Glyphosate resistance in mobile weeds across land uses: implications for area wide management of weeds

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Summary Weeds that have the ability to rapidly disperse across the landscape can create ongoing problems for individual land managers. Efforts by land managers to reduce the risk of a weed problem can be negated by such mobile weeds moving across boundaries. When mobile weeds are easy to control using current practices, there is little incentive for more coordinated action among land managers. However, for more difficult to control weeds (or resistant weeds) there may be a case for collective action through an area wide weed approach. This project examined the distribution of glyphosate resistant weeds at a local scale in the Sunraysia, the Riverina and the Darling Downs. Based on local land manager consultation, the study focused on the three most concerning mobile weeds: *Conyza bonariensis* (flaxleaf fleabane) in the Sunraysia, flaxleaf fleabane and *Lolium rigidum* (annual ryegrass) in the Riverina and *Chloris virgata* (feathertop Rhodes grass) in the Darling Downs. Geo-referenced weed seed samples, plants in the case of annual ryegrass, were collected from a variety of land uses in each district in 2019-2020 and 2020-2021, including public land and roadsides, and the samples tested for resistance to glyphosate. Samples with survivors to glyphosate were considered to be resistant. High frequencies of glyphosate resistance were identified in all weeds and in all districts ranging from 5% of flaxleaf fleabane samples in the Sunraysia in 2021 to 81% of annual ryegrass samples in the Riverina in 2021. Spatial patterns of resistance status were analysed for each weed and region. Resistance occurred across the area sampled and was present on multiple land uses, including non-farm land. As land managers from multiple sectors already need to manage these resistant weeds, the incorporation by all land managers of practices that reduce seed production, and hence potential for spread of these weeds would be beneficial.

Keywords Area wide management, mobile weeds, glyphosate resistance.

INTRODUCTION

Area wide pest management (AWM) is an approach to pest management where the pest is managed collectively across a geographic area, rather than treating individual fields for pests. AWM offers benefits through all populations of the pest within the geographic region being suppressed, rather than allow some populations to expand and risk infesting already treated or new areas (Kruger 2016). AWM has been used successfully for the management of many insect species, including *Helicoverpa armigera* Hübner and various fruit flies in Australia (Fitt 2000, Lloyd et al. 2010). AWM is most feasible where pest populations are mobile and there is an economic penalty from the pest on a large number of land holders (Lloyd et al. 2010, Kruger 2016).

There are few examples of AWM being used for weeds. Most AWM programs involving weeds are eradication programs where there are additional regulatory options for ensuring cooperation (Panetta 2014). Some of the challenges of employing AWM for weeds are the view that weeds are not very mobile, the perception that weeds are relatively easy to manage and land managers often managing a suite of weed species (Ervin and Frisvold 2016). In addition, area wide management is logistically complex (Kruger 2016).

Glyphosate is a herbicide widely used across many land uses in Australia. The intensive use of glyphosate has resulted in the evolution of resistance to this herbicide in 21 weed species in Australia (Heap 2022). Resistance occurs in situations where glyphosate is repeatedly used including grain production, horticulture, irrigation channels and roadsides (Preston 2010a). Mobile weed species with glyphosate resistance are likely to be challenging for land managers to control. Weeds such as these would be ideal to explore the benefits of AWM for weeds.

In this study, localised physical surveys in the Sunraysia, Riverina and Darling Downs of weed species were conducted to identify the proportion of weeds resistant to glyphosate and their location across land uses.

MATERIALS AND METHODS

Sample collection Weed species sampled were identified following consultation with land managers in each of the regions to identify mobile weeds of concern for land managers to control. Surveys focused on *Conyza bonariensis* (L) Cronq. (flaxleaf fleabane) in the Sunraysia, flaxleaf fleabane and *Lolium rigidum* Gaud. (annual ryegrass) in the Riverina and *Chloris virgata* Sw. (feathertop Rhodes grass) in the Darling Downs.

During 2019-2020, project participants in each of the regions collected seed heads from individual, geo-referenced plants of flaxleaf fleabane and feathertop Rhodes grass and sent these to the University of Adelaide for testing. For annual ryegrass in the Riverina, 6 to 10 plants from a geo-referenced location were collected when well-tillered and sent to the University of Adelaide for testing. A second collection of weeds was made in 2020-2021 from the Sunraysia and Riverina only.

