive and on-going, whereas most sheep readily graze ragwort (Betteridge *et al.* 1997) without becoming poisoned. Conversely, even small amounts of ragwort are highly toxic to cattle (Cheeke 1998) where the pyrrolizidine alkaloids (PAs) in ragwort cause cumulative, irreversible liver damage both to livestock and can lead to death.

McEvoy & Rudd (1993) have shown that RFB

(*Longitarsus jacobaeae* (Waterhouse)) reduced ragwort biomass in an Oregon trial by up to 99% within 4 years. Biocontrol with RFB has been spectacular on many New Zealand farms, but on some, where RFBs have established, ragwort still persists. Egg laying by RFB commences in southern Hawkes Bay from early March and declines from around May to very low levels in early spring (Page 2000). RFB larvae can be found in ragwort leaf petioles, stems and roots from around April. They then pupate in the soil after developing through three instars. In this region there may be more than one RFB life cycle per year (Page 2000) although RFB are commonly thought to emerge only in late spring (Philip & Syrett 1988). Most ragwort damage is done by larvae, and it is during this period that farm management practices may impact on them. The aims of this study were to compare herbicide and RFB control of ragwort and to find if 2,4-D reduced RFB efficacy. Liver histology results from culled cows, blood profiles (indicators of health status) from two herds, and composite milk PA concentration from the herd exposed to, the herd protected from ragwort were measured to determine whether alkaloids were impacting on cow health or potentially entering the food chain.

Materials and methods

Site and treatments

The trial was on a dairy farm near Dannevirke, southern Hawkes Bay, on which ragwort was extremely dense even though RFB had been established there 5 years earlier. The farmer sprayed all pastures with 2,4-D herbicide each winter and spot-sprayed new plants with Preside® during summer to reduce the risk of poisoning cows and to increase the grazing area.

Using 10 m square plots within four adjacent blocks (replicates), four treatments were applied: (1) herbicide alone (H) to kill young ragwort; (2) insecticide alone (I) to determine ragwort growth in the absence of RFB; (3) treatment H+I helped estimate the effect of RFB on seedlings that established after the winter application of herbicide; and (4) neither H nor I (Control). In treatments H and H+I herbicide (2,4-D 3l/ha) was boom sprayed once only, in June 1999, with no further treatments being applied to later germinating plants. In treatments I and H+I, insecticide Orthene® (100g/100l) was sprayed once in June and once in July 1999, after which Hallmark® was sprayed 3-weekly through to August 2000. Ragwort

density was determined by using as many randomly placed 0.5 m² quadrats as required to locate at least 10 ragwort plants (including seedlings). RFB larval numbers on whole ragwort plants (root+top) were counted in the laboratory by exposing washed plants to a temperature gradient for 48 hours, so that larvae moved towards and fell through a funnel into cool water, from which they were counted (Page 2000). These plants were then dried at 80°C for 12 hr and weighed. Adult RFB density/plant was determined by sucking beetles from 10 moderate – large ragwort plants within each plot, using a motorised leaf blower. Ragwort seeds were extracted (AgriQuality Seed Testing Station) from thirty (25 mm diam. and 25 mm deep) soil cores from within the trial area. Potential seed set was estimated by counting flower buds, flowers and mature seed heads of two single-stem and one 6-stemmed ragwort plants, in the laboratory, in February 2001 and relating these to ragwort plant numbers in each treatment. Each flower was estimated to produce 66 seeds.

Statistical analysis

Blocks were combined into single values/treatment because of the large number of zero values in the plant, RFB and RFB larval data. Plant density was analysed by log linear regression analyses; RFB/plant by general linear regression of $\log(x+1)$ transformed data and RFB/g DM by regression of $\log(x+1)$ data using Genstat 6.0. Plant density was the dependent variable and treatment and date were the independent variables. Treatments are

significantly different where 83% confidence interval (CI) bars between pairs of mean values do not overlap (Ms Z Park-Ng pers. comm.).

Animal health and milk

In autumn 2000, two livers, and in autumn 2001 eight livers from slaughtered cull cows, selected by the farmer as poor producers, were histologically examined for PA lesions (Dr F. Hill, Animal Health Laboratory, Palmerston North). In autumn 2001, blood samples from 10 cows on the ragwort-infested farm and 10 cows from a nearby farm without ragwort were screened for pepsinogen, albumin, gamma globulin and total protein (Animal Health Laboratory, Palmerston North). One whole-herd composite milk sample was collected from each of these farms for PA analysis, using a high pressure liquid chromatography-mass spectrometry technique (Dr S. Colegate, CSIRO Animal Health, Geelong, Australia).

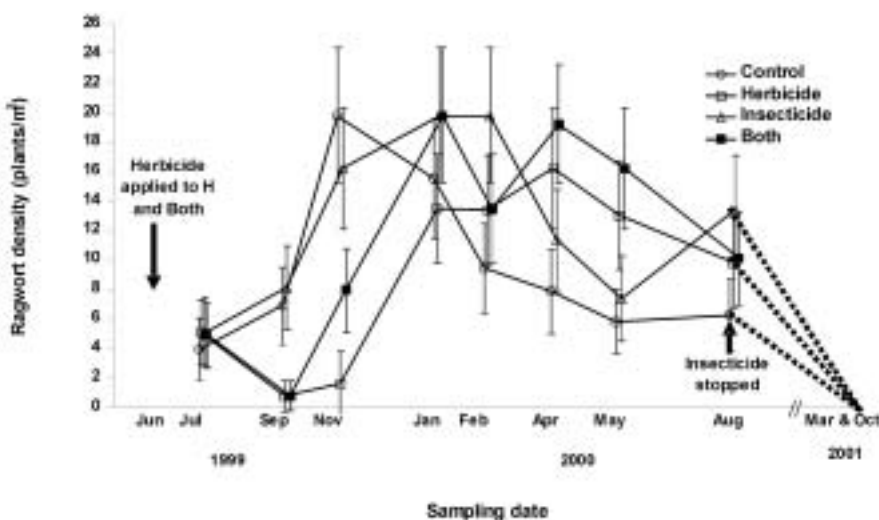
Results

Mean daily maximum and minimum air temperatures were 26°C in February and 4°C in June respectively. Average annual rainfall is 1500 mm.

Ragwort density

There was a significant date by treatment interaction (Figure 1; $P < 0.01$). From July 1999 to November 1999, ragwort density (all growth stages) increased substantially but was unaffected by RFB in either the

Figure 1 Back-transformed mean (83% CI) ragwort density (including seedlings) in pastures where ragwort was sprayed with 2,4-D herbicide (H, June 1999), or sprayed with insecticide (I) at 3-4 week intervals.



In Figures 1-3, treatments are significantly different where 83% confidence interval (CI) bars between pairs of mean values do not overlap. Sampling dates have been off-set to more clearly reveal CI bars.

Control (RFB present) or in I-alone-treated plots (few RFB larvae; Figure 3). By contrast, in February 2000 plant density in Control declined more quickly than in I. In treatments H and H+I, herbicide initially removed ragwort from the pasture but ragwort density (predominantly seedlings) then increased during the next year to give a greater plant density in H and H+I than in Control and sometimes in I (Figure 1). In November 1999 H had fewer plants than H+I – a non-significant trend continuing through to May 2000. In February 2000, most ragwort plants in H and H+I treatments were seedlings, whereas in Control and I there were also many single- and multi-stemmed flowering plants. Neither in March nor in October 2001 (end of trial) could ragwort be found, and in May 2002 (data not shown) few ragwort plants of any age remained in the trial area or the surrounding paddocks.

RFB per plant

There were 3-4 adult RFB per plant in late March and May 2000 in Control and H compared to <1 per plant in I and H+I. Few beetles were present in August 2000 (Figure 2). Control (March and May) and H (May) had more RFB than either I or H+I treatments ($P < 0.01$). The insecticide treatments removed many, but not all, RFB from ragwort.