Testing for resistance to glyphosate Seed of flaxleaf fleabane and feathertop Rhodes grass were sown onto the surface of potting mix and watered. Once the seed had germinated, plants were transplanted into 0.55 L square pots (Masrac, SA) containing potting mix with 5 plants per pot, and grown outdoors at the Waite Campus, University of Adelaide (Boutsalis et al. 2012). At the 6 to 8-leaf stage for flaxleaf fleabane and 4 to 6-leaf stage for feathertop Rhodes grass, duplicate pots for each sample were treated with 1080 g a.e. ha^{-1} glyphosate (WeedMaster® Argo®, 540 g L^{-1} glyphosate present as the potassium and isopropylamine salts, Nufarm Australia) using a moving boom laboratory spray cabinet (Boutsalis et al. 2012). At 28 days after application survival was assessed. Samples with survivors were recorded as resistant and samples where all plants were killed were recorded as susceptible. Susceptible control populations were completely controlled by this rate of glyphosate.

Annual ryegrass resistance to glyphosate was assessed using the Quick Test method (Boutsalis 2001). Plants of annual ryegrass were divided into 3 pieces containing 2 to 4 tillers each. The plants were trimmed and potted into 0.55 L square pots containing potting mix. The three pieces of each plant were placed in the same position in three pots. A week after transplanting, when plants had produced new green leaf tissue, two pots of each population were treated with 540 g ha^{-1} glyphosate as above. The final pot was an untreated control. A single pot of susceptible annual ryegrass plants was also treated with glyphosate. Plants were assessed for survival at 28 days after treatment. Samples with survivors were recorded as resistant and samples where all plants were killed were recorded as susceptible.

Spatial pattern of resistance Resistant and susceptible samples for each of the regions were plotted on maps using the geo-references. The land use was recorded at the time of collection. Further analysis of spatial patterns was supplemented by other freely available datasets like land cover, transport and hydrology networks.

RESULTS

Not all samples collected could be tested for resistance to glyphosate. Some seed samples of flaxleaf fleabane and feathertop Rhodes grass failed to germinate. In addition, some annual ryegrass samples failed to grow once transplanted.

Resistance to glyphosate was identified in each of the weed species tested in 2020 (Table 1) and in 2021 (Table 2). There were differences in the percentage of glyphosate resistant samples within a region between years. Glyphosate resistance status by land use was analysed for fleabane in the Riverina and resistance was found across the range of land uses. This includes samples from within agricultural land (crop, orchard, vineyard) where 60% of samples were resistant and from the edge of roads, tracks and channels where 54% of samples were resistant.

Flaxleaf fleabane was sampled in Sunraysia and Riverina in both years. In 2020, the frequency of glyphosate resistance was high in both regions with 42% of samples tested from the Sunraysia resistant and 64% of tested samples from the Riverina resistant (Table 1). The frequency of resistance in this species was lower in both regions in 2021 (Table 2).

Table 1. Percentage of tested weed samples resistant to glyphosate in each of the weed species collected in Sunraysia, Riverina and Darling Downs during 2019-2020.

Region	Weed species	Samples tested	Samples with resistance to glyphosate (%)
Sunraysia	Flaxleaf fleabane	50	42
Riverina	Flaxleaf fleabane	64	64
	Annual ryegrass	18	67
Darling Downs	Feathertop Rhodes grass	36	50

Table 2. Percentage of tested weed samples resistant to glyphosate in each of the weed species collected in Sunraysia and Riverina in 2020-2021.

Region	Weed species	Samples tested	Samples with resistance to glyphosate (%)
Sunraysia	Flaxleaf fleabane	55	5
Riverina	Flaxleaf fleabane	57	37
	Annual ryegrass	16	81

Annual ryegrass was sampled in the Riverina in both years. The frequency of glyphosate resistance was high in both years of sampling, 67% of samples in 2020 (Table 1) and 81% in 2021 (Table 2). Feathertop Rhodes grass was sampled only in the Darling Downs in 2020 and 50% of the samples were resistant.

The spatial distribution of glyphosate resistant samples of each species was dispersed across the collection region and interspersed with susceptible samples. Figure 1 illustrates the distribution of flaxleaf fleabane in the Riverina in the two years. Resistant samples of each weed species were found across all land uses.