September 1999, but in July 2000 rose in Control and H whereas they remained very low in I and H+I, showing the effective RFB control by Hallmark®. During the trial period the mean larval density (regression of log mean number larvae/plant) was higher for Control and H than for treatments including insecticide ($P < 0.001$).

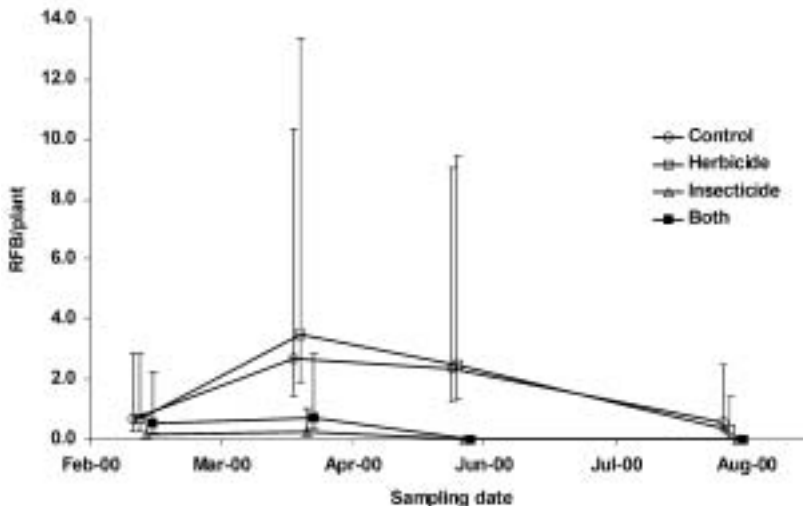
Potential seed set and buried seed

Two single-stemmed and one multi-stemmed plants, sampled in February 2001, were estimated to be capable of producing 25 000, 62 000 and 279 000 seeds respectively. In December 1999, buried seed in the top 3 cm averaged 7 700/m² (range: 1 000- 40 000) and averaged 9 000/m² in December 2001.

Animal health

Mean assays of the four blood parameters of 10 cows from the herd exposed to ragwort-dense pastures and from the neighbouring herd eating pastures without ragwort were similar and within the 'normal range' (data not presented). Histological examination of livers of all 10 cull cows from the problem ragwort farm, showed lesions consistent with exposure to mild to moderate exposure to PAs (Hill 2000). Milk samples contained no PAs (Dr S. Colegate, pers. comm.).

Figure 2 Back-transformed mean (83% CI) number of adult RFB/plant in contrasting treatments.



RFB larvae/g ragwort dry matter

There was a non-significant trend for mean larval number to be higher in Control in July 1999 (7.2/g DM, range 0-25.8; Figure 3) than in H. In July 1999 the I-alone (Orthene®) treatment was only partially effective (2.4/g DM) in removing larvae, whereas H alone reduced larvae to only 1/g DM. Numbers were generally low in

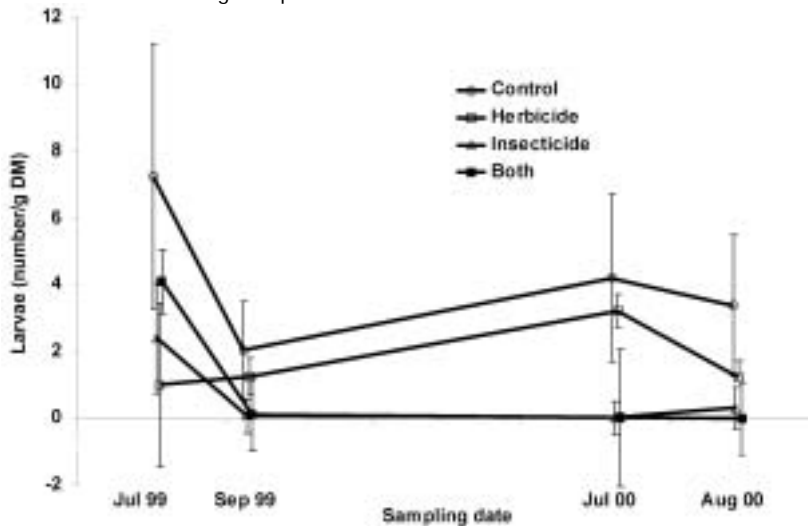
Discussion

RFB appears to have removed ragwort, older than seedlings, from pasture within 2 years of stopping the use of 2,4-D herbicide application (H and H+I). This occurred in spite of the high density of ragwort 9 months earlier. Over the same period, ragwort had ceased to be a problem across the whole farm following the cessation of winter 2,4-D spraying. During the 5 years prior to