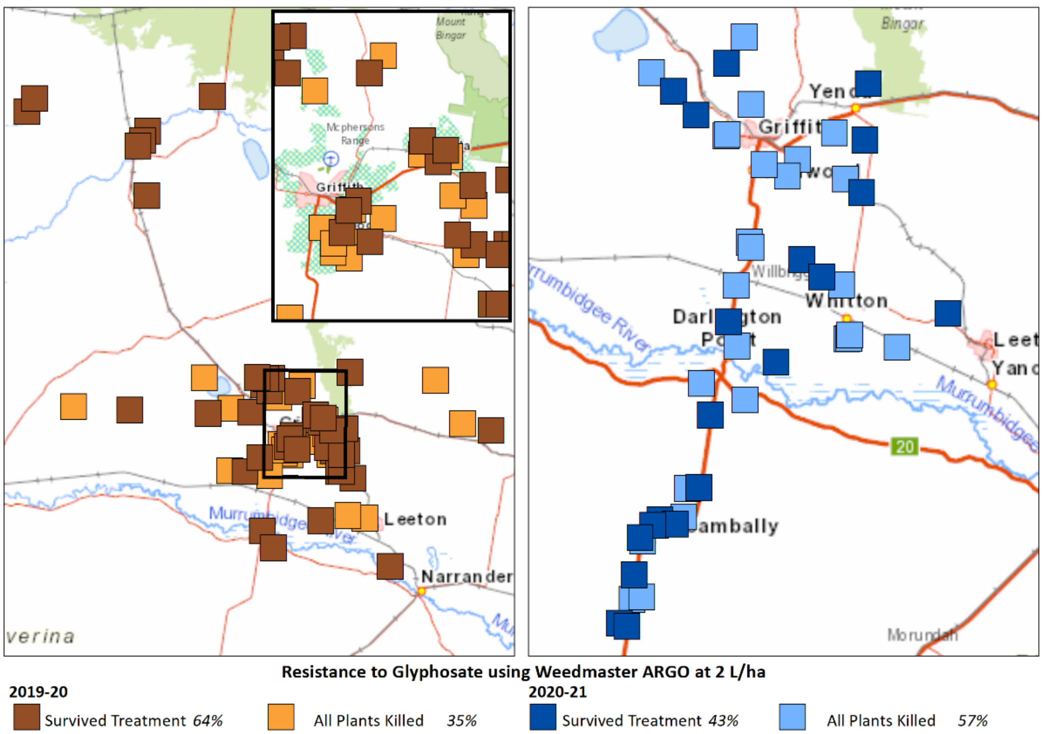

Figure 1. Distribution of glyphosate resistant samples of flaxleaf fleabane (dark symbols) and susceptible samples (light symbols) in the Riverina collected in 2019-2020 (Left) and 2020-2021 (Right).

DISCUSSION

High frequencies of resistance to glyphosate in flaxleaf fleabane, annual ryegrass and feathertop Rhodes grass was identified in the localized surveys conducted here. Glyphosate resistant weed samples were dispersed across the landscape and occurred in multiple land uses. These are all relatively mobile weed species where glyphosate resistance had previously been identified in multiple land uses (Preston 2010b, Malone et al. 2012, Ngo et al. 2018, Aves et al. 2020).

There were variations between years in the frequency of glyphosate resistant samples identified, particularly for flaxleaf fleabane. In 2019-20, 42% of all flaxleaf fleabane samples collected in Sunraysia were resistant to glyphosate (Table 1), but only 5%

of samples collected in 2020-2021 (Table 2). While sampling was conducted across a localized area, the same geo-referenced locations were not sampled in both years (Figure 1). Sunraysia had a severe drought in 2019 resulting in fewer weeds being present and those were concentrated along water courses and road sides. Considerably more rainfall occurred in 2020 resulting in a more dispersed distribution of weeds. This suggests that some land uses may have higher likelihood of glyphosate resistant summer weed occurrence, which is being further investigated.

As glyphosate resistance in each of the three regions sampled occurred across multiple land uses, many land managers are already having to manage glyphosate resistant weeds. Land managers who fail to control these glyphosate resistant weeds on their land could have populations increase and spread to neighbouring land (Hereward et al. 2022). This creates an opportunity for collective management where all land managers could reduce seed set of the weeds and thereby reduce spread of glyphosate resistance. The challenge will be to encourage all land managers to participate and demonstrate the benefits that could arise from joint action (Height et al. 2022).

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