this trial, winter application of 2,4-D was an annual event that, in concert with RFB, had no apparent long term effect. The comparatively mild climate at the site was unlikely to have affected RFB's efficacy as RFB are effective in much cooler and high altitude zones and in very warm low altitude zones in Oregon (Dr D. Isaacson pers. comm.).

We believe that the regular use of 2,4-D herbicide prevented the RFB population reaching a critical density needed to control ragwort, as one month after the single herbicide application, the larval density in H-sprayed plants was greatly reduced, compared to Control plants. The herbicide impact on larvae was apparently a direct effect, rather than one mediated through loss of RFB's food source, since H-treated plants took at least 2 months to die. This herbicide appeared to have been as toxic to larvae as Orthene® (Figure 3), although subsequent treatment with Hallmark® gave better RFB larvae control.

Figure 3 Back-transformed mean (83% CI) number of RFB larvae (n/g DM) in rosette ragwort plants.



Adult beetles were also deterred from visiting Hallmark-sprayed ragwort. Had herbicide continued to be used each winter, as many farmers desire, or are required by Regional Councils to do, we believe that biocontrol of ragwort would never have been achieved.

The declining ragwort density in Control from November 1999 to May 2000 would have been due to both the natural death of plants following seeding and the influence of RFB, since ragwort density in Control declined sooner than in insecticide treatments where RFB were absent. The more rapid rise in plant density in November 1999 and to a less extent in January 2000 in the herbicide treatment without RFB (H+I), than in H, suggests that RFB in the latter were to some extent limiting the establishment of small ragwort plants. This is

supported by the greater number of RFB/plant (April and May 2000) and RFB larvae/g DM (July and August 2000) in plants not sprayed with insecticide.

The peak in RFB density/plant in April-May 2000, preceding the July peak in larval density, is consistent with results reported from a nearby trial by Page (2000), yet it is in late autumn-early winter that farmers often apply herbicide for ragwort control.

By August 2000, in spite of the indications of a RFB effect described above, there was no sign that ragwort was soon to disappear from the pastures. Although there were no large ragwort plants in plots sprayed with 2,4-D 15 months earlier, even the small ragwort plants that established following herbicide use were not being greatly impacted by RFB. Yet by March 2001, ragwort could not be found in any treatment. As there was no ragwort present at this time, it was not possible to measure larval or adult RFB densities. However, on a few plants found

nearby, both larvae and adult RFB were found. Thus, while it appears clear that winter 2,4-D herbicide application to pastures will prevent the RFB population reaching some threshold density needed to kill ragwort, it was impossible to prove this point with the use of the single herbicide application at the start of the trial. Further, because the farmer stopped the herbicide boom-spraying of his whole farm at the same time, it is probable that the RFB population in the surrounding pastures also increased. If this was so, it was not apparent in the larval density count in August 2000, compared to count in the previous September.

The highest average larval density/g DM (7.2 in Control plants) was found in July 1999, when the range amongst the 20 plants was 0 to 28.4 larvae/g DM. One large, multi-stemmed, rotting plant elsewhere on this farm had only 0.7 larvae/g DM, while another dying plant had only 0.28 larvae/g DM. The apparently low larval populations which resulted in the eventual disappearance of ragwort from pastures in this trial contrast with the mean population density of 19.6 larvae/g DM in 1987 reported by McEvoy & Rudd (1993) that resulted in the near extinction of ragwort cohorts in 1988. By contrast, they reported that the density of only 1.2 larvae/g DM in

plants with near-complete protection from biocontrol insects in 1987, increased to only 3.6 larvae/g DM one year later, and the ragwort in their Oregon trial remained in the pasture. Our infrequent sampling may have missed the highest RFB larvae density that occurred prior to ragwort's disappearance, but it seems that a density of <10 RFB larvae/g DM indicates that ragwort will soon be killed, providing that herbicides are not used.

The mean 9 000/m² ragwort buried seed count in December 2001, arising from many years of very high potential seed set by individual mature plants, indicates that seedling ragwort will continue to appear for many more years. James & Rahman (2000) reported ragwort viability of buried seed in the 0-2 cm soil horizon declined to 1% after 10.9 to 14.6 years, reaching this low viability sooner in wetter soils.

The wide range in confidence intervals for many of the measured mean values in this trial indicates the highly variable distribution of ragwort in the field and of RFB larvae and adults amongst ragwort plants. High variability in ragwort density, that included seedlings, was undoubtedly due to the high potential seed set from unevenly distributed mature ragwort plants within the pasture. The highly variable larval density amongst plants may reflect the finding that oviposition by RFB is independent of RFB eggs already present on a plant (Page 2000). This variability may be accentuated where there are insufficient RFB to visit and oviposit all plants in a pasture community containing a high ragwort density. Similar weed and biocontrol studies should establish more than the four treatment replications we used, in order to detect significant differences that appeared only as trends between treatments in this trial.

While the biocontrol agent cinnabar moth (*Tyria jacobaeae*) was present during December and January each year, it was found close to hedge rows and its impact on ragwort survival was negligible. It is unlikely that winter herbicide application impacted on cinnabar moth larvae as they are only found on ragwort in summer and they typically pupate under debris on the soil surface (Syrett 1983), wood lots and hedges, so would not be exposed to winter-sprayed herbicide. Research in north-western USA found that cinnabar moth causes little reduction in ragwort density (McEvoy & Rudd 1993).

Animal health and milk quality

The culled cows were selected solely on their low milk production, with none displaying clinical signs of ragwort toxicity, yet each had histological lesions in the liver, consistent with ragwort poisoning. Given the high density of ragwort across this farm, it is likely that cows would have consumed some ragwort while grazing pasture. Because ragwort poisoning is cumulative and irreversible (Cheeke 1998) the liver damage observed

might have occurred at any stage during the cows' life and most likely would have contributed to reduced milk production. Without histological examination of the liver samples, casual observation of the damaged liver might easily have been attributed to facial eczema (Dr F. Hill, pers. comm.). The fact that the blood profiles were 'in the normal range' from cows in the herd exposed to ragwort may reflect a very low frequency of subclinical ragwort toxicity within the herd, or that the tests were insensitive to this level of poisoning. It therefore seems unlikely that dairy farmers can do a simple blood test to detect sub-clinical ragwort poisoning in their herds.

Although no ragwort alkaloids were detected in the single milk sample from cows on this farm, it cannot be assumed that milk at other times of the year, or milk from other farms with ragwort, will not be contaminated with PAs. But, because cows are so susceptible to PA poisoning (Cheeke 1998) and because they avoid eating ragwort wherever possible (personal observation), it seems unlikely that these toxic alkaloids will show in milk without clinical symptoms also showing in the herd.

Conclusion

To enable RFB to become the primary method of ragwort control on farms the farmer must first establish a population of these beetles. The use of boom-sprayed 2,4-D should be stopped, at least in one or two paddocks, to allow the RFB population to establish and expand. Once ragwort is being controlled in these paddocks, then herbicide spraying of surrounding paddocks should also cease. At all times a high pasture density is required to ensure other weed species do not establish in the gaps left by ragwort.

ACKNOWLEDGEMENTS

We gratefully acknowledge Mr & Mrs V. Payne who provided pastures for the trial, co-operated fully with our requests and showed great interest in these studies. Dr Steve Colegate, Plant Toxins Unit, CSIRO Animal Health, Victoria, Australia, kindly analysed the milk samples. Dr Fred Potter, AgResearch, provided valuable statistical assistance. This research was funded by the New Zealand Foundation for Research Science and Technology.

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